

Indian Journal of Plant Genetic Resources

SPECIAL ISSUE

Genetic Resources Management: Road Ahead

*An International Journal for
Conservation and Use of Plant Diversity*



Indian Society of Plant Genetic Resources
New Delhi, India

INDIAN SOCIETY OF PLANT GENETIC RESOURCES

(Registration No. S/18336)

The Society was founded in 1987 with the following objectives:

- To serve and promote the scientific cause and to advance academic interest in the field of plant genetic resources.
- To disseminate knowledge relating to various aspects on plant genetic resources.
- To provide a forum for organizing symposia/conferences with a view to develop close relationship among the scientists engaged and interested in plant genetic resources activities.

Indian Journal of Plant Genetic Resources, the official publication of the Society, is published thrice in a year. The contribution to the journal, except for invited papers, is open to the members of the society only.

Membership to the society is open to all the individuals/institutions interested in various aspects of plant genetic resources. The membership fee is as follows:

Membership*	Inland	Foreign
Life Member	Rs. 5,000	US\$ 1,500
Annual Member	Rs. 1,000	US\$ 100
Institutional (Annual)	Rs. 20,000	US\$ 1,000
Institutional (5 Years)	Rs. 90,000	US\$ 4,500
Institutional (10 Years)	Rs. 1,75,000	US\$ 8,500

Issues of the journal published earlier are also available.

Limited space is available for advertisement of interest to botanists/geneticists/plant breeders and all those concerned with plant genetic resources.

CORRESPONDENCE relating to membership, advertisement and other related matters should be addressed to the General Secretary, Indian Society of Plant Genetic Resources, ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110 012, India (E-mail: ispgr2015@gmail.com).

COMMUNICATIONS regarding the publication of research papers should be addressed to Editor-in-Chief, Indian Journal of Plant Genetic Resources, Indian Society of Plant Genetic Resources, C/o ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110 012, India (E-mail: ispgr2015@gmail.com).

ISSN: 0971-8184
Online ISSN: 0976-1926

Indian Journal of Plant Genetic Resources

Vol.35 No.3 2022

80

Dr RS PARODA BIRTH ANNIVERSARY
SPECIAL ISSUE

Special Issue GENETIC RESOURCES MANAGEMENT: ROAD AHEAD

Editors

Sunil Archak
Anuradha Agrawal
Anil K Saxena
Kuldeep K Lal
BP Mishra
M Nagesh



INDIAN SOCIETY OF PLANT GENETIC RESOURCES

NBPGR CAMPUS, NEW DELHI-110012, INDIA

Web: ispgr.nbpgr.ernet.in

INDIAN SOCIETY OF PLANT GENETIC RESOURCES

Patrons	MS Swaminathan, RS Paroda, Mangala Rai
Honorary Fellows	MS Swaminathan, GS Khush, S Rajaram, RS Paroda, Mangala Rai, Emile Frison, RB Singh, SK Datta, HY Mohan Ram, KS Gill

EXECUTIVE COUNCIL (2022-24)

President	RS Paroda (New Delhi)	
Vice Presidents	RK Tyagi (New Delhi); Jai Chand Rana (New Delhi)	
General Secretary	Anuradha Agrawal (New Delhi)	
Joint Secretary	Manjusha Verma (New Delhi)	
Treasurer	Sanjeev Kumar Singh (New Delhi)	
Ex Officio Members	Immediate Past President, ISPGR; Director, ICAR-NBPGR	
Councillors	North Zone : Monika Singh (New Delhi)	Kuldeep Tripathi (New Delhi)
	East Zone : Mohan Lal (Jorhat)	Veerendra Kumar Verma (Umiam)
	West Zone : Sandip K. Bera (Junagadh)	Anil Patidar (Jaisalmer)
	South Zone : N. Sivaraj (Hyderabad)	Rose Mary Francies (Thrissur)
	Central Zone : Shailesh Tiwari (Varanasi)	Surendra Kumar Barpete (Madhya Pradesh)

Editor's Note

Indian agricultural researchers are facing enormous challenges of enhancing the agricultural productivity in the face of climate change, achieving nutritional security, increasing the farm income and ensuring the soil health – simultaneously. Genetic resources can provide practical solutions to many problems, provided the resources are conserved, characterized and accessible.

Genetic resources management is inherently inter-disciplinary. Each activity of the management – augmentation, quarantine, conservation, characterization, evaluation, use, etc. – requires trained human resource with specialized skill sets and adequate infrastructure. Activities need to be institutionalized and supported by suitable policy structure. India had been fortunate to have visionaries who led the genetic resources programmes from nascent stage with aplomb. From time to time, responding to the global developments, India expanded the genetic resources management programme from a small unit at IARI, New Delhi to a full-fledged National Bureau of Plant Genetic Resources followed by four other Bureaus on animal, fish, microbes and insect genetic resources. Contributions of many researchers and policy makers have made this growth and development possible.

The person who provided the leadership to genetic resources setup in India at critical junctures is Dr Rajendra Singh Paroda. He was instrumental in orienting the genetic resources research and genebanking in India to global standards and scale. In all his professional capacities, he has been an institution builder (National Genebank at NBPGR, *inter alia*, bears testimony); encouraged and supported capacity building and trainings; left no stone unturned to garner funding support for conservation activities in India and globally; advocated preparedness and inclusive deliberations to develop operational policies; organized numerous conferences, symposia, brain-storming, etc. on various topics to ensure researchers meet, network and collaborate.

He founded the Indian Society of Plant Genetic Resources and started Indian Journal of Plant Genetic Resources (IJPGR) as a publishing forum for plant genetics resources researchers, when there was no dedicated journal in the world! The IJPGR would like to celebrate the 80th birth anniversary of the visionary personality in a manner befitting his enormous contributions over the last five decades. This special issue “Genetic Resources Management: Road Ahead” is published in Dr Paroda’s inimitable style of brevity and focus on challenges and action points. Leading researchers from across the domains have contributed to this issue of IJPGR on diverse topics, making it a collector’s edition.

I acknowledge the role of my fellow editors Dr Anuradha Agrawal (plant genetic resources section), Dr BP Mishra (animal genetic resources section), Dr Kuldeep K Lal (fish genetic resources section), Dr Anil K Saxena (microbial genetic resources section) and Dr M Nagesh (insect genetic resources section) in accomplishing a daunting task of putting together an impressive collection of articles.

We believe that this special issue meets the expectations of Dr Paroda and all the readers.

Sunil Archak
Editor-in-Chief

CONTENTS

INTERVIEW			
Genetic Resources Must be Accessible and Used: Dr RS Paroda	SUNIL ARCHAK	...	1
Reorienting the Plant Genetic Resource Management for Enhanced Utilization	TRILOCHAN MOHAPATRA and SUNIL ARCHAK	...	8
Farmers' Rights and Plant Breeders' Rights under Indian Regulations of Plant Variety Protection: A TRIPS Compliant Harmonised Coexistence	RAJ GANESH, ARUN KUMAR, DS PILANIA, AK SINGH, DR CHOUDHURY, TK NAGARATHNA, DK AGARWAL and KV PRABHU	...	13
Climate-Ready Genebanks for Climate-Smart Agriculture	SUNIL ARCHAK and HIMANSHU PATHAK	...	18
The Role of CGIAR in the Global System of PGRFA Conservation and Use	KULDEEP SINGH, MICHAEL HALEWOOD, CHARLOTTE LUSTY and HD UPADHYAYA	...	22
Genomics-Driven Application of Plant Genetic Resources for Sustainable Agriculture	RAJEEV K VARSHNEY, ABHISHEK BOHRA and VANIKA GARG	...	28
Access to Plant Genetic Resources: Policy Vista	PL GAUTAM and SUNIL ARCHAK	...	34
A Novel <i>Circa situm</i> Approach to Conserve Forest Genetic Resources of the Western Ghats	R VASUDEVA	...	37
Empowering Farmers with Blockchain-based Transparency Throughout the Value Chain	GAURAV SOMWANSHI, PRATAP DESHMUKH, GANESH ANANTWAR, MANISH VERMA, AREEB MOHAMMAD KHAN, AARTI TIWARI, DANISH SIRAJ, GUNVANT SARPATE, MANDAR DARADE, VIKAS TAKLIKAR, NETAN MANGAL, SYED BURAIR, RENUKA PATURKAR, GAJENDRA SAHU and ROHIT DHIVARE	...	41
Agroecology-based Biodiversity Management	KS VARAPRASAD and V SANDHYA KUMARI	...	46
New Paradigms in Agricultural Education in India	RC AGRAWAL	...	50
Networks and Partnerships are Tools for Efficient PGR Management	RK TYAGI	...	53
Contributions of Dr RS Paroda to the Field of Genetic Resource Management	ANURADHA AGRAWAL	...	57
PLANT GENETIC RESOURCES			
Use of Plant Genetic Resources for Food and Nutritional Security	VIRINDER SINGH SOHU and BALDEV SINGH DHILLON	...	65
Mini Core Germplasm Collections for Developing Climate Resilient Crop Cultivars and Achieving Enhanced Genetic Gains in Plant Breeding Programs	HARI D UPADHYAYA and ANDREW H PATERSON	...	69
Biofortified Crops – Boon for Nutritional Security	SHUVOBRATA MAJUMDER, KARABI DATTA, DIPAK GAYEN, SOUMITRA PAUL, NUSRAT ALI, SUBHRAJYOTI GHOSH, ARITRA KARMAKAR, SANANDA BHATTACHARYA, SHINJINI SENGUPTA and SWAPAN KUMAR DATTA	...	74
Status of Genetic Resources in Oilseed Crops and Their Potential Use for Making India Atmanirbhar in Edible Oils	RASHMI YADAV, PARTHA RAY CHOUDHURY, ANJALI KAK KOUL, VEENA GUPTA, ASHOK KUMAR and DEVENDRA K YADAVA	...	85
International Year of Millets 2023: Opportunity for Enhancing the Use of Indian Millets Germplasm	M ELANGOVAN, KARNAM VENKATESH, SUSHIL PANDEY and CHITRA DEVI PANDEY	...	90
Genome Editing Technologies for Efficient use of Plant Genetic Resources	KC BANSAL, SOMNATH ROY and BASUDEV GHOSHAL	...	95
Building Value Chains for Enhanced PGR Utilization and Sustainable Food Systems	JC RANA, SK MALIK and ELEONORA DE FALCIS	...	100

Contribution of Technology in Enhancing and Preserving PGR RAM KAUNDINYA, SHIVENDRA BAJAJ and RATNA KUMRIA	...	107
Vegetable Genetic Resources to Mitigate Nutritional Insecurity in India TUSAR KANTI BEHERA, JAGDISH SINGH, RAKESH KUMAR DUBEY and SHAILESH KUMAR TIWARI	...	110
Exploration and Collection of Plant Genetic Resources in India: Status and Priorities SP AHLAWAT, KC BHATT, DP SEMWAL, K PRADHEEP and OP DHARIWAL	...	117
Crop Wild Relatives in India: Inventorization, Prioritization and Conservation K PRADHEEP, SP AHLAWAT, S NIVEDHITHA and VEENA GUPTA	...	124
Plant Taxonomy and Biosystematics in PGR Collecting and Conservation K JOSEPH JOHN, K PRADHEEP and E ROSHINI NAYAR	...	131
Application of Geographical Information System for PGR Management N SIVARAJ, SR PANDRAVADA, ANURADHA AGRAWAL, V KAMALA, V CELIA CHALAM and K ANITHA	...	136
Role of Plant Quarantine in Preventing Entry of Exotic Pests V CELIA CHALAM, KAVITA GUPTA, MC SINGH, Z KHAN, J AKHTAR, BH GAWADE, POOJA KUMARI, PARDEEP KUMAR, BR MEENA, AK MAURYA and DS MEENA	...	141
Large-scale Characterization for Management and Utilization of Indian National Genebank Collections ASHOK KUMAR, KULDEEP TRIPATHI and JYOTI KUMARI	...	147
Plant Genetic Resources for Crop Improvement: The North-Western Himalayan Perspective SK SHARMA, NIKHIL MALHOTRA and MOHAR SINGH	...	151
Management of Horticultural Genetic Resources in India: Recent Advances UMESH SRIVASTAVA	...	154
Conservation and Use of Underutilized Crops: Challenges and Way Forward BHAG MAL	...	162
Use of Crop Wild Relatives (CWRs) of Wheat in Disease Resistance Breeding NIRANJANA M, SAHARAN MS, JHA SK, NIHARIKA MALLICK, RAGHUNANDAN K and VINOD	...	169
Plant Germplasm Registration System in ICAR – Achievements ANJALI KAK and VEENA GUPTA	...	172
Role of Genebanking in the Era of Genomics VEENA GUPTA and SHERRY RACHEL JACOB	...	176
Sustainability of <i>In Vitro</i> Genebanks and Cryogenebanks ANURADHA AGRAWAL, R GOWTHAMI, SUBHASH CHANDER and VARTIKA SRIVASTAVA	...	180
Repository of <i>Allium</i> Genetic Resources at ICAR-NBPGR: Prospects and Challenges for Collection and Conservation ANJULA PANDEY, PAVAN KUMAR MALAV, DP SEMWAL, SUBHASH CHANDER, R GOWTHAMI and KM RAI	...	185
Grain Legumes Diversity of Indian National Genebank: A Potential Resource for Food and Nutritional Security PADMAVATI G GORE, NEETA SINGH and VEENA GUPTA	...	191
Genomic Tools in Plant Genetic Resource Management DHAMMAPRAKASH P WANKHEDE, RAKESH SINGH and S RAJKUMAR	...	194
Role of Bioinformatics in the Development of Plant Genetic Resources TANWY DASMANDAL, DWIJESH CHANDRA MISHRA and ANIL RAI	...	200
Genomic Resource Generation in Medicinal and Aromatic Plants RAKESH SINGH and RAMESH KUMAR	...	204
Policies Impacting Access to Plant Genetic Resources in Last Four Decades PRATIBHA BRAHMI, VANDANA TYAGI, PRAGYA and ANURADHA AGRAWAL	...	213
PGR Informatics: Sustaining PGR Management and Supporting PGR Policy SUNIL ARCHAK, KULDEEP SINGH and RC AGRAWAL	...	218
ANIMAL GENETIC RESOURCES		
Animal Genetic Resources (AnGR) Diversity in India BP MISHRA and SK NIRANJAN	...	221
Cattle and Buffalo Genetic Resources of India RK PUNDIR	...	229
Goat and Sheep Genetic Resources of India RAJEEV AK AGGARWAL, HK NARULA, SP DIXIT, REENA ARORA, DINESH KUMAR YADAV and INDRAJIT GANGULY	...	237

Poultry Genetic Resources of India and its Role in Rural Poultry Production	AK MISHRA	...	242
Conservation for Effective Management of AnGR in India	MS TANTIA, RAK AGGARWAL and REKHA SHARMA	...	247
Valorization of Indigenous Livestock and Poultry – An Approach towards their Conservation	REKHA SHARMA, REENA ARORA and SONIKA AHLAWAT	...	251
Uniqueness of Animal Genetic Resources Adapted to High Altitude Environment of Leh-Ladakh	MANISHI MUKESH, SAKET NIRANJAN, MONIKA SODHI, RS KATARIA, RAHUL BEHL, MOHAMMAD IQBAL, VIJAY K BHARTI, TSEWANG DORJAY and BP MISHRA	...	258
Genetic Uniqueness for Immune Response Traits in Native Animal Genetic Resources	RS KATARIA, ANKITA GURAO, RAVINDER SINGH, SK NIRANJAN and BP MISHRA	...	264
A1/A2 Milk Research in Indian Cattle	MONIKA SODHI, M MUKESH, RS KATARIA, SK NIRANJAN and BP MISHRA	...	269
Breed Registration and the Gazette Notification: A Unique National Framework for Protecting Native Animal Germplasm	SK NIRANJAN, RK PUNDIR and BP MISHRA	...	279

FISH GENETIC RESOURCES

FAO Resources for Strategic Planning	GRAHAM MAIR and DANIELA LUCENTE	...	283
Informatics as Future for Fish Genetic Resources Management	SIMON WILKINSON	...	289
The New Finfish Genetic Resource Discoveries from Indian Waters – Need for Enhancing Taxonomy Capacity	TK TEENA JAYAKUMAR, KK BINEESH, TT AJITH KUMAR and KULDEEP K LAL	...	292
A Review on Ichthyofaunal Diversity and Distribution of North-East India	KANGKAN SARMA, RAJDEEP DAS, RATUL CHANDRA BHARALI and DANDADHAR SARMA	...	298
Diversity of Fish Genetic Resources below the Species Level, Characterization and Applications in Resource Management	RAJEEV K SINGH, PR DIVYA, VINDHYA MOHINDRA and KULDEEP K LAL	...	301
Genomic Research in Fishery Sector: Current Status and Future Prospects	MURALI S KUMAR, RAVINDRA KUMAR, BASDEO KUSHWAHA, RAJEEV K SINGH, VINDHYA MOHINDRA	...	305
Reservoirs Fisheries Development and its Trade-offs with Conservation of Natural Fish Genetic Resources	VASU SUGUNAN and VVR SURESH	...	308
Geo-Spatial Tools for Science-Based Management of Inland Aquatic Habitats and Conservation of Fish Genetic Resources	P KRISHNAN, G KANTHARAJAN, REJANI CHANDRAN, A ANAND and VINDHYA MOHINDRA	...	312
Mariculture Development in India: Status and Way Forward	A GOPALAKRISHNAN, BOBY IGNATIUS and VVR SURESH	...	317
Fishing Technologies for Conservation of Marine Fish Genetic Resources	CN RAVISHANKAR and VR MADHU	...	322
Boosting Widespread Adoption of Sustainable Agriculture – New Metrics and the Role of Science	NATALIA ESTRADA CARMONA, SUNAYANA SHARMA, JAI RANA and KULDEEP K LAL	...	325

MICROBIAL GENETIC RESOURCES

Microbial Genetic Resources: Some Aspects and Prospects	MANOHARACHARY CHAKRAVARTHULA	...	333
Conserving Microbial Diversity: Practices, Trends and Beyond	ALOK KUMAR SRIVASTAVA, HILLOL CHAKDAR, PRAMOD KUMAR SAHU and MURUGAN KUMAR	...	338
Diversity of Root Nodule Bacteria: Strengthening R&D and Inoculant Supply	DLN RAO	...	343
Cyanobacterial Diversity Assessment Under Diverse Environments: A Molecular Approach	KRISHNA KUMAR RAI, RUCHI RAI, SHILPI SINGH and LC RAI	...	346
Synthetic Microbial Community (SynCom) for Sustainable Agriculture	NUNNA SAI APARNA DEVI and DANANJEYAN BALACHANDAR	...	351

Microbiome and Plant Health PUSHP LATA, KIRAN BALA, PIYUSH GOEL, UTKARSH SOOD, PRINCY HIRA, VATSALA DWIVEDI, ABHILASH KUMAR, CHARU DOGRA RAWAT and RUP LAL	...	355
Interlinking Soil Microbial Diversity and Rhizodeposition for Enhancing Nutrient Uptake and Productivity KK PAL and RINKU DEY	...	360
Microbial Nitrification Paradox: A Paradigm Shift on Nitrogen Uptake by Rice BABANPREET KOUR and BALASUBRAMANIAN RAMAKRISHNAN	...	365
Current Status and Recent Developments in Microbial Pesticide Use in India HB SINGH	...	369
Blue Green Algae for Secondary Agriculture HILLOL CHAKDAR, SHALOO VERMA and SUNIL PABBI	...	375

INSECT GENETIC RESOURCES

Role of Invertebrate Diversity with Special Reference to Insects in Agro-biodiversity and Ecological Services – Indian Efforts M NAGESH and K SREEDEVI	...	383
Insect Diversity and International Cooperation in Invasive Species Research RICH A VARSHNEY, RS RAMYA and OMPRAKASH NAVIK	...	386
Diversity of Scarab Fauna (Coleoptera: Scarabaeidae) – Importance and Implications KOLLA SREEDEVI, JUDITH COROLYN and M NAGESH	...	389
Conserving Floral and Faunal Diversity of Rice Paddies CHITRA SHANKER and JHANSI RANI BILLA	...	393
Diversity of Bee Pollinators – Global and Indian Perspective AMALA UDAYAKUMAR, KESAVAN SUBAHARAN and TM SHIVALINGASWAMY	...	397
Ecological Implications on Insect Biodiversity AMALA UDAYAKUMAR and KESAVAN SUBAHARAN	...	401
Insect Collections as a Reservoir of Insect Genetic Resources ANKITA GUPTA	...	406
Arthropod Germplasm Information System (AGIS) in India M PRATHEEPA, RICH A VARSHNEY and GANDHI GRACY	...	408
Guidelines to Authors	...	410

INTERVIEW

Genetic Resources Must be Accessible and Used: Dr RS Paroda

By **Sunil Archak**

Q1. Everyone knows about your immense contributions to Indian agriculture in general and agricultural research, education and extension in particular. However, your passion and unending quest for genetic resources has altogether a different dimension. When and how did your interest in genetic resources begin?

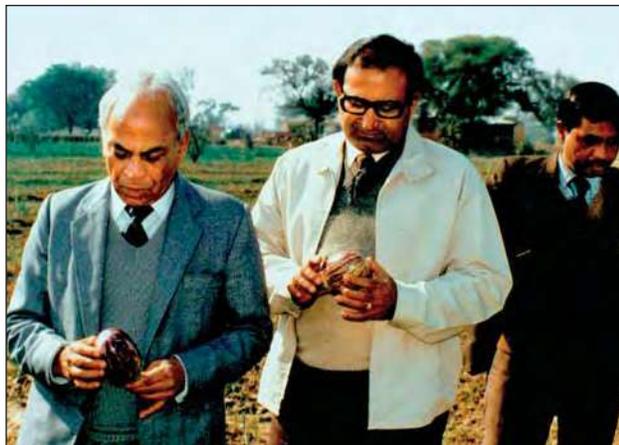
Hailing from the Thar desert of Rajasthan, while helping often my mother in farm operations, I understood the importance of plants, crops, animals, especially for the livelihood and survival of the people right from my childhood. Later, as student of Genetics and Plant Breeding, I got fascinated by Darwin's theory of evolution and Mendel's laws of inheritance. While being a Ph.D. student at IARI, under the guidance of Dr AB Joshi, I could appreciate the importance of genetic resources for crop improvement. At that time, I was impressed by Dr Harbhajan Singh's valuable work while heading the Division of Plant Introduction. Later, while working for my post-doctorate with Prof. Hugh Rees, FRS at University of Wales, Aberystwyth on cytogenetics of Eu-sorghums, I got more convinced of the importance of related wild species as useful source of resistance to biotic and abiotic stresses. The success of nobilization of sugarcane by TS Venkataraman at SBI, Coimbatore had fascinated me more towards the importance and management of genetic resources. The first multi-cut

sorghum variety SSG 59-3, released at the national level, was an outcome of sorghum × sweet Sudan grass hybridization work carried out at HAU, Hisar when I was working as Forage Breeder. The successful release of more than ten varieties at the national level of sorghum, oats, cowpea, guar and berseem by our team was mainly due to the use of rich germplasm that we had, many of those accessions were obtained from NBPGR.

My interest in genetic resources further got intensified after I visited the deserts of Australia, America and Central Asia. I realized that Thar desert is fortunate to have rich genetic resources of plants, trees and animals. However, there is an urgent need to collect, evaluate and conserve them before they become extinct in view of harsh climate and vagaries of weather.

As destiny would have it, I got an opportunity in 1985 to lead NBPGR – an institute on plant genetic resources. However, I understood immediately that for improved efficiency the system needed good infrastructure facilities, including the construction of a modern Genebank, which was indeed a great challenge since neither land nor funds were available then.

Q2. World has witnessed a paradigm shift in the way we access and use plant genetic resources. You have not only observed these changes but you also provided



With Dr AB Joshi: Teacher, guide and mentor



As forage breeder with Dr MS Swaminathan

policy guidance at national and global levels. Could you let the readers know about your first-hand experience? Have we achieved what we set-out for?

As a student of genetics and plant breeding, two cardinal principles were taught to us. These are: i) Genetic resources are common heritage of mankind (humankind) and ii) Genetic resources are exchanged freely.

Somehow, after the Convention on Biological Diversity (CBD) came into force in 1993, with a call to nation members to have their *sui generis* system to protect biodiversity, both these principles hold no good. There is though better awareness of rights now but in general lack of appreciation for mutual exchange of genetic resources. The current protectionist approach will be counter-productive. No doubt, we shall have to adapt to paradigm shift from free to better understood and agreed access and benefit sharing (ABS) system for bilateral exchanges as per Nagoya Protocol. On the contrary, in retrospect, had such restrictions been there earlier, our food basket would have not been so diversified as today. The current challenge is to protect what we possess but then we need to be wise to have in place ABS mechanisms by which needed exchange continues uninterrupted for future food, nutrition and environmental sustainability.

Fortunately, for multilateral exchanges, a well understood inter-governmental mechanism exists under FAO in the form of International Treaty on Genetic Resources for Food and Agriculture - ITPGRFA (signed 2001), which is in harmony with CBD and guaranteeing food security through the conservation, exchange (multilateral) and sustainable use of world's plant genetic resources. I was personally involved as head of Indian delegation in very long debates and meetings to put its framework in place, especially to have an agreement on definition of farmer's rights. During the Third Extraordinary Session of the CGRFA (mid-December 1996), I was made the chairman of Working Group on Farmers' Rights and had experienced fierce long debates, often inconclusive, for almost two years to finally have an agreement on farmer's rights on genetic resources, considered on par with those of breeder's rights. Similarly, it was not easy to agree on the list of 64 crops under Annex I of the Treaty that was approved in 1995. I was personally disappointed that despite strong justifications, crops like Soybean and Cotton were not included. Since then, almost 25 years have passed but



Representing India at an ITPGRFA session

the list of crops under Annex I has not been extended despite the presumption that Treaty would ensure this to happen. Hope the 9th Governing Body meeting being held in New Delhi next month would succeed in expanding it. Another concern that I have is about contributions towards Gene Fund, for which provision was made under the Treaty. Unfortunately, not much has happened since then. A significant development is, however, seen in the form of standard Material transfer agreement (SMTA) for the exchanges of genetic resources held in the Genebanks of International Centers under the umbrella of this treaty. Many developing countries, including India, have adopted this STA for multilateral exchanges of 64 crops listed under Annex I. This is likely to help considerably in opening the doors for sharing of genetic resources.

Q3. You are credited with building modern infrastructure for genetic resources conservation and management. Was it not thinking ahead of time and how difficult it was to



Project negotiation with USAID team

get government support to accomplish your dreams? When you look at these Bureaus, how you would consider their achievements?

When I joined as Director, NBPGR in 1985, the institute was located in an old building. It needed good research laboratories, equipment and the Genebank to conserve existing valuable genetic resources. With the help of colleagues and within available meagre budget, we tried to renovate the laboratory facilities. For building the Genebank, neither there were funds nor the land on Pusa campus. When I approached Dr. N.S. Randhawa, the then Director General and Dr. M.V. Rao, DDG (Crop Science), ICAR for additional funds, both expressed helplessness and advised to explore the options of external grant. Accordingly, considering it my first priority, I started negotiating with organizations like JICA and USAID for needed support.

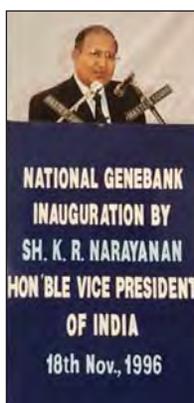
Fortunately, I could approach Dr. Nyle C. Brady, former Director General, IRRI, who then was Assistant Administrator for Science and Technology in USAID, Washington. Besides him, I received good support of Dr. D.S. Athwal, then working with Winrock International as Vice-President. In fact, both of them had worked closely at IRRI. After intense negotiation, and keeping in mind the facilities and infrastructure at the Genebanks at Fort Collins, USA and at Tsukuba, Japan, we submitted a project for the construction of institute building, scientific equipment and also projected the requirement of training in the US labs of most NBPGR scientists. As luck would have it, I could get the project finally sanctioned in 1987 with an outlay of US \$25 million. This led to the allocation of land and start of construction of a new campus of NBPGR housing the most modern Genebank with capacity to store one million accessions, which then was completed and inaugurated during the 2nd

International Crop Science Congress in November, 1996 by the then Vice President KR Narayanan. Today, this Genebank is the second largest with over 0.45 million seed samples stored safely, which has a capacity of 1 million accessions. In addition, facilities for Tissue Culture and Cryo-preservation Bank for vegetatively propagated plants were also created. Besides, a centre of excellence on DNA Fingerprinting was established at NBPGR.

Soon after joining as Director General, ICAR and being fully convinced of the importance and need of institutional support for genetic resources other than plants, I could convince policy makers to get sufficient funds and create the Bureaus of Animals, Fish, Microorganisms and Useful Insects. The National Bureau of Fish Genetic Resources at Lucknow was built in a record time of five years and it was inaugurated by the then Hon'ble Prime Minister Atal Bihari Vajpayee. Similarly, The Bureau of Animal Genetic Resources at Karnal was built in five years. As a result, all these four institutions, besides NBPGR, are serving a great cause and jointly they form a unique national network on management of genetic resources for posterity. Today, we do not find such a strong national program elsewhere in the world. Hence, we can legitimately feel proud of such a strong network of genetic resources.

It must also be mentioned that considering the need for trained Human Resource, a post-graduate program in the discipline of PGR was initiated for the first time in 1997, under which 73 M.Sc. and 31 Ph.D. degrees have been conferred.

With regard to my impression about the functioning of these bureaus, over the last two decades, I think overall they have done a good job. This was evident from an excellent display of their activities in the



Inauguration of Indian National Genebank, a dream project of Dr Paroda



At NBPGR genebank module

exhibition that was organized during the 1st International Agrobiodiversity Congress in November, 2016. The foreign participants from several countries were highly impressed by the quality of papers presented by our scientists.

However, an aggressive well monitored network effort in a Mission mode is warranted, as was done when a Mission on exploration and conservation was undertaken under NATP when we almost doubled (from 2 lac to 4 lac) the accessions in just five years. In retrospect I must say that if these institutions were not created then, we would have needed them now. We are better placed as they already exist and it is our responsibility to nurture them well.

As the Chairman of the Working Group on Farmers' Rights of FAO Commission on Plant Genetic Resources (1995-97), I could get the Farmers' Right defined and agreed by the Commission. While serving as the member of the International Committee constituted for Plant Genetic Resources by the Board on Agriculture, National Academy of Sciences, Washington for the period 1990-



Naming of ICRISAT genebank after RS Paroda



Felicitating Dr APJ Abdul Kalam during Indian Science Congress

94, I got recommended the much-needed renovation and strengthening of the Genebanks. I also worked passionately and closely with Dr Ismail Serageldin, the then Chairman, CGIAR and Dr Geoff Hawtin, the then Director General, IBPGR and succeeded in convincing the Funders, through constant dialogue and events at the meetings that I chaired, to support the cause of conserving global genetic resources. These efforts eventually led to the establishment of Global Crop Diversity Trust (GCDT) in 2004. In fact, it was Dr Jacques Diouf, the then Director General, who kindly agreed to our request to host GCDT at FAO, Rome. It is sad that the primary objective of extending support to the national Genebanks for rejuvenation and conservation of genetic resources in the developing countries has not yet been realized. Instead, focus got shifted to fund the Genebanks of CGIAR Centers. Another disappointment was that its headquarters got shifted to Germany.

Q4. You have organized important national and international conferences, symposia, brainstorming on PGR and have brought out world-class policy



Establishment of Kazak Genebank: Named as Raj Paroda Genebank

documents. Also, you are credited to have established some PGR networks. Do you see them as opportunities for strengthening R&D on PGR? Is there awareness among researchers about the significance of enabling policy environment for the management of genetic resources?

I have always felt strongly about the importance of establishing links and partnerships among agricultural institutions at state, national, regional and international levels. Hence, I had organised a number of networks on PGR and also the conferences, symposia, expert consultations, workshops and brainstorming sessions considering them important for strengthening research and development.

To ensure a platform for publishing scientific work of researchers, it was considered necessary to establish Indian Society of Plant Genetic Resources (ISPGR) at NBPGR in 1986. This Society has served a great cause of publishing scientific papers through its Journal IJPGR and by organising a number of national and international conferences. It also published a number of proceedings and promoted scientific collaboration among its more than 850 members.

It also organised a number of national symposia and conferences and brought out useful publications and proceedings. The First National symposium on PGR was held in March 1987 at IARI, New Delhi; which was inaugurated by Dr BP Pal, former Director General, ICAR and attended by Dr AB Joshi and Dr MS Swaminathan. Later, in 2013, in collaboration with BI, it had organised a Global Consultation on Use and Management of Agrobiodiversity for Sustainable Food Security at New Delhi. In November, 2016, it also organised successfully the 1st International Agrobiodiversity Congress (IAC)

with 1,000 participants from 60 countries at New Delhi, which was inaugurated by the Hon'ble Prime Minister Narendra Modi. Besides its proceedings, the conference also adopted "Delhi Declaration on Agrobiodiversity Management" concerning various aspects of conservation, management, access and use of agrobiodiversity and the Declaration is being used now for defining a 'Road Map' for future management of Agrobiodiversity.

It is also a great satisfaction that IAC has now become an important rolling event. The 2nd IAC was hosted in Rome in November, 2021 and was funded by the Government of Italy. It is expected that 3rd IAC will be hosted by CAAS in China. In my view, such scientific conferences are the best ways to expose young scientists to recent advances and catalyse them to pursue quality research in their career.

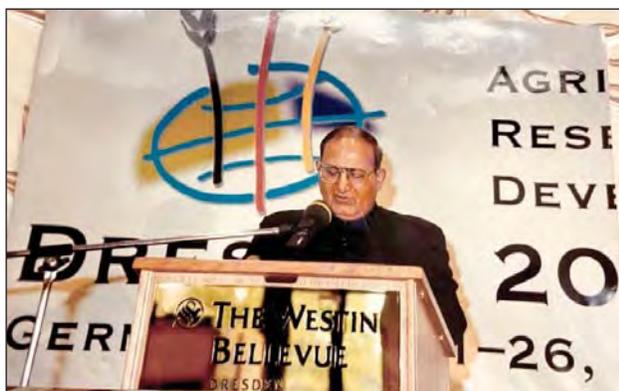
While working as Regional Plant Production and Protection Officer, FAO, Bangkok (1992-1994), and later as Executive Secretary, APAARI, I could initiate a number of regional PGR networks for evaluation and use of genetic resources in different countries. These were: on maize -TAMNET involving FAO and CIMMYT; on cereals and legumes – CLAN involving ICRISAT, ICARDA and AVRDC; on rice – CORRA involving IRRI; on cotton – INCANA involving AARINENA, APAARI and CACAARI; and on fish - GoFAR involving WorldFish Center. For these networks, an effort was made to involve more NARS and strengthen three regional PGR networks of IBPGR for i) South Asia, ii) Southeast Asia and iii) the Pacific region. These networks helped in joint evaluation and exchange of useful genetic resources by concerned NARS and derive mutual benefits. It was



Dr BP Pal at the inauguration of First National Symposium on PGR at IARI in 1987



Meeting on Strengthening PGR Networks at FAO, Bangkok



First Global Conference by GFAR in Dresden

great satisfaction to see the willingness of NARS partners to collaborate and identify useful varieties and hybrids that can enhance productivity of these crops.

As founder Chairman of the Global Forum on Agricultural Research (GFAR) from 1998-2002, I encouraged partnership building among NARS and the Agricultural Research for Development (AR4D) stakeholders. During the first Global Conference on AR4D organized by GFAR in 2000, a special session was held in collaboration with Bioversity International where the “Dresden Declaration on Plant Genetic Resources for Food and Agriculture” was adopted, defining clearly a Road Map for strengthening PGR activities at the global level. Later, as Executive Secretary, Asia Pacific Association of Agricultural Research Institutions, I could hold in collaboration with BI and other CG Centers, an International Symposium on Agrobiodiversity in Suwon in 2010 which led to the adoption of ‘The Suwon Agrobiodiversity Framework’, catalysing all stakeholders

in the Region to accelerate activities to conserve their valuable genetic resources through use. Subsequently, both GFAR and CGIAR decided to organize jointly every two years the Global Conference on Agricultural Research for Development (GCARD). As chairman of the Program Committee of GFAR, I had the privilege to organize GCARD 1 at Montpellier, France in 2010 and GCARD 2 at Punta del Este in Uruguay in 2012. As an outcome, we could get overwhelming endorsement of ‘GCARD Road Map’.

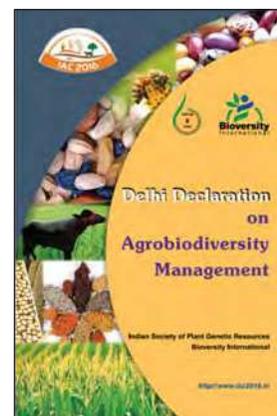
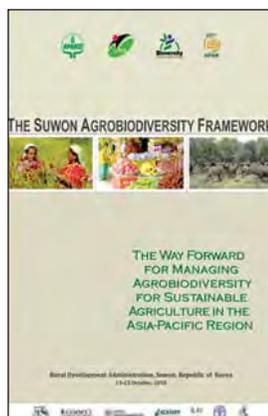
To promote regional cooperation and scientific networks, I took the major initiative to organize a number of Regional Expert Consultations. These were: Regional Roundtable Meeting on Implementation of the International Treaty for Plant Genetic Resources for Food and Agriculture (ITPGRFA) in 2005 in Bangkok; Expert Consultation on Progress of Research Networks at ICRISAT in 2007; Regional Workshop for Implementation of Suwon Agrobiodiversity Framework in 2011 in Kuala Lumpur; Regional Consultation on Genetic Resources in the Pacific held at Fiji (2012); and the Regional Consultation on Medicinal Plants in 2013 in Bangkok.

Q5. It is rare to see five dedicated Bureaus working on different genetic resources in any other country. Do you believe these Bureaus have done justice to their existence? Can these be the global research Centers of genetic resources?

As stated earlier, a well monitored mission mode approach is needed to make these bureaus more effective at global level. India is fortunate to have presently five Bureaus on Genetic Resource management of its rich agrobiodiversity, besides a unique *sui generis* system



1st International Agrobiodiversity Congress organized with 1,000 participants from 60 countries (New Delhi, 6-9 Nov 2016), which was inaugurated by the Prime Minister Narendra Modi.



Dresden Declaration (2000); Suwon Framework (2010); GCARD Roadmap (2012); Delhi Declaration (2016)

through two acts approved by the Parliament, namely, i) Protection of Plant Varieties and Farmers' Rights Act (2001), and ii) National Biodiversity Act (2002). As head of ICAR, and in view of my personal interest in the subject, I was actively involved in drafting and getting these bills approved. These Acts and Bureaux are today serving a great cause for the conservation and use of country's valuable agrobiodiversity. Surely, we can feel proud of having such a strong national system with more than 200 scientists actively engaged in various aspects of genetic resources. They need to have real passion for the genetic resource management activities. Moreover, in my view there is no room for complacency just because we have a strong system now in place.

Q6. Do you think modern technologies like gene editing will make genebanks redundant? What is the role of genebanks in combating climate change? In other words, how can the Indian genebank be a valuable resource to address climate change?

I do not think so. In fact, genome editing is just a technique to accelerate the breeding process –it is rather a disruptive innovation helpful in transferring desired

genes in good agronomic background. In any case, gene editing will need new genes for specific traits to be incorporated. Hence, success would obviously depend on whether we have the desired genes for editing. As such, the importance of Genebanks, especially the collections of wild and weedy species, shall remain. On the contrary, there will be an acceleration of conservation through use now than ever before. With regard to the role of Genebanks to combat climate change, India is again much better placed globally in view of existing rich germplasm that has the best-known tolerance to stresses – both abiotic (cold, heat, salinity, drought, flooding etc.) and biotic (diseases, pests, weeds etc.). We are known to have the best salt, heat and drought tolerant materials of wheat, rice sorghum, pearl millet and a number of fruits, vegetables, grasses and trees. These would be valuable sources for breeding crops and animals to adapt well to climate change.

To harness the benefits of genome editing, much would depend on enabling policies and institutional support, besides enhanced funding so critical for harnessing rich genetic resources.

Reorienting the Plant Genetic Resource Management for Enhanced Utilization

Trilochan Mohapatra^{1*} and Sunil Archak²

¹President, National Academy of Agricultural Science, Pusa Campus, New Delhi-110012

²ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012

Context

Since mid-20th century, India transformed itself as a country that depended on food aid to a net food exporter, with nearly six-fold increase in food grain production from 50 million tonnes in 1950-51 to 314.51 million tonnes in 2021-22, due to the adoption of technologies. By 2050, India's population will reach 1.7 billion people, creating the most populated country in the world. Food demand will increase by 70 per cent. Globally, the demand for food is expected to rise by 50 per cent and yields may decline by 2050 in the absence of adequate climate action. Thus, India faces the formidable challenge of ensuring a guaranteed and adequate supply of nutritious and healthy food produced in an economically, culturally, socially and environmentally sustainable manner.

The United Nations' 2030 Development Agenda includes 17 for Sustainable Development Goals (SDGs) and those related to agriculture include: 1) No poverty, 2) Zero hunger 3) Good health and well-being for people 12) Responsible consumption and production, 13) Climate action, 14) Life below water, and 15) Life on land. However, addressing several of SDGs in an increasingly challenging environment and burgeoning population would be a formidable challenge. It is here that genetic resources play a crucial role.

Role of Genetic Resources

Genetic resources are the biological cornerstone of global food security. The agricultural diversity and genetic resources that support agriculture need to be used efficiently both to maintain current levels of food production and to confront future challenges. Adapting crop varieties to local ecological conditions can reduce risks induced by climate change. Thus, identifying adapted germplasm requires reoriented emphasis on augmentation, characterization and evaluation of germplasm in genebanks. In fact, conservation is the

beginning of the solution in terms of trait-discovery (Fig. 1). Science-based interventions are required at various stages of genetic resources use.

Increasing yields of major food crops – or even maintaining them – in the face of climate change will depend on combining genetic traits found in materials of a wide range of origins, including wild species. Unfortunately, wild species are especially vulnerable to climate change because they do not receive management interventions that help them adapt to changing conditions. Narrowly adapted species and endemics are especially vulnerable to the direct effects of climate change. Some of the centres of landrace diversity exist in areas under

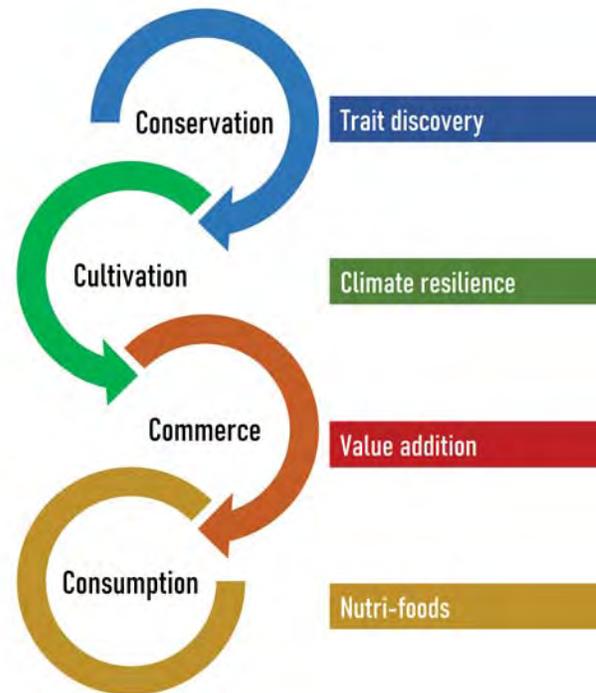


Fig. 1. Science-based interventions required at various stages of genetic resources use

*Author for Correspondence: Email-tmnrcpb@gmail.com

Prioritization of crop wild relatives

NBPGR shortlisted 861 Indian CWR taxa belonging to 769 species for 171 crops (of ICAR-mandate) falling under 14 crop groups (based on overall closeness with crop; potential use). Prioritisation resulted in 292 taxa (257 species) belonging to 85 crops (level of closeness to crop, economic traits or wide hybridisation work under progress and extent of distribution/ threat). Of 292 prioritised taxa, only 167 are conserved in the National Genebank. Conserved species lack representative samples from across a geographical and ecological range.

considerable climate risk, where diversity is valued for its resilience. It is, however, poorly understood how the increase of climate risk, and change in the climate baseline might impact the current diversity in landraces found in situ. Impacts are likely to be both positive and negative on landrace diversity depending on the region, but a priority for research and monitoring activities is to ensure that more diversity is not lost.

Status of PGR Management and Utilization

Germplasm augmentation: Climate change calls for change in priorities for action with respect to genetic resources. Today there is urgent need for consolidating Indian genebank collections of wild species, including crop wild relatives (CWR), due to increased likelihood of extinction for narrowly adapted and endemic species. Novel and increased demands on germplasm in genebanks for adapting

agriculture to climate change, including the need to screening for different characters. By carrying out focused collecting missions, a total of 132 exploration and collecting missions during 2016-2021 resulted in collection of more than 9700 (Fig. 2). About 30% of the accessions belonged to the focus group of CWR. In the NEH region alone, as many as 28 explorations were conducted and more than 2000 accessions. Use of all modern tools viz. GIS, digital surface and soils maps, satellite imagery, etc. must be routine practice of exploration missions. Collecting, compiling and documenting ITKs related to PGR is also significant.

Plant quarantine: A systematic step-wise strategy followed for testing of each sample imported for presence of any unwanted pests and all samples found infested/infected were salvaged and if they could not be salvaged, they were rejected and destroyed by suitable means.

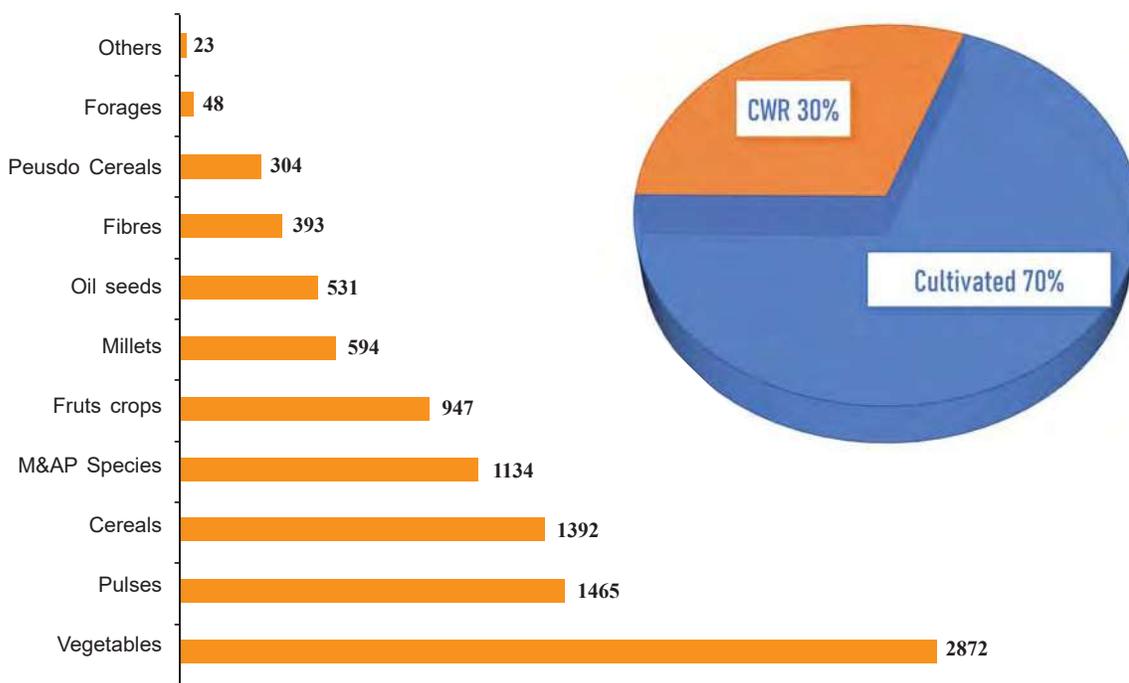


Fig. 2. Crop-group wise germplasm collected by NBPGR through targeted explorations during the period 2016-21

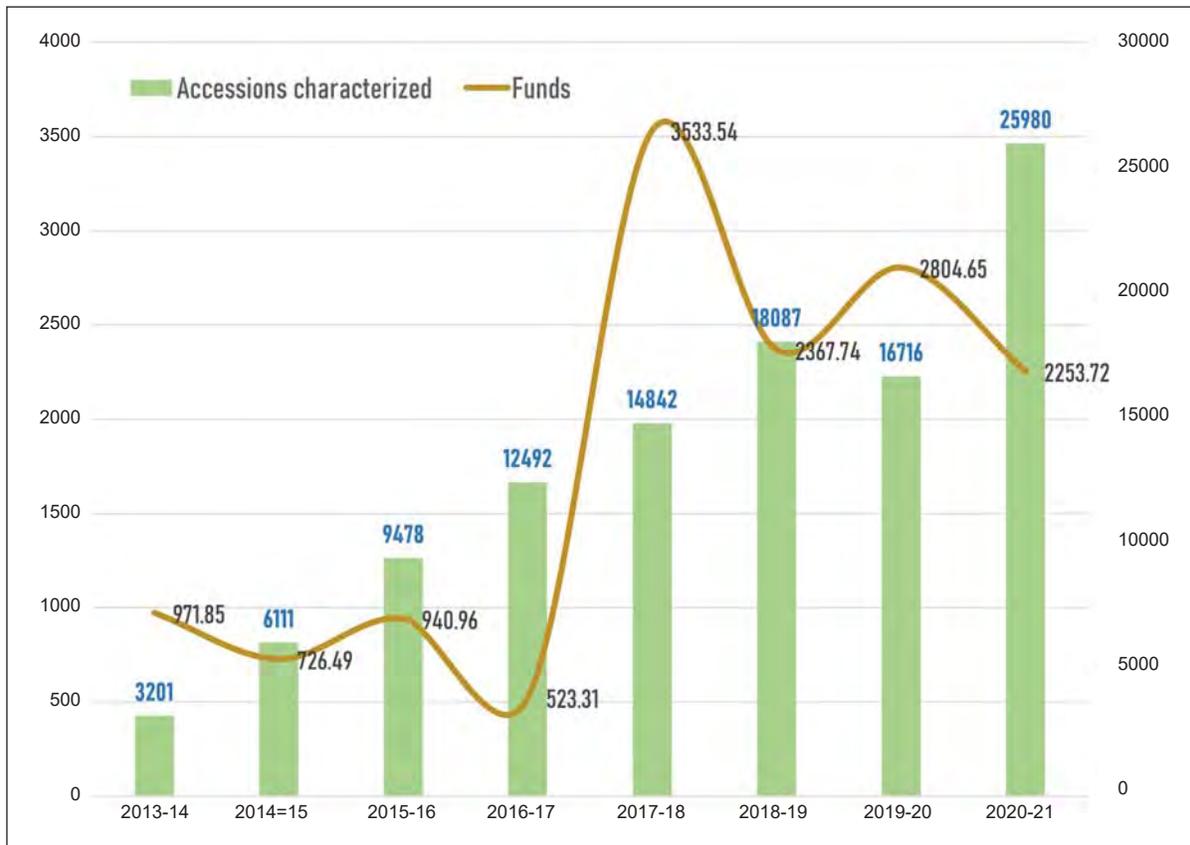


Fig. 3. Institutional funding (line, in lakh rupees) accelerating the characterization of germplasm (bar, number of accessions).

Estimated losses in India averted by interception of quarantine pests in introduced germplasm is estimated to be around Soybean (Rs.45 crore), Maize (Rs.47 crore), Rapeseed- mustard (Rs.39 crore) and Groundnut (Rs.33 crore). This calls for arming the quarantine set-up with modern tools and carrying out supportive research and documentation. Generation of awareness about importance of quarantine among public and policy makers is also essential.

Conservation: Globally more than 7 million accessions are conserved *ex situ* (WIEWS data) and about the same number are in the safety back-up at the Svalbard Global Seed Vault. Data of only 2.5 million are available on Genesys-PGR portal. Indian genebank conserves about 0.45 million accessions collected from habitats within India (Table 1) or imported from other countries. Despite global holdings being redundant, enough scope of augmentation and enough room in the seed genebank exist. Number of accessions of CWR added to seed genebank has jumped from 17 per year to about 120 per year since 2014. NBPGR has the best of cryo and in vitro genebanks. But field genebanking of horticultural

crops requires attention and coordination with institutes and universities. Working with custodian farmers has great advantage.

Germplasm characterization: Genebanks carry out characterization as a routine activity. However, large-scale phenotyping of the germplasm accessions in one-go can facilitate designation of core collections allowing increased use of germplasm. ICAR's initiatives in supporting the field characterization of several crops under an ambitious project "Consortia Research Platform on Agrobiodiversity" provided much needed funds that led to forming networks and carrying out multi-location experiments. The dedicated financial support gave an impetus to take out entire genebank collections of various crops for characterization (Fig. 3). A total of >280 crore rupees was invested by ICAR through mega projects like CRP-Agrobiodiversity and NICRA causing a dominion effect and attracting about 150 crore rupees as extra-mural grants. Advantages of such initiatives include: (i) involvement of plant breeders and specialists from the outset guaranteeing the use of identified trait specific resources and (ii) creation of an institutional as well

as organic partnerships for long term associations. For instance, varieties as many as 78 vegetable, 29 spices, 26 millets, 24 pulses, 21 cereals, 17 forage, 13 oilseeds among others have been developed using the indigenous germplasm in the last decade (2011-2020).

Germplasm sharing: India, though NBPGR and other partner institutions, has ensure that germplasm is available for research in the country. In fact, procedures are laid out for sharing germplasm with foreign researchers. It is important to ensure that germplasm is exchanged for research purpose without hurdles and also follow legal requirements nationally and compliance internationally. In collaboration with Kirkhouse Trust, India has shared >250 accessions of pulses with ten African countries.

Use of modern technologies: India has the advantage of being endowed with high degree of genetic variability accessible from ex situ genebank. There is now need to lay greater emphasis on employing novel technologies including genomics for large scale genotyping and associating with phenotypic variations. Trait discovery, identification of best germplasm accessions with multiple trait advantage will ultimately lead to their use in developing climate resilient and nutri-rich varieties.

Action Points

Keeping in mind the changing climatic conditions, changing needs and advancements in technologies, it is essential to calibrate our PGR management procedures and prioritize the resources for enhanced utilization of PGR (Fig. 4). Some suggestions given below:

Efficient use of gene pool: Augmentation of CWRs by collecting or by importing; developing descriptors where not available and characterization; trait-specific screening; pre-breeding; genotyping and identification of loci. Effective use genomic tools for genotyping and associating the data with phenotypic trait and contributing the material/information to varietal development programmes.

Map R&D and PGR services onto visions and programmes: A number of significant activities are being carried out by NBPGR and partners. They need to be mapped onto the relevant SDGs and national vision documents to make the PGR activities look relevant and contemporary. Excellent activities can be planned and implemented under the aegis of national programmes aiming at farmers welfare as well as NEH/TSP specific activities.

Table 1. Germplasm accessions of various crops collected from different states and conserved in Indian genebank

State	Count
Andaman and Nicobar Islands	622
Andhra Pradesh	13765
Arunachal Pradesh	3573
Assam	6136
Bihar	5711
Chhattisgarh	14881
Delhi	6912
Goa	352
Gujarat	8476
Haryana	6049
Himachal Pradesh	8950
Jammu and Kashmir	3732
Jharkhand	4125
Karnataka	8246
Kerala	4653
Madhya Pradesh	14069
Maharashtra	17545
Manipur	1825
Meghalaya	1975
Mizoram	1585
Nagaland	1978
Odisha	10159
Others	45580
Punjab	4621
Rajasthan	10889
Sikkim	1239
Tamil Nadu	7204
Telangana	8290
Tripura	603
Unknown	70444
Uttar Pradesh	14817
Uttarakhand	12378
West Bengal	2753
Others	114

Documentation and awareness: Without an efficient information system, data on useful germplasm and traits cannot be accessed by researchers curtailing the utilization potential. In addition to the impactful research papers aiming at scientists, semi-technical and general publications must be brought out periodically. They will provide necessary information to policy makers and funding agencies about the ongoing programmes and success stories. PGR is localized and therefore leaflets and brochures in vernacular languages must be made available.

Collaboration and networking: Every activity of PGR management is multi-disciplinary. Inter-institutional collaborations within India have shown good results in

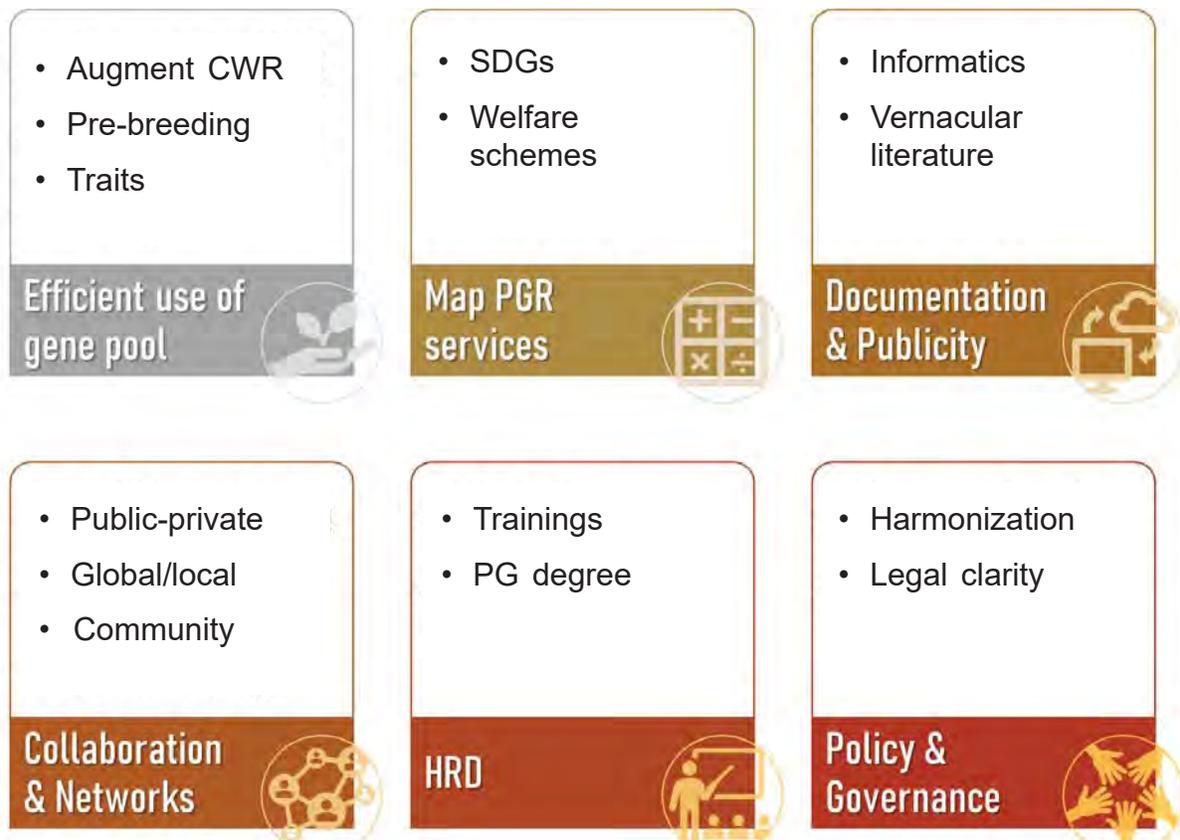


Fig. 4. Five action areas for the future of genetic resources utilization

phenotyping and genotyping. International collaborations, particularly in screening for biotic and abiotic stress factors will save valuable years in preparation. Public-Private partnerships are woefully less and needs strategic planning. Working with custodian farmers and conserving communities will provide not only practical lessons but also access to hitherto uncollected populations of lesser-known crops.

Capacity building and HRD: PGR management requires specialized skills and therefore training programmes to various stakeholders must be a regular activity. Topic-

specific trainings in taxonomy, genotyping, quarantine, informatics, etc. are required for upgradation of skills. It is important to generate human resource that will be well-prepared to continue PGR programmes in future through post-graduate programmes.

Acknowledgement

Information has been drawn from various reports of ICAR-NBPGR and thanks are due to all the researchers of the Bureau and network partners.

Farmers' Rights and Plant Breeders' Rights under Indian Regulations of Plant Variety Protection: A TRIPS Compliant Harmonised Coexistence

Raj Ganesh, Arun Kumar, DS Pilania, AK Singh, DR Choudhury, TK Nagarathna, DK Agarwal and KV Prabhu*

*Protection of Plant Varieties and Farmers' Rights Authority, (A Statutory Authority by an Act of the Parliament)
Ministry of Agriculture and Farmers' Welfare, Pusa Campus, New Delhi-110012, India*

India's 'Protection of Plant Varieties and Farmers' Rights (PPV&FR) Act, 2001' was enacted by its Parliament in accordance to the Article 27.3(b) of Trade Related Intellectual Property Rights (TRIPS) Agreement. In a significant deviation from the contemporary legislations around the world and also the International Union for the Protection of New Varieties of Plants (UPOV) Acts (1973 and 1991), the PPV&FR Act is a *sui generis* option, that provides an effective system for the protection of plant varieties, the rights of farmers and plant breeders that encouraged development of new varieties as well as those which were in common knowledge of the farmers or were in the possession of farmers traditionally. This article explains how the Indian legislation is the best fit to match the requirements farmers as well as plant breeders, keeping in view the local agricultural situations, societal constitution and livelihood mechanisms, especially for small-holder farmers.

Introduction

In compliance to the Trade Related Intellectual Property Rights (TRIPS) Agreement, the Government of India adopted the *sui generis* option to provide for the establishment of an effective system for the protection of plant varieties, the rights of farmers and plant breeders that encouraged development of new varieties as well as those which were in common knowledge of the farmers or were in the possession of farmers traditionally. 'The Protection of Plant Varieties and Farmers' Rights (PPV&FR) Act, 2001' was enacted by the Parliament of India in accordance to the Article 27.3(b) of TRIPS Agreement. In a significant deviation from the contemporary legislations around the world and also the International Union for the Protection of New Varieties of Plants (UPOV) Acts (1973 and 1991), the Parliament considered it necessary to recognize and to protect the rights of the farmers in respect to their contributions made at any time in conserving, improving and making available plant genetic resources (PGR) for the development of new plant varieties in a predominantly agriculture driven economy, industry and livelihood options in a country that had more than 70% of its population living on agriculture directly of whom, more than 85% were small holder farmers (Agriculture Census, 2015-16, <https://agcensus.nic.in>). After two decades of the enactment, with global

commerce & trade reforms under WTO and other such international negotiations in place, more than 50% of India's population still continues to be that of farmers (Annual Report 2020-21, Department of Agriculture, Cooperation & Farmers' Welfare, Ministry of Agriculture & Farmers' Welfare, Government of India).

The Indian legislation has made sufficient provisions to protect the interests of plant breeders from both private and public sectors with overarching recognition of the farmers as conservers of traditional plant varieties, landraces, wild relatives of crops for specific traditional practices of saving, producing, sharing or exchanging seed amongst farmers as their farm produce while fully recognizing the contributions of both commercial plant breeders and farmers in plant breeding activity in developing new varieties (PPVFRA, 2001).

It is this unique balance between plant breeders' rights and farmers' rights that is maintained by the Indian legislation that has been interpreted as contradictory or conflicting approach in protecting plant breeders' rights by several legal or regulatory consultants or practitioners. Without exception, such contradictions are based primarily on their conviction about UPOV Acts of 1973, more appropriately of 1991 as the premise, any deviation from which is held as a contradiction to international standards, despite conformity to TRIPS as well as accepted laws of the land. We explain here

*Author for Correspondence: Email-chairperson-ppvfra@nic.in; Website: <http://plantauthority.gov.in>

how the Indian legislation is the best fit to match the requirements of the beneficiaries of the products of the Act, majority of whom are farmers followed by the plant breeders. Globally, plant breeders' rights centric regulations are the obvious best fit only to the plant breeders; however such regulations disregard the local agricultural situations, societal constitution and livelihood mechanisms among small holder farmers dominated populations.

What is the Meaning of Protection of Plant Varieties in India?

The protection exercised through registration of a plant variety by its designation by the PPV&FR Authority, means no one can sell, export, import or produce the seed or propagating materials of such protected (registered) plant variety without the registered breeder's (owner's) permission.

Protection of a plant variety is an intellectual property right (IPR) that the breeder (who could be an individual, farmer, community of farmers, institution or a government) enjoys over the variety along with designation in the notified plant species as the said variety's owner, who is entitled to plant breeders' rights on the variety with the exception of farmers' rights on the variety as provided in the Act. This shall remain with the registered breeder till he assigns it to anyone else, for the entire period of protection for the purpose of preventing any other person from making economic benefit from it or from assuming any of the rights to which the breeder is entitled to, without permission of the registered breeder.

The protection of a plant variety with its denomination also means that no one including the registered breeder can sell any other variety with the same denomination or sell the protected variety with any other denomination, commitment of which is also a punishable act of infringement.

Farmers' Rights on Plant Varieties in India: Integral to National Agriculture and Food Security

India is the only country which has provided IPR to farmers or communities of farmers over the traditional varieties, wild relative species of cultivated traditional crops, landraces conserved by them. This is to protect their rights on these materials in order to prevent utilization of such genetic resource by plant breeders without sharing any benefits realised from such varieties with the conserver farmers or farming communities, in

Section 26 of PPVFR Act (2001). In doing so, India at once recognizes existing plant varieties also, which are not novel but are in trade and in the common knowledge of farmers, since a period not beyond the duration of the period of maximum protection, retrospectively from the date of registration (these are generally the varieties in possession of plant breeders from public or private sectors), or those regionally well-adapted traditional varieties with their own value in trade, cultivation and usage with the farming communities of the country. This category involves the traditional and landrace varieties. In contrast, other regulatory legislations globally (broadly following either UPOV 1973 or UPOV 1991 Acts) only recognize new plant varieties while all other beyond the novelty acceptable period are relegated to a status of being in public domain for anybody to access.

It is through this exception for acknowledging the existing non-novel varieties as well as traditional or landrace varieties, the Indian law facilitates formal entitlement of farmers as well as plant breeders to their rights on such already existing varieties within the protectable period of 15-18 years before the date of application by the plant breeders concerned; or exclusive farmers' rights on those varieties known to be existing traditionally as heritage or as landraces in the community for generations, to exercise their rights on the materials for a further period of 15 or 18 years for their rights on these varieties, from the date of registration. In all other countries where protection of plant varieties is in practice, they however, categorise all the latter groups of landraces as well as varieties in common knowledge as varieties in public domain for anyone to access and use freely.

Some Specific Features of Indian PVP and FR Exercised by Plant Breeders and Farmers of India unlike their Counterparts in Other Countries

- Voluntary option to register for Plant Breeders' Rights
- Voluntary option for breeder to market without either getting the seed certified or released under Seed Act procedures (Seed Act, 1966)
- Voluntary registration under Seed Act procedures for seed quality can be practiced equally independently without protection of the plant breeders rights
- Registered breeder can authorise others to exercise any of his rights or engage any agency or licensee to exercise any of the plant breeders' rights

- The rights on variety of the plant breeder limit only to its use as seed and not as a produce or harvested product from the seed/seeding materials
- Farmers have their own rights on the registered protected varieties during period of protection
- Farmers who infringe the plant breeders rights out of genuine ignorance are given benefit of not being guilty to infringement owing to their ignorance on the protected status of the particular variety
- Farmers can claim compensation on less than minimum performance expected from the variety upon following standard mentioned package of practices on a plant variety whose seed has been legitimately procured by him through authorised seller of the registered variety in the current season.
- Farmers have the right to get the seed of protected varieties on the reasonable price without being subjected to predatory market practices
- Farmers have right to get the assured supply of seed through compulsory licensing, in case the original licensees fail to do so
- Farmers have the provision of getting recognition and rewards for their contribution in conserving and adding value to traditional genetic diversity

Prevailing Situation of Seed Trade and Marketing Network in India

In India, the existing seed trade and marketing network does not facilitate a shift completely to formal seed system for distribution, sale and marketing of protected varieties. Presently, seed distribution, production and marketing through informal system is 50-55% with the average seed replacement rate hovering from 27% in groundnut to 61% in pigeon pea during 2019-20 (Fig. 1) among major crops (DAC&FW, MoA&FW). In traditional varieties and region-specific local crop species, there is hardly any such possibility due to lack of seed related activities among private or public sectors. Varietal Replacement Rate (VRR) is one of the important factors in realizing higher crop productivity. The pace of progress in food production is largely dependent upon the progress of seed programmes that enable supply of quality seed of high yielding varieties with superior genetics. In India, barring a few commodities such as wheat wherein the share of seed replaced varieties released in the last five years was 59.5% while the varieties released in the last ten years made up 92.3% of the replaced seed during 2022-23, the situation in other crops though gradually improving is as not as rosy for example rice wherein share of varieties, notified during last five years and ten years in total breeder seed indent were only 28.4% and 64.8 % during 2022-23 as compared to 12.8 % and

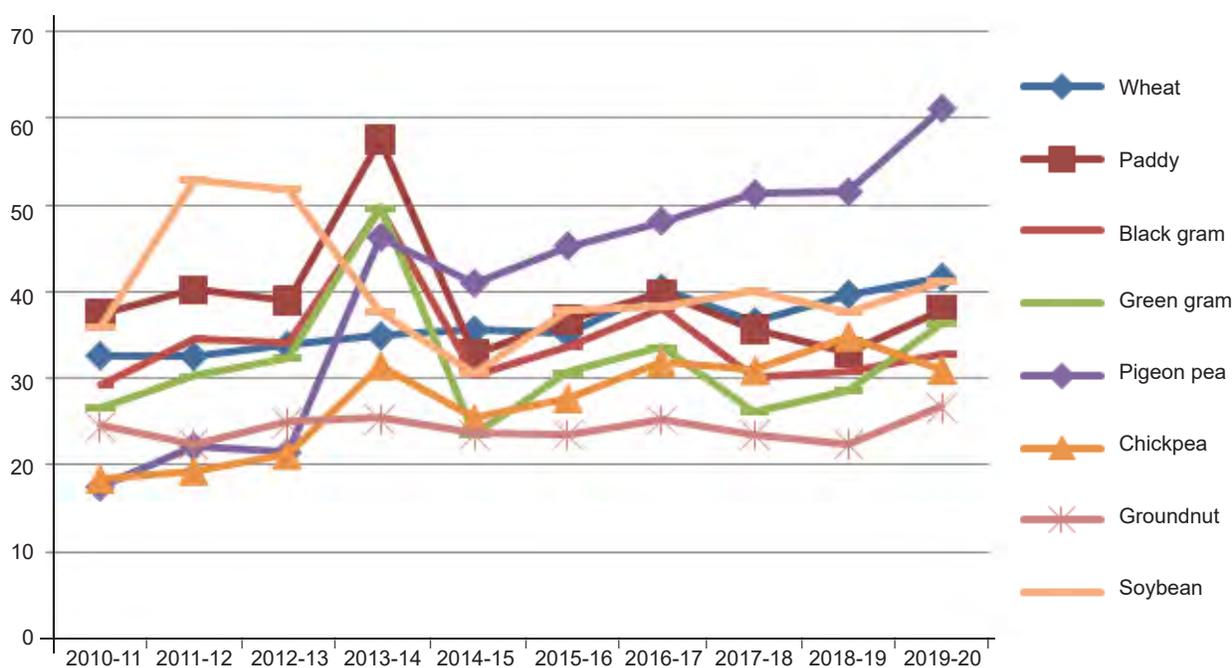


Fig. 1. National seed replacement rates among major crops (%)

43.2 % during 2018-19. If one accounted to average seed replacement over the entire country to 45%, then the magnitude of recent varieties in cultivation will amount to an estimated 26.8% and 41.4% of total area of wheat, of the varieties less than five and ten years old, respectively.

The total quality seed availability is 48.37 million tons (Agricultural Statistics at a Glance, 2021) in the country which as per varying demand and seed supply situation of various states would not at all suffice the seed requirements of the country. Registration for protection of new plant varieties is compulsory prior to marketing of a variety in all UPOV following regulation systems. If the same were to exclusively be practiced in India without the above described farmers' rights, consequences would be as follows:

- a. A huge vacuum in total food production compromising country's food and nutritional security would happen by taking out the informal seed system from seed trade which will mean non-availability of authorised seed to plant a major chunk of 200 million ha planted over multiple seasons with 140% cropping intensity.
- b. It would take more than 4 to 5 times the current investment for the entire area to be covered by formal seed system that would disbalance the entire economics of agricultural seed trade in India.
- c. The sketchy and unorganized seed distribution would hike the demand based increased seed price to an extent that may make the seed unaffordable or much more dearer than today to a farmer willing to invest in new seed.
- d. UPOV 1991 exemptions to allow self-saved seed would not have enabled more than 70% of seed-deprived farmers to plant seeds as less than 30% of farmers have any facility for saving their seed.
- e. Extant plant varieties in trade beyond 15 years of their release or notification or development make up for lion's share among seed trade in India. If these were not to be included, as legal seed as UPOV 1991 or 1973 apply only the new varieties, then the unavailability of quality seed of only new plant varieties would have further escalated the demand for new seed while further increasing the estimated number of farmers who would be deprived of the new variety's seed. The dimension is unimaginably large for most of the crops, whether sown in less

than one million ha or more than 10 million ha purely because of the small size individual planting spread over a large geographic area of each crop, making it difficult for any seed producer to reach the spots in time.

The above scenario that is highly probable would not only cause a large gap between the potential and actual production in different commodities including those which are necessary for food security ingredients as per the National Food Security Act (NFSA), 2013 and for nutrition management as committed to the Sustainable Development Goals (SDGs) due to gross reduction of production in general and litigation related non-marketability due to prosecution proceedings or legal proceedings likely to escalate from claims on violation to the plant breeders right.

Farmers Rights Under PPV&FR Act

Integral to prevailing Indian agriculture, are the farmers' rights to sow, resow, share, exchange and sell seed as farm produce of varieties including protected varieties, under Section 39(iv) of the PPV&FR Act. This provision only facilitates the survival with enrichment through better plant varieties of the informal seed sector. It also enhances the fast spread of the new varieties protected because of the sharing or selling of self-saved seed or farm produce of a new variety in a given region where the registered breeder may not have a reach or agency to cover seed selling

It also allows an inbuilt mechanism of evaluation of performance by the degree of economic satisfaction a farmer receives by growing of crop raised from quality seed obtained from the genuine authorised source that automatically becomes the base for comparison of the quantity or quality of the produce from self-saved or neighbour produced seed in the next season. The word of mouth spread of this information motivated investment for new seed after first experience of use self-saved seed facilitated by the right to resow or exchange or share or sell farm produce as seed.

It goes without saying that even the farmer who has saved seed of previous season crop has an access in time to plant new seed from the formal authorised agency; he would on first priority procure new quality seed before choosing his own seed. There is no exception to it. When the farmer knows that his right to claim for compensation is operational only on new seed procured legitimately from authorised seed seller of a plant variety,

even if available with himself or neighbourhood, he is bound to first procure the available new seed from the seller than use his or neighbour's seed. The farmer also is aware that the new quality seed has assured germination, purity and viability which he cannot take for granted on his own saved seed even in the case of self-pollinated crop species.

The above points are only a few which immediately ensure that the provision of free-access to protected unbranded seed known as "brown bag seed" in trade, is more of generation of knowledge and experience that is also shared along with shared seed on the variety's performance that comes as bonus to the registered breeder who does not have to invest in laying out a demonstration.

Conclusion

Thus, for a country with historical agriculture that is the backbone of its culture and economics, it is most essential to respect the rights traditionally enjoyed by the farmers who are the custodians by heritage of the plant and crop diversity as well as production through

generations, while encouraging them to adopt new and improved plant varieties in different crops with the aim to improve their livelihood support systems and income. The PPV&FR Act is thus, a balancing act to take care of its humongous agrarian base.

References

- Agricultural Statistics at a Glance (2021) Department of Agriculture, Cooperation & Farmers' Welfare, Ministry of Agriculture & Farmers' Welfare, Government of India, New Delhi.
- Annual Report (2020-21) Department of Agriculture, Cooperation & Farmers' Welfare, Ministry of Agriculture & Farmers' Welfare, Government of India, New Delhi.
- PPVFRA (2001) Protection of Plant Varieties and Farmers' Rights Act No. 53 of 2001. October 30, 2001. New Delhi, India.
- UPOV (1973) International Convention for the Protection of New Varieties of Plants, of December 2, 1961. Additional Act of November 10, 1972, Amending the International Convention for the Protection of Varieties of Plants adopted by the Diplomatic Conference on November 10, 1972. April 1, 1973, Geneva, Switzerland.
- UPOV (1991) International Convention for the Protection of New Varieties of Plants, of December 2, 1961 Amendment Act of November 10, 1972 revised and amended on March 19, 1991, Geneva. Switzerland.

Climate-Ready Genebanks for Climate-Smart Agriculture

Sunil Archak¹ and Himanshu Pathak^{2*}

¹ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

²Secretary (DARE) and Director General (ICAR), Krishi Bhavan, New Delhi-110001, India

Climate-smart agriculture demands development of new crop varieties involving the use of a wider range of intra-specific diversity so as to increase adaptability and resilience, and improve ecosystem services. Indian agriculture is well served by availability of disease-resistant or heat/drought tolerant. However, varietal development programmes, need to be constantly fed by genotypes with excellent adaptation potential. The best way to identify germplasm suitable for abiotic stress tolerance is to evaluate them based on specific traits. The challenge is to prepare the genebanks to be climate-ready in terms of availability of resilient germplasm accessions and planning and executing collection and conservation activities. Genebank housed at ICAR-NBPGR, needs to consider working on systematic identification of germplasm accessions that are multiple-stress tolerant using all possible methodologies including climate-analogue studies and field/lab evaluation.

Introduction

Climate has been the most important determinant of agricultural operations and yields in India. Indian farming, with more than 50% area being rainfed, is exposed to severe and multiple biotic and abiotic stresses making it vulnerable to fluctuating climate. Climate change makes Indian agriculture riskier and unsustainable (Pathak, 2022). India is considered to be one of the most vulnerable regions to witness climatic changes at a large scale. The unseasonal rain, drought and flood are rising while the duration of rainfall is reducing (IPCC, 2019). It is well known that, if climate goes wrong, Indian agriculture and consequently Indian economy stare down the barrel. Reports show that the changing climate can have multiple negative effects (individually or in combination) including (i) decline in yields of major crops by 3-18% (Naresh Kumar *et al.*, 2020); decline in cultivable area (e.g. rainfed rice area to decline by 15- 40%); (iii) significant change in areas of cultivation (e.g. coconut plantations to gain in west-coast but lose in the east-coast); (iv) Significant change in orchard altitude (e.g. apple belt shifts from 1250 msl to 2500 msl); (v) loss of economic product (e.g. lower output in Assam tea and Arabica coffee); (vi) reduction in quality of output (e.g. protein content in wheat to decrease by 1%; Zn/Fe to lower in many

food grains) (Pathak, 2022). It is also possible to have some positive impacts of climate change viz. expected yield gain due to elevated CO₂ level or increased arable areas due either to water availability in hot dry areas or to increased temperature in cold dry areas (Pathak, 2022).

Climate change exacerbates biotic stress faced by Indian agriculture in terms of pest/pathogen load, emergence of new strains and races, diminished scope for change in sowing time, choice of varieties, change in cropping systems and land use, etc. (Naresh Kumar *et al.*, 2020). Changing temperature regimes are expected influence the interactions among plants-pests-natural predators leading to excessive feeding on foliage, additional generations and increased pest load on crops, emergence of new invasive species (e.g., Asian fruit fly, blackfly, American tomato moth, fall armyworm and mango fruit borer) and intensified desert locust attack on crops. India can face a silent attack on nutritional status of low-income households because increased pests and diseases on home gardens, which provide substantial supplementary nutrition to rural families.

Indian Agriculture Needs to be Climate-smart

To address the emerging challenges and harness a few benefits of climate change, Indian agriculture needs to be climate-smart, which is defined as “an

*Author for Correspondence: Email-hpathak.iari@gmail.com

integrated approach for developing technical, policy and investment conditions to achieve sustainable agricultural development for food security under climate change” (FAO, 2013). Technologies and policy options for climate smart agriculture include weather (forecasting, agro-advisory, geo-ICT delivery, eco-regional crop planning), crop, water (increased efficiency in micro-irrigation, rainwater harvesting, drainage and approaches like direct-seeded rice), nutrient management, (site-specific Integrated nutrient management, neem-coated urea and bio-fertilizer), livestock (stress tolerant breeds, reviving small ruminants, managing feed, shelter and health), fisheries (composite/cage/wastewater culture and diversification), energy (conservation agriculture/protected cultivation, energy plantation), policy (contingency plan, insurance, credit, incentivization, seed bank) (Fig. 1, top frame) (Pathak, 2021). Among these options, working with the “crops” option requires access to plant genetic resources (PGR) in all their forms (multi-stress tolerant and input-efficient varieties, diversification, cultivation of new crops) (Fig. 1, middle frame). Many other researchers have also identified genetic solution to develop varieties that are inherently robust and resilient.

When various strategies and technologies advocated by experts were compared for their potential benefits to

achieve climate smart agriculture in India, the genetic resources-based technology —revolving around adoption of new crops/development of stress tolerant varieties/diversification —performed the best. Novel genetic resources would infuse excellent adaption benefit, productivity gain, income gain along with ease of implementation and suitability to small farmers (Agrawal *et al.*, 2021).

PGR for the Rescue

On one hand, climate change may render cultivated crop varieties and perennial trees inadequate to survive or to be economically viable underscoring the importance of access to PGR sources from different regions and countries. For instance, after centuries of breeding, wheat has a narrow genetic base making it difficult to breed new varieties with increased yields tolerance to biotic and abiotic stresses that are anticipated with climate change. Genebank accessions and crop wild relatives are potential sources of new genetic diversity (Kumar *et al.*, 2016). A systematic approach (including pre-breeding) is required to identify suitable accessions. On the other, climate change has brought the wild, neglected and underutilized species into focus. Neglected and underused crops are domesticated plant species that have been traditionally used for centuries in India for

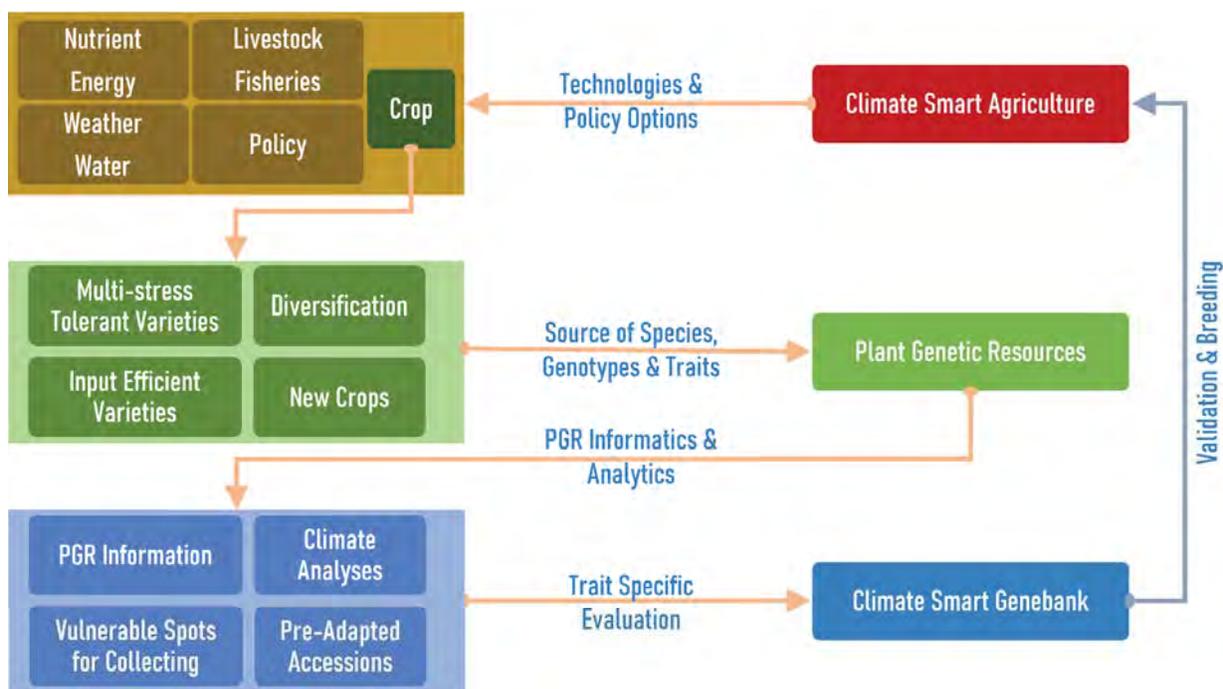


Fig. 1. Climate smart genebank an important prerequisite for achieving climate smart agriculture in India

their food, fibre, fodder or medicinal properties, but have been reduced in importance over time because of preponderance of “green revolution” crops. Fortunately, India has a number of such species distributed across different agroecologies and habitats.

ICAR-NBPGR houses one of the top three genebanks in the world. Many programmes on evaluating germplasm to identify accessions tolerant to biotic stresses or suitable for abiotic stress conditions. For instance, multi-location evaluation of wheat germplasm conserved in the Indian National Genebank to identify sources of resistance to powdery mildew and resistance to rust and spot blotch diseases (Kumar *et al.*, 2016; Vikas *et al.*, 2020) was successfully carried out with funding from NICRA funding of ICAR. However, such trait specific evaluation studies are challenging because of large number of accessions. Alternative approaches, including genomics or climate analysis, to choose a small sample for detailed studies need to be explored.

Climate Analysis of PGR

It is possible to link specific geographic origins of germplasm accessions with current and future climatic data. By effectively accessing and interpreting such information, one could shortlist prospective germplasm accessions that are pre-adapted (value addition to genebank collections) to predicted changes in climate. This approach could improve the resilience and capacity of agricultural systems to adapt to environmental changes in India. It also meant that germplasm collection activities could be planned based on climate analysis and identification of sites immediately vulnerable to climatic changes (Fig. 1, bottom frame).

A pilot study carried out by NBPGR researchers has presented realistic possibilities of the approach (NBPGR 2015). Passport information on 64,467 accessions belonging to two each of cereals (rice and wheat), millets (sorghum and pearl millet), pulses (pigeon pea and chickpea), oilseeds (brassica and sesame) and vegetables (capsicum and brinjal) were georeferenced based on their collection sites and the locations were mapped. Using climatic data from the Worldclim database for current climate (1950-2000) and from UKMO HADCM3 Climate Model for near future (2010-2039), analyses for the changes in the mean maximum temperatures during the cropping season for each of the ten crop species were carried out. Climate maps, depicting the possible locations of germplasm occurrence on current and future

temperature maps, could be generated for all the ten crops. Based on locations (source and test sites), climate matching, available agronomic performance data and seed availability in the genebank, 12 wheat accessions, 875 rice accessions, 150 sorghum accessions, 822 pearl millet accessions, 82 chickpea accessions, 43 pigeon pea accessions, 99 accessions of sesame wild relatives, 198 chilli-pepper accessions and 12 accessions of brinjal wild relatives could be designated as pre-adapted. As many as 2039 Taluks for wheat, 912 for rice, 593 for pearl millet, 541 for sorghum (*kharif*), 1174 for sorghum (*rabi*), 1445 for chickpea, 728 for pigeon pea 178 for oil seed brassica, 912 for sesame CWR 616 for chilli-pepper and 563 for brinjal CWR were predicted as vulnerable and to draw immediate attention to conduct collection missions.

Conclusion and Action Points

Genebanks need to leverage all the available tools and technologies ranging from genomics to hyperspectral analyses to maximize the identification of trait specific germplasm for their immediate use. Climate-smart practices will be well served with a bouquet of options in terms of choice of germplasm and trait combinations. Authors recommend the following actions in this regard:

1. Establish robust PGR databases and build PGR analytics tools to link habitat data, climate data and trait data.
2. Exploit the power of GIS to link multiple sets of information.
3. Explore the power and potential of climate analog tools for value addition to the conserved germplasm as well as for the identification of vulnerable sites.
4. Coordinate with breeders and domain experts to plan and implement experiments aimed at trait specific evaluation for biotic stresses.
5. Establish standard procedures and infrastructure for screening germplasm against abiotic stress factors.

References

- Agrawal *et al.*, 2021 J Roy, H Pathak, SN Kumar, B Venkateswarlu, A Ghosh and D Ghosh. (2022). Managing Climatic Risks in Agriculture. In: Chand, R., Joshi, P., Khadka, S. (eds) Indian Agriculture Towards 2030. India Studies in Business and Economics. Springer, Singapore. https://doi.org/10.1007/978-981-19-0763-0_4
- IPCC. 2019. Executive Summary. In Climate Change and Land: An IPCC Special Report on Climate Change, Desertification, Land Degradation, Sustainable Land Management, Food

- Security, and Greenhouse Gas Fluxes in Terrestrial Ecosystems (P. Shukla, J. Skea, E. Calvo Buendia, V. Masson-Delmotte et al., Eds.). Intergovernmental Panel on Climate Change.
- Kumar S, Archak S et al (2016). Evaluation of 19,460 wheat accessions conserved in the Indian national genebank to identify new sources of resistance to rust and spot blotch diseases PLoS ONE **11(12)**: e0167702.
- NBPGR (2015). Utilization of *ex situ* collections and climate analogues for enhancing adaptive capacity to climate change: Project Report. National Bureau of Plant Genetic Resources, New Delhi. India. pp. 96.
- Pathak H (2021). Making Indian Agriculture Climate Smart. In “Challenges in fertilizer challenges in fertilizer and agriculture” Papers presented at the FAI Annual Seminar December 1-3, 2021.
- Pathak H (2022) Impact, adaptation and mitigation of climate change in Indian agriculture. Environmental Monitoring and Assessment (In press).
- Vikas VK, Kumar S, Archak S, et al (2020). Screening of 19,460 genotypes of wheat species for resistance to powdery mildew and identification of potential candidate using FIGS approach Crop Science DOI: 10.1002/csc2.20196.

The Role of CGIAR in the Global System of PGRFA Conservation and Use

Kuldeep Singh^{1*}, Michael Halewood^{2*}, Charlotte Lusty³ and HD Upadhyaya⁴

¹Head Genebank, International Crops Research Institute for the Semi-Arid Tropics, Hyderabad-500030, Telangana, India

²Bioversity International-CIAT Alliance

³Global Crop Diversity Trust

⁴Plant Genome Mapping Laboratory, University of Georgia

Introduction

This short paper is dedicated to celebrating the 80th birthday of Dr RS Paroda, who has been champion of the genetic resources' conservation in India and a strong supporter of the CGIAR genebanks, especially the ICRISAT genebank that is named after him. This team wishes him many more years to his healthy life. This paper provides a high-level overview of the globally coordinated system for plant genetic resources for food and agriculture (PGRFA) conservation and sustainable use under the framework of the United National Food and Agriculture Organization (UN FAO). The paper also considers the role that CGIAR Centers, particularly their genebanks, have historically played within that system, and how recent reforms to the CGIAR system could influence the role it plays. Finally, the paper ends with a few recommendations for further development of the global system.

The Evolving Global System for PGRFA

For decades, under the auspice of the United Nations Food and Agricultural Organization, the international community has repeatedly committed itself to supporting a globally coordinated system for the conservation and sustainable use of plant genetic resources for food and agriculture. Over time, there has been a gradual accretion of international policies, standards, tools, and guidelines adopted by UN FAO, and by other organizations, that have formed a part of that global system. Some of the most important developments in this regard were creation of the FAO Commission on Plant Genetic Resources and adoption of the International Undertaking on Plant Genetic Resources for Food and Agriculture in 1983; and the adoption of the International Treaty on Plant Genetic Resources for Food and Agriculture (Plant Treaty) in

2001; adoption of the genebank standards, and revised genebank standards in 1994 and 2014 respectively.¹ These instruments create a platform for countries, international organizations, crop and genetic resources networks, and farmer, civil society and private sector organizations to compare notes, coordinate activities, hash-out contentious issues, develop best practices and standards, and ultimately collectively contribute to: increasing the scope of plant genetic resources that are safely conserved, exchanged, and sustainably used; increasing the quantity and quality of information about those PGRFA that is globally, publicly available, and increasing the equitable sharing benefits derived from the use of PGRFA.

Since the earliest efforts under FAO to promote a global system, the focus has tended to be on three fundamental components: i) an internationally linked system of *ex situ* PGRFA collections; ii) creation of a global information system comprising information about those collections and iii) creation of a fund to support strengthening of the capacity of developing countries to play an active role in conserving and using and benefitting from the PGRFA in the global system. There has been comparatively little energy and resources dedicated to *in situ* conservation and local level management of plant genetic diversity as part of the global system. Recent efforts to recognize and promote Farmers Rights are starting to address these lacunae, at least partially. Indeed, the 9th session of the Governing Body of the Plant Treaty to be held in India in September 2022 will consider (and we hope) adopt a document entitled *Options for encouraging, guiding,*

¹ Indeed, there are many relevant policies, guidelines and standards adopted by the UN CGRFA and Governing Body of the Plant Treaty to list here.

*Author for Correspondence: Email-kuldeep.singh@cgiar.org; m.halewood@cgiar.org

and promoting the realization of Farmers’ Rights, and develop a program to support implementation of those options within countries over the next biennia.

There has also been comparatively little international attention or resources dedicated – at the level of the Plant Treaty’s Governing Body, or the UN FAO CGRFA or FAO, more generally under the rubric of global system, on supporting plant breeding *per se*. This may be due to a perception that plant breeding is already being effectively addressed and practiced by national, international, public, and private sector organizations (or the hope that it will be), or it could simply be a reflection of the fact that supporting national, or sub-regionally organized plant breeding programs has been beyond the resource constraints of the UN FAO.

The Role of CGIAR within the Global System

The international community has gone to considerable lengths to ensure that the CGIAR Centers and the collections they host are part of the global system. This is important given that these collections are among the most genetically diverse, and most used, in the world. In 1994, with the approval of the UN FAO Commission on Plant Genetic Resources, FAO entered into legal agreements with CGIAR Centers hosting *ex situ* collections (the FAO-CGIAR In Trust Agreements) to confirm the legal status of those collections as being held in trust by the CGIAR Centers on behalf of the international community, making them available without restriction for conservation and agricultural research and development. The Plant Treaty includes an invitation to CGIAR Centers (in Article 15) to enter into agreements with the Governing Body of the Plant Treaty to agree to manage those collections under the Plant Treaty framework, making materials available under the terms and conditions of the multilateral system of access and benefit sharing. In 2006, the Centers concluded such Article 15 agreements and have been managing their collections and taking policy advice from the Governing Body ever since. The Centers’ genebanks act like global PGRFA pumps, acquiring and conserving materials from all over the world, and then redistributing it to the recipients all around the world along with associated information. In this sense, the Centers’ operations reflect, and perpetuate, the interdependence of all countries in the world on PGRFA. Those Centers currently conserve, and make available, using the standard material transfer agreement (SMTA), a total of 722,525 accessions of

crop, tree, and forage germplasm, as set out in Tables 1 and 2, below.

Table 1. PGRFA conserved and made available by CGIAR Centers’ genebanks pursuant to their Article 15 agreements with the Governing Body of the Plant Treaty

Center	Crop	Accessions available with SMTA
AfricaRice	Rice	19,696
Bioversity	Banana	1,682
CIAT	Beans	37,934
	Cassava	5,965
	Forages	22,662
CIMMYT	Maize	28,694
	Wheat	135,021
CIP	Andean roots and tubers	1,178
	Potato	7,367
	sweet potato	6,143
ICARDA	Barley	31,843
	Chickpea	15,230
	faba bean	9,594
	Forages	25,358
	Grasspea	4,301
	Lentil	14,295
	Pea	4,593
	Wheat	41,967
ICRAF	fruit trees	8,246
	multipurpose trees	6,744
ICRISAT	Chickpea	20,258
	Groundnut	15,237
	pearl millet	24,355
	pigeon pea	13,534
	small millets	11,683
	Sorghum	42,750
IITA	Banana	392
	Cassava	3,184
	Cowpea	17,051
	Maize	1,561
	misc legumes	6,747
	Yam	5,929
ILRI	Forages	3,918
IRRI	Rice	127,413
	Total	722,525

Source: Global Crop Diversity Trust/CGIAR On-line Reporting Tool, covering period up to December 31, 2021.

Table 2. CGIAR Centers' transfers of PGRFA with SMTAs, 2007 to 2021/22. PUD stands for PGRFA under Development

Center	SMTAs	Samples	PUD	From	To
AfricaRice	598	53,351	30,714	2007-03-05	2022-01-29
Bioversity	554	8,595	831	2007-01-24	2021-11-18
CIAT	3,077	298,291	36,646	2007-01-05	2022-02-24
CIMMYT	26,644	3,211,789	-	2007-03-16	2021-12-21
CIP	788	22,503	12,221	2007-01-19	2022-05-05
ICARDA	11,531	992,246	873,178	2007-02-13	2021-12-21
ICRAF	358	1,719	-	2011-09-03	2022-03-03
ICRISAT	6,048	297,367	100,532	2006-12-07	2021-11-26
IITA	1,172	48,891	-	2007-03-07	2022-02-09
ILRI	944	13,712	-	2007-02-22	2022-02-14
IRRI	9,317	764,594	441,382	2007-01-04	2022-02-09

Source: Global Crop Diversity Trust/CGIAR On-line Reporting Tool, covering period up to December 31, 2021

The CGIAR centers also host very important crop and forage improvement programs focusing on a wide range of species/genera, including maize, wheat, rice, pigeon pea, cowpea, sorghum, finger millet, pearl millet, barnyard millet, proso millet, kodo millet, little millet, potato, cassava, banana, chickpea, lentils, barley, ground nut, yam, potato, sweet potato, lathyrus, grass pea, etc.

In the 16 years, since signing their Article 15 agreements with the Plant Treaty's governing body, the CGIAR Centers have distributed over 6 million

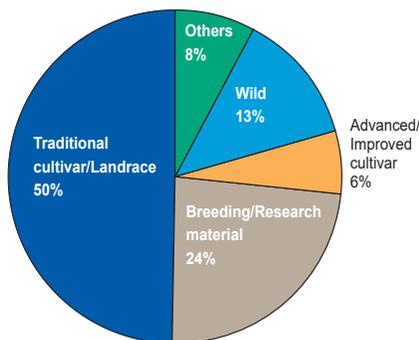
PGRFA samples to recipients around the world under the Plant Treaty's framework (Table 2). The majority of the PGRFA frequently requested and distributed between 2017 and 2019 were traditional landraces/cultivars (50%), followed by breeding materials (24%), and wild relatives (13%) (Fig. 1). Eighty percent of the materials were sent to recipients in developing countries. As far as regional distributions are concerned, the largest percentage of materials from CGIAR centers (29%) were sent to recipients in Asia; 19% distribute to

Box 1. ICRISAT genebank case study

The germplasm supplied by CGIAR centers has played a critical role in supporting NARS crop breeding programs and agriculture. To cite an example, the germplasm supplied by ICRISAT Genebank, 114 germplasm accessions have been directly released as 153 cultivars in 52 countries. Some of the germplasms released as cultivars have significant economic impact. For example, ICP 8863, a pigeonpea landrace resistant to fusarium wilt was released in 1986 as Maruthi for cultivation in Karnataka state of India. The value of benefits from ICP 8863 was estimated at US\$ 75 million by 1996 with 73% internal rate of return. The Crop Trust Impact story of direct germplasm release of ICP 8863 <https://www.genebanks.org/news-activities/news/maruthi/>.

Enhancing use of germplasm in crop improvement is an important research area where ICRISAT genebank has played seminal role. Having recognized that less than 1% germplasm has been used in most crop improvement programs, and development of core collections (10% of entire collection) was not helpful where number accession was large, Upadhyaya and Ortiz, 2001 (*Theor. Appl. Genet.* **102**: 1292–98) postulated concept of mini core (10% of core or 1% of entire collection) and proposed development process, to produce resources recognized globally as an "International Public Good" (IPG). New diverse sources of genetic variation have been identified from mini core collections of various crops for multiple traits including biotic and abiotic stresses, agronomic and nutritional traits for use in crop improvement programs (Upadhyaya et al., 2013 – *Crop Sci.* **53**: 2506–2517; Upadhyaya et al., 2014 – *Crop Sci.* **54**: 679-693; Upadhyaya et al., 2019 – *Agriculture* **9**, 121). These multiple trait diverse sources would be extremely useful in developing high yielding climate resilient and nutrition rich crop cultivars with a broad genetic base and having greater genetic gains.

(a) All Centres



(b) By Center

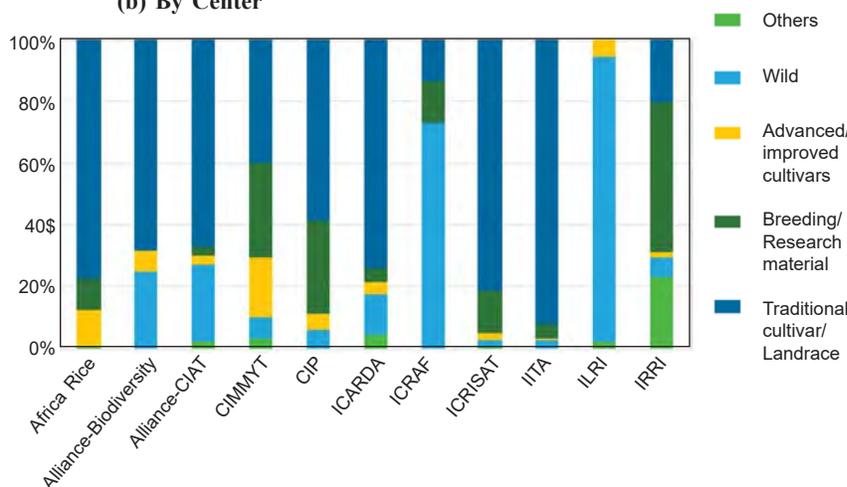


Fig. 1. Types of germplasm distributed by CGIAR genebanks to all centers (a) and by the centers (b), 2017–2019 (Michael *et al.*, 2020 - *Plants*, 9, 1296).

recipients in Africa; 19% to recipients in Europe, 14% to recipients in Latin America and the Caribbean; 11% in the Near East and 7% in North America. Transfers from CGIAR Centers account for 89% of all reported transfers under the Plant Treaty’s multilateral system of access and benefit sharing. The germplasm distributed through genebanks has been evaluated and released as varieties after direct selection from the germplasm (Box 1) as well as used as parents for crossing or sources for important traits/genes.

The CGIAR works through partnerships, primarily with national research and development organizations in developing countries. As such, technology transfer, capacity strengthening, information sharing, – all important forms of non-monetary benefit sharing associated with PGRFA – are built into CGIAR Centers’ modus operandi and reflected in their day-to-day operations. In recent years, Centers have been asked to report to the CGIAR System Office on how they have been promoting Farmers’ Rights in particular in the context of their day-to-day R&D activities. Highlights are included in the Centers’ biennial reports to the Plant Treaty, and in the annual CGIAR Intellectual Asset Management reports.

Recent CGIAR Reforms

The CGIAR is currently in a system-wide reform, including the development of a ‘One CGIAR’ governance arrangement, aiming to increase efficiency and effectiveness in response to evolving global challenges (Fig. 2). As part of those reforms, CGIAR adopted

‘CGIAR 2030 Research and Innovation Strategy: Transforming food, land, and water systems in a climate crisis’. Under the new governance structure, most research and development initiatives working directly with genetic resources for food and agriculture (including the work of genebanks, breeding programs and seed systems R&D) take place under ‘Genetic Innovation’. This arrangement is conceived to getting faster reflections/ feedback/ participation from leaders who collaborate across research groups, and from top CGIAR leadership on important evolving issues, including CGIAR genetic resources management policies. *In situ* conservation and dynamic *in situ/ex situ* PGRFA conservation are still not explicitly integrated into the work of the Genebank Initiative. Some such work is included in the Initiative entitled Nature Positive Solutions, but one could argue that more targeted support - including strong involvement of NARS would be useful and justified.

The ‘OneCGIAR’ reform process has involved considerable realignment and integration of the governance and operational structure of the CGIAR System overall, but the CGIAR Centers will maintain their legal status as independent legal entities in their own right, and that their Article 15 agreements with the Governing Body of the Plant Treaty will remain in place. CGIAR considers the ‘in trust’ plant genetic resources maintained by the CGIAR genebanks to be of inestimable value for the international community. CGIAR remains committed to supporting the continued management of collections hosted by International Agricultural Research

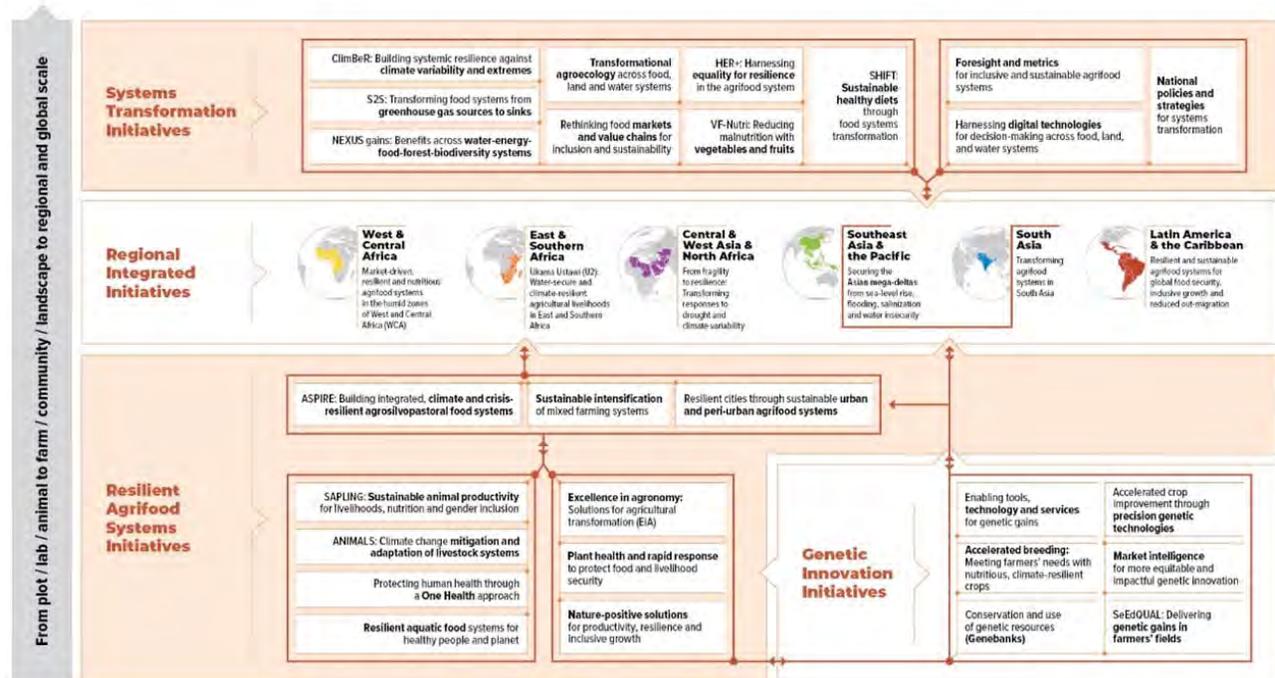


Fig. 2. One CGIAR Research Initiatives

Centers (IARCs), including within 'OneCGIAR' under the framework of the Plant Treaty.

Although CIFOR-ICRAF and ICRISAT have not adopted for 'OneCGIAR' unified governance arrangements at this time, they remain CGIAR Centers that hold Article 15 agreements with the Plant Treaty's Governing Body. ICRISAT and ICRAF will continue to ensure their genebanks are operated and maintained as per international standards. ICRISAT and ICRAF will continue to conserve the germplasm for the international community, under the auspices of the UN FAO and the Plant Treaty, as per the agreements signed in 1994 and 2006, and will continue to deliver their missions to international agriculture, food systems and landscape stewardship.

One of the consequences of opting out is that these Centres do not have access to the same financial support from the OneCGIAR Trust fund as Centers that have joined the OneCGIAR governance arrangement. Funding from the Crop Trust, however, is not affected by the OneCGIAR reform. Alternative sources of funding are actively being sought as well as additional funds from the Global Crop Diversity Trust (Crop Trust). The long-term grants already in place with nine CGIAR Centre genebanks continue to provide funding for genebank operations. In addition, as genebanks

reach performance targets they become eligible for a larger Long Term Partnership Agreement (LPA), which provides enough funding to cover all essential genebank operations. ICRISAT is among a number of international genebanks that are approaching eligibility for an LPA. ICRISAT, CIFOR-ICRAF and the CGIAR senior leadership are also engaged in discussions to work out the conditions for future cooperation, coordination, and mutual supportiveness. ICRISAT and CIFOR-ICRAF continue to be international institutions and CGIAR Centers. Their long-standing Article 15 agreements with the Plant Treaty's governing body are not affected.

Going Forward in the Context of an Expanding, more Efficient, Global System

Moving forward in the further evolution of the global system, it will be important for all article 15 organizations and other international institutions hosting PGRFA under the Plant Treaty framework to work more closely together, developing stronger network ties, to enhance their collective contributions. These organizations include CGIAR Centers inside and outside the new OneCGIAR governance arrangement, International Center for Biosaline Agriculture, FAO Mutant Germplasm Repository, International Coconut Genebank for the South Pacific, International Coconut Genebank for African and the Indian Ocean, International

Cocoa Genebank – Centro Agronomico Tropical de Investigacion u Ensenanza (CATIE), Centre for Pacific Crops and Trees (CePACT), World Vegetable Center, and SADC Plant Genetic Resources Center (SPGRC).

Efficient functioning of the global system will also depend on stronger linkages and better-defined sharing of responsibilities for all actors including national and international collection holders, and organizations involved in promoting *in situ* conservation and sustainable use. As part of that effort, we flag the following important initiatives:

- Coordinated analysis of gaps in globally accessible national and international collections
- Work on the West and Central African regional hub (possibly other hubs)
- Global Plant Cryopreservation Initiative
- Participation in the Crop Trust’s Global Genebank Partnership,
- Capacity building for a much wider range of actors to both conserve, access and to use PGRFA and related information for addressing pressing local to global challenges including climate change, and nutrition deficits
- Promote development and operationalizing a more supportive policy environment through enhancement of the Plant Treaty’s multilateral system of access and benefit sharing, and mutually supportive implementation of the Plant Treaty, Nagoya Protocol, IPPC, etc
- Contribute to international discussions/negotiations with respect to setting targets for, and monitoring, biodiversity conservation, generating and sharing benefits (and monitoring that sharing) through the use of digital sequence information
- Developing pangenebank (IARC and NARS genebanks) unified collections using high throughput genomics approaches

Genomics-Driven Application of Plant Genetic Resources for Sustainable Agriculture

Rajeev K Varshney*, Abhishek Bohra and Vanika Garg

Murdoch's Centre for Crop and Food Innovation, State Agricultural Biotechnology Centre, Food Futures Institute, Murdoch University, Murdoch, Western Australia, 6150, Australia.

Genebanks preserve the valuable genetic diversity in the form of huge collections of plant genetic resources (PGR). Dr Raj S. Paroda has spearheaded the modernization of one of the world's largest genebanks, the ICAR-National Bureau of Plant Genetic Resources (NBPGR), New Delhi, India, which currently houses nearly 462,466 accessions. Dr Paroda's notable contributions in the field of PGR management have earned him the popular name of "Genebank Guru". As Dr Paroda turns 80 on August 28, 2022, we commemorate the occasion by synthesizing an article in line with Dr Paroda's forward-thinking vision for PGR management and utilization. The present article highlights how modern genetic technologies have expanded our capacity to characterize and exploit the unexplored genetic diversity from genebanks. Extensive molecular and phenotypic characterisation support efficient strategies to minimize large PGR collections to workable sizes, thus allowing targeted search of plant diversity for sustainable agriculture. As the technological innovations transform genebanks into "bio-digital resource centres", availability of detailed genotype-phenotype maps and genomic predictions for large PGR collections will guide selection and breeding decisions in crop improvement programmes. Effective preservation and use of PGR collections is imperative to impart climate change adaptation to crop production systems and safeguarding our future food supply.

Key Words: Accession, Genebank, Genome, Phenotype, Plant genetic resources, Trait

Introduction

Plant genetic resources (PGR) harbour genetic variation that serves as raw material for breeding and selection decisions in crop improvement programmes. Padma Bhushan Dr Raj S. Paroda [Chairman, Trust for Advancement of Agricultural Sciences (TAAS) and Former Secretary DARE & DG, ICAR] has played a pivotal role in transformation of Indian national PGR system, represented by the ICAR-National Bureau of Plant Genetic Resources (NBPGR), New Delhi. To recognize his immense contribution in the field of PGR management, two genebanks have been named after him: 'RS Paroda Genebank' at ICRISAT (<https://www.genebanks.org/genebanks/icrisat/>) and genebank at the Agricultural Research Institute in Kazakhstan.

In recent years, evolving DNA sequencing and plant phenotyping have facilitated detailed characterisation and evaluation of PGR. For instance, we recently sequenced the genomes of 3,366 chickpea accessions from 'RS. Paroda Genebank' and provided genomic variations and haplotypes for future chickpea improvement

(Varshney *et al.*, 2021). Conforming to the vision of Dr Paroda, our article discusses how technological developments have enhanced efficiency of PGR management and their use in plant breeding and research. We begin with presenting a brief overview of major germplasm holdings across the globe. We then discuss the role of evolving DNA sequencing/genotyping and phenotyping systems in developing strategies for sustainable use of PGR. We also underscore the importance of targeted collection of under-represented germplasm from "hotspot" regions to bridge the conservation gaps in genebanks.

Germplasm Holdings Across Major Genebanks Worldwide

Protection of plant genetic diversity is essential to sustainable increase in crop production and food security. *Ex situ* collection constitutes the major form of germplasm conservation that aims to conserve germplasm in controlled environments, away from their native habitats. An alternative germplasm conservation strategy

*Author for Correspondence: Email-rajeev.varshney@murdoch.edu.au

stores accessions *in situ* in their natural habitats, allowing creation of new genetic variation because of continuous evolution. The growing realization of the urgent need of protecting genetic diversity for sustainable future has led to the establishment of the Svalbard Global Seed Vault by the Government of Norway (<https://www.seedvault.no/>). As a backup facility to the world's crop diversity, the global seed vault currently secures 1,165,041 seed samples from different genebanks including ICRISAT (Fig. 1 a, b).

According to the Second Report on The State of the World's Plant Genetic Resources for Food and Agriculture, a total of 7.4 million accessions are preserved in 1,750 genebanks worldwide. The 11 genebanks under the Consultative Group on International Agricultural Research (CGIAR) network contain 773,112 accessions. Though the accessions held in CGIAR genebanks represent 10% of the total accessions, the CGIAR network accounts for nearly 94% total germplasm shared in line with the International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA) framework. The Convention on Biological Diversity (CBD) and ITPGRFA constitute the mechanism of germplasm exchange across the globe. Besides, several national repositories also conserve germplasm accessions that protect genetic diversity for current and future use of breeders, researchers, farmers and other stakeholders. The other major national repositories National Plant

Germplasm System (NPGS), USA; ICAR-National Bureau of Plant Genetic Resources (NBPGR), India; Institute of Crop Germplasm Resources (ICGR)-Chinese Academy of Agricultural Sciences (CAAS), China; N.I. Vavilov All Russian Institute of Plant Genetic Resources (VIR), Russia; National Agriculture and Food Research Organization (NARO), Japan; Leibniz-Institute of Plant Genetics and Crop Plant Research (IPK), Germany; and Canadian National Plant Germplasm System (CNPGRS), Canada collectively hold 2,282,718 accessions. The ICAR-National Bureau of Plant Genetic Resources (NBPGR), India represents the second largest genebank in the world that currently houses 462,466 accessions (http://www.nbpgr.ernet.in/Research_Projects/Base_Collection_in_NGB.aspx).

Generation of Molecular Data on Large-scale Collections

Advancements in DNA sequencing technologies have led remarkable expansion in our capacity to perform in-depth molecular characterization of large PGR collections. In different crops, tens of thousands of genetic markers have been assayed on large PGR collections to elucidate the vast amount of genetic diversity they contain. Earlier, simple sequence repeat (SSR) markers were used to examine the genetic diversity of genebank accessions. For instance, the analysis of genotyping data and genetic structure of a total of 3,367 accessions from global



Fig. 1. a) Entrance to the Svalbard Global Seed Vault and b) Transferring seed boxes containing seeds of ICRISAT crops to the Seed Vault (Prof. Rajeev Varshney and Dr. Hari D. Upadhyaya, the then Research Program Director- Genetic Gains, and Head, Genebank, respectively at ICRISAT).. The global seed vault stores 1,165,041 backup seeds of 5947 species from 91 genebanks. The global seed vault provides seed backups of world's several genebanks. ICRISAT has deposited nearly 90% of its genebank seed as backup in the vault.

Table 1. Characterization of diverse germplasm collections based on whole genome sequence information in some crop species

Crop	Total accessions sequenced	Cultivated	Landraces	Wild	Others	Reference
Chickpea (<i>Cicer arietinum</i>)	3,366	548	2,439	195	184	Varshney et al., 2021
	429	144	268	7	10	Varshney et al., 2019
Pigeonpea (<i>Cajanus cajan</i>)	292	117	166	7	2	Varshney et al., 2017a
Soybean (<i>Glycine max</i>)	2,898	1,747	1,048	103	-	Liu et al., 2020
	1,110	228	723	157	2	Bayer et al., 2022
Sunflower (<i>Helianthus annuus</i>)	493	287	17	189	-	Hübner et al., 2019
Common bean (<i>Phaseolus vulgaris</i>)	683	154	529	-	-	Wu et al., 2020
Pearl millet (<i>Pennisetum glaucum</i>)	994	963	-	31	-	Varshney et al., 2017b
Rice (<i>Oryza sativa</i>)	3,010	3,010	-	-	-	Wang et al., 2018
	1,143	1,143	-	-	-	Lv et al., 2020
Maize (<i>Zea mays</i>)	103	60	23	19	1	Chia et al., 2012
Lettuce (<i>Lactuca sativa</i>)	445	131	-	314	-	Wei et al., 2021
Sorghum (<i>Sorghum bicolor</i>)	44	17	18	7	2	Mace et al., 2013
Tea (<i>Camellia sinensis</i>)	81	58	20	3	-	Xia et al., 2020
Cotton (<i>Gossypium</i> L.)	419	419	-	-	-	Ma et al., 2018
	318	271	35	12	-	Fang et al., 2017
Grape (<i>Vitis vinifera</i>)	472	329	-	143	-	Liang et al., 2019
Spinach (<i>Spinacia oleracea</i>)	305	295	-	10	-	Cai et al., 2021
Tartary Buckwheat (<i>Fagopyrum tataricum</i>)	510	-	478	32	-	Zhang et al., 2021

composite germplasm collection represented majorly by landraces (89.5%) with 41 SSR markers allowed formation of ‘Reference Set’, a smaller representative subset of accessions for future use (Billot et al., 2013). A paradigm shift in marker genotyping assays and sequencing technologies has reflected in genetic profiling of plant genotypes (Garg et al., 2021). Table 1 shows the examples of genetic profiling large number of accessions as elucidated by whole genome resequencing. Next generation sequencing (NGS)-based methods based on reduced representation are particularly suitable for genetic profiling of large genebank collections. Notable examples include genetic profiling of 21,405 IPK barley accessions including domesticated and wild barley using genotyping-by-sequencing (GBS), thus revealing genetic variations in the form of 171,263 SNPs (Milner et al., 2019). Detailed genetic profiles of PGR offer opportunity to understand the molecular basis of

crop domestication and evaluation. The high-density genotyping datasets can be combined with historical evaluation records to develop genotype-phenotype maps for the PGR collections and genetically dissect the variation in agriculturally important traits using genome-wide association studies (GWAS). Examination of the genetic similarities based on genome-wide marker data may help revealing the redundancies within and between collections besides aiding in identification of collection gaps and mislabelling of biological status in the historical records in genebanks across the globe (Bohra et al., 2022a). Recent genome sequencing of 3,366 chickpea accessions allowed identification and correction in labelling of the chickpea accessions ICC 16369 (Varshney et al., 2021). The mislabelling of ICC 16369 as cultivated was evident owing to the presence of wild-specific allele of the *SHATTERPROOF2* homolog.

Next-generation Phenotyping to Assist PGR Characterization

Characterization and evaluation of PGR is crucial to realize their true potential for genetic research and breeding. Plant phenotyping has always remained a great challenge in crop management and improvement. The traditional methods of monitoring phenotypic changes in plant morphological traits are cumbersome, destructive, costly, and inaccurate. Furthermore, the substantial influence of genotype \times environment interaction on phenotypic expression challenges the efficacy of the traditional methods of PGR characterization and evaluation. In recent years, advances in imaging and sensor technology have enabled automation of measurements of anatomical, physiological and biochemical changes in plants. Availability of such high-throughput phenotyping platforms have facilitated rapid plant phenotyping at different growth stages in accurate, cost-effective and non-invasive manner. Different kinds of imaging platforms including RGB, thermal, fluorescence, laser scanning, tomography, multispectral and hyperspectral offer plant phenotyping at varying scales i.e. microscope, laboratory, glasshouse, field and satellite (Nguyen *et al.*, 2022). Several national (<https://www.plantphenotyping.org/>; <https://www.plantphenomics.org.au/>) and international networks (<https://www.plantphenotyping.org/>) have been established across the globe to allow scientists/stakeholders in academia and industry accessing cutting-edge tools, facilities and analytics of plant phenomics. In the wake of growing sequence information, the acquisition of high-quality data opens exciting avenues for identification of beneficial alleles preserved in the large genebanks. Association of the high-throughput phenotypes with genetic loci has been demonstrated in various crops including rice, wheat, maize, barley, soybean, sorghum, cotton, rapeseed etc (Xiao *et al.*, 2022).

Enhancing PGR use for Future Genetic Research and Crop Improvement

Generation of Trait-specific Subsets for Efficient Use

Huge collections of PGR accessions held in genebanks are the reservoir of valuable traits, which may be essential to impart climate adaptation and stress tolerance to future crop varieties. For example, Isleib *et al.* (2001) documented the economic value of the tomato spotted wilt virus (TSWV) resistance introgressed from PI

203396 into peanut (*Arachis hypogaea*) cultivars in the USA, which amounted to more than \$200 million annually. Despite this, the use of PGR from the large genebank collections remains limited largely to the poor morphological and molecular characterization of the germplasm. Less than 10% of the germplasm accessions held in genebanks have been used so far in breeding programs (Nguyen and Norton 2020). Lack of trait-specific subsets of crop germplasm has also been a major limitation in the selection and use of appropriate accessions in pre-breeding and varietal development. Strategies based on the development of manageable germplasm diversity subsets have been proposed to enhance the use of PGR from genebanks. Brown' core collection concept aimed to capture 70% diversity in a smaller collection comprising 10% of the total accessions. Core collections have been developed in various crops including rice, maize, wheat, soybean. Later, the concept of mini core collection gained popularity given the fact that the size of core collections still poses challenge in terms of evaluation and management, especially in crops having large germplasm holdings (Anglin *et al.*, 2018). Phenotyping data in combination with genetic information has allowed construction of mini core collections comprising 1% of the total accessions in different crops including chickpea, pigeonpea, and several other crops of semi-arid regions.

Another strategy for efficient use of large PGR collection is the focused identification of germplasm strategy (FIGS) that uses environmental information for customization of the germplasm sets to a workable size (Bhullar *et al.*, 2009). FIGS is based on the premise that suitable germplasm for adaptive traits can be sampled from the sites experiencing selection pressures for the particular trait (Bohra *et al.* 2022b, c). In wheat, authors could narrow down on 1,320 landraces from the large collection of 16,089 accessions through focusing on 323 geographic sites having strong selection pressure for powdery mildew (Bhullar *et al.*, 2009). This FIGS-customized set of wheat facilitated targeted search for useful genetic variation for the resistance (*Pm3 gene*) against powdery mildew disease. More recently, Haupt and Schmid (2020) made 'trait-focused' panels of 183 and 366 accessions in soybean for environmental adaptation by combining core collection and FIGS in a collection of >17,000 accessions, representing introduced landraces from the USDA Soybean Germplasm Collection. A combination of targeted approaches could prove highly

useful for identification of functional diversity for adaptation traits.

Genomic Predictions to ‘Turbocharge’ Genebanks

Availability of genotype and phenotype data on germplasm sets may help train prediction models to compute the genetic worth of millions of accessions archived in the genebanks. Genomic selection bypasses the need of repeated phenotyping, and predicts the worth of an unobserved individual based on the models trained on individuals having both genotype and phenotype scores. Yu *et al.*, 2016 recently demonstrated a proof-of-concept for genomic prediction as a promising strategy to aid selection and breeding decisions. The high density GBS genotyping enabled molecular characterization of sorghum reference set comprising 962 accessions with 340,496 single nucleotide polymorphisms (SNPs). The authors selected a set of 299 accessions as training population to predict yield of the 633 untested lines of the reference set. The study recorded high prediction accuracies for the untested lines of the reference set and a 200-accession validation set. Relatively poor prediction accuracies for an independent set of 580 exotic sorghum accessions suggested less representation of alleles from exotic lines in the training population derived from the reference set. The advances in genotyping and phenotyping would help extending the genomic prediction strategy from small subsets of accessions to whole genebank collection to enable efficient use of PGR.

Analysis of the Existing Gaps to Inform Future Collections

A systematic analysis of the representativeness of different taxa and species distributions in large *ex situ* collections is required for identification of the critical conservation gaps and setting of collection priorities (Ramírez-Villegas *et al.*, 2010). Advances in Geographic Information Systems (GIS) technologies have supported conservation planning efforts. With the help of GIS-modelled distribution, Ramírez-Villegas *et al.* (2010) identified *Phaseolus* wild relatives that are underrepresented in *ex situ* collections and the authors suggested ‘hot spot’ regions that should be targeted for future collection of high-priority germplasm.

Conclusions and Future Directions

Approaches to accessing the suitable germplasm are greatly benefited by the growing acquisition of molecular information and phenotyping data on large-

germplasm collections. Extensive phenotypic and genetic characterization will be crucial to unlock the full potential of PGR held in genebanks via revealing the genomic regions controlling traits that are important to biotic and abiotic stress adaptation (Haupt and Schmid, 2020). The high-volume and complexity of the phenotyping data offers a new set of challenges in analysis; such as the need for data/image processing algorithms to analyse the data acquired from various imaging systems. The adoption of the new-generation plant phenotyping will rely on standardization of the high-volume datasets, which in turn highlights the growing requirement of new digital datasets to remain adhered with the guidelines or the checklists provided in the Findability, Accessibility, Interoperability, and Reuse (FAIR; <https://www.go-fair.org/fair-principles/>) and Minimum Information About a Plant Phenotyping Experiment (MIAPPE; <https://www.miappe.org/>). The rising availability of big data on PGR resulting from latest innovations in genetic and phenotyping technologies poses new challenges in terms of networking and equitable sharing of the resources and information. In this context, coordinated efforts are needed to address the access and benefit sharing issues particularly in relation to the developing countries.

Acknowledgements

Authors are thankful to Food Futures Institute, Murdoch University (Australia) for financial support. RKV is thankful to all colleagues at ICRISAT and its collaborators, and donors for their contributions to germplasm research during his tenure at ICRISAT.

References

- Anglin NL, A Amri, Z Kehel, and D Ellis (2018) A case of need: linking traits to genebank accessions. *Biopreserv. Biobank.* **16**: 337-349.
- Bayer PE, B Valliyodan, H Hu, JI Marsh, Y Yuan, TD Vuong, *et al.* (2022) Sequencing the USDA core soybean collection reveals gene loss during domestication and breeding. *Plant Genome* **15**: e20109.
- Bhullar NK, K Street, M Mackay, N Yahiaoui, and B Keller (2009) Unlocking wheat genetic resources for the molecular identification of previously undescribed functional alleles at the Pm3 resistance locus. *Proc. Natl. Acad. Sci. U.S.A.* **106**: 9519-9524.
- Billot C, P Ramu, S Bouchet, J Chanterreau, M Deu, L Gardes, *et al.* (2013) Massive sorghum collection genotyped with SSR markers to enhance use of global genetic resources. *PLoS One* **8**: e59714.

- Bohra A, Bansal KC, Graner A (2022a) The 3,366 chickpea genomes for research and breeding. *Trends Plant Sci.* **27**: 217-219.
- Bohra A, B Kilian, S Sivasankar, M Caccamo, C Mba, SR McCouch, *et al.* (2022b) Reap the crop wild relatives for breeding future crops. *Trends Biotechnol.* **40**: 412-431.
- Bohra A, A Tiwari, P Kaur, SA Ganie, A Raza, M Roorkiwal, *et al.* (2022c) The key to the future lies in the past: insights from grain legume domestication and improvement should inform future breeding strategies. *Plant Cell Physiol.* doi:10.1093/pcp/pcac086.
- Cai X, X Sun, C Xu, H Sun, X Wang, C Ge, *et al.* (2021) Genomic analyses provide insights into spinach domestication and the genetic basis of agronomic traits. *Nat. Commun.* **12**: 7246.
- Chia JM, C Song, PJ Bradbury, D Costich, N de Leon, J Doebley, *et al.* (2012) Maize HapMap2 identifies extant variation from a genome in flux. *Nat. Genet.* **44**: 803-807.
- Fang L, Q Wang, Y Hu, Y Jia, J Chen, B Liu, *et al.* (2017) Genomic analyses in cotton identify signatures of selection and loci associated with fiber quality and yield traits. *Nat. Genet.* **49**: 1089-1098.
- Garg V, O Dudchenko, J Wang, AW Khan, S Gupta, P Kaur, *et al.* (2021) Chromosome-length genome assemblies of six legume species provide insights into genome organization, evolution, and agronomic traits for crop improvement. *J. Adv. Res.* doi:10.1016/j.jare.2021.10.009.
- Haupt M, Schmid K (2020) Combining focused identification of germplasm and core collection strategies to identify genebank accessions for central European soybean breeding. *Plant Cell Environ.* **43**: 1421-1436.
- Hübner S, N Bercovich, M Todesco, JR Mandel, J Odenheimer, E Ziegler, *et al.* (2019) Sunflower pan-genome analysis shows that hybridization altered gene content and disease resistance. *Nat. Plants* **5**: 54-62.
- Isleib TG, CC Holbrook, and DW Gorbet (2001) Use of plant introductions in peanut cultivar development. *Peanut Sci.* **28**: 96-113.
- Liang Z, S Duan, J Sheng, S Zhu, X Ni, J Shao, *et al.* (2019) Whole-genome resequencing of 472 *Vitis* accessions for grapevine diversity and demographic history analyses. *Nat. Commun.* **10**: 1190.
- Liu Y, H Du, P Li, Y Shen, H Peng, S Liu, *et al.* (2020) Pan-genome of wild and cultivated soybeans. *Cell* **182**: 162-176.
- Lv Q, W Li, Z Sun, N Ouyang, X Jing, Q He, *et al.* (2020) Resequencing of 1,143 *indica* rice accessions reveals important genetic variations and different heterosis patterns. *Nat. Commun.* **11**: 4778.
- Ma Z, S He, X Wang, J Sun, Y Zhang, G Zhang, *et al.* (2018) Resequencing a core collection of upland cotton identifies genomic variation and loci influencing fiber quality and yield. *Nat. Genet.* **50**: 803-813.
- Mace ES, S Tai, EK Gilding, Y Li, PJ Prentis, L Bian, *et al.* (2013) Whole-genome sequencing reveals untapped genetic potential in Africa's indigenous cereal crop sorghum. *Nat. Commun.* **4**: 2320.
- Milner SG, Jost M, Taketa S, Mazón ER, Himmelbach A, Oppermann M, *et al.* (2019) Genebank genomics highlights the diversity of a global barley collection. *Nat. Genet.* **51**: 319-326.
- Nguyen GN and SL Norton (2020) Genebank Phenomics: A Strategic approach to enhance value and utilization of crop germplasm. *Plants* (Basel) **9**: 817.
- Nguyen VD, R Sarić, T Burge, O Berkowitz, M Trtilek, J Whelan, *et al.* (2022) Noninvasive imaging technologies in plant phenotyping. *Trends Plant Sci.* **27**: 316-317.
- Ramírez-Villegas J, C Khoury, A Jarvis, DG Debouck, and L Guarino (2010) A gap analysis methodology for collecting crop genebanks: A case study with phaseolus beans. *PLoS One* **5**: e13497.
- Upadhyaya HD and R Ortiz (2001) A mini core subset for capturing diversity and promoting utilization of chickpea genetic resources in crop improvement. *Theor. Appl. Genet.* **102**: 1292-1298.
- Varshney RK, RK Saxena, HD Upadhyaya, AW Khan, Y Yu, C Kim, *et al.* (2017a) Whole-genome resequencing of 292 pigeonpea accessions identifies genomic regions associated with domestication and agronomic traits. *Nat. Genet.* **49**: 1082-1088.
- Varshney RK, C Shi, M Thudi, C Mariac, J Wallace, P Qi, *et al.* (2017b) Pearl millet genome sequence provides a resource to improve agronomic traits in arid environments. *Nat. Biotechnol.* **35**: 969-976.
- Varshney RK, M Thudi, M Roorkiwal, W He, HD Upadhyaya, W Yang, *et al.* (2019) Resequencing of 429 chickpea accessions from 45 countries provides insights into genome diversity, domestication and agronomic traits. *Nat. Genet.* **51**: 857-864.
- Varshney RK, M Roorkiwal, S Sun, P Bajaj, A Chitikineni, M Thudi, *et al.* (2021) A chickpea genetic variation map based on the sequencing of 3,366 genomes. *Nature* **599**: 622-627.
- Wang W, R Mauleon, Z Hu, D Chebotarov, S Tai, Z Wu, *et al.* (2018) Genomic variation in 3,010 diverse accessions of Asian cultivated rice. *Nature* **557**: 43-49.
- Wei T, R van Treuren, X Liu, Z Zhang, J Chen, Y Liu, *et al.* (2021) Whole-genome resequencing of 445 *Lactuca* accessions reveals the domestication history of cultivated lettuce. *Nat. Genet.* **53**: 752-760.
- Wu J, L Wang, J Fu, J Chen, S Wei, S Zhang, *et al.* (2020) Resequencing of 683 common bean genotypes identifies yield component trait associations across a north-south cline. *Nat. Genet.* **52**: 118-125.
- Xia E, W Tong, Y Hou, Y An, L Chen, Q Wu, *et al.* (2020) The reference genome of tea plant and resequencing of 81 diverse accessions provide insights into its genome evolution and adaptation. *Mol. Plant* **13**: 1013-1026.
- Xiao Q, X Bai, C Zhang, and Y He (2021) Advanced high-throughput plant phenotyping techniques for genome-wide association studies: A review. *J. Adv. Res.* **35**: 215-230.
- Yu X, Li X, Guo T, Zhu C, Wu Y, Mitchell SE, *et al.* (2016) Genomic prediction contributing to a promising global strategy to turbocharge gene banks. *Nat. Plants* **2**: 16150.
- Zhang K, M He, Y Fan, H Zhao, B Gao, K Yang, *et al.* (2021) Resequencing of global Tartary buckwheat accessions reveals multiple domestication events and key loci associated with agronomic traits. *Genome Biol.* **22**: 23.

Access to Plant Genetic Resources: Policy Vista

PL Gautam^{1*} and Sunil Archak²

¹Former Director (ICAR-NBPGR); Chairman (NBA and PPV&FRA); DDG (Crop Science, ICAR); Vice Chancellor (GBPUA&T)

²ICAR National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

Introduction

Plant genetic resources (PGR) is the term used to refer to any plant material *containing functional units of heredity of actual or potential value* (UNEP 1992). PGR include cultivated varieties (extant and obsolete), landraces, wild and weedy crop-related species and all other potential sources such as breeder's lines, populations and mutants. PGR are, thus, the raw material of crop improvement – traditional or molecular plant breeding or genome editing. There is global interdependence on PGR as no nation is self sufficient (Table 1). Hence, sharing/exchange of PGR is of paramount importance in securing sustainable food, fibre, fuel, nutrition, health, livelihood, environmental security, etc.

Genebanks are institutions conserving PGR *ex situ*, making them available for current use as well as for

posterity. In fact, genebanks carry out a host of operations including exploration and collecting, characterization and evaluation, import and quarantine, etc. Therefore, experts refer to genebanks as genetic resources centres. Such centres have a responsibility of helping users identify and obtain the most suitable material. Most genebanks provide web-based access to accession level and trait level information about the collections. Systematically organized information has dramatically improved the task of identifying desirable material and eventually potential use of PGR in research.

Prior to the implementation of CBD, the biodiversity was considered as common heritage of mankind and exchange of PGR was unrestricted and facilitated. Subsequently, however, accessing PGR for use has become increasingly challenging. Reasons include:

Table 1. Top providers and recipient countries based on germplasm shared from CG genebanks between 1985–2009 (from Galluzzi *et al.*, 2016).

Provider country	Total samples provided	Accessions provided	Recipient countries
India	188,911	48,635	144
Peru	67,899	16,216	158
Ethiopia	40,143	13,683	120
United States	36,652	6294	156
Iran	29,829	9779	87
Turkey	29,579	9634	83
Syria	26,029	7487	78
Sudan	24,262	3457	61
Philippines	21,626	4016	109
China	18,559	7225	125
Recipient country	Total samples received	Accessions received	Provider countries
India	284,454	115,849	181
United States	45,992	39,963	178
China	33,690	18,664	151
Ethiopia	28,863	17,572	150
Australia	20,218	17,566	150
Japan	17,628	12,022	141
United Kingdom	17,231	14,283	144
Morocco	16,362	14,618	97
Philippines	16,332	8798	107
Iran	13,083	12,301	135

*Author for Correspondence: Email-plgautam47@gmail.com

(i) increased awareness about the value of PGR; (ii) global agreements aimed at facilitating access and benefit sharing have ended up restricting access (e.g., Convention on Biological Diversity, CBD; International Treaty on Plant Genetic Resources for Food and Agriculture, ITPGRFA); (iii) increased compliance with the phytosanitary measures related to PGR movement (The Commission on Phytosanitary Measures, CPM; Sanitary and Phytosanitary Agreement, SPS); (iv) national legislations concerning import and use of genetically modified organisms; (v) increased footprint of private seed industry with IP protected cultivars. India responded with legislating and institutionalizing the PGR access and exchange process (Fig. 1). By the turn of the century, these regimes in combination have made it rather complicated to obtain and use the genetic resources.

New Technologies and New Restrictions?

Post-CBD era has witnessed fast and intense technological developments in plant biology and agricultural research

led by genomics. Accessibility to huge amount of DNA sequence data in the public domain, computational ability to store and mine big data, availability of genome editing technologies *inter alia* have made it practically possible to use PGR in a dematerialized manner. Consequently, international forums —including CBD and the Nagoya Protocol, ITPGRFA and the FAO Commission on Genetic Resources for Food and Agriculture, PIP Framework of WHO —are deliberating on *Digital Sequence Information* (DSI), its definition and access and benefit-sharing aspects of DSI. There could be serious implications on the access and benefit-sharing of PGR (Brink and van Hintum 2022).

Countries have option to consider DSI-PGR to be in scope of the Nagoya Protocol (bilateral access and benefit-sharing systems), either by mentioning the inclusion of DSI in their legislation. As a consequence, access to and use of DSI from this country may not be free anymore (in principle at least), as potential users are bound by the national laws of the country irrespective of global agreements. A study by CBD shows that

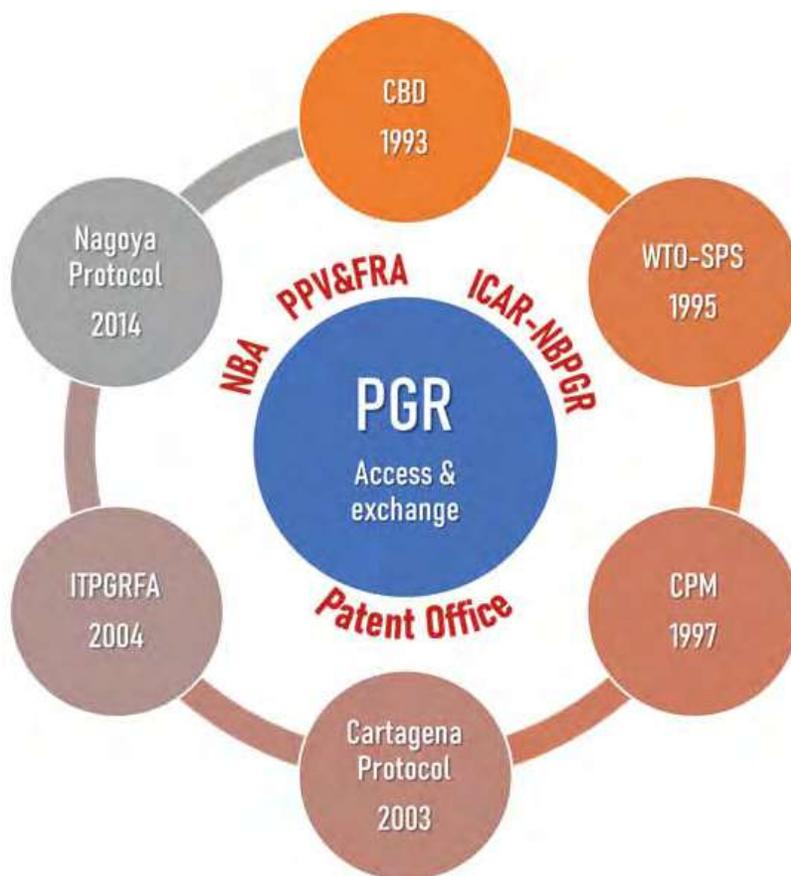


Fig. 1. International agreements and Indian institutions involved in access to PGR and their exchange

many countries are considering including DSI in their access and benefit-sharing legislation (https://www.cbd.int/abs/DSI-peer/Study4_domestic_measures.pdf). This might mean the need of country-labels for DSI on genetic resources uploaded in public databases to ensure compliance, a serious challenge for implementation. Add to that country-shopping for PIC and MAT. Alternative scenarios have also been proposed where access is free but benefits are shared with the specific provider countries of the genetic resources to which the DSI applies (including application of blockchain technology or commons licenses). Following a multilateral system can avoid tracking and tracing as well as country-shopping by decoupling access and benefit-sharing. Access continues from public databases and benefits are shared through a multilateral fund via say subscription systems. In contrast, countries argue that DSI should be kept out of the Nagoya Protocol or ITPGRFA jurisdiction by not including DSI in the definition of genetic resources. As a result, no specific monetary obligations are connected to the access to or utilization of DSI. Supporters of this option argue that free availability of information in itself is a form of benefit-sharing. “Open access” to valuable information from indigenous accessions is practically equivalent to “PGR being heritage of mankind” and against the premises of CBD – sovereign ownership.

The DSI conundrum reiterates the fact that national policy and legislation affecting plant genetic resources should support both international obligations and national objectives (Engels *et al.*, 2000). Countries have been unsuccessful to achieve a consensus on DSI either at CBD or at ITPGRFA for lack of clarity and practical examples. However, keeping India’s interests in mind and respecting the international obligations, authors recommend that India consider the following:

- i. Continue DSI negotiations under the aegis of ITPGRFA for crop genetic resources rather than opting for Nagoya option;
- ii. Decouple access and benefit-sharing; safeguard open access to researchers from public databases

and ensure that benefits are shared through Treaty multilateral fund via subscription systems.

India is at a critical juncture with respect to PGR policy options. Failing to comprehend the enormity of the state of affairs and forsaking proactive approach will be disastrous to the PGR stakeholders. Following activities may be prioritized:

Action Points

1. Harmonization of legislations and cross-talk between institutions regulating PGR access (e.g., National Biodiversity Authority, Protection of Plant Variety and Farmers’ Rights Authority, Indian Patent Office, National Bureau of Plant Genetic Resources (along with focal point in the Ministry of Agriculture) to minimize paperwork and expedite access (supply, import, statutory approvals).
2. Build a policy for digitalization of import permits, MTAs, release and feedback process; Provision of click wrap MTAs for germplasm supply.
3. Building bilateral and regional processes (with policy support) within a multilateral system for effective exchange of PGR of common interest.
4. Awareness generation among stakeholders as well as capacity building of PGR researchers in PGR policy issues and procedures including recent developments such as DSI.

References

- Brink M, van Hintum T (2022) Practical consequences of digital sequence information (DSI) definitions and access and benefit-sharing scenarios from a plant genebank’s perspective. *Plants People Planet*. **4**: 23–32. [10.1002/ppp3.10201](https://doi.org/10.1002/ppp3.10201)
- Galluzzi, G., Halewood, M., Noriega, I.L. et al. Twenty-five years of international exchanges of plant genetic resources facilitated by the CGIAR genebanks: a case study on global interdependence. *Biodivers Conserv* **25**, 1421–1446 (2016). <https://doi.org/10.1007/s10531-016-1109-7>
- Engels, J.M.M., Lyndsey Withers, Ruth Raymond and Hareya Fassil. 2000. Issues and options for national genetic resources programmes. Ten fact sheets on plant genetic resources. IPGRI, Rome, Italy. Pp. 10.

A Novel *Circa situm* Approach to Conserve Forest Genetic Resources of the Western Ghats

R Vasudeva

Department of Forest Biology, College of Forestry, University of Agricultural Sciences, Dharwad, Sirsi-581401, Karnataka, India

The Western Ghats of India is a rich repository of forest genetic resources in terms of wild flora, wild relatives of crop plants, edible fruits, medicinal plants and myriad agro-forestry systems. This paper discusses the importance and potentiality of novel approaches such as ‘Forest gene banks’ and ‘*circa situm* conservation’. It also emphasizes creation of range-wide, spatially explicit big-database on *circa situm* conservation value of agro-forestry systems as well as to develop guidelines for establishment of *circa situm* gene banks across the country.

Key Words: Big-data, Forest gene bank, Medicinal plants, On farm conservation

Western Ghats: Region with Outstanding Universal Value for Biological Diversity

India harbours four of the 34 hotspots of biological diversity of the world. The Western Ghats, a rolling ribbon of mountains spread along the Southern-Western part of India, is regarded as one of the world’s eight ‘hottest hotspots of biological diversity’. Geologically, it is older than the Himalayas. This mountain range contains unique flora and fauna and is declared as one of World’s Natural Heritage Sites by the UNESCO. The Western Ghats of India is also a rich repository of about 5000 species of flowering plants; over 200 tropical edible fruit species, 200 undomesticated vegetable species and also well-known for harbouring a rich variety of medicinal plants and wild relatives of crop plants. It is regarded as the “world’s best pharmacy” as it supports several coded systems of local health traditions. The agro-forestry systems of the Western Ghats have some of the most amazing diversity of tropical fruit trees (Vasudeva *et al.*, 2015).

Forest Genetic Resources of the Western Ghats and its Valuation

Forest Genetic Resource (FGR) is a component of the genetic diversity which is of actual or potential use either for production systems or for the ecosystem functioning in a forest. Conservation of FGRs is fundamental to the sustainable and productive management of the forest ecosystems. Genetic diversity occurs at various levels

of organization from the ecosystem, their sub-specific populations, and individual genotypes to the molecular level, of the gene. Hence, it is essential that all levels of genetic diversity be considered in the utilization and conservation prioritization process.

Although the economic value of the products derived from forest genetic resources (e.g., NTFPs, genes for the development of new crops and bio-molecules as a source for the development of new drugs) is difficult to assess, their value has been estimated to touch many billions of dollars annually (Vasudeva, 2016). One of the earliest rough estimates of global value of the myriad uses of genetic resources was at US \$ 500 to 800 billion (Kate and Laird, 1999). The volume of global medicinal plants market has been estimated at US\$ 60 billion per year, growing at a rate of 7% annually. Global demand for herbal products, mostly derived from forest ecosystems in recent years has experienced a quantum jump in volume of plant material traded within and outside the countries of origin. Further, globally, NTFPs generate social benefits and play an important role to improve livelihoods of local communities which are involved in their harvesting, processing and trade.

Further, the forest ecosystems contain many potentially useful new resources. Genetic resources derived from forest ecosystems represent a rapidly growing and highly promising source of new drugs, agricultural products, and other fruits of biotechnology. For instance, ants found in forests may provide novel

*Author for Correspondence: Email-vasudevar@uasd.in; vasukoppa@gmail.com

antibiotics that are important for human medicine, silk from spiders may provide the lightest but high tensile fibre which may be stronger weight-for-weight than steel, *etc* (Kate and Laird, 1999). Services provided at free of cost by forest ecosystems around the globe are benefiting humankind. Examples include oxygen production, climate control by forests, nutrient cycling, water purification, natural pest control, and pollination of crop plants. In 1997, these services were valued at US\$ 33 trillion per year, almost double the US\$18 trillion annual global national product.

Converting Biodiversity into the Biological Resources: Prerequisite for Utilization

Today biological diversity of a country is regarded as its “resource capital”. Understanding spatial distribution of this plant resource is a prime pre-requisite for its conservation and sustainable utilization. Unfortunately, primary data on quantitative estimates of the resources, their geographical spread and the levels of regeneration in their natural habitats did not exist even for an important eco-region such as the Western Ghats till recently.

Further, the impact of unscrupulous extraction of some of these resources is also not very well understood. Earlier work on understanding the occurrence and distribution of plant resources are scanty, isolated and never done adopting a uniform sampling strategy throughout the Western Ghats at a fine geographic scale. Hence there was a critical need to map the plant resources at local, regional and national levels throughout the country adopting uniform sampling strategy and develop a database.

In this context, mapping of these resources reflecting the spatial distribution would greatly help in understanding their structure and dynamics in natural habitats, allow assigning conservation values of different sites/recognizing hotspots of plant diversity within an eco-region, and eventually in formulating strategies for sustainable utilization of plant resources (Ganeshiaiah, *et al.*, 2012). Most importantly this would also offer a perspective on the economic value of forest resources in the Western Ghats.

Realizing this importance, National Bio-resource Development Board (NBDB), of the Department of Biotechnology, New Delhi commissioned a national initiative which broadly aimed at: a) quantitatively assessing the geographic distribution and population status of the plant resources of the Western Ghats,

b) identifying the threats to these plant resources and c) setting up a Western Ghats eco-region specific database of plant resources (Ganeshiaiah, *et al.*, 2012).

The programme also aimed at developing thematic maps of the density and distribution for all the economically important species along the Western Ghats as well as analyzing the spatial and temporal patterns of change in specific plant resources along the Western Ghats. A survey of 3132 grids, each of 6.25 km x 6.25 km geographical area along the Western Ghats, undertaken in the project period perhaps represents one of finest-scale vegetation surveys of any hotspot of biological diversity in the world. This is the largest primary data ever assembled on the Western Ghats vegetation in the Indian history which can be accessed at <https://betaibin.iirs.gov.in>. A total of 3132 kilometer-transects of grueling field work spreading across six states has been undertaken. The overall gamma diversity for the entire Western Ghats eco-region as computed by Shannon’s diversity was at 5.31 (Ganeshiaiah, *et al.*, 2012). However, there is a critical need to map and generate such fine scale spatially explicit big-database on the agro-forestry resources of the country.

Novel Approaches Conserve Forest Genetic Resources

Central to the genetic resource utilization and conservation is the maintenance of sufficient inter and intra-specific genetic variability. However it is a challenge in those species that are subjected to intense harvesting pressures. There have been number of isolated approaches proposed to address the utilization and conservation of genetic resources. Unfortunately, most approaches are aimed at conserving the “resources” *per se* and have seldom taken into account the genetic diversity. *Ex-situ* and *in-situ* methods followed in the conservation of crop genetic resources have been extended to forest genetic resource conservation without critically evaluating their appropriateness.

New approaches are therefore needed to integrate national conservation activities for maximum effect in both production forests and fully protected area systems. Complementary Conservation Methods (CCM) are increasingly being viewed as newer approach to utilize and conserve genetic resources, which is essentially a decision-making process to adopt an appropriate method, protocol to achieve a lasting conservation. In CCM, *in situ* conservation of genetic resources are reinforced with

the efforts of *ex situ* and vice versa while an enabling policy environment is created. Newer approaches such as 'Forest Gene Banks' and '*circa situm* conservation' of resources is far more participatory in nature and effective in involving communities for domestication and utilization.

Forest Gene Bank

Forest Gene Bank (FGB) is an alternative strategy to the existing conventional *ex situ* and *in situ* methods of conservation. FGB is essentially an *in situ* site where a target species occur naturally that has been augmented and genetically upgraded through introductions of individuals from other populations. Essentially this technique enhances the genetic diversity through interventions and thus enables the population to survive for a longer time periods (Uma Shaanker and Ganeshiah, 1997). This approach takes into account the spatial distribution of the species and the underlying genetic variability of a species.

In this model, the forest gene banks would be located within the distributional space of the species. These *in situ* sites would serve as sink sites and would have the propagules introduced from the neighboring source sites which harbor unique and rare alleles. Further, this model would also take into account the phenotypic variations existing within each of the species and bring them to the sink sites. Thus, these sites would maintain a regional set of both genotypic as well as phenotypic variations. These sites would, serve as gene banks for facilitating continuous turnover of the genetic material within and among populations (Uma Shaanker and Ganeshiah, 1997). Forest gene banks for two endangered tree species viz., *Dysoxylum malabaricum* and *Semecarpus kathalekanensis* have been established near Sirsi in the Central Western Ghats.

Circa Situm Conservation

Circa situm conservation is the preservation of planted and/or remnant trees and wildlings in farmland where natural forest or woodland containing the same trees was once found, but where natural vegetation has been lost or modified significantly through agricultural expansion (Dawson *et al.*, 2013). *Circa situm* conservation emphasizes on maintaining higher tree species diversity to conserve the processes of evolution and adaptation (Jarvis, 1999). Forest remnants in cleared agricultural landscapes, sacred groves and coffee plantations may be

extremely important as tree breeding stocks and are the places for *circa situm* conservation (Boshier *et al.* 2004).

Circa situm conservation has a very important role in agricultural areas with high demographic pressure and significant deforestation where alternative approaches to conservation are limited and traditional agroforestry systems like coffee/areca agro-forests, home gardens and sacred groves are known to harbour high levels of plant diversity (Maxted *et al.*, 1997). Motivation to adopt *circa situm* conservation by farmers is largely based on the perceived use value of the species. Maheswarappa *et al.* (2021) have shown small farmers of Kodagu region of Karnataka tend to conserve highest number of useful tree species and 95 per cent of them retain native tree species than big and medium category farmers.

In a study by Maheswarappa *et al.* (2019), the extent of genetic diversity of tree populations of *Acrocarpus fraxinifolius* occurring in natural forests, sacred groves and coffee plantations of Kodagu in Southern Karnataka was analyzed using ISSR markers. Interestingly, populations of sacred grove and coffee plantations recorded higher diversity than natural forest population. This pattern was also similar in two other tree species. The authors have also shown that the traditional agroforests host substantial diversity of useful trees suggesting a huge potential to be considered as *circa situm* sites. Understanding the pattern of tree diversity, species conservation concern, genetic diversity of *Circa situm* areas of different agroforestry areas is poorly attempted in India (Maheswarappa *et al.* 2020).

Apart from recognizing potential sites for *circa situm* conservation, augmenting these sites with additional diversity to convert them as gene banks can be attempted. In an attempt to establish the first *circa situm* gene bank of trees in the country, Maheswarappa *et al.* (2021) have planted *Ficus racemosa*, *Ficus reliogiosa*, *Pongamia pinnata*, *Toona ciliata*, *Pavetta indica*, *Artocarpus hirsutus*, *Oroxylum indicum*, *Kydia calysina*, *Erythrina indica*, *Artocarpus heterophyllus*, *Mangifera indica*, *Terminalia tomentosa*, *Dalbergia latifolia*, *Acrocarpus fraxinifolius* and *Ficus racemosa* in a coffee farm near Ponnampet, Kodagu, Karnataka, India.

New tools, technologies, protocols are required to rapidly and precisely characterize/monitor diversity, assess the dynamics/loss across landscapes and as well as to achieve lasting conservation. New technologies offer unprecedented abilities to monitor change, create new

ways of collecting data. Information and virtual tools, space technologies, and genomic data are some of the newer and rapidly evolving technologies. Ubiquitous devices such as cell phones and people connected to cloud computing systems will revolutionize the types of data we collect for effective utilization and conservation.

Action Research Points

There is a need for the establishment of a key technical secretariat that involves practicing foresters, scientists, technocrats, information scientists, policy makers, *etc.* to build a range-wide, spatially explicit big-database on *circa situm* conservation value of agro-forestry systems as well as to develop guidelines for establishment of *circa situm* gene banks across the country.

References

- Boshier David H, Gordon, James E and Barrance, Adrian J (2004) Prospects for *circa situm* tree conservation in meso-american dry-forest agro-ecosystems". In: Biodiversity Conservation in Costa Rica: Learning the Lessons in a Seasonal Dry Forest, edited by Gordon W. Frankie, Alfonso Mata and S. Bradleigh Vinson, Berkeley: University of California Press, 2004, pp. 210-226.
- Dawson IK, Guariguata MR, Loo J, Weber JC, Lengkeek A, Bush D, Cornelius J, Guarino L, Kindt R, Orwa C, Russell J, Jamnadass J (2013) What is the relevance of smallholders' agroforestry systems for conserving tropical tree species and genetic diversity in *Circa situm*, in situ and ex situ settings? A review. *Biodivers Conserv* **22**: 301-324.
- Ganeshaiyah KN, Ganesan R, Vasudeva R, Kushalappa CG, Menon APP, Patawardhan A, Yadav SR, Uma Shaanker R (2012) Plants of Western Ghats, Vol 1 and 2, Published by Department of Biotechnology, GOI, New Delhi.
- Jarvis D (1999) Strengthening the scientific basis of *in situ* conservation of agricultural biodiversity on farm. *Bot. Lith Suppl* **2**: 79-90.
- Maheswarappa V, R Vasudeva, Ramakrishna Hegde, Ravikanth G and Pownita KV (2019) ISSR Analysis of genetic diversity in *Acrocarpus fraxinifolius* from three landscape elements of transition forest belt of Kodagu district, Karnataka, India. *Int. J. Curr. Microbiol. App. Sci.* **8**: 1611- 1624. doi: <https://doi.org/10.20546/ijemas.2019.809.1184>
- Maheswarappa V, R Vasudeva, Ramakrishna Hegde and Ravikanth G (2020) Genetic diversity analysis of *Ficus racemosa* using ISSR markers from three landscape elements of dry deciduous forest belts in Kodagu, Karnataka, India. *Inter. J. of Genet.* **12**: 768-772.
- Maheswarappa V, R Vasudeva Ramakrishna Hegde (2021) *Circa situm* conservation of coffee agroforests: farmer's perception in Kodagu landscape of Karnataka, India. *Tropical Ecology* **63**, pages 104-112. <https://doi.org/10.1007/s42965-021-00141-w>
- Maxted N, Ford-Lloyd BV, Hawkes JG (1997) Contemporary conservation strategies. In: Maxted N, Ford-Lloyd BV, Hawkes JG (eds) Plant genetic conservation: an in-situ approach. Chapman and Hall, London, pp 20-55.
- Kerry ten Kate and Sarah A Laird (1999) The Commercial Use of Biodiversity - Access to Genetic Resources and Benefit Sharing. Routledge Library Edition. EarthScan Publications, London.
- Uma Shaanker R and Ganeshaiyah KN (1997) Mapping genetic diversity of *Phyllanthus emblica*: Forest gene banks as a new approach for in situ conservation of genetic resources. *Current Science* **73**: 163-168.
- Vasudeva R, V Bhat, GV Nayak, B Sthapit and V Ramanatha Rao (2015a). Patterns of diversity and drivers of on-farm/ in situ conservation of native mango varieties in the Central Western Ghats, India. *Acta Hort.* 1101. ISHS. DOI 10.17660/ActaHortic.2015.1101.21
- Vasudeva R, Bhuwon Sthapit, I Salma, Suchitra Changtragoon, Idha W Arsanti, D Gerten, Nataya Dum-ampai, S Rajan, MR Dinesh, IP Singh, Sanjay Kumar Singh, BMC Reddy, VA Parthasarathy and V Ramanatha Rao (2015b). Use Values and Cultural Importance of Major Tropical Fruit Trees: An Analysis from 24 Village Sites Across South and South-East Asia Indian *J. Plant Genet. Resour.* **28(1)**: 17-30.
- Vasudeva R (2016) Developing framework for economic valuation of forest genetic resources and bio-resources under the access and benefit sharing regime. In: Kumar VP (ed.) "Economic Valuation of Forest Genetic Resources and Bio-Resources under the Access and Benefit Sharing" Published by the Karnataka Biodiversity Board. pp 11-28.

Empowering Farmers with Blockchain-based Transparency Throughout the Value Chain

Gaurav Somwanshi*, Pratap Deshmukh, Ganesh Anantwar, Manish Verma, Areeb Mohammad Khan, Aarti Tiwari, Danish Siraj, Gunvant Sarpate, Mandar Darade, Vikas Taklikar, Netan Mangal, Syed Burair, Renuka Paturkar, Gajendra Sahu and Rohit Dhivare

CEO & Cofounder, Emer Tech Innovations (AgroTrust: Blockchain platform), Mumbai-400076, Maharashtra, India

Majority of the Indian farmers growing diverse local crops and traditional cultivars are small small-landholding farmer. Lower yields and challenges to find a value chain are accentuated by the effects of climate change. Since 2003, a new provision has been made available to farmers to come together and form ‘Farmer Producer Organization’ and ‘Farmer Producer Company’, in which farmers themselves are shareholders and external entities aren’t allowed to become investors. Currently, there are 7,000 such registered groups in India, and it’s being hailed as a promising step towards farmer liberation. Many farmer collectives, especially Farmer Producer Companies, are eagerly adopting emerging technologies for their operations, scaling, value-chain integration, and marketing purposes. This paper will seek to explore a case study of India’s largest farmer collective of fruits and vegetables regarding their usage of Blockchain technology in bringing transparency and efficiency across their integrated value chain.

Key Words: Blockchain, Farmer producer company, Farmer producer organisation, Traceability, AgroTrust

Introduction

Income earned by a small and marginal landholding farmer in India depends on multiple extraneous factors thereby making farming a financially perilous profession. Lack of infrastructure, less bargaining power, lack of storage facilities and marketing resources have been the prime challenges for the small landholding Indian farmer. Thus, economy of scale is becoming important for basic sustainability which can be enabled with collectivisation, more so for those intending to grow traditional specialty crops/cultivars. At the same time, modern consumers are also becoming health conscious and socially aware regarding farming practices and ensuring farmer equity.

But lack of transparency leads to food fraud which costs the global food industry an estimated \$30-40 billion annually (PricewaterhouseCoopers 2016). Consumers want to know more about journey (harvest-to-shelf), freshness, authenticity, integrity and ingredients of the food they consume (Allied Market Research 2019). On the other hand, the small-landholding Indian farmer has always been operating in a domain where lack of transparency dictates nearly every facet of life, right from isolation from market trends till exploitation by

middlemen. The initiative by Farmer Producer Companies to form integrated value chains to ensure that fair equity is ensured among the value chain partners is critical for betterment of the entire rural economy.

This paper will explore a case study of how Blockchain technology is being leveraged by one such Farmer Producer Company, Sahyadri Farmers Producer Company Limited, to bring transparency throughout the value chain from farmer till the consumer. The research questions to be answered in the paper are: (1) How can collectivization of farmers ensure better equity to the small landholding Indian farmer? (2) How can Blockchain technology be leveraged by farmer groups to bring transparency in the value chain?

Problems Faced by a Small-landholding Indian Farmer

Lack of accessibility to the benefits of mechanization, modern irrigation, and other investment-based technological improvements leading to suboptimal productivity. A perpetual debt cycle, post-harvest losses, exploitation by middlemen, increasing input costs, fluctuating supply resulting in price volatility, are some of

*Author for Correspondence: Email-gauravsomwanshi@gmail.com

the major problems faced by the Indian farmer. To help ease this situation, Government of India gives guidelines to buy 23 crops from farmers at a Minimum Support Price (MSP). But as per Shanta Kumar Committee Report 2015, only 5.8% of Indian farmers sell their produce to Government agencies at MSP (Food Corporation of India, Government of India (2015, January). However, benefits of MSP are not country-wide and is limited to some states and for limited crops (Tiwari 2020). In order to overcome these challenges, farmers are now coming together to form a collective, to aggregate their produce, invest in common infrastructure, minimize exposure to market fluctuations, reduce risks, and increase their bargaining power.

Farmer Producer Company/Farmer Producer Organization (FPC/ FPO)

As explained by Ministry of Agriculture, Government of India, in the ‘Policy and Process Guidelines for Farmer Producer Organizations’, ‘Collectivization of producers, especially small and marginal farmers, into producer organizations has emerged as one of the most effective pathways to address the many challenges of agriculture but most importantly, improved access to investments, technology and inputs and markets. Department of Agriculture and Cooperation, Ministry of Agriculture, Govt. of India has identified farmer producer organization registered under the special provisions of the Companies Act, 1956 as the most appropriate institutional form around which to mobilize farmers and build their capacity to collectively leverage their production and marketing strength’ Ministry of Agriculture, Government of India. (2013). A Farmer Producer Organization (FPO) or a Farmer Producer Company (FPC) enables every farmer to have equal voting rights regardless of their shareholding, thereby bringing democratization. Only farmers can become shareholders within an FPO or FPC, although an FPO or FPC can invest and hold equity in other organizations (Ministry of Corporate Affairs, Government of India, 2013). As per the latest information, there are more than 7000 registered Farmer Producer Companies in India, and the government is making policies and plans to boost their number.

Blockchain can Mitigate Challenges Faced by Farmer Collectives

Despite a growing number of farmer collectives being registered as Farmer Producer Companies, not all of

them are able to achieve sustainability. Primary reasons are (i) Losses due to huge wastage (>40% post-harvest food loss) and (ii) Lack of transparency (inadequate professional management, risk of frauds). Traceability (ability to follow the movement of a feed or food through specified stages of production, processing and distribution) allows farmers to get maximum leverage of the value unlocked at the retail end. There are documented cases where the small landholding Indian farmers haven’t even received any remuneration for selling their produce as the middlemen unduly charged them with transport and rent to subtract from the prices. On the other side, the consumer has no way to discern how much actually ends up reaching the farmer. In such a scenario, a radical outlook towards maximum transparency can help both the farmers and consumers and help strengthen the fragmented value chain.

Blockchain to Enable Traceability and Transparency

In order to establish trust throughout the value chain from the small landholding farmer till the end consumer, it becomes prudent to achieve this through a Blockchain-based platform, as described in Fig. 1. The very properties offered by Blockchain ensures both trust over the data being trustworthy, or at least acts as a deterrent for any future misleading data entry owing to its immutability. The critical decision over here, is rather, non-technical; which parts of the value chain is one being transparent about? We explore this in the case study of Sahyadri Farmers Producer Company Limited.

Sahyadri Farmers Producer Company Limited (Sahyadri Farms) and Sahyadri Farms Sustainable Grassroots Initiative (SFSGIL-RU brand) Case Study

Sahyadri Farms was registered as a Farmer Producer Company (FPC) in 2010 under the provision introduced in Companies Act, 1956, Government of India. It’s a collective that is owned and managed by farmers. It is India’s largest Farmer Producer Company and has highest share of grape exports from India compared to any other. Majority of the farmers within the collective are small landholding farmers. Sahyadri Farms partnered with Emertech Innovations Pvt Ltd to onboard the value-chain allied activities on a Blockchain platform named AgroTrust, as described in Fig. 2. The project for Fruits & Vegetables traceability began in April 2019, and lasted

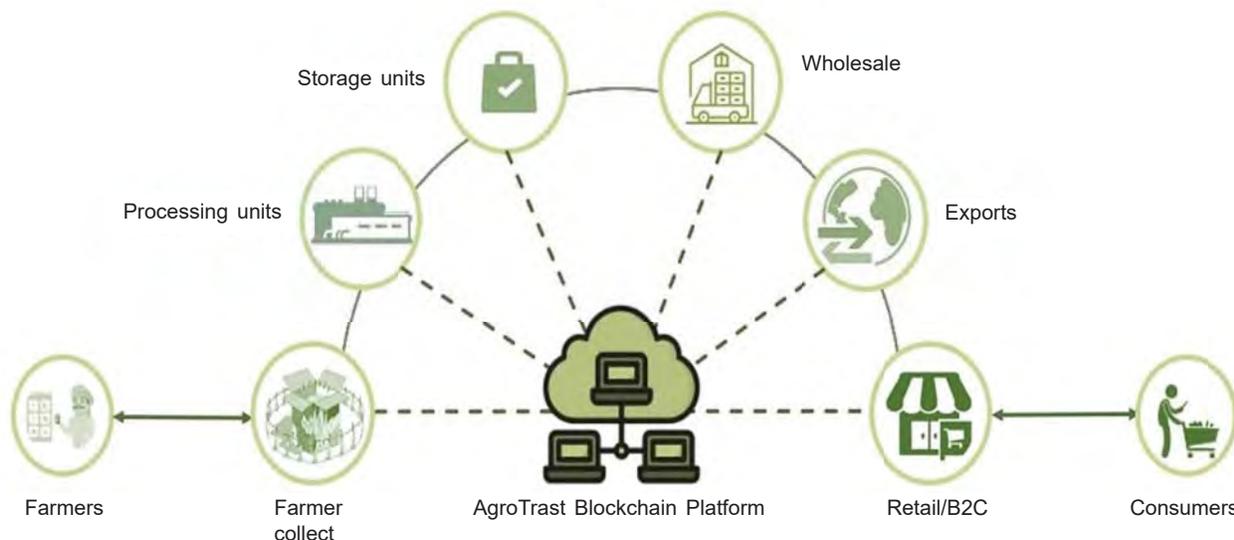


Fig. 1. Blockchain-based platform of a farmer collective

until June, 2022. A total of 5.2 million Blockchain-backed QR codes were printed during the project duration. The project for cotton-to-cloth initiative, under SFPCL’s allied initiative of Sahyadri Farms Sustainable Grassroots Initiative Limited (SFSGIL) with the commercial brand name of ‘RU’, began its live operations in February, 2022 and is ongoing (at the time of submission of this paper), with over 35,000 Blockchain-backed QR codes being printed on T-shirts.

Process Flow

For implementing this solution for fruits and vegetables for SFPCL, four attributes of every unit of produce

needed to be transparently recorded at stock keeping unit (SKU) level across multiple operational locations: origin, pricing, provenance, quality & safety through certificates. Each location captured this information during operational processes and stored it onto the blockchain. Real life movement of produce necessitated a four-location design. The value chain began at cultivated plots called origins where the produce was harvested. Information about origins was recorded onto a blockchain verified registry of farmers and their fields. Harvested produce was purchased by Sahyadri Farms and transported to a production and packaging location that performed quality assessment. Critical information

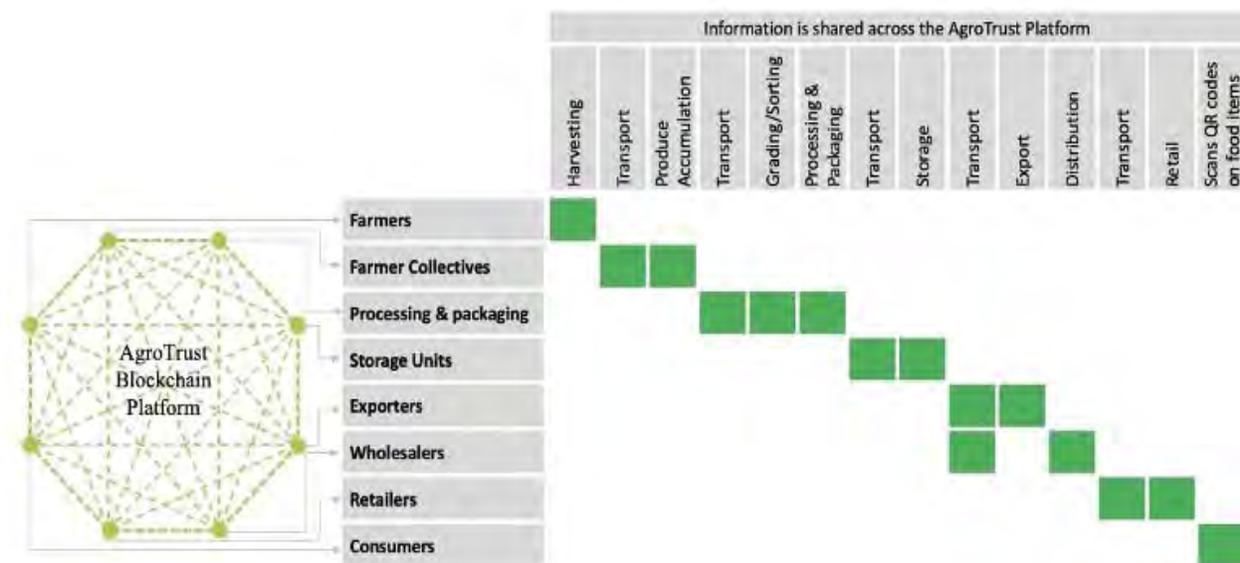


Fig. 2. Agro-Trust blockchain-based platform

about the price given to the farmers and transport timestamp was recorded onto the blockchain. Accepted produce quantities were then moved to assembly lines for production into freshly packaged SKUs (stock keeping unit). At this stage, information about the production costs and timestamp were calculated as per the weight of each SKU and recorded onto the blockchain. Finished SKUs were then dispatched to multiple distribution centers that serviced either physical retail stores or direct delivery to consumers in designated geographical zones. Transport timestamps and storage costs incurred by distribution centers and retail stores were calculated as per the weight of each SKU and recorded onto the blockchain. Due to the perishable nature of the products being tracked, it was extremely important that data was captured in real time or near real time, validated as per predefined business rules and committed to blockchain.

For the ongoing project (at the time of submission of this paper) with SFSGIL, there are integrations with 'Farm Setu', SAP, and solution by Vesatogo initiative which are used to bring attributes to traceability on T-shirts. The project seeks to incrementally add more information to the QR codes with subsequent integrations and development of new modules. AgroTrust blockchain transparency solution is a consortium blockchain network based on the principle of 'One Crop, One Blockchain'. It is implemented using the Multichain protocol which is a built by taking of Bitcoin client and enhanced for permissioned blockchains. Consensus mechanism used in this protocol is a randomized validation based on distributed agreement between permissioned block validators similar to PBFT. Multichain allows fine grained permissions control at address or node level and implements streams which are on-chain key-value data stores with timestamps and publisher identity. Streams are used to record and retrieve immutable data onto a blockchain.

Consumer Transparency Service and impact

Upon scanning the QR code of the item, customer can see - Know Your Farmer, Know Your Money, Know Your Journey and Know Your Food, along with Sahyadri Farms brand logo and 'Why Blockchain?' content for making consumers aware about the technology. A cropped sample of a real QR code scan of an SKU is provided in Fig. 3., in which one can see the amount received by the farmer with respect to how much the end consumer is paying for the final product. Owing to

the nature of the data availability and data capture, the QR codes could show varying degrees of information. Transparency regarding how much money goes to the farmer defined per SKU is a first of its kind initiative in India. Consumer awareness and consciousness is now being channelized by offering them visibility of what happens in the journey of the food that they consume. The rising awareness, along with maintenance of food quality and safety standards, leads to more consumers for the farmer collective which in turn enables the collective to offer better prices to the farmers. As of 28th June, 2022, more than 5.2 million QR codes have been printed with a range of 5000 to 24,000 QR codes depending upon supply and consumer demand of fruits and vegetables. For SFSGIL-RU brand, over 35,000 T-shirts are equipped with a Blockchain-backed QR code. The successful implementation has been covered by the mainstream media for Blockchain-based traceability and transparency (Wadke 2020).

Action Points for On-farm Conservation of Genetic Resources

1. The campaign of bringing small landholding and marginal farmers under Farmer Producer Companies/ Farmer Producer Organizations is a promising step



Fig. 3. Agro-Trust blockchain-based platform

towards ensuring a life of dignity and sustainability.

2. The adoption of technologies in agricultural practices is also proving to be a boon. Among these, the use of Blockchain in providing radical traceability and transparency throughout the value chain can help highlight the crucial aspects of the value chain.
3. It is the domain experts, who make Blockchain a relevant and saucerful solution. Therefore, pilot projects must be launched as a collaborative effort between farmer community-NGOs-genebank

Acknowledgments

We thank the leaders of the Farmer Producer Company, Sahyadri Farmers Producer Company Limited, primarily Mr Vilas Shinde who has pioneered the project of uplifting fate of small landholding farmers. Mr Abhijeet Kale, CEO of SFSGIL, also has been pioneering in this regard. We also received crucial support and guidance

from Society for Innovation and Entrepreneurship (SINE), IIT Bombay that enabled the implementation of this project.

References

- PricewaterhouseCoopers (2016, October) *Fighting \$40bn food fraud to protect food supply*. PricewaterhouseCoopers Malaysia. <https://www.pwc.com/my/en/press/160127-fighting-40bn-food-fraud-to-protect-food-supply.html> (accessed November 01, 2020)
- Allied Market Research (2019, May) *Food Traceability Market Outlook - 2025*. Shankar Bhandalkar/Debojyoti Das. <https://www.alliedmarketresearch.com/food-traceability-tracking-market> (accessed November 05, 2020)
- Ministry of Agriculture, Government of India (2013) *Policy and Process Guidelines for Farmer Producer Organisations*. https://mofpi.nic.in/sites/default/files/fpo_policy_process_guidelines_1_april_2013.pdf (accessed November 03, 2020)
- Tiwari S (2020, December 9) *MSP and Public Procurement*. PRSIndia. <https://www.prsindia.org/theprsblog/msp-and-public-procurement> (accessed December 15, 2020)

Agroecology-based Biodiversity Management

KS Varaprasad^{1*} and V Sandhya Kumari²

¹Revitalising Rainfed Area Network, Hyderabad; Independent Consultant (APAARI and IGGAARL)

²Junior Research Coordinator, RySS, Guntur, Andhra Pradesh-522034, India

The Green Revolution helped India to avoid famine and destruction and ensured food security in most developing countries. Currently, India is not only self-sufficient in food production but also has foodgrain buffer stocks. This agricultural growth revolved around harnessing genetic potential of major food crops developing via high input responsive improved varieties. In the long run, these agricultural technologies, that contributed to food security of an ever-growing population, led to soil degradation, water and air pollution and loss of biodiversity. In order to tackle these negative consequences, agroecology-based agriculture is catching up globally for sustainable development. The agroecology principles are relevant to organic farming, regenerative agriculture, conservation agriculture, nature-based agriculture and several other traditional agri-food systems. Common principle among these traditional agri-food systems is protecting the natural resources including vulnerable bioresources. Using rich crop and landrace/farmer varietal diversity that are nutrient dense, pest resistant and resilient to climate aberrations is a sustainable approach. It is in this context, the need for identification and enhanced use of genetic resources that demand minimal or no chemical inputs yet possess the potential to meet the growing food demand.

Introduction

The extreme and indiscriminate adoption of the modern agricultural technologies for higher crop yields has shown its baneful effect over the years. Consequences are deterioration of soil fertility (75% of the earth's land is degraded losing 36 billion tons of soil annually). The Food and Agricultural Organisation indicated that due to soil degradation, only 60 harvesting years are left and this is a major concern for the growing population (FAO, 2016). Fertiliser consumption has increased from 12.4 kg/ha in 1969 to 133.4 kg/ha in 2020. The imbalanced use resulted in a declining fertiliser crop response ratio from 123.47 kg grain/kg in 1970 to 3.70 kg grain/kg in 2005. During 2020-21, the Government of India spent Rs 127,921 crore on fertiliser consumption and the nation consumes pesticides worth Rs 6,000 crores. The extreme consumption of agrochemicals has led to contamination of water, soil and air, thereby reducing crop productivity (Smith *et al.*, 2013). Conventional chemical farming increased the cost of cultivation and led Indian farmers to debt trap. Climate change has further aggravated the situation and yield losses have led to 1.5% loss in GDP on annual basis. Do we have alternatives to arrest plant biodiversity loss? Can we revive natural systems through enhanced use of crops and landraces diversity?

Challenges

Modern agricultural practices have promoted monocropping in only five major crops compared to fifty crops commonly cultivated in traditional agricultural systems about eight decades ago. Traditional cultivars and landraces have largely replaced with improved varieties and hybrids. Thus, several wild relatives of crops, neglected and underutilized crop species are now conserved only in ex-situ genebanks and have almost disappeared from the cultivation. India is one of the eight important *Vavilovian* centers of origin and crop diversity. India has 8% of the total global biodiversity with an estimated 49,000 species of plants of which 4900 are endemic (Anil *et al.*, 2014). In addition to several sacred groups maintained by indigenous communities, institutional efforts have led to establishment of 514 wildlife sanctuaries and 102 national parks including 18 biosphere results covering about 5% of the total geographical area. India has institutionalized agrobiodiversity management with five Bureaus under ICAR with the mandate of collection, conservation, evaluation and documentation. In spite of conscious institutionalized efforts to conserve, there has been severe biodiversity loss at ecosystem level and at farm level.

*Author for Correspondence: Email-prasadksv@gmail.com

Components of Agroecology and the importance of Biodiversity

Agroecology is the holistic integration of concepts and principles of ecology, science and technology, practices, economics, political, and social processes for the sustainable management of agriculture and food from production to consumption (FAO, 2022). It provides a strategy to increase the diversified agro-ecosystem, i.e., by promoting the effectiveness of plant, animal biodiversity, and nutrient recycling through the use of natural resources systems, like, legumes, trees, livestock, etc. and thus making the food and societal system sustainable (Tripathi et al., 2015). This system can support the huge production in quantity and quality of diverse food, fabric, and medical crops both for family consumption and market, as this reduces the risk of nutrition as a whole. Agro-ecosystem on which our food and agriculture production system depends is provided by biodiversity or in general it is biodiversity-based agriculture, addressing the current crisis. These two are central for food security and climate resilience and guide us in managing and mitigating health and climate shocks and crises.

An experiment conducted in all districts of Andhra Pradesh to see whether the birds returned to the natural farming adopted landscapes revealed a substantial difference in bird visits between farms that use agroecological techniques and those that use chemical farming methods. Natural farming practices increased the micro-climate for birds with lower temperatures compared to outside temperatures and a variety of bird species lay eggs and stay for longer periods in natural farming fields (Hussain et al., 2022). Shift from intensive farming to adopting agroecology or natural farming can increase the soil organic carbon pool, enhance soil quality, and increase soil resilience to adjust to the extreme climate effects. Similarly, recycling of nutrients, forage yield, pollination, resistance to weed infestation, etc. are some of the ecosystem services and ecological functions in agroecosystems (Dumont et al., 2013).

Traditional agricultural systems and promoting landraces

Several localized traditional agricultural systems have evolved over millennia of years suited to the soils, weather and local food and fodder needs. These systems were eco-friendly and sustainable. An innovative collaborative

effort is being made by Watershed Support Services and Activities Network (WASSAN), an NGO, to document and experiment to understand the principles behind these systems (Table 1). Traditional agricultural systems that have the potential to be recognized as Heritage systems in India are documented with details of locations of practice and their unique features (Singh and Rana, 2019).

Table 1. Localized traditional agricultural systems documented and experimented by WASSAN (<https://www.wassan.org>)

Hangadi Kheti, Barediyo, or Hath-hangdo	Tribal junction of the state of Gujarat, Rajasthan, and Madhya-Pradesh
Akkadi Saalu/Saalu Bele/ Mishra Bele	Karnataka
Olya, Sat-Gajra	Madhya Pradesh
Nau Anja	Himachal Pradesh
Pata	Maharashtra
Kurwa	Jarkhand
Rammol	Gujarat
Panamkuthu	Kerala
Misa Chassa	Odisha

Lakhs of indigenous varieties have been permanently lost (John and Babu 2021). Diversification of agriculture refers to the shift from the regional dominance of one crop/varieties to regional production of a number of crops/varieties. It aims to improve soil health and a dynamic equilibrium of the agroecosystem. Crop diversification takes into account the economic returns from different value-added crops. Diversified farms are usually more economically and ecologically resilient.

These technologies have continued to exist only in rainfed areas. Nutrient dense landraces evolved over thousands of years in the rainfed ecosystem with local relevance to cultural and organoleptic needs of the communities. They have distinct advantage of resilience to climate change effects. Neglected and underutilized species and minor crops remained out of the formal seed system. Hence there is a need to develop an alternative seed system to meet the requirements of rainfed areas. Such an alternative seed system should aim to enhance crop and varietal diversity; promote landraces that are adopted to local soil types and multiple cropping systems; access to seed in times of contingencies; reasonable price margin between seed and grain; intrinsic conservation of crop diversity in the ecosystem and timely access to quality indigenous seed by encouraging local Farmer Procedure Organizations (FPOs). Operationalization of

such an alternative seed system will bring the following benefits to rainfed farmers.

1. Resilient diversified cropping systems; availability of quality seeds
2. Enhanced farmer income [e.g. Navdanya crop system in Anantapur, the most drought prone district, increased the farmer income by about rupees 15000 per acre]
3. Increased productivity [e.g. Ragi local (*Pedda Chodi*) productivity improved from 4 quintals to 8 quintals per acre in Andhra Pradesh and Odisha; paddy landrace Kalachampa promoted by Department of Agriculture Odisha yielded more than local checks].
4. Enhanced market share with premium price [landraces viz. black rice, brown rice, short grain scented rice, etc.]

Enhanced household food and nutritional security [due to enhanced access to pulses, oilseeds and millets owing to multiple and mixed cropping systems]

New Paradigm for Indian Agriculture

The Government of India has come up with an approach for Indian agriculture to shift from agroindustry to agroecology farming. Five main areas through which agroecology can point a new way for agriculture are 1. Reduced reliance on pesticides, 2. Enriching biodiversity, 3. Revitalising small farms, 4. Creating alternative animal production systems and 5. Enhancing urban agriculture (Neelam Patel *et al.*, 2022). It is also important modify and revise varietal release criteria with:

- Focus on evaluation in eco-geographic regions/districts from where the landrace cultivated/collected/originated
- Farmers' yields based on crop cutting experiment documented by the State Department of Agriculture considered
- Nutrition, local preference, cropping system and resilience to climate and biotic stresses value considered
- A comprehensive on ground pilot study encompassing all components of alternate seed system is needed.

Conclusion and Action Points

There is an urgent need to shift from the current chemical based agri-food system to a system that aims to provide

a *one-health ecosystem to plants, animals and humans*. In a large chunk of landrace diversity is still with rural and tribal communities. These communities are holding this diversity and conserving them on-farm through use for reasons of taste, religious use, food preference and other organoleptic properties.

1. Participation of communities and general public in conservation through use and exchange of material/knowledge with the genebanks/seed banks needs to be facilitated through appropriate policies in place.
2. Granting rights to group or individual claims on farmer varieties by the Plant Variety Authority is illogical and all the farmer varieties shall remain as nation's property. There is a need to revise policy and define process of establishing sovereign rights of India linking contributing communities.
3. Changing genebank's approach on accession-oriented collecting, evaluation and conservation to respecting communities' wisdom precipitated through several generations.
4. Benefit sharing implementation needs to be strengthened; the logic of pre-CBD collections not attracting benefit sharing is illogical.
5. Substantial increase in investments by governments on community seed banks facilitating access to quality seeds of farmers' varieties that are locally produced and locally consumed.
6. National Agricultural Research System to invest more time and resources on mainstreaming farmer varieties particularly in rain-fed areas.
7. Science based examination of traditional systems that encourage use of local biodiversity for sustainability, ecological stability and one-health.

References

- Dumont B, Fortun-Lamothe L, Jouven M, Thomas M, and Tichit M (2013) Prospects from agroecology and industrial ecology for animal production in the 21st century. *animal*, **7(6)**: 1028-1043.
- FAO (2016). The State of Food and Agriculture. <https://www.fao.org/3/i6030e/i6030e.pdf>
- FAO (2022). Retrieved from <https://www.fao.org/agroecology/overview/en/>
- Hussain Z, Boppana B, Anisetti H, Soma R and Gangisetty S (2022) Do Birds Return to Agricultural Landscapes through Adoption of Natural Farming Practices? A Comparison of Natural Farming vs. Chemical Farming in Andhra Pradesh. *Agric. Sci*, **13(3)**: 358-377.

- John, DA., & Babu, GR (2021). Lessons from the after maths of the green revolution on the food system and health. *Frontiers in sustainable food systems*, 5, 644559.
- MNV Anil,* Kanchan Kumari1 and S R Wate1 (2014) Loss of Biodiversity and Conservation Strategies: An Outlook of Indian Scenario. *Asian J. Conserv. Biol.*, Vol. 3 No. 2, pp. 105-114.
- Neelam Patel, Bruno Dorin and Ranveer Nagaich (2022) A New Paradigm for Indian Agriculture from Agroindustry to Agroecology. NITI Aayog.
- Singh,AK and Rana,RS (2019). Nationally important Agricultural Heritage Systems in India: Need for Characterisation and Scientific Validation. *Proc Indian Natn Sci Acad*, **85(1)**: 229-246.
- Smith, P, & Gregory, PJ (2013). Climate change and sustainable food production. *Proceedings of the Nutrition Society*, **72(1)**: 21-28.
- Tripathi N, Singh RK, Pal D & Singh RS (2015) Agroecology and sustainability of agriculture in India: An overview. *EC Agriculture*, **2(1)**: 241-248.

New Paradigms in Agricultural Education in India

RC Agrawal

Deputy Director General (Agricultural Education), Indian Council of Agricultural Research (ICAR), Krishi Anusandhan Bhavan II, Pusa Campus, New Delhi-110012, India

The New Education Policy-2020 (NEP-2020) of India provisioned that “the design of agricultural education will have to be strengthened towards developing professionals with the ability to understand and use local knowledge, traditional knowledge and emerging technologies, while being cognizant of critical issues of declining profitability and/or productivity but enhanced economic aspirations of farmers, climate change, food sufficiency, etc.”. Based on the principles and philosophy of NEP-2020, a roadmap and Implementation Strategy for NEP-2020 in Agricultural Education System has been prepared and released on 28 September, 2021 during the Annual Vice-Chancellors Conference in New Delhi.

Introduction

The New Education Policy-2020 (NEP-2020) of India provides an opportunity and has opened the gateway to introduce various changes in the education system, including higher agriculture education. Some of the major highlights of NEP 2020 include enhancing the gross enrollment ratio (GER), defining minimum standards of quality of agricultural education and ensuring their adherence by all stakeholders, improvement in research contributions, importance of staying relevant and providing placement along with right skills. Starting with multiple exit and entry points into higher education, relaxation of the residential requirements of UG, PG and PhD programmes, restructuring and reformulation of the UG curriculum in accordance with the new system advised by NEP, compliance with Academic Bank of Credits as per the directives of the Ministry of Education, Deemed universities of ICAR may initiate process for transforming them into Multidisciplinary Education and Research University (MERU).

Accordingly, the Indian Council of Agricultural Research (ICAR) constituted a national level Committee of Vice-Chancellors to develop an implementation strategy to comply with various provisions of the NEP-2020. The committee was asked to assess and recommend requirements of higher agricultural education in the present set of education/research/extension and suggestion for consideration of agricultural education in the categories of Medical and Legal Education under NEP-2020. In this direction, several meetings

and consultations were held to deliberate with various stakeholders. The draft report was also circulated to all the Vice-Chancellors and Chief Secretaries for their input and comments. Various timelines for implementation of NEP by AUs were also defined by the committee.

The preparation of professionals in agriculture and veterinary sciences under NEP-2020 is through programmes integrated with general education. Most

NEP-2020 and Higher Education in Agriculture –Salient Targets

- By 2022-23, common entrance test may be conducted by ICAR for admission of the students in all the AUs, and AUs to start increasing seats on annual basis by 10% until the target is achieved.
- By 2025-2030 all institutions, located in the same premises, offering either professional or general education may aim to organically evolve into multi-disciplinary institutions/ clusters offering higher education both seamlessly, and in an integrated manner.
- By 2035, achieving 50% Gross Enrolment Ratio (GER) in higher agricultural education including vocational education.
- ▮ All higher education institutions (HEIs) should aim to become multidisciplinary institutions by 2040.

*Author for Correspondence: Email-ddg.edu@icar.gov.in

importantly, NEP-2020 has indicated that “*the design of agricultural education will have to be strengthened towards developing professionals*” with the ability to understand and use local knowledge, traditional knowledge and emerging technologies, while being cognizant of critical issues of declining profitability and/or productivity but enhanced economic aspirations of farmers, climate change, food sufficiency etc. Based on the principles and philosophy of NEP-2020, a roadmap and implementation strategy for NEP-2020 in Agricultural Education System has been prepared and released on 28 September, 2021 during the Annual Vice-Chancellors Conference in New Delhi. The major recommendations of the committee are as given below.

Restructuring of Agricultural Universities under NEP-2020

As per the main thrust of NEP-2020, it has been recommended to end the fragmentation of higher agricultural education by transforming Agricultural Universities/Colleges into large multidisciplinary universities, colleges, and Higher Education Institute (HEI) clusters/Knowledge Hubs with 3,000 or more students. The canvas of multi-disciplinarity of agricultural education shall encompass academic programmes of basic sciences, social sciences and allied disciplines of agricultural sciences. Therefore, the single stream universities under the ICAR-AU system need to move towards multidisciplinary institutions by 2030 while continuing the focus on agriculture.

Several affiliated colleges exist in large numbers in both public and private domain. As per NEP-2020, they need to be brought under the new norms of higher education. In compliance with the proposed policy, collective steps have been proposed to work in coordination with the universities to eliminate “affiliation” by 2035.

By taking the advantage of the available expertise and resources, the necessary steps will be taken to translate ICAR Deemed Universities into multi-disciplinary research-intensive universities.

For each university it is necessary that it works on preparing a blue print of its future called, Institutional Development Plan (IDP) and use it to seek/attract funding from different sources as well for planned growth and development.

Academic Restructuring of Agriculture Education

It has been proposed to revamp academic program structure with an innovative system of multiple entry and exits with options to award certificate, diploma, UG degree general, or degree research, and one or two years of Master’s degree. The residential requirements of UG, and PG programmes will be relaxed so that the students wishing to exit/enter may be able to do so irrespective of any time limit.

A Deans’ committee may be constituted by ICAR for restructuring UG curricula in compliance with the provisions of NEP-2020 (based on the recommendation, ICAR has already constituted the 6th Deans committee). Based on the demand, the universities may enhance the intake of UG so that the exits of few students with Certificate/Diploma do not hamper the numbers of degree pass-out students. Further, the agricultural universities may be given time to make this restructured four year UG programme functional by 2025. A separate curriculum and admission criteria may be devised for the admissions to one year certificate and two year diploma in Agriculture.

In PG programmes, it is proposed to follow multidisciplinary approach with an option to choose major and minors as per the choice of the student. Teaching assistantship shall be encouraged to PhD students to gain the required experience and also to address the shortage of faculty in many institutions/universities.

Policy directives have been enlisted to enhance Gross Enrolment Ratio (GER) in agricultural universities with at least increase of 10% seats from 2021-22 academic session on annual basis; the scores of the common entrance test conducted by ICAR through National Testing Agency (NTA) for admission of the students in all the AUs for UG/PG/PhD may be used by AUs for the admissions. Entrance Examination for UG may be conducted in regional languages and compliance with Academic Bank of Credits (ABC) as per the directives of the Ministry of Education.

Role of ICAR in Regulation of Agricultural Education

As per the recommendations of NEP-2020, the ICAR shall act as a Professional Standard Setting Body (PSSB) for Agricultural Education to develop the curricula and lay

down academic standards for the agricultural education. As a member of the proposed General Education Council (GEC), the ICAR shall take necessary steps through PSSB for making a uniform academic structure across the country both in public and private institutions offering academic programmes in agricultural sciences.

As member of the GEC, the ICAR will be able to contribute in the regulation of agricultural education through single regulatory body to be enacted as a National Higher Education Regulatory Council (NHERC), the first vertical of the proposed Higher Education Commission of India.

As per NEP-2020, the National Accreditation Council (NAC) shall act as a meta Accrediting body and shall assign the task of functioning as recognized accreditor to appropriate number of institutions.

The task to function as a recognized accreditor shall be awarded to an appropriate number of institutions by NAC. In this perspective, the NAEAB of ICAR may be recognized as an Accreditor for the accreditation of Universities/Colleges offering agricultural education.

To achieve the goal of 'internationalization at home' by maintaining global quality standards and attracting greater numbers of international students, an International Students Office shall be created to coordinate all matters relating to welcoming and supporting students arriving from abroad.

Research/teaching collaborations and faculty/student exchanges with high-quality foreign institutions will be facilitated, and relevant mutually beneficial MOUs with foreign countries will be signed.

With the available alternative modes of quality education, we need to complement/enrich traditional and in-person modes of education.

The necessary steps should be taken to avail the existing e-learning platforms such as SWAYAM, DIKSHA, SWAYAMPURABHA, etc and also to develop e-courses in agriculture and allied sciences.

Attracting Talent in Higher Agricultural Education

In order to attract and retain the talent in higher agricultural education and achieve educational excellence, ICAR provides financial assistance through various

scholarships/ fellowships to the admitted students. In addition, a national level examination was started ICAR-All India Entrance Examination for Admissions (AIEEA), in the year 1996-97, to address the challenges of quality manpower development and meet the requirement of much needed talented pool of human resource in the field of agriculture and allied sciences. The primary objective of conducting the exam is curtailing academic inbreeding in agricultural education and promoting national integration by providing a window of opportunity for the students to take admissions outside their domicile states in reputed institutions having state-of-the-art infrastructure and facilities, infuse merit, encourage talent and promote uniform examination standards across the agricultural universities, thereby leading to an overall improvement in the quality of Higher Agricultural Education.

The SAUs annually provide 15% (UG) and 25% (PG and PhD) of their existing seats as ICAR quota seats for filling up through this entrance examination. However, ICAR fills 100% PG and PhD seats at ICAR-DUs, CAUs. To align with NEP-2020 and reduce the burden on students for appearing in multiple entrance examination for admissions in State Agricultural Universities (SAUs), ICAR has taken an initiative and offered the SAUs to utilize NTA scores for UG, PG and PhD admissions. During academic session 2021-22, 05 SAUs for UG, 07 for PG and 08 for PhD admissions utilized NTA scores for state quota seat admissions.

National Talent Scholarship (NTS): The NTS UG and NTS PG were initiated to discourage inbreeding to pursue undergraduate and post-graduate studies to all those candidates who are admitted through ICAR-AIEEA and take admission outside their state of domicile and maintain prescribed academic standards.

PG Scholarship: Based on their merit in AIEEA (PG), the ICAR-PG Scholarship, is awarded to 600 candidates per year for pursuing Masters' degree for a period of two years who get admission in AUs located in states other than those from where they had graduated.

Junior/Senior Research Fellowship (JRF/SRF): 300 students are awarded this scholarship every year for pursuing Ph.D. degree program in ICAR-AU System subject to the condition that they meet the other prescribed eligibility requirements.

Networks and Partnerships are Tools for Efficient PGR Management

RK Tyagi

Asia-Pacific Consortium on Agricultural Biotechnology and Bioresources (APCoAB), Asia-Pacific Association of Agricultural Research Institutions (APAARI), Bangkok 10100, Thailand

Agriculture is facing unprecedented challenges from a rapidly expanding human population and an unstable and changing cultivation environment. With the global system facing a multitude of crises across the social, health, environmental, and peace and security spectrum, there is an urgent need for scaling up international, regional cooperation and partnerships to find lasting solutions to ensure food and nutritional security. Plant genetic resources (PGRs) can be used for breeding improved crop varieties, providing food and nutrition for ever-increasing population of animals and human beings. An attempt is made briefly to discuss about the importance of network and partnership and key role, feature and challenges for sustaining the network and partnership on long-term basis for management of PGRs.

Introduction

Agriculture is facing unprecedented challenges from a rapidly expanding human population and an unstable and changing cultivation environment. To increase food production sustainably in the face of these challenges multiple tools ranging from the application of novel technologies in breeding and farming to the broadening of the genetic base are needed. The plant genetic resources (PGRs) include wild relatives of cultivated species, varieties and hybrids, as well as breeding material. PGRs can be used for breeding improved crop varieties, providing food and nutrition for ever-increasing population of animals and human beings. It is well known that agriculture affects natural biological resources, but it also uses these resources to obtain varieties and hybrids, the reciprocal relationship leading to increased economic benefits and sustainability.

Sustainable Development Goals and International Partnership

The 2030 Agenda for Sustainable Development, adopted by all United Nations Member States in 2015, provides a shared blueprint for peace and prosperity for people and the planet, now and into the future. At its heart there are the 17 Sustainable Development Goals (SDGs), which are an urgent call for action by all countries – developed and developing – in a global partnership (<https://sdgs.un.org/goals>). It is clearly evident that biodiversity plays a significant role in achieving several SDGs, particularly SDG # 2 (Zero

Hunger), SDG # 3 (Good Health and Well-being), SDG # 12 (Responsible Production and Consumption, SDG # 13 (Climate Action) and SDG # 15 (Life on Land). SDG # 17 (Partnership for the Goals) deals with strengthening the means of implementation and revitalize the global partnership for sustainable development (<https://sdgs.un.org/goals>) to: (i) enhance international support for implementing effective and targeted capacity-building in developing countries to support national plans to implement all the sustainable development goals, including through North-South, South-South triangular cooperation; (ii) enhance the global and partnership for sustainable development, complemented by multi-stakeholder partnerships that mobilize and share knowledge, expertise, technology and financial resources and (iii) encourage and promote effective public, public-private and civil society partnerships, building on the experience and resourcing strategies of partnerships. However, according to SDG Progress Report (2021), the interconnected global economy requires a global response to ensure that all countries, developing countries in particular, can address compounding and parallel health, economic and environmental crises and recover better. Strengthening multilateralism and global partnerships is more important than ever as about 63% of low-income and lower-middle-income countries are need of data and information sharing and additional funds particularly to face challenges posed by the pandemic (<https://unstats.un.org/sdgs/report/2021/The-Sustainable-Development-Goals-Report-2021>).

*Author for Correspondence: Email-tyaginbpgr@gmail.com

pdf). With the global system facing a multitude of crises across the social, health, environmental and peace and security spectrum, there is an urgent need for scaled up international, regional cooperation and partnerships to find lasting solutions to ensure food and nutritional security.

Networks and Partnerships

Although individuals or organizations have been getting together to collaborate and to share information and resources for a very long time. However, agricultural research networks have been proliferating in recent past, and these networks are seen as mechanisms for more efficient and cost-effective exchange of resources, information and research results. As for as PGR management is concerned, the Second Global Plan of Action for Plant Genetic Resources for Food and Agriculture (PGRFA), adopted by the FAO Council in November 2011, is the main reference document for national, regional and global efforts to conserve and use PGRFA.

Key Features

Networks are commonly used to increase the effectiveness of international agricultural research by involving a number of individuals, organizations or countries in sharing information and other resources at regional and global level. However, success of any PGR network or partnership depend on some key features as discussed below:

1. All partners should identify the clear objectives of common interest for sustainability of the partnership.
2. All partners need to ensure mutual equality and equity in a network.
3. Role/responsibility of each partner needs to be defined clearly and exchange/sharing of data, information or genetic material must take place for vibrancy of the network.
4. Each associated partner should have capacity to utilize the information and resources to maintain continued interest to fulfill common objectives.
5. A coordinating committee comprised of network members needs to be established for developing a work plan and periodically monitoring the progress.
6. Depending upon the progress of results/outputs, the objectives may be evolved. Therefore, there must be flexibility to make modifications and changes in

pathway in the interest of each partner.

7. Sharing of intellectual property rights be fair and equal, as due to any partner.
8. Trust-building and ethical values to be respected by each partner in a network for long-term sustainability.

Key Roles

The role of PGR networks is stressed by almost all international agreements, including the Convention on Biological Diversity (CBD) and the International Treaty on Plant Genetic Resources for Food and Agriculture, to fulfill the respective objectives under obligations of a mutual agreement. A network is generally interdependent on each other partner(s) to develop the synergies and respect their mutual requirement related to exchange and management PGR and related information. At the same time networks not only facilitate the exchange of PGR, but also provide a platform for scientific discussion, sharing responsibilities and information, technology transfer and research collaboration (Maggioni and Engels, 2014) .

PGR management requires functional convergence of global policy and regulatory frameworks that deal with PGR to contribute significantly to food and nutritional security. Regional initiatives need to bring neighboring countries together to address the common goals in PGR management. Regional networks can play significant role for management of PGR, as discussed below [adopted from Tyagi (2019)]:

1. *Prioritization of Research*

As discussed, role of networks is also to provide platform for scientific discussion for various stakeholders engaged in PGR management by organizing High Level Policy Dialogues, Expert Consultations, Workshops, Seminars, Symposia and Brainstorming Sessions to priorities the research agenda at national/regional level.

2. *Capacity Development*

Capacity development is an important area to conserve and utilize the PGR. The major issues are to identify the areas of capacity development and the stakeholders. Therefore, mapping the existing capacity and need of capacity development is pre-requisite to build the capacity not only for technical but soft skills also. Capacity also needs to be developed to create robust impact pathways, prepare logical frameworks, measure indicators, from research to extension, policy, and livelihoods impact on farmers' livelihoods. Functional capacity development is

also required to build awareness among policy makers, media and the general public.

3. *Public Awareness*

Network can play significant role in creating public awareness by sharing of experiences, and best practices, indigenous knowledge, success stories across various communities, regions and countries.

4. *Policy Advocacy*

By engaging concerned PGR stakeholders in High Level Policy Dialogues and Expert Consultations, enabling and supportive policy environment may be created by developing policy legal frameworks including policy briefs, development of the value chain and exploration of markets at local, national and international levels.

5. *New Partnership Development*

Networks are instrumental to facilitate increased cooperation at regional and international levels to develop synergies by building new partnerships and networking of groups and institutions including non-governmental organization, community-based organizations and civil society organizations for implementing the programmes efficiently.

6. *Mobilization of Investment*

Success and sustainability of any network will depend on continuous availability of adequate financial resources. Donors from private sector play an important role to mobilize the funds for conservation and utilization of PGR at national and regional level. Networks may be instrumental to develop mechanisms for partnerships by greater involvement of private sector, philanthropists and international donors by brokering of partnerships.

Key Challenges

Limited trained human, financial and physical resources are major challenges for establishing the PGR network. A pre-requisite of a strong network is that the individual partners are strong. In addition, there are other challenges are also faced for long-term sustainability of the network as mentioned below:

1. *Low Level of Coordination of Activities*

Despite the best efforts and enthusiasm for establishing the regional and global networks for PGR management, it has been observed that there was still a low level of coordination activities on long-term basis. The expectation is that breeders and other users will be

able to have easy access to well-characterized and well-maintained genetic resources among the associate member institutions. Therefore, established networks should consistently and actively grow and sustain for long-term for mutual benefits of the associated partners.

2. *Technical Impediments for Sharing Information and Data (Halewood, 2018)*

- (i) Data are fragmented and dispersed across organizations and international borders and are not managed following the FAIR principles of Findability, Accessibility, Interoperability and Reusability (Wilkinson *et al.*, 2016);
- (ii) Inadequate systems exist for logging and tracking PGR as well as metadata related to PGR, for example, there is no universally agreed-upon system for permanently and uniquely identifying PGR (*e.g.* publications do not provide traceable, permanent unique identifiers for PGR); and,
- (iii) Radically different approaches to data management and sharing within and across public and private sectors due to fundamentally different objectives and low levels of mutual trust.

3. *Political and Institutional Impediments*

Until the late 1960s, PGRs were generally treated as 'global public goods'. In the decades that followed, after Convention on Biological Diversity 1993 (CBD; <https://www.cbd.int/>) technologically advanced countries pushed for the international recognition of intellectual property protection for living materials through various agreements, treaties, protocol. In light of this recent history, it is perhaps not surprising that some country and regional representatives, civil society and farmers' organizations have voiced concerns that technological breakthroughs in genomic breeding, gene editing, and gene synthesis will widen the technology gap, and concomitant economic disparities, between the developed and developing countries.

4. *Trust-deficit in Global South and Global North*

There is a worry that the major gap of capacity of understanding and use of advanced knowledge and new technologies (expertise), infrastructure (research for development facilities) and enabling policies dealing with new legal instruments between developed and developing countries for utilization of PGR, will exacerbate tensions associated with the unrealized expectations of monetary benefits accruing from access and benefit

sharing laws. These stakeholders note that at present the requisite technological capacities principally reside in elite research institutions in the global North. They are skeptical that these new capacities will be used to develop technologies targeted at resource-poor farmers working in vulnerable agricultural systems. This has led to demands (by some civil society organizations and developing countries) that research organizations stop providing unregulated open access to genetic sequence data until benefit sharing issues can be addressed (Hammond, 2016; The International Civil Society Working Group on Synthetic Biology, 2016).

5. Continuity of Financial Resources

Generally, developed countries are well-placed in terms of expertise, infrastructure and availability of funds but poor in occurrence or availability of genetic resources. At the same time converse is true in case of developing or least-developed countries who are rich in availability of PGRs. This situation likely to warrant the availability of adequate finance resources to carry out the mutually accorded activities in a network, if the terms and references are not made transparent. Even otherwise, it is observed that there is less investment is made by private sector for the basic research for utilization of PGRs by the breeders. For sustainability of the network and partnership, assurance of availability of financial resources are very important be engaging public and private partners on mutually agreed, equal and transparent terms of references to achieve the defines objectives.

Conclusions

Networking and partnerships are potentially effective tools for sharing and exchanging of information, germplasm resources, conservation techniques and tool through which institutional and capacity building can be developed and strengthened. It is through networking arrangements that PGR activities or programmes can be properly facilitated, coordinated and implemented. This is important because, with limited resources, no individual institute can really afford to carry out all the operations. Joining hands and sharing resources is the way into the future for the effective conservation and efficient use of PGR. As the network develops and becomes sustainable, the responsibility of running the network may be more formalized through a regional organization. A successful and viable network needs a coordinator with networking

experience as well as its technical and political expertise, which is well positioned to offer its contribution to this process and to actually assume more responsibilities for a co-ordinated involvement of the all associated institutions with mutual trust and respect. If efforts are not made to enhance trust and inter-stakeholder cooperation between developing and developed countries to establish and sustained new networks at South-South and North-South levels, such situations might ultimately undermine the development of an open-science culture, slowing the rate of the scientific advancement in PGR management and consequently crop improvement for food and nutritional security and climate challenges.

References

<https://sdgs.un.org/goals>

<https://www.cbd.int/>

- Halewood M, T Chiurugwi, RS Hamilton, B Kurtz, E Marden, E Welch, F Michiels, J Mozafari, M Sabran, N Patron, P Kersey, R Bastow, S Dorius, S Dias, S McCouch and W Powell (2018) Plant genetic resources for food and agriculture: opportunities and challenges emerging from the science and information technology revolution. *New Phytologist* **217**: 1407–1419 doi: 10.1111/nph.14993
- Hammond E (2016) Digital genebankers plan to ignore UN request on the impact of genomics and synthetic biology on access and benefit sharing: a preliminary report. Penang, Malaysia: Third World Network.
- The International Civil Society Working Group on Synthetic Biology (2016) Synthetic biology and the CBD: five key decisions for COP 13 & COP-MOP 8. [WWW document] URL https://www.boell.de/sites/default/files/2016-11-icswgsb_synbio_brief_cop13_.pdf
- Tyagi RK (2019) Dryland Agrobiodiversity for Adaptation to Climate Change: Role of Regional Organizations. In: Agrawal A, S Rajkumar, JC Rana, RK Tyagi, OP Yadav, K Singh and RS Paroda (eds) Proceedings and Recommendations of the Satellite Symposium on ‘Dryland Agrobiodiversity for Adaptation to Climate Change’. Indian Society of Plant Genetic Resources, New Delhi, Bioversity International, New Delhi, Asia-Pacific Association of Agricultural Research Institutes, Bangkok, held at Jodhpur, Rajasthan, India, February 13, 2019, pp 39-42.
- Wilkinson MD, M Dumontier, IJJ Aalbersberg, G Appleton, M Axton, A Baak, N Blomberg, J-W Boiten, LB da Silva Santos, PE Bourne et al. (2016) The FAIR Guiding Principles for scientific data management and stewardship. *Scientific Data* **3**: 160018.
- United Nations (2021) The Sustainable Development Goals Report 2021. 64p. <https://unstats.un.org/sdgs/report/2021/The-Sustainable-Development-Goals-Report-2021.pdf>

Contributions of Dr RS Paroda to the Field of Genetic Resource Management

Anuradha Agrawal

Indian Council of Agricultural Research, Krishi Anusandhan Bhavan 2, Pusa Campus, New Delhi-110012, India

Dr RS Paroda has been a champion for the cause of genetic resources (GR) management for over five decades. His passion for the subject has led to massive reforms in infrastructure, capacity building and projects/programs in the arena of agrobiodiversity management and use, which has a direct bearing in increasing agricultural productivity leading to food and nutritional security. The Indian Society for Plant Genetic Resources (ISPGR) established in 1987, was his brainchild and as the Founder (1987-88) as well as current (2022-24) President of the society, he has immensely contributed in its overall growth and visibility. This paper gives a brief overview on the contributions of Dr RS Paroda in the area of GR management.

Key Words: Genebank, ISPGR, National Bureau, Regional and Global Networks

Introduction

Genetic resources are the building blocks for global food, nutrition and environmental security. Accordingly, their efficient management such as exploration, evaluation, exchange and conservation through use is fundamental for the survival of humanity and for posterity. Recognising this and being an ardent supporter, Dr Rajendra Singh Paroda has made extra-ordinary contributions in the field of genetic resource (GR) management at the national, regional and global level, as elaborated below.

GR Contributions at National Level

As Director, National Bureau of Plant Genetic Resources (NBPGR) from 1985-1987, Dr Paroda took major initiative to build one of the most modern Indian Genebank at New Delhi, seeking then the funding support of US \$ 25 million from the United States Agency for International Development (USAID). The Genebank was inaugurated by the then Hon'ble Vice-President of India, during the II Crop Science Congress organised by Dr Paroda in 1996. This Genebank, the second largest in the world, currently houses more than 460, 000 valuable accessions of different crops.

Considering the importance of capacity building in the field of genetic resources, he not only got all 100 scientists of NBPGR trained in the USA, but was also instrumental in initiating MSc (PGR) course in 1997 and PhD (PGR) from 2004 onwards at the Post-Graduate School of Indian Agricultural Research Institute (IARI), New Delhi.

For promotion of PGR science and realising the importance of publishing scientific literature, Dr Paroda established the Indian Society of Plant Genetic Resources (ISPGR) in 1986 which got registered in 1987 and has made significant contributions, including holding of a few national and international conferences. These include: i) National Symposium on Plant Genetic Resources held in New Delhi in 1987; ii) National Symposium on Conservation of Agrobiodiversity held in 1997; iii) Role of Science and Society towards Plant Genetic Resources Management – Emerging Issues, held in 2005, New Delhi; iv) National Symposium on Recent Global Developments in the Management of Plant Genetic Resources held at New Delhi, 2009 v) Brainstorming Meeting on Strategies for Implementation of Delhi Declaration for Agrobiodiversity Management in New Delhi in 2017; vi) National Webinar on Implementation of Access to Plant Genetic Resources and Benefit Sharing (ABS) in 2020. The proceedings of these events have served effectively the purpose of knowledge dissemination among PGR scientists.

Convinced of the importance of genetic resources, Dr Paroda as Director General, Indian Council of Agricultural Research (from 1994-2001) created four new National Bureaux on: (i) Animals, (ii) Fish, (iii) Micro-organisms, and (iv) Insects. He also launched a National Action Plan on PGR management in 1999, which spurred collections (almost doubled in five years) and evaluation of genetic resources through inter-institutional collaboration at the national level.

*Author for Correspondence: *Email-Anuradha.Agrawal@icar.gov.in



Dr RS Paroda, President, ISPGR (2022-24) along with dignitaries, invitees, awardees and ISPGR members at Award Function held on August 20, 2022

Under his visionary leadership, two important national laws relating to genetic resources were drafted, which were enacted by the Parliament in record time. These are: i) Protection of Plant Varieties and Farmers' Rights Act (2001), and ii) Biological Diversity Act (2002). He also served from 2011-2014 as the Chairman, National Advisory Board on Management of Genetic Resources and ensured effective coordination and collective action by all the Bureaux.

Recognising his significant contributions in the field of genetic resource management and agricultural development, Dr Paroda was awarded the most prestigious national civil honour 'Padma Bhushan' by the Hon'ble President of India in 1998. He also received the famous 'Dr Norman Borlaug Award' from President of India in 2006 and 'Dr MS Swaminathan Award' in 2020. Recognising his enormous support for genetic resource programs, ICRISAT honoured him by naming its Genebank as 'Rajendra S. Paroda Genebank' in 2005.

GR Contributions at Regional Level

At the regional level, while working as Regional Plant Production and Protection Officer, FAO, Bangkok (1992-1994), and as Executive Secretary, Asia Pacific Association of Agricultural Research Institutions (APAARI) for more than 22 years, Dr Paroda could

initiate a number of regional PGR networks for evaluation and use of genetic resources in different countries. These were on:

- i. Tropical Asian Maize Network (TAMNET) involving International Maize and Wheat Improvement Center (CIMMYT), Mexico;
- ii. Cereals and Legumes Asia Network (CLAN) involving the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), India, International Centre for Agricultural Research in the Dry Areas (ICARDA), Syria and World Vegetable Center (AVRDC), Taiwan.
- iii. Council for Partnership on Rice Research in Asia (CORRA) involving International Rice Research Institute (IRRI), Philippines;
- iv. Inter-regional Network on Cotton in Asia and North Africa (INCANA) involving Association of Agricultural Research Institutions in the Near East & North Africa (AARINENA), APAARI, Central Asia and the Caucasus Association of Agricultural Research Institutions (CACAARI), Agricultural Research, Education and Extension Organization (AREO) Iran and ICARDA.
- v. Inter-Regional Network on Cotton in Asia and North Africa (INCANA) involving Association of

Agricultural Research Institutions in the Near East and North Africa (AARINENA), APAARI and CACAARI;

- vi. Asia-Pacific Group of Fisheries and Aquatic Research (GoFAR) involving World Fish Center, Malaysia.
- vii. Three regional PGR networks for South Asia, Southeast Asia and the Pacific region facilitated by Bioversity International.

These networks helped in joint evaluation and exchange of useful genetic resources by concerned NARS in each sub-region.

As Head of CGIAR Consortium for Sustainable Agriculture in Central Asia and the Caucasus (CAC) region, Dr Paroda catalysed the process of strengthening the Genebanks in Azerbaijan and Uzbekistan and created new Genebanks in Armenia, Georgia, Kazakhstan, Kyrgyzstan, Tajikistan, and Turkmenistan. The one established at Krasnyvadopad in Kazakhstan has been named as ‘Raj Paroda Genebank’. For these extra-ordinary efforts, the CAC team under the leadership of Dr Paroda had won the prestigious “King Baudouin Award” of CGIAR.

To promote regional cooperation and scientific networks, Dr Paroda took major initiative to organise a number of Regional Expert Consultations as follows:

- i. Regional Roundtable Meeting on Implementation of the International Treaty for Plant Genetic Resources for Food and Agriculture in 2005 in Bangkok;
- ii. Expert Consultation on Progress of Research Networks at ICRISAT in 2007;
- iii. International Symposium on Agrobiodiversity in Suwon in 2010 which led to adoption of The Suwon Agro-Biodiversity Framework
- iv. Regional Workshop for Implementation of Suwon Agrobiodiversity Framework in 2011 in Kuala Lumpur;
- v. Regional Consultation on Genetic Resources in the Pacific held at Fiji (2012);
- vi. Regional Consultation on Medicinal Plants in 2013 in Bangkok.



A highly decorated scientist, Dr RS Paroda is recipient of many awards. Seen here with three Presidents of India in various award ceremonies, including Padma Bhushan (1998) (center)



Inaugural session of the '1st International Agrobiodiversity Congress (IAC2016)' in the presence of Hon'ble Prime Minister of India

GR Contributions at Global Level

At the global level, Dr Paroda served as the Chairman of the Working Group on Farmers' Rights of FAO Commission on Plant Genetic Resources (1995-97) when he could get the Farmers' Right defined and agreed by the Commission. He also served as member of the International Committee constituted for Plant Genetic Resources by the Board on Agriculture, National Academy of Sciences, Washington for the period 1990-94 and recommended renovation and strengthening of the Genebank.

As founder chairman, Global Forum on Agricultural Research (GFAR), FAO, Rome, Dr Paroda had organised a Global Conference in 2000, in collaboration with Bioversity International in which "Dresden Declaration on Plant Genetic Resources for Food and Agriculture" was adopted, which defined a road map for strengthening PGR activities at the global level. Dr Paroda as Chair of GFAR also worked passionately and closely with Dr Ismail Serageldin, Chairman, CGIAR and Dr Geoff Hawtin, Director General, IBPGR to convince donors to support the cause of conserving global genetic resources. These efforts eventually led to the establishment of Global Crop Diversity Trust (GCDT) in 2004.

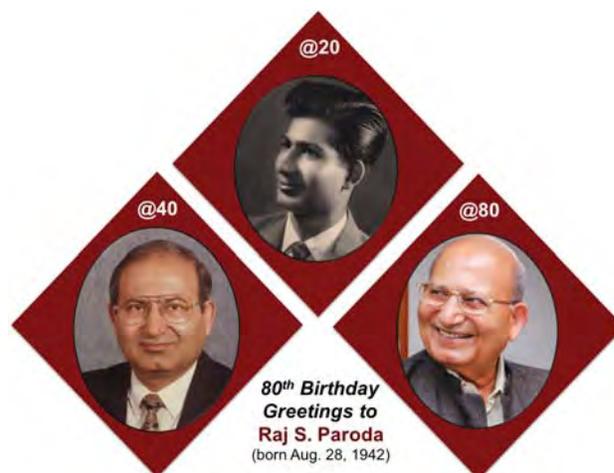
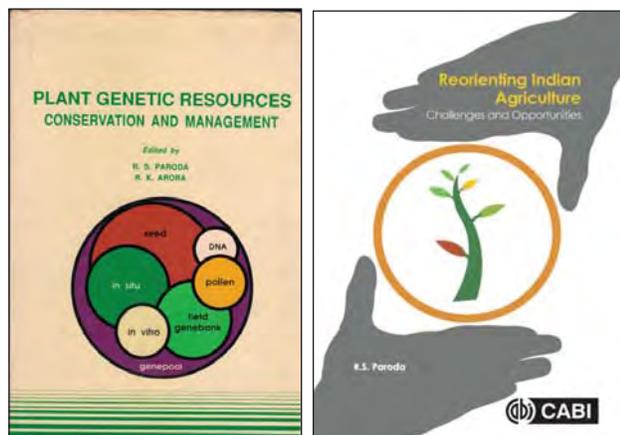
As Chairman, TAAS, Dr Paroda organised a 'Global Consultation on Use and Management of Agrobiodiversity for Sustainable Food Security' in 2013 at New Delhi. As President, ISPGR (2016-18),

he further visualised and organised very successfully the '1st International Agrobiodiversity Congress (IAC)' with 1,000 participants from 60 countries during 6-9 November, 2016 at New Delhi, which was inaugurated by the Hon'ble Prime Minister Mr Narendra Modi. Besides its proceedings, the conference also adopted "Delhi Declaration on Agrobiodiversity Management". The IAC has now become a regular event as 2nd Conference was held in November, 2021 in Rome organised by the Government of Italy in collaboration with Bioversity International and ISPGR. The third one is expected to be held in China in 2025.

Publications on GR

Dr Paroda brought out several important publications including books, strategy papers and reports for creating awareness on conservation and use of agrobiodiversity. Significant ones include:

- i. Life Support Plant Species – Diversity and Conservation. National Bureau of Plant Genetic Resources, New Delhi. Eds. Paroda RS, Kapoor Promila, Arora RK and Bhag Mal. 1987. 190p;
- ii. Plant Genetic Resources-Indian Perspective, Eds. – Arora RK, Chandel KPS and Paroda RS, National Bureau of Plant Genetic Resources, New Delhi 1988. 545p;
- iii. Plant Genetic Resources – Conservation and Management Concepts and Approaches. Eds - Paroda



RS and Arora RK, International Board for Plant Genetic Resources, 1991, 392 p.

- iv. Agro-Biodiversity, Conservation, Management and Use. Eds.-Paroda RS, Rai M. and Gautam PL, 1998.
- v. Status of Plant Genetic Resources Conservation and Utilization in the Asia-Pacific Region: Regional Synthesis Report. Eds. – Paroda RS and Chandel KPS, 2000;
- vi. Strategy Paper on Implementing the International Treaty to Address Current Concerns about Managing our Plant Genetic Resources by Dr. RS Paroda, January, 2012;
- vii. “Reorienting Indian Agriculture” by Dr RS Paroda published by CABI, London, 2018. 296p, which has a Section on Managing Plant Genetic Resources encompassing three chapters (a) The International Treaty–Current Concerns; (b) Agrobiodiversity: Dynamic Change Management, and (c) Managing Agrobiodiversity Through Use: Changing Paradigms.

Conclusion

Dr RS Paroda has been a champion for the cause of genetic resources management for over five decades. Rightly nicknamed as “Genebank Guru”, his passion for the subject has led to massive reforms in infrastructure, capacity building and projects/programs in the arena of agrobiodiversity management and use, which has a direct bearing in increasing agricultural productivity leading to food and nutritional security. The ISPGR established in 1987, was his brainchild and as the Founder (1987-88) as well as current (2022-24) President of the society, he has immensely contributed in its overall growth and visibility. Indian agriculture is fortunate to have a globally recognized visionary like him. On the occasion of his 80th birthday on August 28, 2022, the genetic resources fraternity wishes this octagerian and living legend many more years of healthy life and happiness, and he continues to remain a torch-bearer for generations ahead.

ARTICLES ON
PLANT GENETIC RESOURCES



ARTICLES ON PLANT GENETIC RESOURCES

Title	Page No.
Use of Plant Genetic Resources for Food and Nutritional Security	65
Mini Core Germplasm Collections for Developing Climate Resilient Crop Cultivars and Achieving Enhanced Genetic Gains in Plant Breeding Programs	69
Biofortified Crops – Boon for Nutritional Security	74
Status of Genetic Resources in Oilseed Crops and Their Potential Use for Making India Atmanirbhar in Edible Oils	85
International Year of Millets 2023: Opportunity for Enhancing the Use of Indian Millets Germplasm	90
Genome Editing Technologies for Efficient use of Plant Genetic Resources	95
Building Value Chains for Enhanced PGR Utilization and Sustainable Food Systems	100
Contribution of Technology in Enhancing and Preserving PGR	107
Vegetable Genetic Resources to Mitigate Nutritional Insecurity in India	110
Exploration and Collection of Plant Genetic Resources in India: Status and Priorities	117
Crop Wild Relatives in India: Inventorization, Prioritization and Conservation	124
Plant Taxonomy and Biosystematics in PGR Collecting and Conservation	131
Application of Geographical Information System for PGR Management	136
Role of Plant Quarantine in Preventing Entry of Exotic Pests	141
Large-scale Characterization for Management and Utilization of Indian National Genebank Collections	147
Plant Genetic Resources for Crop Improvement: The North-Western Himalayan Perspective	151
Management of Horticultural Genetic Resources in India: Recent Advances	154
Conservation and Use of Underutilized Crops: Challenges and Way Forward	162
Use of Crop Wild Relatives (CWRs) of Wheat in Disease Resistance Breeding	169
Plant Germplasm Registration System in ICAR – Achievements	172
Role of Genebanking in the Era of Genomics	176
Sustainability of <i>In Vitro</i> Genebanks and Cryogenebanks	180
Repository of <i>Allium</i> Genetic Resources at ICAR-NBPGR: Prospects and Challenges for Collection and Conservation	185
Grain Legumes Diversity of Indian National Genebank: A Potential Resource for Food and Nutritional Security	191
Genomic Tools in Plant Genetic Resource Management	194
Role of Bioinformatics in the Development of Plant Genetic Resources	200
Genomic Resource Generation in Medicinal and Aromatic Plants	204
Policies Impacting Access to Plant Genetic Resources in Last Four Decades	213
PGR Informatics: Sustaining PGR Management and Supporting PGR Policy	218

Use of Plant Genetic Resources for Food and Nutritional Security

Virinder Singh Sohu¹ and Baldev Singh Dhillon^{2*}

¹*Principal Wheat Breeder and Head, Department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana-141027, Punjab, India*

²*Former Vice Chancellor, Punjab Agricultural University, Ludhiana-141027, Punjab, India*

Plant genetic resources (PGR) have been extensively used to develop new cultivars for meeting the emerging challenges and needs of the society. These have played a pivotal role in assuring food and nutritional security of the humankind. Our National Agricultural Research System, particularly the National Bureau of Plant Genetic Resources, has made dedicated efforts to collect indigenous germplasm and introduce exotic germplasm and conserve the same. Indigenous germplasm has been used to develop crop varieties and even now traditional varieties/landraces act as rich source of genes of economic importance (submergence tolerance in FR13A rice, salinity tolerance in Pokkali rice and Kharchia wheat). Exotic germplasm has been used to develop several landmark varieties including IR 8 rice; Kalyansona and Sonalika wheats, the cultivation of which resulted in Green Revolution. Hybridization of indigenous and exotic germplasm to combine their desirable traits has led to remarkable achievements in enhancing crop productivity coupled with adaptation to prevalent agro-ecologies. The overdependence on improved cultivars of few crops (to ensure food security of increasing human population) for human diet markedly changed the food consumption pattern causing hidden hunger, and also making these crops more vulnerable to biotic and abiotic (climate change) stresses. In view of these developments, research on nutritional security and adaptation to changes in agro-ecologies has been getting greater attention in recent past with appreciable successes. In these research programmes, unadapted germplasm of same crop species and wild relatives are being increasingly used. More focussed efforts are needed on acquisition, characterization and conservation of PGR as well as their use. The International Treaty on Plant Genetic Resources for Food and Agriculture needs to be implemented in letter and spirit and pre-breeding research on wild relatives be strengthened to develop usable PGR and be strongly linked with cultivar development.

Introduction

Plant genetic resources (PGR) are the genetic material of plants having value as a resource for present and future generations. PGR include land races and primitive cultivars, modern varieties, parental lines of hybrids, genetic stocks with known desirable attributes and wild and weedy relatives of crop plants. Being the building blocks of improved crop varieties, the availability of PGR is pivotal requirement in cultivar development programmes. Earlier there was sufficiently wide food basket that ensured balanced nutrition of human beings. As civilization progressed, the ever-increasing population and changes in lifestyle necessitated research programmes for productivity enhancement. Commendable achievements of these programmes in some crops led to overdependence on wheat, rice, maize, potato and cassava which meet 75% caloric requirement of our society. The narrowed food basket resulted in nutritional deficiencies. The spread of improved varieties also narrowed on-farm diversity accentuating the damage due to insect-pests and diseases. Recently, the climate

change and volatility have added further dimensions. For addressing these problems, the focus has been greatly enhanced on search, development and use of PGR for traits like tolerance to temperature and moisture extremes, high protein, zinc and iron concentration, and even traits like low glycemic index and low gluten concentration to tackle the increasing incidence of diabetes and gluten intolerance.

Indian centre is one of the eight centres of origin and one of the 12 mega gene centres of the world. Many crops, including important ones, namely rice, chickpea, pigeonpea, mungbean, urdbean, sugarcane, *desi* cotton, jute, eggplant, cucumber, mango, citrus, banana have originated in this centre. India is enormously rich in plant wealth and lot of variability for these crops has been collected, conserved and utilized.

Collection of Indigenous Germplasm and its Use

In India, initial sporadic collection of indigenous germplasm were made in wheat (Howard and Howard, 1906-10, D Milne), jute (Burkill and Finlow, 1907),

*Author for Correspondence: Email-dhillonbaldevsingh@gmail.com

legumes (Shaw *et al.*, 1931-33, rice (Govindaswamy, 1955-60, RH Richharia, 1965, 1977-79; SD Sharma and SVS Sastry, 1967-1972) and sugarcane (Thuljaram Rao, 1968). Systematic and elaborate work on collection and maintenance of germplasm was started in 1946 in IARI, New Delhi by Dr. Harbhajan Singh and co-workers. The National Bureau of Plant Genetic Resources (NBPGR), New Delhi, established in 1976 is the nodal agency for PGR management in the country.

The systematic plant breeding work in India started with collection, classification and purification of local landraces leading to release of wheat varieties T9 and 8A in the 2nd decade followed by rice varieties *Jhona* 349 and *Basmati* 370 in 3rd decade of 20th century. Selection from local landraces continues to yield new varieties with specific traits like aromatic rice variety Kalanamak from Nepal/India, low glycemic index variety Chhattisgarh Madhuraj Paddy-55 developed by refinement of local cultivar Chaptigurmatiya and Sona Moti variety of native wheat (*T. sphaerococcum*). Traditional varieties/landraces are proving to be rich sources of genes/QTLs for traits of economic importance, e.g., in rice, the *sub1* gene from FR13A landrace from Ghaghara Ghat of Uttar Pradesh, salinity tolerance from *Pokkali* of Kerala, the *Pup1* locus for phosphorous deficiency tolerance in *aus* type rice variety Kasalath. Many improved rice varieties like Swarna *sub1* have been developed by utilizing these resources.

Germplasm Exchange

Export of Indian Germplasm

The rich wealth of PGR in India attracted collectors from across the globe. The scientists from India and foreign countries conducted joint explorations to collect germplasm of rice, wild rice, wheat, barley, wild barley, tropical forages, *Brassica* spp., wild crucifers, minor millets, sesame, cucumber, melons, banana, etc. William Farrer introduced Indian wheats in Australia in 1890 for imparting earliness, drought tolerance and rust resistance in the Australian wheats. Wheat variety C591 with excellent grain quality was used in Canadian wheat breeding programme. Wheat variety PBW 65 has been used extensively in breeding programme by International Maize and Wheat Improvement Center, Mexico as a source of resistance to Karnal bunt and loose smut. Chickpea variety C 235 was released in Australia (as Tyson) and USA (as Farha). Maize composite

Vijay was released in Nepal (as Rampur Yellow) and in Pakistan (as J1).

Introduction of Exotic Germplasm

Germplasm exchange, facilitated by human migration, cross-border trade, foreign invaders as well as domestic requirements, has been happening since long. Some important crops like wheat, maize, American cotton, sunflower, etc. were introduced. There are several examples of successful deployment of introduced hallmark germplasm (global elite varieties) as cultivars namely, wheat Sonora 64, Lerma Rojo 64, and rice IR 8, etc. Mungbean variety SML 668, identified from the material received from Asian Vegetable Research Development Centre, Taiwan, revolutionized mung production during spring season in North India.

Secondary Introductions – Selections from Introduced Material

These have made a major impact as exemplified by development of wheat varieties PV18, Kalyansona, Sonalika during late 60s through selections in introduced germplasm, the adoption of which resulted in Green Revolution. Such selections have continued to revolutionize crop production (wheat mega-varieties PBW 343, DBW 187). Selected from germplasm introduced from International Rice Research Institute, Philippines (IRRI), rice variety PR 106 released in Punjab in 1976 ruled the state for three decades. Rice variety PR126 released in 2017, is high yielding, short duration variety. It fits well in the multiple cropping system and has become the most sought-after variety in North-west India. *Gobhi sarson* (*Brassica napus*) was introduced during early 1980s and owing to its higher productivity, oil content, and resistance to white rust, it became an important crop in North-west India. Tomato varieties Punjab Tropic and Punjab Varkha Bahar 4 were developed through selection from segregating material received from the USA and Taiwan, respectively. Chilli varieties Punjab Surkh and Punjab Guchhedar were selections from germplasm received from Indonesia.

Hybridization of Indigenous and Exotic Germplasm

The breakthroughs in yield potential have also been achieved by crossing introduced germplasm with locally adapted germplasm. Introduced Mexican semi-dwarf wheat varieties crossed with local germplasm resulted

in development of landmark varieties including WL 711. Thereafter, wheat introductions carrying 1B/1R translocation (from rye) possessing higher grain yield and resistance to biotic and abiotic stresses were used extensively. In Indian mustard, two varieties (JM 1 and JM 2) were derived by utilizing white rust resistant accessions L 4 and L 6 from Canada.

Nutritional Security

Tremendous progress in productivity in certain crops led to their predominance over vast area. Though, this progress ensured food security to increasing human population but caused nutritional insecurity (hidden hunger) owing to overdependence on few crops for human diet and other changes in food consumption behaviour (processed, junk food). To achieve nutritional security and address human health concerns, research on identification of novel genes in diverse PGR and their mobilization to develop biofortified varieties has been greatly strengthened in recent years. Some nutritionally superior crop varieties released in India are, high protein rice CR Dhan 310; high Zn rice DDR Dhan 49; high Zn wheat PBW 1 Zinc, WB 2; high protein wheat HD 3226, PBW 752; high lysine and tryptophan Maize Vivek QPM 9; high provitamin A maize Pusa Vivek QPM 9 improved, Pusa HQPM 7 Improved; high iron Pearl millet RHB 234, HHB 311; high iron lentil Pusa Ageti Masoor; Kunitz Trypsin inhibitor free Soybean NRC-127; lipoxygenase-2 free soybean NRC 132; high oleic acid Soybean NRC 147; high oleic acid peanut Girnar 4 and Girnar 5; high linolenic acid linseed TL99; high provitamin A Cauliflower Pusa Beta Kesari 1; high anthocyanin potato Kufri Manik; high provitamin A Sweet potato Bhu Sona; high Fe, Zn, Vitamin C Pomegranate Solapur Lal. Hybridization between Australian genotypes led to development of canola quality Gobhi sarson variety GSC 7 that also became popular variety in North-west India. Low erucic acid varieties of Indian mustard (Pusa Karishma, LET 17, LET 18, LES 1-27) were derived from ZEM 1 and ZEM 2 from Australia.

Use of Wild Relatives

Sometimes genetic variability is not available in adapted germplasm and necessitates the use of unadapted germplasm or related species or even wild weedy relatives. In such cases, prebreeding is needed to develop usable PGR which used to be a long-term and tedious project. However, the biotechnological tools now

available, are being applied to hasten it. High grain weight and grain number per spike has been transferred from diploid progenitor species (*Ae. tauschii*, *T. monococcum*, *T. boeoticum*, *T. dicoccoides*) to bread wheat. Rust resistance genes viz. *Lr 57/Yr40* (*Ae. geniculata*), *Lr 58* (*Ae. triuncialis*), *Lr76/Yr70* (*Ae. umbellulata*) have been characterized and mobilized from wild species to elite wheat background leading to development of varieties Unnat PBW 343, PBW 803, etc. High grain protein gene



Maize germplasm having plant type required for the development of cultivars suited for cultivation under high plant density

Gpc B1 (originally from *T. dicoccoides*) and high grain Fe and Zn QTL's (from *T. monococcum*, *T. boeoticum*) have been transferred to durum and bread wheats. In rice, *O. nivara*, acc. 101508, the only known source of resistance to grassy stunt virus is being widely used as donor. Many genes imparting resistance to brown plant hopper and bacterial blight of rice have been introgressed from related wild species. These include widely used *Xa 21* gene from *O. longistaminata* identified at IRRI and *Xa 38* gene from *O. nivara* at PAU. Resistance to aphid and Sclerotinia stem rot in *B. juncea* has been achieved through introgression from *B. fruticulosa* and *Erucastrum cardaminoides*. In pigeonpea, *C. scarabaeoides* was used as a source of genes for tolerance to *Maruca vitrata* and *Phytophthora* stem blight besides higher productivity. In chickpea, resistance to Botrytis grey mould and Ascochyta blight has been introgressed from *C. pinnatifidum* and *C. reticulatum*.

Nobilization of sugarcane is a classic example of use of wild relatives. It was initiated by CA Barber and Venkataraman in 1912 at Sugarcane Breeding Institute, Coimbatore. Local strains of *Saccharum barberi*, (unproductive species that grows wild in North India) was crossed with tropical noble cane *S. officinarum* (thick soft stem, high sucrose content but unsuited to North India). The resultant hybrid was crossed with wild cane *S. spontaneum*, and tri-hybrid canes with high sucrose content suitable for North India were developed.

Future Perspectives and Action Points

- PGRs are the backbone of plant breeding programmes and are vital for food and nutritional security of

human being. However, after the establishment of WTO, germplasm exchange and use of PGR obtained from other sources is becoming increasingly difficult and complex. The 'International Treaty on Plant Genetic Resources for Food and Agriculture' needs to be implemented in letter and spirit and be accompanied by the efforts to facilitate acquisition of germplasm.

- The IPRs on PGR needs to be balanced with 'benefit sharing' and 'farmers rights'.
- The database on linking acquisition, characterization and conservation needs to be strengthened and regularly updated to facilitate PGR use.
- The transformation of multi-institutional CG system to 'One CG' should not adversely affect the germplasm exchange between CG and national agricultural research systems.
- National Genetic Stock Nurseries needs to be strengthened.
- Dedicated prebreeding programmes need to be strengthened as an integral part of crop breeding programmes at least at a few centres to promote the utilization of genes of interest from wild relatives.
- Deposition of precious breeding lines developed under multi-institutional collaborative ad hoc research projects with NBPGR and their sharing should be made mandatory.

Mini Core Germplasm Collections for Developing Climate Resilient Crop Cultivars and Achieving Enhanced Genetic Gains in Plant Breeding Programs

Hari D Upadhyaya* and Andrew H Paterson

Plant Genome Mapping Laboratory, University of Georgia, Athens, GA 30605, USA

Crop germplasm diversity contributes to developing improved crop cultivars aimed at increasing crop productivity to meet future food security needs of world. The large size of germplasm collections and lack of reliable information on traits of economic importance has resulted in low use (<1%) of germplasm. To facilitate realization of the potential benefits of germplasm collections to broaden the genetic base of cultivars in many crops, the mini core (~1% of the entire collection) concept was postulated and a development process proposed, to produce resources recognized globally as an “International Public Good” (IPG). Mini core collections have been developed in several crops and new diverse sources of genetic variation identified for biotic and abiotic stresses, agronomic and nutritional traits for use in crop improvement programs. Using genetically diverse sources scientists have developed breeding lines with enhanced trait expression and higher genetic gains. Molecular characterization of germplasm has revealed that phenotypic traits were as effective as molecular markers in sampling allelic diversity to form mini core collections.

Introduction

Germplasm is the total gene pool of a species consisting of landraces, advanced breeding lines, elite cultivars, and wild and weedy relatives. It forms the raw material for any crop improvement program and plays an important role in developing cultivars with the intrinsic genetic potential to mitigate the adverse effects of climate change on global agriculture. Nikolai Ivanovich Vavilov, was an early and exemplary advocate of the importance of genetic diversity for crop improvement and organized extensive germplasm collections of various crops from their ‘centers of origin’ and distribution for conservation. Since then the germplasm collections of major crop plants continued to grow in number and size. Globally, 7.4 million germplasm accessions are conserved in about 1,750 genebanks. The vulnerability of genetically uniform modern varieties, which are often closely related to one another and planted to large areas, to new pests, diseases, climatic conditions and changes in market needs is widely acknowledged. Diverse landraces, exotics and wild relatives hold a wealth of genes/alleles which, if included in breeding programs, can help raise the yield ceiling as well as enhance stress resilience of agronomically superior cultivars.

Definition and Development of Mini Core Collection

Less than 1% of assembled germplasm has been used in breeding programs globally (Upadhyaya *et al.*, 2006). The elite cultivars of many major crops trace to very few founder genotypes – for example, 50% of wheat cultivars trace to 9 genotypes; 75% of potato to 4, and 50% of soybean to 6 (World Conservation Monitoring Centre, 1992). In chickpea, 41% of cultivars from hybridization have Pb7 as a parent; in pigeonpea 41% have T1 and T190; in urdbean 64% have T9 and in mung bean 35% have T1. Limited numbers of germplasm lines were used in chickpea breeding programs including those of ICRISAT and ICARDA. At ICRISAT, only 91 germplasm lines have been used to develop 3,548 advanced varieties during 1978-2004. L-550 (909 times) and K 850 (851 times) were the most widely used accessions (Upadhyaya *et al.* 2006). The main reasons for low use of germplasm include large size of collections and lack of reliable data on traits of economic importance, which show high genotype x environment interaction (Upadhyaya *et al.*, 2013). To overcome these problems, Frankel (1984) proposed the concept of core collections (10% of entire collection) that can be evaluated more extensively to identify

*Author for Correspondence: Email-hari.upadhyaya@uga.edu

promising germplasm. Core collections of various crops were developed at ICRISAT (Table 1). However, it was soon realized that even the size of core collections was unwieldy for replicated multilocal evaluation to identify sources of variation for traits which show genotype \times environment interaction, as the large field areas required to evaluate thousands of lines inherently added to environmental variation. To overcome this, Upadhyaya and Ortiz (2001) proposed the concept of mini core collections (10% of core or 1% of entire collection)

Upadhyaya and Ortiz (2001) proposed a two stage strategy for developing mini core collections, as follows:

1. Development of a core collection (~10%) from the entire collection;
2. Evaluation of the core collection for various morphological, agronomic and quality traits or need specific characters and selecting a further subset of about 10% of accessions that sample phenotypic diversity.

At both stages, standard clustering procedures are used to identify groups of similar accessions (Fig. 1), to guide sampling of diversity from the entire collection in the core or mini core entries. Following this strategy mini core collections have been developed at the ICRISAT (Table 1) and several other countries.

Mini Core Collections Meet the Needs of Plant Breeders for Variation in Multiple Traits

Plant breeders often work on many traits simultaneously and prefer sources of variation for multiple traits. The mini core collections developed at ICRISAT have been evaluated for various biotic and abiotic stresses, for nutritional traits and for their agronomic performance in multilocal replicated trials. Genetically diverse sources for individual as well as for multiple traits have been reported in chickpea, groundnut and sorghum (Upadhyaya et al 2013, 2014, 2019). Such diverse germplasm with multiple trait-specific sources in agronomically desirable backgrounds offer breeders opportunities to combine several genes (Upadhyaya *et al.*, 2014).

Mini core collections have helped increase use of germplasm in crop breeding. Pedigree analyses of chickpea breeding lines developed at ICRISAT revealed 47% greater use of germplasm and use of 7 unique germplasm accessions during 2005-08. A similar pattern was observed in groundnut.

Trait Enhancement and Greater Genetic Gains through Use of Mini Core Sources

The use of diverse groundnut germplasm lines identified from a mini core collection resulted in developing exceptionally high oil (up to 63%, compared to ~48% in control cultivar) and high-yielding breeding lines,

Table 1. Core and mini core collections developed for ICRISAT mandate crops

Crop	Accessions	Traits	Core/mini core	Accessions	Reference
Chickpea	16,991	13	Core	1,956	<i>Crop Sci.</i> 41 : 206-210.
	1956	22	Mini core	211	<i>Theor. Appl. Genet.</i> 102 : 1292-1298
Groundnut	14,310	14	Core	1,704	<i>Genet. Resou. Crop Evol.</i> 50 : 139-148
	1704	47	Mini core	184	<i>Crop Sci.</i> 42 : 2150-2156
Pigeonpea	12,153	14	Core	1,290	<i>Genet. Resou. Crop Evol.</i> 52 : 1049-1056
	1,290	33	Mini core	146	<i>Crop Sci.</i> 46 : 2127-2132
Sorghum	22,473	20	Core	2,247	<i>Crop Sci.</i> 41 : 241-246
	2,247	21	Mini core	242	<i>Crop Sci.</i> 49 : 1769-1780
Pearl millet	20,766	12	Core augmented	2094	<i>Crop Sci.</i> 49 : 573-580
	2,094	18	Mini core	238	<i>Crop Sci.</i> 51 : 51 :217-223
Finger millet	5,940	14	Core	622	<i>Genet. Resou. and Crop Evol.</i> 53 : 679-685
	622	20	Mini core	80	<i>Crop Sci.</i> 50 : 1924-1931
Foxtail millet	1,474	23	Core	155	<i>Plant Genet. Resou.</i> 7 : 177-184
	155	21	Mini core	35	<i>Field Crops Res.</i> 124 : 459-467
Proso millet	833	20	Core	106	<i>Crop Pastu. Sci.</i> 62 : 383-389
Barnyard millet	736	21	core	89	<i>Crop Sci.</i> 54 : 2673-2682
Little millet	460	20	core	56	<i>Crop Sci.</i> 54 : 2673-2682
Kodo millet	656	20	core	75	<i>Crop Sci.</i> 54 : 2673-2682

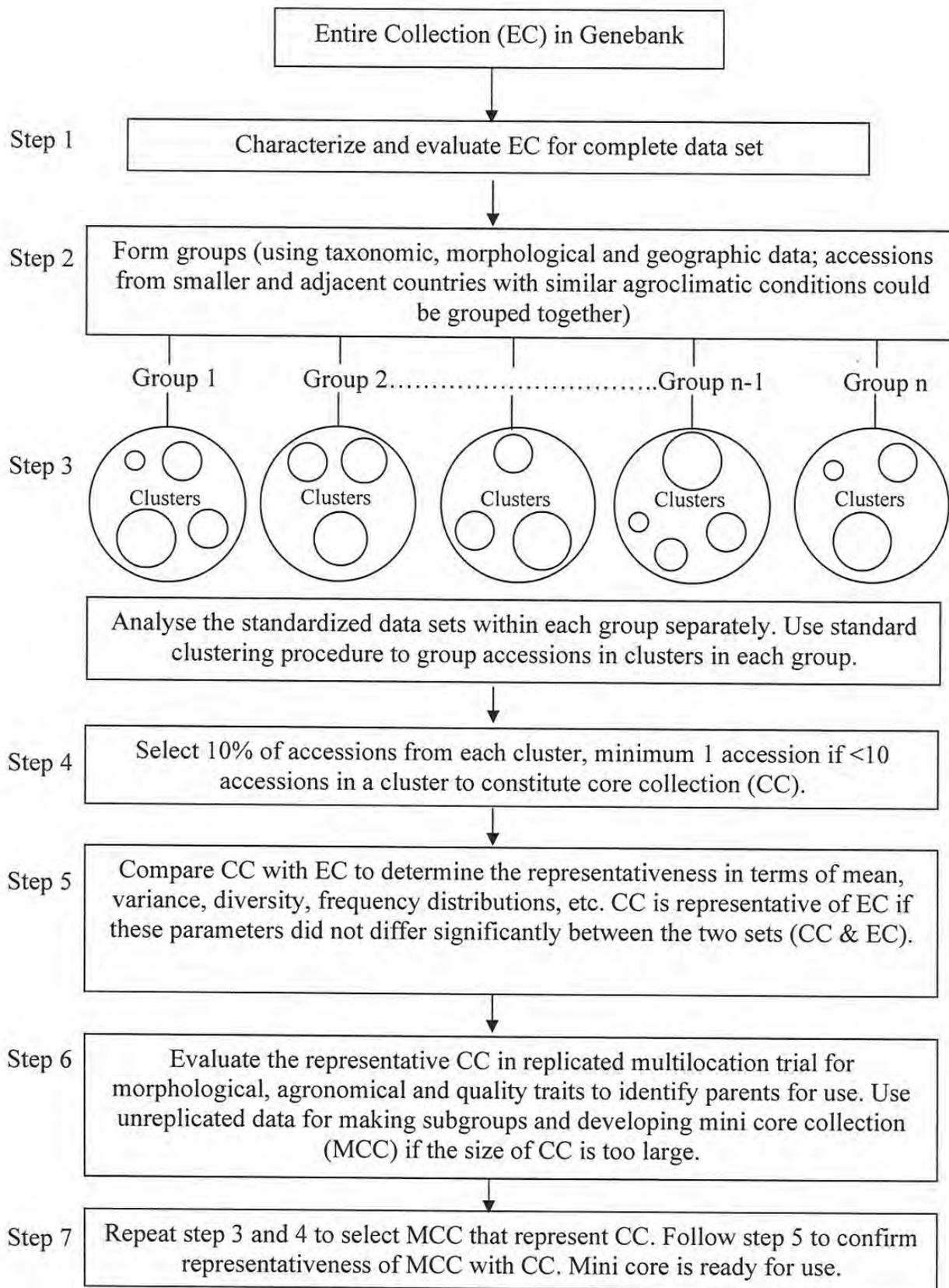


Fig. 1. Flow diagram to establish core and mini core collections in a crop species (adapted from Upadhyaya *et al.*, 2009)

indicating that new germplasm sources contribute to enhancing genetic gains.

Comparison of Mini Core Collections Developed using Phenotypic Traits and those Based on Molecular Markers

In early 2000, when this author (Hari Upadhyaya) started developing mini core collections of the ICRISAT mandate crops and presented outcomes in international conferences, scientists often asked about the efficacy of phenotypic traits *vis-à-vis* molecular markers in sampling diversity to form the mini core collection. Our collaborative work at ICRISAT with the Generation Challenge Program helped us to genotype a large number of germplasm accessions of chickpea, sorghum (both 3,000 accessions), groundnut, pigeonpea, pearl millet and finger millet (1000 accessions each), and foxtail millet (500) with 20-50 simple sequence repeats (SSR). The aim was to study genetic diversity, population structure and to establish reference sets of 200-400 genetically diverse accessions. To cite an example, the genetic structure, diversity and allelic richness in a world composite collection of chickpea (3,000 accessions), using 48 SSR markers, was assessed and a reference set of 300 accessions was established at ICRISAT (Upadhyaya *et al.*, 2008). The 48 SSR markers detected 1,683 alleles in 2,915 accessions, of which 935 were considered rare, 720 common and 28 most frequent. The composite collections were also characterized for qualitative and quantitative traits at ICRISAT Center, Patancheru, India. Reference sets based on SSR markers, qualitative traits, quantitative traits and their combinations were formed and compared for allelic richness and diversity. In chickpea, for example 48 SSR based reference set captured 78.1% of 1683 alleles in the composite collection compared to 73.5% of alleles in the reference set based on seven qualitative traits. The reference sets based on SSR and qualitative traits captured 80.5% (1,354) of alleles in the composite collection. Similarly, in groundnut the SSR-based reference set captured 95.1% (466) of alleles in the composite collection (490) compared to 93.3% (457) of alleles in the reference set based on 14 qualitative traits. The reference sets based on SSR and qualitative traits captured 95.9% (470) of alleles in the composite collection. In pigeonpea, a reference set based on SSR data and consisting of 300 most diverse accessions, captured 187 (95%) of the 197 alleles in the composite collection.

Another reference set based on qualitative traits captured 87% of the alleles in the composite set. This demonstrated that SSR and qualitative traits were similarly efficient in capturing the allelic richness of composite collections, and thus mini cores that were selected using phenotypic traits were as good as those based on SSRs.

Mini Core Collections as Association Mapping Panel

A sorghum mini core collection has been used as an association mapping panel for various traits such as plant height, maturity, kernel weight and number, resistance to grain mold, rust, anthracnose, germination at low temperature, saccharification yield and other traits.

Conclusion

To meet unprecedented challenges to global agriculture in the 21st century, new genetic variation for developing climate-resilient crops is required. Further, climate resilient cultivars of many taxa, for example pulses, must be nutritionally dense to mitigate adverse effects of malnutrition. Enhanced use of germplasm is needed to develop such cultivars. The concept and process of forming mini core collections, which was recognized as an International Public Good, is very useful in identifying promising sources. Genetically diverse parents provide opportunities to identify transgressive progeny with greater trait expression and enhanced genetic gains.

References

- Frankel OH (1984) Genetic perspective of germplasm conservation. In: W Arber, K Limensee, WJ Peacock and P Stralinger (eds) *Genetic Manipulations: Impact on Man and Society* Cambridge University Press, Cambridge, UK, pp. 161-470.
- Upadhyaya HD and R Ortiz (2001) A mini core subset for capturing diversity and promoting utilization of chickpea genetic resources in crop improvement. *Theor. Appl. Genet.* **102**: 1292-98.
- Upadhyaya HD, CLL Gowda, HK Buhariwalla and JH Crouch (2006) Efficient use of crop germplasm resources: identifying useful germplasm for crop improvement through core and mini core collections and molecular marker approaches. *Plant Genet. Resour.* **4**: 25-35.
- Upadhyaya HD, SL Dwivedi, M Baum, RK Varshney, SM Udupa, CLL Gowda, D Hoisington and S Singh (2008) Genetic structure, diversity and allelic richness in composite collection and reference set in chickpea (*Cicer arietinum* L.). *BMC Plant Biology* **8**: 106. <http://www.biomedcentral.com/1471-2229/8/106>.
- Upadhyaya HD, N Dronavalli, SL Dwivedi, J Kashiwagi, L Krishnamurthy, S Pande, HC Sharma, V Vadez, S Singh,

- RK Varshney and CLL Gowda (2013) Mini core collection as a resource to identify new sources of variation. *Crop Sci.* **53**: 2506-2517.
- Upadhyaya HD, SL Dwivedi, V Vadez, F Hamidou, S Singh, RK Varshney and B Liao (2014) Multiple resistant and nutritionally dense germplasm identified from mini core collection in peanut. *Crop Sci.* **54**: 679-693.
- Upadhyaya HD, M Vetriventhan, AM Asiri, V CR Azevedo, HC Sharma, R Sharma, SP Sharma and Y-H Wang (2019) Multi-trait diverse germplasm sources from mini core collection for sorghum improvement. *Agriculture* **121**. doi:10.3390/agriculture9060121
- Upadhyaya HD, RPS Pundir, SL Dwivedi and CLL Gowda (2009) Mini core collections for efficient utilization of plant genetic resources in crop improvement programs. *Information bull.* No. **78**, ICRISAT, India, pp. 42.
- World Conservation Monitoring Centre (1992) In: Groombridge B (ed) *Global Biodiversity: Status of the earth's living resources*, Chapman & Hall.

Biofortified Crops – Boon for Nutritional Security

Shuvobrata Majumder, Karabi Datta, Dipak Gayen, Soumitra Paul, Nusrat Ali, Subhrajyoti Ghosh, Aritra Karmakar, Sananda Bhattacharya, Shinjini Sengupta and Swapan Kumar Datta*

Laboratory of Translational Research on Transgenic Crops, Department of Botany, University of Calcutta, Kolkata-700019, India

Micronutrient deficiencies of iron, zinc, and vitamin-A are such serious global health issues, that it affects one out of every three people worldwide. The intensity of this “hidden hunger” compels us to acknowledge global nutritional security issues. Plant based food are the most popular and trending choices for people of all rungs of the social ladder. Biofortification is a sustainable and promising process of improving nutrition in plant based food through different agronomic approaches, conventional plant breeding and genetic engineering. Biofortified crops have been developed with high iron, high zinc, vitamin-A, with other nutritional quality enhancements and these crops have already proved to be a ‘boon for nutritional security’. This review highlights some selected Biofortified crops with special reference to rice (*Oryza sativa*), as 50% of the global population relies on it.

Key Words: Hidden hunger, Biofortification, Nutritional Security, Rice, Wheat, Maize, Cassava, Legumes

Introduction

Human population is ever increasing and is currently more than 7 billion (Fig. 1A). According to a United Nations new report, we will be 9.8 billion by 2050 and 11.2 billion by 2100 (www.un.org/en/desa, accessed on July 18, 2022). An increase in the global food production (Fig. 1B) is the need of the hour to ensure food security. Food may satisfy hunger but only nutritionally enriched food can satiate “hidden hunger”.

Malnutrition has affected more than 2 billion people around the world, mostly in countries of South Asia, Africa and Latin America (FAO, 2015). It stealthily compromises the immune system, declines growth of the mind and body and increases chances of mortality. Daily, more than 24,000 people die globally owing to malnutrition (Fiaz *et al.*, 2019). Iron deficiency anaemia (IDA), zinc deficiency, and vitamin-A deficiency (VAD) are common among the malnourished population. Children of sub-Saharan Africa, South and Southeast Asia are suffering from VAD and its associated risks more than other countries in the world (Wu *et al.*, 2021). Such deficiency makes these children vulnerable to VAD associated infectious, diarrheal diseases, blindness, sensory losses, and premature death (Schmitz *et al.*, 2012).

Adequate supply of nutrient enriched diversified food, and food supplements may not be sustainably available to the affected malnourished population. One of the sustainable approaches to combat malnutrition in all the affected parts of the world is through nutrition enriched food crops (Welch, 2002). Cereal crops contribute the most in the daily energy intake in malnourished populations. People from Asia and Africa depend on such staple crop meals to meet 60–80% of their per day energy requirement (Fig. 1C). Rice, wheat (*Triticum aestivum*) and maize (*Zea mays*) are the most popularly produced cereal crops worldwide (Fig. 1D). It is understandable that nutritional improvement of these crops could have a massive positive health impact on the world population.

Crops that have been developed for the purpose of fighting against malnutrition are called biofortified crops. These can be developed by means of conventional breeding and genetic engineering. As many as 35 countries have accepted and released such biofortified crops (conventionally bred) for cultivation and were consumed by a population of 40 million (Fig. 1E) (Mishra *et al.*, 2022).

Recently, a high provitamin-A rice variety (Golden Rice) developed through genetic engineering has been released in the Philippines (Majumder *et al.*, 2022).

*Author for Correspondence: Email-swpndatta@yahoo.com

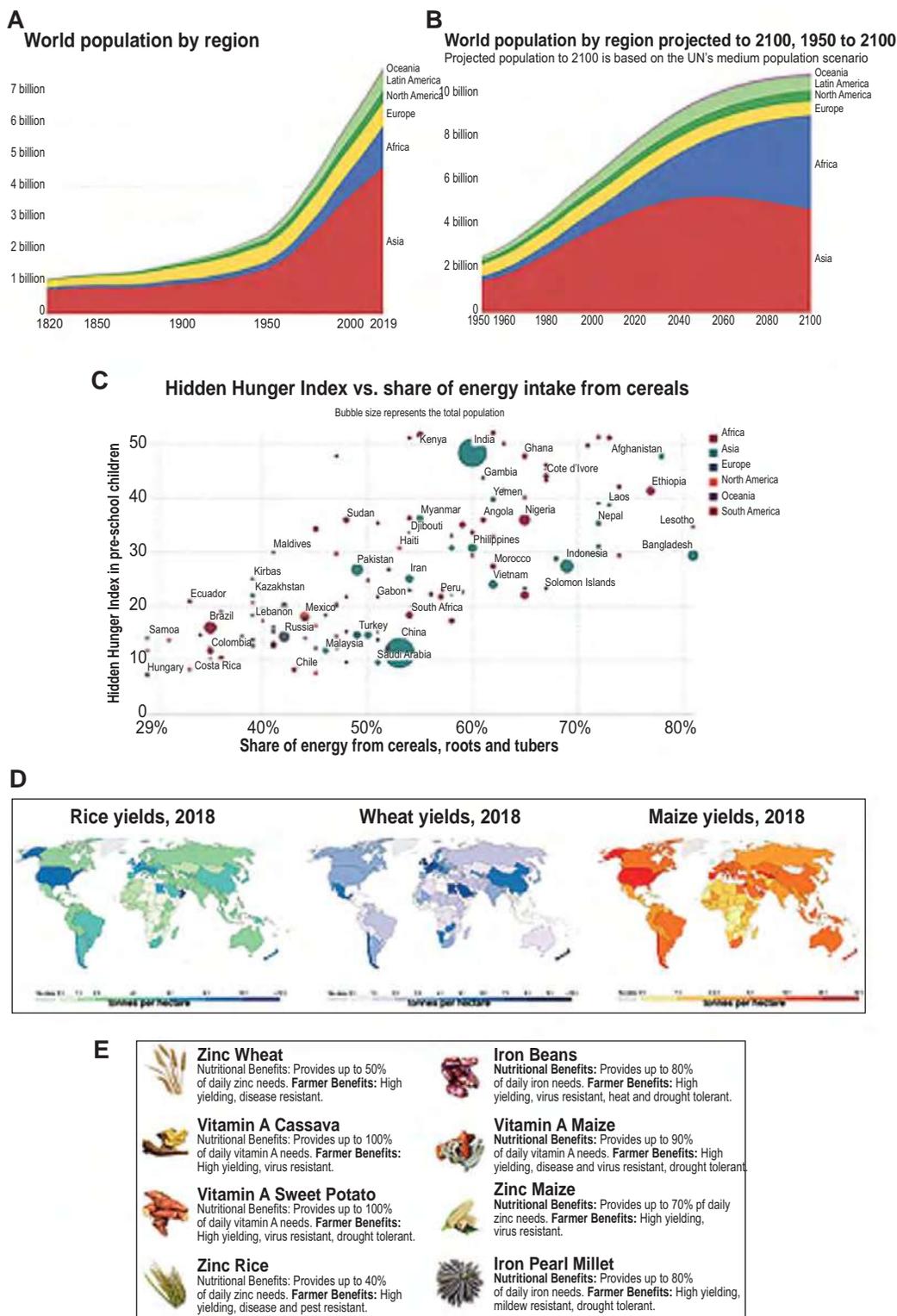


Fig. 1. The world population and cereal nutrition. Current world population by region (A). World population by region projected upto 2100 (B). Share of energy intake from cereals (C). Worldwide yield (tonnes/hectare) of three major cereal crops- rice, wheat, and maize according to 2018 data (D). Released biofortified crops created by conventional breeding approaches (E). This infographic is created based on the information from <https://www.harvestplus.org/> and <https://ourworldindata.org/micronutrient-deficiency>, accessed on July 20, 2022.

Biofortified Rice

Rice is undisputedly the leading cereal crop as a major part of the global population rely on it for upto 70% of their daily calories (Mishra *et al.*, 2018). Modern techniques of molecular breeding were essential to the development of many biofortified rice varieties. The provitamin-A enriched Golden Rice, is one such variety which was recently approved in the Philippines and hopefully it will be also approved soon in Bangladesh (Wu *et al.*, 2021). Different approaches have been successfully applied to developed high iron and high zinc biofortified rice. Recently, the CRISPR/Cas9 genome editing tool was used to develop high oleic rice (Abe *et al.*, 2018).

Golden Rice

Invention of golden rice was a much celebrated application of metabolic engineering. Bacterial and plant genes were introduced into rice to produce β -carotene, a pro-vitamin A precursor. Synthesis of β -carotene in rice endosperm, specifically in non-photosynthetic tissue like endosperm (edible part), requires overexpression of phytoene synthase (PSY) from daffodil (*Narcissus pseudonarcissus*) and bacteria (*Erwinia uredovora*) phytoene desaturase (CRT-I) (Datta and Datta, 2020). Presence of β -carotene in the endosperm adds the golden (yellow orange) colour (Fig. 2).

Introduction of Daffodil *PSY* gene into japonica rice variety (Taipei-309), a carotenoid synthesis pathway intermediate photoene was induced in rice endosperm Burkhardt *et al.* (1997). This proved the principle that provitamin-A can be synthesised in a non-photosynthetic, carotenoid-lacking plant tissue (endosperm). This report is considered as the first golden rice (golden rice-1) development. The same research group also transformed Daffodil *PSY* and lycopene β -cyclase (*LCY*) with bacterial (*E. uredovora*) *CRT-I* into Taipei-309 giving results of 1.6 $\mu\text{g/g}$ total carotenoids (Ye *et al.*, 2000). Combination of these genes gave similar amounts of total carotenoids (1.05 $\mu\text{g/g}$) in other rice cultivars of IR 64, BR 29, Nang Hong Cho Dao and Mot Bui as reported by Datta *et al.* (2003). Till date, 37.0 $\mu\text{g/g}$ of total carotenoids have been reported (golden rice-2) using maize *ZmPSY* and *E. uredovora CRT-I* genes in japonica rice cultivar-Asanohikari (Paine *et al.*, 2005).

In the last two decades, continuous improvement has been observed in golden rice research (Fig. 3). Initially, golden rice was developed using different japonica rice varieties but about 90% of the rice consumers preferred indica rice worldwide (Khush 2001). Therefore, Golden indica rice development was emphasised by multiple research groups with different promoters and gene combinations for better accumulation of carotenoids

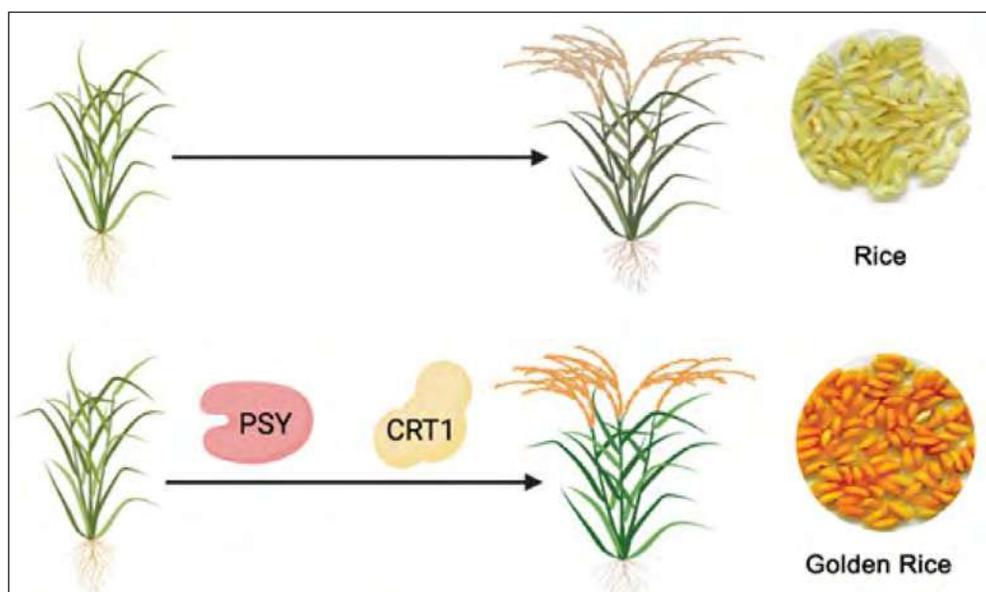


Fig. 2. Metabolic engineering in rice and β -carotene expression in rice grains. Conventional rice is unable to produce β -carotene in its endosperm but golden rice harbours *phytoene synthase* (PSY), and *phytoene desaturase* (CRT-I) genes from other sources and can produce β -carotene in the endosperm. Source of the golden rice picture is from Datta *et al.*, 2007.

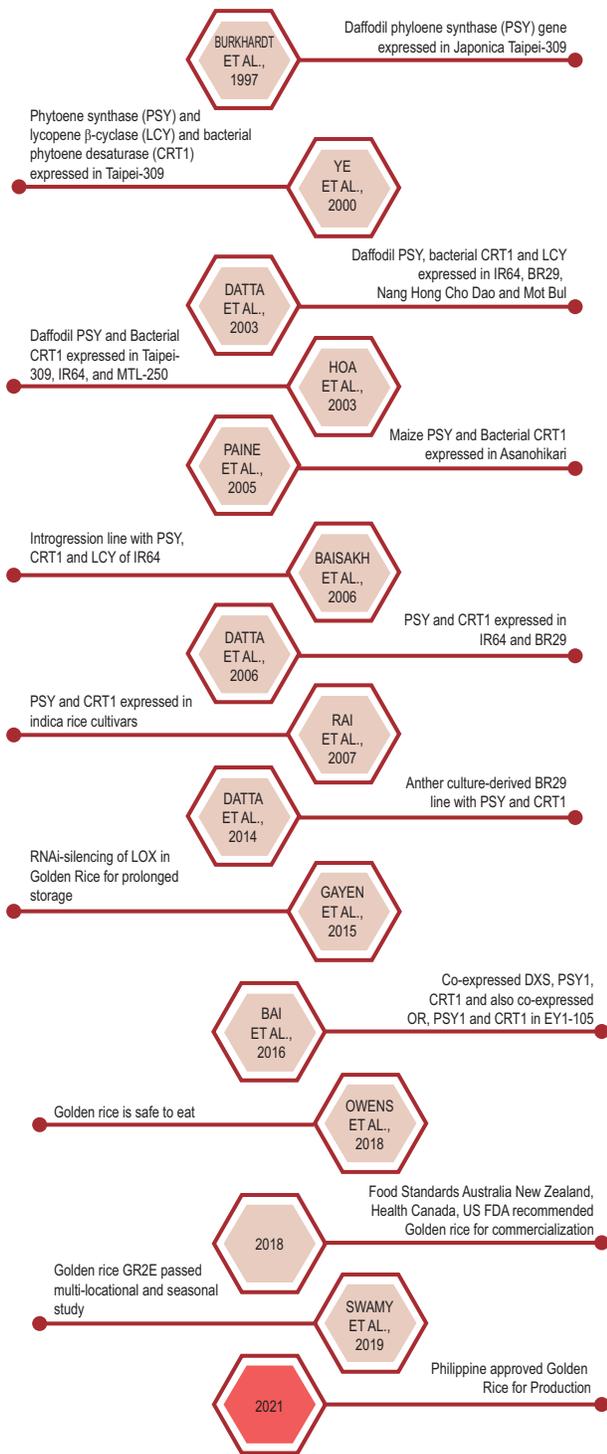


Fig. 3. Timeline (1997-2021) of Golden rice development and its advancement research

in the rice seeds (Datta 1999; Datta *et al.*, 2003, 2006, 2007, 2014; Parkhi *et al.*, 2005; Rai *et al.*, 2007; Rehana *et al.*, 2018; Ghosh *et al.*, 2019).

One limitation of golden rice is deterioration of its carotenoid level during storage (Datta *et al.*, 2021). This is caused by lipoxygenase (LOX) that catalyses lipid peroxidation resulting in the seed quality deterioration. LOX produces hydroperoxy fatty acids which oxidise carotenoids and decolourize seeds (Gayen *et al.*, 2015). Diminishing LOX activity in golden rice seeds can give better stability to carotenoids during storage. Seed specific RNAi-mediated *LOX* gene silencing in golden rice showed less deterioration of carotenoids and stabilised seed quality (Gayen *et al.*, 2014, 2015, 2016).

Recently the three distinguished agencies for international food safety regulation - the Food Standards Australia New Zealand, the Health Canada, and the United States Food and Drug Administration, highly approved ‘Golden Rice’ for commercialization (Datta and Datta, 2020). The Philippines, is the first country in the world for planting and cultivation of golden rice (Wu *et al.*, 2021). Positive perception is gradually developing towards commercialising golden rice with the availability of positive biosafety data on it (Owens *et al.*, 2018).

High Iron Rice

According to the World Health Organization (WHO) estimation, around 800 million children and women are affected by anaemia (WHO, 2015). Developing countries (regions like South–East Asia, Eastern Mediterranean and African) endure IDA more than the higher–income zones (North America, Europe and some parts of Central Asia) (Majumder *et al.*, 2022). Iron biofortified rice could save many lives from these regions. High iron rice has been achieved by multiple strategies some of which are discussed here.

Overexpression of ferritin gene

Upto 4500 iron atoms can form a complex (non-toxic) with the multi-subunit protein ferritin and from this complex the human intestine can absorb iron ions (Andrews *et al.*, 1992). Soybean (*Glycine max*) ferritin genes *SoyferH1* and *SoyferH2* were found to be useful for developing iron biofortified rice (Kok *et al.*, 2018). Soybean ferritin gene was expressed in rice with the help of globulin and glutelin promoters (Goto *et al.*, 1999; Lucca *et al.*, 2002; Vasconcelos *et al.*, 2003; Qu *et al.*, 2005; Khalekuzzaman *et al.*, 2006; Oliva *et al.*, 2014). Rice ferritin gene (*Osfer1*) was transferred into Pusa Sugandhi II rice under the control of a glutelin promoter that accumulated over 2-fold iron in milled

rice grains (Paul *et al.*, 2012). Upto 4.4-fold increment of iron content has been reported in IR64144 rice using the Soybean *ferritin* gene (Vasconcelos *et al.*, 2003) and upon being introduced into the Swarna rice variety, this gene produced 2.54-fold more iron in milled rice than the control (Paul *et al.*, 2014).

Silencing of Phytic Acid in Rice Grains

Phytic acid (myo-inositol-1,2,3,4,5,6-hexakisphosphate or IP₆ or InsP₆) is commonly found in cereals. It predominantly accumulates in the seeds as insoluble complex salt of mineral ions, known as phytate. It is highly potent in chelating divalent ions like iron (Fe²⁺), zinc (Zn²⁺), magnesium (Mg²⁺), and calcium (Ca²⁺), thereby reducing their bioavailability in the body (Majumder *et al.*, 2019). Down regulation of *myo*-inositol-3-phosphate synthase (MIPS), which catalyses the initial step of phytic acid biosynthesis pathway in rice seeds, successfully generated low phytate seeds. Initially the down regulation process was driven by constitutive CaMV35S promoter (Feng and Yoshida, 2004) and later by seed specific promoters like Glutelin B-1 (Kuwano *et al.* 2006) and Oleosin 18 (Kuwano *et al.*, 2009 and Ali *et al.*, 2013a) to eliminate the undesirable effects on vegetative tissue. Reduction of phytic acid level in rice seeds was also achieved by tissue specific downregulation of *myo*-inositol trisphosphate kinase (ITPK) in Khitish rice cultivar (Karmakar *et al.*, 2020 and Sengupta *et al.*, 2021) and *myo*-inositol pentakisphosphate-2-kinase (IPK1) in Pusa Sugandhi II (Ali *et al.*, 2013b). Recently, Sengupta *et al.* (2021) reported that seed specific downregulation of rice *ITPK-2* gene reduces 37% of phytic acid content in the transgenic seeds.

Degradation of Phytic Acid

Phytase is an enzyme that can degrade phytate (a salt form of accumulated Phytic acid) and release chelated minerals, including phosphate. Unfortunately, the digestive tract of monogastric animals, including humans, possess negligible or no phytase activity (Colombo *et al.*, 2020). Due to that, monogastric animals can break down only about 10% of the phytate during digestion and causes major unavailability of important divalent ions of iron, zinc, magnesium, and calcium. Fortunately, degradation of phytate was achieved in rice by seed specific expression of the phytase gene- *Aphytase*, derived from fungi (*Aspergillus fumigatus*) (Wirth *et al.*, 2009; Boonyaves *et al.*, 2016, 2017) and the phytase gene-*appA*, derived from bacteria (*Escherichia coli*)

(Bhattacharya *et al.*, 2019). Recent study in Khitish indica rice cultivar showed that seed specific expression of the *E. coli appA* gene not only increased twofold iron content but also increased zinc by threefold and inorganic phosphorus (Pi) levels by fourfold in rice grains (Bhattacharya *et al.*, 2019). Such rice biofortification strategies can improve the bioavailability of multiple, nutritionally important, divalent ions at a time.

Improvement of Iron Bioavailability

Studies have shown that absorption of non-haem iron can be increased by cysteine-rich metallothionein (MT) (Taylor *et al.*, 1986; Hsieh *et al.*, 1995). A combination of MT gene and *Aphytase* iron-biofortified Taipei-309 rice was developed that could complete the degradation of phytic acid (Lucca *et al.*, 2001). Thereby genes or combination of genes can be used to ensure better absorption of divalent ions to improve bioavailability.

Chelation-based Strategy

When the iron concentration is low, roots of graminaceous staple crops (including rice) secrete phytosiderophores (PS) like mugenic acid (MA) and avenic acid at a low concentration which chelates iron or zinc from the soil (Romheld and Marschner, 1990; Marschner and Romheld, 1994). Two genes – *nicotianamine synthase* (NAS) (*OsNAS1*, *OsNAS2*, and *OsNAS3*) and *nicotianamine transferase* are responsible for PS synthesis in rice (Huguchi *et al.*, 1999; Nozoye *et al.*, 2011). The rice NAS gene was overexpressed to develop high iron biofortified rice (Lee *et al.*, 2009; Johnson *et al.*, 2011; Lee *et al.*, 2012). In some cases NAS genes (*HvNAS1*) from Barley (*Hordeum vulgare*) and Barley's *IDS2* and *IDS3* genes increased iron content in grains of polished rice (Masuda *et al.*, 2008, 2009; Suzuki *et al.*, 2008). This seed specific expression of PS (chelation based) strategy provided double iron concentration in biofortified rice.

Zinc Biofortified Rice

The zinc-regulated, iron-regulated transporter-like proteins (ZIP) family protein in rice facilitate Zn and Fe uptake and homeostasis. Overexpressing the ZIP family genes *OsIRT* and *MxIRT* elevated zinc and iron concentration in rice grain (Lee and An, 2009; Tan *et al.*, 2015). Zinc accumulation in seeds was increased by using a combination of *AtIRT1*, *AtNAS1* with *Pvferritin* gene expression in biofortified rice (Boonyaves *et al.*, 2017). Approaches for rice iron biofortification also gave positive results of zinc accumulation in seeds.

Strategies of expressing the *Osfer2* gene (Paul *et al.*, 2012), and silencing of important genes of the phytic acid pathway- *MIPS* (Ali *et al.*, 2013a), *IPK1* (Ali *et al.*, 2013b), and *ITPK* genes (Karmakar *et al.*, 2020; Sengupta *et al.*, 2021) in rice was accompanied by increased zinc content in the seeds along with iron.

High Oleic Rice

Oleic acid (18:1, monounsaturated) is one of the fatty acids found in rice grains that has many health benefits. It helps maintain healthy blood pressure, prevent heart attack, stroke and other cardiovascular diseases (Lopez–Huertas, 2010). Four fatty acid desaturase 2 (FAD2) genes – *OsFAD2-1*, *OsFAD2-2*, *OsFAD2-3*, and *OsFAD2-4* have been identified in rice. *OsFAD2-1* is directly involved in oleic acid to linoleic acid conversion (Zaplin *et al.*, 2013). Down regulation of the *OsFAD2-1* gene can help maintain a higher amount of oleic acid in rice by preventing conversion to linoleic acid. CRISPR/Cas9 knockout *OsFAD2-1* gene from Nipponbare rice confirmed this hypothesis as the concentration of oleic acid in rice seeds surged by twofold (Abe *et al.*, 2018). Rice bran oil (RBO) made from such CRISPR-ed (knockout *OsFAD2-1*) rice cultivars can have high commercialization potential as RBO is favoured by many Asian households as ‘healthy cooking oil’.

Wheat Biofortification

After rice, wheat is the second most popular and produced cereal crop in the world (Fig. 1D). It is a staple food for around 30% of the world population (Lobell *et al.*, 2011). Agronomic, genetic approaches and available genomic resources for wheat biofortification have been recently reviewed in multiple articles (Ludwig and Slamet-Loedin 2019; Saini *et al.*, 2020; Ali and Borrill, 2020). Research on wheat biofortification is limited compared to rice.

Provitamin-A biofortification was achieved in wheat, by maize *PSY* and bacterial (*E. uredothora*) *CRT-I* gene introduction into an elite wheat variety EM12 (Cong *et al.*, 2009). In another report provitamin-A content increased 76-fold upon expressing the bacterial *CRT-B* and *CRT-I* genes in Bobwhite wheat (Wang *et al.*, 2014). When the *OsNAS2* gene was made to overexpress under a maize ubiquitin promoter in wheat, the iron content increased by 2.1-fold and zinc content by 3.7-fold in the grains (Singh *et al.*, 2017). Similar results were reported in another study where overexpression of the *OsNAS2* gene in wheat increased iron content by 1.4-

fold (Beasley *et al.*, 2019). Seed specific expression of the soybean *ferritin* gene in wheat resulted in 40 µg/g of iron accumulation in the wheat foliage (Drakakaki *et al.*, 2000) and 44.5 µg/g surged iron in the biofortified wheat endosperm (Borg *et al.*, 2012). A vacuolar-iron transporter- *TaVIT2* gene was expressed under a seed-specific promoter, which resulted in a two-fold increase in iron content than its control (Connorton *et al.*, 2017).

Maize Biofortification

Provitamin-A (β-carotene) biofortification in maize through transgenic approaches was successfully implemented. The bacterial *CRT-B* and *CRT-I* genes, expressed in maize, surged up 34-fold (9.8 µg/g) more β-carotene in the endosperm than control seeds (Aluru *et al.*, 2008). In another study, corn *PSY-I* and the bacterial (*E. uredothora*) *CRT-I* genes were introduced in M37W maize variety under wheat glutenin promoter and barley D-hordein promoter, respectively that accumulated 60 µg/g β-carotene in biofortified maize (Naqvi *et al.*, 2009). Updates on maize biofortification for provitamin-A, zinc, vitamin-E, high protein through conventional and molecular breeding, and country wise commercial availability of such biofortified maize varieties have been recently reviewed by Prasanna *et al.* (2020).

Cassava (*Manihot esculenta*) Biofortification

More than 800 million people in the world consume cassava and the sub-Saharan African population gets 50% of the daily calories from it (Howeler *et al.*, 2013). Biofortification for iron, zinc, provitamin-A have been achieved in cassava. For iron biofortified cassava development algal iron assimilatory protein FEA1 has been introduced into cassava and resulted in a threefold increase in iron content (Ihemere *et al.*, 2012). When the *Arabidopsis* vacuolar iron transporter *VIT1* gene was transferred into cassava it gave four-times increase in iron accumulation in the root (Narayanan *et al.*, 2015). Combined expression of three genes – *AtIRT1* (iron transporter), *FER1* (ferritin) and *VIT1* resulted in 18-times higher iron accumulation and 10-times higher zinc accumulation in the biofortified cassava (Narayanan *et al.*, 2019). For zinc biofortification, *Arabidopsis* zinc transporter gene *AtZIP1* and *AtMTP1* were transferred into cassava (Gaitán-Solís *et al.*, 2015). Carotenoid accumulation upto 6.67 µg/g in cassava has been achieved by expressing the bacterial *CRT-B* gene (Welsch *et al.*, 2010).

Legumes Biofortification

Legumes are an excellent source of nutrition, having high protein, minerals, dietary fibre, and complex carbohydrates. The amount of certain minerals, like zinc and calcium, is low in legumes (Wang *et al.*, 2003). Plant growth-promoting bacteria (PGPB) play a significant role in the biofortification of legumes (Roriz *et al.*, 2020). Iron content of mung beans (*Vigna radiata*) has been increased by 3.4-fold in association with the strain of *Pantoea dispersa* (Patel *et al.*, 2018). The lysine content of pigeon pea (*Cajanus cajan*) was increased significantly in the transgenic line (Thu *et al.*, 2007). Genome-editing technology has been used successfully in several legume crops for biofortification (Bhowmik *et al.*, 2021). Fatty acid and isoflavone content of soybean was increased by CRISPR-Cas9 mediated genome editing technology (Zhang *et al.*, 2020). Amino acid and proteins have been increased by traditional breeding in mungbean (Abbas *et al.*, 2019).

Conclusion

Nutritional security can be achieved sustainably through biofortified crops. The global catastrophe of losing human lives to VAD, IDA, and zinc deficiency and related health problems can be extensively reduced by including provitamin-A, high iron, and zinc biofortified cereals in the diet. Not only do malnourished people need biofortified crops, nutritional enriched foods are important for everyone. Nutrition is crucial for better immunity and to fight the ongoing COVID-19 pandemic that made it evident to the world the urgent necessity for nutritionally enriched, sustainable biofortified foods. World governments and policymakers must find a way to accelerate biofortified crops adoption specifically in those countries where hidden hunger is engulfing the vulnerable masses. The approval of Golden rice cultivation in the Philippines has inspired the world to shun taboos related to GMOs and accept the simplicity of scientific evidence for the noble cause of human wellbeing.

Acknowledgements

“Distinguished Biotechnology Research Professor Award” by the Department of Biotechnology (DBT), Government of India to Prof. Swapan K. Datta. “M. K. Bhan Young Researcher Fellowship Award” by DBT, Government of India to Dr. Shuvobrata Majumder. DBT and Indian Council of Agricultural Research (ICAR),

Government of India for financial support of research projects on rice biofortification and crop improvement at the University of Calcutta, Kolkata, India.

References

- Abbas G, M Ahsan, MJ Asghar, M Rizwan and M Hussain (2019) Genetic improvement in nutritional quality of mungbean through interspecific hybridization with mashbean. *ISST*. **43(5)**:2173-2184.
- Abe K, E Araki, Y Suzuki, S Toki and H Saika (2018) Production of high oleic/low linoleic rice by genome editing. *Plant Physiol. Biochem.* **131**:58–62.
- Ali N, S Paul, D Gayen, SN Sarkar, SK Datta and K Datta (2013a) RNAi mediated down regulation of myo-inositol-3-phosphate synthase to generate low phytate rice. *Rice* **6**:12.
- Ali N, S Paul, D Gayen, SN Sarkar, K Datta and SK Datta (2013b) Development of low phytate rice by RNAi mediated seed-specific silencing of *inositol 1,3,4,5,6-pentakisphosphate 2-kinase* gene (IPK1). *PLoS ONE* **8**:e68161.
- Ali, MW and P Borrill (2020) Applying genomic resources to accelerate wheat biofortification. *Heredity* **125**:386-395 doi:10.1038/s41437-020-0326-8
- Aluru M, Y Xu, R Guo, Z Wang, S Li, W White, K Wang and S Roderme (2008) Generation of transgenic maize with enhanced provitamin A content. *J. Exp. Bot.* **59**:3551–3562.
- Bai C, T Capell, J Berman, V Medina, G Sandmann, P Christou and C Zhu (2016) Bottlenecks in carotenoid biosynthesis and accumulation in rice endosperm are influenced by the precursor-product balance. *Plant Biotechnol. J.* **14**:195–205.
- Baisakh N, S Rehana, M Rai, N Oliva, J Tan, D Mackill, GS Khush, K Datta and SK Datta (2006) Marker-free transgenic (MFT) near-isogenic introgression lines (NILs) of ‘golden’ indica rice (cv IR64) with accumulation of provitamin A in the endosperm tissue. *Plant Biotechnol. J.* **4**:467–475.
- Beasley JT, JP Bonneau, JT Sánchez-Palacios, LT Moreno-Moyano, DL Callahan, E Tako *et al.* (2019) Metabolic engineering of bread wheat improves grain iron concentration and bioavailability. *Plant Biotech. J* **17**:1514–1526.
- Bhattacharya S, S Sengupta, A Karmakar, SN Sarkar, G Gangopadhyay, K Datta and SK Datta (2019) Genetically engineered rice with *appA* gene enhanced phosphorus and minerals. *J. Plant Biochem. Biotechnol.* **28**:470–482.
- Bhowmik P, D Konkin, P Polowick, CL Hodgins, M Subedi, D Xiang, B Yu, N Patterson, N Rajagopalan, V Babic and DK Ro (2021) CRISPR/Cas9 gene editing in legume crops: Opportunities and challenges. *Legume Sci.* **3(3)**:96.
- Boonyaves K, T-Y Wu, W Gruissem and NK Bhullar (2017) Enhanced grain iron levels in Rice expressing an iron-regulated metal transporter, nicotianamine synthase, and *ferritin* gene cassette. *Front. Plant. Sci.* **8**:130.
- Boonyaves K, W Gruissem and NK Bhullar (2016) NOD promoter-controlled *AtIRT1* expression functions synergistically with

- NAS and FERRITIN genes to increase iron in rice grains. *Plant. Mol. Biol.* **90**:207–215.
- Borg S, H Brinch-Pedersen, B Tauris, LH Madsen, B Darbani, S Noeparvar and PB Holm (2012) Wheat ferritins, Improving the iron content of the wheat grain. *J. Cereal Sci.* **56**:204–213.
- Burkhardt PK, P Beyer, J Wunn, A Kloti, GA Armstrong, M Schledz, J von-Lintig, and I Potrykus (1997) Transgenic rice (*Oryza sativa*) endosperm expressing daffodil (*Narcissus pseudonarcissus*) phytoene synthase accumulates phytoene, a key intermediate of provitamin A biosynthesis. *Plant J.* **11**:1071–1078.
- Colombo F, D Paolo, E Cominelli, F Sparvoli, E Nielsen and R Pilu (2020) MRP transporters and low phytic acid mutants in major crops: main pleiotropic effects and future perspectives. *Front. Plant Sci.* **11**:1301.
- Cong L, C Wang, L Chen, H Liu, G Yang and G He (2009) Expression of phytoene synthase1 and carotene desaturase *crtI* genes result in an increase in the total carotenoids content in transgenic elite wheat (*Triticum aestivum* L.), *J. Agric. Food Chem.* **57**:8652–8660.
- Connorton JM, ER Jones, I Rodríguez-Ramiro, S Fairweather-Tait, C Uauy and J Balk (2017) Wheat vacuolar iron transporter TaVIT2 transports Fe and Mn and is effective for biofortification. *Plant Physiol.* **174**:2434–2444.
- Datta K, G Sahoo, S Krishnan, M Ganguly and SK Datta (2014) Genetic stability developed for β -carotene synthesis in BR29 rice line using dihaploid homozygosity. *PLoS One* **9**:e100212.
- Datta K, M Rai, V Parkhi, N Oliva, J Tan and SK Datta (2006) Improved ‘golden’ indica rice and post-transgeneration enhancement of metabolic target products of carotenoids (β -carotene) in transgenic elite cultivars (IR64 and BR29). *Curr. Sci.* **91**:935–939.
- Datta K, N Baisakh, N Oliva, L Torrizo, E Abrigo, J Tan, M Rai, S Rehana, S Al-Babli, P Beyer, I Potrykus and SK Datta (2003) Bioengineered ‘golden’ indica rice cultivars with beta-carotene metabolism in the endosperm with hygromycin and mannose selection systems. *Plant Biotechnol. J.* **1**:81–90.
- Datta SK (1999) Transgenic Cereals: *Oryza sativa* (rice). In: I Vasil (ed) *Molecular improvement of cereal crops*. Springer, Dordrecht, pp 149–187.
- Datta SK and K Datta (2020) Golden Rice. In: A Costa de Oliveira, C Pegoraro and V Ebeling Viana (eds) *The Future of Rice Demand: Quality Beyond Productivity*. Springer, Cham. doi:10.1007/978-3-030-37510-2_6
- Datta SK, K Datta, V Parkhi, M Rai, N Baisakh, G Sahoo, S Rehana, A Bandyopadhyay, M Alamgir, MS Ali, E Abrigo, N Oliva and L Torrizo (2007) Golden Rice: introgression, breeding, and field evaluation. *Euphytica* **154**:271–278.
- Datta SK, S Majumder, K Datta (2021) Molecular Breeding for Improved β -carotene Synthesis in Golden Rice: Recent Progress and Future Perspectives. In: MA Hossain, L Hassan, K Md. Iftekharuddaula, A Kumar and R Henry. (eds) *Molecular Breeding for Rice Abiotic Stress Tolerance and Nutritional Quality*, First Edition. pp. 287-303.
- Drakakaki G, P Christou and E Stoger (2000) Constitutive expression of soybean ferritin cDNA in transgenic results in increased iron levels in vegetative tissues but not in seeds. *Transgenic Res.* **9**:445–452.
- Feng X and KT Yoshida (2004) Molecular approaches for producing low-phytic-acid grains in rice. *Plant Biotechnol.* **21**:183–189.
- Fiaz S, S Ahmad, MA Noor, MA Noor, X Wang, A Younas, A Riaz, A Riaz and F Ali, (2019) Applications of the CRISPR/Cas9 system for rice grain quality improvement: Perspectives and opportunities. *Int. J. Mol. Sci.* **20**: 888.
- Food and Agriculture Organization FAO; International Fund for Agricultural Development IFAD; World Food Programme WFP. *The State of Food Insecurity in the World 2015*; Food and Agriculture Organization of the United Nations: Rome, Italy, 2015.
- Gaitán-Solis E, NJ Taylor, D Siritunga, W Stevens and DP Schachtman (2015) Overexpression of the transporters AtZIP1 and AtMTP1 in cassava changes zinc accumulation and partitioning. *Front. Plant Sci.* **6**:492.
- Gayen D, N Ali, M Ganguly, S Paul, K Datta and SK Datta (2014) RNAi mediated silencing of *lipoxygenase* gene to maintain rice grain quality and viability during storage. *Plant Cell Tiss. Org. Cult.* **118**:229–243.
- Gayen D, N Ali, SN Sarkar, SK Datta, and K Datta (2015) Down regulation of *lipoxygenase* gene reduces degradation of carotenoids of golden rice during storage. *Planta* **252**:353–363.
- Gayen D, S Ghosh, S Paul, SN Sarkar, SK Datta and K Datta (2016) Metabolic regulation of carotenoid-enriched golden rice line. *Front. Plant Sci.* **7**:1622.
- Ghosh S, K Datta and SK Datta (2019) Rice vitamins. In: Bao J (ed) *Rice, chemistry and technology*, 4th edn. Elsevier, Rice, pp 195–220. doi:10.1016/978-0-12-811508-4.00007-1
- Goto F, T Yoshihara, N Shigemoto, S Toki, and F Takaiwa (1999) Iron fortification of rice seed by the soybean *ferritin* gene. *Nat. Biotechnol.* **17**:282–286.
- Hoa TTC, S Al-Babli, P Schaub, I Potrykus and P Bayer (2003) Golden indica and japonica rice lines amenable to deregulation. *Plant. Physiol.* **133**:161–169.
- Howeler R, N Lutaladio and G Thomas (2013) *Save and Grow: Cassava. A Guide to Sustainable Production Intensification* (Food and Agriculture Organization of the United Nations, Rome, 2013).
- Hsieh H, W Liu and PC Huang (1995) A novel stress-inducible metallothionein-like gene from rice. *Plant Mol. Biol.* **28**:381–389.
- Huguchi K, K Suzuki, H Nakanishi, H Yamaguchi, NK Nishizawa and S Mori (1999) Cloning of *nicotamine synthase* genes, novel genes involved in the synthesis of phytoalexins. *Plant Physiol.* **119**:471–479.

- Ithemerec UE, Narayanan NN and Sayre RT (2012) Iron biofortification and homeostasis in transgenic cassava roots expressing the algal iron assimilatory gene, *FEAI*. *Front. Plant Sci.* **3**:171.
- Jha AB and TD Warkentin (2020) Biofortification of Pulse Crops: Status and Future Perspectives. *Plants* **9**:73 doi:10.3390/plants9010073
- Johnson AAT, B Kyriacou, DL Callahan, L Carruthers, J Stangoulis, E Lombi and M Tester (2011) Constitutive overexpression of the *OsNAS* gene family reveals single-gene strategies for effective iron- and zinc-biofortification of Rice endosperm. *PLoS ONE* **6**:e24476.
- Karmakar A, S Bhattacharya, S Sengupta, N Ali, SN Sarkar, K Datta and SK Datta (2020) RNAi-mediated silencing of *ITPK* gene reduces phytic acid content, alters transcripts of phytic acid biosynthetic genes, and modulates mineral distribution in rice seeds. *Rice Sci* **27**(4): 315–328.
- Khalekuzzaman M, K Datta, N Oliva, M Alam and S Datta (2006) Stable integration, expression and inheritance of the *ferritin* gene in transgenic elite indica rice cultivar BR29 with enhanced iron level in the endosperm. *Indian J. Biotechnol.* **5**:26–31.
- Khush GS (2001) Green revolution: the way forward. *Nat. Rev. Genet.* **2**:815–821.
- Kok AD, LL Yoon, R Sekeli, WC Yeong, ZNB Yusof and LK Song (2018) Iron biofortification of rice: Progress and prospects. In: F Saha et al. (ed) *Rice Crop—Current Developments*, Intech Open: London, UK. pp. 25–44.
- Kuwano M, A Ohshima, Y Tanaka, T Mimura, F Takaiwa and KT Yoshida (2006) Molecular breeding for transgenic rice with low-phytic acid phenotype through manipulating myo-inositol 3 phosphate synthase gene. *Mol. Breeding.* **18**:263–272.
- Kuwano M, T Mimura, F Takaiwa and Yoshida KT (2009) Generation of stable ‘low phytic acid’ transgenic rice through antisense repression of the *1 D-myoinositol 3-phosphate synthase* gene using the 18-kDa oleosin promoter. *Plant Biotechnol. J.* **7**:96–105.
- Lee S and G An (2009) Over-expression of *OsIRT1* leads to increased iron and zinc accumulations in rice. *Plant Cell Environ.* **32**: 408–416.
- Lee S, US Jeon, SL Lee, YK Kim, DP Persson, S Husted, JK Schnorring, Y Kakei, H Masuda, NK Nishizawa et al. (2009) Iron fortification of rice seeds through activation of the *nicotianamine synthase* gene. *Proc. Natl. Acad. Sci. USA* **106**:22014–22019.
- Lee S, YS Kim, US Jeon, YK Kim, JK Schjoerring and G An (2012) Activation of rice *nicotianamine synthase 2 (OsNAS2)* enhances iron availability for biofortification. *Mol. Cells* **33**: 269–275.
- Lobell DB, W Schlenker and J Costa-Roberts (2011) Climate trends and global crop production since 1980. *Science* **333**:616–620.
- Lopez-Huertas E (2010) Health effects of oleic acid and long chain omega-3 fatty acids (EPA and DHA) enriched milks. A review of intervention studies. *Pharmacol. Res.* **61**:200–207.
- Lucca P, R Hurrell and I Potrykus (2001) Genetic engineering approaches to improve the bioavailability and the level of iron in rice grains. *Theor. Appl. Genet.* **102**:392–397.
- Lucca P, R Hurrell and I Potrykus (2002) Fighting iron deficiency anemia with iron-rich rice. *J. Am. Coll. Nutr.* **21**:184S–190S.
- Ludwig Y, and IH Slamet-Loedin (2019) Genetic biofortification to enrich rice and wheat grain iron: from genes to product. *Front. Plant. Sci.* **10**:833.
- Majumder S, K Datta and SK Datta (2019) Rice biofortification: high iron, zinc, and vitamin-A to fight against “hidden hunger”. *Agronomy* **9**:803.
- Majumder S, K Datta and SK Datta (2022) Transgenics for biofortification with special reference to rice. In: S Kumar, HK Dikshit, GP Mishra, A Singh (eds) *Biofortification of Staple Crops*. Springer, Singapore. pp. 439–460. doi:10.1007/978-981-16-3280-8_17
- Marschner H and V Romheld (1994) Strategies for plants for acquisition of iron. *Plant Soil* **165**:261–274.
- Masuda H, K Usuda, T Kobayashi, Y Ishimaru, Y Kakei, M Takahashi, K Higuchi, H Nakanishi, S Mori and NK Nishizawa (2009) Overexpression of the barley *nicotianamine synthase* gene *HvNAS1* increases iron and zinc concentrations in rice grains. *Rice* **2**:155–166.
- Masuda H, M Suzuki, KC Morikawa, T Kobayashi, H Nakanishi, M Takahashi, M Saigusa, S Mori and NK Nishizawa (2008) Increase in iron and zinc concentrations in rice grains via the introduction of barley genes involved in phytosiderophore synthesis. *Rice* **1**:100–108.
- Mishra GP, HK Dikshi, Priti, B Kukreja, M Aski, DK Yadava, A Sarker, and S Kumar (2022). Historical Overview of Biofortification in Crop Plants and Its Implications. In: S Kumar, HK Dikshit, GP Mishra, and A Singh (eds) *Biofortification of Staple Crops*. Springer, Singapore. doi:10.1007/978-981-16-3280-8_2
- Mishra R, Joshi RK and Zhao K (2018) Genome editing in rice: recent advances, challenges, and future implications. *Front. Plant Sci.* **9**:1361.
- Naqvi S, C Zhu, G Farre, K Ramessar, L Bassie, J Breitenbach, D Conesa, G Ros, G Sandmann, T Capell, and P Christou (2009) Transgenic multivitamin corn through biofortification of endosperm with three vitamins representing three distinct metabolic pathways. *Proc. Natl. Acad. Sci. USA* **106**:7762–7767.
- Narayanan N, G Beyene, RD Chauhan, E Gaitán-Solís, J Gehan, P Butts, et al. (2019) Biofortification of field-grown cassava by engineering expression of an iron transporter and ferritin. *Nat. Biotech.* **37**:144–151.
- Narayanan N, G Beyene, RD Chauhan, E Gaitán-Solís, MA Grusak, N Taylor and P Anderson (2015) Overexpression of *Arabidopsis VIT1* increases accumulation of iron in cassava roots and stems. *Plant Sci.* **240**:170–181.
- Nozoye T, S Nagasaka, T Kobayashi, M Takahashi, Y Sato, N Uozumi, H Nakanishi and NK Nishizawa (2011)

- Phytosiderophore efflux transporters are crucial for iron acquisition in graminaceous plants. *J. Biol. Chem.* **286**:5446–5454.
- Oliva N, P Chadha-Mohanty, S Poletti, E Abrigo, G Atienza, L Torrizo, R Garcia, C Duenas Jr, MA Poncio, J Balindong et al. (2014) Large-scale production and evaluation of marker-free indica rice IR64 expressing *phytoferritin* genes. *Mol. Breed.* **33**:23–37.
- Owens B (2018) Golden Rice is safe to eat, says FDA. *Nat. Biotechnol.* **36**:559–560.
- Paine JA, CA Shipton, S Chagger, RM Howles, MJ Kennedy, G Vernon, SY Wright, E Hinchliffe, JL Adams, AL Silverstone, et al (2005) Improving the nutritional value of Golden rice through increased pro-vitamin A content. *Nat. Biotechnol.* **23**:482–487.
- Parkhi V, M Rai, J Tan, N Oliva, S Rehana, A Bandyopadhyay, L Torrizo, V Ghole, K Datta and SK Datta (2005) Molecular characterization of marker free transgenic indica rice lines that accumulate carotenoids in seed endosperm. *Mol. Genet. Genomics* **274**:e325–e336.
- Patel P, Trivedi G and Saraf M (2018) Iron biofortification in mungbean using siderophore producing plant growth promoting bacteria. *Environ. Sustain.* **1**:357–365 doi:10.1007/s42398-018-00031-3
- Paul S, N Ali, D Gayen, SK Datta and K Datta (2012) Molecular breeding of *Oser2* gene to increase iron nutrition in rice grain. *GM Crops Food* **3**: 310–316.
- Paul S, N Ali, SK Datta and K Datta (2014) Development of an iron-enriched high-yielding Indica rice cultivar by introgression of a high-iron trait from transgenic iron-biofortified rice. *Plant Foods Hum. Nutr.* **69**:203–208.
- Prasanna BM, Palacios-Rojas N, Hossain F, Muthusamy V, Menkir A, Dhliwayo T, Ndhlela T, San Vicente F, Nair SK, Vivek BS, Zhang X, Olsen M and Fan X (2020) Molecular Breeding for Nutritionally Enriched Maize: Status and Prospects. *Front. Genet.* 10:1392. doi:10.3389/fgene.2019.01392
- Qu LQ, T Yoshihara, A Ooyama, F Goto, and F Takaiwa (2005) Iron accumulation does not parallel the high expression level of ferritin in transgenic rice seeds. *Planta* **222**:225–233.
- Rai M, K Datta, V Parkhi, J Tan, N Oliva, HS Chwla, SK Datta (2007) Variable T-DNA linkage configuration affects inheritance of carotenogenic transgenes and carotenoid accumulation in transgenic indica rice. *Plant Cell Rep.* **26**:1221–1231.
- Rehana S, N Baisakh, K Datta, N Oliva, E Abrigo, MA Mazil, SA Alam, MR Uddin and SK Datta (2018) Engineering Provitamin A synthesis pathway with β -carotene metabolism in rice endosperm of a restorer line BR827R. *Arc. Agri. Environ. Sci.* **3**(2):103–108.
- Romheld V and H Marschner (1990) Genotypical differences among graminaceous species in release of phytosiderophores and uptake of iron phytosiderophores. *Plant. Soil* **123**:147–153.
- Roriz M, SM Carvalho, PM Castro and Vasconcelos MW (2020). Legume biofortification and the role of plant growth-promoting bacteria in a sustainable agricultural era. *Agronomy*, **10**(3):435.
- Saini DK, P Devi and P Kaushik (2020) Advances in genomic interventions for wheat biofortification: a review. *Agronomy* **10**:62.
- Schmitz J, KP West Jr., SK Khattry, L Wu, SC Leclercq, et al. (2012) Vitamin A supplementation in preschool children and risk of hearing loss as adolescents and young adults in rural Nepal: Randomised trial cohort follow-up study. *BMJ* **344**:d7962.
- Sengupta S, S Bhattacharya, A Karmakar, S Ghosh, SN Sarkar, G Gangopadhyay, K Datta and SK Datta (2021) RNAi-mediated down-regulation of ITPK-2 enhanced inorganic phosphorus and minerals in the transgenic rice. *J. of Biosciences* **46** (2), 1-13.
- Singh SP, B Keller, W Gruissem and NK Bhullar (2017) Rice NICOTIANAMINE SYNTHASE 2 expression improves dietary iron and zinc levels in wheat. *Theor. Appl. Genet.* **130**:283–292.
- Suzuki M, KC Morikawa, H Nakanishi, M Takahashi, M Saigusa, S Mori and NK Nishizawa (2008) Transgenic rice lines that include barley genes have increased tolerance to low iron availability in a calcareous paddy soil. *J. Soil. Sci. Plant. Nutr.* **54**:77–85.
- Swamy BPM, M Samia, R Boncodin, et al. (2019). Compositional analysis of genetically engineered GR2E “golden rice” in comparison to that of conventional rice. *J. Agric. Food. Chem.* **67** (28): 7986–7994.
- Tan S, R Han, P Li, G Yang, S Li, P Zhang, WB Wang, WZ Zhao and LP Yin (2015) Over-expression of the *MxIRT1* gene increases iron and zinc content in rice seeds. *Transgenic Res.* **24**:109–122.
- Taylor PG, TC Martinez, EL Romano and M Layrisse (1986) The effect of cysteine-containing peptides released during meat digestion on iron absorption in humans. *Am. J. Clin. Nutr.* **43**:68–71.
- Thu TT, E Dewaele, LQ Trung, M Claeys, M Jacobs, and G Angenon (2007) Increasing lysine levels in pigeonpea (*Cajanus cajan* (L.) Millsp) seeds through genetic engineering. *Plant Cell Tissue Organ Cult.* **91**(2):135-143.
- Vasconcelos M, K Datta, N Oliva, M Khalekuzzaman, L Torrizo, S Krishnan, M Oliveira, F Goto and SK Datta (2003) Enhanced iron and zinc accumulation in transgenic rice with the *ferritin* gene. *Plant Sci.* **164**:371–378.
- Wang C, J Zeng, Y Li, W Hu, L Chen and Y Miao (2014) Enrichment of provitamin A content in wheat (*Triticum aestivum* L.) by introduction of the bacterial carotenoid biosynthetic genes *CrtB* and *CrtI*. *J. Exp. Bot.* **65**:545–556.
- Wang TL, C Domoney, CL Hedley, R Casey and MA Grusak (2003) Can we improve the nutritional quality of legume seeds? *Plant Physiol.* **131**(3):886-891.
- Welch RM (2002) Breeding strategies for biofortified staple plant foods to reduce micronutrient malnutrition globally. *J. Nutr.* **132**:495S–499S.
- Welsch R, J Arango, C Bar, B Salazar, S Al-Babili, J Beltran, P Chavarriaga, H Ceballos, J Tohme and P Beyer (2010)

- Provitamin A accumulation in cassava (*Manihot esculenta*) roots driven by a single nucleotide polymorphism in a *phytoene synthase* gene, *Plant Cell*. **22**:3348–3356.
- WHO (2015). The global prevalence of anaemia in 2011. Geneva: World Health Organization; 2015
- Wirth J, S Poletti, B Aeschlimann, N Yakandawala, B Drosse, S Osorio, T Tohge, AR Fernie, D Gunther, W Gruissem, et al. (2009) Rice endosperm iron biofortification by targeted and synergistic action of nicotianamine synthase and ferritin. *Plant Biotechnol. J.* **7**:631–644.
- Wu F, J Wesseler, D Zilberman, RM Russell, C Chen, and AD Dubock (2021) Allow Golden Rice to save lives. *PNAS* **118**(51):e2120901118 doi:10.1073/pnas.2120901118
- Ye X, S Al-Babili, A Klöti, J Zhang, P Lucca, P Beyer and I Potrykus (2000) Engineering the provitamin a (β -carotene) biosynthetic pathway into (carotenoid-free) rice endosperm. *Science* 287:303–305
- Zaplin ES, Q Liu, Z Li, VM Butardo, CL Blanchard and S Rahman (2013) Production of high oleic rice grains by suppressing the expression of the *OsFAD2-1* gene. *Funct. Plant Biol.* **40**:996–1004.
- Zhang P, H Du, J Wang, Y Pu, C Yang, R Yan, H Yang, H Cheng and D Yu (2020) Multiplex CRISPR/Cas9-mediated metabolic engineering increases soya bean isoflavone content and resistance to soya bean mosaic virus. *Plant biotechnol. J.* **18**(6):1384–1395.

Status of Genetic Resources in Oilseed Crops and Their Potential Use for Making India Atmanirbhar in Edible Oils

Rashmi Yadav¹, Partha Ray Choudhury², Anjali Kak Koul¹, Veena Gupta¹, Ashok Kumar¹ and Devendra K Yadava^{2*}

¹ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

²Indian Council of Agricultural Research, Pusa Campus, New Delhi-110012, India

Plant Genetic Resources play an important role in the genetic enhancement of different crops leading to food and nutritional security of the nation. India is genetic diversity rich country with more than 4.62 lakh accessions of 2065 crop species conserved National Gene Bank at National Bureau of Plant Genetic Resources, New Delhi. Oilseeds are integral part of the various crop breeding programmes under National Agricultural Research System. In case of nine edible oilseed crops grown in India 57977 accessions of 13 species are conserved in the National Gene. In general, it has been observed that the utilization of germplasm conserved in the National Gene Bank is very low. Characterization of the germplasm and identification of trait specific donors is pre-requisite for successful utilization in the various breeding programmes. In this direction efforts have been made by the Oilseed Breeders in the country and in addition to release and notification of 956 varieties of nine oilseed crops since 1969, 193 novel genetic stocks of different oilseed crops for different agro-morphological, biotic and abiotic stress tolerance, high seed meal and oil quality traits have been registered with NBPGR. In addition, a large number of exotic collections have also been procured and being utilized in breeding programmes and more trait specific germplasm of all the oilseed crops has been identified along with their source country. For Atmanirbharta in edible oils in our country, we need to follow two-pronged strategy i.e. horizontal expansion targeting more area under oilseed crops and vertical expansion by deploying high yielding varieties and adoption of good agricultural practices. In vertical expansion breeding high yielding varieties is the main component which can be achieved with the use of diverse germplasm available globally. Hence, characterization and wider use of trait specific diverse germplasm of oilseed crops will be of great help in making the country Atmanirbhar in edible oils.

Background

Oilseed crops are great gift of nature to mankind which are source of food (salad, oil, margarine, vanaspati, shortening, cooking, and bakery), feed and industrial raw material (pharmaceutical products, soap, paints and resins, coatings, linoleum, cosmetics, lubrication, chemicals, plastic coatings, and ethanol). In India total nine oilseed crops viz., soybean (*Glycine max* Merr.), groundnut (*Arachis hypogaea* L.), rapeseed mustard Brassica spp.), sunflower (*Helianthus annuus* L.), safflower (*Carthamus tinctorius* L.), sesame (*Sesamum indicum* L.), niger (*Guizotia abyssinica* Cass.), linseed (*Linum usitatissimum* L.) and castor (*Ricinus cummunis* L.) are being grown. Among these nine oilseed crops; soybean (35%), groundnut (26%), rapeseed & mustard (30%) contribute to more than 91% of total oilseeds production and has more than 80% share in indigenous edible oil with major share of mustard (35%), soybean (23%) and groundnut (25%). Other edible oilseeds like sesame, linseed, safflower and sunflower are constantly losing

their acreage, as well as their share in gross cropped area. In totality, oilseeds have registered continuous growth in area, production and productivity since 1950-51 which has about 2.70 times increase in area, 7.00 times increase in production and 2.60 times increase in productivity (Fig. 1). The major breakthrough in area, production and productivity was witnessed after implementation of Technology Mission in Oilseeds and pulses during 1985-86. However, the fluctuations in area and variation in yield is a major factor for production variability during all decades, which is an indication of risks associated with oilseeds.

During 2020-21, against the total demand of 23.46 million tons of edible oils in India; 8.97 million tons was met through domestic production and 14.60 million tons through imports which is attributed to increasing population, enhanced per capita consumption and low productivity. The low productivity of oilseeds is due to many reasons including cultivation of these crops on low and marginal lands, impact of climate change

*Author for Correspondence: Email-adgseed.icar@nic.in

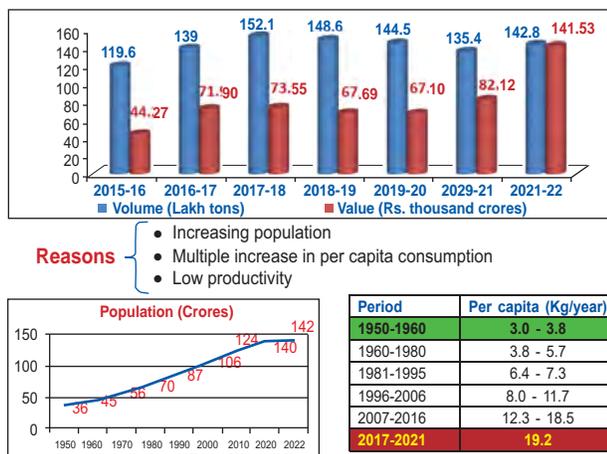


Fig. 1. Ever increasing edible oil imports: A challenge

like recurring drought and water logging, temperature fluctuations, frost and various emerging biotic stresses in the form of diseases, insect-pests and weeds. In comparison to average global productivity and countries with highest productivity, except castor we are far behind (Fig. 2).

It clearly indicates that ample scopes exist for genetic enhancement of different edible oilseeds crops for making the country Atmanirbhar in edible oils which can be achieved by breeding resistant/ tolerant varieties for the various biotic and abiotic stresses with globally competitive enhanced seed yield, oil content and quality of seed meal and oil is the need of the hour to reduce the constantly increasing import bills on edible oils. Hence, serious research efforts are required to enhance the production and productivity using latest breeding tools to mine and utilize the germplasm with desirable traits.

Present Status

Plant genetic resources (PGR) play an important role as these are the basic constituent in any varietal development programme using naturally occurring variability for different agro-morphological traits. For strengthening genetic resources management; evaluation, conservation and documentation of germplasm, exchange under appropriate quarantine measures and distribution of germplasm for utilization as well as medium- and long-term conservation of valuable germplasm in National Gene Bank for posterity of mankind are essential activities. Characterization and evaluation are important activities under plant genetic resources programmes. The genetic resources can be broadly classified into landraces,

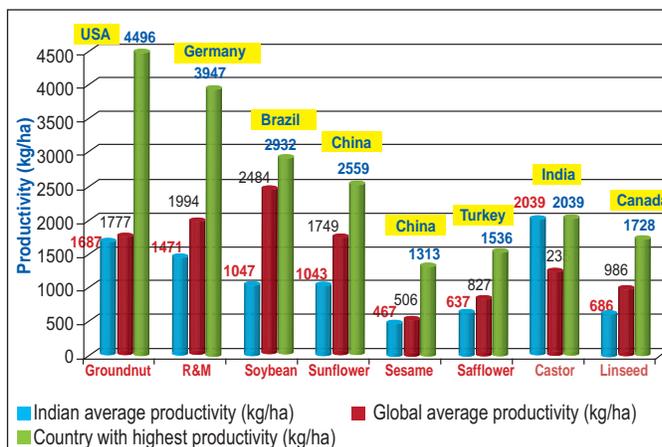


Fig. 2. Comparative productivity status of Oilseeds – India World and Country with highest productivity

wild relatives of crop species, genetic and cytogenetic stocks, breeding lines, modern cultivars and mapping populations which provide basic raw materials to crop improvement programmes and act as reservoir of genes for resistance to various biotic and abiotic stresses. Over 462,606 accessions of various crop plants belonging to 2065 species are currently stored in the repository (as on 31.07.2022), of which 57977 accessions belong to different oilseed crops representing 13 genera and 77 species (Table 1).

In comparison to the total germplasm conserved in the Nation Gene Bank, use of germplasm by breeders is inadequate. For the use of conserved germplasm and efficient utilization of available genetic diversity; large-scale systematic evaluation is generally a pre-requisite. However, except a few crops, the information on the

Table 1. Status of oilseed germplasm base collections in National Genebank (-18°C)

Crop	No. of Species	No. of Accessions
Groundnut (<i>Arachis speices</i>)	20	13514
Brassica species	20	11470
Taramira (<i>Eureca species</i>)	2	389
Crambe/ abyssinian mustard (<i>Crambe abyssinica</i>)	1	20
Garden Cress (<i>Lepidium sativum</i>)	1	73
Safflower (<i>Carthamus species</i>)	4	7014
Sesame (<i>Sesamum species</i>)	10	10121
Soybean (<i>Glycine species</i>)	3	5195
Sunflower (<i>Helianthus species</i>)	10	1974
Linseed (<i>Linum species</i>)	3	2833
Castor (<i>Ricinus communis</i>)	1	2672
Niger (<i>Guizotia abyssinica</i>)	1	2378
Perilla (<i>Perilla frutescens</i>)	1	324
Total	77	57977

Table 2. Oilseeds crop varieties released (1969-2021) and germplasm registered (till 2021) in National Gene Bank at NBPGR

Crop	Trait specific germplasm (Donors) registered till 2021						Total trait specific germplasm	Varieties released
	Abiotic	Biotic	Breeding value	Agronomic	Nutritional	Quality		
Soybean		1		4	4	5	14	160
Brassica	19	13	17	7	14	8	78	218
Groundnut	2	20	6	13	3	14	58	211
Sunflower		2	2	2		1	7	65
Sesame			1	3	1		5	92
Safflower	1	1	2	1	1	1	7	46
Niger								24
Linseed		1	1	2	2	1	7	88
Castor	2	19	12	2	1	1	37	52
Total	14	57	41	34	26	31	193	956

sources of resistant donors from germplasm conserved in the National Gene Bank at NBPGR, New Delhi and their genetic diversity status based on agronomic traits is also limited. Trait discovery and identification of novel traits is one of the important activities of NBPGR and commodity based ICAR Institutes and ICAR - All India Coordinated Research Projects Centres operating in Central and State Agricultural Universities. Total 193 novel genetic stocks of different oilseed crops, thus, developed and characterized for different agromorphological, biotic and abiotic stress tolerance, high seed meal and oil quality traits are registered by Plant Breeders and other researchers working in the National Agricultural Research System (NARS) with NBPGR for their use in future breeding programmes. In addition during 1969 to 2021, 956 improved crop varieties of different edible oilseed crops have also been released and notified for cultivation by the farmers (Table 2).

To expedite the process of characterization of available germplasm and for traits discovery, a Consortium Research Platform (CRP) on 'Agrobiodiversity' was launched by the Indian Council of Agricultural Research so that germplasm conserved at NBPGR could be effectively linked with enhanced use in crop improvement under National Agricultural Research System (NARS). Under this programme, Indian mustard was prioritized for detailed evaluation at different hotspots for different agro-morphological traits, biotic and abiotic stresses under field conditions and further validate the resistance under laboratory conditions so that the accessions carrying resistance against diseases along with agronomic superiority are identified for use in breeding for disease resistant and high yielding varieties in Indian mustard. Other than the

CRP, multi-location evaluation trails in many crops are also carried by ICAR-NBPGR and many trait specific germplasm lines are identified to register and share them as donors for various important traits. During past more than one-decade efforts have been made to identify abiotic stress tolerant germplasm in selected oilseed crops (Table 3).

Access to the exotic germplasm: In general, in most of the crop improvement programmes, it has been observed that use of exotic germplasm leads to higher heterosis as well as transgressive segregants, however some linkage drags are always a limiting factor which can be eliminated by various breeding and biotech tools. In India, during 2019-21, 4377 accessions of ten oilseed crops from USA, Netherlands, Argentina, Canada, Uganda, Niger, Netherlands, United Kingdom, Ethiopia, Kenya, Japan, Czech Republic, France, Switzerland, Australia were introduced based on the indents of public and private sector organizations for use in the different breeding programmes (Table 4).

Way Forward

To increase the total production of oilseeds for making India Atmanirbhar in edible oils, the two approaches are discussed widely viz., 1) Horizontal expansion i.e. Area Expansion under Oilseeds including searching new avenues, crop diversification, oilseeds in non-traditional areas and enhancing cropping intensity by including short duration varieties of oilseed crops; and 2) Vertical increase i.e. Improving Productivity of oilseed crops including breeding of high yielding hybrids/varieties using diverse germplasm and modern biotech tools like marker assisted selection, transgenics and gene editing; crop management and soil and moisture management.

Table 3. Promising germplasm lines of different oilseed crops germplasm identified during past one decade

Crop	Trait	Accessions
Rapeseed mustard	Salt tolerance	CS-52, CS-54, CS-234-2-2, CS-56, RH-8814 (IC-401570), CS-58, CS-1100-1-2-3-5-1, CS-1500-1-2-2-2-1, BPR-540-6
	Drought tolerance	RH-781, RH-819, RH-406, RB-50, RH-725, RVM-2, Pant Rai-20, RGN-298, Aravali, Geeta, Shivani, DRM-541-44
	Heat tolerance	Pusa Vijay (NPJ-93), Pusa Mustard 25 (NPJ-112), Pusa Mustard 27 (EJ-17), Pusa Mustard 28 (NPJ-124), BPR-540, BPR-541-4, BPR-543-2, BPR-549-9, Pant Rai-18, RH-406, RGN-229, RGN-236, RGN-298
	Frost tolerance	RH-819, RGN-48
Groundnut	Low and high temperature tolerance	NRCGs 14480, 14324, 14367, 14414, 14333, 14492, 14454 and released groundnut varieties viz., ALR 2, ALR 3, GJG 9, GG 13, Somnath, KRG 1, JGN 3, LGN 1, TAG 24, JL 220, Narayini, ICGV 00350, ICGV 87846, TG 1, TG 17
	Drought tolerance	NRCGs 14390, 14395, BG 2, ICGV 86590, Kadiri 3, M 197, DRG 12, BG 3, Kadiri 2, TG 1, DSG 1, GAUG 10
Soybean	Drought tolerance	JS 97-52, EC 538828, NRC 7, EC 602288, JS 71-05
	Water logging tolerance	JS 97-52, PK 472, JS 20-38
	Heat tolerance	JS 97-52, EC 538828, NRC 7
	Photo-insensitivity	MACS 330, EC 325097, EC 33897, EC 34101, EC 325197, EC 390977
	Long Juvenility	AGS 25
Sunflower	Drought tolerance	AKSF-42-1, M-1026, 298-R, GMU-351 and hybrids; Laxmi-225, CO-2, CSFH-12205, DRSH-1, KBSH-44
Castor	Drought tolerance	RG298, RG1437, RG1826

Table 4. Exotic germplasm introduced in India during 2019-21

Crop	Country	Number of accessions		
		Public sector	Private Sector	Total
Sunflower	USA, Netherlands, Argentina	12	60	72
Indian Mustard	Canada	364	364	728
Groundnut	Uganda, Niger	4		4
Crambe/ abyssinian mustard	USA	18		18
Linseed	Netherlands, USA, Canada, United Kingdom	220		220
Niger	Ethiopia, USA, Kenya	24		24
Safflower	USA	278		278
Sesame	USA, Kenya, Taiwan, Japan, Czech Republic	2378		2378
Taramira	USA	247		247
Wild Sesame	USA	3		3
Brassica	USA, France, Switzerland, Australia	157	65	222
Castor	USA		133	133
Total oilseeds		3755	622	4377

Genetic enhancement through use of diverse germplasm available in India and globally is one of the best options for genetic enhancement for yield and other traits and needs to be exploited systematically. The different exotic germplasm of oilseed crops and their crop wild relatives (CWR) with specific traits available globally have been identified by the commodity Institutes (Table 5), which need to be procured on priority for their use in yield enhancement *per se*, trait improvement and for development/ diversification of Cytoplasmic Male

Sterility systems for hybrid breeding programmes in various oilseed crops.

To fully utilize the available genetic resources of various oilseed crops, large scale screening is required under different agroclimatic zones to identify the potential lines for using them in breeding programme, as has been done in case of wheat. The promising lines for various traits identified under field screening should further be characterized under controlled stress conditions (phenomics/ phytotronics) and the molecular markers

Table 5. Trait specific germplasm and crop wild relatives of oilseed crops their availability globally

Crops	Country	Specific traits
Groundnut	China, USA, Korea, Brazil	High oil content ($\geq 57\%$), Resistant to diseases (<i>Aspergillus flavus</i> , bacterial wilt, aflatoxins, bacterial wilt, Sclerotinia blight, Pod rot, Leaf spot, web blotch, stem rot); and early maturing landraces (80-90 days)
Soybean	USA, Taiwan, Japan, Brazil, China	Drought, heat, salinity and waterlogging tolerance, mechanical damage tolerant, Near iso-genic lines for photoinsensitivity and long juvenility, high Protein (55-58%), Triple Null lox (1, 2 & 3), High oleic acid (80%)
Mustard	China, Canada, Australia	High yield (>4.0 t/ha), oil content ($>50\%$), oleic acid ($>60\%$), >20 seeds/ siliqua, Sclerotinia stem rot, white rust and Alternaria blight resistant lines, Orobanche tolerant lines
Sunflower	Russia, Serbia, Argentina, France	High seed yield (>60 g/plant) and high oil content ($>55\%$), dwarf and short duration, Powdery mildew and Alternaria resistant lines
Sesame	China, Ethiopia, Korea	Male-sterility, Drought resistance, Water logging tolerance, Determinate Disease resistance, Earliness
Lentil	Lebanon/ Morocco, Australia, Iran, USA	Mechanized harvesting, Herbicide tolerance, Early maturity, bold and small seed size, high protein and high biomass, tolerance to pod dehiscence and number of pods/ plant, tolerant to heat, drought and boron, high grain Fe and Zn, phosphorus use efficient
Mungbean	Thailand, USA, Japan	Resistant to Powdery mildew resistance, MYMV and bruchid; sprout specific genotypes, pre harvest sprouting, bold seeded, long pods, 15-16 seeds/pod, high seed protein, mechanized harvesting, herbicide tolerance
Chickpea	Australia, USA	Super nodulating chickpea lines, Mechanical Harvesting lines, Salinity tolerant lines, Non nodulating Mutants, Ultra high protein chickpea lines
Pigeonpea	Kenya, Tanzania, Mzmbq, Malawi, Uganda,	Early duration, large seed size, long pod, high number of pods per plant, wilt resistance, SMD resistance and pod borer tolerance

linked to traits of interest should be identified for their use in precision breeding for introgression of these traits. More systematic efforts are required to collect, conserve and regenerate the germplasm, local landraces and wild relatives from the different agroclimatic zones of the country.

References

- Yadava D.K., Choudhury P.R. and Yadva Rashmi 2019. Genetic resources of oilseeds crops adaptable to climate change: Issues and way forward. Presented during Satellite Symposium on Dryland Agrobiodiversity for adaptation to climate change. Organised by ISPGR, New Delhi, Bioversity International, New Delhi and APAARI, Bangkok at Jodhpur during on February 13, 2019. P 54-56.
- Yadava D.K., Sujata Vasudev., Singh Naveen, Mohapatra T. and Prabhu K.V. 2011. Breeding major oilcrops: Present status and future research needs. Technical innovations in major Oilseed crops, Vol. 1 Breeding (Ed. S.K. Gupta). Springer. pp 17-52.
- Yadava D.K., Yadav Rashmi, Vishwakarma Harinder, Yashpal, Yadav Sangita, Saini Navinder and Vasudev Sujata. 2021. Genetic diversity characterization and population structure in *Brassica juncea*. In *The Brassica juncea* genome (Eds. Chittaranjan Kole and Trilochan Mohapatra) Springer, Singapore. pp. 73-84.
- Yadava Devendra Kumar, Yashpal, Saini Navinder, Nanjundan Joghee and Vasudev Sujata. 2022. *Brassica* Breeding. In *Fundamentals of Field Crop Breeding* (Eds. Yadava et. al.) Springer, Singapore. pp 779-835.
- <http://agricoop.nic.in/faq.html>
- <http://seednet.gov.in/>
- http://www.nbpgr.ernet.in/PGR_Databases.aspx

International Year of Millets 2023: Opportunity for Enhancing the Use of Indian Millets Germplasm

M Elangovan^{1*}, Karnam Venkatesh¹, Sushil Pandey² and Chitra Devi Pandey²

¹ICAR-Indian Institute of Millets Research (IIMR), Hyderabad-500030, Telangana, India

²ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

The global as well as Indian agriculture has witnessed tremendous growth in food production in the recent decades to meet the growing population, however it is also facing challenges such as climate change and malnutrition. Millets provide a great opportunity due to their climate resilient hardy nature and can be chosen to replace the conventional crops in the dry and marginal areas. In addition, millets are also rich in several vital micronutrients and vitamins necessary for reducing malnutrition and provide multiple health benefits. The National Genebank at ICAR-NBPGR, New Delhi and Millets Genebank at ICAR-IIMR, Hyderabad conserves approximately one lakh accessions of millets germplasm which can be of great use in identification and development of improved millets cultivars. This article enlists the different aspects of importance of millets in reducing the risk of climate change and malnutrition, SWOT analysis for utilization of millets germplasm resources, future prospects and action points for greater and efficient use of millet germplasm in India.

Introduction

The global as well as Indian agriculture has witnessed tremendous growth in food production in the recent decades, however it is also facing challenges such as climate change and malnutrition (Sharma *et al.*, 2015; Kumar *et al.*, 2018). The nation has witnessed over exploitation of irrigated agriculture lands in the past however there is a need to change the focus towards dry and marginal lands to address the adverse effects of ongoing and future climate change scenario. Generally, the lower fertility of dry and marginal lands makes it difficult to achieve higher production from conventional crops such as rice and wheat. Millets provide a great opportunity due to their climate resilient hardy nature and can be chosen to replace the conventional crops in the dry and marginal areas. In addition to their climate resilience, millets are also rich in several vital micronutrients and vitamins necessary for reducing malnutrition (Hariprasanna *et al.*, 2014; Elangovan *et al.*, 2022). Millets also known as nutri-cereals are reported to be the treasure house of vitamins, minerals, essential fatty acids, phyto-chemicals and antioxidants that can help to eradicate the hidden hunger. Due to the richness of millets in polyphenols and other biological active compounds, they are also considered to impart role in lowering rate of fat absorption, slow release of

sugars (low glycemic index) and thus reducing risk of heart disease, diabetes and high blood pressure (Kumar *et al.*, 2018).

The major millets are Pearl millet (*Pennisetum glaucum*, with synonyms of *P. americanum*, *P. typhoides*, and *P. typhoideum*), Foxtail millet (*Setaria italica*), Proso millet or white millet (*Panicum miliaceum*), and Finger millet (*Eleusine coracana*). Minor millets include Barnyard millet (*Echinochloa spp.*), Kodo millet (*Paspalum scrobiculatum*), Little millet (*Panicum sumatrense*) and Browntop millet (*Urochloa ramosa/Brachiaria ramosa/Panicum ramosum*) (Upadhyaya *et al.*, 2006).

Millets are Climate Resilient Crops

- Pearl millet and finger millet can grow up to a soil salinity of 11–12 dS/m
- The rainfall requirement of pearl millet and proso millet is as low as 20 cm, which is several folds lower than the rice
- Most of the millets mature in 60–90 days after sowing which makes them a water saving crop
- Millets fall under the group of C4 cereals, have better photosynthetic efficiency, water use efficiency compared to C3 crops

*Author for Correspondence: Email-elangovan@millets.res.in

Nutritional Profile of Millets

Millets being rich in gluten-free proteins, high fibre content, low glycemic index and bioactive compounds have made them a suitable health food

- **Carbohydrates:** The average carbohydrates content of millets varies from 56.88 to 72.97 g/100 g
- **Protein:** The protein content of all the millets is comparable to each other with an average protein content of 10 to 11%, except finger millet, which contain protein in the range of 4.76 to 11.70 g/100 g (Kumar *et al.*, 2018). Finger millet protein is rich in essential amino acids like methionine, valine and lysine, and of the total amino acids present, 44.7% are essential amino acids. Proso millet has the highest protein content among millets and the amount of essential amino acids are higher compared to conventional cereals
- **Fibres:** Millets are richest source of fibres, i.e. crude fibre as well as dietary fibre. Barnyard millet is the richest source of crude fibre with an average content of 12.8 g/100 g. The highest dietary fibre content, i.e. 38% and 37%, has been reported to be in little millet and kodo millet respectively
- **Calcium:** Calcium content of finger millet is about eight times higher than wheat and being the richest source of calcium (348 mg/100 g) it has the ability to prevent osteoporosis
- **Iron:** Barnyard millet and pearl millet are the rich source of iron, and their consumption can meet the iron requirement of pregnant women suffering from anaemia. The iron content of barnyard millet is 17.47 mg/100 g which is only 10 mg lower than the required daily value.
- **Zinc:** Foxtail millet contains highest content of zinc (4.1 mg/100 g) among all millets and is also a good source of iron (2.7 mg/100 g)
- **Thiamine:** The highest thiamine content in millets, i.e. 0.60 mg/100 g, is found in foxtail millet
- **Riboflavin:** Barnyard millet (4.20 mg/100 g) has the highest content of riboflavin followed by foxtail millet (1.65 mg/100 g) and pearl millet (1.48 mg/100g)

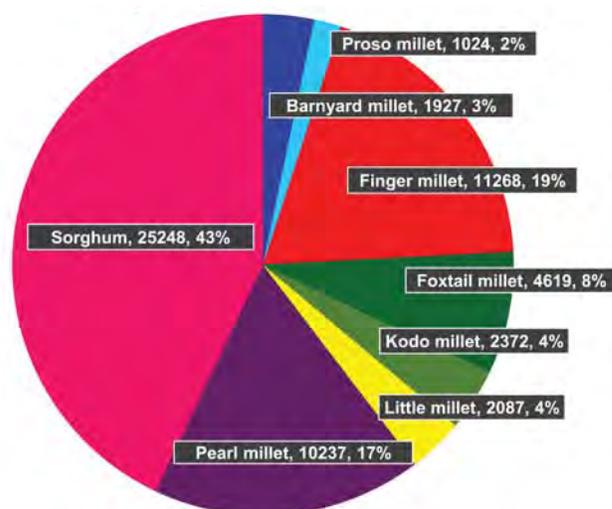
Status of Millets Germplasm holdings at Global, National and Institutional Genebanks

Vast collections of millets germplasm from 92 countries are conserved at ICAR-Indian Institute of Millets

Research (IIMR), Hyderabad; ICAR-National Bureau of Plant Genetic Resources (NBPGR), New Delhi and International Crops for Semi-Arid Tropics (ICRISAT), Patancheru genebanks. The status of millets genetic resources in the genebanks are given in Table 1.

Table 1. Status of millets genetic resources in the Genebank

Crop name	ICRISAT	NGB-ICAR-NBPGR	MGB-ICAR-IIMR
Sorghum (IS)	42,788	20,376	27,140
Pearl millet (IP)	24,390	7,841	4,128
Finger millet (IE)	7,519	11,587	7,806
Foxtail millet (ISc)	1,542	4,244	4,653
Kodo millet (IPs)	665	2362	344
Little millet (IPmr)	473	1885	694
Proso millet (IPm)	849	1005	2128
Barnyard millet (IEc)	749	1888	1705
Total	78,975	51,188	48,598



Status of millets germplasm in National Genebank

Contribution of Indian accessions to the global minicore collections

The Indian millet germplasm accessions contributed significantly to the global germplasm diversity which can indirectly be measured by assessing the proportion of Indian accessions in the global millet minicore collections (Upadhyaya *et al.*, 2011, 2019; Babu *et al.*, 2013). The contribution of Indian millet germplasm accessions to the global minicore collections varies from 6.6 % (7 acc.) in proso millet to 100 % (total 75 acc.) in kodo millet (Table 2).

Table 2. Contribution of Indian accessions to the global minicore collections

Crop	No. of acc. in Minicore	Indian accessions	% contribution by Indian origin acc.
Sorghum	242	30	12.4
Barnyard millet	89	49	55.1
Finger millet	80	17	21.3
Foxtail millet	35	12	34.3
Kodo millet	75	75	100.0
Little millet	57	55	96.5
Pearl millet	238	60	25.2
Proso millet	106	7	6.6

Millet Species Diversity and Variability

Millets species diversity is high in many Indian states viz., Uttar Pradesh (21), Odisha (16), Rajasthan (15), Bihar (14), Telangana and Himachal Pradesh (12), Chhattisgarh and Jammu & Kashmir (11), Jharkhand (10), Arunachal Pradesh (9), Punjab and Haryana (8), Kerala (6), West Bengal and Nagaland (5) etc. Tremendous genetic variability in the indigenous millets germplasm accessions has been recorded through characterization efforts carried out at ICAR-IIMR, ICAR-NBPGR, ICRISAT and AICRP centres under Sorghum, Pearl millet and Small millets programme.

Sorghum [*Sorghum bicolor* (L.) Moench]: Sorghum is being grown in two important seasons viz., *kharif* (rainy) and *rabi* (post-rainy) and season-specific Indian landraces are collected and conserved. Overall, the variability in Indian sorghum germplasm for days to 50 % flowering ranged between 41 and 141 days with a mean value of 75 days. Flowering is an important phenological trait helping the breeders in selecting varieties suited to different agro-ecological zones and climatic conditions. The plant height ranged between 30 and 480 cm with a mean of 213 cm. The variability in plant height provides opportunity for selection of germplasm suiting to different end uses like, grain, fodder, high biomass, dual purpose (grain and fodder) etc. The grain yield (g plant^{-1}) varied from 10.0 to 144.5 g with a mean of 30.2 g whereas, 100-seed weight is varying from 0.5 g to 6.2 g with an average value of 2.74 g.

Pearl millet [*Pennisetum glaucum* (L.) R. Br.]: Pearl millet in India is being grown in *kharif* and summer season in parts of Gujarat, Rajasthan and Uttar Pradesh and during the *rabi* season at a small scale in Maharashtra and Gujarat (Yadav and Rai, 2013). In general, the variability in the Indian pearl millet germplasm for days to 50 %

flowering varied from 33 to 159 days with a mean value of 72.7 days. The number of productive tillers attained by each plant ranged between 1 and 19 tillers with an average of 2.07 tillers per plant. The yield attributing traits like panicle length and width also recorded huge diversity. The panicle length recorded a minimum value of 5 cm and a maximum of 134 cm with a mean of 28.8 cm whereas the panicle width had a minimum of 8 cm to 58 cm with a mean of 23.9 cm. The plant height showed a minimum height of 30 cm and a maximum height of 490 cm with an average of 248 cm. Another yield attributing trait, 100-seed weight also has recorded huge variability between lowest of 0.15g and highest of 2.11 g per with an average value of 0.85 g.

Finger millet [*Eleusine coracana* (L.) Gaertn]: The finger millet is a major staple crop among tribal farmers and it can adapt to adverse climatic conditions, require minimal inputs and possess superior nutritional properties (Pradhan *et al.*, 2019). In general, the Indian finger millet variability for days to 50 % flowering ranged from 33 to 143 days with a mean of 87.7 days. The productive tillers varied from 1 to a maximum of 32 tillers per plant with an average of 9.2 tillers. The finger number on the main head ranged between 2.3 and 16.3 fingers with a mean value of 7.3 fingers per ear head. The individual finger length and width also showed tremendous diversity, the length of finger ranged between 2.5 and 19.5 cm with a mean of 7.17 cm while the width of the finger varied between 0.3 cm and 8.7 cm with a mean of 1.26 cm. The potential grain yield in Indian finger millet germplasm ranged between 1 g and 177.3 g per plant. The 100-seed weight also depicted a vast variability with a minimum value of 0.1 g and a maximum value of 1.35 g and a mean value of 0.25 g.

Foxtail millet [*Setaria italica* (L.) P. Beauv]: Foxtail millet is an important grain crop cultivated for food in some parts of China, India and Japan. The foxtail millet grains are a rich source of protein, minerals (calcium, iron, potassium, magnesium, and zinc) and vitamins (Upadhyaya *et al.*, 2011). The variability in the conserved germplasm accessions of Indian origin for days to 50 % flowering ranged between 34 and 99 days with a mean flowering days of 50.7 days. The number of basal tillers recorded the range of 1.3 to 27 tillers with a mean value of 6.4 tillers. The stem diameter ranged between 0.2 cm and 1.2 cm with an average of 0.4 cm. The yield attributing traits such as panicle length and panicle width showed tremendous variation. The panicle length ranged

between 3.4 cm and 37.8 cm with a mean of 17.9 cm and panicle width varied from 0.3 cm to 4.3 cm with a mean of 1.5 cm. The grain yield harvested from a single plant varied from a minimum of 1g to a maximum of 154.5 g with a mean of 25.2 g. The 100-seed weight of the foxtail millet grains showed variability between 0.03 g to 0.85 g with a mean of 0.26 g.

Proso millet (*Panicum miliaceum* L.): Proso millet is widely cultivated in India, China, Nepal, Africa, Russia, Ukraine, Belarus, Middle East, Turkey and Romania. The shallow root system (90–120 cm) and very short growing season (60–90 days) make it an ideal dryland crop and can fit into cropping system approach (Das *et al.*, 2019). In general, the variability of the Indian proso millet accessions for the number of basal tillers varied from 2.5 to 24 tillers per plant with a mean of 7.5 tillers. The days to 50 % flowering showed from very early flowering days of 38 days to very late flowering with 60 days and the mean of all the accessions was 48.2 days. The panicle length of the proso millet accessions varied between 12.4 cm and 46.7 cm with a mean of 25.9 cm while the panicle width ranged between 1.3 cm and 6.8 cm with an average value of 3 cm. The days to maturity of the proso millet accessions showed a diversity varying between 59 days and 89 days with a mean of 76 days. The grain yield harvested from each plant varied between 1 g to 18.4 g with an average harvest of 4.6 g from each plant. The 100 seed weight varied between 0.06 g and 0.8 g with a mean value of 0.4 g.

Barnyard millet (*Echinochloa frumentacea* Link): Barnyard millet is mainly grown in India, China, Japan, and Korea for human consumption as well as fodder (Upadhyaya *et al.*, 2016). The grains have superior nutritional properties including high micronutrients, dietary fiber content, and low glycemic index (GI) with potential health prospective (Upadhyaya *et al.*, 2016). In general, the variability in number of basal tillers ranged between 1 and 16.5 tillers per plant with a mean of 4.4 tillers. The days to 50 % flowering showed a minimum flowering day of 46 days as compared to the accession which showed delayed flowering in 86 days and the mean of all the accessions was 58 days. The panicle length of the barnyard millet accessions varied between 10.4 cm and 31.1 cm with a mean of 19.3 cm while the panicle width ranged between 1.5 cm and 6.8 cm with an average value of 3.7 cm. The plant height of the barnyard millet accessions showed a diversity varying between 60 cm and 291 cm with a mean of 122

cm. The grain yield harvested from each plant varied between 1.2 g and 37 g with an average harvest of 12 g from each plant. The 100-seed weight of the Indian barnyard millet accessions varied between 0.16 g and 0.47 g with a mean value of 0.27 g.

Little millet (*Panicum sumentranse* Roth): Little millet is largely cultivated throughout India by tribal people in small areas for food and fodder. It is a rich source of several micronutrients and vitamins. The variability present in the Indian little millet germplasm can be of great use for plant breeders. In general, the number of basal tillers among the accessions ranged between 3 and 46 tillers per plant with a mean tiller number of 14.6. The thickness of the culm varied from 2 mm to 18 mm with an average diameter of 6.6 mm. The minimum number of days required for the 50 % flowering was 39 days whereas the maximum days required for flowering were 138 days with a mean of 57.9 days. The panicle length was ranged between 27 mm and 500 mm with a mean of 282 mm. The width of the panicle was varying from 10 mm to 480 mm with an average width of 147.6 mm. The grain yield harvested from each plant varied between 2.95 g and 14.26 g with an average harvest of 7.37 g from each plant (Vetriventhan *et al.*, 2021). The 100-seed weight of the Indian little millet accessions varied between 0.14 g and 0.32 g with a mean value of 0.22 g.

SWOT Analysis of Utilization of Indian Millets Germplasm

Strength: The vast diversity of millets germplasm characterized and conserved in the National Genebank, Millets Genebank and AICRP Centres.

Weakness: Lack of data on multi-location evaluation of potential millets germplasm, nutritional characterization of nutri rich millets germplasm and difficult crop for breeding.

Opportunities: Awareness under International Year of Millets 2023 (IYoM) created a demand for value addition in millets and need to develop conventional breeding approaches

Threats: Millets cultivation area is decreasing due to occupation of commercial crops. Non-availability of processing machineries and minimum support price for the millets produce are the biggest concerns of the millet growing famers.

Kodo millet (*Paspalum scrobiculatum* L): Kodo millet is cultivated by tribal people across India for food and fodder. The Indian Kodo millet germplasm accessions depict a vast diversity for key traits of importance. In general, the accessions recorded a minimum of 2 basal tillers and maximum of 48 basal tillers per plant with a mean of 15.1. The days to 50% flowering ranged between 51 days and 112 days with a mean value of 77.3 days. The number of leaves ranged between 3 and 15 with a mean value of 5.7 leaves per plant. The plant height varied from 30 cm to 97 cm with an average height of 54.7 cm. The raceme number varied from 1 to 8 racemes per plant with a mean of 3 racemes. The grain yield harvested from each plant varied between 5.37 g and 31.37 g with an average harvest of 15.19 g from each plant (Nirubana *et al.*, 2017). The 100-seed weight of the Indian Kodo millet accessions varied between 0.24 g and 0.49 g with a mean value of 0.36 g.

Future Prospective and Action Points

- India being the primary origin for Little millet, Kodo millet and Barnyard millet; secondary origin for Sorghum. Finger millet being the major crop in the semi-arid regions, the adaptation and diversity of these millets germplasm are exclusive to India for some extent
- Re-introduction of millets local landraces in the region of collections to rejuvenate its natural habitat capability
- More emphasis on the characterization of millets for nutritional parameters to know the nutritional status of all available millets germplasm
- Evaluation of millets germplasm in all adverse agro-climatic conditions in the country to identify the best germplasm and supportive evidence for millets as climate resilient crop
- The CVRC should encourage breeders to release nutritional-specific millet varieties wherein nutrition is the primary trait followed by grain and fodder yield
- Millets demand is created through awareness campaign on the benefits of millets and diverse products by the start-ups in the markets. However, afford to buy millets by the poorer comes only through Public Distribution System. Need to diversify the plate of the PDS of the country with nutri-cereals.

References

- Babu TK, RP Thakur, HD Upadhyaya, PN Reddy, R Sharma, AG Girish and NDRK Sarma (2013) Resistance to blast (*Magnaporthe grisea*) in a mini-core collection of finger millet germplasm. *Eur. J. Plant Pathol.* **135**: 299-311.
- Das S, R Khound, M Santra and D Santra (2019) Beyond bird feed: Proso millet for human health and environment. *Agriculture* **9**: 64
- Hariprasanna K, V Agte, M Elangovan and J Patil (2014) Genetic variability for grain iron and zinc content in cultivars, breeding lines and selected germplasm accessions of sorghum [*Sorghum bicolor* (L.) Moench]. *Indian J. Genet. Plant Breed.* **74**: 42-49
- Kumar A, V Tomer, A Kaur, V Kumar and K Gupta (2018) Millets: a solution to agrarian and nutritional challenges. *Agric Food Security* **7**: 31.
- Maruthamuthu E, K Venkatesh, A Bellundagi, S Pandey and CD Pandey (2022) Assessment of variability in sorghum [*Sorghum bicolor* (L.) Moench] germplasm for agro-morphological traits. *Electron. J. Plant Breed.* **13**: 1-10.
- Nirubana V, K Ganesamurthy, R Ravikesavan and T Chitdeshwari (2017) Genetic variability studies for yield and yield components in kodo millet (*Paspalum scrobiculatum* L.). *Electron. J. Plant Breed.* **8**: 704.
- Pradhan A, AK Panda and RV Bhavani (2019) Finger millet in tribal farming systems contributes to increased availability of nutritious food at household level: Insights from India. *Agric. Res.* **8**: 540-547.
- Sharma I, B Tyagi, G Singh, K Venkatesh and O Gupta (2015) Enhancing wheat production-A global perspective. *Indian J. Agric. Sci.* **85**: 3-13.
- Upadhyaya HD, C Gowda, H Buhariwalla and J Crouch (2006) Efficient use of crop germplasm resources: identifying useful germplasm for crop improvement through core and mini-core collections and molecular marker approaches. *Plant Genet. Resour.* **4**: 25-35.
- Upadhyaya HD, M Vetriventhan, SL Dwivedi, SK Pattanashetti and SK Singh (2016) Proso, barnyard, little, and kodo millets. In: *Genetic and Genomic Resources for Grain Cereals Improvement*. Elsevier, pp. 321-343.
- Upadhyaya HD, M Vetriventhan, AM Asiri, V CR Azevedo, HC Sharma, R Sharma, SP Sharma and YH Wang (2019) Multi-trait diverse germplasm sources from mini core collection for sorghum improvement. *Agriculture* **9**: 121.
- Upadhyaya HD, CR Ravishankar, Y Narasimhudu, NDRK Sarma, SK Singh, SK Varshney, VG Reddy, S Singh, HK Parzies, SL Dwivedi, HL Nadaf, KL Sahrawat and CLL Gowda (2011) Identification of trait-specific germplasm and developing a mini core collection for efficient use of foxtail millet genetic resources in crop improvement. *Field Crops Res.* **124**: 459-467.
- Vetriventhan M, HD Upadhyaya, VC Azevedo, V Allan and S Anitha (2021) Variability and trait-specific accessions for grain yield and nutritional traits in germplasm of little millet (*Panicum sumatrense* Roth. Ex. Roem. & Schult.). *Crop Sci.* **61**: 2658-2679.
- Yadav OP and KN Rai (2013) Genetic Improvement of Pearl Millet in India. *Agric. Res.* **2**: 275-292.

Genome Editing Technologies for Efficient Use of Plant Genetic Resources

KC Bansal*¹, Somnath Roy² and Basudev Ghoshal³

¹National Academy of Agricultural Sciences, Pusa Campus, New Delhi-110012, India

²ICAR-National Rice Research Institute, Central Rainfed Upland Rice Research Station, Hazaribag-825302, Jharkhand, India

³Agriculture and Agri-Food Canada, Summerland Research and Development Centre, Summerland, British Columbia, Canada

Plant genetic resources (PGRs) are the basis for food and nutritional security, and enhanced utilization of these resources is of paramount importance for genetic improvement of crops. Till date the use of PGRs has been limited in conventional breeding programmes for developing widely adapted crop varieties. In the recent past, genome-wide association studies, genomics and functional genomics approaches using diverse germplasm accessions have facilitated discovery of novel quantitative trait loci (QTLs), genes and alleles associated with useful agronomic traits for use in crop improvement through molecular breeding and genetic engineering. More recently, genome editing has emerged as a new plant breeding technology with great potential for enhancing the use of PGRs, for addressing the challenges of climate change, malnutrition, environmental security, achieving SDGs by 2030, and to sustainably feed 10 billion people by 2050. In this article, an attempt has been made to highlight the use of CRISPR-based genome editing technologies for crop improvement through the use of PGRs.

Introduction

Plant genetic resources (PGRs) have contributed significantly to the development of modern high-yielding cultivars, through the use of conventional breeding methods, which have contributed to the dramatic increase in productivity of major crops since the middle of the 20th century. In recent times, the importance of PGRs has increased manifold for achieving climate resilience and ensuring sustainability in crop production. For instance, the rice landrace FR13A from Odisha, India was identified as a source for introducing *SUB1* (*SUBMERGENCE1*) quantitative trait locus (QTL) into mega-varieties for the development of Sub1 rice with submergence tolerance (Bailey-Serres *et al.*, 2010).

While conventional plant breeding methods have played a vital role in developing new crop varieties for increasing food production, advances in genomics, genetic engineering, molecular breeding and the recent development of new plant breeding technologies have enabled breeders to address the challenges of climate change, malnutrition and environmental security. The remarkable progress in plant genomics, sequencing and bioinformatics offers enough opportunities for mining germplasm collections, discovering new genes, elucidating gene function, and identifying superior alleles for use in the new breeding technologies like genome editing (Katiyar *et al.*, 2012, Halewood *et al.*,

2018). Genome editing, which was invented in 2012, is leading to a new revolution by accelerating the pace of genetic improvement of crops, and it is turning out to be indispensable technology for achieving climate resilience and sustainable agricultural development in this 21st century. Genome editing is already revolutionizing crop improvement by introducing desired changes in the plant's native genes with a high level of precision, accuracy and efficiency for developing new crop varieties with improved traits, without the need for introduction of foreign genes.

With the advent of genome editing as the next generation crop breeding technology, plenty of opportunities are now available to develop varieties with increased use efficiency of nutrients, water and radiation, and to create crops with inbuilt resistance to emerging pathogens and environmental stresses on a much faster timescale, not practically feasible with the use of conventional breeding approaches. Further, gene editing technologies could simplify the use of PGRs including crop wild relatives (CWR) and landraces in breeding programmes for expanding specific allelic variations and also eliminating linkage drag of undesirable traits.

Germplasm, Genomics and Pangenomics Facilitate Genome Editing for Crop Improvement

Over 7 million accessions of different plant species are conserved globally in various genebanks. However,

*Author for Correspondence: Email-kcbansal27@gmail.com

utilisation of this valuable resource is a major concern today by breeders with only < 1% use reported in crop improvement programmes. Genome editing offers a great promise to enhance the use of PGRs for crop improvement (Fig. 1).

The next-generation genome sequencing (NGS) technologies are contributing significantly to characterise the genebank accessions at genetic and molecular level, and to identify the genetic variation for enhancing their use in crop breeding. To capture the entire genetic variation in crop germplasm collections including landraces and wild species, advances in sequencing technologies have permitted whole genome sequencing of the diverse accessions of different crop species. In the recent past, efforts have been made to re-sequence 3,000 accessions of rice and over 3,000 accessions of chickpea generating pangenome/gene sequencing data across species with a view to linking genetic variation with traits/phenotypes of agronomic importance (Wang

et al., 2018; Varshney et al., 2021). For pangenomics, multiple accessions of a crop species are re-sequenced to determine structural and copy number variations associated with traits such as resistance to biotic and abiotic stresses that are useful for improving crop productivity in variable environments. Since a vast number of genes are not captured in a single reference genome, pangenomics assumes significance (Zanini et al., 2022). Information on pangenomics in major food crops is already being utilised in crop improvement programmes. Efforts are currently in progress in crops like rice, maize, soybean, chickpea, *Brassica* species, tomato and potato. More studies on pangenomics need to be conducted in other crop species, including CWR for identifying relevant genes and alleles associated with stress resistance and other agronomic traits. This will allow application of genome editing for the enhanced use of crop diversity for bringing novel genes/alleles from CWR and landraces to the cultivated gene pool for addressing the issues of climate adaptation of crops, shrinking natural resources, and increasing input use-efficiency, and for improving nutritional quality and architectural features of crops (Gasparini et al., 2021). Additionally, introducing the yield determining attributes from wild species to the cultivated crop genotypes or neo-domestication of wild species is now technically feasible using the CRISPR-based genome editing, base editing, prime editing and/or CRISPR-Combo approaches (Fig. 2).

Further, during the process of domestication several deleterious alleles have accumulated in the breeding lines and crop cultivars, affecting the fitness of the cultivated crops. Such deleterious alleles, for instance, have been identified by Varshney and his group in a recent study in chickpea including *Cicer* species, landraces, and superior cultivars, which can now be precisely removed through the use of various genome editing approaches (Varshney et al., 2021, Bohra et al., 2021).

Additionally, PGRs can also serve as a source for exploring epigenomic variation for crop improvement. The epigenome refers to the set of chemical modifications (e.g., DNA methylation, Histone acetylation) in the genome involved in regulating gene expression and holds great potential for crop improvement. It can be a target for the manipulation of regulation of gene expression and, consequently, the phenotype. As pangenome studies on PGRs are conducted to better understand the genetic variation, similar “pan (epi)genome” studies on PGRs can

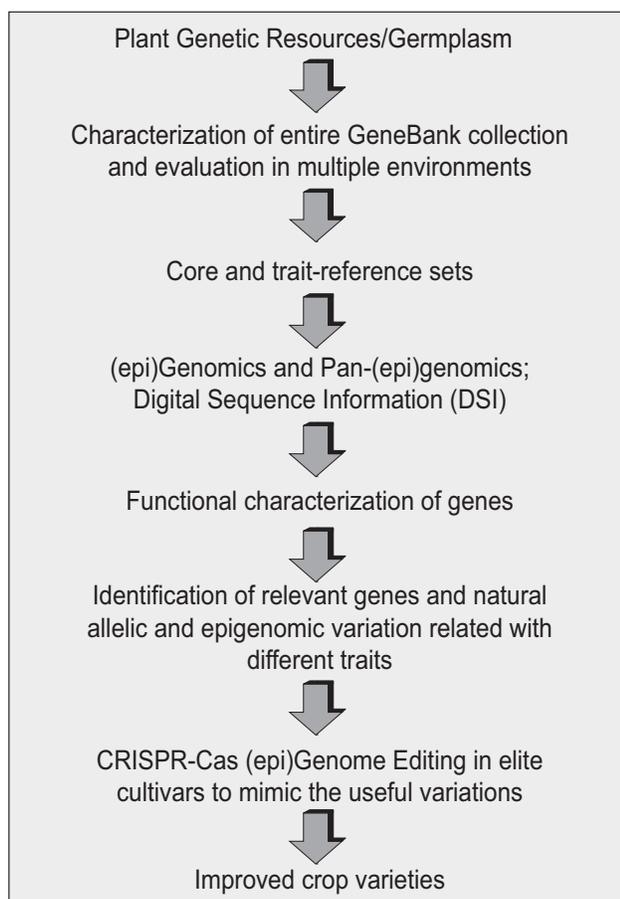


Fig. 1. Plant genetic resources as source of genomic information for crop improvement through (epi)genome editing

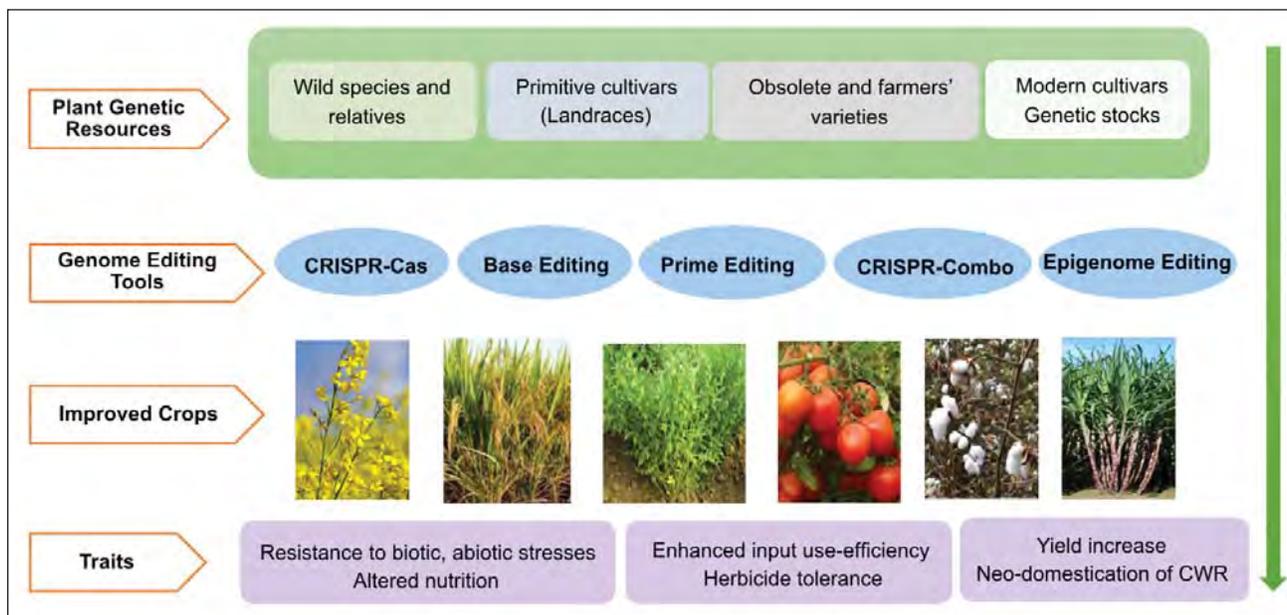


Fig. 2. Utilization of plant genetic resources for crop improvement through genome editing and epigenome editing

be useful to pave the way for modifying the epigenome and exploring the PGRs exhaustively (Fig. 2).

CRISPR-based Approaches for Crop Improvement

The CRISPR-Cas system creates a double-strand DNA break, which is repaired by cell's DNA repair machinery called NHEJ (Non homologous end joining). Through this repair mechanism, deletion or insertion of random nucleotides occurs, knocking out the gene function. The technology also allows precise substitution of nucleotides and insertion (knock-in) of DNA sequences at a predefined position using homologous template sequences through a homology-directed repair (HDR) pathway.

However, HDR-mediated genome editing is less efficient. To circumvent these issues, researchers prefer using base editing to introduce precise point mutations to introduce a trait, as recently shown by Gao and her group to confer resistance to a variety of herbicides in wheat (Zhang *et al.*, 2019). Similarly, a precise single-nucleotide change in *eIF4E1* gene using cytosine base editor rendered *Arabidopsis* plants immune to a disease. In base editing, one single base is converted to another through base editors without the double strand DNA break. Cytosine base editors for transforming C-G to T-A and adenine base editors for converting A-T to G-C are currently in use for crop improvement. This editing process has vast potential for incorporating useful agronomic traits in diverse crop species, thus enhancing

the use of PGRs for crop improvement (Molla *et al.*, 2021).

Another approach called prime editing has been developed with ability to perform sequence deletion, addition and substitution. This method requires no donor DNA as template or double-strand breaks. It employs catalytically impaired Cas9 endonuclease (nCas9), that nicks only one DNA strand, fused to reverse transcriptase and the prime editing guide RNA (pegRNA), which carries specified edits and the target site information. This allows direct transfer of new genetic information as desired edits from the pegRNA into a specified genomic site (Molla *et al.*, 2021). To overcome the low efficiency of this system, a new version of prime editing, enpPE2 has been recently developed and tested in rice (Li *et al.*, 2022).

Further, to overcome limitation of the classical CRISPR-mediated genome editing system of deletion or insertion confined to single genes, a novel breeding approach has been developed more recently by Yiping Qi and his group and called it CRISPR-Combo for multiple gene editing and simultaneously altering gene expression of other native genes without any deletion/insertion (Pan *et al.*, 2022). This system is also useful for enhancing *in-vitro* regeneration efficiency of recalcitrant crops varieties, in addition to simultaneously editing multiple genes, as demonstrated in poplar cells (Pan *et al.*, 2022).

Besides the above-mentioned approaches, epigenome editing is another novel approach for crop improvement (Gardiner *et al.*, 2022). Targeted manipulation of epigenetic marks is called epigenome editing and is achieved by combining sequence-specific DNA binding modules with effectors that can add or remove these marks from the genome (Gardiner *et al.*, 2022). Several epigenome editing tools have been developed using CRISPR-Cas9 systems to activate or repress gene expression by modifying the epigenetic marks in plants (Gallego-Bartolome *et al.*, 2018; Ghoshal *et al.*, 2021; Lee *et al.*, 2019; Papikian *et al.*, 2019). For example, inactive catalytic versions of Cas9 (dCas9) were combined with catalytic domains of DNA methyl transferases to silence gene expression by adding repressive DNA cytosine methylation marks (Ghoshal *et al.*, 2021; Papikian *et al.*, 2019). In a similar approach, the catalytic domain of human TEN-ELEVEN TRANSLOCATION 1 enzymes is combined with dCas9 to develop tools for activating gene expression by removing DNA cytosine methylation (Gallego-Bartolome *et al.*, 2018). Initially, these tools were tested and generated in the model plant *Arabidopsis thaliana*; currently, research is underway to expand their use to crops.

Conclusions and Future Perspective

In conclusion, it may be highlighted that genome editing has great potential for the enhanced use of crop diversity for bringing novel resistance genes/alleles from CWR and landraces to the cultivated gene pool for addressing the issues of climate adaptation of crops, shrinking natural resources, and increasing input use-efficiency. Direct genome editing in elite cultivars is likely to replace backcross breeding for altering alleles and creating optimal genetic variation required for desired traits.

However, to harness the full potential of these technologies, urgent attention will be needed for QTL/gene discovery through functional genomics using the diverse accessions of major crops for key traits (Das *et al.*, 2016). This requires that enough emphasis is given for characterisation of entire germplasm collections followed by genomics and pangenomics, and functional characterisation of genes for establishing precise links between phenotype and the genomic regions/genes or genetic elements in a given crop (Archak *et al.*, 2016; Kumar *et al.*, 2017; Phogat *et al.*, 2020).

Further, it is desired that simplified, reproducible *in-vitro* regeneration and genetic transformation systems are

developed in a range of agriculturally important crops and elite cultivars for efficiently generating genome-edited crop events. Similarly, successful genetic transformation of wild crop species and innovations in developing simplified methods for introducing genetic material will prove useful for accelerating crop domestication with improved traits related to climate resilience. These novel breeding technologies in due course of time could pave the way for enhanced utilisation of germplasm accessions conserved globally in different genebanks for sustainable food and nutritional security.

Acknowledgement

Authors thank Dr Kutubuddin A Molla, ICAR-NRRI, Cuttack for his valuable comments on the manuscript.

References

- Archak S, RK Tyagi, PN Harer, LB Mahase, N Singh, OP Dahiya, MA Nizar, M Singh, V Tilekar, V Kumar, M Dutta, NP Singh and KC Bansal (2016) Characterization of chickpea germplasm conserved in the Indian National Genebank and development of a core set using qualitative and quantitative trait data. *Crop J.* **4**: 417-424.
- Bailey-Serres J, T Fukao, P Ronald, A Ismail, Heuer S and D Mackill (2010) Submergence tolerant rice: *SUB1*'s Journey from landrace to modern cultivar. *Rice* **3**: 138-147.
- Bohra A, KC Bansal and A Graner (2022) The 3366 chickpea genomes for research and breeding. *Trends Plant Sci.* **27**(3): 217-219. doi: 10.1016/j.tplants.2021.11.017.
- Das S, M Singh, R Srivastava, D Bajaj, MS Saxena, JC Rana, KC Bansal, AK Tyagi and SK Parida (2016) mQTL-seq delineates functionally relevant candidate gene harbouring a major QTL regulating pod number in chickpea. *DNA Res.* **23**: 53–65, <https://doi.org/10.1093/dnares/dsv036>
- Gallego-Bartolome J, J Gardiner, W Liu, A Papikian, B Ghoshal, HY Kuo, JM Zhao, DJ Segal and SE Jacobsen (2018) Targeted DNA demethylation of the *Arabidopsis* genome using the human TET1 catalytic domain. *Proc. Natl. Acad. Sci. USA* **115**: E2125-E2134.
- Gardiner J, B Ghoshal, M Wang and SE Jacobsen (2022) CRISPR-Cas-mediated transcriptional control and epi-mutagenesis. *Plant Physiol.* **188**: 1811-1824.
- Gasparini K, J Moreira, L Peres and A Zsögön (2021) De novo domestication of wild species to create crops with increased resilience and nutritional value. *Curr. Opin. Plant Biol.* **60**: 102006.
- Ghoshal B, CL Picard, B Vong, S Feng and SE Jacobsen (2021) CRISPR-based targeting of DNA methylation in *Arabidopsis thaliana* by a bacterial CG-specific DNA methyltransferase. *Proc. Natl. Acad. Sci. USA* **118**: e2125016118.
- Halewood M (2018) Plant genetic resources for food and agriculture: opportunities and challenges emerging from the science and information technology revolution. *New Phytol.* **217**: 1407–1419. doi: 10.1111/nph.14993

- Katiyar A, S Smita, V Chinnusamy, DM Pandey and KC Bansal (2012) Identification of miRNAs in sorghum by using bioinformatics approach. *Plant Signal. Behav.* **7**: 246-259. DOI: 10.4161/psb.18914
- Kumar S, S Archak, RK Tyagi, J Kumar, VK Vikas, SR Jacob, K Srinivasan, J Radhamani, R Parimalan, M Sivaswamy, S Tyagi, M Yadav, J Kumari and KC Bansal (2017) Evaluation of 19,460 wheat accessions conserved in the Indian National Genebank to identify new sources of resistance to rust and spot blotch diseases. *PLoS One* **11**: e0167702. doi:10.1371/journal.pone.0167702
- Lee JE, M Neumann, DI Duro and M Schmid (2019) CRISPR-based tools for targeted transcriptional and epigenetic regulation in plants. *PLoS One* **14**: e0222778.
- Li J, L Chen, J Liang, R Xu, Y Jiang, Y Li, J Ding, M Li, R Qin, P Wei (2022) Development of a highly efficient prime editor 2 system in plants. *Genome Biol.* **23**: 161 <https://doi.org/10.1186/s13059-022-02730-x>
- Molla KA, S Sretenovic, KC Bansal and Y Qi (2021) Precise plant genome editing using base editors and prime editors. *Nat. Plants* **7**: 1166–1187. doi: 10.1038/s41477-021-00991-1.
- Pan C, G Li AA Malzahn, Y Cheng, B Leyson, S Sretenovic, F Gurel, GD Coleman and Y Qi (2022) Boosting plant genome editing with a versatile CRISPR-Combo system. *Nat. Plants* **8**: 513–525. <https://doi.org/10.1038/s41477-022-01151-9>
- Papikian A, W Liu, J Gallego-Bartolome and SE Jacobsen (2019) Site-specific manipulation of *Arabidopsis* loci using CRISPR-Cas9 SunTag systems. *Nat. Commun.* **10**: 729.
- Phogat BS, S Kumar, J Kumari, N Kumar, AC Pandey, TP Singh *et al.* (2020) Characterization of wheat germplasm conserved in the Indian National Genebank and establishment of a composite core collection. *Crop Sci.* **61**: 604-620 <https://doi.org/10.1002/csc2.20285>
- Varshney RK, M Roorkiwal, S Sun, P Bajaj, A Chitikeni, M Thudi, NP Singh *et al.* (2021) A chickpea genetic variation map based on the sequencing of 3,366 genomes. *Nature* **599**: 622-627.
- Wang W, R Mauleon, Z Hu, D Chebotarov, S Tai, Z Wu *et al.* (2018) Genomic variation in 3,010 diverse accessions of Asian cultivated rice. *Nature* **557**: 43–49. <https://doi.org/10.1038/s41586-018-0063-9>
- Zanini SF, PE Bayer, R Wells, RJ Snowdon, J Batley, RK Varshney, HT Nguyen, D Edwards and AA Golicz (2022) Pangenomics in crop improvement - from coding structural variations to finding regulatory variants with pangenome graphs. *Plant Genome* **15**: e20177.
- Zhang R, J Liu, Z Chai, S Chen, Y Bai, Y Zong, K Chen, J Li, L Jiang and C Gao (2019) Generation of herbicide tolerance traits and a new selectable marker in wheat using base editing. *Nat. Plants* **5**: 480–485.

Building Value Chains for Enhanced PGR Utilization and Sustainable Food Systems

JK Rana^{1*}, SK Malik² and Eleonora De Falcis³

¹The Alliance of Bioversity International and CIAT, India Office, NASC Complex, Pusa Campus New Delhi-110012, India

²ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

³The Alliance of Bioversity International and CIAT, Rome, Italy

Introduction

The World Food System is characterized by a high level of concentration and dependency. It is estimated that out of the above 5500 plant-known edible species, only 12 makeup of more than 75% of what we eat. Cereals provide the highest share in terms of calorie intake, with only three crops – rice, wheat, and maize – providing more than 50% of calorie intake. This impressive concentration on a few staple crops exposes the agricultural production system and their markets to vulnerability. Furthermore, recent UN report on World Population Prospects–2022 has made the whole World worry especially the countries like India where the population by 2050 would cross 1.66 billion and surpass the population of China (WPP, 2022). In such circumstances meeting the Sustainable Goals and providing healthy and nutritious food to increasing population would be a big challenge. Ensuring food and nutritional security side by side reducing pressure on natural resources, mitigating climate change and controlling price volatility due to conflicts and other disturbances is also a matter of deep concern for developing world.

Diversification of the production base could incorporate local landraces/cultivars/farmers varieties which however needs policy shift and investing in developing value chains. India is endowed with vast diversity of plant genetic resources with 167 cultivated species and 329 wild relatives of crop plants (Arora, 1991). In India around 1000 wild edible plant species have been exploited by native tribes and these include 145 species of roots and tubers, 521 of leafy vegetables/greens, 101 of buds and flowers, 647 of fruits and 118 of seeds and nuts (Arora and Pandey, 1996). Large number of land races about 30,000-50,000 of various crops, fruits and vegetables like rice, wheat, maize, millets, pseudo cereals, pigeon pea and other legumes, cucurbits, mango

and other fruits, turmeric, ginger, sugarcane etc. are exiting in India. Several of these landraces were grown by the farmers for their own consumption, however, era of green revolution and commercialization of agriculture led to the diversion of farmers to high yielding newer technologies. Several of these landraces are still being cultivated by the small and marginal farmers for their special attributes in India.

In the era of hybrid technology, gene transformation and gene editing one can think how the local landraces of crop plants nurtured, conserved and utilized by the native farming communities for centuries are even highly relevant today. These local landraces are not only rich in nutritional attributes but also have several genes tolerant to biotic and abiotic stresses (Duc *et al.*, 2010; FAO, 2019) These farmers landraces or varieties were the integral rather major part of agricultural production system in the past. Invention and spread of hybrid technology to increase productivity and income of farmers changed the scenario of farms especially small and marginal farms. It is the earnest fact that in indirect way farmers were involuntarily diverted from their traditional crops and local cultivars. This practice led to the erosion of several of indigenous landraces from the farmers' field and many local crops that were previously common in some areas, today are hard to find. However, still in remote, hilly terrains and fragile areas farmers with small land holdings prefer to grow some of these traditional crops especially small millets, pseudocereals, indigenous vegetables and other important crops for local use Bajracharya *et al.*, 2010

Need for Building Value Chains

In India agriculture provides livelihood to about 58% of population and contributes about 20% to the total economy of India as per the data released in 2021

*Author for Correspondence: Email-j.rana@cgiar.org

(MoAFW, 2021). Food and grocery market of India is also very large and stands at sixth position in the World. This is the position where more than 80% of the farmers are having less than 2 Hectares of land holding and agriculture is mostly for self-sufficiency. During the green revolution our emphasis was just on increasing the agricultural production to meet the food security, however, self-sufficiency in most of the food commodities has now led to the greater emphasis on nutritionally rich crops and value added products. There has been drastic increase in demand for the healthy food, and an expansion of the demand for fruits and vegetables. In India, for instance, food grain production in 2020-21 reached to 296 MT while horticultural production has crossed 330 MMT (MoAFW, 2021). Major initiative of government of India on food processing through Kisan Sampada Yojana where farmers are encouraged to cultivate processing varieties of fruits/vegetables and emphasis on organic agriculture has prompted increased investment of private players and also investment through foreign sources.

Farming community and consumers are now realizing the importance of local landraces. Many of them are rich in nutrients required systematic production, value addition and market value chain to reach to the peri-urban and urban markets. Small and marginal farmers growing these landraces sometimes lack skills to add value, resources and accessibility to the market for their product. Intervention of NGO's, Governmental agencies and corporate houses is often required to systemically achieve these goals (Gauchan *et al.*, 2020). Value chain has been defined as a set of activities that are linked to adding value to the product at each stage by bringing all the actors on a common platform to deliver a high-quality product and also generating more income among the participants. Agricultural value chain comprises farmers and farming inputs-seeds, planting material and other inputs; preharvest operations-crop and farm management, harvest; supply chain-processing, storage, packaging, transport and marketing and consumers. In the agri-value chain farmer the prime factor is having no visibility and not rewarded for his major contribution. However, when we talk of value chain for local landraces farmers, farming community and demography is at the centre along with trait may be nutritional value, taste, culinary attribute or any other specificity of the produce. Systematic development of value chain leads to sustainable food system and adds

to the livelihood of farmers and recognition to the area. Local landraces, farmers varieties and cultivars have been awarded or could qualify for Geographical Indicator (GI) have specificity for some trait and link to the territory as well as local traditions. GI Tags to local commodities could help in preserving genetic diversity, indigenous knowledge and enhancing farmers livelihood by recognizing their specificities and increasing their value added and contributing to increase awareness.

In India there are several examples of successful value chain development with the farmers participation besides commercial agri-value chains developed and acquired by the food companies and Agri-start-ups. Some of the successful case studies are given below:

Rice Value Chain SuccessStory of GEF Project

Several Indian rice-eating states have vast diversity of local rice varieties/landraces rich in nutrition, flavour, taste and texture that have been grown for centuries. These landraces were being cultivated using traditional methods caring for soil health also and sustain for future generations. The Kola Joha or Black Husked is rich in nutrients such as protein, minerals and contains high levels of antioxidants that protect cells, tissues, and vital organs. Traditionally this type of rice is cooked for pregnant women to full fill nutritional needs.

Under the GEF-UNEP project on "Mainstreaming agricultural biodiversity conservation and utilisation in the agricultural sector to ensure ecosystem services and reduce vulnerability", 24 heritage rice varieties including Kola Joha were identified and selected, after nutritional profiling, for revival across Assam under the Native Basket brand by Guwahati-based NGO Foundation for Development Integration (FDI). These traditional rice landraces are being marketed since December 2020, traditional rice growers are now targeting the increasing share of health-conscious Indian middle and upper class as their clients. Some of these rice landraces native to Assam has been revived from an almost-lost status to being currently farmed by hundreds of smallholders. Successful value chain has been developed for these highly nutritious landraces and products are available in the urban markets. In Assam, under the Native Basket brand, farmers have learnt to not just grow their rice but to independently handle market linkages after the Alliance, ICAR-NBPGR and FDI aided in registering a new brand to protect intellectual property rights (Fig. 1).

Similar efforts have been made by a group of 20 indigenous women farmers in Surguja district of Central-eastern Indian state Chhattisgarh realising the threats to the survival of their traditional rice variety called Jeeraphool by local name, translating to 'Cumin-Flower' taking its name from its small cumin-shape and pleasant aroma. After registering Jeeraphool with Plant Varieties and Farmers' Rights Authority of India, with technical support of the Alliance, it has also been awarded a Geographical Indication tag as the Jeeraphool variety is primarily grown only in Surguja district. The heritage rice has now found its place on India's food export list. The women self-help groups are striding ahead with their success, linking up with companies and local-level government offices to produce and market alternate products from the rice. From 120 hectares and 180 tonnes of Jeeraphool grains in 2005 they have more than tripled cultivation to 400 hectares harvesting over 1,000 tonnes in 2020 in Surguja district. The agricultural heritage has traversed a long journey to victorious survival.

The small and marginal farmers have also been organised into Farmer Producer Organisation and a new brand was registered. The rice demand and brand recognition increased, price fetched 50 percent more at 1,550 rupees (\$22) per quintal. Their aromatic rice brought in up to 20 percent higher. Over 2,000 farmer families are benefiting the whole gamut of activity from production to processing and sale. In the eight different locations where the Alliance is working, 19 community seed banks currently conserve, maintain and provide farmers ready access to over 2,000 traditional varieties of different crops.

Millet Value Chain–ICAR-IIMR Success Story

Millets are generally grown in 131 countries of the World (<https://milletadvisor.com/importance-of-millets-in-india/>). Millets are the traditional food of approximately 590 million people of Asia and Africa. India may be treated as the home of Millets with the production of >170 lakh ton which is the 80% production of Asia's and about 20% of global production. In India, these millets are represented by six species, namely, finger millet (*Eleusine coracana*), kodo millet (*Paspalum scrobiculatum*), foxtail millet (*Setaria italica*), little millet (*Panicum sumatrense*), proso millet (*Panicum miliaceum*) and barnyard millet (represented by two

species, viz., *Echinochloa crusgalli* and *E. colona* latter domesticated in India)

ICAR-Indian Institute of Millet Research (ICAR-IIMR) is the premier institute undertaking the research, development, extension and value chain development of millets. ICAR-IIMR through its several projects and initiatives promoted not only the cultivation of millets but several innovative interventions in value chain building have led to development of nutritious products meeting consumers' demand. These efforts have led to the millet revolution in India and given new life to the rather elapsed crops. ICAR-IIMR through its Nutrihub (<https://www.nutrihubiimr.com/about-nutrihub>) is sharing and demonstrating processing Infrastructure facilities for entrepreneurs incubated along with 60+ value added technologies for millet value added products backed by extensive research and Centre of Excellence. The Agribusiness Incubator (ABI) and Technology Business Incubator (TBI) are providing lot of support to entrepreneurs and farming community. More than 175 Stratups have been supported and above 190 technologies related to millets have been transferred to industries and entrepreneurs (<https://www.nutrihubiimr.com/about-nutrihub>; Rao and Tonapi, 2021). ICAR-IIMR is also supporting 31 FPOs to undertake and promote millets and their value-added products. These FPOs are of great help in supporting millet farmers in cultivating, processing and marketing of their produce. These efforts are helping in popularising millet products and liking of these nutri-rich foods is increasing in health-conscious rural and urban population. These efforts are also adding to livelihood of small farmers by increasing their disposable income and providing them better nutrition. Study undertaken as a part of global project on NUS species amply demonstrated that currently marginalized crops, such as minor millets, can in fact contribute to the nutrition security of rural and urban poor people in India, while at the same promoting economic development and the empowerment of women and other vulnerable groups (Padulosi et al., 2015).

Jackfruit Value Chain and Benefit Sharing (<https://www.iihr.res.in/linking-biodiversity-livelihood-security-jackfruit>)

There are several examples where farmers and community have been benefited by conserving varieties or cultivars having some special traits and higher economic value due to special liking by the consumers. In most of the

cases farmers are unable to recognise this or do not have capacity to gain extra benefit out of these elite germplasm or farmers cultivar. Vast genetic diversity of crops and horticultural species is available in the backyard gardens and farmer's field. Such farmers or community holding unique diversity have been recognised by PPVF&RA, ICAR, SAUs and other governmental agencies and NGOs as "Custodian Farmers" during last two decades. ICAR-Indian Institute of Horticulture Research (ICAR-IIHR), identified two superior jackfruit genotypes with attractive coppery red coloured flakes in the traditional jackfruit growing tracts of Southern Karnataka. These varieties were given wide visibility through a combination of media advertisement and fairs. This cultivar received tremendous response for the planting material by farmers. Hence, ICAR-IIHR created a model for commercialization in 2017, after recognising and honouring the farmers for conserving this variety as "Custodians of Genetic Diversity". Revenue generation was also shared between the Licensor (jackfruit farmer) in such a way that 75% of the earning would go to the farmer and 25% would go to the Institute, if the price per sapling is Rs 150 then Rs 112 would go to the farmer and Rs. 38 for the Institute. The model is well established and custodian farmers are profiting through benefit sharing and also the elite germplasm is conserved and utilized. Such innovative models of linking biodiversity with livelihood security are helping farmers in increasing their income.

Custard Apple Value Chain in Rajasthan (NAIP Project Report, 2013 and Shailza et al., 2020)

Development of custard apple value chain is a successful example where a wild fruit of the tribal areas of Rajasthan could be benefited. This fruit is widely growing particularly in the tracts of Udaipur, Chittorgarh, Jhalawar, Dungarpur, Bhilwara and Rajsamand districts. Freshly harvested fruits by tribals were sold at very less price due to difficulty in transport and also the perishable nature of fruits. There were several common issues tribal farmers face in handling and marketing most of the underutilized fruits. Some of these are harvest and post-harvest losses, lack of processing technologies and value addition and unorganized market (Kaushik, 2013). These challenges have been successfully addressed in custard apple and various processing options, value added products and marketing channels were established. The processing of raw fruit into various innovative products like pulp, powder, etc. promotes market acceptability

and gives the products high economic value (Kaushik, 2013). These innovations of value addition greatly helped in post-harvest management of fruits, income and employment generation of tribal farmers. All this started with the training of farmers on harvesting, standardized rejuvenating technology, pulp extraction equipment development and adopting browning free technology for the processing of custard apple, which helped in increasing the shelf life of finished product from a few days to more than a year. It involved the establishment of collection centre at village level, processing unit, a storage unit and marketing network to promote sale of the produce (Shailza *et al.*, 2020). Interventions involving improved cultivation practices and reduction in post-harvest losses would increase the production level by 50 per cent and 20 per cent, respectively, whereas value addition activities through processing would give better returns to the processors as well as to the tribal harvesters (Shailza *et al.*, 2020). Technology has been transferred and successfully adopted by many start-ups and tribal farmers, and all farmers, particularly women, are benefited by earning additional livelihood (Fig. 2).

Challenges in Building Value Chains

Value chain development encompass significant challenges at farm, pre- and post-harvest, supply chain, and consumer level. Building successful value chains especially for landraces depends upon success at all these levels. Encouraging farmers to grow the local landraces on their farm and developing value chains need concerted efforts by all the stakeholders involved, and political will. There are several examples where such successful attempts have been made and through community participation conservation and use of indigenous landraces and local vegetables and fruits have been ensured. Several value chains have been developed and demonstrated under various projects involving NGOs and result of studies have been published showing substantial gains and benefits, however, practically these value chains have not been successfully implemented or adopted by the farmers and consumers. Value chains developed by the private business houses where ample investment and able backstopping with deep market studies are involved have shown the good success. Innovations in food retailing has increased the involvement of the private sector in agriculture and also an emphasis on development and refinement in agriculture value chains. However, value chain development starting from the scratch and involving small farmers



Fig. 1.



Fig. 2

face enormous challenges. Supporting farmers in the cultivation, access and use of landrace, improve market linkages and prices, through value added and mitigate losses in productivity. Bringing farmers in to the FPO fold and taking advantage of various governmental schemes is beneficial to some extent but needs greater involvement. The recent initiative by Government of India like promotion of Millets through “Initiative for Nutritional Security through Intensive Millets Promotion (INSIMP)”, ‘Poshan Abhiyaan 2018-22’, Poshan Month, including celebrating 2018 as Year of Millet and UN would observe 2023 as International Year of Millets due to Indian spearheading. Inclusion of millets and other nutri-foods in the Midday Meal programme and PDS would also promote the cultivation of these crops leading to enhanced farmers income.

Plant Genetic Resources Conservation and Utilization

Genetic diversity in the form of diverse landraces belonging to cereals, millets, pseudo cereals, pulses, vegetables, and indigenous fruits needs urgent conservation. The genetic diversity of landraces is an important part of global crop biodiversity and is considered of paramount importance for future world production (Wood and Lenne, 1997). The utilization of local crop landraces is intensely entangled into the food habit, socio-religious, traditional and cultural dynamics of most of the rural and tribal communities of the World. Most of these local landraces and their uses are limited to small area and native population. Mostly landraces are dynamic populations of genetically diverse, locally adapted cultivated plant species that have historical origin, distinct identity, no formal crop improvement,

and are often times associated with traditional sustainable farming systems (Camancho *et al.*, 2005). Most of the indigenous landraces being grown by the small and marginal farmers have now been either vanished from the farmers field or only being grown for domestic consumption. The reduced use of landraces can be traced back to the beginnings of the “Green Revolution” (Walters, 2018). Landraces possess vast genetic diversity and due to limited use by the farmers and market value are fast disappearing, their use and continued cultivation is critical near the primary and secondary centres of diversity due to alleles present and evolved during the centuries of evolution at these habitats (Brush, 1994). During the last two decades concerted efforts are being made to protect these indigenous landraces by not only conserving in the Genebank but also promoting their use by adding value to these to enable small farmers to remain with these crops. Although attempts have been made during last 3-4 decades to collect, characterise and conserve these genetic resources in seed genbanks, in vitro repositories, cryobanks and field genebanks. However, in situ on farm conservation is now preferred to conserve these genetic resources with the farmers participation. Some of the landraces of major cereals like wheat, rice, maize, barley, pulses and millets are still widely grown and used by local communities. This effort of farmers to grow these important landraces in the farmers field would also protect and conserve the valuable genetic resources. Establishing Community Seed Banks (CSBs) at Panchayat or Block level may be of great advantage in facilitating farmers in conserving, utilizing and selling their produce. CSBs also can be elevated to the status of small training hub for value addition marketing activities of local products. This

practice of conservation through use of landraces would require handholding of farmers to enable them to get suitable value of their produce. This would only be possible by developing successful and reliable value chains for these landraces or their value-added products.

Sustainable Food Systems

The global food system is, at present, based on extremely low genetic diversity, this is impacting dietary quality of vast population especially in the developing world. Beyond their relevance to diets and nutrition, food systems also play an important role in promoting environmental sustainability (climate change adaptation and mitigation, biodiversity, soil and water degradation), inclusivity (viability for smallholder farmers, indigenous peoples, gender equity), livelihood and productivity (increased production of nutritious foods, economic development). The Sustainable Development Goals support making the food system more productive, environmentally sustainable and resilient.

Almost half of the calories consumed by humans come from just four crops, namely wheat, rice, sugar and maize (FAO, 2018); consumed food is becoming more energy-dense and nutrient-poor (Khoury *et al.*, 2014); and fruits and vegetables are under-consumed in all regions of the world, except for China, Japan and South Korea (Berners-Lee *et al.*, 2018). Low intake of fresh fruits, vegetables, nuts and seeds and whole grains is associated with an increased risk of disease, especially cardiovascular disease, type II diabetes and cancer, and affects the poorest populations in particular (Afshin *et al.*, 2019). Reversing these trends requires a redesign of the global food system, which in turn necessitates a thorough understanding of which foods have the potential to simultaneously deliver environmental, nutritional and livelihood benefits at local and global levels.

Extreme climate events are going to impact our food security and in turn resilience of existing food systems. Traditional food system which was more robust and were climate resilient have largely disturbed due to the changing demand for food and policies of governments to grow more. There is a deep contradiction among the policy makers, one side there is an advocacy for the production centric agriculture to meet the increasing food demand and another side need for resilient and sustainable food systems to ensure environmental and nutritional security is picking up the momentum. Transforming

food systems for improved resilience has been discussed by the Committee on World Food Security at the UN, IPES-Food, International Assessment of Agricultural Knowledge, Science and Technology for Development, and the UN Food Systems Summit (UNFSS), however, still opinions are divided (Mehrabi *et al.*, 2022). However, the demand for the more proteinaceous food now even in the developing markets due to high rate of obesity is changing the farm priorities. Diversification of food would enhance number of crops in the farm and would provide opportunity to farmers for crop rotation, incorporation of native crops in farming system and also the enhanced income.

In the 21st century agriculture is going under immense transformation entry of educated and tech-savvy youth entrepreneurs in to an attractive vast agricultural economy to grasp some share through modern tools is evident. The wave of start-ups coming in the Agri-sectors and vast investment coming from domestic and foreign investors in farmers as well as consumers centric business is changing the entire landscape of agri-business. During 2017 to 2020, India received ~US\$ 1 billion in agritech funding. With significant interest from the investors, India ranks third in terms of agritech funding and number of agritech start-ups. By 2025, Indian agritech companies are likely to witness investments worth US\$ 30-35 billion (IBEF, Report, 2022). Substantial investment is going in to the value chain development benefiting small and marginal farmers. There is a great opportunity for farming community to encash upon this trend and diversify to the nutri-rich, climate resilient crops to meet the market demand and to strengthen sustainable agriculture and food systems. Diversification of production base and developing value chains for indigenous landraces of food and horticultural crops is the only answer to meet the food and nutrition demand of increasing population.

References

- Afshin A *et al.* (2019) Health effects of dietary risks in 195 countries, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. *The Lancet*, [http://dx.doi.org/10.1016/S0140-6736\(19\)30041-8](http://dx.doi.org/10.1016/S0140-6736(19)30041-8)
- Arora RK (1991) Plant diversity in Indian gene centre. *In*: Paroda RS, Arora RK (Eds.). *Plant Genetic Resources - Conservation and Management*. New Delhi, India: IPGRI, Regional Office for South Asia, 1991;25-54.
- Arora RK and A Pandey (1996) *Wild edible Plants of India: Diversity, Conservation and use*. National Bureau of Plant Genetic Resources, New Delhi, India

- Bajracharya J, RB Rana, D Gauchan, BR Sthapit, DI Jarvis, & JR Witcombe (2010) Rice landrace diversity in Nepal. Socio-economic and ecological factors determining rice landrace diversity in three agro-ecozones of Nepal based on farm surveys. *Genetic Resources Crop Evolution* **57**(7): 1013–1022
- Berners-Lee M *et al.* (2018) Current global food production is sufficient to meet human nutritional needs in 2050 provided there is radical societal adaptation. *Elem Sci Anth*, **6**: 52. DOI: <https://doi.org/10.1525/elementa.310>
- Brush SB (1994) In situ conservation of landraces in centers of crop diversity. *Crop Sci* **35**(2): 346–354.
- Camacho Villa TC, ZN Maxted, MA Scholten and BV Ford Lloyd (2005) Defining and identifying crop landraces. *Plant genet. resour.* **3**(3): 373–384
- Duc G, S Bao, M Baum, B Redden, M Sadiki, , MJ Suso *et al.* (2010) Diversity maintenance and use of *Vicia faba* L. genetic resources. *Field Crop Res.* **115**: 270–278. doi: 10.1016/j.fcr.2008.10.003
- FAO (2019) Voluntary Guidelines for the Conservation and Sustainable Use of Farmers' Varieties/Landraces. Rome: FAO.
- Final Report. National Agricultural Innovation Project (Indian Council of Agricultural Research) (2013) A Value chain on commercial Exploitation of Underutilized Fruits of Tribal Zones of Rajasthan.
- Food and Agriculture Organization of the United Nations, International Fund for Agricultural Development, International Organization for Migration, & World Food Programme (2018) The linkages between migration, agriculture, food security and rural development: technical report. <https://www.fao.org/documents/card/en/c/CA0922EN/>
- Gauchan D, BK Joshi, B Bhandari, HK Manandhar and DI Jarvis (eds.) (2020) Traditional Crop Biodiversity for Mountain Food and Nutrition Security in Nepal. Tools and Research Results of the UNEP GEF Local Crop Project, Nepal. NAGRC, LI-BIRD and the Alliance of Bioversity International and CIAT; Kathmandu, Nepal.
- <https://www.iihr.res.in/linking-biodiversity-livelihood-security-jackfruit>
- <https://www.ibef.org/industry/agriculture-india>
- <https://milletadvisor.com/importance-of-millets-in-india/>
- <https://www.pib.gov.in/PressReleasePage.aspx?PRID=1741942>
- https://agricoop.nic.in/sites/default/files/Web%20copy%20of%20AR%20%28Eng%29_7.pdf
- <https://www.nutrihubiimr.com/about-nutrihub>
- <https://tracextech.com/category/case-studies/>
- Mehrabi Z *et al.* (2022) Research priorities for global food security under extreme events. *One Earth* **5**, 756–766.
- Kaushik RA (2013) Technology on custard apple processing and value chain: A step towards doubling the income of tribals. *Manual of RCA Alumni Association, Udaipur* **1**(3): 72–75
- Khourya CK, AD Bjorkmanc, H Hannes Dempewolf, J Julian Ramirez-Villegasa, L Guarinof, A Jarvisa, LH Riesebergd and PC Struikb (2014) Increasing homogeneity in global food supplies and the implications for food security. *PNAS*, DOI: 10.1073/pnas.1313490111.
- Rao BD and VA Tonapi (2021) A Compendium of Millet Start-ups' Success Stories, ICAR-Indian Institute of Millets Research, Rajendranagar, Hyderabad. PP. 204
- Sangappa RD, Manjuprakash, Ashok Sajjan, B Laxmi, K Srinivas Babu and Vilas A Tonapi (2022) Profiles of Millets Farmer Producer Organizations Promoted by ICAR-IIMR. Indian Institute of Millets Research, Hyderabad-500030, Telangana, India.
- Shailza Sharma L, SS Burark, RA Kaushik and GL Meena (2020) Prospects of custard apple value chain development in Rajasthan. *Economic Affairs* **65**(2): 207–212.
- Stefano Padulosi, Bhag Mal, Oliver I King and Elisabetta Gotor (2015) Minor Millets as a Central Element for Sustainably Enhanced Incomes, Empowerment, and Nutrition in Rural India Sustainability, **7**: 8904–8933; doi:10.3390/su7078904
- Walters SA (2018) Essential role of crop landraces for world food security. *Modern Concepts & Developments in Agronomy* **1**: 91–94.
- Wood D and JM Lenné (1997) The conservation of agro biodiversity on-farm questioning the emerging paradigm. *Biodiversity and Conservation* **6**(1):109–129.
- United Nations Department of Economic and Social Affairs, Population Division (2022) World Population Prospects 2022: Summary of Results. UN DESA/POP/2022/TR/NO. **3**: 207–212.

Contribution of Technology in Enhancing and Preserving PGR

Ram Kaundinya*, Shivendra Bajaj and Ratna Kumria

Federation of Seed Industry of India, 10A, Vandhana Building, 11 Tolstoy Marg, New Delhi-110001, India

Agriculture begins with seed and most improvements in agriculture employ plant genetic diversity. Agricultural trait improvement and genetic gain rely on germplasm diversity and associated information. Given the pace of germplasm loss, technology is critical in germplasm management and preservation. Traits that improve crop productivity and quality of produce also indirectly contribute towards germplasm conservation. Digital information can also help to monitor and conserve genetic diversity.

Introduction

Plants form the basis for all other life on our planet. They balance the various ecosystems and provide food, fibre and fuel for the growing human population, as well as habitat for most other organisms. Human intervention with their growing number, urbanization and industrialization have led to plant ecosystems being disturbed resulting in loss of plant biodiversity. Plant scientists have been working to preserve and conserve the diversity of plant genetic resources across the globe. Conservation efforts have been taken up both locally as well as globally with contributions from communities and nations. The diversity of plants needs to be preserved as it is the only way forward for upgrading marginal or degraded regions and for ecological sustainability. Technology and agriculture are interfused with and make use of diversity of germplasm and can also contribute to its preservation.

Efforts so Far are Still not Adequate

The largest seed bank for long term storage of seeds was established in 2008 near arctic circle, the Svalbard Global Seed Vault. India is home to the second largest gene bank set up by National Bureau of Plant Genetic Resources (NBPGR) in 2021. Most of the countries and CGIAR institutions have their own banks for plant genetic resources. However, these efforts are inadequate, given the immense diversity of the plant varieties and races spread across the globe in varied environments. In India, NBPGR and National Biodiversity Authority (NBA) are the authorities that manage germplasm conservation and access.

Modern Technology Provides Solutions

In the era of genome sequencing and genomics, physical access to germplasm is not the only way to utilize and benefit from the diversity, the sequence data associated with the germplasm is also a treasure trove of knowledge that may be used for better understanding a trait, improving it or modifying it with all the new available breeding tools. Therefore, we need to invest in generating gene sequence or protein data for the available accessions in seed banks and devise a mechanism to manage the access for such data. Promoting open exchange of Digital Sequence Information (DSI) will support conservation, fosters research into technological solutions to tackle societal challenges and benefit the global population as a whole. Germplasm or DSI access is a necessity for scientific advancement and technological development. Any barriers to the sharing and use of DSI would discourage innovation and scientific research. The benefits of open access and sharing were demonstrated well in case of tracking and monitoring of SARS-CoV-2 across the globe, that helped in containment, management of the spread and vaccine development for the virus.

Global and Local Approaches Needed

International Treaty on Plant Genetic Resources for Food and Agriculture (ITPGRFA) was adopted by FAO in 2001 and focuses on sustainable use of all plant genetic resources for food and agriculture. India is a signatory of this treaty as well as Convention on Biological Diversity (CBD). These treaties recognize the contribution of farmers to the diversity of crops while establishing a global system for access and benefit sharing. The open

*Author for Correspondence: Email-ram@kaundinya.in

access and monitoring of benefit sharing will require use of digital technologies across the globe, with cross access across databases, so that duplication of efforts in data monitoring is avoided. This will reduce costs and ensure fair monitoring.

GM and Gene Edited Crops

Developers of GM and gene edited crops rely on germplasm diversity and allelic variation for trait improvement. Without ample sequence information and study of variants and wild species, none of the biotechnology tools would be effective. The cultivation of the improved crops positively impacts associated ecosystems as well as field resources. It is well documented that GM cultivation has reduced land conversion and reduced environmental impact of agriculture. For instance, if crop biotechnology had not been available to farmers in 2018, maintaining global production levels that year would have required the planting of an additional 14 percent of the arable land in the United States, or roughly 38 percent of the arable land in Brazil or 16 percent of the cropping area in China. These technologies utilize plant genetic resources and also contribute towards their conservation and improvement.

Policy Imperatives

Continuous crop improvement is an imperative to meet growing demand for food, feed and fuel. Governments in each country, while trying to protect their own native germplasm, have to adopt facilitating policies which allow cross border exchange of germplasm, open access to germplasm by both public and private sector players and conservation of local germplasm by communities. Private sector has a crucial role to play in the crop improvement programs of different countries. It is very important to clearly define the parameters of access and benefit sharing for germplasm. Biodiversity conservation efforts of different organizations and communities should be rewarded and encouraged. Private industry may be encouraged to help local communities to conserve their genetic resources through professional management of local seed banks. Harmonization of policies across nations for conservation and exchange of plant genetic material through global treaties is very important and they should be put in place sooner than later. Farmers who conserve natural resources including soil and biodiversity should be rewarded on the basis of clearly defined and measurable parameters. Short term and long-term storage

infrastructure for germplasm may be created in remote areas of the countries by the governments. Professional cataloguing along with gene sequence and protein data of all the germplasm stored in such facilities needs to be facilitated by the government.

Sustainable Future Scenario

We are moving from productivity centric agriculture to sustainable agriculture that requires diversity in food crops and promoting local germplasms. Though plant breeders have multiple modern tools available with them, they all rely heavily on the historic records and genetic sequence information. Be it MAS, GWAS or gene editing they all can improve the breeding process but need genetic sequence information about breeding population or allelic variants. Breeding for climate resilience and de-novo domestication will not only require access to available diversity but will also contribute to germplasm diversity for various crops adapted to local environments.

Plant genetic resources preservation entails intense, long-term investment and is spearheaded by national governments or multi-nation consortia. But the resources, be it seed or DSI is accessed by researchers, plant breeders and trait developers from universities, public institutions and private industry and in many cases, they collaborate and contribute towards enhancing the preservation and conservation efforts. An equitable germplasm benefit sharing system can further support conservation or efforts of a community. Open access to plant genetic resources and collaboration between various stakeholders will support plant breeding for global food and nutritional security and maintain the diverse niche farming systems. Local seed banks are needed for better preservation and access to germplasm as well as involvement of the local community and stakeholders. Infrastructure development or upgradation is critical for these efforts and it can be supported by stakeholders accessing the germplasm from the seed bank

Global acceptance of policies and harmonization across nation is also critical for adequate plant genetic resource management, besides the use of digital technologies, infrastructure development and use of new breeding tools.

Indiscriminate destruction and misuse of natural resources can be balanced by re-planting degraded areas and conserving existing habitats in view of climate change in a sustainable manner. This can only be achieved if

diverse germplasm is preserved and are available for use across the globe.

References

- Giller KE, Anderson JA, Corbeels M, Kirkegaard J, Mortensen D, Erenstein O and Vanlauwe B, Beyond conservation agriculture, 2015, *Frontiers in Sci.* (6) (<https://doi.org/10.3389/fpls.2015.00870>)
- Svalbard Global Seed Vault- <https://www.regjeringen.no/en/topics/food-fisheries-and-agriculture/svalbard-global-seed-vault/id462220/>
- ITPGRFA- <https://www.fao.org/plant-treaty/overview/en/Promoting-sustainable-use-and-conservation-of-biodiversity-through-open-exchange-of-Digital-Sequence-Information,2019,7-joint-stakeholder-statement-on-DSI-ICC.https://pgeconomics.co.uk//efaidnbmnnnibpcajpcglclefindmkaj/https://pgeconomics.co.uk/pdf/globalimpactfinalreportJuly2020.pdf>

Vegetable Genetic Resources to Mitigate Nutritional Insecurity in India

Tusar Kanti Behera*, Jagdish Singh, Rakesh Kumar Dubey and Shailesh Kumar Tiwari

ICAR-Indian Institute of Vegetable Research, Post Bag No. 01, Post Office Jakhini, Shahanshapur, Varanasi-221305, Uttar Pradesh, India

Vegetable crops are key sources of essential micronutrients required for good health. They add fiber, flavor, taste, and nutritional quality to human diets. Increasing production and consumption of vegetables constitutes a direct and affordable way to deliver better health and overcome malnutrition. Vegetable production has the potential to generate more income and employment than any other segment of the agricultural economy. Vegetables can be grown on small areas of land, close to the consumers in urban and peri-urban settings, and they do not necessarily need advanced technologies to grow them. To realize those benefits, governments and donors need to give more weight and support to the ex situ, on-farm, and in situ conservation of genetic resources including farmers' varieties, landraces, and wild related species of global, as well as traditional, vegetables.

Key Words: Alliums, Brassicas, Chillies, Cucurbits, Eggplant, Lettuce, Micronutrients, Spinach, Tomatoes, Vegetable legumes

Introduction

Climate change and population growth in many developing countries impede progress toward achieving food and nutritional security. The historic success of the Green Revolution in terms of yield gains, together with lower food prices, ensured adequate quantities of staple cereal grain, thereby drastically reducing the problem of famine. However, after years of steady decline, the trend in world hunger is now slowly increasing in terms of under-nutrition and malnutrition. Hunger and under-nutrition among children, pregnant ladies, lactating mothers particularly in the economically weaker strata of the society is one of the pervasive health problem in developing country contributing to malnutrition and mortality at younger stage. Malnutrition is the single most important risk factor for disease. Diet-related diseases such as diabetes, cardiovascular disease, hypertension, stroke, cancer, and obesity are escalating at a global level. Despite of being the direct outcome of the food habit of any individual, nutrition has been perceived as a minor factor by policy planners in our country. Despite India being self-sufficient in food grain production, it was home to nearly one fourth of world population of the under-nourished people.

Of-late, diversified diets based on a range of crop species, are essential for nutritional security which could be achieved by shifting the policies of

“calories fundamentalism” to “nutrition sensitive” ones. Importance of fruits and vegetables in the daily human diet promotes and recommends the consumption of 400 g of fruits and vegetables per day to provide necessary nutrient lacking in other food groups (WHO 2003). With the increasing epidemiological support on benefit of vegetable consumption on health, Indian Council of Medical Research (ICMR) recommended 300 g vegetable consumption (125 g leafy vegetable, 100 g root and tuber and 75g fruit vegetable) per capita per day. The diversity and quality of food produced and consumed is a decisive factor when addressing the triple burden of malnutrition, i.e., undernutrition, micronutrient deficiency, and over-nutrition. Although fruit and vegetables are usually mentioned jointly when addressing malnutrition, this article focuses mainly on the compilation and review the role of both global and traditional vegetables in addressing nutritional security and human health.

Economic Importance of Vegetables

Vegetables comprise a wide range of genera and species and are important component of a healthy diet. They ensure nutrition security through the provision of vitamins, antioxidants, minerals, fiber, amino acids, and other health-promoting compounds, while enhancing diversity, flavor, and taste of many otherwise bland staple dishes. Vegetables are rich sources of diverse group of

*Author for Correspondence: Email-tusar@rediffmail.com

nutraceuticals with specific health benefit. Being high value cash crops and an excellent source of vitamins and micronutrients, vegetables not only generate employment and income, but immensely contribute to gender equity, better livelihoods, and alleviate malnutrition from imbalanced diets in developing countries as well as in developed countries through increased vegetable consumption. Amongst various food groups, vegetables have been considered as important towards addressing the chronic issue of nutritional security. If integrated in the daily diet, several indigenous vegetables are rich source of micronutrients and have potential to contribute to nutritional security.

Diversity in Vegetable Species

A world vegetable survey showed that 402 vegetable crops representing 230 genera across 69 families are cultivated worldwide. The most dominant vegetables are tomatoes, cucurbits (pumpkins, squashes, cucumbers, and gherkins), alliums (onion, garlic, shallot), and chillies (sweet and hot pepper; *Capsicum* spp.). Other major vegetable crops based on farm gate value of global production, are spinach, brassicas (cabbages, broccoli, rape), vegetable legumes, eggplants, lettuce and chicory, carrots and turnips, and asparagus. Production statistics usually do not list indigenous or traditional vegetables as these are often produced in home or family gardens or collected from the wild for family consumption, and they are, in general, only offered in local markets.

The term “indigenous vegetables” primarily refers to plants grown in their centers of origin or diversity, but also encompasses plant species introduced from other geographical areas that adapted well, naturalized, and evolved in the new environment. Indigenous vegetables are often more nutrient-dense than global vegetables, require low levels of external inputs, and cope well with abiotic and biotic stresses if grown on a small scale and in mixed cropping systems as is the case in their centers of origin. However, data on nutritional profiles of indigenous vegetables in raw and cooked forms are scarce.

With a few exceptions, private and public sector investments in vegetable research are mainly focusing on the development of hybrid cultivars of predominantly global vegetables, while indigenous or traditional vegetables are being neglected. High adaptability of these indigenous vegetable crops to biotic and abiotic stresses makes them a source of income and nutrition to the

farmers during any such stress by combating malnutrition and poverty. Several organizations in NARS including the National Bureau of Plant Genetic Resources, New Delhi, ICAR-Indian Institute of Vegetable Research, Varanasi have a highly diverse collection of indigenous vegetables, and are engaged in multiplying selected lines of a wide range of crops while conserving their genetic resource base.

Since Independence, there has been marked increase in production, per capita availability and consumption of fruits and vegetables. A combination of agriculture research and efficient implementation of government policies have resulted in a shift in land use from staple crop farming to commercial farming particularly the cultivation of vegetables as cash crops and off-season cultivation under protected structures has ensured availability of adequate quantity and quality of safe vegetables of a particular season area throughout the year for consumption. This has in turn resulted in an increased supply of non-staple foods including vegetables, with a positive impact on the nutritional status of the country. Blessed with varied agro-climatic zones amenable to grow a wide range of vegetable crops, India is the second largest producer of vegetable crops in the world with 191.77 MT production next only to China with productivity of 18.52 t/ha (2nd advance estimate for 2019-20 of National Horticulture Board). Vegetables contribute 41.32% area and 59.32% production in total horticultural crops. Consumers are now offered more diversity and convenience but at the same time cheaper, less healthy, and highly processed food with high content of fats and sugars has been made more easily accessible and affordable and is, therefore, in high demand. However, increasing awareness of the importance of nutritious diet including fruits and vegetables is required to attain long-term outcome of such efforts and policies.

Nutritional Potential of Vegetables

Though the nutrient content in vegetables varies with the variety and hybrids, Toensmeier *et al.* (2020) have reviewed and classified the nutrient concentration cases in major vegetables based on the reference crop nutrient levels, thereby identifying multi-nutrient species to address traditional malnutrition. Each vegetable group contains a unique combination and amount of these phytochemicals, which distinguishes them from other groups for example the *Apiaceae* family (e.g. celery, parsley, carrot) is rich in flavonoids, carotenoids, vitamin

C, and vitamin E. Celery and parsley for example are among the best vegetables sources for the flavonoid apigenin and vitamin E, and carrots have an unique combination of three flavonoids: kaempferol, quercetin, and luteolin. The *Asteraceae* or *Compositae* family (e.g. lettuce, chicory) is rich in conjugated quercetin, flavonoids, and tocopherols. The *Cucurbitaceae* family (e.g. pumpkin, squash, melon, cucumber) is rich in vitamin C, carotenoids, and tocopherols. In a survey of 350 melon accessions of *Cucurbita melo* a 50-fold variation in ascorbic acid content, ranging from 0.7 mg to 35.3 mg/100g of fresh fruit weight was observed. Ascorbic acid and β -carotene content ranged from 7.0 to 32.0 mg/100g and 4.7 to 62.2 μ g/100g, respectively in sweet melons. Bitter gourd (*Momordica charantia*) has anti-diabetic properties and can be used to ameliorate the effects of type-2 diabetes. The *Chenopodiaceae* family (e.g. spinach, Swiss chard, beet greens) is an excellent source of folate and have been shown to inhibit DNA synthesis in proliferating human gastric adenocarcinoma cells.

All the legumes (*Fabaceae* or *Leguminosae* family; e.g. bean, pea and soy-bean), seeds are good sources of dietary fiber and iso-flavonoids. Some legumes are also rich in iron. Cruciferous vegetables (*Brassicaceae* or *Cruciferae* family) which include, cabbage, broccoli, cauliflower, Brussels sprouts, kales, Chinese cabbage, turnip, rutabaga, radish, horseradish, watercress, mustards, among other vegetables, provide the richest sources of glucosinolates in the human diet. Based on one of the largest and most detailed reviews of diet and cancer, the World Cancer Research Fund in USA concluded that a diet rich in crucifers is likely to protect humans against colon, rectum, and thyroid cancers, and when consumed with vegetables rich in other phytonutrients. Crucifers rich in glucosinolates including broccoli, cabbage, Brussels sprouts, and kale have been shown to protect against lung, prostate cancer, breast cancer, and chemically induced cancers.

Alliums vegetables (*Alliaceae* family) include, garlic, onion, leek, chive, Welsh onion, among other vegetables. They are rich in a wide variety of thiosulfides, which have been linked to reducing various chronic diseases. Owing to the presence of prebiotic polysaccharides, which are poorly degraded by the gut enzymes, and the presence of flavonoids, onions have been shown to possess antidiabetic potential. Consumption of *Allium* vegetables has been found to retard growth of several

types of cancers. For instance, there appears to be a strong link between the consumption of onions and the reduced incidence of stomach and intestine cancers.

A study of the factors determining micronutrient bioaccessibility in leafy vegetables revealed that the pectin content of the leaves impaired carotenoid bioaccessibility. Leafy vegetables are rich in condensed tannins, such as drumstick tree (*Moringa oleifera*), had exceptionally low content of pectin and were characterized by high micronutrient bioaccessibility. Therefore, selection and development of cultivars with high micronutrient and low pectin content is a good approach to improve absorption of micronutrients by the human gut.

In the present scenario it is important for diversification towards traditional crops because the current dependence on a few major crops may result in food scarcity. Traditional, underutilized crops, especially those which are locally available and culturally acceptable, are ideally placed to play a much greater role in contributing to improved nutrition and health. These underutilized crops are also referred by other terms such as orphan, under-exploited, underdeveloped, new, novel, promising, neglected, alternative, pseudocrops or local crops. Some of these crops include *Chenopodium*, amaranth, *oca*, *kalazeera*, buckwheat, basella, fenugreek, moringa etc. which are resistant to adverse climatic conditions and can be used to improve food supply as well as income to the growers.

Fresh leaves and tender shoots are consumed as vegetable throughout the world. Several leafy vegetables are produced and consumed in India viz., Amaranth, Spinach beet, Basella, fenugreek, bathua, mustard and other minor crops grown regionally across the country. Green leafy vegetables are rich source of nutrients such as minerals (Fe, Zn, Ca, K, P and Mg), vitamins (vitamins A, K, C, E, and B complex), essential amino acids, antioxidants, phytochemicals and dietary fibre which are beneficial for the maintenance of good health, and prevention of diseases and malnutrition. These foods are ideal for weight management as they are typically low in calories. They are useful in reducing the risk of cancer and heart disease since they are low in fat, high in dietary fiber, and rich in folic acid, vitamin C, potassium and magnesium, as well as containing a host of phytochemicals, such as lutein, β -cryptoxanthin, zeaxanthin, and β -carotene. Because of their high magnesium content and low glycemic index, green leafy

vegetables are also valuable for persons with type 2 diabetes. The high level of vitamin K in greens makes them important for the production of osteocalcin, a protein essential for bone health. Green vegetables are also a major source of iron and calcium for any diet. Green leafy vegetables are rich in beta-carotene, which can also be converted into vitamin A and also improve immune function.

Perennial vegetables are neglected and underutilized class of crops with potential to address current nutritional challenges. They represent 33–56% of cultivated vegetable species, and occupy 6% of world vegetable cropland. Despite their distinct relevance to climate change mitigation and nutritional security, perennial vegetables receive little attention in the scientific literature. Compared to widely grown and marketed vegetable crops, many perennial vegetables show higher levels of key nutrients needed to address deficiencies. Trees with edible leaves are the group of vegetables with the highest levels of these key nutrients. Individual “multi-nutrient” species are identified with very high levels of multiple nutrients for addressing deficiencies. Under exploited vegetables are increasingly been recognized as essential for food and nutritional security. Global diversity in vegetable crops is estimated at about 400 species, with about 80 species of major and minor vegetables are reported to have originated in India. It provides a promising economic opportunity for reducing rural poverty and unemployment and providing farm diversification strategies. The nutritional composition studies could be used to develop strategies to promote the consumption, acculturation and commercialization of these vegetables.

Minor vegetable have immense potential for contribution to a particular pocket's of food production because they are well adapted to existing as well as adverse environmental conditions and are generally resistant to pests and pathogens. Minor legumes may be a cheap, alternate source of protein and can alleviate protein malnutrition among preschool children in rural areas. Nutritious pods of *Parkia roxburghii* are consumed as staple legume vegetable in the NEH region of the country. Similarly, *Mucuna pruriens* is considered one of the most preferred legume vegetables in tribal people of Uttar Pradesh, Bihar and Jharkhand. A vast reservoir of leafy vegetables belongs to group of underutilized vegetables, which are rich source of vitamins, minerals, fiber and diversity in the diet. Furthermore, they have

been a traditional part of cropping systems, especially in home gardens. Their cultivation, utilization and acceptability should not be a problem. Minor vegetables as a whole can, therefore, make an impact on the nutritional status of population, yet among food crops, they are neglected. They are generally low in energy and dry matter content, but immensely important as source of protective nutrients, especially vitamins, mineral and phyto-chemicals. Vegetables are the most important source of vitamin A, which is deficient in most part of the world where rice based diets predominate, blinding thousands of children, annually.

Phytochemicals in Vegetables

Phytochemicals/ phytonutrients / phytonutriceuticals are organic compounds derived from plants that have health promoting properties. Besides the common nutrients such as carbohydrates, amino acids and protein, there are certain non- nutrient phytochemicals in vegetables that have biological activity against chronic diseases. They are low in fat and, like all plant products, contain no cholesterol. Most phytochemicals are found in relatively small quantities in vegetable crops. However, when consumed in sufficient quantities, phytochemicals contribute significantly towards protecting living cells against chronic diseases. Major phytochemicals have been classified in to ten different classes based on their biological activities including carotenoids (α - and β -carotene, β -cryptoxanthin, lutein, lycopene, and zeaxanthin), glucosinolates (sulforaphane, indole-3 carbinol), inositol phosphates (phytate, inositol tetra and penta phosphates), cyclic phenolics (chlorogenic acid, ellagic acid, and coumarins), phyto-estrogens (isoflavones, daidzenin, genistein, and lignans), phytosterols (campesterol, β - sitsterol, and stigmasterol), phenols (flavanoids), protease inhibitors, saponins, and sulfides and thiols.

Vegetables have been shown to protect against specific types of cancer for example, the crucifers (*Brassicaceae*) including Broccoli, Brussels sprouts, Kale and Cabbage have been shown to protect against lung and chemically induced cancers. The alliums (*Liliaceae*), including garlic, chive, and onion have been shown to protect against stomach cancer, the solanaceous vegetables (*Solanaceae*) including tomatoes and pepper have been shown to protect against esophageal, gastric, and prostate cancers. The chenopods (*Chenopodiaceae*) including spinach and chard have been shown to

inhibit DNA synthesis in proliferating human gastric adenocarcinoma cells. There is increasing evidence for a link between antioxidant nutrients (e.g. vitamin C, vitamin E, β -carotene and selenium) in fruits and vegetables and lower risk of cardiovascular disease. Studies have found that 34 per cent reduction in mortality is due to cardiovascular disease among those who consumed vegetables rich in vitamin E and C. A 30-40 per cent reduction in risk of colon cancer in populations with higher vegetable consumption, especially garlic and dietary fiber has also been reported. In addition to reducing cancer and cardiovascular diseases, a diet high in vegetables has also been linked to reducing rheumatoid arthritis, anemia, diabetes, macular degeneration and gastric ulcer. The carotenoids, Vitamin E, and Vitamin C are now firmly established as protective dietary antioxidants with additional beneficial functions. Polyphenols and flavonoids are also gaining prominence and the protective role of folate is above dispute. All of these components are uniquely found in fresh and cooked vegetables, which underline the importance of vegetables in healthy diets. Major antioxidants and polyphenolic compounds present in different vegetable crops has been provided in Table 1.

Source for Minerals and Micronutrients Supplements

Wide ranges of minerals and trace element are present in vegetables. Leafy vegetables and crucifers are rich source

Table 1. Major antioxidants and polyphenolic compound in vegetable crops

Major antioxidants	Sources
β -Carotene	Carrot, fennel, kale, mustard greens, pumpkin, red pepper, lettuce, spinach, sweet potatoes, swiss chard, winter squash.
Vitamin C	Broccoli, brussels sprouts, watermelon, cauliflower, green pepper, red cabbage, red pepper, potatoes
Lutein/ zeaxanthin	Kale, broccoli, spinach, winter squash, brussels sprouts, celery, leeks, mustard greens, peas, green onions, summer squash
Lycopene	Tomato, watermelon
Vitamin E	Dark green leafy vegetables, sweet potatoes
Lipoic acid	Dark leafy green vegetables, especially spinach and broccoli
Flavonoids	Onions, soybeans Onion, lettuce, endive, horse radish, tomato and beans.
Polyphenols	Grapes, nuts, oranges, strawberries, green tea, black tea, red wine
Anthocyanin	Red cabbage, purple broccoli, brinjal, rhubarb, radish, black carrot, onion
Flavones	Celery, tomato, brinjal, garlic, onion
Isoflavone	Soybean, pea, broccoli, asparagus, alfalfa, okra

of minerals and trace element. Calcium, phosphorus and iron are important minerals have major role in bone health and prevention of anaemia. The important trace element found in vegetables are Zn, Cu, Mn, Se played an important role in immune function, body defence against oxidative stress. Interest in selenium as a nutraceuticals has increased dramatically in the last three decades as a results of several studies that demonstrated that increased risk of cancer with low selenium intake. Broccoli and garlic are rich source of selenium when grown on high selenium rich soil. Chromium is another trace element which may be effective in optimizing insulin metabolism and lowering plasma cholesterol levels. On an average a man excretes daily about 20 to 30 g of mineral salts, consisting mostly of chlorides, sulphate and phosphates of sodium, potassium, magnesium and calcium and this output must be recuperated by the intake of food stuffs. In case of the growing body, provision must be made for additional amounts of many of the elements to ensure adequate growth of the tissues. Underutilized vegetables are considered to be a reservoir of various mineral nutrients. Looking into the prevalence of high level of micronutrient malnutrition among the vulnerable sections in the developing countries and the increasing prevalence of chronic degenerative diseases globally, the need for exploration of underutilized foods is significant to overcome the nutritional disorders. The diet and food based approach in combating micronutrient malnutrition is essential for its role in increasing the availability and consumption of micronutrient rich foods. Increasing the utilization of green leafy vegetables (GLV) in our diet, known to be rich sources of micronutrients as well as dietary fiber can be a food-based approach for ensuring the intake of these nutrients. It is essential that the locally available GLV, which are inexpensive and easy to cook, be used in the diets to eradicate micronutrient malnutrition and also to prevent the degenerative diseases.

Medicinal Importance of Vegetables

Naturally occurring nutraceuticals have potentiality to overcome growing problem of non-communicating diseases like cancer, cardiovascular disease, alzheimer, diabetes and so on through there multiple health promoting function. Vegetables are rich sources of diverse group of nutraceuticals with specific health benefit.

Number of minor vegetables possesses several desired medicinal properties. Drumstick is known for its medicinal properties since time immemorial and its

leaves are used by physicians of traditional medicine for the treatment of hypertension. Hypotensive action of alcoholic extract obtained from the dried drumstick leaves in patients with moderate to severe hypertension. The anti-perkinsonian and prolactin reducing effects of minor legume vegetable, *Mucuna pruriens* have been reported. Eating leaves of *Polygonum plebeium* as vegetable improves lactation. Macro and micro-scopic studies on the underutilized leafy vegetable *Alternanthera sessilis* and *Portulaca quadrifida* and the inflorescence analysis of its powder, quantitative microscopy and micro chemical tests have been reported. Antifibrinolytic activity of the roots of minor leafy vegetable *Boerhaavia diffusa* has been found to be due to a phenol glucoside, punarnvoside. It was found effective in controlling IUD induced bleeding in monkeys. List of minor vegetables reported to have significant pharmacological activity is given in Table 2.

Table 2. Underexploited vegetables possessing medicinal properties

Vegetables	Family	Activity
<i>Boerhaavia diffusa</i> L.	Nyctaginaceae	IUCD
<i>Carica papaya</i> L.	Caricaceae	Anti- androgenic
<i>Cissus quadrangularis</i> L.	Vitaceae	Analgesic
<i>Coleus forskohlii</i> (Willd.) Briq.	Labiatae	CNS depressant, Hypotensive Spasmolytic
<i>Costus speciosus</i> (Koen. ex Retz.) Sm	Costaceae	Hypotensive
<i>Cyamopsis tetragonoloba</i> (L.) Taub	Papilionaceae	Hypoglycaemic, Hypolipidaemic
<i>Gymnema sylvestre</i> (Retz.) Schult	Asclepiadaceae	Hypoglycaemic
<i>Mollugo cerviana</i> (L.) Ser.	Molluginaceae	Cardiostimulant
<i>Momordica charantia</i> L.	Cucurbitacea	Hypoglycaemic, Hypolipidaemic
<i>Trianthema portulacastrum</i> L.	Aizoaceae	Analgesic, Antipyretic, CNS depressant

The benefits of cultivating indigenous crops and conserving their germplasm over the introduced HYVs include cultivation of indigenous crops can make agriculture more genetically diverse and sustainable, consumption of domestically cultivated indigenous crops can reduce the carbon footprints and imports, the indigenous crops are highly adapted to the climatic conditions of the land, and consumption of indigenous foods contribute to food diversity and enrichment of diet with micronutrients provides health benefits due to the interactions between the inherited genes and food

nutrients. However, there may be few challenges in reviving indigenous species, which may include farmers' willingness in the propagation of indigenous varieties, identifying the farmers with traditional knowledge of crop cultivation, encouraging the farmers with large landholdings to cultivate indigenous crops, awareness among the consumers and stakeholders about the ecological and health benefits of indigenous varieties, support of the government to the farmers for the propagation of these crops in small and large scale, and development of mechanization suitable for processing indigenous crops, as the existing machines are designed for the HYVs, and employing the same techniques for the processing of indigenous crops may lead to the loss of micronutrients and phytochemicals.

Conclusions

The effective utilization of vegetable genetic resources in breeding programs and testing and deployment of newly developed varieties with tolerance to abiotic stresses and resistance against multiple diseases and insect pests in farmers' fields will ultimately benefit the farming community and consumers. By doing so, a significant reduction or, even better, a complete elimination of the obvious and persistent gap between WHO-recommended and actual intake levels of fruits and vegetables would make a significant contribution to the achievement of Sustainable Development Goals related to food and nutrition security and good health, in particular aiming at doubling agricultural productivity and incomes of small-scale food producers, ensuring sustainable food systems, and maintaining genetic diversity.

References

- Agarwal A, Pandey A, Varaprasad KS, Tyagi RK and Khetrapal RK (2018) Regional Expert Consultation on Underutilized crops for food and nutritional security in Asia and the Pacific. *Indian J. Plant Genet. Resour.* **31(2)**: 194-195.
- Bamji MS, Murty PVVS and Sudhir PD (2021) Nutritionally Sensitive Agriculture-An approach to reducing hidden hunger. *Eur. J. Clin. Nutr.* **75**: 1001-1009.
- Ebert AW (2020) The Role of Vegetable Genetic Resources in Nutrition Security and Vegetable Breeding. *Plants*, **9**, 736; doi.org/10.3390/plants9060736
- <http://nhb.gov.in/StatisticsViewer.aspx?enc=MWoUJibk35dW2g36TUJWAoZqESmAYFi7h2irlsmjIINtCf11rG/kLbq8ZQbWUvuM>
- Jose S, Gulati A and Khurana K (2020) Achieving nutritional security in India: Vision 2030. NABARD Reseach Study-9.
- Nelson ARLE, Ravichandran K and Antony U (2019) The Impact of the Green Revolution on indigenous crops of India. *J.*

- Ethnic Foods*, 6:8; doi.org/10.1186/s42779-019-0011-9.
- Pradyumna A, Mishra A, Utzinger J and Winkler MS (2021) Health in food systems policies in India: A Document Review. *Int. J. Health Policy Manage.* Doi 10.34172/ijhpm.2021.18.
- Rajasekharan PE (2019). Vegetable Genetic Resources: Diversity, Distribution and Conservation. *In: PE Rajsekharan , VR Rao (Eds). Conservation and Utilization of Horticultural Genetic Resources.* Springer Nature Singapore Pte Ltd., Pp: 117-138.
- Singh J, Kalloo G and Singh KP (2001). Vegetable Crops: Nutritional Security. Indian Institute of Vegetable Research Technical Bulletin No. 6.
- Technical Bulletin 83, 84 and 95.
- Toensmeier E, Ferguson R and Mehra M (2020) Perennial vegetables: A neglected resource for biodiversity, carbon sequestration, and nutrition. *Plos ONE* doi.org/10.1371/journal.pone.0234611.
- Ulian T *et al.* (2020) Unlocking plant resources to support food security and promote sustainable agriculture. *Plants People Planet.* DOI: 10.1002/ppp3.10145.

Exploration and Collection of Plant Genetic Resources in India: Status and Priorities

SP Ahlawat*, KC Bhatt, DP Semwal, K Pradheep and OP Dhariwal

ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

Plant exploration and germplasm collection is the foremost activity in PGR management system aimed at augmenting genetic resources for *ex-situ* conservation and utilization by end users. Organized PGR collecting was initiated in India way back in 1946, and so far, 2.82 lakhs accessions representative of nearly 1,695 crops/species have been collected through 2,838 explorations undertaken across the country mainly in collaboration with ICAR crop-based institutes and SAUs. Till two decades back, the focus has been shifted mainly on the collection of cultivated variability which has resulted in fair representative collections for important crops from across India. Now, increasing importance has been entrusted to crop wild relatives, minor fruits and wild edibles species, which has resulted in a phenomenal share of wild germplasm (18%) in the total collection. This communication highlights the collection trends, salient achievements made, emerging issues, and the priority areas which need adequate addressing for meaningful germplasm collections.

Key Words: Crop landraces, Diversity-rich areas, Gap analysis, Germplasm augmentation, Trait-specific germplasm

Introduction

The Indian subcontinent represents four mega biodiversity hotspots, one among 12 megacentres of crop plant diversity in the world, and about 166 crop taxa have originated and/or developed diversity here (Zeven and de Wet, 1982). There are 861 taxa of crop wild relatives documented in India, which includes wild/weedy forms or wild populations of 150 crop taxa in this country (Pradheep *et al.*, 2021). Crops such as rice, sugarcane, green gram, black gram, jute, mango, citrus, banana, cucumber, snake gourd, yam, taro, turmeric, ginger, cardamom, black pepper, jack fruit, horse gram, sesame, okra, and muskmelon, etc. have originated and evolved here. In addition, since antiquity, several new crops were also introduced in Indian agriculture at various times, resulting in constant enrichment of cultivated species, besides many semi-domesticates. A total of 480 crop species of food and agricultural importance and >1000 wild edible plants were documented in this country (Arora and Pandey, 1996; Nayar *et al.*, 2003).

Plant genetic resources (PGR) are one of the most important components of present and future crop breeding programmes. The pattern of crop diversity depends on interactions between plants' genetic makeup and environmental factors including biotic and human factors.

Augmentation of germplasm is the first and foremost activity in the PGR management system. Germplasm collectors rationally apply scientific principles during collection to arrive at an optimum number of samples from a particular site, which is manageable by curators/breeders. More the scientifically-collected diversity is conserved and made available for future use, better the chances of fulfilling future demands. For a successful germplasm collection programme, considerable knowledge of diverse disciplines such as phylogeography, agro-ecology, plant taxonomy and ethnobotany, the biology of crops, and the allied fields is of paramount significance. The threat to loss of diversity, particularly in the wake of anthropogenic pressures and emerging vagaries of climate change, necessitates collecting representative diversity for conservation lest it is lost forever. Besides, germplasm collecting is also aimed for use in breeding programmes, immediate use (e.g., wild edibles), and filling the gaps in existing *ex situ* collections. The germplasm may represent any of the following kinds, namely, wild form, weedy race, landrace/primitive cultivar, obsolete and advanced cultivars, ecotype, botanical form/variety, subspecies, wild progenitor, crossable wild relatives, etc.; this indicates that an explorer generally aims to collect genepool samples. In recent years, increasing interest

*Author for Correspondence: Email-Sudhir.Ahlawat@icar.gov.in

was shown in germplasm collection of wild species of current and potential use.

There were reports of sporadic collections of indigenous crop germplasm during the earlier part of the 20th century. However, systematic plant exploration and collection work was initiated in India with the establishment of a nucleus Plant Exploration and Collection Unit in 1946 in the then Division of Botany, Imperial Agricultural Research Institute, New Delhi under the leadership of Dr Harbhajan Singh, who is also known as 'Indian Vavilov', and had significantly contributed to the field of PGR collection. The period between 1946 and 1976, has also witnessed several national and international collaborative explorations in the country, fetching a sizeable collection of over 31,225 acc. comprising cereals, millets, legumes, oilseeds, vegetables, fibre yielding and other economic plants.

The exploration activities have been more systematized with the creation of NBPGR in 1976. The Bureau with its 10 Regional Stations coordinates/undertakes exploration with crop-based institutes of ICAR, AICRPs and SAUs. The majority of exploration missions during the initial years were of multi-crop/region-specific surveys and collection; however, during the 1980s, collaborative explorations with crop-based institutes received a greater impetus. Priorities were set at both crop/ species level as well as phytogeography/habitat level. After the implementation of the Convention on Biological Diversity in 1993, germplasm was considered sovereign property of the nation. This demanded the urgent need for survey, inventorisation, collection, conservation and documentation of native PGR. Collection activity has gained momentum through the World Bank-aided National Agricultural Technology Project (NATP) on Sustainable Management of Plant Biodiversity, operational at ICAR-NBPGR from September 1999 to 2005 (as reflected in Fig. 1). During the past two decades, the number of annual explorations remains almost unchanged (30-40 in number), but priority has shifted towards collecting trait-specific germplasm and wild germplasm including CWR and minor fruits, which resulted in a significant increase of wild germplasm (18%) and 890 wild species are collected. At the same time, accessions collected per exploration trip was more than 100 per trip till 1992, becoming less than 100 subsequently (see Fig. 1) indicating focused collecting.

Collection Status

Since the inception of NBPGR, a significant amount of diversity in the majority of crops and their wild relatives have been assembled from all the phytogeographic/agroecological regions of the country. So far, a total of 2,81,759 accessions comprising cultivated (2,11,574) and wild species (38,950) have been assembled through 2,838 explorations conducted in collaboration with ICAR crop-based institutes, SAUs, KVKs and other stakeholders (Fig. 1). Crop group-wise holdings indicate that the majority of collections have been made in the cereals (61,496) followed by vegetables (55,353) and pulses (42,162). Collections in other crop groups include 22,879 in millets, 7,447 in pseudocereals, 25,659 in oilseeds, 5,707 in fibres, 14,525 in fruits, 2,046 in fodder species, 28,317 in spices, condiments and M&AP, 2,107 in agroforestry species and 1,235 in sugarcane. The germplasm collections for major crops are given in Table 1.

Details of areas/regions from where germplasm collections made (phytogeography/ crop/ crop-group wise), gap areas (geographic region/crop/crop-group wise), and targeted priority traits, if any, were documented then and there in the past works (Arora, 1988; Malik and Srivastava, 2004; Ahlawat *et al.*, 2015). The Western Ghats, Western Himalayas, arid and semi-arid regions and Andaman & Nicobar Islands were reasonably well explored for crops, while remote and tribal areas of the NEH region, central and eastern India, Eastern Ghats, and Jammu & Kashmir, as well as distinct ecosystems like coastal areas and cold-arid tracts, exhibited some collection deficits, which are being addressed in a phased manner. Collected germplasm represents distinctly named landraces/local cultivars (e.g., rice-14,491; maize-316; pigeonpea-97). In general, more emphasis was given to augmenting orthodox seed-bearing field crops (including vegetables, and seed spices) as they can be bankable in the NGB. However, the functioning of the cryobank facility and the development of adequate protocols have led to the collection of recalcitrant species like citrus, and minor or under-utilized fruits as well. For collection and conservation of perennial horticultural crops (like fruit, plantation, and other tree crops) whose conservation is mainly made through field gene bank, the Bureau is maintaining a strong linkage with the respective NAGS. Salient collection achievements are given in Box 1.

Table 1. Germplasm collection status of major crops

S. No.	Crop-group	Crops (accessions)*
1.	Cereals	Rice (103,538), wheat (21,107), maize (14,221), barley (7,889), oats (619)
2.	Millets	Sorghum (13,742), pearl millet (9,074), finger millet (12,918), foxtail millet (6,142), kodo millet (2,744), barnyard millet (2,510), proso millet (1,854), little millet (765)
3.	Pseudocereals	Grain amaranth (6,817), buckwheat (1,909), chenopod (379), job’s tears (344)
4.	Grain legumes	Chickpea (16,300), pigeonpea (14,436), greengram (7,534), blackgram (5919), French bean (7,295), horsegram-(3,614), lentil (5,028), mothbean (2,672), adzuki bean (162), cowpea (8,807)
5.	Oilseeds	Groundnut (12,014), sesame (10,275), rapeseed & mustard (8,089), soybean (4,185), linseed (3,077), niger (2,857), perilla (583)
6.	Vegetables	Chilli (8,069), okra (4,462), lablab bean (4,115), tomato (1,549), brinjal (6,251), onion (2,931), garlic (1,952), <i>Colocasia</i> (2,739), bittergourd (<i>Momordica</i> spp., 2,360), ridge-gourd (1,593), sponge gourd (1,385), bottle gourd (2,011), pointed gourd (163), watermelon (804), cucumber (1,746), snapmelon (356), pea (5,245), winged bean (305), drumstick (234), yam (81), <i>Dioscorea</i> spp (1,750), pumpkin (1,898), ivy gourd (310)
7.	Fibre	Cotton (14,202), jute (2,948), sunnhemp (419), <i>roselle</i> (1,291)
8.	Fruits and nuts	Bael (127), custard apple (378), chironji (140), <i>Capparis</i> (68), <i>Cordia</i> (130), aonla (183), phalsa (187), karonda (29), khirni (103), <i>Salvadora</i> (147), pomegranate (360), ber (112), sapota (57), peach (115), walnut (764), apple (110), citrus (847), jackfruit (581), water nut (764), papaya (189), apricot (664), pear (102)
9.	Fodder spp.	Sain grass (264), lucerne (352), Sorghum (13,742)
10.	Spices and condiments	Dill (99), celery (129), caraway (45), black caraway (16), coriander (1,778), cumin (560), fennel (181), <i>Nigella</i> (51), anise (19), fenugreek (558), turmeric (2,822), ginger (2,525)
11.	M&AP	Aloe (454), ashwagandha (97), giloe (317), <i>Ocimum</i> (633), kalmegh (222), bhang (27)
12.	Agroforestry spp.	<i>Acacia nilotica</i> (134), <i>Casuarina equisetifolia</i> (4), <i>Eucalyptus</i> (5), <i>Leucaena leucocephala</i> (13)

*This collection figure includes collections from all stakeholders processed for IC numbers

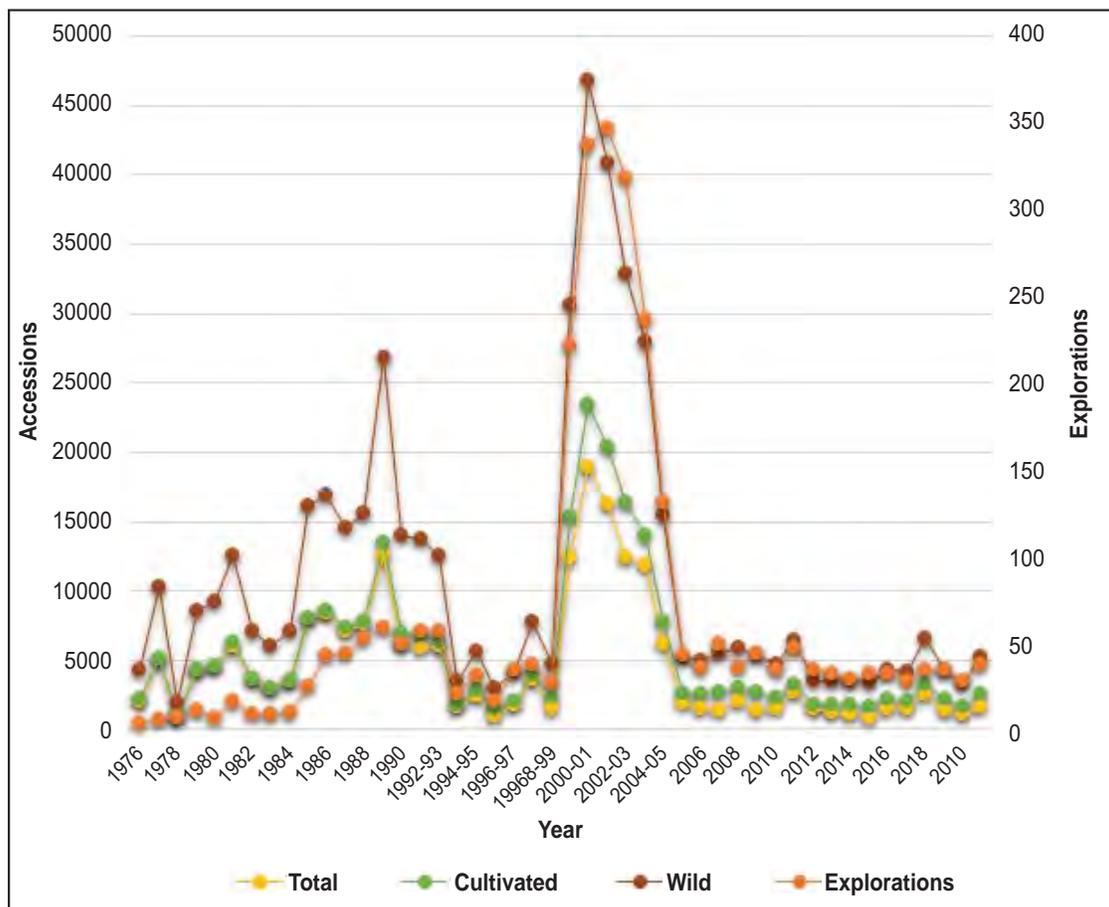


Fig. 1. Trends in germplasm collection since the inception of NBPGR

Box 1. Significant Achievements at a Glance

- From 1966 to prior to the adoption of CBD, joint explorations in collaboration with other countries and CGIAR centres were undertaken under PL-480 scheme, Indo-Australian mission, Indo-Canadian mission, Indo-Japanese mission, INDO-USAID, USIF, G-15, IPGRI, IRRI, ICRISAT, etc. A total of 50,205 samples were collected within India (37,580) and from foreign countries (12,625).
- **Implementation of NATP-Plant Biodiversity:** a unique effort at the national level, wherein more than 130 organizations representing all crop-based Institutes, SAUs, NGOs, etc. collaborated for the national cause of collecting and conserving agro-biodiversity in mission mode approach. A total of 1,733 explorations were undertaken and 1,05,949 acc. were augmented.
- **Trait-specific collections:** Exclusive explorations for biotic and abiotic stress in tolerant germplasm are conducted for rice, wheat, maize, brassica, oilseeds, etc. crops. Some examples include drought/terminal heat tolerant wheat landraces (Kharchia, Lal gehun, Wazia, Khattia) from areas adjoining the southern Thar Desert; grey mildew immune cotton from Tamil Nadu; orange-fleshed cucumber from Manipur and Mizoram; spineless bael from Jaunpur; deep water rice landraces from Assam; salt-tolerant rice landraces from West Bengal. The germplasm for quality traits are collected generally based on the information from farmers.
- **Diversity-rich areas explored/Special Exploration Missions executed:** 16 special missions have been undertaken in different inaccessible areas like A&N islands, Brahmaputra River island (Assam), Sunderbans delta (West Bengal), Valley of Flowers (Uttarakhand), Zanskar valley, Western Ghats (Maharashtra and Karnataka) etc.
- **Rescue missions:** cyclone-hit areas (1999-2000) and flood-affected areas (2008) in Odisha; Tehri dam catchment areas (2000) and natural calamity-affected areas (2013) in Uttarakhand; Earthquake-hit areas of Kachchh (Gujarat, 2001); Sardar Sarovar catchment areas (Gujarat, MP and Maharashtra, 2001-2003).
- **Difficult/disturbed areas explored:** Long duration (45 days) exploration in Nicobar; Nyoma and Zanskar (>3500m) in Ladakh; Gurez and Kistwar in J&K; Malkangiri hills (Odisha); Gadchiroli district (Maharashtra); Bastar, Sukma and Konta (Chhattisgarh).
- **Use of GIS in gap analysis:** Geo-referencing and mapping of diversity vis-à-vis germplasm collected and conserved has been completed in 21 major crops (rice, wheat, maize, chickpea, pigeonpea, *Sesamum*, pearl millet, sorghum, fox-tail millet, finger millet, proso millet, little millet, kodo millet, black gram, mothbean, green gram, cowpea, ricebean, tomato, brinjal, *Allium*) and accordingly gap areas were identified up to district/taluk level. This activity also facilitates the identification of duplicates and updating of passport data. Trait-specific germplasm and potential sites are being identified through grid mapping.
- **PGR Map:** The PGR Map is a web-based and mobile application that provides three uses: “*What’s around me*” helps to obtain the accessions collected and conserved in the genebank from a particular location in India from where the user is accessing; “*Search the map*” helps to list the accessions collected and conserved in the genebank; “*Search for species*” helps to map the collection sites of a species.
- **National Herbarium of Cultivated Plants-:** This Index-Herbariorum accredited herbarium has 25,283 herbarium specimens representative of 267 families, 1,547 genera and 4,380 species of important taxa of PGR relevance including over 500 crop taxa and 550 CWRs. Sixteen new taxa, not represented earlier were added in 2022. Additionally, 3,181 seed samples and 756 economic products/ carpological samples provide representative/referral collections as supplementary holdings. Twenty-five type specimens of 18 taxa (belonging to 7 genera and 5 families) are represented. *Digital images of herbariums* (10,441) were added to the virtual digital database of NHCP at website of NBPGR (<http://www.nbpgr.ernet.in:8080/nhcp/>) and available for reference and studies.
- **Taxonomic/Biosystematic studies:** Studies on the germplasm assembled through various explorations have advanced the knowledge of CWR and their relationship with cultivated species, helped in correcting misidentifications, updating their nomenclature. This has resulted in 24 new plant discoveries and 10 new distribution records in the country, besides 45 records on new distribution in various Indian states. Also, 10 new records of cultivation of economically important species (e.g., *Plukenetia corniculata*, *Inula racemosa*, *Allium fasciculatum*) were made. Illustrated guides for species identification in *Vigna*, *Abelmoschus*, *Cucumis*, *Allium* and *Sesamum* were prepared.
- **Ethnobotanical information compiled:** Ethnobotanical observations and new uses of plants, especially those collected from tribal-dominated tracts, are recorded and published.

Analysis of gaps in the collection in a scientific manner (keeping in view actual variability/ diversity present in habitats, the conserved material, material under regeneration, and best utilization of GIS tools) and pinpointing the pattern of infraspecific diversity through a mission-mode approach is on the way. So far, gap analysis work has been completed for 21 crops and the reports are available online (<http://www.nbgr.ernet.in/Publications.aspx>) gap areas identified through this procedure are being explored/re-explored through Annual Exploration Plans in a phased manner. Pradheep *et al.* (2021) have identified gap areas for 292 high priority-wild taxa (related to 85 crops) at the district level; of them, more than 100 taxa are yet to be augmented from natural habitats.

***In situ* Conservation of PGR**

Locating and declaration of Citrus Gene Sanctuary in Garo hills of Meghalaya was the first attempt of NBPGR on documentation of diversity rich site for *in-situ* conservation during 1980s (Singh, 1981). Subsequently, there was a long gap on this activity, however, PPV&FRA has recognized custodian farmers and awarded them by Genome Saviour Awards. In last three years, the NBPGR has identified high density population sites of wild rice (*Oryza rufipogon*) in Assam, wild pigeonpea (*Cajanus cajanifolius*) in Bailadila range of Chhattisgarh and for guava in Uttarakhand.

The ICAR-NBPGR has located an ideal habitat having good population of wild rice- *Oryza rufipogon* between Borjuli Bagan and Misamari in Sonitpur district of Assam, for its perpetuation and long-term conservation in nature through protection under “The



Biological Diversity Act 2002” involving all stakeholders like State Forest Department, Assam State Biodiversity Board (SBB) and National Biodiversity Authority (NBA), Biodiversity Management Committee, Village Panchayat/ local body. The Government of Assam has declared (on 7th May, 2022) it as “Biodiversity Heritage Site of wild rice”. Probably this may be first initiative of the ICAR on *in-situ* conservation of CWR through BHS.

During an on-farm survey in Pithoragarh district of Uttarakhand, the natural stands of guava in forests and farmer’s fields spreading along the East Ramganga River valley between Muwani to Nachani was located during 2019. Occurrence of guava stands in forests, beyond its cultivation areas in hills was unusual observation. Elderly persons of area could not guess history of its introduction in that area. Detailed survey conducted in 2020 and 2021, revealed naturalised distribution in large area in cultivated and abandoned fields, hill tops and along river bank of Saryu and Kali river valleys, surrounding about 50 villages at 780 m to 1350 m amsl. A large variability in fruit shape, fruit size/fruit weight (100-300g), fruit colour, pulp colour (pink/reddish and



Habitat of naturalised guava population in Pithoragarh district (Left) and fruit variability (Right)

normal) and taste etc. was recorded. A large quantity of fruits is transported by villagers to Pithoragarh, Lohaghat, Champawat and local markets of surrounding areas for sale. The possibility of its *in-situ* conservation and diversity enhancement by introducing other species of *Psidium* is being explored involving all the stakeholders.

Emerging Issues

In recent years, collection activities are being suffered due to the following issues, which may be circumvented at the earliest in the interest of this noble cause of conservation for posterity.

- As farmers are increasingly aware of the registration scope of elite germplasm/unique landraces under the PPVFRA regime, convincing and acquiring germplasm is often a challenge. Also, in some states, vigilant members of the State Biodiversity Boards directing explorers to get permission from their end. Also, the hassles imposed by the forest department to survey and collect PGR from Protected Areas.
- Lack of trained human resources in this field calls for an emphasis on developing skilled manpower and the use of modern tools. Cases on re-demanding the IC numbers to germplasm conserved in FGBs on account of no records, are being received particularly from young or incumbent scientists.
- Sometimes, ‘fancy’ names are assigned to the landraces for gaining popularity, and marketability (apart from different names for the same landrace and vice versa). This necessitates thorough verification through various sources which often consumes time.
- Visible sensitiveness of phenology of wild species due to changes in climate, therefore, making it difficult to strategize the collection activities.
- Losing importance of infraspecific classification in crops (and non-recognizing of infraspecific taxa by taxonomists) resulting in loss of valuable information, which has a huge role in the collection, categorization, characterization, and maintenance of a larger number of samples.
- Lack of interest in multiplication of cross-pollinated crops particularly in vegetables, large fruit with low seeds is a deterrent in germplasm collection of such crops, though criteria of OP vegetable crops have been relaxed from 4000 to 1000 seeds for submitting in NGB.

Priorities and Thrust Areas

- Reducing the collection-to-conservation gaps (Operational gap) through proper coordination, active monitoring and reporting systems, and strengthening the regeneration facilities at the multiplication site, especially for cross-pollinated crops. It is strongly advised to gather an adequate sample size in the field itself, wherever possible.
- Studies on improving storage conditions (including packing) of vegetative propagules and live plants during transport are the need of the hour, as more recalcitrant species, fruit crops and tree species are on the priority list for collection.
- As the importance of collecting wild species including CWR is gaining momentum, it is imperative to develop illustrative field identification keys using leaf, fruit and seed characters.
- Enhancing the effectiveness of collecting trait-specific germplasm through the adoption of FIGS in identifying sites for exploration, encouraging the use of portable equipments/kits (TSS meter, P^H meter, lysimeter, chlorophyll fluorescence meter, NIR food analyser, seed driers, etc.) and the thorough knowledge of habitat/ecological/micro-climate conditions, (pest) hotspot areas, and morphological traits associated with the trait of interest.
- Augmenting representative diversity of relatively less-attended crop groups such as M&AP, forages, ornamentals and green manure, agro-forestry species and also plants belonging to RET species, nutraceutical and industrial value, after required prioritization.
- Similar to the Botanical Survey of India involved in the preparation of flora, development of the PGR Inventory/ checklist for states and country, involving inputs from all possible sources and stakeholders. This would enable identifying meaningful gaps in collections, changes in diversity distribution over time, and aid during collecting. Also, the valuable data lying in the form of field notebooks, reports of collecting missions and collectors’ notes need to be brought out to the benefit of the user.
- Need for a unified collection database involving details of collections by all the stakeholders (ICAR institutes, SAUs and other govt. organizations,

including collections made under different international centres); this only will ensure a better understanding of the germplasm collections on hold, besides meaningful gap analysis.

Other priorities include strengthening expertise in techniques of *in vitro* germplasm collection (which has the advantage of safe movement of germplasm), and the use of species distribution models (like Maximum Entropy, MaxEnt, BioClim, and EcoCrop) for possible localities of populations of rare CWR and RET species. It is also desired to undertake collaborative explorations in foreign countries having primary and secondary centres of diversity of priority crops, for which low diversity exists in national genebanks.

Conclusion

In the current scenario, germplasm collection for utilization is the prevailing trend, and breeders are interested in a few but selected materials showing desirable traits. Hence, it is required to adopt the recent but feasible technologies to maximize the coverage and efficiency of germplasm collection. Though time-consuming, detailed documentation in the field itself during collection is preferred for the fullest utilization of germplasm, besides protecting them legally. Germplasm collections should also need to address specific common problems, which arise then and there (e.g., search for immunity boosting plants/accessions during COVID). Collection priorities need to be reassessed at regular intervals; accordingly, the exploration plan shall be formulated after meaningful gap analysis. A mature decision needs to be taken between recollection and regeneration of accessions with inadequate sample size; and between germplasm collecting from farmers' fields or natural habitats or keeping it *in situ*/on-farm, keeping

in view the pros and cons. Educating citizens through PGR awareness campaign, and partnership approach with grassroot level workers (like those in KVK, and NGO) would help in acquiring desired germplasm in remote and inaccessible areas.

References

- Ahlawat SP, KC Bhatt and K Pradheep (2015). Plant exploration and germplasm collection at ICAR-NBPGR, New Delhi: Status and priorities. In: Pradheep K, A Pandey, KC Bhatt, SP Ahlawat, DP Semwal and KC Bansal (Eds). *Training Manual: Crop Wild Relatives: Identification, Collecting and Utilization*. ICAR-National Bureau of Plant Genetic Resources, New Delhi. pp 1-17.
- Arora RK and Pandey A (1996) *Wild Edible Plants of India: Diversity, Conservation and Use*. National Bureau of Plant Genetic Resources, New Delhi, India.
- Arora, RK (1988) The Indian gene centre-Priorities and prospects for collection. In: RS Paroda, RK Arora and KPS Chandel (eds). *Plant Genetic Resources: Indian Perspective*, NBPGR, New Delhi, pp. 66-75.
- Malik SS and U Srivastava (2004) Germplasm collection, achievements, gaps and opportunities. In: Dhillon BS, RK Tyagi, A Lal and S Saxena (eds) *Plant Genetic Resource Management*. Narosa Publishing House, New Delhi, pp 153–171.
- Nayar ER, A Pandey, K Venkateswaran, R Gupta and BS Dhillon (2003) *Crop Plants of India: A Checklist of Scientific Names*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, India, 48 p.
- Pradheep K, SP Ahlawat, S Nivedhitha, Veena Gupta and Kuldeep Singh (2021) *Crop Wild Relatives in India: Prioritisation, Collection and Conservation*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, 181 p.
- Singh B (1981) Establishment of First Gene sanctuary for Citrus in Garo Hills. Concept Publ. Co., New Delhi, p. 182.
- Zeven, AC and JMJ de Wet (1982) *The Dictionary of Cultivated Plants and their Region of Diversity*. PUDOC. Wageningen.

Crop Wild Relatives in India: Inventorization, Prioritization and Conservation[#]

K Pradheep^{1*}, SP Ahlawat², S Nivedhitha³ and Veena Gupta²

¹ICAR-National Bureau of Plant Genetic Resources, Regional Station, Thrissur-680656, Kerala, India

²ICAR-National Bureau of Plant Genetic Resources, Pusa Campus-110012, New Delhi, India

³ICAR-National Bureau of Plant Genetic Resources, Regional Station, Hyderabad-500030, Telangana, India

Identified as a critical component for food and nutritional security and environmental sustainability in the current century, crop wild relatives (CWR) warrant species-level prioritization and meaningful germplasm conservation. A rough-and-ready approach placing all other species of a crop genus as CWR is unacceptable, as it would lead to the listing of many unrelated species, also conversely, in some crops, the search has gone beyond the crop genera. Initially, based on overall closeness with the crop as well as their potential for use, a total of 861 Indian CWR taxa (769 species) were prioritized for 171 ICAR-mandated crops falling under 14 crop groups. Further prioritization was made on shortlisted taxa, based on the economic importance of crops *per se*, level of closeness to crops (cytogenetically/morphologically/molecularly), possessing traits of breeders' interest, or already under wide-hybridization programme, and the extent of distribution/threat. This resulted in the identification of 292 taxa (257 species) belonging to 85 crops. These high-priority taxa were further analyzed for conservation gaps, if any. Of 292 taxa, only 167 were conserved in the National Genebank. While 28 taxa were represented with only one accession and 81 with <10 accessions, only 40 were with a fair number of accessions (≥ 50); however, they too lacked representative samples from across a geographical and ecological range. This communication highlights the constraints involved in the process including insufficient information on threat status, gap areas for future collection, and thrust areas.

Key Words: Gap analysis, Genebank, Germplasm collection, Threat status assessment, Wild species

Introduction

India is endowed with a rich diversity in crops and it is one of the 12 centres of crop diversity in the world (Zeven and de Wet, 1982). About 166 crop taxa have originated and/or developed diversity in India (Arora, 1991). Indian Gene Centre has particularly contributed to the origin and evolution of crops such as rice, sugarcane, green gram, black gram, jute, mango, citrus, banana, cucumber, snake gourd, yam, taro, turmeric, ginger, cardamom, black pepper, jack fruit, etc. Recently, a few more crops, *viz.*, horse gram, sesame, okra, and muskmelon were added to this list. The rich occurrence of close relatives of these native crops in this country forms important evidence in this regard. In addition, Indian agriculture has been consistently enriched by the introduction of new crops since antiquity, and many species are in the process of domestication. Nayar *et al.* (2003) inventoried 480 crop species in this country.

As long as the breeders' needs are incessant, the available germplasm base in most crops is proven to be insufficient, especially for the stress-related traits such as tolerance to biotic pests (insects, nematodes, pathogens, and weeds) and abiotic factors (salinity, drought, cold, and heat). In this context, wild species related to crops, which are surviving in harsh environmental conditions, marginal lands, and field boundaries, would play a crucial role in offering these much-needed traits. Also, some wild relatives can contribute to yield and quality traits as well. Often, they serve as rootstock to impart resistance/tolerance to abiotic and biotic stresses, increase crop productivity, and help to induce flowering. Though, there has been a steady increase in the rate of release of cultivars containing genes from CWR, most of the CWR are not only in peril in natural habitats in the wake of man-made disasters but are highly under-represented in the *ex situ* genebanks.

[#] This work has been presented at the Second International Agrobiodiversity Congress held online in Nov. 2021, and largely excerpted from the book entitled "Crop Wild Relatives in India: Prioritisation, Collection, and Conservation" authored by Pradheep *et al.* (2021), and published by ICAR-NBPGR

*Author for Correspondence: Email-K.Pradheep@icar.gov.in

According to Maxted *et al.* (2006), CWR can be defined as “a wild plant taxon that has an indirect use derived from its close genetic relationship to a crop”. The closer the species is related to crops, the more the possibility/practicality to get their traits incorporated. Wild forms (i.e., wild but distinct morphotypes belonging to the same taxon in which crop is grouped) or wild populations (i.e., wild plants morphologically indistinguishable from cultivated partners) of crops, wild progenitors, and wild taxa closely related to crop plants, all constitute CWR. Maxted *et al.* (2006) were of the view that closer wild relatives could be found within genepool GP_{1B} (based on crossability) or Taxon Group 1 & 2 (based on infrageneric classification; *genus-tribe-section-series*).

It is to be noted that the common approach of considering all the species of the same crop genus as CWR is not justifiable, especially for large genera encompassing diverse kinds of plants (e.g., *Crotalaria*, *Dioscorea*, *Ficus*, *Ipomoea*, *Panicum*) and in well-researched crops (e.g., barley, wheat, maize, sugarcane); here species of related genera often utilised). Therefore, their prioritization is crucial for meaningful conservation. From the crop improvement angle, the genepool concept needs to be given priority, nevertheless, complete information on crossability is hardly available for most crops. The existence of natural hybrids and successful experimental wide hybridizations indicates that they are closely related. Different species (within the genus) exhibiting the same chromosome number and homology would be rather related. An integrated approach involving morphology, cytogenetics, and molecular systematics, supplemented with allied evidence (graft compatibility, palynology, chemotaxonomy, micromorphology) would help establish the level of relatedness of wild species with the crop.

Earlier, on the basis of evidence from morphology, cytology, crossability, and utility, Arora and Nayar (1984) reported the occurrence of 326 wild relatives in India, which needs a revisit due to a lot of dynamics in species distribution, species concept, threat status, discovery of new taxa since then, and the growing importance of unattended crop-groups such as forages, ornamentals, medicinal and aromatic plants. Latter works (Singh *et al.*, 2013; Pradheep *et al.*, 2014; Singh, 2017), focused more on the distribution and usefulness of the wild species related to crops. The development of checklists and inventories is vital in any national strategy for the

conservation and use of plant diversity (Brehm *et al.*, 2008). Analysing the gap existing between available CWR (in nature) *vis à vis* how many and to how much extent they are conserved (Seed /Cryo/In Vitro/Field Genebank) would help in formulating future exploration plans rationally, including revisiting. Keeping this in view, the research questions set out to answer are as follows:

- How many meaningful CWR occur in India?
- How many of them are conserved *ex situ* and to what extent?
- What issues and constraints are associated with the prioritization and conservation of CWR in India?

Materials and Methods

The information available on wild species related to crops (Arora and Nayar, 1984; Pradheep *et al.*, 2014; Singh, 2017) and new literature (published till February 2021) formed the base for this work. Based on the overall closeness of wild taxon with the ICAR-mandated crops and their usefulness in crop breeding, initial prioritization was made. Those prioritized taxa were further subjected to criteria – economic importance of crops *per se*, level of closeness to crops (cytogenetically/ morphologically/ molecularly), possessing traits of breeders’ interest, or already under the wide-hybridization programme, and the extent of distribution/threat. Wherever feasible, feedback from crop experts was utilized in the prioritization process. The scoring technique adopted for prioritization of CWR taxa (Pradheep *et al.*, 2016) was not followed in this analysis as only incomplete information is available on the criteria such as the extent of distribution/threat (as comprehensive flora of India is yet to be published, and only one-fifth of prioritized taxa were subjected to threat status assessment), and level of closeness. Therefore, only subjective assessments have been made to pinpoint the high-priority CWR taxa. Gap areas for future collection of high-priority taxa were identified as per the steps mentioned in Fig. 1.

Results and Discussion

Initial prioritization of CWR resulted in a total of 861 CWR taxa (769 species) in 171 ICAR-mandated crops (Table 1; for the entire list, see Pradheep *et al.*, 2021), which accounts for about 4% of flowering plants in India. There are 150 crop taxa having wild/weedy forms or populations occurring in India (Table 1), which means these crops have either originated from here or developed

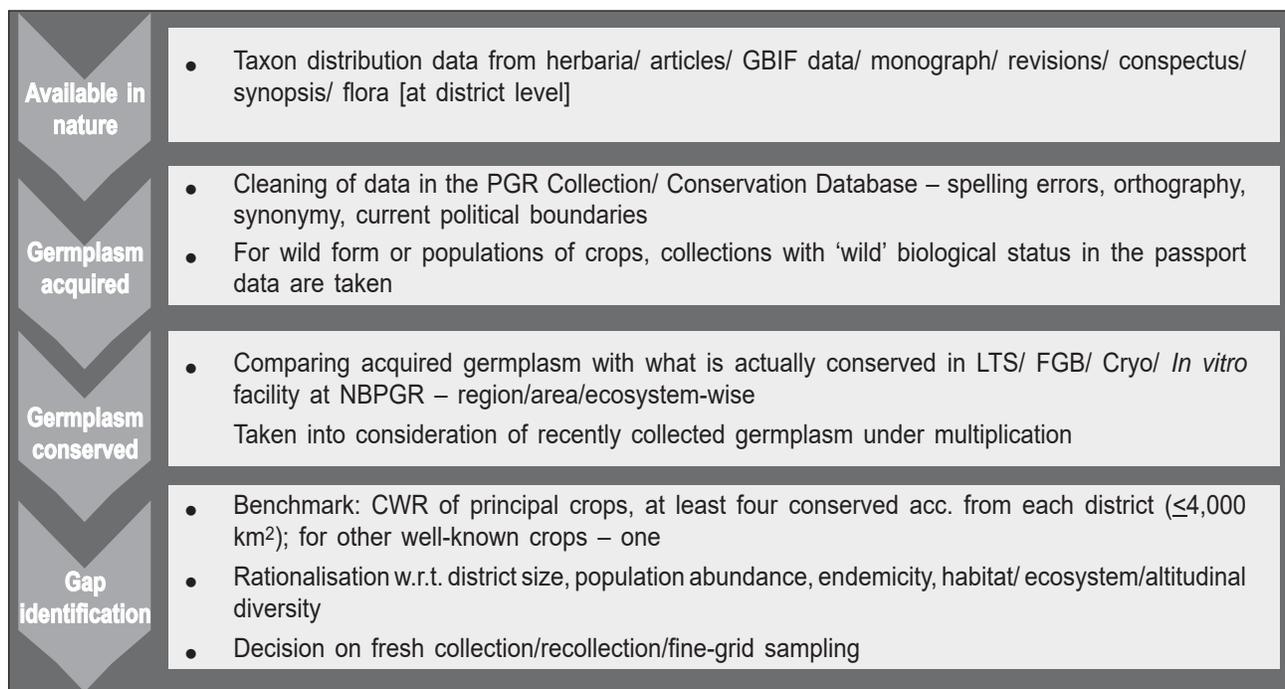


Fig. 1. Gap identification strategy for germplasm collection in high-priority CWR

Table 1. Summary of crop-group wise prioritized crops and their wild relatives for India

S. No.	Crop-group*	No. of crops		CWR species		CWR taxa	
		Initially prioritized	Further prioritized	Initially prioritized**	Further prioritized	Initially prioritized	Further prioritized
1.	Cereals	5	3	52 (2)	46	58	50
2.	Millets	8	5	23 (1)	8	27	9
3.	Pseudocereals	3	1	14 (1)	1	14	1
4.	Grain legumes	10	9	51 (4)	27	59	30
5.	Oilseeds	5	4	13 (1)	10	14	10
6.	Fibres	5	4	19 (3)	9	21	9
7.	Forages	16	4	58 (14)	4	63	5
8.	Fruits and nuts	36	14	130 (17)	55	148	65
9.	Vegetables	26	21	87 (13)	46	102	54
10.	Spices and condiments	12	7	58 (8)	22	62	24
11.	Ornamental plants	13	2	141 (59)	5	153	8
12.	Medicinal & aromatic plants	20	7	74 (19)	8	85	11
13.	Plantation crops	3	1	12 (0)	1	14	1
14.	Others	9	3	37 (8)	15	41	15
Total		171	85	769 (150)	257	861	292

*One crop may involve more than one species **Figures in parenthesis are crop taxa having wild/weedy form(s) or wild populations occurring in India, which are also included for counting as CWR. Source: Pradheep et al. (2021)

a secondary centre of diversity. Crop groups like fruits and nuts, vegetables and medicinal and aromatic plants exhibited ≥ 20 priority crops, while a higher number of meaningful CWR taxa were found in ornamental plants

(141), followed by fruits and nuts (130), vegetables (87) and medicinal and aromatic plants (74), owing to the involvement of higher numbers of crops as well as closely related taxa to these crops.

Further prioritization based on four criteria resulted in the identification of 292 high-priority taxa (257 species) belonging to 85 crops (Table 1; for the list, see Pradheep *et al.*, 2021). Here, crop representatives from groups such as forages, ornamental plants, and fruit and nuts were kept minimum, keeping in view the meagre attention currently given to utilizing wild species in the improvement of crops under these groups. Nevertheless, in forage grasses, a separate exercise was made, resulting in prioritizing 44 wild taxa for 15 crops. In general, biodiversity hotspot regions such as Western Ghats, North Eastern India, Himalayas, Andaman & Nicobar Islands tend to have a greater number of CWR.

Out of high-priority 292 taxa, only 167 are conserved in the National Gene Bank (NGB) of ICAR-NBPGR (including collections at Regional Stations), indicating the need for augmenting other wild relatives, with correct taxonomic identity (Fig. 2). Of yet-to-be collected 125 taxa, a minimum of 30 are endemic, niche-specific, rare and threatened taxa, and some are highlighted in the Box 1. Further, collected species are largely underrepresented, as almost half the conserved species are with <10 accessions on hold. While 28 taxa were represented with only one accession and 15 with just two accessions, only 40 were with a fair number of accessions (between 50 and 559) which includes species like *Abelmoschus tetraphyllus*, *Oryza nivara*, *O. rufipogon*, *Saccharum spontaneum*, *Sesamum indicum* subsp. *malabaricum*, *Solanum insanum*, *Trifolium repens*, *Vigna sublobata* and *Withania somnifera*. However, the latter taxa too lacked representative samples from across a geographical and ecological range. Similar is the scenario in the world as well, and according to Castaneda-Alvarez *et al.* (2016), over 70% of CWR species in the world need collection and conservation in the genebank, and over 95% are insufficiently represented indicating the need for systematic representation from the full geographic and ecological range of species. In the Indian context, huge collection gaps exist even in crucial crop groups (e.g., cereals and pulses), in protected areas (PA) and in fragile ecosystems such as coastal and cold-arid regions.

Some Observations on Various Crop-groups

A perusal of Fig. 2 indicates that even the easy-to- conserve crop groups like cereals, legumes, and vegetables, see huge gaps in species representation. For example, out of 50 prioritised taxa for cereal crops, only 18 were represented. This difference arises mainly due

Box 1. Some rare and endemic CWR yet-to-be represented in NGB

1. *Agropyron thomsonii*
2. *Allium farctum*
3. *Cajanus sericeus*
4. *Cajanus trinervius*
5. *Corchorus urticifolius*
6. *Elymus gangotrianus*
7. *Fagopyrum tataricum* subsp. *potaninii*
8. *Gossypium stocksii*
9. *Hordeum* × *lagunculciforme*
10. *Hordeum spontaneum*
11. *Macrotyloma uniflorum* var. *stenocarpum*
12. *Mangifera sylvatica*
13. *Medicago sativa* notho subsp. *varia*
14. *Musa acuminata* subsp. *manipurensis*
15. *Musa balbisiana* var. *sepa-athiya*
16. *Oryza malampuzhaensis*
17. *Solanum multiflorum*
18. *Sorghum propinquum*
19. *Trichosanthes cucumerina* subsp. *villosula*
20. *Vigna nepalensis*

to a vast number of wild Triticeae members (*Aegilops*, *Agropyron*, *Elymus*, *Eremopogon*, *Hordeum*, *Leymus*) occurring in the Western Himalayas, however, only a few had so far been represented in NGB. When coming to the total wild accessions collected (Fig. 3), vegetables and cereals dominated the collections, as are easily bankable through seeds. Fruits and nuts, ornamental plants, and medicinal and aromatic plants are difficult-to- conserve groups and often require maintenance in FGB. The collection to conservation gap is particularly higher in vegetatively propagated germplasm.

In rice, GP₂ and GP₃ species such as *Oryza meyeriana*, *O. officinalis*, and *O. coarctata* need thorough representation in NGB. In millets, barring *Setaria viridis* (a progenitor of foxtail millet), there is a need to assemble at least the primary genepool of crops, though sizeable numbers of unidentified wild germplasm do exist in *Panicum* and *Pennisetum* genera. There is a need to systematically collect the secondary and tertiary genepools of pigeonpea, apart from its endemic progenitor, *Cajanus cajanifolius*. Only two collections are on hold for the latter species, however, need to have a meaningful collection of at least 25 accessions. *Sesamum prostratum*, a coastal strand vegetation species,

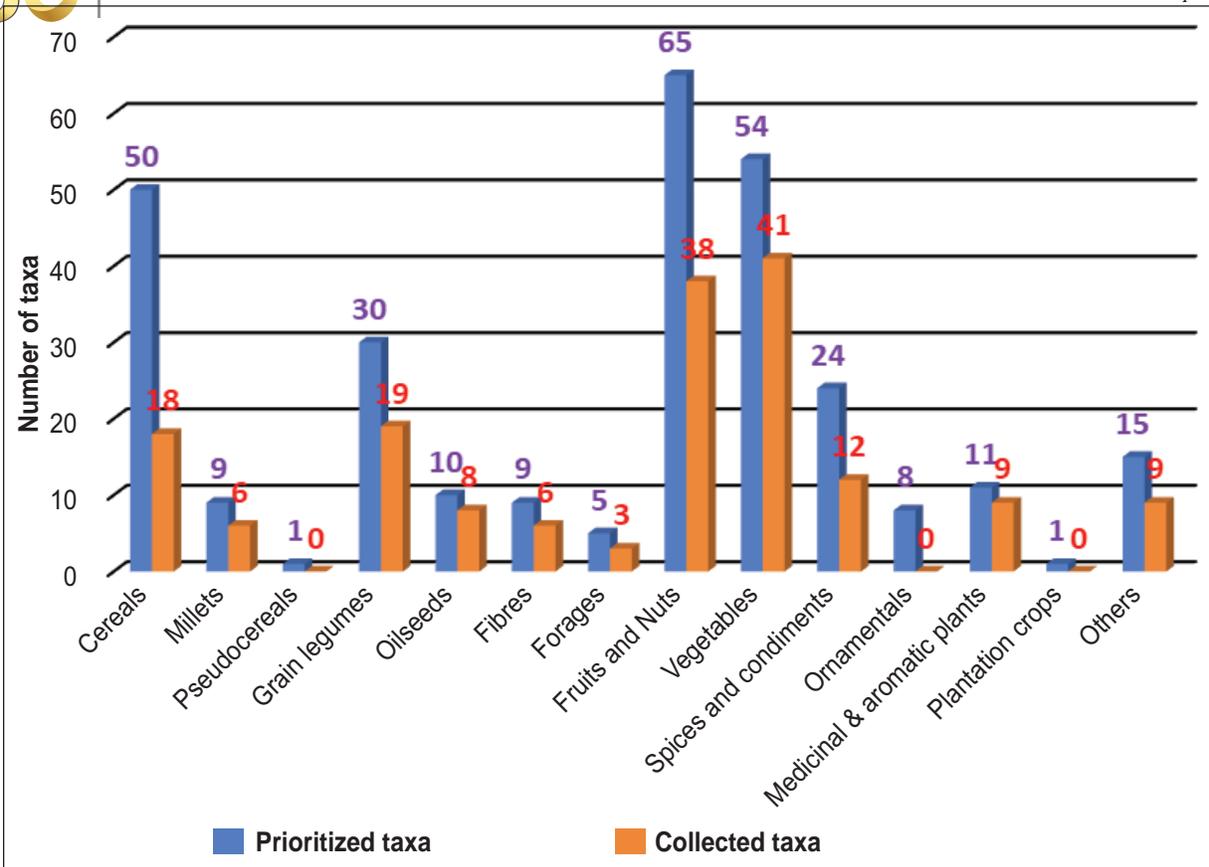


Fig. 2. A crop-group-wise comparison of highly-prioritised vs collected CWR taxa

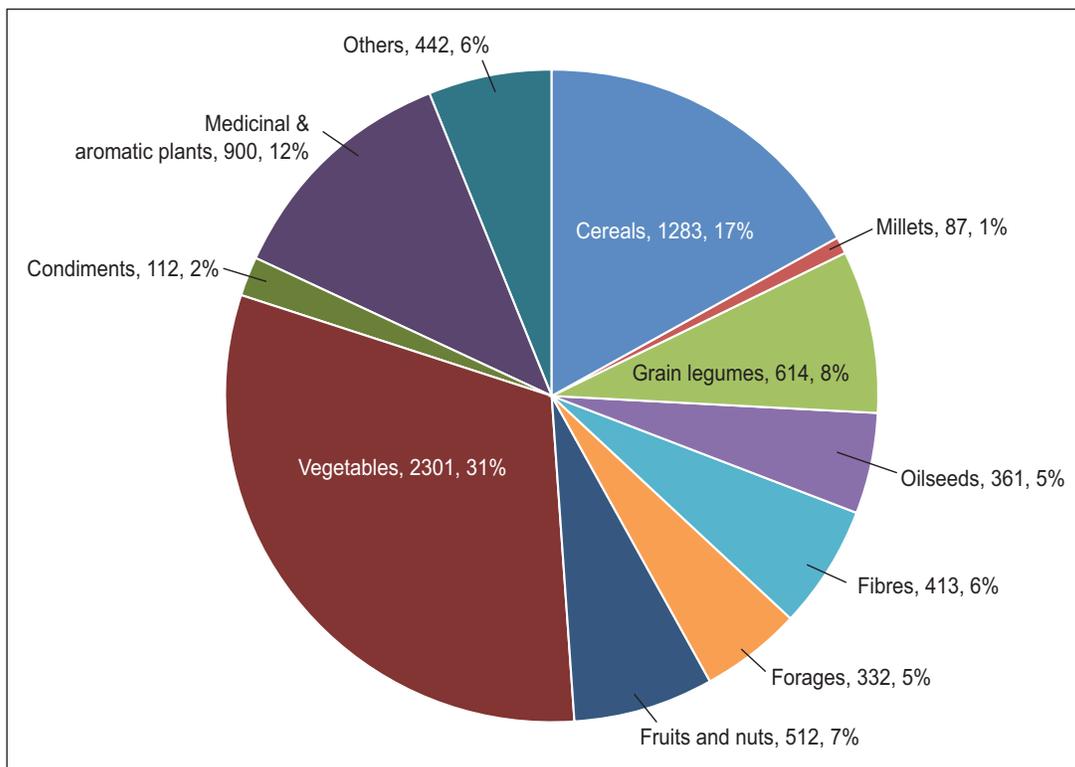


Fig. 3. A break-up of crop-group-wise collected CWR accessions

endemic to the East Coast of southern Andhra Pradesh and northern Tamil Nadu and of Sri Lanka, needs fine grid collecting. In fibre crops, >95% germplasm belongs to wild relatives of jute. A relative of Levant cotton, *Gossypium stocksii* was reported to grow in the Kachchh and Dwarka areas of Gujarat, needing extensive herbaria studies coupled with field surveys to locate and collect.

There exists a huge conservation gap in the case of CWR of horticultural crops (barring vegetables and spices) and forages, as in these cases, even systematic collecting of crop germplasm *per se* needs to be achieved. Often, crops of these groups rarely underwent the domestication process, therefore cultivated forms are hardly distinguishable from the wild ones and are often destructively harvested in the wild (esp. medicinal plants and orchids). In fruit crops, thanks to the systematic explorations undertaken for augmenting *Citrus* gene pool, at the same time, nearly 25 *Musa* taxa are yet to find a place in NGB. In the case of brinjal gene pool, confusion in taxonomic identity of otherwise distinct taxa – *Solanum violaceum* (with *S. anguivi*), *S. lasiocarpum* (with *S. ferox*), *S. insanum* (with *S. incanum*), is often reflected in the collections, besides reports on germplasm utilisation. Systematic studies at Bureau in the crop genera – *Momordica*, *Abelmoschus*, *Cucumis*, *Trichosanthes*, *Allium*, *Luffa* and leafy amaranths had advanced the knowledge of CWR and their relationship with cultivated species.

Important Target Areas for CWR Collection

Taxa-wise collection gaps (292 taxa) were worked out at the district level, and a total of 75 exploration trips were suggested across the country for the next five years (Pradheep *et al.*, 2021). This includes fragile ecosystems such as the cold-arid Himalayas, coastal areas, A&N islands, and Thar deserts, besides some diversity-rich gap areas (see Box 2). Based on geographical significance/uniqueness and richness of wild relatives, 15 PA were also identified for exploration and collection. Apart from representative samples, focused collection through fine-grid sampling shall be undertaken from hotspot areas (for biotic stress tolerance); cold arid Himalayas (cold), Thar desert (heat and drought), and coastal areas (salinity). Keeping in view the nature of wild species (habit/ versatility) and the remoteness of collection locality, the choice shall be made between revisit or seed regeneration in case of germplasm with insufficient seeds aimed for NGB conservation.

Box 2. Some CWR diversity-rich gap areas

- Coastal tracts, esp. East Coast, Gujarat coast
- Himalaya and NEH Region
- A&N, especially unexplored islands
- Western Ghats, esp. central & northern tracts
- Eastern Ghats, especially of Odisha, Karnataka and Tamil Nadu
- Desert areas, esp. Thar desert
- Semi-arid areas (northern and central Karnataka, adjoining Deccan Plateau, semi-arid Tamil Nadu)
- Bundelkhand region
- Duars & terai belt
- Bastar-Vizag-Malkangiri-Koraput-ranges
- Vindhya-Satpura Ranges in central India
- Chotanagpur belt of Jharkhand

Important Issues and Thrust Areas

- Shortage of trained manpower in basic disciplines such as taxonomy and cytogenetics resulting in taxonomic misidentification, and paucity of chromosomal data and crossability, respectively. Taxonomic studies/revisions and the development of illustrated keys for field identification of various wild species are worth-considering.
- Incomplete knowledge of the level of closeness of CWR, their distribution, threat status and usefulness. Crossability studies would aid in the realization of the gene pool concept in crops, helpful in knowing the closest relatives. IUCN Red List has data of just 19.5% of prioritized taxa, therefore threat status assessment needs to be geared up for remaining wild species.
- Wild forms of crops need a clear entry in passport data (either in the 'biological status' field of passport data or distinguished as different infraspecific taxon). New information emanated from the studies needs to be incorporated then and there, particularly the taxonomic identity corrections including new species if any described from using existing germplasm holdings.
- Often, not all the collected/studied germplasm go for long-term conservation due to multifarious reasons.

Pradheep *et al.* (2015) identified bottlenecks leading to low seed turnout during CWR collecting and suggested means to minimise the same. Rigorous monitoring/interaction between the stakeholders during seed regeneration/ initial field establishment (in the case of vegetative propagules) would be helpful in its realisation.

Other areas which need strengthening include knowledge of the population structure of wild species, studies on conservation/storage behaviour, protocol standardization for *in vitro* (RET spp.) and pollen cryopreservation (helpful in wide hybridization). Getting blanket permission for PA entry and effective use of GIS tools would aid in representative sampling across diverse habitats. The development of an integrated database of germplasm holdings inclusive of the SAUs and allied stakeholders would aid in comprehensive gap analysis.

Conclusion

In the context of the requirement of novel genes for crop improvement, sophisticated biotechnological tools amenable to transfer useful traits to crops, increasing anthropogenic pressure and negative impact of climate change on wild species, and meagre *ex situ* germplasm collections, any effort to inventorize, prioritize, collect and conserve CWR is of paramount significance in agricultural research works. This paper identifies 861 priority taxa (769 spp.) of CWR value for 171 Indian crops and further narrowed 292 high-priority taxa for 85 important crops. About 43% of high-priority taxa were yet to be represented while 15% were represented by 1-2 acc., and 28% with <10 acc. in genebank indicating the lack of adequate representative collections in most of the taxa. Huge collection gaps were identified even in cereals and pulses, in PA and in fragile ecosystems, and accordingly, exploration trips were proposed. Working on the thrust areas identified herewith would be of help in the proper management of CWR germplasm in the country.

References

- Arora RK and ER Nayar (1984) *Wild Relatives of Crop Plants in India*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, India, 90 p.
- Arora, RK (1991) Plant Diversity in the Indian Gene Centre. In: RS Paroda and RK Arora (eds) *Plant Genetic Resources Conservation and Management: Concepts and Approaches*, International Board for Plant Genetic Resources, New Delhi, pp. 25-44.
- Brehm JM, N Maxted, BV Ford-Lloyd and MA Martins-Loucao (2008) National inventories of crop wild relatives and wild-harvested plants: case-study for Portugal. *Genet. Resour. Crop Evol.* **55**: 779-796.
- Castañeda Álvarez NP, CK Khoury, HA Achicanoy, V Bernau, H Dempewolf, RJ Eastwood, L Guarino, RH Harker, A Jarvis, N Maxted and JV Müller (2016) Global conservation priorities for crop wild relatives. *Nature Plants* **2**: 1-6.
- Maxted N, B Ford-Lloyd, SL Jury, SP Kell and MA Scholten (2006) Towards a definition of a crop wild relative. *Biodiv. Conserv.* **14**: 1-13.
- Nayar ER, A Pandey, K Venkateswaran, R Gupta and BS Dhillon (2003) *Crop Plants of India: A Checklist of Scientific Names*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, India, 48 p.
- Pradheep K, DC Bhandari and KC Bansal (2014) *Wild Relatives of Cultivated Plants in India*. Indian Council of Agricultural Research, New Delhi, 728 p.
- Pradheep K, KC Bhatt and ER Nayar (2015) Problems in collection and conservation of some crop wild relatives in India: An analysis. *Int. J. Biol. Sci. Eng.* **6**: 73-77.
- Pradheep K, KC Bhatt, DP Semwal and ER Nayar (2016) Prioritisation of crop wild relatives for collection and conservation from coastal areas of India. In: T Ramkumar, G Ramesh and S Vasudevan (eds.) *Coastal Groundwater: Modern Observations*. Orchid Books Pvt. Ltd., Chennai. pp. 153-165.
- Pradheep K, SP Ahlawat, S Nivedhitha, V Gupta and K Singh (2021) *Crop Wild Relatives in India: Prioritisation, Collection and Conservation*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, India, 181 p. Accessible at <http://www.nbpg.ernet.in/Downloadfile.aspx?EntryId=9221>
- Singh AK (2017) *Wild Relatives of Cultivated Plants in India: A Reservoir of Alternative Genetic Resources and More*. Springer, Singapore, 309 p.
- Singh AK, RS Rana, B Mal, B Singh and RC Agrawal (2013) *Cultivated Plants and their Wild Relatives in India – An Inventory*. Protection of Plant Varieties & Farmers' Rights Authority, New Delhi, 215 p.
- Zeven, AC and JMJ De Wet (1982) *Dictionary of Cultivated Plants and their Regions of Diversity*, 2nd ed. Centre for Agricultural Publishing and Documentation. Wageningen, The Netherlands, 263 p.

Plant Taxonomy and Biosystematics in PGR Collecting and Conservation

K Joseph John¹, K Pradheep¹ and E Roshini Nayar²

¹ICAR-National Bureau of Plant Genetic Resources, Regional Station, Thrissur, Kerala-680656, India

²Ex. ICAR- National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

A sound understanding of the principles and practices of plant taxonomy is essential for plant genetic resources (PGR) conservation. Identification of new crop wild relatives (CWR) taxa, designating appropriate taxon status for discontinuous variants representing a geographical area, developing descriptors for new crops, and recruiting wild economic plants and semi-domesticates into cultivation, all require a good taxonomic base. The taxonomy of cultivated plants is essentially governed by a different set of guidelines under International Code of Nomenclature for Cultivated Plants (ICNCP). Biosystematic analysis helps to elucidate the genepool relationship of wild relatives with the crop, thereby promoting its utilization in crop improvement.

Key Words: Crop wild relatives, Genepool, Germplasm, ICNCP, Plant identification

Introduction

Plant taxonomy is the practice and science of identification, naming and classification based on well-laid-out principles. Correct botanical identity of a taxon is a prelude to its utilisation in research programmes as the wrong taxonomic identity of a plant specimen may lead to spurious results and utter confusion. With the current emphasis to include more and more wild economic plants into the domain of agriculture and forestry, correct botanical identity becomes more important for research information pertaining to the taxon, and their efficient utilisation. The Indian subcontinent with four mega biodiversity hotspots (<https://www.cepf.net>) has 861 taxa of crop wild relatives recorded at present (Pradheep *et al.*, 2021). Ensuring the correct botanical identity of the specimen and ensuring universality in the nomenclature of newly collected material is important in the utilisation of this treasure for agricultural improvement. The wrong botanical identity of genebank samples could have far-reaching consequences as the ‘original error’ is repeated at many levels by varied users over a long period of time. In order to avoid such problems and in the light of the rapidly decreasing number of expert taxonomists, and in the present era of access to digital records of historic herbarium specimens and literature, a germplasm collector needs to develop plant identification skills and have an understanding of nomenclatural rules, to ensure accuracy of passport data and other initial records.

Taxonomic issues for a plant genetics resource

(PGR) worker with agricultural science background are many. The foremost is designating unique morphological variants of CWR taxa into appropriate taxonomic entities and determining their taxonomic status as distinct species or infraspecific taxa. Ignoring discontinuous variants is dangerous in genebank conservation as precious variants may get unrepresented in core collections as a result of generalisation. Frequent nomenclatural changes on the basis of molecular study and phylogenetic analysis are another taxonomic issue daunting the non-classical taxonomists dealing with CWR taxa. Many are not aware of these nomenclatural changes in economically important species (e.g., tomato, pearl millet, Chinese potato, etc.). More important, the substantiating evidence for merging taxa, needs to be thoroughly checked by the PGR scientist (discussed below), especially in the case of genera with very high species diversity, rare species which are poorly represented in herbaria, taxa with minute floral parts (e.g., Poaceae), species complexes (e.g., *Oryza rufipogon-nivara*- f. *spontanea*), cryptic species and the like.

Understanding patterns of variation in crop plants and PGR, in general, was dependent on the collection and study of cultivated plants superimposed by that of wild and weedy forms (de Candolle, 1883), domestication traits, and use of wild taxa and progenitors leading to defining centres of diversity of crop taxa (Vavilov, 1922); thus, a nearly 150-year long knowledge of PGR diversity in relation to geography forms the basis of collection and

*Author for Correspondence: Email-josekattukunnel@gmail.com

exchange of germplasm, its use in breeding programmes and bringing into cultivation a wide range of locally used taxa in 12 mega-centres of agro-diversity all over the world (Zeven and Zhukovsky, 1975). Phylogenetic study and biogeographic analysis based strongly on newer evidence provide further clues to understanding the cultivation and domestication processes over space and time. Genepools of crop plants as defined by Harlan and de Wet (1971), though having an experimental basis related to a relative degree of crossability among crop and wild species of seed-producing predominantly annual crops, provided a concept that used morphological, cytological, phytochemical and molecular evidences to determine the wild relatives belonging to a wide range of crop genepools.

Vavilov's Law of Homologous Series (Vavilov, 1922), determined regularities in the expression in respect of the morphological and physiological characters, which distinguish varieties and races (Nanjundiah *et al.*, 2022) as well as parallel variations seen in species of the same genus; he demonstrated its predictive value for variation across related genera, *viz.*, wheat, barley and rye, a trend observed later in the systematic study of other taxa, *viz.*, *Phaseolus* and *Vigna* (Smartt, 1990).

Precise taxonomic data helps to estimate the ecogeographic representativeness of crop genepool captured in a genebank (Dempewolf *et al.*, 2014), thereby helpful in devising future collection strategies, as CWR are, in general, under-represented in genebanks across the globe (Castañeda-Álvarez *et al.*, 2016). Updated taxonomic knowledge helps in the prioritization of taxa, sorting out spelling errors, orthography, mismatches in common vs scientific names, synonymy, wrong identity, misapplied names in passport data and collection, and conservation databases, including the PGR portal. Taxonomy has a clear role in preparing species inventories, red lists, identification of protected areas, gene sanctuaries, and biodiversity heritage sites. At the same time, taxonomic data is critical as well, for example, IUCN Red List or CITES has earlier named a species under their list, but because of synonymy, if this species has been merged with the related species, funding (if any) to protect its population would be in question.

Taxonomy and PGR Management

A sound understanding of plant taxonomic principles is essential in PGR management, right from exploration and

collection of germplasm to conservation/documentation. Plant identification in their natural habitats needs good observation skills, field knowledge, and regular practice. Explorers perceive a wide range of taxa while on survey and collection. They need to distinguish, on the spot, target species from umpteen number of other species at the collection site. In this regard, key or spot characters in combination with habitat information available in past collection records (*viz.* in floras, monographs and treatises) are an aid to the collector. For example, if an explorer intended to collect germplasm of wild species of *Trichosanthes*, he could look for medium-sized climbers with fringed white flowers and red cricket ball-sized fruits, which are very unique and discernible traits even at a distance, and sitting in a fast-moving vehicle.

Expertise in field identification, even in the absence of flowers or fruiting specimens, rather based on available plant parts is often required for CWR collection as in many cases, by the time the seed matures, flowering might have stopped or the plant may have started to wilt and dry. The study of herbarium specimens and spirit collections/ carpological specimens are essential for the plant exploration of a specific CWR group. Publication of handy field aids like seed atlas, fruit atlas, and illustrated field guides with keys will be useful for CWR explorers (Pandey *et al.*, 2011). Habitat information including associated species is very important, especially when collecting niche-specific taxa/wild species necessitating the need for the taxonomic expertise of a collector.

Recircumscription of taxa as a result of revisionary and systematic studies is a common feature in taxonomy. PGR scientist has to make a decision based on their taxonomic knowledge and as Charles Darwin rightly suggests that “in determining whether a form should be ranked as a species or variety, the opinion of naturalists having sound judgement and wide experience seems the only guide to follow”.

Morphological variations are the basis of taxonomic classification. In fact, characterization is essentially describing an accession based on a set of morphological characters called descriptors and descriptor states. The development of descriptors for crop species needs a sound understanding of morphology, which is essentially a taxonomic element. A sound understanding and application of taxonomic /morphological knowledge will help to take out unique variants and non-matching entities during the process of characterization.

Field-level practices like identification and removal of weedy or off-type plants/rogues, mechanical admixtures, rootstock vs scions, and pollinizers, all require taxonomic knowledge. Undesirable genetic contamination due to crossability with progenitors/close CWRs (especially transgenic crops) can be conveniently sorted out using taxonomic expertise. Traditional taxonomy is still the forerunner, though the latest developments in molecular techniques (such as DNA barcoding and molecular systematics) provide substantiating and supporting evidence, and in cases such as introgression, disjunct variants within a taxon help to resolve confusion as well as determine the probable evolutionary trend. Selecting *in situ* conservation sites, demarcating genetic reserves within protected sites, and environmental impact analysis, all require adequate knowledge of the region's flora.

Taxonomic knowledge on invasive and quarantine weeds in terms of their accurate identification is a must to enable the blacklisting of species for entry to the country. Adequate awareness is essential of the changes in nomenclature, and familiarity with common synonyms used worldwide for a particular species. Here, the updated seed atlas of weeds of quarantine significance and mimicry weeds is helpful.

Updated and precise taxonomic identification and labelling of accessions form the foundation for various genebank management activities such as organizing germplasm collections and retrieving the accessions. Taxonomic units such as genus, and species, are part of the documentation systems of genebanks and information retrieval. Wherever taxonomic information up to the

infraspecific level is available, documentation systems should be in a position to accommodate the same. Similarly, it is always a good practice to include author citation as a part of documentation systems including journals/ reports as there are cases with the same name proposed by different authors. There needs a mechanism to swiftly correct the taxonomic identity of germplasm if later proved to be misidentified or there is a need to redesignate with the new taxon name (in the case kept under a 'broad' species name). Correct spelling (including avoiding orthographic errors, and correct terminal ending of epithet) is a must for database entry (for that IPNI offers the best), otherwise, more than one entry will be there for the same species. Knowledge of synonyms and adequate awareness of changes in nomenclature is a must for the proper documentation of germplasm collections. Guzzon and Ardenghi (2018) suggested a taxonomic and nomenclatural peer-review process to ensure taxonomic authenticity before making each new accession accessible to different stakeholders.

ICAR-NBPGR has been instrumental in pursuing taxonomic work on CWR taxa for the past four decades which led to 24 new plant discoveries and 10 new distribution records in the country, besides numerous records on new distribution in various Indian states (Box 1; Table 1). The enormous number of diverse species represented in two major facilities at the Bureau, *viz.*, National Genebank (2,000 species; 0.5 million germplasm accessions) and National Herbarium of Cultivated Plants (4,300 species; 25,000 herbarium specimens) further signifies the role of taxonomists in PGR management works.

Table 1. First report by NBPGR of PGR taxa in India based on their natural occurrence

S.No.	Taxon	Remarks
1.	<i>Corchorus pseudo-olitorius</i> Islam & Zaid	Described from Pakistan; now from Delhi and Tamil Nadu; a CWR of jute
2.	<i>Cucumis muriculatus</i> Chakrav.	Described from Myanmar, now reported from Mizoram; a CWR of cucumber
3.	<i>Dioscorea piscatorum</i> Prain & Burkill	Lesser Sunda Islands native species was reported from Nicobar; wild edible tuber used by Nicobaris
4.	<i>Fagopyrum gracilipes</i> (Hemsl.) Dammer ex Diels	Chinese native species reported from Arunachal Pradesh; weed in buckwheat fields
5.	<i>Momordica subangulata</i> Blume subsp. <i>subangulata</i>	Indo-Chinese taxon was located in Arunachal Pradesh and Nagaland; a CWR of teasel gourd
6.	<i>Rubus praecox</i> Bertol.	European species reported from Jammu & Kashmir; a minor fruit related to blackberry
7.	<i>Trichosanthes dunniana</i> H.Lév.	Indo-Chinese species documented from Manipur, Mizoram, Nagaland
8.	<i>Trichosanthes tricuspidata</i> Lour.	Indo-Chinese species reported from A&N islands; <i>T. bracteata</i> was earlier misidentified in Indian literature
9.	<i>Trichosanthes wallichiana</i> subsp. <i>subrosea</i> (CY Cheng & CH Yueh) K Pradheep & KJ John	Indo-Chinese taxon reported from NE India. New nomenclatural combination made
10.	<i>Ziziphus subquinquenervia</i> Miq.	Indonesian species was reported from Great Nicobar; minor fruit

Box 1. New Taxa Described by NBPGR

1. *Abelmoschus angulosus* Wight & Arn. var. *mahendragiriensis* RC Misra
2. *Abelmoschus enbeepeegearensis* K.J. John, Scariah, Nissar, KV Bhat & SRYadav
3. *Abelmoschus palianus* Sutar, KV Bhat & SR Yadav
4. *Abelmoschus pungens* var. *mizoramensis* KJ John, Krishnaraj & K Pradheep
5. *Allium negianum* A.Pandey et al.
6. *Begonia bachulkarii* Aitawade, Kattuk & SR Yadav
7. *Cucumis melo* subsp. *melo* var. *alwarensis* A Pandey & S Rajkumar
8. *Curcuma amada* var. *glabra* Velay, Unnikr, Asha & Maya
9. *Curcuma karnatakensis* Amalraj, Velay & Mural
10. *Curcuma kshonapatra* Velay
11. *Curcuma kudagensis* Velay, VS Pillai & Amalraj
12. *Curcuma longa* var. *vanaharidra* Velay, Pandrav, JK George & Varap
13. *Curcuma malabarica* Velay, Amalraj & Mural
14. *Curcuma nilamburensis* Velay, Mural, Amalraj, PL Gautam & S Mandal
15. *Curcuma thalakaveriensis* Velay, Amalraj & Mural
16. *Curcuma vellanikkarensis* Velay, Mural, Amalraj, PL Gautam & S Mandal
17. *Herpetospermum operculatum* K Pradheep, A Pandey, KC Bhatt & ER Nayar
18. *Momordica cochinchinensis* (Lour.) Spreng. subsp. *andamanensis* Kattu, Roy & Krishnaraj
19. *Momordica sahyadrica* KJ John & VT Antony
20. *Momordica sahyadrica* subsp. *anamatayana* KJ John, K Pradheep & Krishnaraj
21. *Piper pseudonigrum* Velay & Amalraj
22. *Trichosanthes dunniana* H Lév. subsp. *clarkei* K Pradheep
23. *Vigna konkanensis* Latha, KV Bhat, IS Bisht, Scariah, KJ John & Krishnaraj
24. *Vigna sahyadriana* Aitawade, KV Bhat & SR Yadav

Cultivated Plant Taxonomy

Taxonomy and nomenclature of cultivated plants especially within the crop taxon is an entirely different proposition from that of wild plants. While up to species level cultivated plant nomenclature is governed by ICN guidelines like any other plants, designating infraspecific categories like ‘cultivars’ man-made hybrids, chimaeras etc. are governed by an entirely different set of rules under the ICNCP. With private sector breeding initiatives progressing at a faster pace, many new

cultivars/varieties in perennial horticultural plants and vegetatively propagated crops like tuber crops, banana, potato, sugarcane, fruit trees and ornamentals are bound to be developed and along with that IPR related legal issues, which demand proper naming of new cultivars.

Capacity Building

A strong plant taxonomic base was felt and stressed by expert committees like RAC & QRT of ICAR-NBPGR. Considering these suggestions, recently ICAR-NBPGR has signed an MoU with Botanical Survey of India (BSI) for fostering mutual cooperation in various aspects of plant taxonomic research related to PGR. Further, in order to enhance the taxonomic knowledge base of PGR scientists, especially plant identification skills, the ICAR-NBPGR conducted a one-week online training programme on Plant Taxonomy for the benefit of entry-level scientists of the national agricultural system.

Suggestions for Furthering Taxonomic Studies in PGR

1. An attachment training on taxonomy and morphology for all newly recruited PGR workers commissioned to work in the PGR discipline. About three months of attachment in premier institutes/labs (such as BSI, CSIR-NBRI, FRI, Shivaji University, Kolhapur, and other centres of excellence in plant taxonomy) is desirable. A collaborative programme between scientific societies such as ISPGR and the Indian Association of Angiosperm Taxonomy (IAAT) may be resorted to bridge the knowledge gap in applied taxonomy.
2. Currently working PGR scientists need hands-on taxonomy training (2-3 weeks) at regular intervals (in 5 years) in key deficit areas such as plant identification skills including short-cut keys for discerning different families and genera, spot characters for identification of CWR taxa in the field, botanical nomenclature, and cultivated plant taxonomy. International taxonomy training needs to be encouraged for a few senior-level scientists of the bureau. NBPGR authorities need to groom strong expertise in the taxonomy of CWR/cultivated plants and their infraspecific classification, and an MoU with appropriate institutes (such as CSIR-NBRI, IPK-Gatersleben) may be contemplated. A designated taxonomist at the national level positioned at NHCP-NBPGR/PPVFRA is a dire need to address taxonomic issues in cultivated plants as well as

- promote research in this comparatively neglected branch.
3. In collaboration with the expert institutes, it is dire need of the hour to bring out authentic checklists, interactive keys and illustrated plant identification guides (with the inclusion of fruit, seed, and vegetative characters) for different plant/ crop groups/ economically important genera to faster realization of correctly-identified wild germplasm for using in PGR and crop improvement works. It includes redesignating existing germplasm collections with correct taxonomic identity and accepted names. Also, focus on identification using seeds, seedlings and vegetative characters for use in genebank management may be given attention.
 4. Revamping PG syllabus for PGR course, especially incorporating more credit hours for enhancing plant identification skills as a sizeable percentage of fresh ARS recruits for the discipline of Economic Botany and PGR are from agricultural science background.
 5. Undertaking comparatively basic taxonomic studies (especially systematic studies for delineating CWR taxa) in unattended crop groups such as medicinal and aromatic plants, ornamentals and forages, after adequate prioritization as part of dissertation work of M.Sc. (PGR). Voucher herbarium specimen of experimental study material needs to be safeguarded to ascertain their taxonomic identity in future.
 6. Digital databases, viz., World Checklist of Vascular Plants (WCVP), Plants of the World Online (POWO) and Global Biodiversity Information Network (GBIF) are currently being used as a 'ready reckoner' for accepted plant names. There is a need to recognise that these, in contrast to a data collator such as International Plant Names Index (IPNI), are compilations of historical data

on plant nomenclature, and as such should be used as a source of information, rather than authentic and verified taxon names. PGR workers should be trained to make use of these sources to check the latest nomenclature.

References

- Candolle A de (1886) *The Origin of Cultivated Plants*. International Scientific Series **49**: Second Ed. 1904, London.
- Castañeda-Álvarez NP, CK Khoury, HA Achicanoy, V Bernau, H Dempewolf, RJ Eastwood, L Guarino, RH Harker, A Jarvis, N Maxted and JV Müller (2016) Global conservation priorities for crop wild relatives. *Nature Plants* **2**(4): 16022.
- Dempewolf H, RJ Eastwood, L Guarino, CK Khoury, JV Müller and J Toll (2014) Adapting agriculture to climate change: a global initiative to collect, conserve, and use crop wild relatives. *Agroecol. Sustain. Food Syst.* **38**(4): 369–377.
- Guzzon F and NM Ardenghi (2018) Could taxonomic misnaming threaten the *ex-situ* conservation and the usage of plant genetic resources? *Biodivers. Conserv.* **27**(5): 1157–1172.
- Harlan JR and De Wet JMJ (1971) Toward a rational classification of cultivated plants. *Taxon* **20**: 509-517.
- Nanjundiah V, R Geeta and VV Suslov (2022) Revisiting N.I. Vavilov's "The Law of Homologous Series in Variation" (1922) *Biol. Theory*. <https://doi.org/10.1007/s13752-022-00403-3>
- Pandey A, DC Bhandari and K Pradheep (2011) Plant taxonomy in plant genetic resource management. In: R Gupta (ed) *Plant Taxonomy: Past, Present, and Future*. The Energy and Resources Institute (TERI), New Delhi, pp. 129-140.
- Pradheep K, SP Ahlawat, S Nivedhitha, V Gupta and K Singh (2021) *Crop Wild Relatives in India: Prioritisation, Collection and Conservation*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, 181 p.
- Smarrt J (1990) *Grain Legumes: Evolution and Genetic Resources*. Cambridge University Press, Melbourne, 379 p.
- Vavilov NI (1922) The law of homologous series in variation. *J. Genetics* **12**: 47–89.
- Zeven AC and PM Zhukovsky (1975) *Dictionary of Cultivated Plants and Their Centres of Diversity*. Wageningen, Netherlands: Centre for Agricultural Publishing and Documentation, 219 p.

Application of Geographical Information System for PGR Management

N Sivaraj^{1*}, SR Pandravada¹, Anuradha Agrawal², V Kamala², V Celia Chalam³ and K Anitha¹

¹ICAR-National Bureau of Plant Genetic Resources, Regional Station, Hyderabad-500030, Telangana, India

²Agricultural Education Division, ICAR Headquarters, Pusa Campus, New Delhi-110012, India

³1-19-62, HIG B-1, A.S. Rao Nagar, Hyderabad-500062, Telangana, India

⁴ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

Geographical information system (GIS) tools have enormous applications in management of plant genetic resources (PGR). GIS in conjunction with passport/herbarium/gene bank databases serves in geo-referencing, diversity distribution mapping and predicting suitable sites for future collection of crop species and threatened taxa. They have been used in ecogeographic surveys for locating diversity, planning field exploration and collecting of PGR. They are also useful for identification of PGR conservation areas and individual species, species-rich areas and vegetation types that are not represented or under represented under conservation programs. In terms of biosecurity, GIS applications can be effectively used in locating hotspots, spread of pests and pathogens, develop early warning systems, build risk assessment models, assist in site specific protection measures etc. These technologies and applications developed in response to a wide range of needs are nowadays an indispensable tool in the management of PGR.

Introduction

Plant genetic resources (PGR) are most important for the continued existence and interests of humanity. A vast number of domesticated plant species are crucial to global food security, while other species are of great importance for purposes such as wood and bio-fuel production. In addition to the cultivated species, many wild plants still play an important role in meeting local needs for food, fuel, medicine and construction materials; crop wild relatives are also of special interest for crop breeding programmes. There are currently hundreds of underutilized plant species and varieties displaying traits of interest to meet present and future needs, while the value of many other plant species is yet to be discovered. Amongst the plethora of new age tools available for PGR management, geographical information system (GIS) is of immense importance for the improved understanding and monitoring of germplasm.

A GIS integrates hardware, software, and data for capturing, managing, analyzing, and displaying all forms of geographically referenced information. GIS allows one to view, understand, question, interpret, and visualize data in many ways that reveal relationships, patterns, and trends in the form of maps, globes, reports, and charts (www.esri.com). Thus, GIS is a database management system that can simultaneously handle data representing spatial objects and their attribute data. GIS

can be effectively used in PGR management particularly in a) inventorisation/ mapping, b) collecting strategies, c) conservation strategies, d) characterization and evaluation, e) crop expansion strategies, f) documentation and g) plant quarantine.

Inventorisation/Mapping Strategies

Inventorisation/ Mapping Strategies Priority Action 1 of Global Plan of Action (GPA) calls for increased surveying and inventorying of plant genetic resources for food and agriculture. Datasets of PGR with attributes, identity and geo-reference data of relevant point locations are prerequisites for GIS mapping. Some of the GIS Software used for mapping are Arc-GIS, FloraMap, DIVA-GIS, Quantum GIS, GARP, ECOSIM, Degree, Climex, MaxEnt, HyperNiche, SAM, SPADE, gvSIG, Geo Da, ECOGEO, GEOQUAL etc.

GIS mapping has been successfully used in assessing biodiversity and in identifying areas of high diversity in *Phaseolus* bean (Jones *et al.*, 1997); coconut (Bourdeix *et al.*, 2005); wild potatoes (Hijmans and Spooner 2001); wild arachis (Jarvis *et al.*, 2003); horsegram (Sunil *et al.*, 2008); *Jatropha curcas* (Sunil *et al.*, 2009); linseed (Sivaraj *et al.*, 2009; 2012); sesame (Spandana *et al.*, 2012), blackgram (Babu Abraham *et al.*, 2010); rapeseed-mustard (Semwal *et al.*, 2012); piper (Parthasarathy *et al.*, 2008); *Canavalia* fatty acids (Sivaraj *et al.*, 2010); medicinal plants (Varaprasad *et al.*, 2007); *Spondias*

*Author for Correspondence: Email-N.Sivaraj@icar.gov.in

(Miller *et al.*, 2006), common bean (Sheikh Sultan *et al.*, 2014) and agrobiodiversity in general for South East coastal India (Varaprasad *et al.*, 2008).

Collecting Strategies

Germplasm exploration and collecting are planned generally based on available databases of passport information. Passport information includes an identity to the collection, specific location of collection, details of habit/ habitat and other reference data. GIS can be effectively used in preparing distribution maps of species, probable location of the collection sites, gap analysis, analyzing diversity rich pockets etc. GIS can be used to link the passport database with district and state map layers to analyse what has been explored and collected and from where, and what are the gaps in germplasm collection (Fig.1). GIS and other specialized computer program (e.g. FloraMap) along with associated data can be used to map the predicted distribution of plant species or areas of possible climatic adaptation of organisms in the wild (Jones *et al.*, 2002). Also, GIS can play an important role in the management of large and complex PGR datasets (Guarino *et al.*, 2001). Guarino (1995) discusses the use of GIS to develop strategies for collecting germplasm. For example, collecting regions can be mapped to identify areas with desired ecogeographic attributes such as acid soils or climate extremes (Hart *et al.*, 1996). Thus, to plan future exploration programs which are trait specific/region specific GIS can be used effectively (Jones *et al.*, 1997; Greene *et al.*, 1999). Remote sensing satellite temporal data (time interval) in digital form can be used in impact

assessment (by overlaying of different geospatial layers) studies, temporal changes to pinpoint status of collected threatened taxa diversity to find gaps and predict new areas for collecting.

Mapping the spatial distribution of target species along with the prevailing knowledge systems of communities can be effectively carried out using GIS. Traditionally tribal communities and farmers have been the custodians of PGR and developed huge knowledge systems over years. Indigenous traditional knowledge (ITK) associated with PGR, their time of cultivation, system of cultivation, its relation to the environment form a vital part of the tribal communities. Such knowledge systems which co-evolved with the nature provide the food and nutritional security of the tribal communities. Documentation followed by validation of PGR related ITK would make available such secure sources of ethnic systems to be harnessed for benefit of all.

Conservation Strategies

Complementary conservation strategies include the protection of wild species, plant populations and traditional crop varieties where they have evolved (*in situ* conservation), with the collection and preservation of inter- and intraspecific diversity in gene banks and botanical gardens (*ex situ* conservation). *Ex situ* genetic resource collections maintain germplasm in the form of seed or live plants, representing current, obsolete and primitive crop varieties, wild and weedy relatives of crop species, and wild species collected or augmented from around the world. GIS can be effectively used for genetic resources conservation in (i) identifying gaps for

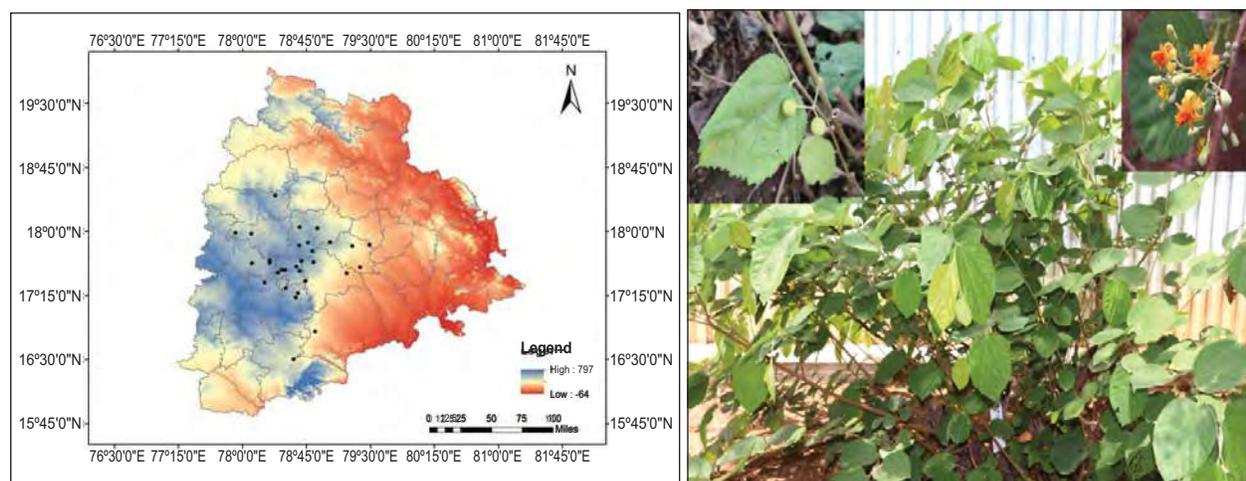


Fig. 1. Collection sites of *Phalsa*, an underutilized fruit crop from Telangana State, India

conservation in both *ex-situ* and *in-situ* and (ii) design and management of *on-farm in-situ* conservation sites. Further GIS, climate change models and geographical distribution data of crop plants and their wild relatives may be used to predict the impact of a changing climate on PGR, conservation and use. Geographical prediction of threatened plant distribution is important to genetic resource conservation planning and regional management decisions. Geospatial technologies are useful in predicting the spatial distribution of crop landraces and other target threatened species. It assesses multiple interdependent abiotic factors, e.g., solar radiation, air temperature, precipitation, and soil properties affecting plant distribution, models the environmental niches of target plants and refines their distribution maps for conservation planning.

PGR Characterization and Evaluation

Precise characterization and evaluation of PGR are a pre-requisite for their utilization. Germplasm utilization requires accurate characterization, evaluation and documentation of the material. PGR characterization and evaluation adds value to the germplasm and thereby, facilitates its utilization (Gautam, 2004). Largely germplasm evaluation involves the whole range of activities including germplasm multiplication, characterization, preliminary and detailed evaluation, regeneration, maintenance and documentation (Gautam,

2004). In all these activities GIS can play a crucial role by way of managing large data sets, identifying suitable locations for multiplication and evaluation of germplasm. In addition, identifying suitable locations for evaluating crops introduced from other countries could be done using GIS. Morphological descriptors/genetic variations may be linked with environmental attributes using GIS for selecting potentially useful germplasm accessions (Pederson *et al.*, 1996). Data on physiological, morphological or genetic descriptors can be added to a punt map to investigate the spatial distribution of diversity (Guarino, 1995)

Crop Expansion Strategies

GIS can play a crucial role by way of managing large data sets, identifying suitable locations for multiplication and evaluation of germplasm introduced from other countries. Morphological descriptors/genetic variations may be linked with environmental attributes using GIS for selecting potentially useful germplasm accessions for crop expansion (Pederson *et al.*, 1996). Ecological niche modelling studies on horticultural crops (Ceylon spinach, sorrel, roselle) using GIS had been worked out by Reddy *et al.* (2015a, b, c). Exotic germplasm of several agri-horticultural crops (tef, kiwi, olive, dragon fruit etc.) can be introduced into suitable agro-climatic regions of our country after assessment using GIS (Fig. 2)

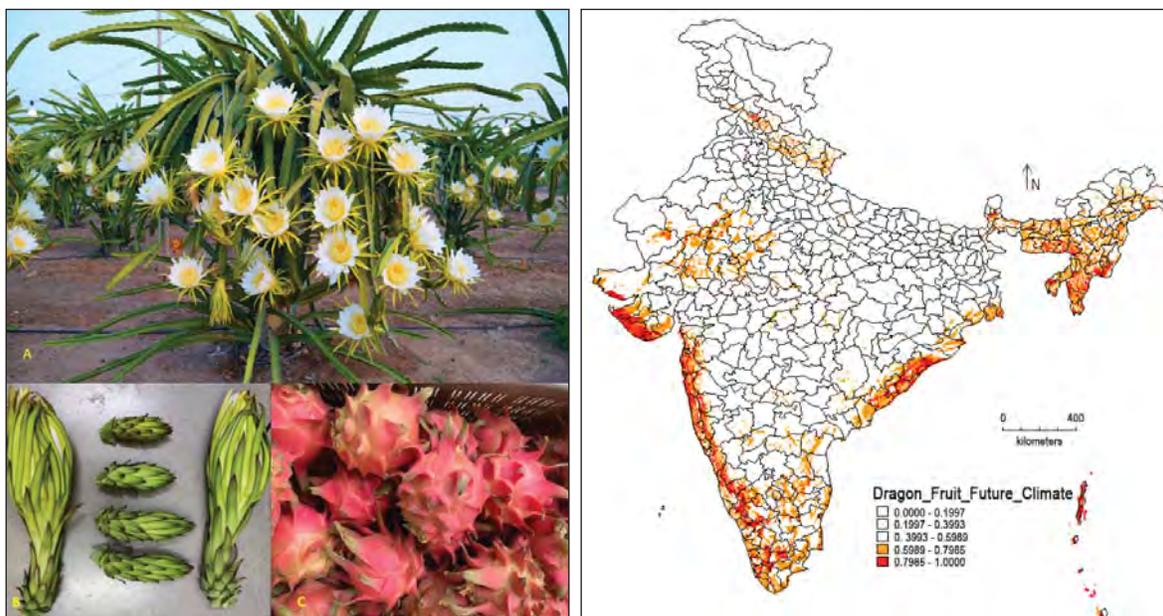


Fig. 2. Ecological niche model of climate suitable regions for dragon fruit cultivation in India (Future climate)

PGR Documentation

Stephanie L. Greene and Thomas Hart state that “*Genetic resource databases could potentially serve as an information resource to a broader scientific community, providing relatively independent global data sets to meet the unique needs of environmental modelers. Multi-disciplinary collaboration would ensure the evolution of a set of databases useful to a broad range of professions*” (<http://www.ars-grin.gov>.) Genetic resource databases available with ICAR-NBPGR, ICAR-IIHR, and other agri-horticultural crop-based institutes in India could potentially serve as an information bank to the PGR researchers, providing the large PGR datasets to meet the requirements of breeders, environmental specialists. The information from these datasets can be used to generate and validate agro-ecological models developed and to predict the agronomical potential of germplasm accessions of agri-horticultural crops. Based on the existing records indicating the historic availability of the various crops and their wild relatives, the distribution and use of crop species including adoption of new cultivars or new crop species could be assessed. Passport information of the germplasm accessions including the presence of associated species can be useful in preparation of predicted distribution maps of wild or cultivated species.

Plant Quarantine Strategies

GIS has been applied for pest monitoring and detection through data visualization and query, survey data collection, management, and risk pathway analysis. It can also help to determine areas that have highest risk for a pest introduction. Prediction models can be generated for detection in case of invasive alien species and to quantify area change in their spread. The use of GIS in PQ is important as it provides economic benefit, is a proactive approach in safeguarding agriculture, helps in quality control assistance and decision support system. Recent examples include (i) Adult grasshopper hazard analysis (in USA) to identify patterns in grasshopper survey that may predict future population increases, as related to environmental conditions (*i.e.*, climate, soil, vegetation) (ii) to identify areas most likely to have Asian gypsy moth and to improve and/or validate existing AGM trapping locations (in USA) (iii) Sugarcane woolly aphid spread in southern India and (iv) mapping of pest distribution data (from pest interceptions of import quarantine)

Future Thrusts

- GIS can be used in gap analysis, planning and execution of future exploration programs at national level for effective conservation and utilisation of threatened PGR.
- PGR passport data information, satellite data spectral signature and climate analogue tools could be used in diversity distribution mapping and prediction of diversity rich areas.
- GIS coupled with hyperspectral remote sensing can be used in distinguishing and identification of threatened PGR for effective conservation measures.
- High spatial resolution (60 cm) satellite data can be used in mapping of disease symptoms in threatened species at fine grid level.
- Ecological niche models/species distribution models for all the crop landraces and other plant genetic resources to be constructed for effective conservation in the changed climatic regime.

References

- Abraham B, Kamala V, Sivaraj N, Sunil N, Pandravada SR, Vanaja M, Varaprasad KS (2010) DIVA-GIS approaches for diversity assessment of pod characteristics in black gram (*Vigna mungo* L. Hepper). *Curr. Sci.* **98(5)**: 616-619.
- Bourdeix R, Guarino L, Mathur PN and Baudouin L (2005) Mapping of coconut genetic diversity. In: Pons Batugal, Ramanatha Rao V, Jeffrey Oliver (eds) *Coconut Genetic Resources*. IPGRI. pp. 32-43.
- Dikshit N, Abdul Nizar, M and Sivaraj N (2012) Evaluation and diversity analysis of safflower germplasm in relation to morpho-agronomic characteristics. *J. Oilseeds Res.* **29** (Special Issue): 17-23.
- Gautam PL (2004) Trends in plant genetic resource management. In: Dhillon BS, Tyagi RK, Arjun Lal (eds) *Plant genetic resource management*. Narosa Publishing House, New Delhi pp 18-30.
- Greene S, Hart T and Afonin A (1999) Using geographic information to acquire wild crop germplasm: II. Post collection analysis. *Crop Sci.* **39**: 843-849.
- Guarino L (1995) Geographic information systems and remote sensing for the plant germplasm collector. In: Guarino L, Ramanatha Rao V and Reid R (eds) *Collecting Plant Genetic Diversity. Technical Guidelines*. CAB International, Wallingford, UK, pp. 315-328.
- Guarino L, Jarvis A, Hijmans RJ and Maxted N (2002) Geographic information systems (GIS) and the conservation and use of plant genetic resources. In: Engels JMM, Rao VR, Brown AHD, Jackson MT (eds) *Managing Plant Genetic Diversity*. IPGRI, Rome, Italy pp 387-404.

- Guarino L, Maxted N and Sawkins M (1999) Analysis of geo-referenced data and the conservation and use of plant genetic resources. In: Greene SL and Guarino L (eds) *Linking Genetic Resources and Geography: Emerging Strategies for Conserving and Using Crop Biodiversity*. CSSA Special Publication No. 27. ASA and CSSA, Madison, Wisconsin, pp. 1-24.
- Hart TS, Greene SL and Afonin A (1996) Mapping for germplasm collections: Site selection and attribution. Proc. Third Int. Conf. 'Integrating GIS and environmental modeling'. NCGIA, Santa Barbara, CA.
- Hijmans RJ and Spooner DM (2001) Geographic distribution of wild potato species. *Amer. J. Bot.* **88**: 2101-2112.
- Hijmans RJ, Guarino L, Cruz M and Rojas E (2001) Computer tools for spatial analysis of plant genetic resources data 1. DIVA-GIS. *Plant Genetic Resourc. Newslett.* **127**: 15-19.
- Jarvis A, Ferguson ME, Williams DE, Guarino L, Jones PG, Stalker HT, Valls JFM, Pittman RN, Simpson CE and Bramel P (2003) Biogeography of wild *Arachis*: assessing conservation status and setting future priorities. *Crop. Sci.* **43**: 1100-1108.
- Jones PG, Beebe SE, Tohme J, Galway NW (1997) The use of geographical information systems in biodiversity exploration and conservation. *Biodiversity and conservation* **6**: 947-958.
- Jones PG. and Gladkov A (1999) FloraMap: A Computer Tool for the Distribution of Plants and Other Organisms in the Wild. CIAT, Cali, Colombia.
- Jones PG, Guarino L, Jarvis A (2002) Computer tools for spatial analysis of plant genetic resources data: 2. FloraMap. *PGR Newsletter* **130**: 1-6.
- Miller, Allison J, Knouft JH, (2006) GIS-based characterization of the geographic distributions of wild and cultivated populations of the Mesoamerican fruit tree *Spondias purpurea* (Anacardiaceae). *Am. J. Bot.* **93**: 1757-1767.
- Parthasarathy U, George J, Saji KV, Srinivasan V, Madan MS, Mathur PN, Parthasarathy VA (2008) Spatial analysis for Piper species distribution in India. *Plant Genetic Resources Newsletter*. 147:1-5.
- Pederson GA, Fairbrother TE, Greene SL (1996) Cyanogenesis and climate relationships in U.S. white clover germplasm collection and core subset. *Crop Sci.* **36**: 427-433 99
- Reddy MT, H Begum, N Sunil, SR Pandravada, N Sivaraj and S Kumar. 2015a. Mapping the climate suitability using maxent modeling approach for Ceylon spinach (*Basella alba* L.) cultivation in India. *J. Agric. Sci.* **10(2)**: 87- 97.
- Reddy MT, Begum H, Sunil N, Pandravada SR, Sivaraj N. (2015b) Assessing climate suitability for sustainable vegetable Roselle (*Hibiscus sabdariffa* var. *sabdariffa* L.) cultivation in India using MaxEnt model. *Int. J Agric. Biol.* **1(2)**: 62-70.
- Reddy MT, Begum H, Sunil N, Pandravada SR, Sivaraj N and Kumar S. (2015c) Predicting potential habitat distribution of sorrel (*Rumex vesicarius* L.) in India from presence-only data using Maximum Entropy Model. *Open Access Library Journal*, 2: e1590. <http://dx.doi.org/10.4236/oalib.1101590>
- Sheikh Mohammad Sultan, Sher Ahmad Dar, Suheel Ahmad D and Natarajan Sivaraj (2014) Diversity of common bean in Jammu and Kashmir, India: a DIVA geographic information system and cluster analysis. *Int. j. appl. nat. sci.* **6(1)**: 226-233.
- Semwal DP, Bhandari DC, Bhatt KC and Ranbir Singh (2012) Diversity distribution pattern in collected germplasm of Rapeseed-Mustard using GIS in India. *Indian J of Plant Gene. Resour.* **26(1)**: 76-81.
- Sivaraj N, Sunil N, Pandravada SR, Kamala V, Vinod Kumar, Rao BVSK, Prasad RBN, Varaprasad KS (2009) DIVA-GIS approaches for diversity assessment of fatty acid composition in linseed (*Linum usitatissimum* L.) germplasm collections from peninsular India. *India. J. Oilseeds Res.* **26**: 13-15.
- Sivaraj N, Sunil N, Pandravada SR, Kamala V, Rao BVSK, Prasad RBN, Nayar ER, Joseph John K, Abraham Z, Varaprasad KS (2010) Fatty acid composition in seeds of Jack bean [*Canavalia ensiformis* (L.) DC] and Sword bean [*Canavalia gladiata* Jacq.]DC] germplasm from South India: A DIVA-GIS analysis. *Seed Technol* **32(1)**: 46-53.
- Sivaraj N, Sunil N, Pandravada SR, Kamala V, Vinod Kumar, Babu Abraham, Rao, BVSK, Prasad RBN and Varaprasad KS (2012) Variability in linseed (*Linum usitatissimum*) germplasm collections from peninsular India with special reference to seed traits and fatty acid composition. *Indian J. Agric. Sci.* **82 (2)**: 102-105.
- Sivaraj N, Pandravada SR, Dikshit N, Abdul Nizar M, Kamala V, Sunil N, Chakrabarty SK, Mukta N, Varaprasad KS (2012) Geographical Information System (GIS) approach for sustainable management of Safflower (*Carthamus tinctorius* L.) genetic resources in India. *J. Oilseeds Res.* **29** (Special Issue): 45 - 49.
- Spandana B, Sivaraj N, John Prasanna Rao G, Anuradha G, Sivaramakrishnan S, Farzina Jabeen (2012) Diversity analysis of sesame germplasm using DIVA-GIS. *Journal of Spices and Aromatic Crops* **21(2)**: 145-150.
- Sunil N, Sivaraj N, Anitha K, Babu Abraham, Vinod Kumar, Sudhir E, Vanaja M and Varaprasad K S (2009) Analysis of diversity and distribution of *Jatropha curcas* L. germplasm using Geographic Information System (DIVA-GIS). *Genetic Res. Crop Evol.* **56**: 115-119.
- Sunil N, Sivaraj N, Pandravada SR, .Kamala V, Raghuram Reddy P, Varaprasad KS (2008) Genetic and geographical divergence in horsegram germplasm from Andhra Pradesh, India. *Plant Genetic Resources: Characterization and Utilization* **7(1)**: 84-87.
- Varaprasad KS, Sivaraj N, Mohd Ismail. Pareek SK (2007) GIS mapping of selected medicinal plants diversity in the Southeast Coastal Zone for effective collection and conservation. In: Janardhan Reddy K, Bir Bahadur, Bhadraraiha B, Rao MLN (eds) *Advances in Medicinal Plants*. Universities Press (India) Private Ltd, pp 69-78.
- Varaprasad KS, Sivaraj N, Pandravada SR, Kamala V, Sunil N (2008) GIS mapping of Agrobiodiversity in Andhra Pradesh,. Proceedings of Andhra Pradesh Akademi of Sciences. Special Issue on Plant wealth of Andhra Pradesh pp 24-33.
- Vavilov NI (1951) *The Origin, Variation, Immunity and Breeding of Cultivated Plants*. Ronald Press Company, New York.

Role of Plant Quarantine in Preventing Entry of Exotic Pests

V Celia Chalam*, Kavita Gupta, MC Singh, Z Khan, J Akhtar, BH Gawade, Pooja Kumari, Pardeep Kumar, BR Meena, AK Maurya and DS Meena

Division of Plant Quarantine, ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

Plant quarantine is a government endeavour enforced through legislative measures to regulate the introduction of planting material, plant products, soil and living organisms, etc. in order to prevent inadvertent introduction of pests (including fungi, bacteria, viruses, nematodes, insects and weeds) harmful to the agriculture of a country/state/region, and if introduced, prevent their establishment and further spread. ICAR-National Bureau of Plant Genetic Resources (ICAR-NBPGR) is authorized to issue import permit and undertake quarantine of plant germplasm including transgenics and for issue of Phytosanitary Certificate for material under export. Adopting a systematic workable strategy, using a combination of conventional and modern techniques, over the past forty-five years, a total of 78 exotic pests including fungi (6), viruses (19), insects/ mites (26), nematodes (9) and weeds (18) of great quarantine significance to India have been intercepted. All efforts are made to salvage the infested/ infected materials; however, if the material is unsalvageable, it was incinerated. The interceptions made signify the potential dangers in import of planting material if proper quarantine measures had not been followed. ICAR-NBPGR, also makes concerted efforts to develop and customize the modern detection and eco-friendly salvaging techniques to minimize the risk of escape in quarantine processing. Scientifically sound and transparent risk analysis prior to import is an important tool, so that, our agricultural production is not jeopardized. The Indian phytosanitary regulations provide a fragmented legislative system which needs to be harmonized and integrated to holistically deal with national biosecurity while complying with international norms. Also, there is a need to support research, training, capacity-building, networking and information sharing activities at both national and regional levels.

Plant Quarantine at ICAR-NBPGR

International exchange of plants/planting material carries an inadvertent risk of introduction of exotic pests or their new virulent races/strains into new areas. History has witnessed several examples of dangerous pests introduced along with plants/planting material/plant products that have led to serious socio-economic consequences. Plant quarantine is a mandatory requirement to regulate the entry of seed/planting material, plant products, living organisms and soil etc. so as to prevent unintended entry of pests across nations. International exchange of plant genetic resources (PGR) is important to broaden the genetic base of crops in order to develop improved crop varieties. The Government of India has legislated the Plant Quarantine (Regulation of Import into India) Order 2003, [hereafter referred to as PQ Order] to regulate the import of plant material. Under the PQ Order, ICAR-National Bureau of Plant Genetic Resources (ICAR-NBPGR) has been delegated powers to issue Import Permit and to carry out quarantine processing of imported PGR including transgenics and for issue of Phytosanitary Certificate for PGR meant for export.

The Division of Plant Quarantine at ICAR-NBPGR, New Delhi has developed an efficient and systematic methodology for quarantine processing for pest diagnostics, salvaging and containment to ensure biosecurity during exchange of PGR and to be transparent during exchange internationally. Stringent quarantine examination of 49,97,795 samples of PGR during 1976–2021 of which 1,78,507 samples were found infested/infected by insects (1,08,615); pathogens (42,123); Nematodes (23,952) and weeds (3,817). A systematic step-wise strategy is being followed for testing of each of the samples imported for presence of any unwanted pests Fig 1.

Over the years, during quarantine processing, a large number of pests have been intercepted in imported germplasm and other research material. The intercepted pests can be categorized as:

- (i) Pests not known to occur in India
- (ii) Pests with different races/biotypes/strains not known to occur in India

*Author for Correspondence: Email-celia.chalam@icar.gov.in; mailcelia@gmail.com

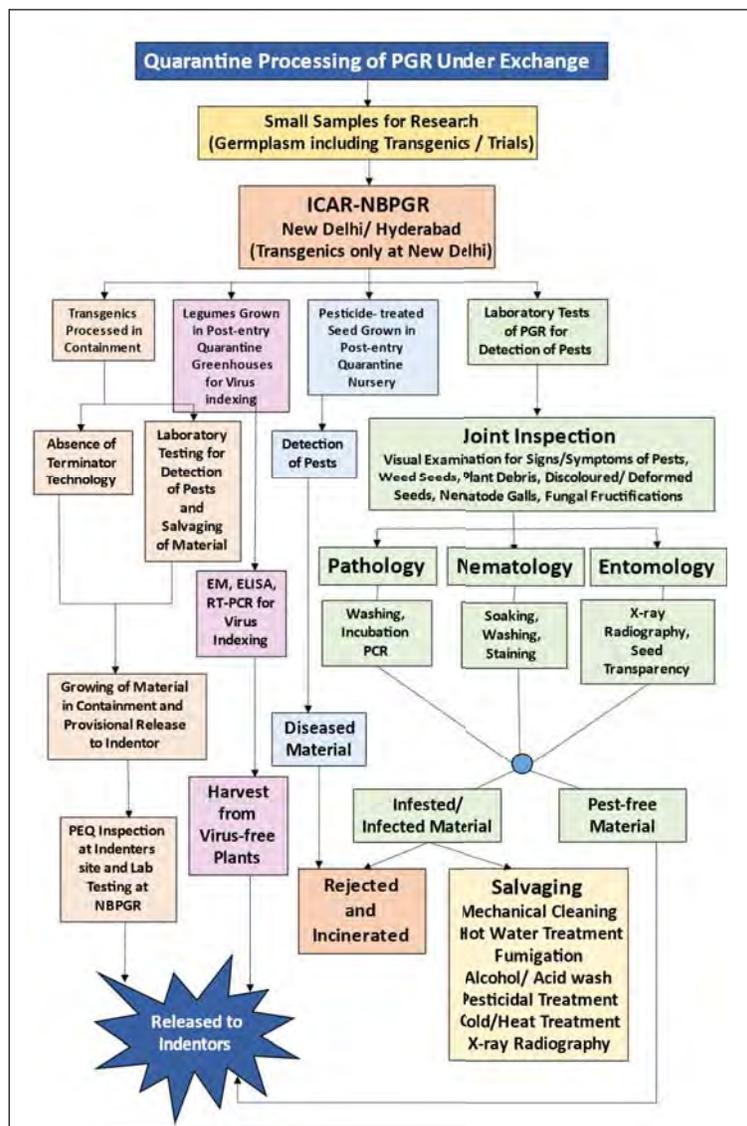


Fig. 1. Schematic presentation of Quarantine Processing

- (iii) Pests intercepted on a new host or are from a country from where they have never been reported before
- (iv) New pest species hitherto unreported in science
- (v) Pests reported to be present in India

These interceptions, especially of pests and their variability not yet reported from India [Category (i) and (ii)] signify the importance of quarantine in preventing the introduction of destructive exotic pests. The categories (iii) and (iv) pests are not expected in the sample as per the PRA which is literature-based and since no records are available on the pest/ host their presence is unexpected and hence, important from quarantine view point. The last category (v) – pests reported to be

present in India are also important as they could have a wide host range and become invasive in case they find suitable biotic and abiotic environment (Khetarpal and Gupta, 2008). A total of 78 exotic pests belonging to Category (i) including insects/ mites (26), fungi (6), viruses (19), nematodes (9) and weeds (18) that are not yet reported from India have been intercepted so far.

The samples found infested/ infected with pests viz., were salvaged using suitable methods and if they could not be salvaged, they were rejected and destroyed by appropriate means (Fig. 2). Had any of these exotic pests not been intercepted and had escaped, they could have entered and established in India and subsequently caused devastation to Indian agriculture.

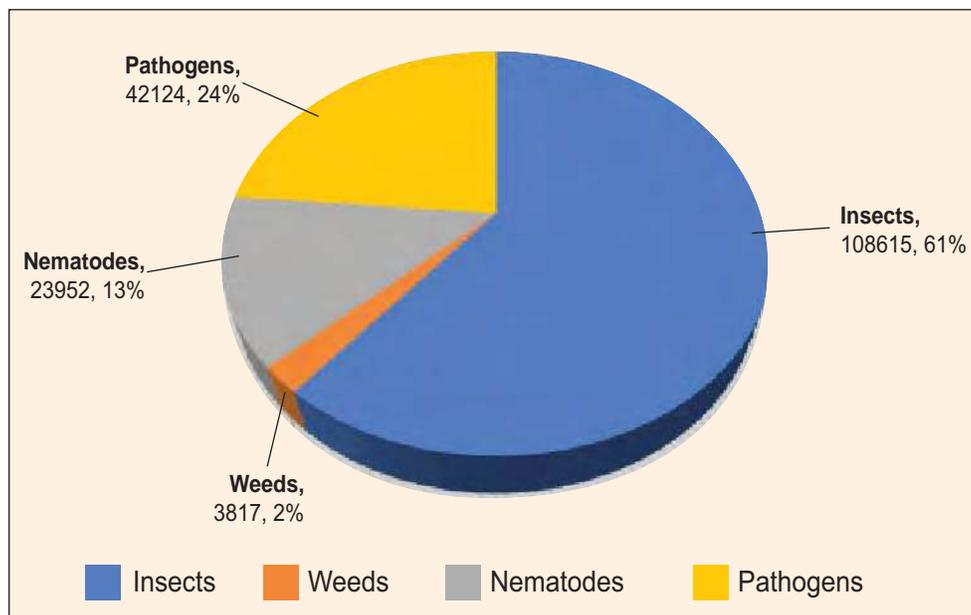


Fig. 2. Samples Infested/Infected (1976-2021)

Presently, exchange of PGR has become more difficult under the Convention on Biological Diversity, hence, all attempts were made to salvage the germplasm and over 99 % of samples were salvaged and less than 1% samples were rejected. ICAR-NBPGR has *state-of-the-art* facilities to properly undertake quarantine. However, more wide-ranging efforts are needed in the national plant quarantine system to develop adapt and adopt the latest detection and eco-friendly disinfestation/disinfection techniques to minimize the risk of pest escape in quarantine.

Issues in Quarantine of Germplasm and Action Points

There are issues related to quarantine in exchange of PGR both legislative and related to quarantine processing and methodology (Khetarpal *et al.*, 2006; Chalam and Khetarpal, 2008). The legislative issues pertaining to India are as follows:

The national quarantine legislation needs to be in harmony with the international norms laid down by International Plant Protection Convention (IPPC) and the countries in the region are trying to gear up their activities to comply with it. India has done so by bringing out the PQ Order 2003 and through its various amendments. Under this the imports have been classified as:

(a) Prohibited plant species (Schedule IV);

(b) Restricted species where import is permitted only by authorized institutions (Schedule V);

(c) Restricted species permitted only with additional declarations of freedom from quarantine/ regulated pests and subject to specified treatment certifications (Schedule VI) and;

(d) Plant material imported for consumption/ industrial processing permitted with normal Phytosanitary Certificate (Schedule VII).

Under the PQ order, a pest risk analysis (PRA) has been made mandatory for all material being imported into the country other than those present in Schedules V, VI and VII. The various schedules V, VI and VII of the PQ Order give lists of crops for which a generic PRA is given and detailed PRA is not required. In case of germplasm, a large number of species of cultivated crops (and their wild relatives/land races) with useful traits are imported. Such wild relatives, land races of germplasm whose pest profile is not adequately recorded hampers PRA preparation and consequently their import. However, in 2007 the legislation has been amended whereby ICAR-NBPGR is empowered to undertake PRA for germplasm material for pest-free import. This is more relevant in the present context when access to germplasm is becoming more and more difficult under the Convention on Biological Diversity, 1992.

Another difficulty, which is faced during import of certain material, is the additional declarations being sought under the Schedule VI (Species permitted only with additional declarations of freedom from quarantine/regulating pests and subjected to specified treatment certifications). Many countries from where a pest is not reported find it difficult to certify in the phytosanitary certificate the freedom from those pests. To solve this problem, the recent amendments of the PQ Order with more number of crops give country-specific requirements under additional declarations and special conditions for import. This would greatly help the indentors in procuring germplasm of their interest from varied sources (Dubey *et al.*, 2021).

The technical issues include issues pertaining to quarantine processing i.e. methodology for detection and salvaging of pests are enumerated follows:

- The amount/size of the germplasm sample is very crucial from quarantine processing point of view, as sampling procedures meant for bulk material cannot be adopted. Also, the technique applied should enable detection of miniscule amounts of pest in the samples drawn and also be non-destructive. The sample size may not be enough for direct testing of seed and post-entry quarantine testing and release from virus-free plants after testing may take one crop season.
- Diagnostic reagents such as antisera for viruses/bacteria and the reference collection for insect pests are often not available for exotic pests. A repository of antisera needs to be established as one has to deal with exotic pests in quarantine (Chalam, 2020). Expertise is also required in the field of taxonomy and biosystematics as there is a continuous need to identify unknown/ new pathogens or strains.
- There is an urgent need to have National Plant Pests Diagnostic Network (NPPDN) linking the research laboratories with seed/vegetative planting material testing laboratories and quarantine stations, which would be the backbone for strengthening the programme on biosecurity from plant pests (Chalam and Maurya, 2018).
- Strengthening research on the development of sensitive and non-destructive detection and salvaging techniques as well as on:
 - Increased use of radiation and other eco-friendly non-chemical frontier techniques as effective mitigation treatment as alternatives to methyl bromide fumigation
 - Development of user-friendly serological/molecular diagnostic protocols/kits for prognostic detection of exotic pests and their variants and also low levels of pest infections.
 - Development of digitized biosystematic keys for pest identification.
 - Epidemiological studies including survey and surveillance of diseases/pests to prepare database on endemic pests, identify pest free areas and target IPM for reducing threats.
 - Developing models for risk analysis for exotic pests, diseases, invasive weeds and genes.
 - Developing standard operating procedures through relevant Handbooks/Manuals for survey and surveillance and checking major diseases/pests including invasive weedy species.
 - Studies on factors influencing climate change and pest diversity and virulence.
 - Studies on factors affecting potential of pest survival under different transport conditions of, modes of dispersal, availability of hosts/alternate hosts at port of entry, establishment potential, strategy for reproduction and mode of pest survival, vectors and natural enemies of the concerned pest in the new area.
 - Simulated evaluation of mitigation options to deal with epidemics/pandemics.
 - Need for special focus on management of indigenous diseases, pests and invasive weeds with a potential to impact food security, environment including biodiversity, and trade.
 - Development of national dynamic biosecurity database of insect pest, diseases, invasive weeds and their management.

National Plant Quarantine System: Way Forward

The liberalized trade under WTO has made quarantine personnel highly accountable on accurately achieving the objectives of eliminating alien pests or to take up eradication actions. The threat of pest introduction and the ways to stop the establishment of such pests into India continues to be a major fear. Unless proper phytosanitary procedures are adopted, pests could be transferred all over the world, get established into new

areas and impact agricultural production (Khetarpal and Gupta, 2008). On the other hand, this also presents a tremendous opportunity for farmers to export agricultural products/produce to boost our economy if they meet the quality standards and overcome phytosanitary restrictions at international level. Active participation in meetings of IPPC and Codex Alimentarius by India to negotiate for realistic standards would also give a necessary push to trade in agriculture. All this is feasible only if the government and the organizations working on SPS issues work hand-in-hand with each other, to accomplish international standard of agricultural produce that would eventually boost Indian exports at the same time safeguarding our crops and environment from ingress of exotic pests (Khetarpal and Gupta, 2007).

Also, awareness about the biosecurity among general public and scientific community is greatly required for generating respect for quarantine/ biosecurity regulations. Besides, the domestic quarantine regulations already promulgated for nine destructive pests (fluted scale, San José scale, coffee berry borer, codling moth, banana bunchy top and mosaic viruses, potato cyst nematode, potato wart and apple scab) seem to be a complete failure as there are reports of spread of some of these pests to neighbouring states and also to different parts of the country. Extensive eradication measures and surveys are needed in different regions of the country that would finally contribute in identifying the pest free areas, a task that is also required to be accomplished as per the international norms for fulfilling the commitments made under the SPS Agreement. There is also an urgent requirement to prevent not only spread of the pests above but also to develop domestic quarantine regulations against certain recently introduced/ detected pests in the country and which might spread fast (Gupta and Dubey, 2017). More resources need to be diverted towards developing appropriate legal and protection measures against such pests to save the country.

Unlike in USA and Australia, there is a total absence of organized plant quarantine services at state borders in India. It is essential to set-up plant quarantine checkpoints at inter-state border roads/inland container depots/ rail roads manned by trained personnel to control the movement of planting material, transport carriers and containers so as to have effective check of the pests especially those covered under domestic quarantine regulations.

Indian plant quarantine system has certainly come a long way starting with the legislation viz., Destructive Insects and Pests Act way back in 1914. However, modernizing quarantine mechanism is much more crucial today in the light of globalization not only to protect the crops from invasive foreign pest species but to safeguard the economy by ensuring enhanced productivity and farm incomes. The Agricultural Biosecurity Bill (2013) which provided for instituting of a mechanism for holistic prevention, eradication and management of pests of plants and, pests and diseases of animals and undesirable organisms for ensuring biosecurity in agriculture and to comply with our international commitments, for enabling exports and imports of plants and their products, animals and their products, aquatic beings and regulation of microorganisms of agricultural importance and for matters connected therewith or incidental thereto needs to be revived and put in place.

References

- Bhalla S, VC Chalam, B Singh, K Gupta and SC Dubey (2018) *Biosecuring Plant Genetic Resources in India: Role of Plant Quarantine*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, 216 p +vi.
- Chalam VC (2020) Elimination of plant viruses by certification and quarantine for ensuring biosecurity. In: *Applied Plant Virology Advances, Detection and Antiviral Strategies*. Awasthi LP (eds) Academic Press., USA, pp 749-762.
- Chalam VC and AK Maurya (2018) Role of quarantine in ensuring biosecurity against transboundary plant viruses. *Agric. Res. J.* **55**: 612-626.
- Chalam VC and RK Khetarpal (2008) A critical appraisal of challenges in exclusion of plant viruses during transboundary movement of seeds. *Indian J. Virol* **19**: 139-149.
- Dubey SC, K Gupta, J Akhtar, VC Chalam, MC Singh, Z Khan, SP Singh, P Kumar, BH Gawade, Raj Kiran, T Boopathi and P Kumari (2021) Plant quarantine for biosecurity during transboundary movement of plant genetic resources. *Indian Phytopathol.* **74**: 495–508. <https://doi.org/10.1007/s42360-021-00375-7>
- Gupta K and SC Dubey (2017) Biosecurity Policies Influencing International Exchange of PGR. *Indian J Plant. Genet. Res.* **30(3)**: 258-266
- Khetarpal RK and K Gupta (2008) Plant quarantine in India in the wake of international agreements: A review. Scientific Publishers (India), Jodhpur, *Rev. Plant Pathol.* **4**: 367-391.
- Khetarpal RK (2004) A critical appraisal of seed health certification and transboundary movement of seeds under WTO regime. *Indian Phytopathol.* **57**: 408-421.
- Khetarpal RK and K Gupta (2007) Plant Biosecurity in India- Status and Strategy. *Asian Biotech. Development Rev.* **9**: 39-63.

Khetarpal RK, A Lal, KS Varaprasad, PC Agarwal, S Bhalla, VC Chalam and K Gupta (2006) Quarantine for safe exchange of plant genetic resources. pp 83-108 In: *Hundred Years of Plant Genetic Resources Management in India* (eds. AK Singh, K Srinivasan, S Saxena and BS Dhillon). National Bureau of Plant Genetic Resources, New Delhi, India.

Plant Quarantine (Regulation of Import into India) Order (2003) The Gazette of India Extraordinary, Part II Section 3 Sub-section (ii) No. 1037 Published by Authority. p 314 available at <https://plantquarantineindia.nic.in/pqispub/pdffiles/pqorder2015.pdf>

Large-scale Characterization for Management and Utilization of Indian National Genebank Collections

Ashok Kumar*, Kuldeep Tripathi and Jyoti Kumari

ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

Globally, plant genetic resources (PGR) are the key biological resources having great potential to meet the challenges of food and nutritional security. ICAR-National Bureau of Plant Genetic Resources (NBPGR) has been working with a network of 10 regional stations located in different agro-climatic zones of India for the management of genetic resources. The linkages established with CGIAR centres, national crop-based institutes and state agricultural universities are the backbones of PGR utilization. NBPGR characterizes and evaluates germplasm conserved in the Indian national genebank for different traits and provides material to breeders. The effective utilization of PGR in crop improvement programmes depends mainly on their systematic characterization and evaluation, and identification of potentially useful germplasm. To date, we have characterized more than 2.35 lakh accessions of different agri-horticultural crops and developed core collections for the effective management of a large number of accessions. Presently, our focus is on the large-scale characterization of entire germplasm conserved in the Indian National Genebank and detailed evaluation of prioritized crops; identification of reference sets and core sets for enhanced utilization.

Key Words: Crop wild relatives, Characterization, Evaluation, Genebanks, Germplasm

Introduction

Plant genetic resources (PGR) are the genetic material of plants having immense value as a resource for mankind of present and future generations. The ICAR-NBPGR acts as a nodal institute at a national level for the management of PGR under the national agricultural research system. The Bureau's activities include PGR exploration, collection, exchange, characterization, evaluation, conservation and documentation. Genebanks houses pools of germplasm variability containing vital genes that play a crucial role in breeding programs for sustainable crop production. Efficient and effective use of germplasm in crop improvement depends upon a thorough understanding of the existing genetic variability, and knowledge of the traits and genes present in individual accession, which depends on the comprehensive characterization and evaluation. Without the information on the traits of the accessions conserved in genebanks, they become museums of plant accessions or living herbaria. To identify novel accessions corresponding to the genes with desired traits, it is prudent to characterize the germplasm collections conserved in the genebanks. Thus, the need for characterization and evaluation for sustainable use of agro-biodiversity has been felt universally. However, the characterization of large collections by plant breeders is time- and resource-consuming, therefore genebank

curators have developed a concept of a core set (~10% of the entire collection) from entire collections that are characterized at once to identify the targeted germplasm and use it efficiently (Frankel and Brown, 1984). The core set refers to a minimum set of germplasm that captures the entire range of genetic variability of any crop, with minimum repetitiveness. Being smaller in size and diverse in nature, the core set can be efficiently used as a kickoff point to enhance genetic gains, including the use of phenomics and genomics tools in less time. Further, depending on the number of collections it can be minimized to a mini core set that is ~1% of the entire collection. Also, trait-specific reference set can be identified for introgression, gene discovery and genomics studies.

Therefore, it is essential to use genetic diversity conserved in genebanks and identify novel germplasm, core sets and reference sets for their use in trait-specific breeding programs to enhance cultivar productivity and resilience to climate change.

Status and Strategies of Characterization, Evaluation and Utilization

To date, more than 2.35 lakhs accessions of different crops at ICAR-NBPGR have been characterized and evaluated (Singh *et al.*, 2020). The year-wise information

*Author for Correspondence: Email-ashok.kumar28@icar.gov.in

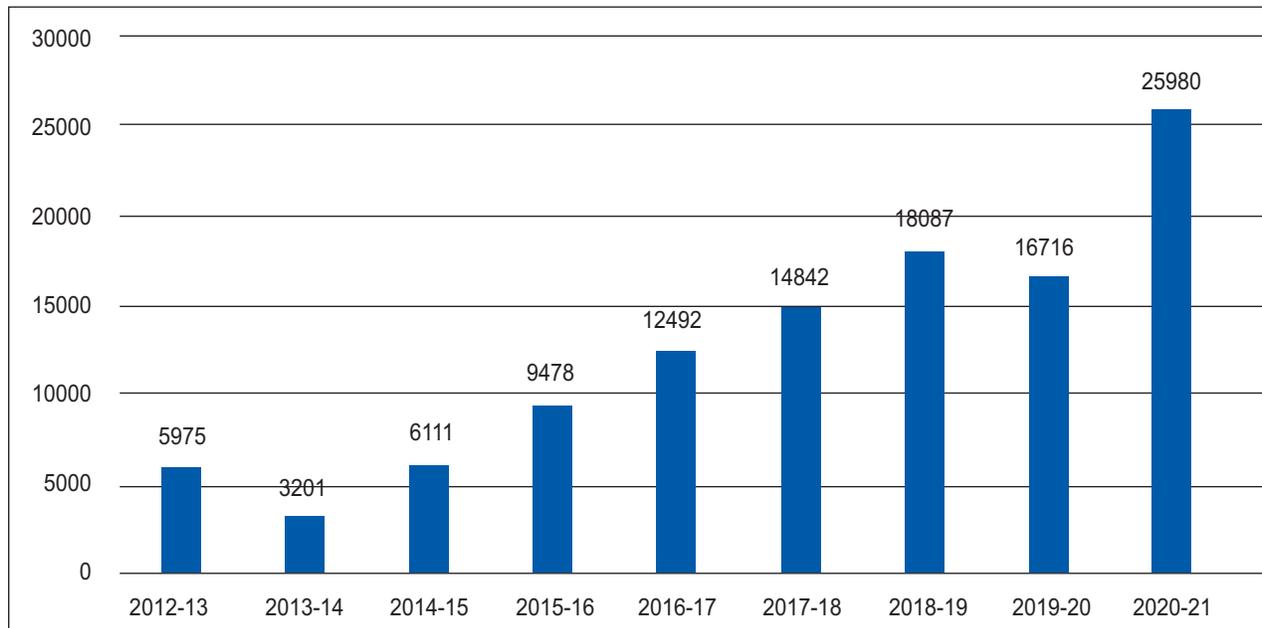


Fig. 1. Characterization of agri-horticultural crops (2012-2022)

on characterized accessions at ICAR-NBPGR have been depicted in Fig. 1.

To characterize the entire accessions conserved in the Indian national genebank (INGB), the mega programme on characterization and evaluation of germplasm was initiated in 2011-12 in which 22,416 accessions of wheat were characterized. Similarly, 18,775 accessions of chickpea were also characterized in 2012-13. These mega-characterization programmes paved the way for other prioritized crops conserved in INGB. Under a focused strategy for enhancing germplasm utilization, ICAR-NBPGR has shifted from piecemeal work to a complete characterization approach in recent years where entire accessions of prioritized crops were characterized in one go using a comprehensive strategic plan (Fig. 2). The large-scale characterization is being carried out under institute projects, Consortium Research Platform on agrobiodiversity and other network projects. The network project on minor pulses, minor oilseeds, rice, wheat and chickpea have been initiated under the mission mode programme supported by the Department of Biotechnology, Government of India. To facilitate germplasm management and utilization, core sets have been developed in different crops at ICAR-NBPGR (Table 1). Moreover, core sets have been developed for cowpea, mungbean, urdbean and horsegram are under evaluation and validation stage. Besides this, trait-specific germplasm (TSG) has been also identified from

large-scale characterization and evaluation programs which are becoming the material of choice for crop breeders nationally. Some trait-specific germplasm has been identified and validated which are mentioned in Table 2.

Future Perspectives

Global interdependence for PGR requirements is a matter of utmost importance to satisfy the needs of countries. There is a need to collaborate at local, national and global levels for the evaluation and utilization of the germplasm. Genebank researchers should work on the strategy of linking every accession conserved in genebanks with traits. Completely characterized genebank collections will facilitate their management and utilization. New tools of phenotyping including drones and digital data loggers need to be deployed to accelerate the phenotyping of the large number of accessions. Further, the characterization and evaluation activity requires substantial inputs and a decentralized evaluation network with the collaboration of NARS partners. Also, there is a need to modify the descriptors for evaluation accordingly and make the search for the desired characteristics in the database as quick and efficient as possible. The core collection concept is a more structured and efficient approach to identifying limited sets of diverse germplasm and utilising the same more effectively. The use of crop wild relatives (CWR) is needed to incorporate novel traits related to

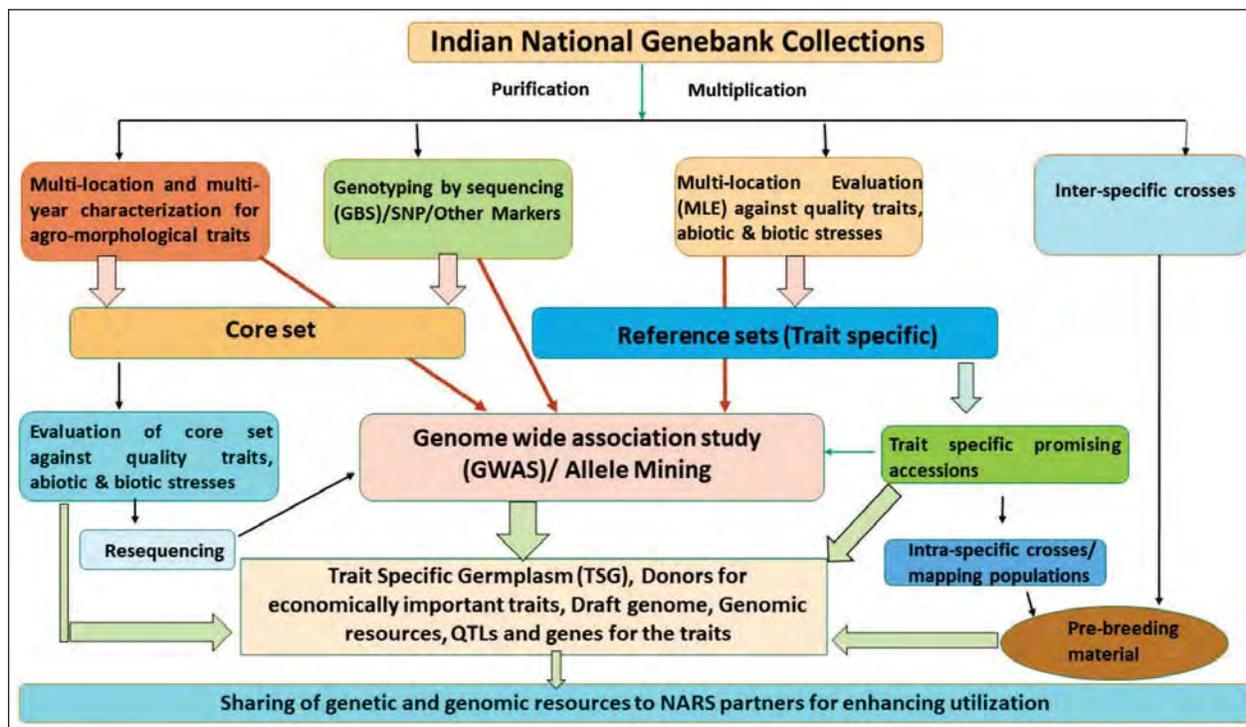


Fig. 2. Flowchart depicting strategies adopted by ICAR-NBPGR for germplasm utilization

Table 1. Large-scale characterization and core sets developed at ICAR-NBPGR during the last decade

Crop	No. of Acc. Characterized/ Descriptors	No. of Acc. in core collection	Reference
Wheat	22,464/34	2,226	Phogat <i>et al.</i> , 2020
Chickpea	14,651/21	1,103	Archak <i>et al.</i> , 2016
Barley	6,778/19	688	Kaur <i>et al.</i> , 2022
Lentil	2,324/26	170	Tripathi <i>et al.</i> , 2022
Wild lentil	405/18	96	Singh <i>et al.</i> , 2014
Cowpea	3,720	425	Under validation
Mungbean	4,100	400	Under validation
Urdbean	2,266	218	Under validation
Horsegram	2,186	300	Under validation

abiotic and biotic stresses, and nutritional quality to bring enhanced levels of productivity and stability of performance, and to provide food and nutritional security.

References

Archak S, RK Tyagi, PN Harer, LB Mahase, N Singh and OP Dahiya (2016) Characterization of chickpea germplasm conserved in the Indian National Genebank and development of a core set using qualitative and quantitative trait data. *Crop J.* 5: 417-424.

Table 2. Glimpses of trait-specific germplasm identified by ICAR-NBPGR

Lentil, IC317520 (INGR 19072) Trait: Extended funiculus (Tripathi <i>et al.</i> , 2019)	
Lentil, IC241473 (INGR21223) Trait: Multi-podding germplasm (Tripathi <i>et al.</i> , 2022)	
Pearl millet, IC283734 (INGR 13056) Trait: High popping yield (Kumari <i>et al.</i> , 2016)	
Wheat, IC529962 (INGR19044) Trait: Highly resistant to spot blotch (Kumari <i>et al.</i> , 2018)	
Chickpea, EC267301(INGR 21137) Trait: Resistant to Ascochyta blight (Gayacharan <i>et al.</i> , 2020)	

- Frankel OH and AHD Brown (1984) Current Plant genetic resources—a critical appraisal. In: JHW Holden and JT Williams (eds) *Crop Genetic Resources: Conservation and Evaluation*, George Allen & Unwin, London, pp. 249–257.
- Gayacharan, U Rani, S Singh, AK Basandrai, VK Rathee, K Tripathi, N Singh, GP Dixit, JC Rana, S Pandey, A Kumar and K Singh (2020) Identification of novel resistant sources for ascochyta blight (*Ascochyta rabiei*) in chickpea. *PLoS One*. doi: 10.1371/journal.pone.0240589.
- Kaur V, J Aravind, Manju, SR Jacob, J Kumari, BS Panwar, N Pal, JC Rana, A Pandey and A Kumar (2022) Phenotypic characterization, genetic diversity assessment in 6,778 accessions of barley (*Hordeum vulgare* L. ssp. *vulgare*) germplasm conserved in national genebank of India and development of a core set. *Front. Plant Sci.* **13**: 771920.
- Kumari J, MK Bag, S Pandey, SK Jha, SS Chauhan, GK Jha, NK Gautam and M Dutta (2016) Assessment of phenotypic diversity in pearl millet (*Pennisetum glaucum* (L.) R. Br.] germplasm of Indian origin and identification of trait-specific germplasm. *Crop Pasture Sci.* **67**(12): 1223-1234.
- Kumari J, S Kumar, N Singh, SS Vaish, S Das, A Gupta and JC Rana (2018) Identification of New Donors for Spot Blotch Resistance in Cultivated Wheat Germplasm. *Cereal Res. Commun.* **46**: 467-479.
- Phogat BS, S Kumar, J Kumari, N Kumar, AC Pandey and TP Singh et al. (2020) Characterization of wheat germplasm conserved in the Indian National Genebank and establishment of a composite core collection. *Crop Sci.* **61**: 604–620.
- Singh K, K Gupta, V Tyagi and S Rajkumar (2020) Plant genetic resources in India: management and utilization. *Vavilovskii Zhurnal Genet Selektii.* **24**(3): 306-314.
- Singh M, IS Bisht, S Kumar, M Dutta, KC Bansal and M Karale (2014) Global wild annual lens collection: a potential resource for lentil genetic base broadening and yield enhancement. *PLoS ONE* **9**: e107781.
- Tripathi K, PG Gore, A Pandey, R Bhardwaj, N Singh, G Chawla and A Kumar (2019) Seed morphology, quality traits and imbibition behaviour study of atypical lentil (*Lens culinaris* Medik.) from Rajasthan, India. *Genet. Resour. Crop Evol.* **66**: 697–706.
- Tripathi K, J Kumari, PG Gore, DC Mishra, AK Singh and GP Mishra et al. (2022) Agro-morphological characterization of lentil germplasm of Indian national genebank and development of a core set for efficient utilization in lentil improvement programs. *Front. Plant Sci.* **12**: 751429

Plant Genetic Resources for Crop Improvement: The North-Western Himalayan Perspective

SK Sharma^{1*}, Nikhil Malhotra² and Mohar Singh²

¹CSIR-Institute of Himalayan Bioresource Technology, Palampur-176061, Himachal Pradesh, India

²ICAR-National Bureau of Plant Genetic Resources Regional Station, Shimla-171004, Himachal Pradesh, India

Himalayas is one of the unique eco-systems, rich in robust indigenous culture and traditional wisdom. Due to its varied climatic conditions, it is gifted with a wide diversity of plant genetic resources (PGRs) of several crop species and their wild relatives. These crops and their wild taxa have contributed significantly to environment and food security of the native people. Recent past has seen some breakthroughs in successful utilization of Himalayan PGRs in cultivar development that could serve as model for future crop improvement programs vis-à-vis climate change and nutritional security.

Introduction

Himalayas is one of the 36 hot spots of diversity identified globally. Plant Genetic Resources (PGRs) in the Himalayas comprise land races and primitive cultivars, obsolete farmers' and old varieties, recently released cultivars, parental line of the released hybrids, genetic stocks, wild and their weedy relatives etc. The farmers of the region grew these land races for their livelihood security since ages. Notwithstanding yield potential, these are invariably characterized by drought tolerance, pest resistance, wider adaptability with desirable quality traits etc. These land races were improved by both selection and hybridization, and a large number of high yielding varieties were developed for the benefit of farmers. In the traditional agro-ecosystem of North-Western Himalayas, these varieties have replaced the traditional cultivars and land races leading to the loss of diversity. There has to be a national action plan for further strengthening their collection, evaluation, conservation and sustainable utilization.

Status of PGRs

Himalayas, including NW Himalayan region is endowed with an array of genetic resources of crop plants, their wild relatives and a plethora of edible wild plants supporting ~85% of the region's population on agriculture for their food and daily needs. The region is very rich in species diversity of cereals, millets, pseudo cereals, pulses, oilseeds, vegetables, fruits, spices and condiments, fibers etc. It represents the richness of PGRs by having around

272 cultivated crop species and 898 wild relatives and related types (Table 1; Sharma and Rana, 2005).

Table 1. Status of PGRs and their wild relatives in NW Himalayas

Crop group	Cultivated species	Wild species	Total
Cereals	5	36	41
Millets	10	44	54
Pseudo cereals	7	20	27
Pulses	19	34	53
Oilseeds	13	11	24
Vegetables	68	58	126
Fruits and nuts	49	65	114
Fibers	9	44	53
Spices and condiments	37	55	92
Forages	8	219	227
Ornamentals	18	130	148
Medicinal and aromatic plants	29	182	211
Grand total	272	898	1170

PGR Utilization

Despite the enormous variability of crop species including wild relatives occurring in the Himalayas, their exploitation in crop improvement programs is still naive. Nevertheless, there are some notable examples demonstrating successful utilization of PGRs for the development of high yielding varieties through either selection or hybridization in NW Himalayan States/UTs. Among cereals, the use of genes from an accession of *Oryza nivara* from NW Himalayas to produce long-lasting resistance to grassy stunt virus is one of the

*Author for Correspondence: Email-skspbg@yahoo.co.in

pioneer examples of using wild germplasm in rice resistance breeding (Khush, 1997). The annual species of wild rice (*Oryza rufipogon*), which grows in the several parts of the NW Himalayas have a high degree of blast resistance (Rathour et al., 2005). Selection from available germplasm has also led to the development of several rice varieties in the region. SKUAST, Jammu, developed Ranbir Basmati and Saanwal Basmati through selection from Basmati 370 and released these for cultivation during 2005 and 2007, respectively. Later, high yielding Basmati 564 was also developed from Basmati 370 through selection during 2014. Recently, it developed Jammu Basmati 118, 123 and 138 from Basmati 370 having high yield potential. Notably, State Variety Release Committee (SVRC) released these varieties for cultivation during 2020. Similarly, pure line selection led to the genetic improvement and uniformity of three most popular rice varieties viz. Mushk Budji, Kamad and Red Rice of Kashmir valley. Of late, red rice grown in the region has also gained considerable attention nationwide due to their unique red pericarp colour and high nutritional values. In Himachal Pradesh, Bhrigu Dhan, the first high yielding red rice variety developed from a cross Chucheng/Deval[®]//Matali was released by the SVRC during 2005. Further, ICAR-VPKAS, Almora developed maize variety VL Baby Corn 1 during 2005 by using a local land race Murali Makkai, a Sikkim primitive maize exhibiting prolificacy and excellent popping capacity.

In pulses, CSK HPKV, Palampur developed three popular varieties of rajmash namely, Baspa (KRC-8) derived from collection from Kinnaur and released as variety for cultivation in Himachal Pradesh. This is very popular variety for its attractive seed colour and field resistance to bean anthracnose and is still under cultivation in many segments of the state. Triloki variety was derived from a land race of Lahaul valley, and Kailash (SRC-74) was selected from the local germplasm collected from Sangla valley and released for cultivation in the dry temperate region of Himachal Pradesh during 2003. The University also developed horsegram Baiju (HPK-4) variety from local germplasm collection. ICAR-VPKAS, Almora developed wilt and rust resistant lentil VL Masoor 514 by crossing VL Masoor 501 × VL Masoor 103 during 2011. VL Masoor 103 is a selection from local collection of Uttarakhand Himalayas. SKUAST, Jammu developed Bharderwah

Rajmash variety (BR-104) through selection from local germplasm and released by the SVRC during 2020.

In potential crops, the notable instances of the effective use of amaranth germplasm from the Himalayas is the development of the high yielding cultivar Annapurna during 1984 which was a direct selection from a local genetic line followed by Durga during 2006 developed from IC35407 by ICAR-NBPGR Regional Station, Shimla. Both these varieties were released by the CVRC. Similarly, ICAR-NBPGR Regional Station, Shimla also developed buckwheat variety Himpriya as a pure line selection from IC13374 followed by Shimla B1 from IC341671 and notified by CVRC during 1991. Further, ICAR-VPKAS, Almora developed VL UGAL 7 variety during 1991 through mass selection. Recently, ICAR-NBPGR Regional Station, Shimla developed Himphara variety of buckwheat valued for high protein content (13.10 %) from IC341589 and was dedicated to the nation by Hon'ble Prime Minister of India on 28th September 2021.

CSK HPKV, Palampur developed a number of varieties in forages for improving temperate grasslands and pastures by involving the local land races and indigenous germplasm collected from different parts of the Himalayas. Napier-Bajra hybrid NB-37, Red clover PRC-3, Setaria grass S-92, Tall fescue grass Hima-1 and Hima-4, and White clover Palampur Composite-1 are some of the major varieties developed during 1987-2005 and released by the SVRC.

Wild species of fruits are growing abundantly in Himalayas and have been used as rootstock for cultivated temperate/sub-temperate fruit crops. These include different *Prunus* species as rootstock for cherry and plum, kainth for pear, crab apple as seedling rootstock for apple, wild apricot or chulli for apricot, hard-shelled almond and walnut for almond and walnut, respectively. SKUAST, Jammu recently developed high yielding walnut and pecan nut varieties Bhusan and SJPP-25, respectively through selection of local germplasm collected from Kishtwar region in Jammu and released by SVRC for cultivation during 2020. Likewise, popular variety of drying apricot namely Halmen cultivated in Kargil, Leh and Spiti was selected from a local land race. Likewise, Racharpo a table purpose apricot variety and the sweetest in the world was selected from a local land race in Kargil.

Future Prospects and Action Points

- Mainstreaming the use of ancient crop varieties and revisiting the diverse regions of the Himalayas harboring them could help in the conservation and long-term sustainable utilization of PGRs.
- Awareness and capacity building programs are required to enable the local people to use highly diversified crop plants inhabiting the region to increase agricultural productivity as per future needs.
- Promoting the registration of traditional land races with PPV&FRA in response to IPR issues will protect the interests of native farming communities.
- An integrated approach of conventional and advanced technologies such as high throughput genotyping

and phenotyping, development of genomic resources and strengthening PGR informatics should be the contemporary trail for the effective utilization of Himalayan PGRs to develop improved cultivars with climate resilient traits along with superior agronomical and nutritional features.

References

- Khush GS (1997) Origin, dispersal, cultivation and variation of rice. *Plant Mol. Biol.* **35**: 25-34.
- Rathour R, V Gaur, RP Kaushik and RS Chauhan (2005) *Oryza rufipogon* – a possible source of novel resistance specificities against rice blast (*Magnaporthe grisea*). *Curr Sci.* **89(3)**: 443-447.
- Sharma BD and Rana JC (2005) *Plant Genetic Resources of Western Himalaya – Status and Prospects*. Bishen Singh Mahendra Pal Singh, Dehradun, India, 467 p.

Management of Horticultural Genetic Resources in India: Recent Advances

Umesh Srivastava

Consultant, Trust for Advancement of Agricultural Sciences, Pusa Campus, New Delhi-110012, India
Former Assistant Director General (Horticulture), ICAR, New Delhi

Horticultural crops comprise diverse economic species ranging from the fruits/nuts, vegetables, spices and condiments, ornamental plants, aromatic and medicinal plants. Besides the tangible materials, horticultural genetic resources (HGR) also encompass the indigenous knowledge accumulated over ages among gardeners, and which surround the use of such biological resources. Global climate change is causing challenges in productivity with the rise in global temperature and reduced amount of precipitation. It is crucial to develop strategies for effective conservation and judicious use of HGR to improve nutritional security and food safety for human health. This paper describes the present status of HGR in India, its scientific management aspects, including their diversity, conservation and sustainable use. It also addresses crucial concerns regarding conservation in India *vis-à-vis* recent advances in management of the HGR.

Key Words: Climate change/environmental stress, Conservation, HGR management, Horticultural crop diversity, Recent advances

Horticultural crops include fruits, vegetables, ornamentals and medicinal crops/species, and these species vary from place to place. In India, horticultural crops include 145 species of root and tuber (e.g., potato, onion, yam, taro), 521 of vegetables/greens (e.g., beans, peas, carrot, brinjal, cauliflower, cabbage, carrot and tomato, amaranth, *palak*), 101 of buds and flowers (e.g., apple, pear, grapes, cherry, peach and apricot), 118 of seeds and nuts (e.g., cashew, almond), medicinal plants (mint, liquorice, foxglove, cinchona, *Hyoscyamus* and others such as hops-*Humulus lupulus*). Thus, both indigenous (e.g., lemon, cucumber, lime, mango, muskmelon, eggplant) and well-adapted exotic species constitute a well-balanced matrix of crop diversity in India. The important fruit crops grown commercially in India are mango, banana, citrus, guava, grape, pineapple, papaya, sapota, litchi and apple which comprise more than 75% of total area under fruit cultivation. There are quite a large number of indigenous and underutilized fruit crops, which are being used by the local inhabitants. Some of the important vegetable crops grown in India are brinjal, tomato, chilli, sweet pepper, cabbage, cauliflower, knol-knol, okra, onion, garlic, long melon, muskmelon, snap melon, watermelon, cucumber, pumpkin, summer squash, bitter gourd, bottle gourd, ridge gourd, round gourd, snake gourd, sponge gourd, ash gourd, carrot, radish, turnip, broad bean, cluster bean, cowpea, lablab bean, french

bean, peas, amaranths, beet root, fenugreek, spinach, lettuce, drumstick and curry leaf. In addition, there are several underutilized vegetables such as *Karembua*-water spinach (*Ipomoea aquatica*), asparagus (*Asparagus officinalis*), *chekurmanis* (*Sauropus androgynus*), *kachnar* (*Bauhinia purpurea*), *poi* (*Basella alba*), elephant foot yam (*Amorphophallus campanulatus*), pointed gourd (*Trichosanthes dioica*), snapmelon, sweet gourd; and underutilized fruits which have market value but not widely grown in the field and rarely found in the market include: jackfruit (*Artocarpus heterophyllus*), *bael* (*Aegle marmelos*), *jamun* (*Syzygium cuminii*), *carambola* (*Averrhoa carambola*), *aonla* (*Emblica officinalis*), *karonda* (*Carissa carandas*) and *phalsa* (*Grewia subinequalis*, *G. asiatica*) (Gupta and Yadav 2016; Rathore *et al.*, 2005). The need for diversification to horticulture sector was acknowledged by the Government of India in mid-eighties by focussing its attention on investment in this sector. Presently horticulture has established its credibility in improving income through increased productivity, generating employment and in enhancing exports. Resultantly, horticulture has moved from rural confines to commercial venture.

Horticultural Diversity Across the Country

Horticulture is potential to raise the farm income, livelihood security and earn foreign exchange. There is

*Author for Correspondence: Email-srivastavaumesh@gmail.com

a need to encourage diversification to high value crops (HVC) at the rate of 5% every year. According to the estimate, the share of the agriculture and allied sector in total GVA has improved to 20.2% in 2020-21 and 18.8% in 2021-22. Horticulture accounts for 30% of India's agricultural GDP from 8.5% of cropped area, and 52% of export earnings in agriculture. India is 2nd largest producer of fruits and vegetables in the world, and world's highest producer of onion, green peas, and cauliflower. India possesses 1000 wild edible plant species including 145 species of roots and tubers, 521 species of leafy vegetables/greens, 101 species of bulbs and flowers, 647 species of fruits, 118 species of seeds and nuts, 9,500 plant species of ethno-botanical uses, 7,500 for ethno-medical purposes, and 3,900 for edible uses of native tribals. A total of 583 species are cultivated, of which 417 belong to hort. crops. Cultivated species include 27 in fruits & nuts, 23 in vegetables, 15 in plantation and tuber crops and 16 in spices and condiments, and Wild relatives included 331 in fruits and nuts, 215 in vegetables, 154 in plantation and tuber crops and 161 in spices and condiments. Thus, the amount of species diversity available offers substantial genetic diversity to meet the future needs, particularly for the present scenario of climate change (Rathore *et al.*, 2005). Details of these diversity scattered in nineteen (19) regions of India, are stated below:

1. **Western Himalayan Region:** Region is inhabited by admixture of Indo-Aryan/Mongolian races. It is secondary centre of origin/ diversity of *Sorbus*, *Rubus*, *Prunus* Apple, pear, peach, plum, almond, apricot, cherry, walnut, and chilli, potato, pumpkin, and primary centres of *Allium* spp., brinjal, cucumber.
2. **Eastern Himalayan Region:** Most of Arunachal Pradesh and Sikkim, and northern tip of West Bengal (Darjeeling and Kalimpong); Tibetan culture, original inhabitants of Sikkim-Lepchas, Adi, Galo, Nishi, Khamti, Monpa, Apatani, and Hill Miris. It is centre of diversity of *Rhododendron*, *Primula*, *Pedicularis*, orchids, spices, *Prunus rufa*, Sikkim-special chilli locally '*Dalle Khorsani*'; *Catharanthus roseus*-anti-cancer/ diabetes; *Centella asiatica*-stomach disorders by different tribes. Wild relatives included *Luffa graveolens*, *Actinidia callosa*, *A. strigosa* (kiwi fruit), *Citrus reticulata* (wild forms), *Mangifera indica* var. *sylvatica*.
3. **Brahmaputra Valley Region:** Most parts of Assam, Meghalaya Plateau, NE Hills, Brahmaputra Valley.

Bhutia and Bodo-the main tribes associated with agriculture, annual rainfall 1,600–2,000 mm with 270 days long growing period. Tea, cucumber, *Musa*, bamboo, brinjal, okra, *Abelmoschus pungens*, welsh onion (*Allium fistulosum*) of China extends up to the region. Ginger and turmeric- many local cultivars with desirable features, and in Citrus- *Citrus reticulata*. Sonowal Kacharis tribe uses *Allium sativum*, *Oryza sativa*, *Cassia sophera*, *Ricinus communis* and *Ananas comosus*. Variability in jackfruit (*Artocarpus heterophyllus*- although native of W Ghats), rich diversity found in Brahmaputra valley, Bihar, Assam Jharkhand, WB, UP; rich in carbohydrates and Vit A.

4. **Malwa Plateau Region:** Western part of Madhya Pradesh and parts of SE Rajasthan. Opium (*Papaver somniferum*) is a traditional crop; wild relatives, like *Abelmoschus tuberculatus*, *A. manihot* ssp *tetraphyllus* var. *megaspermus* (large seed), *A. crinitus*, *A. ficulneus*; *Pusa Nasdar* in ridge gourd from a local landrace from Neemach; *Cucumis* spp.; *Chironji* (*Buchnanania lanzan*); *ber*, *bael*, *karonda*, *khirni*, and custard apple.
5. **Bundelkhand Region:** South of Yamuna between fertile Gangetic plains stretching across northern Uttar Pradesh & southern highlands of MP. Brinjal, *Ziziphus*, *bael* (*Aegle marmelos*)- traditional varieties such as Kagzi Etawah, a known landraces; *chironji* - diversity has been recorded for panicle, fruit size and quality kernels; diversity in *aonla*, *ber*, and *karonda*.



Bhut Jolokia –the world's highest pungency; >1 million SHU.

6. **Garó, Khasi and Jaintia Hills:** Centre of diversity of *Rhododendron*, *Schima*, Zingiberaceae spices, *Citrus*, *Garcinia*; Tura range (Garó Hills)- *Citrus* & *Musa* species diversity; brinjal, *S. khasianum* with resistance to stem and fruit borer, *S.kurzii*-endemic to Garó hills. In chilli, **Bhut Jolokia** with the highest pungency so far; bittergourd, ashgourd, bottlegourd, chow-chow, *Cucumis hardwickii* - the likely progenitor of cucumber; *Mangifera khasiana*-endemic, distinguishable from *M. sylvatica*; in *Citrus*, 8 of the 17 species reported from the NE Region *C. indica*, *C. macroptera*, *C. latipes* naturally occur, presenting rich genetic diversity; *Musa flaviflora* is localized with four additional species. *Artocarpus heterophyllus*, *Litchi chinensis*, *Malus*, *Pyrus*, *Prunus spp.*, *Prunus persica*, *Rubus*, *Sorbus*, *Corylus*, *Castanea sativa*, *Prunus napaulensis*, *Pyrus cerasoides* while *Pyrus pyrifolia*, and *P. serotina* is grown semi commercially.
7. **North-eastern Hills:** Nagaland, Manipur, Mizoram and Tripura and Cachar. Brinjal, *Citrus*, *mango*, tropical and subtropical minor fruits, banana, pineapple, citrus, papaya, plum, peach, apple, cucumber with natural occurrence of *Cucumis hardwickii*, *Cucumis hystrix*, *L. graveolens*, *M. cochinchinensis*, *Trichosanthes ovata*, *T. khasiana*, providing a reservoir of useful genes; brinjal with rich diversity, *S. torvum*, *S. indicum* and *S. khasianum* possess resistance to shoot and fruit borer, and root diseases; *Annona*, *Averrhoa*; wild orange *C. indica* is found in the Naga Hills, whereas, lemon, *C. lemon*, is known with a large number of traditional cultivars, such as Hill lemon.
8. **Arid Western Region and Semi-arid Kathiawar Peninsula:** Western part of Rajasthan, parts of the south- western Haryana and Kathaiawar peninsula of Gujarat. Centre of diversity of *Citrullus*, *Cucumis melo* var *agrestis*, arid fruits, seed spices & *khejri* (*Prosopis cineraria*), marwar teak (*Tecomella undulata*), watermelon, *Citrullus lanatus*, *Cucumis melo* var. *momordica*, *Momordica balsamina*, mandarin (*Citrus reticulata*), kinnow mandarin, sweet orange (*C. sinensis*), *Ziziphus mauritiana-gola*, *seb* and *mundia*; *Punica granatum* – ‘Jodhpur Red’ with wider spread, and Jalore seedless; guar, moth bean; and *Cucumis prophetarum* in Abu Road (Sirohi District) of Rajasthan.
9. **Upper Gangetic Plains:** Northern Punjab, most of

Haryana and western, central and parts of eastern UP, spread from foothills of western Himalayas to Delta in West Bengal. Centre of diversity available are: *Benincasa hispida*, *Citrullus*, *Abelmoschus*, brinjal - *Ramnagar Baingan* (round green fruit type is suitable for Ganga River belt), *Dudhiya* (cluster), *Jethuwa*, *Kuchabuchia* (small clusters), *Jafrabadi*; *Balfahwa Jathuwa Bhanta*; *petha* near Agra, *A. manihot* ssp. *tetraphyllus* (Rampur/Saharanpur), and *A. tuberculatus* (Saharanpur), mango- Bombay Green, *Dashehari*, *Fazli*, *Langra*, *Safeda* Lucknow, *Smarbehisht*, *Chausa*; aonla- *Hathijhool*, *Basanti* red, *Deshi*, *Chakaiya*; jamun, lemon; and water chestnut (*Trapa natans*)- excellent coolant for body, perfect food, high nutrient, low calorie, fat loss, helps in jaundice, thyroid gland, controls diarrhea.

10. **Middle Gangetic Plains:** Eastern UP and parts Bihar, either side of Ganga and Saryu (Ghaghara), and Himalayan foothills and Vindhya ranges. Centre of diversity of cucurbits- *parwal*, (*Trichosanthes dioica*), *satputia* (*Luffa acutangula* var. *satputia*, also known as *Luffa hermaphrodita*); *Momordica*; in *Legeneria*- like *Rajendra Chamatkar* are developed through selection from local landraces; *Luffa echinata*, *Momordica cochinchinensis*, *Momordica subangulata renigera M. dioica*, and more; brinjal, chilli, *Abelmoschus crinitus*; mangoes- *Bathua*, *Bombai*, *Himsagar*, *Kishen Bhog*, *Sukul*, *Langra*, *Sundar Pasand*, *Fazli*, *Gulabkhas*, *Mahmood Bahar* and *Zardalu*’ litchi- North Muzaffarpur, Darbhanga is known for varieties, like *Bedana*, *Calcuttia*, *Purbi*, *Kasba*, *Desi*, *Early Bedana*, *Shahi*; bael, and *makhana* (*Euryale ferox*).



Date palm fruits in cluster

*Cucumis prophetarum* from Abu Road*Cucumis hardwickii* (2n=14); wild progenitor of Cucumber*Cucumis hystrix* (2n=24) amphidiploid

11. **Lower Gangetic Plain or Delta Region:** Parts of WB and Bangladesh where Ganges, Brahmaputra and Meghna meet into Bay of Bengal. *Ipomoea aquatica*, okra, *Momordica*, *Trichosanthes*, *Musa*, ornamentals, *parwal*, elephant foot yam, moringa, *kakoda*, *kostumbari* coriander, brinjal, okra-*Abelmoschus manihot*, *A. crinitus*; mango- *Bombai*, *Himsagar*, *Kishen Bhog*, *Langra*, *Malda*; jackfruit, and *karonda* (*Carissa carandus*).
12. **Chhotanagpur Plateau:** SE plateau of Jharkhand bordering Odisha, WB, Bihar and Chhattisgarh. Variability included in litchi, jackfruit, cucurbits, *Dendrobium* – a epiphytic orchids (over 10 species); ginger- *Maran*, *Kuruppampadi*, *Ernad*, *Wynad*, *Himachal and Nadia*; mango- *Bathua*, *Bombai*, *Himsagar*, *Kishen bhog*, *Gopalbhog*, *Sukul*, *Ranipasand*, *Safed maldah*, *Chausa*, *Fazali*, *Zardalu*, etc. extending from middle and lower Gangetic plain; Jackfruit in the Santhal Parganas; litchi- *Shahi*, *Rose scented*, *China*, *Purbi*, *Early bedana*, *Late bedana*; and lac.

*Momordica cochinchinensis*

Winged bean



Areca nut in cluster



Carambola, Kamrakh

13. **Bastar Region:** Chhattisgarh including Dandakaranya, Bastar, Kanker, Dantewada, Bijapur, Narayanpur, and Abujhmarh. Inhabited by *Abujhmarhias*, *Gonds*, *Muria*, *Maria*, *Dhorla*, *Bhatra*, *Halba*, and *Dhurva* tribes. Mainly root and tuber crops, *Diospyros melanoxylon*; cassava, greater yam, aerial yam, sweet potato, elephant foot yam, *banda*, ginger, turmeric, wild tubers (*Dioscorea*), *pitkanda* (*D. dumetorum*), *kulihakanda* (*D. hispida*);
- other useful tubers- *tikhur* (*Zingiber roseum*), tannia, *vidarikand* (*Pueraria tuberosa*), making the region a very important centre of diversity for root and tuber crops; also *chironji*; *karonda*- rich diversity; melon, cucumber, bottlegourd, ridge gourd, smooth gourd, and a host of medicinal plants, including *keaukanda* (*Costus speciosus*) used to treat cancer.
14. **Koraput Region:** Southern-eastern Odisha and some districts of north-eastern Andhra Pradesh-Nizamabad, Vizagapatnam, Vijanagaram, Srikakulam. Inhabited by *Gonds*, *Khonds*, *Santhals*, *Lahqulas* and *Kinnaras*. Mainly root and tuber crops, cucurbits, *Luffa*, *Momordica*, yam, sweet potato, *Musa balbisiana*, jackfruit, mango, *aonla*, date palm, *karonda*, pomello, tamarind, *tejpat* (*Cinnamomum tamala*), turmeric, pepper, ginger, etc.; brinjal, chillies for fruit colour, shape, size and pungency. *S.indicum*, *S. incanum*, *S. surattense*, *S. pubescens*; *kundru*, *Cucumis hystrix*, *L. acutangula*, *L. graveolense*, *Luffa umbellata*, *Momordica balsamina*, *M. cochinchinensis*, *M. dioica*, *M. tuberosa*, *Trichosanthes bracteata*, *T. cordata*, *Trichosanthes multiloba* and *Trichosanthes himalensis*; okra- rich for fruit and plant types, *A. crinitus*, *A. ficulneus*. In root and tuber crops- *pitharu kanda* (*Dioscorea belophylla*), potato yam (*D. bulbifera*), *pitta kanda* (*D. glabra*, *D. wallichii*, *D. wightii*); Indian Kudzu vine (*Pueraria tuberosa*), and *Vigna vexillata*.
15. **South Eastern Ghats:** Dry areas of southern Andhra Pradesh and Karnataka. Mainly root and tuber crops; brinjal- *Sanna vanga*, *Saara vanga*, *Tella mulaka*, *Tella vanga* and *S. erianthum*, *S. nigrum*, *S. surattense* and in chillies for fruit colour, *Cucurbita pepo*, *Cucumis melo* var. *agrestis*, *C. pubescens*, *M. balsamina* and *M. tuberosa*; *Vigna hainiana*, elephant-foot Yams, *Amaranthus spinosus*, *A. tenuifolius*, *A. dubius*; variability in coriander, mango- *Banganpalli*, *Totapari*, *Cherukuramam*, *Himayuddin*, and *Suvarnarekha*; bael, palmyra palm (*Borassus flabellifer*), *karonda* (*Carissa carandus*), lime, *Commiphora caudata*, wood apple, *aonla*, *Phoenix*, clove, and *Ziziphus horrida*, *Morus alba*; and mulberry- powerhouse of nutrients, excellent source of proteins, reservoir of antioxidants, improves digestion, lowers cholesterol, promotes brain health, and improve immunity.
16. **Kaveri Region:** Kaveri delta of Tamil Nadu- Coromandel plains, South Arcot, North Arcot, Kolli

malai, Pachamalai hills. Mainly *Trichosanthes*, Root and tuber crops, *Syzygium*, banana, bitter gourd, snake gourd, brinjal, chilli, *Canavalia*, moringa, *agathi*, *Cucurbita moschata*, *Cucumis melo* var. *anguria*, *Trichosanthes cucumerina* var. *cucumerina*, *Solanum nigrum*, *S. surattense*, *Momordica balsamina*, *M. tuberosa* and wild okra *Abelmoschus angulosus*; aroids, elephant foot yam, tannia, giant taro, mangosteen, wood apple, clove, mango- *Banganpalli*, *Bangalora*, *Neelum*, *Rumani*, *Mulgoa*; in banana- *Pachable*, *Karpurvalli*, *Monthan*, *Morris*, *Mysore poovan*, *Nendran*, *Pachanadan*, *Rasthali*, and *Robusta*.

17. **North-western Deccan Plateau:** Parts of NW Andhra Pradesh, Western Ghats, extending from Satpura-Mahadeo hills in the north to the Bellary-Dharwad. Mainly occur *Vigna* spp., *Citrus*, *Annona squamosa* (huge variability in custard apple), *Vitis vinifera*, *Tamarindus indica*, *Hibiscus cannabinus*, *H. sabdariffa*; sweet orange, *Flacourtia indica*, wood apple, jamun, grapes, mango- *Alphonso*, *Mankurad*, *Muloga*, *Pairi*, *Banganpalli*, *Totapari*; *Mosambi*, *Sathagudi*, *Malta*; sweet lime (*Citrus sinensis*); Nagpur mandarin, kagzi lime, and yam.
18. **Konkan Region:** Hot humid region of Western Ghats, coastal plains of Maharashtra, Goa, and Uttar Kannada. Centre of diversity of *Vigna* spp. *Vigna* species occur or cultivated in the region are *V. aconitifolia*, *V. angularis*, *V. dalzelliana*, *V. mungo*, *V. radiata* var. *setulosa*, *V. radiata* var. *sylvestris*, *V. sublobata*, *V. trilobata* var. *trilobata*, *V. trilobata* var. *pilosa*, *V. umbellata*, *V. vexillata*, *V. vexillata* var. *stocksii*, *V. khandalensis*, *V. unguiculata* ssp. *sesquipedalis*, offering valuable genetic diversity; spices, mango, *Garcinia*, *Artocarpus heterophyllus*, *A. hybridus*, *A. paniculatus*, *A. polygamus*, *A. spinosus*; bhindi, chillies, *kundru*, lablab bean, bottle gourd, bitter gourd, snake gourd, elephant-foot yam- *A. sylvaticus*, *A. konkanensis*, taro; yam, *Flemingia procumbens*, tapioca, pineapple, coconut, mango, strawberry, *Musa acuminata*, *M. sapientum*; jackfruit; arecanut, *Carum strictocarpum*, turmeric (*Curcuma inodora*), nutmeg, pepper, *Vanilla vatsalana*, ginger, harjodwa (*Cissus quadrangularis*), *Abelmoschus angulosus*, *A. ficulneus*, *A. manihot* ssp. *manihot*, *Cucumis ritchei*, *C. setosus*, *Momordica dioica*, *M. tuberosa*, *Mangifera sylvatica*, *tejpatta*-*C. tamala* and *C. goaense* etc.

19. **Malabar Region:** Hot humid, southern part of Western Ghats and coastal plains ; Dakshin Kannada in the north to Kanyakumari in the south, including whole of Kerala, south of Karnataka. Primary centre of diversity of spices, *Syzygium*, *Garcinia*, *Artocarpus heterophyllus*, coconut, *M. dioica*, *M. tuberosa*; *Trichosanthes anamalayensis*, *T. cucumerina* var. *cucumerina*, winged bean (*Psophocarpus tetragolobus*) , yam bean, cardamom- 3 distinct types (*Malabar*, *Mysore* and *Vazhukka*); wild relatives of *Amomum*, *Cinnamomum*, *Piper*, *Areca Catechu*, *Curcuma*, *Zingiber*, *Myristica* and *Vanilla* offer useful variability for use; banana-red skinned *Kappa*, large yellow skinned *Nedra*, small yellow- *Kadali*, *Rasakadali*, *Poovan*, *Matti*, *Palayamkodan*; mango- *Mundappa*, *Plour*, *Pairi*; in jackfruit *Varikka* landrace has quality fruit;. Coorg Mandarin from Karnataka *Garcinia gummi-gutta* - *Kokam*; wild relatives of *Artocarpus*, *Diospyros*, *Garcinia*, *Syzygium*, and *Vitis* offer significant variability for use.

Vegetable and fruit crops are low in calories but contain high levels of vitamins and minerals (Janick, 2005), making them indispensable for balancing our daily diet. Although the supply of horticultural products is increasing, the diversity and nutritional value of the products are decreasing (Khoury *et al.*, 2014) . These decreases can be partially attributed to the narrow genetic diversity of horticultural crops resulting from domestication and breeding as well as reproductive barriers that inhibit genetic introgression from wild relatives (Kalloo, 1992). Therefore, the generation of genetic resources with diverse and desirable characteristics will be of great value for improving horticultural products. While the conventional approaches can be accomplished by the direct utilization of HGR in breeding program, recent advances in biotechnology have progressively used various tools (such as next generation sequencing, SNP genotyping array and genotyping by sequencing, genome-wide association studies-GWAS, MAS and genomic selection-GS) for selecting potential parents from germplasm collections.

Recent Advances

Horticultural crops are an excellent source of vitamins, antioxidants, and fibers that play an important role in human health. Highlighting the importance of horticultural crops and responding to the main challenges

as well as the use of modern technologies in breeding is of paramount importance to meet the nutritional needs in the light of climate change and the trend for sustainable agriculture. The implementation of multi-omics approaches including genomics, epigenomics, transcriptomics, proteomics, metabolomics, and microbiomics is of great importance in order to reveal quality changes in horticulture crops.

Recent advances in automation and high throughput techniques used in decoding plant genomes play an important role to speed up the genomic research. With the establishment of genome and transcriptome sequencing for several horticultural crops, huge wealth of sequence information have been generated which have been used extensively for analysing and understanding genome structures and complexities, comparative and functional genomics and to mine useful genes and molecular markers. However, certain limitations present a number of challenges for the generation and utilization of genomic resources in many important crops. Given the development and advantages of genome-editing technologies, research that uses genome-editing to improve horticultural crops has substantially increased in recent years. With the advent of CRISPR/Cas9, the application of genome-editing to horticultural crops has greatly advanced. The goal of breeding is to harness genetic variations to introduce desirable traits. These genetic variations can arise in various ways, such as by spontaneous mutation, chemical mutagenesis, and physical mutagenesis (Chen *et al.*, 2018). Gene-editing could be regarded as biological mutagenesis. In comparison with other approaches, genome-editing technology is superior in terms of versatility, efficiency, and specificity. Through genome-editing, desirable traits can be directly introgressed into elite or heirloom lines without compromising other properties, and the resulting lines with targeted improvement will be ready for use in crop production (Zang *et al.*, 2016). The wild relatives of cultivated varieties are also potential materials for genome-editing because they generally present unique features in many important traits.

The horticultural crop management is a fit case for using disruptive technologies. Robotics, AI, and IoT are all technologies that have the potential to radically transform the way we grow food. These are poised to revolutionize horticulture as we know it. Robotics can take over many of the tasks currently performed by human workers, from transplanting and watering to

harvesting and packaging. AI can be used to monitor crops and optimize conditions for maximum yield. And IoT devices can provide real-time data on everything from soil moisture levels to pest infestations. Drones or Unmanned Aerial Vehicles (UAV) with sensor and imaging capabilities can play an increasingly role in identifying and reducing crop damage. In India, over 80 per cent farmers are small and marginal (<1 ha), it is difficult to manage invasive pests. If one field is sprayed, the pests shift to the neighbouring fields. Parameters related to drone-based spraying such as nozzle type, droplet size, drone-type, spread, density, uniformity, deposition, and penetrability should also be a factor during implementation of mitigation strategies. It can be employed in several field operations and is an excellent tools for rapid, reliable, and non-destructive detection of field problems. Not only will these make our crop production more efficient and sustainable, but it will also free up farmers. In combination with vertical farming, these technologies could increase the efficiency and quality of horticultural products. The entire growth process could be digitized and made available in the form of algorithms. This would allow tech companies to get into the food-growing business. And they could probably do it much cheaper and faster.

Although the supply of horticultural products is increasing, the diversity and nutritional value of the products are decreasing. These decreases can be partially attributed to the narrow genetic diversity of horticultural crops resulting from domestication and breeding as well as reproductive barriers that inhibit genetic introgression from wild relatives. Therefore, the generation of genetic resources with diverse and desirable characteristics will be of great value for improving horticultural products.

Future Perspective

There is tremendous scope for enhancing the productivity of Indian horticulture which is imperative to cater to the country's estimated demand of 650 mt of fruits and vegetables by the year 2050.

1. Some of the new initiatives like focus on planting material production, cluster development program, credit push, promotion of FPOs are the right steps in this direction. An integrated holistic approach is needed to increase horticultural productivity by adoption of growth enhancing technologies, pest management systems and 'precision farming', automation (can reduce over-

application of agrochemicals), vertical farming, soil-less horticulture, protected cultivation (better dividends, also brings pride to the profession by attracting youth including women as well). At present, only ~50,000 ha are under protected cultivation in India, whereas China has 2 m ha. There is need to increase 4 times the area (~2,00,000 ha) in the next 4-5 years. It not only provides high water and nutrient use efficiency but it can easily increase the productivity by 3-5 folds over open field cultivation (GoI, 2019).

2. One of the major components in precision agriculture in crop health monitoring, which includes irrigation, fertigation, pesticide sprays, and timely harvest of the crop. In order to accomplish above operations, drones are highly useful for on-site detection of problems so as to act instantly for corrective measures. As labour availability and technical manpower are meagre, drones are gaining popularity. In India, over 80 per cent farmers are small and marginal (<1 ha), it is difficult to manage invasive pests. If one field is sprayed, the pests shift to the neighbouring fields. Parameters related to drone-based spraying such as nozzle type, droplet size, drone-type, spread, density, uniformity, deposition, and penetrability should also be a factor during implementation of mitigation strategies. It can be employed in several field operations and is an excellent tool for rapid, reliable, and non-destructive detection of field problems.
3. Use of plastic mulch (25% more yield than no mulched), crop cover or low tunnels (for early crop and protection from low temperature), walk-in tunnels (for temperate region off-season vegetables), naturally ventilated polyhouses (tomato, cucumber, tomato, flowers), net houses (for large number of vegetables and ornamental plant nurseries), environment-controlled greenhouses (healthy nursery and foliage plants), vertical farming of lettuce, strawberry etc., soil-less farming (hydroponics and aeroponics for vegetables), and vegetable grafts, are some important technological interventions that need to be scaled up and adopted more widely. The use of vertical farming (growing low crops in multiple layers, mostly inside buildings) and urban/peri-urban farming (the growing of plants within and around cities) which contributes for increasing access to food, advancing livelihoods and improving the environment (waste management, reduce CO₂ emissions) combined with technologies such as hydroponics, allows us to make efficient use of space and reduce the distance our food travels to get to consumers. Designing of improved management (storage, packaging, processing and local marketing) will be crucial from a value-chain perspective would create new opportunities for job and income creation, and is also crucial for generating access to fresh and nutritious food to a wider urban population. Another new and interesting development in Japan uses a biopolymer for 'film-farming', which uses 90% less water than conventional farming and offers a viable alternative to resource-intensive horticulture. Promotion of cold-chain management, harvest and post-harvest management would require cluster identification of horticulture produce and creation of infrastructure for aggregation of the produce, pre-conditioning-cleaning, sorting, grading, packaging, transport and/or storage facilities, processing and market linkages. *Mandis* need to have pledged facilities to avoid distress sale. To enhance the delivery effectively, there is a need for innovations to be scaled in PPP mode for better adoption in horticulture. Involvement of youth for technical backstopping, input supply will be helpful. Further, a thrust on secondary agriculture would be beneficial, as farmers can fetch higher price for their produce subjected to value-addition.
4. R&D for using cutting-edge technologies like genomics, proteomics, metabolomics, phenomics for genetic improvement need to be adequately funded. Concerted efforts need to be made to reorient breeding programs to target the traits like enhanced productivity, seedlessness, canopy architecture, nutrient use efficiency, biotic/abiotic stress resistance, shelf-life improvement, and biofortification, etc. Dryland/arid horticulture needs to be given focused attention for which new varieties/hybrids and related cultivation technologies need to be developed on priority.
5. GoI's Mission for Integrated Development of Horticulture (MIDH) is required to address all the activities starting from cultivation till the produce reaches to the consumers in an acceptable form. There is need to establish functional block level resource centres, including facilities for low-cost value-addition (secondary agriculture). Processing

of fruits and canning of vegetables can multiply their value 50 to 500 times. This would not only save post-harvest losses but also add to employment generation at the local level by engaging youth (including women) and creating village level entrepreneurs. Effective coordination is needed among MoA&FW, Ministry of Food Processing Industries and Ministry of Commerce and Industry. For aggregation of farm produce, there is a need for decentralization and modernization of market yards, railway freight operations need to be strengthened through temperature-controlled containers along with loading and unloading facilities, to reduce post-harvest losses and connect land-locked states to export markets. Private entrepreneurs should also be incentivized to establish small farm implement mechanization hubs for every 1,000 ha and big machinery hubs for every 5,000 ha of cultivated area.

Thus, HGR impacts as well as gets impacted by climate change. Crop diversity regimes, especially landraces and farmers' varieties, which must sustainably increase productivity, resilience (adaptation), mitigation (removal of GHG) and biodiversity conservation are needed. These outcomes and the related activities interact in a complex manner and cut across a number of stakeholders, seeking synergistic integration of gene smart, water smart, soil and nitrogen smart, energy smart, carbon smart, weather smart, and knowledge smart development pathways to green the economy. Investment in PGR conservation and its utilization in climate smart agriculture (CSA) should be suitably enhanced and linked with an effective monitoring, evaluation and impact mapping system. Innovative approaches to social safety-nets, including new insurance products, will be needed to augment house hold resilience. The science-policy interface is necessary to sensitize policy makers. As we move towards an Evergreen Economy, crop diversity should be mainstreamed into the national and international policies in mutual harmony with CSA. For increasing production, let us pledge to develop varieties/hybrids/ transgenics that help increasing production by 25% from current levels; varieties and technologies that use fewer resources but permit acceptable or relatively

better output; improve profitability of farming and living conditions of farmers; and involve women and rural youth in agri-horticulture. Feeding hungry is our duty. If we fail to feed the present generation due to pre-conceived fears of frontier technologies, then there would probably increase hunger and under-, malnutrition in future generations. Emerging technologies including biotechnology are not the enemy but hunger is real enemy that affects around one billion and starvation that causes millions of deaths every year. Without adequate food supplies at affordable prices to needy, we cannot expect world health or peace. Judicial blending of traditional and responsible frontier technologies is our future.

References

- Chen L, W Li, L Katin-Grazzini, J Ding, X Gu, Y Li, T Gu, Renwang, X Lin, Z Deng, Richard J McAvoy, Frederick G, Gmitter Jr, Z Deng, Y Zhao and Y Li (2018) A method for the production and expedient screening of CRISPR/Cas9-mediated non-transgenic mutant plants. *Hortic. Res.* **5**: article no 13.
- GoI (2019) Report on Policies and Action Plan for a Secure and Sustainable Agriculture (Chaired by RS Paroda). Principal Scientific Advisor to the Government of India, Vigyan Bhawan Annexe, New Delhi, 198 p.
- Gupta A and Yadav N (2016) Evaluation of nutritional and anti-nutritional activity of indigenous and underutilized green leafy vegetables of North India. *Int. J. Food Nutr. Sci.* **5**: 88-95.
- Janick J (2005) Horticultural plant breeding: Past accomplishments, future directions. *Acta Hortic.* **694**: 61–65. <https://doi.org/10.17660/ActaHortic.2005.694.6>.
- Kalloo G (1992) Utilization of wild species. In: G Kalloo, TR Chaudhury (eds.) *Distant Hybridization in Crop Plants*. Pergmon Press, Oxford UK, pp. 587-604.
- Khoury CK, AD Bjorkman, D Hannes, J Ramirej-Villegas, L Guriano, A Jarvis, LH Rieseberg and PC Struik (2014) Increasing homogeneity in global food supplies and the implications for food security. *Proc. Natl Acad. Sci. USA* **111**: 4001–4006. <http://doi.org/10.1073/pnas.1313490111>
- Rathore DS, Srivastava U, Dhillon BS (2005) Management of genetic resources of Horticultural crops: Issues and Strategies. In: BS Dhillon, RK Tyagi, S Saxena and GJ Randhawa (eds) *Plant Genetic Resources: Horticultural Crops*. Narosa Publishing House, New Delhi, pp. 1-18.
- Zhang Y, Zhen Liang, Yuan Zong, Yanpeng Wang, Jinxing Liu, Kunling c, Jin-Long Qiu and Caixia Gao (2016) Efficient and transgene-free genome-editing in wheat through transient expression of CRISPR/Cas9 DNA or RNA. *Nat. Commun.* **7**: 1261.

Conservation and Use of Underutilized Crops: Challenges and Way Forward

Bhag Mal

Trust for Advancement of Agricultural Sciences (TAAS), Avenue II, Indian Agricultural Research Institute (IARI), Pusa Campus, New Delhi -110012, India

This paper reinforces the importance of underutilized crops and the need for organized research efforts for their improvement. These include species also classified as underexploited crops, under-developed crops, underused crops, abandoned crops, lost crops, orphan crops, minor crops, traditional crops, forgotten crops, vanishing crops, local crops, indigenous crops, niche crops, life support species, poor people's crops, subsistence crops, crops for future, nutri-rich crops, and new potential crops, which have great potential to be used in mainstream agriculture. The domestication of new crops would promote agricultural diversity and could provide a solution to many of the problems associated with intensive agriculture. Being nutritionally very rich, they have proved good potential for food and nutritional security, health and income generation especially for local communities. The use of modern science to improve their productivity, value addition and use by agro-industries are new opportunities that need to be harnessed.

Introduction

The dependence of mankind on plant resources is inevitable. The paradox of agriculture is that there are 300,000 known plant species in the world out of which 100,000 are used by humankind in various ways. About 30,000 species are edible and 7,000 species are used as food at the local level and only 150 species have been commercially cultivated. Only about 30 species provide 90 per cent of plant calories, while only three species – rice, wheat and maize meet 60 per cent demand. These include cereals, legumes, root and other food crops (Paroda, 1988). Notwithstanding the food shortages becoming more acute particularly in the developing countries, an increased dependence on plants rather than animals for the protein in their diet is felt necessary. On the other hand, the ever increasing human population particularly in the developing world is a never diminishing challenge to growers. It has, therefore, become necessary to explore the possibilities of exploiting newer plant resources in order to meet the growing food requirements. There are large number of species known by various names as underutilized crops, underexploited crops, under-developed crops, underused crops, abandoned crops, lost crops, orphan crops, minor crops, traditional crops, forgotten crops, vanishing crops, local crops, indigenous crops, niche crops, life support species, poor people's crops, subsistence crops, crops for future, nutri-rich crops, and new potential crops which have great potential to be exploited.

The concerns focussing attention on new or underexploited crops include resource management, agricultural diversification, self sufficiency, economic gains, germplasm augmentation, conservation, and nutrition. Interests in the underutilized plants are spurred by quite different reasons in developed and developing countries, which need strategic planning. These form a continuum related to crop diversification, production of value added products, rural development and increased income for growers and processors (Williams, 1993). The new crops/underutilized crops continue to be neglected and have to struggle hard in the competition for support and mostly underdeveloped markets.

Sustainable agriculture requires enhancement of the world's productive capacity as well as the conservation of resource base (Walsh, 1991). The traditional crops including lesser known pulses and pseudocereals have been grown for generations on marginal soils under low input management. This situation does not exist today in the same magnitude as even the small farmers are expected to contribute towards increased agricultural production for which new innovations in production technology including the use of alternative or new crops need greater emphasis.

The focus of scientific attention on a particular underutilized crop depends on availability of landraces which can adapt relatively quickly, possess an ability to fit well into cropping systems and capability of giving high yield of acceptable products. These factors need

*Author for Correspondence: Email-bhagml@gmail.com

strategic planning for identification of real promising underutilized crops out of a large number available. The importance of underutilized crops and the need for organized research efforts, for their improvement have been amply emphasized (Arora and Bhag Mal, 1991; Bhag Mal, 1990, 1993; Bhag Mal and Joshi, 1991; Bhag Mal *et al.*, 1992; Bhag Mal and Paroda, 1988; Paroda and Bhag Mal, 1992; Paroda *et al.*, 2022; Williams, 1993).

Asia-Pacific region possesses a rich genetic diversity of several plant species which are presently underutilized but have a great potential to be exploited for food production only in different countries within the region but also elsewhere in the world. The potential underutilized crops which merit urgent attention for improvement include a few pseudocereals, viz., amaranth, buckwheat, chenopods and grain legumes, viz. rice bean, adzuki bean, faba bean, winged bean, moth bean, horse gram, hyacinth bean, grasspea, French bean, lima bean, sword bean, jack bean and zombi pea.

In view of the need for diversification of future agriculture to meet various human needs, it is very timely to seriously consider paying enhanced attention on using the underutilized crops especially grain legumes and pseudocereals. One such approach is to explore non-conventional pathways such as wider adoption of UUCs, as possible future crops due to the fact they are adapted to a range of agro-ecologies, are nutrient-dense and offer better prospects in marginal production areas with low input agriculture. In fact, this approach is an affirmation of sustainable farming systems and human wellbeing known to indigenous local communities for generations. The domestication of new crops would promote agricultural diversity and could provide a solution to many of the problems associated with intensive agriculture. Being nutritionally very rich, they have proved good potential for food and nutritional security, health and income generation especially for local communities. The use of modern science to improve their productivity, value addition and use by agro-industries are new opportunities that need to be harnessed.

Challenges and Opportunities

There are several challenges and constraints which hamper the growth and development of neglected and underutilized crops and need to be addressed. These include: relatively low yield, limited germplasm and lack of improved varieties, lack of improved agronomic

practices, lack of scientific and technical information – production, consumption, utilization; problems associated with taste, cooking and antinutritional traits; lack of interest by researchers, agriculturists and extension workers; lack of awareness on economic benefits and market opportunities; lack of technology for processing, value addition and product development; lack of promotion of specific potential crops; lack of coordination between diverse stakeholders; lack of enabling policies, initiatives and incentives; and lack of credit and investment. Also these crops suffer with several problems relating to development of improved varieties such as asynchronous maturity, small seed size, lodging, seed shattering, difficulty in threshing, weed control, outcrossing, lack of systematic breeding, lack of well-defined seed production system, and lack of resistance to biotic and abiotic stresses.

These species are extremely important for diversification of agriculture as they are immensely helpful in enlarging narrow food base, sustainability of high-input agriculture at risk, rehabilitation of ecosystem, providing protection against climatic changes/unforeseen circumstances, extending cultivation to marginal/degraded lands, balanced nutrition- micronutrient deficiency, meeting the changing human needs, promoting export potential and posterity for future use. There are tremendous opportunities for exploiting the potential of these species due to their manifold benefits such as contributing to poverty alleviation, income generation, widening the food basket, adding nutrients to diet, providing food for low income group people, adapted to fragile environments, contributing to the stability of agroecosystems, meeting new market demands, and value addition and product development.

The underutilized species also play an important role in the subsistence farming and economy of poor people; have high potential for dietary diversification and nutritional enrichment – micronutrients such as vitamins and minerals; provide important environmental service - adapted to marginal soils and diverse climatic conditions; contribute towards food and nutrition security; possess high local importance in consumption and production systems; highly adapted to agro-ecological niches/marginal areas; represented by ecotypes/landraces contributing useful genes; and are cultivated and utilized relying on indigenous knowledge and hence provide enormous opportunities for wider utilization. These crops provide better water foot print, better carbon

foot print and also security in terms of food, feed, fodder and biofuel (green energy). They contribute towards Sustainable Development Goals 2, 3, 12 and 13 which address zero hunger, healthy lives, sustainable consumption and climate change.

Some of these species such as amaranth, buckwheat and chenopods possessing exceptionally high nutritional value attributed to high proteins and minerals (Fig.1) and higher amount of essential amino acids (Table 1) as compared to cereals definitely have the potential to become important components of agriculture. Amaranth, of course, has a great potential to offer for commercial cultivation in the plains as well.

National Program on Underutilized Crops

Amongst the developing countries, India is the only country having well organized national program on underutilized crops. The All India Coordinated Research Project (AICRP) on Underutilized and Underexploited Plants was initiated in 1982 with its headquarters at NBPGR involving 29 crops. Realizing the need for prioritization, the number of crops was reduced to 18. In 2002, the project was converted to network mode as All India Coordinated Research Network (AICRN) on Underutilized Crops with 18 crops. Subsequently, based on the recommendations of the Quinquennial Review Team (QRT), the name of Network was changed

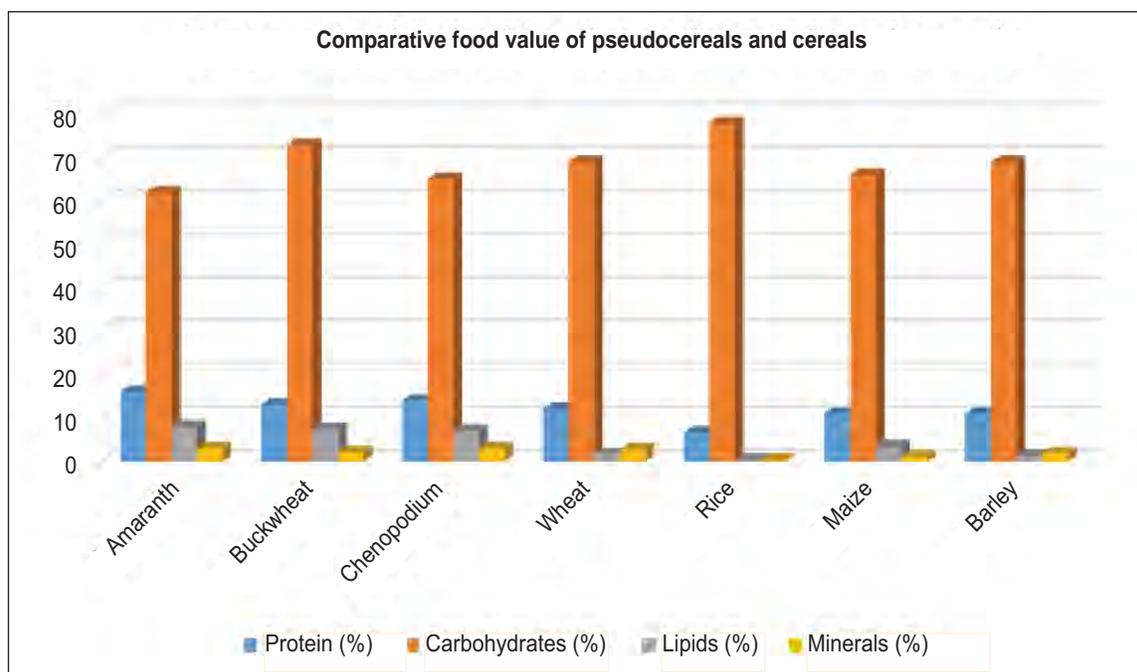


Fig. 1. Comparative food value of pseudocereals and cereals (Bhag Mal, 1994)

Table 1. Amino acid composition of pseudocereals and cereals

Amino acids	Content (g/100g protein)						
	Amaranth	Buckwheat	Chenopod	Wheat	Rice	Maize	Barley
Leucine	4.7	6.2	5.7	5.8	8.5	13.0	7.5
Isoleucine	3.0	3.7	3.3	3.3	4.5	4.1	4.0
Lysine	5.0	6.2	6.0	2.2	3.8	2.9	3.0
Arginine	6.6	10.1	6.9	3.6	3.7	2.9	3.8
Histidine	2.5	2.2	1.8	1.7	1.9	1.8	1.9
Methionine	4.0	1.6	2.2	2.1	3.0	3.4	3.2
Phenylalanine	6.4	4.2	4.1	4.2	8.4	6.4	8.2
Threonine	2.9	3.7	4.0	2.8	3.9	2.7	3.2
Valine	3.6	5.1	4.0	3.6	6.7	5.6	4.7
Tyrosine	6.4	3.2	3.2	8.6	9.1	4.6	8.2
Cysteine	4.0	1.6	1.2	3.7	3.0	3.4	3.7

Bhag Mal (1994)

as AICRN on Potential Crops in 2014 embracing the important priority crops, namely, grain amaranth (*Amaranthus* Spp.), buckwheat (*Fagopyrum* spp.), chenopod (*Chenopodium* spp.), quinoa (*Chenopodium quinoa*), Job's tear (*Coix lacryma Jobi*); adzuki bean (*Vigna angularis*), faba bean (*Vicia faba*), rice bean (*Vigna umbellata*), winged bean (*Psophocarpus tetragonolobus*); perilla (*Perilla frutescens*), paradise tree (*Simarouba glauca*), *Moringa oleifera*; kalingada (*Citrullus lanatus*), kankoda (*Momordica dioica*); *Vigna trilobata*, *Vigna glabrescens*; tumba (*Citrullus colocynthis*), *Vigna marina*. These species were prioritized for intensive research and development.

Significant progress has been made under the AICRN Project for collecting conservation, characterization, evaluation, conservation and utilization over the years. Also, International Workshop on Life Support Species in Asia and the Pacific Region was organized in collaboration with Commonwealth Science Council (CSC), UK at NBPGR, New Delhi, 1987 and the Proceedings were brought out.

Germplasm Conservation

As a result of concerted efforts, a total of 9,348 germplasm accessions of different underutilized crops, namely, pseudocereals, food legumes, oilseeds, vegetables, fodder crops, and crops for problem soils have been presently conserved in the National Genebank at the National Bureau of Plant Genetic Resources (NBPGR) and the field genebanks at different centres. The details of crop groups, crops and number of accessions conserved are given in Table 2.

Varieties Developed and Released

Under the All India Coordinated Program on Underutilized Crops at different centres, intensive selection/breeding programs are underway since its inception in 1982. Till date, a total of 60 varieties in different crops have been released and notified by the Central Variety Release Committee (CVRC) and the State Variety Released Committees. These varieties have been recommended for cultivation in different agro-ecoregions in the country. The details of crops and varieties released are given in Table 3.

The Way Forward

Germplasm Acquisition, Characterization and Conservation

- Ecogeographic surveys need to be conducted to

Table 2. Germplasm of underutilized crops conserved in national genebank/field genebanks

S. No.	Crop Group	Crops	No. of Accessions
1	Pseudoce reals	Amaranth (<i>Amaranthus</i> spp.)	5,815
		Buckwheat (<i>Fagopyrum</i> spp.)	1,095
		Chenopodium (<i>Chenopodium</i> spp.)	204
2	Minor cereal	Job's tears (<i>Coix lacryma jobi</i>)	150
3	Food Legumes	Adzuki bean (<i>Vigna angularis</i>)	200
		Faba bean (<i>Vicia faba</i>)	882
		Winged bean (<i>Psophocarpus tetragolobus</i>)	219
4	Oilseeds	Perilla (<i>Perilla frutescens</i>)	321
		Paradise tree (<i>Simarouba glauca</i>)	1
		Moringa (<i>Moringa</i> sp.)	13
5	Vegetables	Kalingada (<i>Citrullus lanatus</i>)	250
		Kankoda (<i>Momordica dioica</i>)	54
6	Fodder	<i>Vigna trilobata</i>	144
7	Crops of problem soils	Tumba (<i>Citrullus colocynthis</i>)	138
Total			9,348

develop the database on origin, distribution, habitat, agro-climatic requirements, benefits and scientific use of potential underutilized crops. There is also need for intensive studies on basic aspects of research especially on crop biology, nature and extent of pollination, ploidy level and crop husbandry.

- Well organised exploration programmes need to be undertaken to collect the existing diversity for which there is an urgent need to fix priorities for different crops and areas to be explored. The areas having rich diversity of landraces, primitive cultivars and wild relatives of specific crops need to be identified.
- Greater thrust needs to be given for collecting germplasm including locally adapted unique types as well as wild species from hitherto unexplored areas and the niche areas having rich genetic diversity and resistance against diseases, insect-pests, nematodes, lodging, seed shattering, drought, frost, salinity, etc. Germplasm with specific desirable traits also need to be introduced from exotic sources.
- Explorations should also be aimed at collecting material for filling the gaps in existing collections particularly the gene sources for photo-insensitivity, indeterminate growth habit and resistance against

Table 3. Varieties of underutilized crops released

Crop	No.	Name of varieties
Amaranth	19	Annapurna, PRA-2, GA1, GA-2, GA3, GA4, GA5, GA6, Durga, BGA-2, VL Chua 44, VLChua110, RMA-4, RMA-7, KBGA-1, KBGA-4, Phue Kartiki, Chhattisgarh Rajgira-1, Suvadra
Buckwheat	6	Himpriya, VL Ugal 7, PRB 1, Himgiri, Sangla B-1, Him Phaphra
Chenopod (<i>Bathua</i>)	2	Him Bathua, Pusa Green
Qinova	1	Him Shakti
Winged bean	4	AKWB-1, Indira Winged Bean-2, Chhattisgarh Chaudhari Sem-2, Phule Chardhari Wall (PB 11-2)
Faba bean	5	WH 82-1, HFB-1, HFB-2, Suvarna Gaurav, Suvarna Suraksha
Rice bean	11	RBL-1, PRR-2, RBL-6, VRB-3, KBR-1, Bidhan Rice bean-3, Bidhan -1, JRBJ05-2, Shyamla, Surabhi, Jawahar Rice Bean-2
Adzuki bean	1	HPU 51
Kalingada	3	Gujarat Kalingada-1, GK-2, CAZRI Kalingada-1
Kankoda	3	Indira Kankoda-1, Indira Kankoda-2, Chhattisgarh Kankoda-2
Perrila	2	RC Manithoiding 1, RC Manithoiding 2
Tumba	1	RMT 59 (Mansha Marudhara)
Jatropha	1	Chatrapat (SDAUJ 1)
Job's Tear	1	Bidhan Coix 1
Guayule	1	HG-8

Source: Haiger, 2022

diseases, insect-pests, nematodes, lodging, seed shattering, drought, frost, salinity etc. Germplasm with specific desirable traits also need to be introduced from exotic sources.

- The available germplasm needs to be characterized for diverse agro-morphological traits and be conserved on top priority through *ex situ* and *in situ* methods. Germplasm possessing specific desirable traits especially for biotic and abiotic stresses, wider adaptability, climate resilience and nutritional quality traits needs to be identified for use in breeding better varieties.
- Crop descriptor lists which are not available for many of these crops need to be prepared on priority basis. Crop catalogues on the available germplasm should be brought out soonest possible so as to make the information available to the interested researchers. Evaluation efforts should be concentrated on traits of vital importance for evaluation.

Crop Improvement

- Greater emphasis is needed to develop suitable plant-types possessing earliness, photo-insensitivity, high harvest-index, shattering resistance and determinate, bushy growth habit in underutilized grain legumes. Early, bold grained, photo-insensitive and lodging and shattering resistant varieties need to be developed

in pseudocereals. The short duration varieties need to be developed to fit well in the existing cropping systems and to be grown successfully in non-conventional seasons and areas

- Intensive selection and breeding efforts on eco-regional basis need to be made to develop varieties possessing resistance to drought, frost, water logging, diseases, pests and nematodes. Sources of resistance genes should be identified and the resistance be incorporated in high yielding adapted cultivars. Use of related wild and weedy species which possess resistance genes could be important for such purposes.
- The possibilities of exploitation of specific crops for alternate uses need to be explored, for example, winged bean for oil or tubers; chenopods and faba bean for vegetable and for animal feed; amaranth, chenopod, rice bean and hyacinth bean for fodder and buckwheat for vegetables and also many of these crops for use in medicine. Suitable varieties should be developed for such specific purposes in order to have their economic cultivation.
- Basic studies which hitherto remained largely ignored in underutilized grain legumes and pseudocereals require urgent attention. The genetics of plant, fruit and seed characters need to be studied. The causes

of flower and fruit drop also need to be investigated so as to take up remedial measures.

- Timely availability of good quality seed to the farmers at reasonably low price deserves special attention. Sincere research efforts are required to study all aspects of seed production in specific crops. Seed production network involving central and state government agencies and the Research Institutes and State Agricultural Universities needs to be established for production of breeder, foundation and certified seeds.

Production Technology

- In order to integrate the underutilized grain legumes and pseudocereals into the existing agricultural systems, efficient agronomic management is required. Well planned experimentation is needed to determine the package of cultivation practices relating to time and method of sowing, seed rate, plant density and arrangement, irrigation, fertilization and harvesting in different crops.
- Post-harvest technology for each of these crops also needs to be standardised for proper storage of ripe seeds, tubers and immature pods. Systematic research should be carried out on aspects such as chemical treatment of seeds, temperature and humidity control for dry storage, determining optimum moisture content for stored seed and conditioning of stored seed before planting and food purposes.
- Appropriate plant protection measures are required to be developed crop-wise for controlling the diseases, insect-pests and nematodes and also the bird damage. Chemical protectants along with recommended doses need to be specifically determined. Introduction of predators and parasites feeding on harmful insects deserve consideration.

Product Development and Marketing

- In-depth studies on nutritional quality, nutraceutical properties and antinutritional factors need to be given greater thrust. Greater focus needs to be given to processing, value addition, product development and marketing.
- Urgent attention is required for intensive research on the processing and testing of various plant parts for different uses such as ripe seeds for use as food, edible oil and animal feed; plant and leaves for

vegetable and fodder purposes; tubers for food and feed; leaves for protein extraction; ripe seed hull and seed in combination with other on grains to make different food products with improved nutritional value, devising methods of protein extraction from leaves, removing undesirable smell and flavours and easy cooking also require specific consideration.

- Appropriate methods of marketing the products of these crops need to be developed as to ensure adequate production for commercial processing through contract farming among small farmers. In-depth studies are required to estimate the resources required for intensive and extensive production at different locations and the amount of raw product required for efficient manufacture of various products. Any plan designed to increase cultivation of a given crop must be developed concurrently with a program to develop the market for it.

Technology Transfer

- In order to popularise the underutilized grain legumes and pseudocereals, demonstrations and tests are to be organised at no risk to the farmers. Front-line demonstrations should be organised by the Research Institutes/Centres and the State Agricultural Universities (SAUs) at farmers' fields. Adaptive and minikit trials should be conducted by the respective Departments of Agriculture in different States.
- Public awareness of the nutritional and other benefits as well as the profits from cultivation of these crops need to be created through exhibitions and farmers' fairs organised by the Research Institutions and the Departments of Agriculture in the States.
- Suitable publications such as books, brochures, research journals and newsletters on these crops may be brought out for faster dissemination and exchange of information. Preparation of production guides and recipes booklets and educational films with sound tracks in different languages deserve consideration for generating public awareness about the utility and cultivation methods of these crops among the masses.
- Training programmes need to be organised at regular intervals at the level of scientists, extension workers and the farmers in order to appraise them with the latest technology developments relating to particular crops.

Collaboration and Linkages

- Close linkage needs to be established among producers, traders, processors, consumers and other formal and informal sectors in order to develop effective value chain to promote the use of these potential underutilized crops.
- For quality analysis work and for processing, value addition and product development, strong collaboration needs to be developed with the Department of Food Processing/Home Science Department of State Agricultural Universities (SAUs) and concerned institutes and well established laboratories in the country.
- Earlier, there was a strong collaboration of the Indian Program on Underutilized Crops with International Centre of Underutilized Crops (ICUC) at Southampton (UK). Similarly, a close collaboration with the International Centre on Crops for Future (CFF) based in Kuala Lumpur, Malaysia needs to be established.

Policies

- Enabling policies for mainstreaming of use of underutilized crops in food systems needs be given priority attention. Their increased use in mid-day meals of school children needs to be promoted in view of their high nutritional value.
- Appropriate subsidies may be given to the farmers to encourage them for growing the underutilized crops. The cost of quality seed needs to be subsidised and procurement of their produce needs to be assured in the initial stages.
- Local political and administrative support should be developed to encourage cultivation of new crops or expansion of a known but neglected crop. Administrative authorities need to be convinced through proper dialogue about the cultivation of these crops. Farmers should be encouraged for growing the underutilized crops by providing them incentives and subsidies.
- Non-Government organisations (NGO's) may be involved at appropriate levels with a view to give required thrusts to the research and development programmes on underutilized crops.
- Adequate financial and human resource needs to be provided to strengthen the research and development programs on underutilized crops.

References

- Arora RK and Bhag Mal (1991) Genetic resources of forage and food legumes for temperate and cold arid regions of India. In: A Smith and L Robertson (eds) *Legume Genetic Resources for Semi-arid Temperate Environments*. ICARDA, Aleppo, Syria, pp 65-78.
- Bhag Mal and RS Paroda (1988) New potential plants for wasteland development. In: Panjab Singh and RS Pathak (eds) *Rangeland Resources and Management* IGFRI, Jhansi, pp 1-191.
- Bhag Mal (1990) Underutilized plants: A treasurehouse unexplored. *Indian Fmg.* **40**(7): 19-24.
- Bhag Mal and V Joshi (1991) Acquisition, utilization and conservation of genetic resources of underexploited crops. In: *Golden Jubilee Symposium on Genetic Research and Education – Current Trends and the Next Fifty Years*. Indian Society of Genetics and Plant Breeding, New Delhi, Feb. 12-15 (Abstract, Vol. I).
- Bhag Mal, RS Paroda and V Joshi (1992) Status of research on underutilized crops in India. First International Crop Science Congress, Ames, Iowa, USA, (Abstract).
- Bhag Mal (1993) Genetic resources of underexploited plants. In: RS Rana, B Singh, U Srivastava, PN Mathur, M Rai, BM Singh, S Kochhar and IS Bisht (eds) *Exploration, Evaluation and Conservation of Plant Genetic Resources*. NBPGR, New Delhi, India, pp. 158-164.
- Bhag Mal (1994) *Underutilized Grain Legumes and Pseudocereals – Their Potentials in Asia*. RAPA Publications, 162 p.
- Paroda RS (1988) The need for life support species: An Indian perspective. In: RS Paroda, P Kapoor, RK Arora and Bhag Mal (eds) *Life Support Plant Species – Diversity and Conservation*. NBPGR, New Delhi, 190 p.
- Paroda RS and Bhag Mal (1993) Developing a National Programme for Research on Underutilized Crops in India. Proc. First International Crop Science Congress, Ames, Iowa, USA. 14-22 July, 1992.
- Paroda RS, A Agrawal, S Archak, PN Mathur, Bhag Mal, UC Srivastava, , RK Tyagi, and JC Rana (2022) *Proceedings and Recommendations of the National Consultation on 'Plant-based Local Food Systems for Health and Nutrition'*, October 22, 2021. Indian Society of Plant Genetic Resources, New Delhi, India, x+50 p.
- Raiger (2022) Varieties of underutilized crops released during 1982-2021. *Personal Communication*.
- Walsh J (1991) *Preserving the Options: Food Production and Sustainability*, CGIAR Issues in Agriculture No. 2, Consultative Group on International Agricultural Research Washington, DC.
- Williams JT (1993) *Underutilized Crops: Pulses and Vegetables*. Chapman & Hall, London, 247 p.

Use of Crop Wild Relatives (CWRs) of Wheat in Disease Resistance Breeding

Niranjana M¹, Saharan MS², Jha SK¹, Niharika Mallick¹, Raghunandan K¹ and Vinod^{1*}

¹Division of Genetics, ICAR-Indian Agricultural Research Institute, Pusa Campus, New Delhi-110012, India

²Division of Plant Pathology, ICAR-Indian Agricultural Research Institute, Pusa Campus, New Delhi-110012, India

Wheat (*Triticum aestivum* L.) is one of the three major cereal crops contributing 20% calories to the world population. The most serious constraints to wheat production are biotic stresses like rusts, blights, powdery mildew, bunts and smuts. Genetic or host plant resistance is the best way to control these diseases than use of chemical pesticides. Continuous search for novel genes is indispensable to counter the dynamic and rapidly evolving pathogen population. In case of some diseases like spot blotch, Karnal Bunt and Fusarium head blight development of resistant cultivars is not an easy task as the resistance to them found in the germplasm is not satisfactory and none of the commercial cultivars showed resistance. Hence, underutilized crop wild relatives prove to be valuable resource in the search for new disease resistance genes to combat wheat diseases.

Key Words: Fusarium head blight, Karnal bunt, Resistance genes

Introduction

Cereals are the major source of calories for the global human population. Wheat (*Triticum aestivum* L.) is one of the three major cereal crops contributing 20% calories to the world population. The most serious constraints to wheat production are biotic stresses like rusts, blights, powdery mildew, bunts and smuts. Genetic or host plant resistance is the best way to control these diseases than use of chemical pesticides. Through coordinated and concerted research efforts in development and release of resistant varieties, a check has been kept on the management of various diseases and insect pests over past four decades in the country and no epidemics of these occurred. However, new pathogens are always posing challenges and development of new and more virulent pathotypes result in breaking of resistance of popular wheat varieties. The expected onslaught of climate change is also a worrisome aspect and breeding strategies have to be reoriented to mitigate the biotic stresses. Leaf rust caused by *Puccinia triticina* and yellow rusts caused by *P. striiformis* f. sp. *tritici* are serious biotic stresses in the major wheat growing zones like North western plain zone, Northern hill zone and North Eastern plain zone. Stem rust caused by *P. graminis* f. sp. *tritici* is prevalent in hotter climates of Peninsular and Southern India. On the other hand, spot blotch caused by *Bipolaris sorokiniana* has also emerged as one of the most important diseases limiting wheat production

in warmer and humid regions of the world. Another important disease Karnal bunt (KB) caused by *Tilletia indica* has gained importance over the years in NWPZ as most of the varieties recommended for the zone are susceptible to KB. Karnal bunt is an impediment in export of wheat as many importing countries are having zero tolerance to it. Fusarium head blight (FHB) or head scab of wheat caused by *Fusarium graminearum* has also emerged as an important wheat disease worldwide in 21st century. Presently, FHB is a minor disease in India but can cause significant yield loss if rain occurs during mid anthesis.

Crop Wild relatives as Sources of Genes for Disease Resistance

Resistance genes present in current varieties can become obsolete in near future with the emergence of virulence against them. For instance, most of the Indian cultivars have the *Lr* genes *Lr1*, *Lr3*, *Lr9*, *Lr10*, *Lr13*, *Lr14a*, *Lr17*, *Lr19*, *Lr23*, *Lr24*, *Lr26*, *Lr28* and *Lr34*. Out of this *Lr1*, *Lr3*, *Lr10*, *Lr13*, *Lr14a*, *Lr17* and *Lr23* are native to cultivated wheat and have become ineffective due to evolution of new virulence in the pathogen population. Virulence has evolved even against some of the alien genes such as *Lr9*, *Lr19*, *Lr26* and *Lr28* (Niranjana *et al.*, 2017). *Yr* genes in Indian cultivars are *YrA*, *Yr2*, *Yr2KS*, *Yr2* (SKA), *Yr9*, *Yr18*, *Yr27*. Out of these break down of resistance is reported in *Yr9* (Tomar *et al.*, 2014). This suggests that continuous

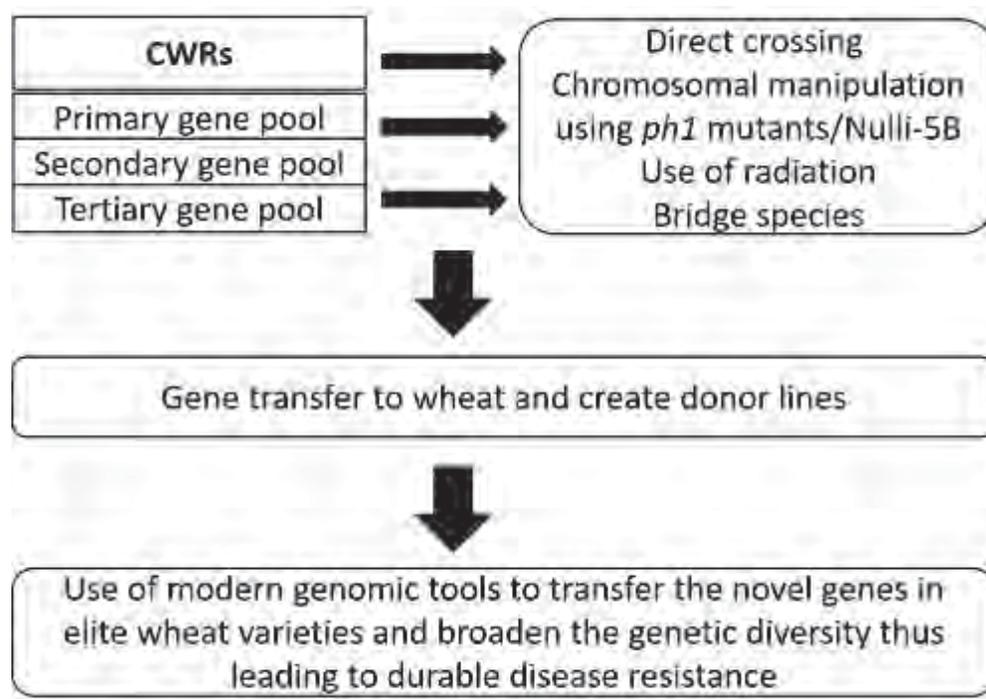
*Author for Correspondence: Email-vinod.genetics@gmail.com

search for novel genes is indispensable to counter the dynamic and rapidly evolving pathogen population. In case of some diseases like spot blotch, Karnal Bunt and FHB development of resistant cultivars is not an easy task as the resistance to them found in the germplasm is not satisfactory and none of the commercial cultivars showed resistance. Hence underutilized CWRs prove to be valuable resource in the search for new disease resistance genes to combat these diseases.

CWRs of wheat are known to possess high level of diversity for both biotic and abiotic stress tolerance and are excellent source for broadening the genetic base for resistance to rusts as well as emerging diseases like Karnal bunt, spot blotch and head scab. Some of the *Lr*, *Sr* and *Yr* resistance genes have been derived from related and distantly related species like *T. monococcum*, *T. dicoccoides*, *T. turgidum*, *T. spelta*, *T. timopheevi* and genera including *Aegilops*, *Agropyron*, *S. cereale*, *Thinopyrum* and *Elymus* (Naik et al., 2015). Resistance to Karnal bunt was identified in *S. cereale*, *Triticale*, several accessions of *Aegilops biuncialis*, *Ae. columaris*, *Ae. crassa*, *Ae. juvenalis*, *Ae. ovata*, *Ae. speltoides*, *T. uratu* and *Ae. squarrosa* (Warham 2009). Amphiploids synthesized from *T. durum* with *T. monococcum*, *T. boeoticum* and *Ae. squarrosa* crosses were screened for KB resistance (Multani et al., 1988). Several markers and quantitative trait loci (QTLs) for spot blotch resistance

have been mapped in wheat (Gupta et al., 2018) but none from CWRs. Similarly, in case of FHB limited sources of resistance known to *Fusarium* spp. causing head scab.

CWRs of wheat can be broadly classified as primary, secondary and tertiary. The primary gene pool of wheat comprises species having homologous genome as in wheat (A, B, D). This includes tetraploid species like *T. dicoccoides*, *T. dicoccum*, *T. durum*, *T. polonicum*; hexaploid species *T. aestivum*, *T. spelta*, *T. compactum*, *T. sphaerococcum* and diploid species with A and D genome like *T. uratu*, *T. monococcum*, *T. boeoticum* and *Ae. squarrosa*. Since their chromosomes are homologous to the cultivated types, they will easily undergo homologous recombination. The primary gene pool has been best utilized for crop improvement as it is relatively easy to transfer genes among them. The secondary gene pool comprises of species which has at least one homologous genome in common with the cultivated species. This includes species like *Ae. speltoides* (SS), *T. timopheevii* and *T. militinae* (AAGG), *Ae. cylindrica* (CCDD). Species included in the tertiary gene pool are distantly related species whose genome is unique from A, B or D genome. This includes species of *Secale* (RR), *Thinopyrum* (EE), *Hordeum* (HH). Despite being a hexaploid, bread wheat acts as an amphidiploid due to the presence of *Ph1* (*pairing homeologous*) locus



which prevents homeologous pairing between A, B, D genomes and promote homologous pairing. For transfer of genes from tertiary gene pool homeologous pairing between wheat and alien genomes should be induced by the inactivation of *Ph1* gene. A single *Ph1* deletion mutant developed in hexaploid wheat cultivar Chinese Spring namely CS *ph1b* has been utilized for this purpose. Since *Ph1* locus is located on 5B chromosome nullisomics lacking 5B (Nulli-5B) chromosome could be utilized. Special strategies such as irradiation, bridge species and gametocidal chromosomes are also used. The availability of such natural diversity provides an excellent starting material for the betterment of wheat genetic base by conventional breeding techniques and chromosome engineering.

Future Prospective and Action Points

1. Development of wheat varieties from diverse parents and exploit genes from underutilized wild relatives of wheat in the breeding cycle which will broaden the genetic base of wheat varieties and prevent genetic erosion.
2. Pre-breeding efforts should be strengthened for the initial transfer of genes from CWRs as it is the trickiest part. Since sterility issues are evident only few progenies could be derived and cytogenetic stability of the introgression lines should be ensured before utilizing them in breeding cycle.
3. Availability of high-quality reference sequence of wheat opens wide vistas in utilization of genes from CWRs. Fine mapping of the genes derived from wild relatives will lead to development of linked markers or better gene-based markers.
4. Once we develop a molecular map and identify the linked markers or gene-based markers they could be utilized in molecular breeding and ensure development of CWR derived elite varieties in a short span of time.

References

- Gupta PK, Chand R, Vasistha NK, Pandey SP, Kumar U, Mishra VK, Joshi AK. (2018) Spot blotch disease of wheat: the current status of research on genetics and breeding. *Plant Path.* **67**: 508–531.
- Multani DS, HS Dhaliwal, Paramjit Singh and KS Gill (1988) Synthetic Amphiploids of Wheat as a Source of Resistance to Karnal Bunt (*Neovossia indica*). *Plant Breeding* **101**: 122–125.
- Naik BK, Vinod, Sharma JB, Sivasamy M, Prabhu KV, Tomar RS, Tomar SMS (2015) Molecular mapping and validation of the microsatellite markers linked to the *Secale cereale*-derived leaf rust resistance gene *Lr45* in wheat. *Mol Breed.* **35**: 61.
- Niranjana M, Vinod, Sharma JB, Mallick N, Tomar SMS, Jha SK (2017) Cytogenetic analysis and mapping of leaf rust resistance in *Aegilops speltoides* Tausch derived bread wheat line Selection2427 carrying putative gametocidal gene(s). *Genome* **60(12)**:1076–1085.
- Tomar SMS, Singh SK, Sivasamy M, Vinod. (2014) Wheat rusts in India: Resistance breeding and gene deployment - A review. *Indian J. Genet.* **74(2)**: 129–156.
- Warham EJ (2009) Screening for Karnal bunt (*Tilletia indica*) resistance in wheat, triticale, rye, and barley. *Canadian J. Plant Path.* 57–60.

Plant Germplasm Registration System in ICAR – Achievements

Anjali Kak Koul* and Veena Gupta

Division of Germplasm Conservation, ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi–110012, India

The Plant Germplasm Registration Committee (PGRC) was constituted by the Indian Council of Agricultural Research in 1996 to provide recognition to researchers involved in identifying, describing and documenting potentially valuable genetic materials and to incorporate these into the public domain for their efficient use by the breeders and researchers. In the last 25 years a total of 48 meetings of PGRC have been held and 1,948 crop genetic materials have been registered belonging to 249 crop species. Among these, highest number of germplasm are registered for resistance against biotic stress (601) followed by quality and nutritional traits (583). In major food crops like cereals, oilseeds and grain legumes, maximum registration is for biotic stresses while in ornamentals, fruit crop, medicinal and aromatic crops, tea and coffee, maximum germplasm is registered for quality traits.

Introduction

Plant genetic diversity is of fundamental importance in the continuity of a species as it provides the necessary adaptation to the prevailing biotic and abiotic environmental conditions, and enables change in the genetic composition to cope with changes in the environment. Climate change leads to the concurrence of a number of *abiotic and biotic stresses*, thus affecting agricultural productivity. Extensive crop improvement programmes are needed to address these challenges. Materials with high potential in breeding for a changing climate need to be assembled in genebank collections. Not only crop wild relatives (CWR), but also useful materials resulting from various research projects, where genes from wild or exotic sources are integrated into adapted material, which have resistance to biotic and abiotic stresses need to be identified and assembled for use in breeding programmes. Accessibility of these unique genetic resources for use by the breeders depends upon the availability of information and material in public domain and equally important is the recognition of the breeder who has developed/identified the unique germplasm. Unlike the breeders/developers of released cultivars, scientists associated with the development of unique trait specific or potentially valuable germplasm and genetic stocks had no mechanism for recognition of their work before 1996. With an important rationale to recognize the efforts of researchers for developing the potentially valuable germplasm and to publically

describe and document new and useful genetic materials and to incorporate these into the public domain for their efficient use by the breeders and researchers, the Indian Council of Agricultural Research (ICAR) constituted a Plant Germplasm Registration Committee (PGRC) under chairmanship of Deputy Director General (Crop Science), to register such unique germplasm. The responsibility of registration and conservation of trait-specific plant germplasm was entrusted to NBPGR by ICAR. The PGRC meets quarterly (normally last month of the quarter) with the concurrence of the Chairman, for consideration of applications and related matters.

In 1996, detailed guidelines and proforma for registration was formalized to facilitate the process of germplasm registration. Taking into consideration the upcoming developments in management of plant genetic resources (PGR) and related policies at global and national level, the guidelines and proforma continue to evolve and accommodate changes suggested by the experts. The guidelines were first published in 2005 and distributed at various fora to give wider publicity of the activity. The major revisions were incorporated in 2014 with respect to submission of application for registration, data requirement and deposition of seed/genetic material in compliance to provisions required as per the changing scenario. Again in 2021, the guidelines were revised to incorporate changes in the data requirement with respect to registration of vegetatively propagated horticultural crops.

*Author for Correspondence: Email-Anjali.koul@icar.gov.in

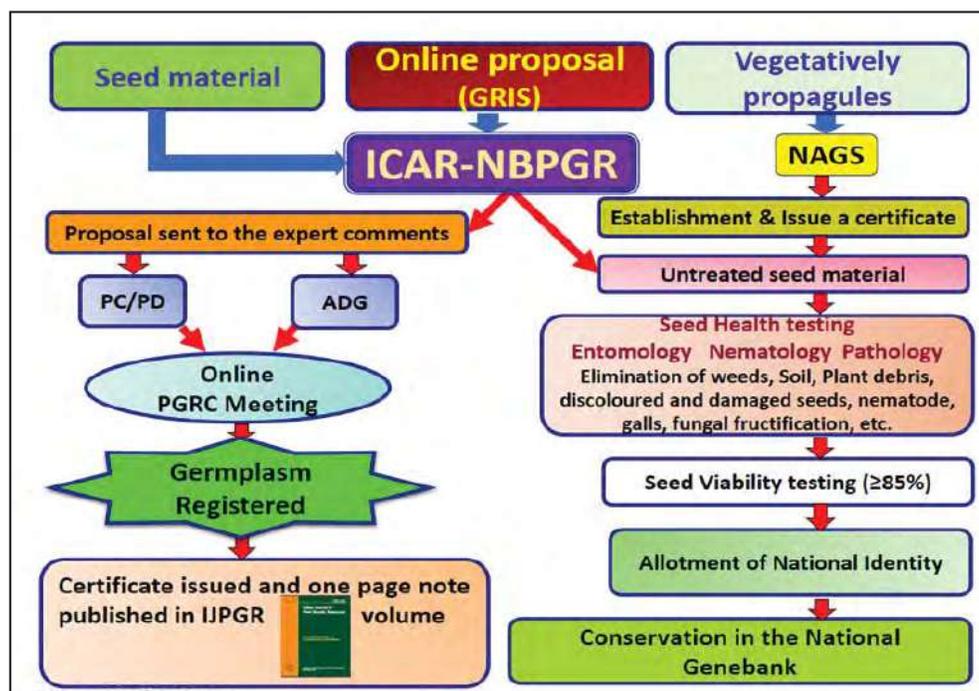
Germplasm/genetic stock/elite material of field, horticultural and other economic crops, including argo-forestry species, spices, medicinal and aromatic plants, ornamental plants, which is unique, uniform, stable and has potential attributes of academic, scientific or commercial value are eligible for registered. Exotic germplasm (imported) can be registered for a trait other than those published or registered. Similarly, selections made from exotic germplasm can also be registered. Selection for unique traits from landraces (other than the landrace is known for) may be considered for registration. The registration is subject to fulfilling of the data requirements as per the guidelines. The guidelines for registration of germplasm are available at NBPGR website (<http://www.nbpgr.ernet.in:8080/registration/Guidelines.aspx>). The registration remains valid for 18 years for trees and vines and 15 years for other plant species, after which the registered germplasm would be national sovereign property. A registration can be repealed by the PGRC in case of false claim(s).

Germplasm Registration Information System (GRIS)

Earlier applications were submitted as hard copies. With the advent of an online system in 2018 the entire process of filling application has become simple and

fast through the Germplasm Registration Information System (GRIS) (<http://www.nbpgr.ernet.in:8080/registration/>). The progress of each application can be tracked through a personal dashboard. Trait-specific germplasm is also searchable *via* the GRIS. Genetic stocks have been provided with a QR Code to access detailed information related to these lines. The online system, thus, provides genebank managers, breeders and plant researchers with hands-on tool for management of germplasm registration process, and to policy makers with a reliable source of information.

Registered germplasm is conserved either in National Genebank (in form of seeds) or at designated crop-based National Active Germplasm Site (NAGS) (especially vegetatively propagated crop germplasm) depending upon the type of the propagule. National Genebank at NBPGR is responsible for long-term conservation of orthodox seed material accompanied by passport and genebank-related information. Untreated seed material received as prescribed seed quantity (minimum 2,000 seeds for self-pollinated and 4,000 seeds for cross-pollinated crops) for registration are subjected to seed health testing for ensuring the germplasm free from quarantine pests (NBPGR, 2005, Guidelines for Registration of Plant Germplasm; <http://www.nbpgr>.



Flow Chart of Plant Germplasm Registration Activity: Conservation, Multiplication and Distribution of Registered Germplasm

ernet.in/download/registration.pdf.). Unique National Identity or IC Number (Indigenous Collection) are assigned, subject to qualifying the international genebank standards (FAO, 2011). After testing of germination following International Seed Testing Association standards (ISTA, 1993), moisture content of the seeds is equilibrated to 5–7% in the walk-in-drying room (15 °C and 15% RH), seeds are packed in tri-layered aluminum foil packets using vacuum sealer, labelled and stored at –18 °C in National Genebank. However, the standards are relaxed for seeds of difficult species, e.g. wild, rare and endangered species on case-to-case basis. Recalcitrant seed and vegetatively propagated crop germplasm are deposited in the field genebank of the concerned crop-based NAGS (<http://www.nbpgr.ernet.in/download/registration.pdf>) for conservation and a certificate to this effect has to be obtained by the developer before the material is proposed for registration. It is obligatory on the part of developer and developing organization to maintain the stock of registered germplasm to share the germplasm in National Agricultural Research System (NARS). Active or working collection(s) may also be maintained by the NAGS with a responsibility for its multiplication and distribution to *bona fide* users in the NARS.

Publication of Registered Germplasm

All germplasm approved for registration is officially communicated to the applicants along with Registration Number. A certificate to this effect is also issued to the applicant, which can be downloaded from GRIS. A brief description of not more than one page (according to instructions given in GRIS) is published in the ensuing issue of Indian Journal of Plant Genetic Resources

– official publication of the Indian Society of Plant Genetic Resources, NBPGR, New Delhi-110012. The compiled information is also published in the form of information bulletins (NBPGR 2006, Kak *et al.*, 2009, Kak and Tyagi 2010, 2015, Kak and Gupta 2017, 2020), to disseminate information among scientists/users. An interactive database of registered germplasm is also available at NBPGR website (<http://www.nbpgr.ernet.in:8080/registration/InventoryofGermplasm.aspx>).

Achievements

Starting in 1996 and continuing for 25 years, total 48 meetings of Plant Germplasm Registration Committee have been held and 1948 crop genetic materials have been registered as of 31st March 2022 belonging to 249 crop species. Considering total number of proposals submitted (4,300) and number of germplasm registered (1,948), 61% proposals are developed/ identified by ICAR crop-based institutes, followed by 23% by SAUs and 6% proposals belonged to ICAR-NBPGR alone. It is to be noted that a very few germplasm accessions were registered by private sector (1%) (Fig 1). The vigorous efforts made to publicize the information on registration during important scientific meetings and increased frequency of PGRC meetings, the rate of qualifying germplasm for registration also increased. Maximum number of the germplasm has been registered for resistance against biotic stress (601) followed by quality and nutritional traits (583). In major food crops like cereals, oilseeds and grain legumes maximum germplasm was registered for biotic stresses. In ornamentals, fruit crop, medicinal and aromatic crops and crops like tea and coffee, maximum number of germplasm was registered, generally, for quality traits.

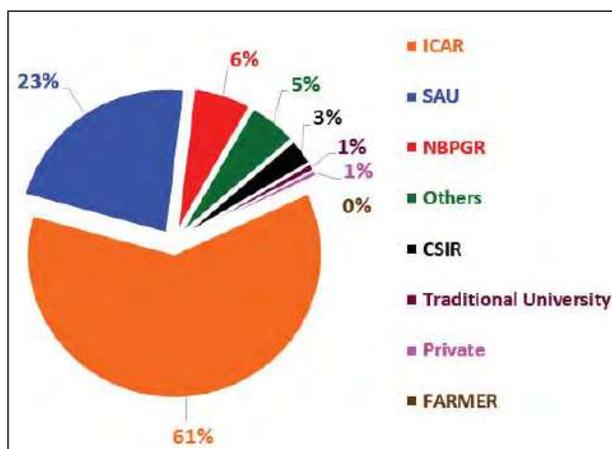


Fig. 1. Institute-wise germplasm registered

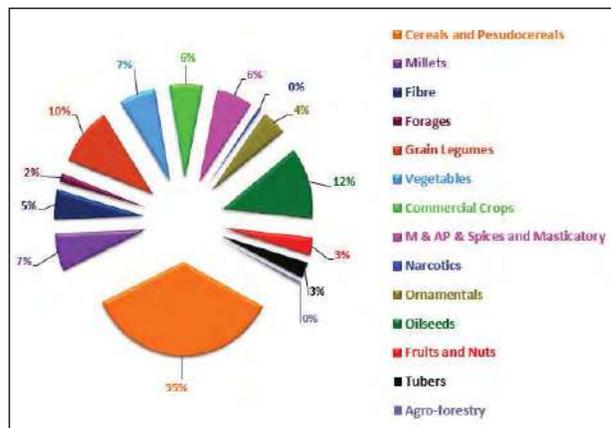


Fig. 2. Crop group-wise representation (%) trait-specific germplasm registered (1996-2022)

NBPGR regularly organize the Germplasm Field Days for displaying the germplasm variability of various crops in the field to select genotypes of interest by the breeders. The registered germplasm accessions of different crops are displayed at suitable locations including in the field genebanks of regional stations of NBPGR located strategically in various agro-climatic regions. Concerned crop breeders select the material and the same is distributed to the breeders to meet their requirement under material transfer agreement (<http://www.nbpgr.ernet.in/download/SMTA.pdf>). The users can indent the germplasm including registered ones of their interest available in NBPGR (<http://www.nbpgr.ernet.in/download.htm>).

In conclusion plant germplasm registration serves the basic intent of registering, conserving, distributing the potentially valuable germplasm and documenting the information to be made available in public domain for further utilization by breeders to develop climate-resilient varieties in crops as genetic variability has been playing and will continue to play significant role in mitigating the challenges due to climate change in future.

References

- FAO (2011) Draft revised genebank standards for conservation of orthodox seeds, *Commission on Genetic Resources for Food and Agriculture Rome*, 18–22 July 2011.
- <http://www.nbpgr.ernet.in/download.htm>
- <http://www.nbpgr.ernet.in/download/SMTA.pdf>
- <http://www.nbpgr.ernet.in:8080/registration/>
- <http://www.nbpgr.ernet.in:8080/registration/Help.aspx>
- <http://www.nbpgr.ernet.in:8080/registration/Guidelines.aspx>
- <http://www.nbpgr.ernet.in:8080/registration/InventoryofGermplasm.aspx>
- ISTA (1993) International rules for seed testing. *Seed Sci. Technol.* 21: 288.
- Kak A, K Srinivasan and SK Sharma (2009) *Plant Germplasm Registration (2005–2008)*. National Bureau of Plant Genetic Resources (NBPGR), Indian Council of Agricultural Research (ICAR), New Delhi, 75 p.
- Kak A and RK Tyagi (2010) *Inventory of Registered Crop Germplasm (2009-2010)*. NBPGR, ICAR, New Delhi.
- Kak A and RK Tyagi (2015) *Inventory of Registered Crop Germplasm (2010-2014)*. NBPGR, ICAR, New Delhi, 101 p.
- Kak A and Veena Gupta (2017) *Inventory of Registered Crop Germplasm (2015-2017)*. National Bureau of Plant Genetic Resources, Pusa Campus, Indian Council of Agricultural Research (ICAR), New Delhi, 71 p.
- Kak A and Veena Gupta (2020) *Inventory of Registered Crop Germplasm (2018-2020)*. NBPGR, ICAR, New Delhi, 96p.
- NBPGR (2005) *Guidelines for Registration of Plant Germplasm*, NBPGR, Pusa Campus, New Delhi.
- NBPGR (2006) *Plant Germplasm Registration*. NBPGR, ICAR, New Delhi.

Role of Genebanking in the Era of Genomics

Veena Gupta and Sherry Rachel Jacob*

Division of Germplasm Conservation, ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

“Cherish variations, for without it life will perish” – Sir Otto Frankel

The global system for conservation of plant genetic resources (PGR) is a pan-continental network integrating more than 2000 national and international genebanks out of which 130 genebanks hold more than 10,000 accessions and eight have more than 100,000 accessions. As per the PGR Genesys platform, accessions conserved in *ex situ* genebanks comprise more than 4 million maintained by over 450 institutes across the globe. Accumulation of huge quantity of germplasm accessions of unknown value and genetic composition within genebanks made its inclusion in crop breeding, economically unmanageable. This article provides some of the new age tools and methodologies being applied for more effective management of genebanks.

Key Words: Digital Sequence Information, Genome-wide association studies, Genomic selection, Machine learning, Pan-genome studies

Introduction

Plant genetic resources (PGR) are the forerunners of crop evolution and they occupy the most diverse ecological habitats on this planet. They have defined and determined the survival and adaptation of all living beings, including mankind and has generously provided for their sustenance. Subsequent crop improvement efforts in selected crops led to preferential cultivation of improved varieties in a very limited number of species. The narrow genetic base in elite varieties resulted in significant diversity loss and the non-preferred species faced extinction. Plant biologists soon realized the impending threat and put forth the need to address ‘diversity’ in a more technical perspective. Starting from 1970s, there were concerted efforts to promote an international network of genetic resources centres to further collection, conservation, documentation, evaluation and use of germplasm (Frankel and Bennett, 1970). Today, the global system for conservation of PGR is a pan-continental network integrating more than 2000 national and international genebanks out of which 130 genebanks hold more than 10,000 accessions and 8 have more than 100,000 accessions. The PGR Genesys platform which hosts compiled data on accessions conserved in *ex situ* genebanks, reports a total of more

than 4 million accessions maintained by over 450 institutes across the globe (<https://www.croptrust.org/>). Over the decades, the accumulation of huge quantity of germplasm accessions of unknown value and genetic composition within genebanks made its inclusion in crop breeding, economically unmanageable. The primary strategy adopted by Genebank researchers to overcome this hindrance was to undertake redundancy assessment and subsequently, identify a priority set of germplasm accessions that can effectively capture the allelic diversity, to the extent required. In the case of Crop Wild Relatives (CWRs) and landraces, the priority was to minimize barriers in mobilizing genetic variation into elite background. The tools and techniques employed for meeting this objective have evolved along with the advances in science and today, PGR management and crop improvement are making parallel progress by harnessing the benefits of cutting edge technologies.

Genomic Tools for Genetic Dissection of Complex Traits

The two major forward genetics approaches used for understanding the genetic basis of traits in germplasm collections are linkage mapping or QTL mapping and association mapping or linkage disequilibrium (LD) mapping (Cadic *et al.*, 2013). In large *ex situ* gene bank

*Author for Correspondence: Email-sherry.jacob@icar.gov.in

collections, an ideal approach for genetic dissection is the association analysis *viz.*, genome-wide association studies (GWAS), which is based on linkage disequilibrium (LD) or the non-random association of alleles at different loci (Kumar *et al.*, 2022). The fundamental principle of GWAS is to analyse and validate marker-phenotype association within a very large, diverse set of individuals, as in the case of germplasm collections. Such collections have preserved historical recombinations and also account for larger number of alleles, thus enabling high-resolution mapping (Alqudah *et al.*, 2020). GWAS is being used extensively to identify allelic variants associated with multiple traits of adaptive and agronomic importance (Akram *et al.*, 2021).

The concept of Genomic Selection (GS) has also gained prominence, w.r.t to understanding the genetic basis of minor alleles. In GS, the genetic value of a set of accessions (breeding population) is predicted based on Genomic Estimated Breeding Value (GEBV) generated on a Training Population (TP) that is related to BP and is both genotyped and phenotyped. Using this prediction model, the marker effects are estimated across the breeding population, without phenotyping or QTL mapping (Heffner *et al.*, 2009; Calus, 2010). As compared to MAS, GS captures larger amount of genetic variation of the specified trait since it involves high density markers with both minor and major effects and distributed throughout the genome (Desta and Ortiz, 2014; Crossa *et al.*, 2017). The developments in the field of Machine Learning (ML) has enhanced genomic prediction accuracy through use of high precision tools for identifying subsets of SNPs having direct link to candidate genes affecting the trait of interest (Li *et al.*, 2018). The GS approach has been attempted in the ex situ genebank collection of CIMMYT, Mexico, for genomic prediction within Mexican and Iranian landrace accessions. The prediction accuracy implied potential scope of this technique in rapid introgression of exotic material including CWRs, into elite background, for effective germplasm enhancement (Crossa *et al.*, 2016).

Moving Ahead of Genomes

The advances in various sequencing technologies have led to generation of high quality reference genomes in several major crops and even in the wild relatives of rice, wheat, barley, soybean and tomato. But, in the context of PGR, this whole genome sequence assemblies had

the serious shortcoming of not addressing Structural Variations (SV) like Copy Number variations (CNVs) which are a major source of genetic variation during domestication and further diversification of crop taxa (Lye and Purugganan, 2019) that can have a pronounced phenotypic impact (Alkan *et al.*, 2011, Mahmoud *et al.*, 2019). The emerging area of pan-genome analysis, effectively addresses this crucial requirement, thus holding significant implications for breakthroughs in genebank germplasm utilization. In pan-genome approach, several reference genome sequences are used as template to map the sequencing data, whereby all possible natural variations get detected, including those lost during domestication and crop improvement (Bohra *et al.*, 2022).

The pangenome work on cultivated Asian rice conserved at IRRRI Genebank could successfully unravel the full repertoire of genetic diversity that exists in the largest ex situ rice Genebank collection (Zhou *et al.*, 2020). Similar work has been reported in hexaploid bread wheat (Montenegro *et al.*, 2017). Recently, the scope of pan-genome studies has been further expanded by including wild relatives, landraces and cultivars in multiple crops (Liu Y *et al.*, 2020). As science progresses in leaps and bounds, the idea of pan-genome has further moved forward into the concept of ‘super-pangenome’, which involves a genus-level approach, by assembling multiple species specific reference genomes. Shang *et al.*, 2022 reports the development of a *Oryza* super pangenome comprising both cultivated and wild species of Asian and African rice that can identify lineage-specific haplotypes for trait-associated genes and can also provide fundamental understanding on the genetic basis of environmental adaptation and domestication of rice. The super pan-genome of genus *Glycine* has also made a remarkable contribution to PGR utilization by unleashing genetic potential from the perennial gene pool for soybean improvement (Zhuang *et al.*, 2022). This strategy will facilitate in fully reaping the benefits of CWRs conserved in genebanks.

Genebank Documentation at the Genebank-Genomics Interphase

Genomics has propelled PGR management into the arena of ‘big data’ science, where data bases are no longer the supportive components, but rather, forms a highly specialized information inter phase that technically

regulates the access to resources. This Digital Sequence Information (DSI) now calls for defined guidelines and management strategies for ensuring fair benefit sharing without restricting access (Aubrey, 2019). Contracting parties to the Convention on Biological Diversity (CBD) and ITPGRFA are starkly divided in their opinions with regard to the rules for accessing PGR DSI (Scholz *et al.*, 2022) and it is currently challenging the basic fundamentals of PGR governance as envisaged in case of physical germplasm collections. Several solutions are being proposed for enhancing transparency in sequence data access. One example is the compulsory tagging of provenance information with the sequence information that has been proposed by the International Nucleotide Sequence Database Collaboration (INSDC), a core infrastructure for storage of global sequence data (Wilkinson *et al.*, 2016). Such endeavours will prove beneficial only if it is effectively complemented with the complete passport data of national genebanks. Thus, as the debate continues, the onus is on genebanks to ensure effective mechanism for extensive documentation of their sovereign genetic resources and also make in-house provisions for feedback and tracking of germplasm utilization by diverse consumers.

References

- Akram S, Arif MAR and Hameed A (2021) A GBS-based GWAS analysis of adaptability and yield traits in bread wheat (*Triticum aestivum* L.). *J. Appl. Genet.*, **62(1)**: 27-41.
- Alkan C, BP Coe and EE Eichler (2011) Genome structural variation discovery and genotyping. *Nat. Rev. Genet.* **12(5)**: 363-376.
- Alqudah AM, A Sallam, PS Baenziger, A Börner, (2020) GWAS: Fast-forwarding gene identification and characterization in temperate Cereals: lessons from Barley – A review, *J. Adv. Res.*, **22**: 119-135, <https://doi.org/10.1016/j.jare.2019.10.013>.
- Aubry S (2019) The Future of Digital Sequence Information for Plant Genetic Resources for Food and Agriculture, *Frontiers in Plant Sci.* 10, DOI=10.3389/fpls.2019.01046
- Bohra A, B Kilian, S Sivasankar, M Caccamo, C Mba, SR McCouch, RK Varshney (2022) Reap the crop wild relatives for breeding future crops. *Trends in Biotechnology.* **40(4)**: 412-431, ISSN 0167-7799, <https://doi.org/10.1016/j.tibtech.2021.08.009>.
- Cadic E, M Coque, F Vear, B Grezes-Besset, J Pauquet, J Piquemal, Y Lippi, P Blanchard, M Romestant, Pouilly N, Rengel D (2013) Combined linkage and association mapping of flowering time in Sunflower (*Helianthus annuus* L.). *Theor. Appl. Genet.* **126(5)**: 1337-1356. doi: 10.1007/s00122-013-2056-2.
- Calus MP (2010) Genomic breeding value prediction: methods and procedures. *Animal: an international journal of animal bioscience*, **4(2)**:157-64.
- Crossa J, D Jarquín, J Franco, P Pérez-Rodríguez, J Burgueño, C Saint-Pierre, P Vikram, C Sansaloni, C Petrolí, D Akdemir, C Sneller (2016) Genomic prediction of gene bank wheat landraces. *G3 (Bethesda, Md.)* **6(7)**: 1819-1834
- Crossa J, González-Camacho, S Pérez-Elizalde, Y Beyene, S Dreisigacker, R Singh, X Zhang, M Gowda, M Roorkiwal, J Rutkoski, R K. Varshney (2017) Genomic selection in plant breeding: Methods, Models, and Perspectives. *Trends in Plant Sci.* **22(11)**: 961-975
- Destá ZA and R Ortiz (2014) Genomic selection: genome-wide prediction in plant improvement, Genomic selection: genome-wide prediction in plant improvement, *Trends in Plant Sci.* **19(9)**: 592-601, <https://doi.org/10.1016/j.tplants.2014.05.006>.
- Frankel OH, and E Bennett (1970) Genetic resources in plants-their exploration and conservation. (Frankel, O.H. and Bennett, E., eds.), pp. 7–17. Blackwell, Oxford
- Heffner EL, ME Sorrells, and JL Jannink (2009) Genomic selection for crop improvement. *Crop Sci.* **49(1)**: 1-2. <https://doi.org/10.1186/s13059-019-1828-7>
- Kumar S, Jacob SR, MR Rouf, VK Vikas, K Pawan, C Tilak, K Satinder, K Uttam, K Suneel, SSHailendra, S Ravinder, P Sai, SA Mahendru, SA Kumar, K Jyoti, MS Saharan, BS Chander, P Manoj, K Sanjay, S Kuldeep (2022) Indian wheat genomics initiative for harnessing the potential of wheat germplasm resources for breeding disease-resistant, nutrient-dense, and climate-resilient cultivars, *Front Genet (13)* DOI=10.3389/fgene.2022.834366
- Li B, N Zhang, YG Wang, AW George, A Reverter, and Y Li (2018) Genomic prediction of breeding values using a subset of SNPs identified by three machine learning methods. *Front Genet.* **(9)** DOI=10.3389/fgene.2018.00237
- Liu Y, H Du, P Li, Y Shen, H Peng, S Liu, GA Zhou, H Zhang, Z Liu, M Shi, and X Huang (2020) Pan-genome of wild and cultivated soybeans. *Cell.* **182(1)**: 162-176. e13, ISSN 0092-8674, <https://doi.org/10.1016/j.cell.2020.05.023>.
- Lye ZN and Purugganan MD (2019) Copy number variation in domestication. *Trends Plant Sci.* **24(4)**: 352-365.
- Mahmoud M, N Gobet, DI Cruz-Dávalos, N Mounier, C Dessimoz, and FJ Sedlazeck (2019). Structural variant calling: the long and the short of it. *Genom. Biol.* **20(1)**: 1-4
- Montenegro JD, AA Golicz, PE Bayer, B Hurgobin, H Lee, CK Chan, P Visendi, K Lai, J Doležel, J Batley and D Edwards (2017) The pangenome of hexaploid bread wheat. *The Plant J.* **90(5)**: 1007-1013.
- Scholz AH, J Freitag, CH Lyal, R Sara, ML Cepeda, I Cancio, S Sett, AL Hufton, Y Abebaw, K Bansal, and H Benbouza (2022) Multilateral benefit-sharing from digital sequence information will support both science and biodiversity conservation. *Nat Commun.* **13(1)**: 1-5.
- Shang L, X Li, H He, Q Yuan, Y Song, Z Wei, H Lin, M Hu, F Zhao, C Zhang and Y Li (2022) A super pan-genomic landscape of rice. *Cell Res.* **12**: 1-9. <https://doi.org/10.1038/s41422-022-00685-z>

Wilkinson MD and IJ Aalbersberg et al. (2016) The FAIR Guiding Principles for scientific data management and stewardship. *Sci Data*. **3(1)**: 1-9

Zhou Y, D Chebotarov, D Kudrna, V Llaca, S Lee, S Rajasekar, N Mohammed, N Al-Bader, C Sobel-Sorenson, P Parakkal, and LJ Arbelaez (2020) A platinum standard pan-genome resource that represents the population structure of Asian rice. *Sci. Data*. **7(1)**: 113 <https://doi.org/10.1038/s41597-020-0438-2>

Zhuang Y, X Wang, X Li, J Hu, L Fan, JB Landis, SB Cannon, J Grimwood, J Schmutz, SA Jackson, and JJ Doyle (2022) Phylogenomics of the genus *Glycine* sheds light on polyploid evolution and life-strategy transition. *Nat. Plants*. **8(3)**: 233-44. <https://doi.org/10.1038/s41477-022-01102-4>.

Sustainability of *In Vitro* Genebanks and Cryogenebanks

Anuradha Agrawal^{1*}, R Gowthami², Subhash Chander² and Vartika Srivastava²

¹Indian Council of Agricultural Research, KAB-2, Pusa Campus, New Delhi-110012, India

²National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

PGRs are essential components of food, nutrition and livelihood security and in the last few decades significant improvements have been made for better and safe conservation in national and international genebanks. There are several species, which cannot be conserved in conventional seed genebanks and needs alternate strategies. Among different strategies *in vitro* and cryogenebanks are the only available approaches for safe conservation of difficult species for short- to medium-term and for long-term respectively. *In vitro* and cryoconservation is in practice for several years, slow growth conservation and cryoconservation protocols are developed in diverse species and many species are successfully and safely conserved in *in vitro* and cryogenebanks in many countries globally. This paper reviews the priority species and future prospects of *in vitro* and cryogenebanks.

Introduction

Plant genetic resources (PGR) are fundamental for life on earth. Globally plant genetic heritage is at greater erosion due to globalization, industrialization, degradation of natural habitats, climate change, replacement of traditional varieties with modern cultivars and hybrids, agriculture intensification, overexploitation from natural habitats, population increase, land degradation etc (Gowthami *et al.*, 2021). Worldwide 17.01 % of the world's vascular plants (3,42,953) are under varying degrees of threat and in the view of 'sixth mass extinction', there is a global alarm for greater need for immediate efforts for conservation of plant diversity for the sustainable future of the planet. Conservation of PGRs in their natural habitat (*in situ*) is the first and primary approach being followed from the time immemorial, but due to vulnerability to natural hazards, a safety backup conservation approaches is required. Then emerged a concept of *ex situ* conservation (conservation away from the natural habitat) as botanical gardens, herbal gardens, arboreta, sacred groves and gene banks. Approximately 92% of angiosperms (3, 30,000) produce orthodox seeds (desiccation-tolerant), which tolerate drying to low moisture content ($\leq 5\%$) without a significant loss of viability. These seeds can be conserved conventionally in seed genebanks, that remains viable for 25-50 years under refrigerated conditions (5-10 °C) and can survive ranged from 100–200 years in the deep freezer (-20 °C). However, conservation of all the seed species are not possible in seed genebanks and is not suitable for all the seed species. Germplasm collections were initiated

in 1920 mainly for breeding purpose and during 1960s & 1970s, major emphasis was given for conservation of major food crops that produce orthodox seeds and research was biased towards these crops, which eventually led to the establishment of present day, nearly 1,750 seed genebanks globally. However, conservation in seed genebanks is not feasible for many plants/crops, which producing difficult to conserve-species (Fig 1A). These species are normally conserved as living collections in field genebanks, but it requires large area, labour intensive, high maintenance cost, easily prone to natural hazards, pest/pathogen attacks etc. The alternative safe and sustainable approaches for the conservation of these difficult-to- conserve species is *in vitro* conservation (short- to medium term) and cryo-conservation (long-term conservation).

In Vitro and Cryogenebank

In vitro genebank represents the conservation of plant genetic resources in the form of *in vitro* cultures under controlled conditions. The potentiality of *in vitro* techniques for safe conservation of difficult-to- conserve species was emphasized in the early 1970s, later in 1980s, International Board for Plant Genetic Resources (IBPGR) formed a specialists working group to evaluate the critical aspects of *in vitro* plant conservation. During 1987-89, the technical and logistical aspects of establishing and running an *in vitro* active genebank using cassava as a model were assessed. Later in 1997, IBPGR status report on *in vitro* conservation techniques was developed and in 2004 technical guidelines for the management

*Author for Correspondence: Email-Anuradha.Agrawal@icar.gov.in

of field and *in vitro* collections was developed. In 2014, with an objective of conservation of PGR with recognized and appropriate standards based on current and available technological and scientific knowledge, genebank standards for PGRFA maintained *in vitro* conservation was developed. In *in vitro* genebanks, PGRs can be conserved for short- to medium-term period (several months to a few years) under normal growth conditions (standard culture room conditions) or subjected to growth-limiting conditions using one or more slow growth conservation strategies *viz.*, low temperature storage, low light storage, use of minimal growth media, use of osmotica in the medium, reduced oxygen concentration, type of culture tube enclosures and culture vessels, desiccation and encapsulation, induction of storage organs etc. (*in vitro* active genebank) and for long-term period under suspended growth (cryoconserved) in liquid nitrogen (*in vitro* base genebank) (Agrawal *et al.*, 2019; Sharma *et al.*,

2020) (Fig. 1B). A huge amount of literature has been published on the application of *in vitro* genebank for sustainable conservation of PGR. Protocols for *in vitro* conservation were developed in a large number of crops (> 1000 species) justifies the successful maintenance of *in vitro* genebanks from several years.

Cryogenebank represents the conservation of PGRs at ultra-low temperature in liquid nitrogen (LN) either in liquid phase (-196 °C) or in vapor phase (-150 to -180 °C). The first scientific report on survival of plant tissues after exposure to liquid nitrogen dates back to 1956, since then major evolutionary developments in the field of plant cryobiology was reported particularly with the evolution of different techniques *viz.*, from classical cryopreservation techniques (slow cooling) to vitrification based techniques (pregrowth, pregrowth-desiccation, vitrification, droplet vitrification, encapsulation-dehydration, encapsulation-

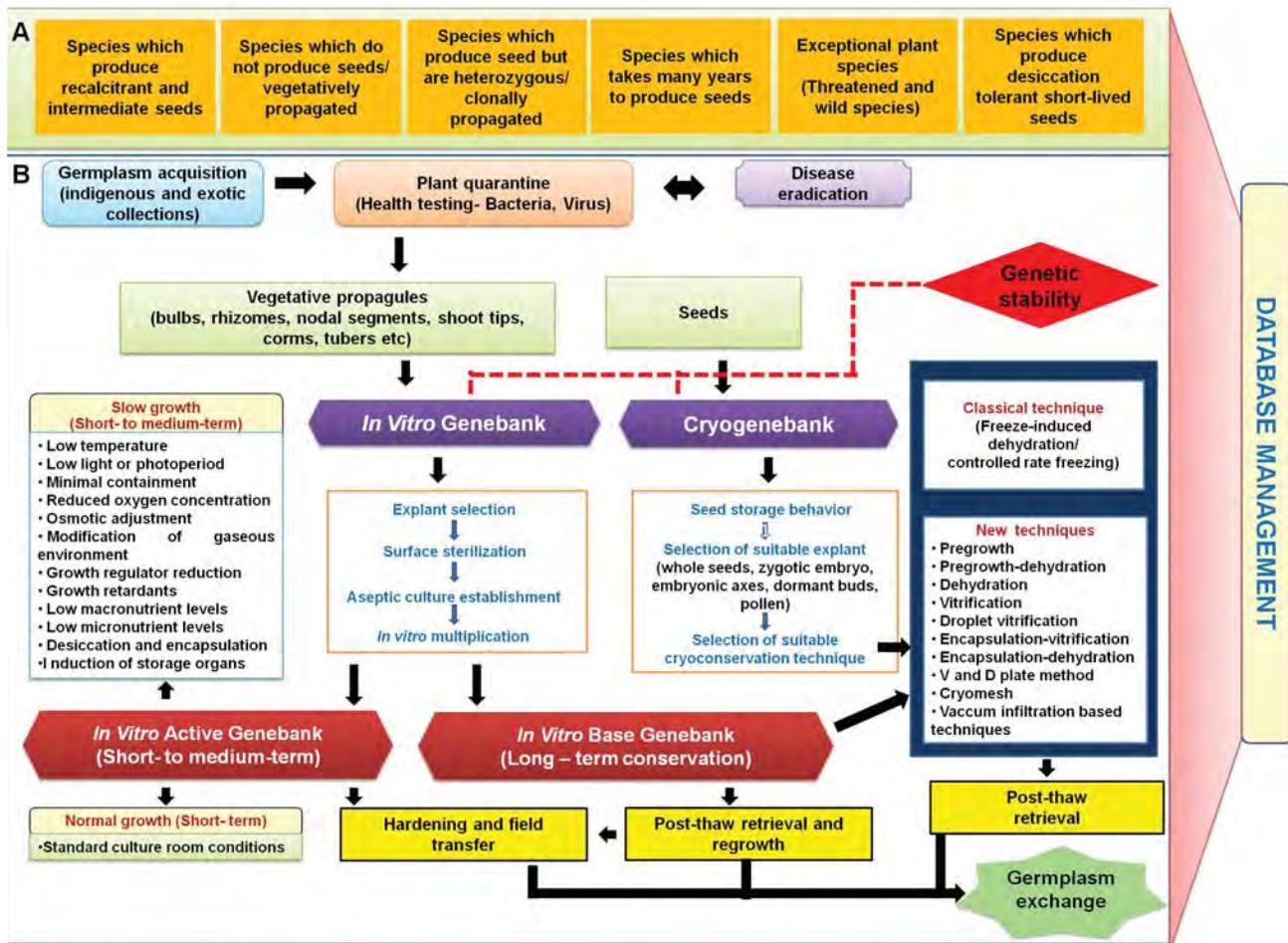


Fig. 1. *In vitro* genebank and cryogenebank (A) Priority species categories for *in vitro* and cryoconservation (B) Schematic representation of different activities in *in vitro* and cryogenebanks

vitrification, V and D cryoplates) and recently developed techniques of cryomesh and vacuum infiltration. So far, cryoconservation is applied to diverse groups of plant tissues *viz.*, whole seeds, zygotic embryos, embryonic axes, dormant buds, shoot tips, hairy roots, shoot meristems, axillary buds, nodal segments, bulbils, somatic embryos, embryogenic cell suspensions etc.). From the six decades after its birth, cryoconservation protocols were developed in diverse species for long-term conservation (Agrawal *et al.*, 2019; Sharma *et al.*, 2020). *In vitro* and cryogenebanks has several advantages than the field genebanks in conservation of difficult-to- conserve species and Fig. 2 provides their SWOT analysis.

***In vitro* and Cryogenebank of ICAR-NBPGR: 36 Years Successful Journey**

In vitro conservation and cryo-conservation of difficult-to- conserve species was initiated at ICAR-NBPGR, New Delhi in 1986 and led to the foundation for National Facility for Plant Tissue Culture Repository-NFPTCR (presently Tissue Culture and Cryopreservation Unit), a state-of -the- art -facility in the area of plant tissue culture (*in vitro* genebank) and cryo-conservation (cryogenebank) representing the unique multi crop repository (Fig. 3) to conserve clonally propagated plant species and recalcitrant seed species with funding from the Department of Biotechnology (DBT), Government of



Fig. 2. Strength, weakness, opportunity and threat (SWOT) analysis of *in vitro* and cryogenebanks



Fig. 3. *In Vitro* and Cryogenebank of ICAR-NBPGR, New Delhi. (A) *In Vitro* Active Genebank (B) *In Vitro* Base Genebank and (C) Cryogenebank

India. Currently, a total of 1,943 accessions belonging to >59 genera and >149 species belonging to six crop groups viz., (a) Tropical fruits (447), (b) Temperate and minor tropical fruits (382), (c) Tuber crops (527), (d) Bulbous and ornamental crops (173), (e) Medicinal & aromatic plants (186) and (f) Spices and industrial crops (228) are maintained either as *in vitro* cultures under normal growth (25 °C temperature, 16/8 h photoperiod regime) in the IVAG (Figure 3A) with a subculture period of 1 to 24 months, depending on the species/genotype. A total of 297 accessions of *Allium* sp., *Bacopa monnieri*, *Colocasia esculenta*, *Dioscorea* sp., *Ensete* sp., *Fragaria* sp., *Garcinia indica*, *Gentiana kurroo*, *Musa* sp., *Rubus* hybrid and *Vaccinium ovatum* were cryoconserved in IVBG (liquid phase of LN) using different cryo-conservation techniques. In addition, a total of 12,274 accessions belonging to more than 820 species have been cryoconserved as seeds, zygotic embryos, embryonic axes, dormant buds and pollen and 2,194 genomic resources (DNA) in Cryogenebank (vapor phase of LN) (Agrawal et al., 2022). With the application of efficient slow growth conservation and cryoconservation protocols, ICAR-NBPGR *In vitro* and Cryogenebank was able to conserve the germplasm without any loss during the recent incidence of COVID-19 pandemic.

Future Prospective and Action Points

- It is estimated that at least 36% of red list species are most likely to produce recalcitrant seeds. Major emphasis should be given to the protocol development for conservation of these species in *in vitro* and Cryogenebank.
 - Development of efficient protocols for vegetatively propagated and non-orthodox seeded crops, with emphasis on under-utilized crop species, rare/endangered plants and their wild relatives.
 - Need to develop generic protocols for cryo-conservation of diverse PGRs.
 - There is a need for National and International collaborations, and networks for delineating the constraints in *in vitro* and cryo-conservation of difficult to conserve species.
 - Database of *in vitro* and Cryogenebanks across the world needs to be developed for efficient knowledge sharing and material transfer.
- *In vitro* genebank and Cryogenebanks receives less importance in comparison to the seed genebanks with respect to funding. Hence there is a need for government support for establishment of *in vitro* and Cryogenebanks for conservation of difficult to conserve species.
 - Emphasis should be given for safety duplication of *in vitro* and cryoconserved germplasm.
 - Development of low-cost slow growth conservation strategies for germplasm conserved in the IVAG.
 - Automation in *in vitro* and Cryogenebank with robotic technology.
 - Capacity building through trainings on *in vitro*/cryoconservation at national and international levels.

Acknowledgements

We extend warm greetings to Padma Bhushan Dr RS Paroda on his 80th birth anniversary on August 28, 2022. We thank him for his vision and leadership in the establishment of NFPTCR facility and program, while serving as Director, NBPGR (1985-87). Authors also acknowledge the immense contributions of all present and past staff of TCCU for efficiently contributing in the sustenance of germplasm collections for the past 36 years.

References

- Agrawal A, S Singh, EV Malhotra, DPS Meena and RK Tyagi (2019) *In vitro* conservation and cryopreservation of clonally propagated horticultural species. In: P Rajasekharan and V Rao (eds) *Conservation and Utilization of Horticultural Genetic Resources*. Springer, New York, pp 529–578.
- Agrawal A, N Sharma, S Gupta, S Bansal, V Srivastava, EV Malhotra, S Chander, R Gowthami and K Singh (2022) Biotechnological applications for plant germplasm conservation at ICAR-National Bureau of Plant Genetic Resources, India—recent achievements. *Acta Hort.* **1339**: 29-42.
- Gowthami R, N Sharma, R Pandey and A Agrawal (2021) Status and consolidated list of threatened medicinal plants of India. *Genet Resour Crop Evol.* **68**: 2235–2263.
- Sharma N, R Pandey and R Gowthami (2020) *In vitro* conservation and cryopreservation of threatened medicinal plants of India. In: PE Rajasekharan and SH Wani (eds) *Conservation and Utilization of Threatened Medicinal Plants*. Springer, India, pp 181-228.

Repository of *Allium* Genetic Resources at ICAR-NBPGR: Prospects and Challenges for Collection and Conservation

Anjula Pandey*, Pavan Kumar Malav, DP Semwal, Subhash Chander, R Gowthami and KM Rai

ICAR-National Bureau of Plant Genetic Resources (NBPGR), Pusa Campus, New Delhi-110012, India

Allium genetic resources repository at the Indian National Genebank (INGB) located at ICAR-National Bureau of Plant Genetic Resources (NBPGR), in India is one of its unique kind which holds germplasm of exotic and indigenous taxa. Under national programme on plant genetic resources, efforts have been made to enrich the repository of *Allium* genetic resources with three components viz. seed genebank, *in vitro* and cryogenebank, and field genebank. In this paper the details on the collecting and conservation efforts of *Allium* species are presented, with action points highlighted for the future conservation and crop improvement programmes in the context of the Indian gene centre.

Introduction

Genus *Allium* L. (family Amaryllidaceae) is widely distributed over the warm-temperate and temperate zones of the northern hemisphere with main centre of evolution concentrated along the Irano-Turanian biogeographical region. The new global methods using cryopreservation mainly of vegetatively propagated germplasm of *Allium* have made genebanking effective and cheaper besides enhancing germplasm health, continuing characterization and evaluation to maximize the utilization held in various genebanks.

Global germplasm holdings of *Allium* maintained at main world centers are represented by country-wise large germplasm holdings (Table 1). The intensified efforts for exploration, collection and conservation of *Allium* have resulted in material conserved in various types of repositories. This paper mainly highlights the national collections in *Allium* genetic resources with information on germplasm conserved in repository by the ICAR-National Bureau of Plant Genetic Resources (NBPGR) in the Indian perspective and national efforts on collection and prioritization for future programmes. The gaps in collection and conservation are reflected with reference to future plan of action in the country.

Why Genetic Resource Programme on *Allium*?

There are many gaps in knowledge on potential of *Allium* genetic resources from the Indian region. Utilization of an accession of wild species *A. roylei* Stearn of Indian origin as a potential donor of genes for resistance to powdery mildew and leaf blight to

cultivated onion *Allium cepa* L. (de Vries *et al.*, 1992), has opened the avenues for search of newer taxa/germplasm (Beetika and Gohil, 2009). Germplasm of cultivated and wild taxa of *Allium* was introduced in late 1980s and was established in ICAR-NBPGR regional station (RS), Bhowali, Uttarakhand. *Allium* genetic resources conservation programme was given high priority under the national programme since 1986 under the leadership of Late Dr KS Negi. Further, intensified efforts towards collection, and conservation of germplasm of cultivated and wild taxa of *Allium* led to establishment of field genebank (FGB) with holding of over 30 species of native and exotic taxa at RS Bhowali (Pandey *et al.*, 2022). During planning of National Programmes (NEP 2020-25) the gap areas were included for collection of germplasm.

For the Indian region, the Himalaya hold two distinct centres of diversity for the genus *Allium* – the western Himalaya and the eastern Himalaya, in contiguity with south China and Pakistan. There is drastic loss in habitat due to anthropogenic activities, climate change and species composition in past decades affecting specific niche/habitat and loss of population due to harvest of wild *Allium* for use. This has resulted in rare, endangered and threatened (RET) status of many *Allium* taxa and thus necessitated gathering of native diversity and crop genepools (Sharma *et al.*, 2020). The Indian gene centre constitutes over 4.5-5 per cent diversity globally represented by 35-40 species of cultivated and wild taxa from temperate to alpine regions of the Indian Himalaya and tropical areas that are broadly distributed

*Author for Correspondence: Email-anjula.pandey@icar.gov.in

in different agro-ecological regions of India. The Indian *Allium* includes over 10 subgenera, 22 sections and 35–40 taxa distributed in different eco-geographical areas of the temperate and alpine regions of Himalaya sharing many taxa of Chinese origin (Astley *et al.*, 1982; Rabinowitch and Brewster, 2018; Pandey *et al.*, 2017, 2022).

From Field to Repository

As evidenced through global germplasm holdings of *Allium* species at main centers of the world, the cultivated *Allium* species are well collected, while the wild taxa are meagerly or under-represented in global collections and species distribution areas are not adequately covered. Global germplasm collections of *Allium* of cultivated and wild taxa (over 300 species) have been built-up at

IPK, Gatersleben, Germany (Keller and Kik, 2018).

In Indian genebanks, the germplasm of cultivated and wild *Allium* species including exotic germplasm is conserved as *ex situ* collection in seed genebank, *in vitro*/ cryo-repository and in field genebank facilities. Germplasm has been assembled from high hills of Uttarakhand, Himachal Pradesh, Jammu and Kashmir (J&K) of the Western Himalayas and the high altitude regions of north-eastern region (Table 2).

Germplasm Collection and Gap Analysis

Gap analysis of the existing holdings in the repository was based on their performance of the material and its conservation status (common, rare/endangered/threatened) and other aspects was taken into consideration

Table 1. Global germplasm holdings of *Allium* spp. at main centers*

Country	Institute/Center	Accessions
<i>Allium cepa</i>, Total = 4,161		
Bulgaria	Institute for Plant Genetic Resources (IPGR), Sadovo	398
Poland	Plant Genetic Resources Laboratory of Research Institute of Vegetable Crops, Skierniewice	144
Germany	Genebank, Institute for Plant Genetic and Crop Plant Research (IPK), Gatersleben	348
Hungary	Ministry of Agriculture and Rural Development (MARD), Budapest	305
India	ICAR-National Bureau of Plant Genetic Resources (NBPGR), New Delhi	1,606
Taiwan	Asian Vegetable Research and Development Centre (AVRDC), Tainan	586
USA	United State Department of Agriculture and Development (USDA) Germplasm Resource Network	774
<i>A. sativum</i>, Total = 3,705		
Bulgaria	IPGR, Sadovo	220
China	Vegetable Crop Gene Resources and Germplasm Enhancement (VCGRGE), Ministry of Agriculture, Beijing	417
Germany	IPK, Gatersleben	485
Hungary	MARD, Budapest	51
India	ICAR-NBPGR, New Delhi	913
Czech Republic	Crop Research Institute, Prague	157
South Korea	National Agrobiodiversity Center (NAAS), RDA, Suwon	1,158
USA	USDA Germplasm Resource Network	304
<i>A. fistulosum</i>, Total = 68		
Germany	IPK, Gatersleben	18
Hungary	MARD, Budapest	24
India	ICAR-National Bureau of Plant Genetic Resources (ICAR-NBPGR), New Delhi, India	26
<i>A. ampeloprasum</i> Total = 111		
Germany	IPK, Gatersleben	91
India	ICAR-NBPGR, New Delhi	9
Netherlands	Centre for Plant Breeding and Reproduction Research (CPRO-DLO), Department of Vegetable and Fruit Crops, Wageningen	11
<i>A. proliferum</i> Total = 154		
Germany	IPK, Gatersleben	154
<i>A. chinense</i> Total = 23		
India	ICAR-NBPGR, New Delhi	23
<i>A. tuberosum</i> Total = 216		
India	ICAR-NBPGR, New Delhi	29
China	VCGRGE, Ministry of Agriculture, Beijing	174
USA	USDA Germplasm Resource Network	13

*updated from individual websites (December 2021)

Table 2. Status of *Allium* species indigenous collections from different states (1976-2022) #

S.No	Cultivated species	Accessions [#]	Wild species	Accessions [#]	Threatened status
1.	<i>A. cepa</i> L. var. <i>cepa</i> L.	2,847	<i>A. carolinianum</i> DC.	25	
2.	<i>A. sativum</i> L.	1,913	<i>A. auriculatum</i> Kunth	12	Endangered
3.	<i>A. cepa</i> var. <i>aggregatum</i> G.Don.	85	<i>A. griffithianum</i> Boiss.	11	
4.	<i>A. fistulosum</i> L.	17	<i>A. wallichii</i> Kunth	7	
5.	<i>A. tuberosum</i> Rottler ex Spreng.*	13	<i>A. consanguineum</i> Kunth	6	
6.	<i>A. chinense</i> G.Don	11	<i>A. przewalskianum</i> Regel	5	
7.	<i>A. ampeloprasum</i> L.	11	<i>A. humile</i> Kunth	5	
8.	<i>A. x proliferum</i> (Moench) Schrad. ex Willd.	10	<i>A. stracheyi</i> Baker	3	Vulnerable
9.	<i>A. hookeri</i> Thwaites	10	<i>A. semenovii</i> Regel	2	
10.	<i>A. cepa</i> var. <i>viviparum</i> (Metz.) Alef.	4	<i>A. fasciculatum</i> Rendle	1	
11.	<i>A. porrum</i> L.	3	<i>A. roylei</i> Stearn	1	Endangered
12.			<i>A. prattii</i> C.H.Wright	1	Rare
13.			<i>A. victorialis</i> L.	1	
	Total	4,924		80	

Semwal *et al.* (2021)

to identify under-represented/least represented taxa/ areas of diversity in the national holdings. *Allium* genetic resources in India with a total of 5,004 diverse accessions of cultivated and wild species of *Allium* were collected through explorations undertaken during 1976-2022, which includes mainly common onion and garlic among the cultivated types. Seeds of wild *Allium* species are preferably conserved in the seed genebank, while vegetatively propagated species are conserved in *in vitro*/ cryogenebank repository at New Delhi and field genebank (FGB) at ICAR-NBPGR, Regional Station, Bhowali, Uttarakhand including eight species of rare, endangered and threatened status (Table 3).

Germplasm Conservation

Ex situ conservation strategies for *Allium* germplasm is determined by reproductive biology of the species, mode of multiplication, threat due to different causal factors and status of occurrence (common or less commonly reported, endemic, rare/ vulnerable, etc.) (Chandel and Pandey 1992). *Ex situ* conservation repository at the ICAR-NBPGR has the following components: a) seed genebank, b) *In vitro* genebank and Cryo-genebank and c) Field genebank (Table 3). Flow of material from collection to conservation and use is depicted in flow diagramme (Fig. 1). Seeds of *Allium* species are orthodox and short lived, and have been conserved in seed genebank at ICAR-NBPGR. Due to short longevity of *Allium* seeds, adequate conservation protocols for seed viability have been developed at the ICAR-NBPGR.

For live collections indigenous and exotic wild species are maintained, characterized and evaluated

at field genebank (FGB) at Bhowali, Uttarakhand. *In vitro*/cryo genebank was established under a project entitled 'National Facility for Plant Tissue Culture Repository' (NFPTCR) (currently Tissue Culture and Cryopreservation Unit, TCCU) jointly by Indian Council of Agricultural Research (ICAR) and Department of Biotechnology (DBT), India. This facility conserves *Allium* under a unique and multi-crop repository.

Meager conservation of wild germplasm in genebanks may be attributed to various reasons like niche-specificity, lack of conservation protocols for *ex situ*, rapid loss of seed viability, poor performance/ non-suitability to climatic conditions of FGB. While collecting, asynchronous maturity and seed shattering pose great problems to gather material in sufficient quantity of desired for conservation.

In vitro conservation of *Allium* species is attained under normal growth conditions or subjected to growth-limiting conditions for short- to medium-term conservation in the *In vitro* active genebank (IVAG) and under suspended growth for long-term conservation using cryopreservation techniques in the *In vitro* base genebank (IVBG) (Pandey *et al.*, 2018). For short-to medium-term conservation (IVAG), different slow growth *in vitro* conservation strategies developed/refined at TCCU for *Allium* spp. use a single or combination of strategies. Whereas long-term conservation (IVAG and Cryogenebank) using *in vitro* explants (shoot tips) was laid emphasis to cryopreserve all the germplasm conserved *in vitro* in the IVAG. Different cryopreservation techniques – vitrification,

droplet-vitrification, encapsulation-dehydration and encapsulation-vitrification are used. Shoot tip cryopreservation using droplet-vitrification is found to be superior, with a varied degrees of post-thaw regrowth (20-60%) among the different species. So far, 159 accessions belonging to 10 different species of *Allium* are conserved in the form of *in vitro* cryopreserved shoot tips in the IVBG (-196°C, LN in liquid form). Genetic stability has been assessed in cryopreserved germplasm of *Allium* using morphological, cytological and molecular markers, and no significant variation observed in the mother plants and cryopreserved plants. In addition, in order to facilitate pre-breeding in *Alliums* crop improvement and as natural propagules of genetic information, pollen have also been conserved in cryogenebank; pollen cryopreservation protocol was standardized in *A. chinense* and *A. tuberosum*.

Field Genebank at RS Bhowali

The “*Allium* Field Genebank” at ICAR-NBPGR RS Bhowali, Uttarakhand, has germplasm of indigenous

as well as exotic species, and was set up by Late Dr KS Negi in 1987. It holds several germplasm collected from difficult areas of Uttarakhand (Niti valley, Milam glacier, and the Valley of Flowers in Western Himalayas), J&K, Himachal Pradesh, Sikkim and Nagaland in the north-eastern region (Negi, 2006). A total of over 150 accessions of *Allium* species are maintained in FGB at RS Bhowali from 1986-2022 and supplied for research to various indentors in the country. Among the major collections *A. tuberosum*, *A. sativum*, *A. negianum*, *A. chinense*, *A. hookerii*, *A. przewalskianum*, *A. schoenoprasum* and *A. ampeloprasum* collected from western and North-eastern Himalayan regions of India are maintained including a few accessions of critically endangered species (*A. stracheyi*). Some of the exotic taxa conserved include *A. tuberosum*, *A. fistulosum*, *A. sativum*, *A. altaicum*, *A. ledebouranum*, *A. lineare*, *A. oreoprasum*, *A. schaninii*, *A. pskemense*, *A. albidum*, *A. fistulosum*, *A. ampeloprasum*, *A. obliquum*, *A. ramosum*, *A. schoenoprasum* and *A. angulosum* from Australia, USSR and Switzerland.

Table 3. Ex situ conservation of cultivated and wild *Allium* species at ICAR-NBPGR

	Ex situ conservation method				
	Seed genebank	Cryogenebank	In vitro genebank	Field genebank	Total
Cultivated <i>Allium</i> Species					
<i>A. cepa</i>	991	9		606	1,606
<i>A. sativum</i>		134		779	913
<i>A. fistulosum</i>	16	3	1	6	26
<i>A. chinense</i>		11	9	3	23
<i>A. tuberosum</i>		7	4	18	29
<i>A. hookeri</i>		2	2	12	16
<i>A. ampeloprasum</i>		2		7	9
<i>A. cepa</i> var. <i>aggregatum</i>		1		3	4
<i>A. fasciculatum</i>			1	14	15
Sub-total	1,007	169	17	1,448	2,641
Wild <i>Allium</i> species					
<i>A. przewalskianum</i> #	1			19	20
<i>A. griffithianum</i>	2	1		8	11
<i>A. carolinianum</i>	3			5	8
<i>A. stracheyi</i> #	2			5	7
<i>A. wallichii</i> #		1		6	7
<i>A. auriculatum</i>	2	1		3	6
<i>A. roylei</i> #	1	1		1	3
<i>A. humile</i>	1			1	2
<i>A. fasciculatum</i>				1	1
<i>A. prattii</i>				1	1
<i>A. victorialis</i>	1				1
Sub-total	13	4		50	67
Total	1,020	173	17	1,498	2,708

Source: Semwal et al. (2021)

Challenges and Action Points

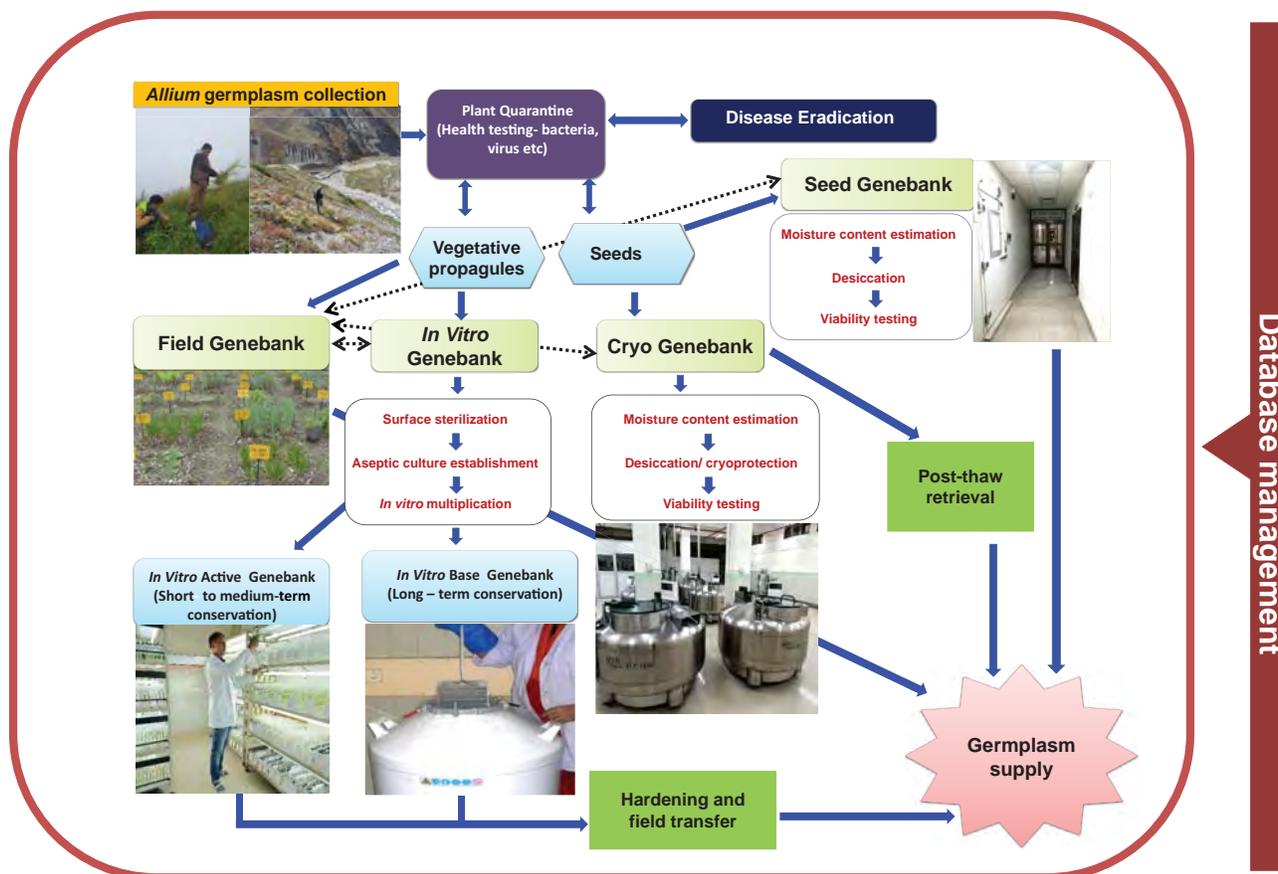
- Difficult access to habitats and wild and threatened species.
- Asynchronized seed maturity/seed shattering of seeds
- Collecting desirable number of seeds/propagules for conservation
- On-spot identification of taxon
- Standardization of cryopreservation protocol
- Limitation in sufficient germplasm for *in vitro* study and lack of manpower/tools for frequent subculturing during *in vitro* conservation
- Establishment problems at FGB and loss of material due to infection/contamination during *in vitro* conservation
- Evaluation and identification/characterization – differential response to agro-climatic conditions leading to no/poor flowering; seed production

- Sexual sterility and poor post-harvest storage response of some species leading to loss of viability during post-harvest process
- Danger of genetic erosion due extensive harvest of the bulbs from wild population.
- Systematics study on identification of genetic resources taxa from subgenus *Cepa* in India
- Identification of *in situ* on-farm sites for conservation

Future Prospective and Action Points Identified

- Prioritization of sites/ areas and underexplored regions for wild *Allium* species
- Focus on wild and hitherto uncollected species diversity including *A. roylei*, *A. farctum*, *A. rhabdotum* which are known for cold hardiness traits and biotic resistance
- Strengthening of repositories: introduction of primary gene pool of onion from native areas

Repository of *Allium* Genetic Resources at ICAR-NBPGR



- Basic studies on reproductive biology of rare/endangered taxa- *A. stracheyi*, *A. wallichii*, *A. auriculatum*, *A. humile* and *A. roylei*.
- Protocol establishment: *in situ* conservation, multiplication, evaluation of germplasm; cryo/*in vitro* conservation study
- Identification of sites: niche specific germplasm (high altitude germplasm that does not establish at lower altitude/ flower/ seed).

References

- Astley D, NL Innes and QP Van der Meer (1982) *Genetic Resources of Allium Species- A Global Report*. IPGRI, Rome.
- Beetika K and RN Gohil (2009) Need to conserve *Allium roylei* Stearn: a potential gene reservoir. *Genet Resour Crop Evol* **56**: 891-893.
- Chandel KPS and R Pandey (1992) Distribution, diversity, uses and *in vitro* conservation of cultivated and wild Alliums: a brief review. *Indian J Plant Genet Resour* **5**(2): 7-36.
- De Vries JN, Wietsma WA and T de Vries (1992) Introgression of leaf blight resistance from *A. roylei* Stearn into onion (*A. cepa* L.). *Euphytica* **62**: 127-133.
- Fritsch R, N Friesen and Bachmann (1999) Research activities in the department of taxonomy, IPK, Gatersleben. In: Maggioni L, Astley D, Rabinowitch H, Keller J and Lipman E. *Report of a working group on Allium*. Sixth Meeting, 23-25 October 1997, Bulgaria, International Plant Genetic Resources Institute, Rome, Italy, pp 76-79.
- Keller ERJ and C Kik (2018) *Allium* genetic resources. In: M Shigyo, A Khar and M Abdelrahman (eds), *The Allium Genomes* (Compendium of Plant Genomes), pp. 23-52. https://doi.org/10.1007/978-3-319-95825-5_3
- NEP (2020-25) National Exploration Plan. ICAR-National Bureau of Plant Genetic Resources, New Delhi.
- Negi KS (2006) *Allium* species in Himalayas and their uses with special reference to Uttaranchal. *Ethnobotany* **18**: 53-66.
- Pandey A, R Pandey, KS Negi and J Radhamani (2008) Realizing value of genetic resources of *Allium* in India. *Gen. Resour. Crop Evol.* **55**: 985-994.
- Pandey A, K Pradheep and KS Negi (2017) Onion and related taxa: ecogeographical distribution and genetic resources in Indian subcontinent. In: Ansari AA, I Gill, S Singh, I Abbas, M Naeem (eds) *Plant Biodiversity: Monitoring, Assessment and Conservation*. Wallingford, Oxfordshire, Boston MA, CABI International, pp 429-442.
- Pandey A, PK Malav, KM Rai and SP Ahlawat (2022) *Genus Allium L. of the Indian Region: A Guide for Germplasm Collection and Identification*. ICAR-National Bureau of Plant Genetic Resources, New Delhi, India, 110p + i-iii.
- Pandey R, R Gowthami, EV Malhotra, N Sharma, R Chandra, DPS Meena, DK Nerwal and A Agrawal (2018) *In vitro* conservation and cryopreservation of clonally propagated crops. In: Rajkumar S, S Kumar, SK Kaushik and A Agrawal (eds) *Compendium for 'National Training Course on Contemporary Approaches to Plant Genetic Resources Management'*. ICAR-NBPGR, New Delhi, Nov. 27-Dec. 17, 2018, pp 73-84.
- Pandey R, R Gowthami, N Sharma, R Chandra and A Agrawal (2019) Cryopreservation techniques in Alliums. In: Agrawal A et al. (eds) *Laboratory Handbook for Eighth International Training Course on In Vitro and Cryopreservation Approaches for Conservation of Plant Genetic Resources*. ICAR-NBPGR, New Delhi, Bioversity International-India, Delhi, APAARI/APCoAB, Bangkok, pp 59-64.
- Rabinowitch HD and JL Brewster (2018) *Onions and Allied Crops*. Publisher CRC Press.
- Semwal DP, A Pandey and SP Ahlawat (2021) Genetic resources of genus *Allium* in India: Collection status, distribution and diversity mapping using GIS tools. *Indian J. Plant Genet. Resour.* **34**(2): 206-215.
- Sharma N, R Pandey and R Gowthami (2020) *In vitro* conservation and cryopreservation of threatened medicinal plants of India. In Rajasekharan PE, Wani SH (eds) *Conservation and Utilization of Threatened Medicinal Plants*. Springer, India, pp 181-228.

Grain Legumes Diversity of Indian National Genebank: A Potential Resource for Food and Nutritional Security

Padmavati G Gore, Neeta Singh* and Veena Gupta

¹ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

In the INGB (Indian National Genebank), a total of 66,283 accessions belonging to 15 genera of 98 species of grain legumes are conserved, which includes 55,757 indigenous collections and 10,526 exotic collections. Many novel and unique traits have been identified in the conserved germplasm including resistance to major pests and disease, agronomic and nutritional value. Conserved legume genetic resources have the potential to mitigate future climate change and revolutionize the legume breeding programme.

Introduction

Legumes are the key protein sources and have always been a part of everyday life, as human food and animal feed. Legumes represent the second most important family of crop plants after Poaceae (grass family), accounting for approximately 27% of the world's crop production. The diversity of legumes comprises around 20,000 species and 700 genera (Lewis *et al.*, 2005). In today's changing climate scenario, to prevent the loss of legume diversity, *ex situ* conservation is essential.

Status of grain legumes conserved in INGB

In the Indian National Gene Bank (INGB), a total of 66,283 accessions belonging to 15 genera of 98 species are conserved. Among them, 84% of accessions were collected from the Indian gene centre while 16% were imported, majorly from the USA, Philippines, Nigeria, Thailand and China. A rich diversity of cultivated and wild *Vigna* species occurs in India. Cultivated species *V. radiate* and *V. mungo* originated in India (Arora, 1985). In the past, enormous efforts have been taken to collect the treasure of this *Vigna* diversity and it is evident from the status of genus *Vigna* conserved in INGB. Out of 15 genera, *Vigna* has the highest number of accessions (15,113) and the number of species (32). Among 15,113 accessions, 13,199 belong to India and 1,914 accession of exotic collections. After genus *Vigna*, *Cicer* has the largest number of accession (15,053) in INGB, belonging to the 10 species, followed by genus *Cajanus* with 11,940 accessions of belonging to seven species (Table 1).

The crop wild relatives (CWRs) are important genetic resources for breeding and crop improvement programs (Vavilov, 1992). As CWRs are typically adapted to different environmental conditions than their domesticated relatives, genetic material from these wild species has the potential to play an important role in breeding for greater abiotic and biotic stress tolerance (Hajjar and Hodgkin, 2007). As climate change and human population expansion threaten global food security, CWRs are poised to play a significant role in mitigating these forthcoming challenges. CWRs are a natural resource and deserved a target for urgent and systematic conservation (Naidoo *et al.*, 2008). In INGB, more than 80 wild relatives of legumes are conserved under long-term storage. Table 2 represents the major cultivated crop legume along with their wild relatives conserved in INGB. In conserved accessions of wild species, unique and novel traits have been identified and registered by the Plant Germplasm Registration Committee of the Indian Council of Agricultural Research (ICAR). For example, the species *Lens orientalis* (Boiss.) Hand. Mazz (EC718515) for resistance against rust (*Uromyces fabae* (Grev.) Fuckel) and powdery mildew (*Erysiphe trifolii*); *Lens nigricans* (M. Bieb.) Webb & Berthel, (EC718266) for resistance against rust (Singh *et al.*, 2020). In genus *Vigna*; *Vigna vexillata* (L.) A.Rich (IC259504) for high protein content (9.5%) in tuber, bold seededness. and fodder type (Tripathi *et al.*, 2021); *Vigna vexillata* (IC248326) for resistance against *Callosobruchus maculatus* L. (Aidbhavi *et al.*, 2022); *Vigna stipulacea* (Lam.) Kuntze (IC336136) for early flowering and early maturity (Gore *et al.*, 2021).

*Author for Correspondence: Email-neeta.singh@icar.gov.in

Table 1. Status of grain legumes conserved in Indian National Genebank

S.No.	Genus	Number of accessions	Number of species	Indigenous collection	Exotic collection
1.	<i>Cajanus</i>	11,940	7	11576	364
2.	<i>Canavalia</i>	132	4	128	4
3.	<i>Cicer</i>	15,053	10	11908	3,145
4.	<i>Cyamopsis</i>	4,308	1	4,270	38
5.	<i>Lablab</i>	1,305	4	1,298	7
6.	<i>Lathyrus</i>	2,747	10	2,557	190
7.	<i>Lens</i>	2,612	7	1,852	760
8.	<i>Macrotyloma</i>	3,157	2	3,146	11
9.	<i>Mucuna</i>	55	2	49	6
10.	<i>Phaseolus</i>	4,154	3	1,730	2,424
11.	<i>Pisum</i>	4,680	1	3528	1,152
12.	<i>Psophocarpus</i>	220	1	84	136
13.	<i>Rhynchosia</i>	21	10	21	0
14.	<i>Vicia</i>	786	4	411	375
15.	<i>Vigna</i>	15,113	32	13,199	1,914
	Total	66,283	98	55757	10526

Table 2. Major cultivated legume and their wild relatives conserved in NGB

Cultivated species (number of accessions)	Wild relatives (number of accessions)
<i>Cajanus cajan</i> (L.) Huth (11,879)	<i>Cajanus albicans</i> (Wight & Arn.) Maesen (6); <i>Cajanus cajanifolius</i> (Haines) Maesen (2); <i>Cajanus mollis</i> (Benth.) Maesen (3); <i>Cajanus platycarpus</i> (Benth.) Maesen (2); <i>Cajanus scarabaeoides</i> (L.) Thouars (47); <i>Cajanus volubilis</i> (Blanco) Blanco (1).
<i>Cicer arietinum</i> L. (14,844)	<i>Cicer bijugum</i> Rech.f. (31); <i>Cicer chorassanicum</i> (Bunge) Popov (2); <i>Cicer cuneatum</i> Hochst. ex A.Rich.(6); <i>Cicer echinospermum</i> P.H.Davis (19) <i>Cicer judaicum</i> Boiss.(54); <i>Cicer microphyllum</i> Royle ex Benth. (38); <i>Cicer pinnatifidum</i> Jaub. & Spach (27); <i>Cicer reticulatum</i> Ladiz. (19); <i>Cicer yamashitae</i> Kitam. (4)
<i>Lens culinaris</i> Medik (2,422)	<i>Lens culinaris</i> subsp. <i>Odemensis</i> (Ladiz.) M.E.Ferguson, Maxted, van Slageren & L.D.Robertson (29); <i>Lens orientalis</i> (Boiss.) Hand. Mazz. (63); <i>Lens culinaris</i> subsp. <i>Tomentosus</i> (Ladiz.) M.E.Ferguson, Maxted, van Slageren & L.D.Robertson (6); <i>Lens ervoides</i> (Brign.) Grande (67); <i>Lens lamottei</i> Czeff. (3); <i>Lens nigricans</i> (M.Bieb.) Webb & Berthel. (22)
<i>Vigna</i> <i>Vigna radiata</i> (L.) R.Wilczek (4,379); <i>Vigna unguiculata</i> (L.) Walp (4,003); <i>Vigna mungo</i> (L.) Hepper (2,297); <i>Vigna umbellate</i> (Thunb.) Ohwi & H. Ohashi (2,235); <i>Vigna aconitifolia</i> (Jacq.) Maréchal (1,530); <i>Vigna angularis</i> (Willd.) Ohwi & H.Ohashi (200)	<i>Vigna glabrescens</i> Maréchal, Mascherpa & Stainier; <i>Vigna khandalensis</i> (Santapau) Sundararagh. & Wadhwa; <i>Vigna membranacea</i> A.Rich.; <i>Vigna minima</i> (Roxb.) Ohwi & H.Ohashi; <i>Vigna reticulata</i> Hook.f.; <i>Vigna gracilicaulis</i> (Ohwi) Ohwi & H Ohashi; <i>Vigna vexillata</i> var. <i>wightii</i> (Benth. ex Bedd.) Babu & SK Sharma (1 accession each). <i>Vigna luteola</i> (Jacq.) Benth.; <i>Vigna parkeri</i> Baker; <i>Vigna racemosa</i> (G Don) Hutch. & Dalziel ex Baker f.; <i>Vigna mungo</i> var. <i>silvestris</i> Lukoki, Maréchal & Otoul; <i>Vigna trinervia</i> (B.Heyne ex Wight & Arn.) Tateishi & Maxted (2 accession each). <i>Vigna nepalensis</i> Tateishi & Maxted; <i>Vigna radiata</i> var. <i>setulosa</i> (Dalzell) Ohwi & H Ohashi; <i>Vigna bourneae</i> Gamble; <i>Vigna pilosa</i> (JG Klein ex Willd.) Bake (4 accessions each). <i>Vigna hainiana</i> Babu, Gopin. & SK Sharma; <i>Vigna marina</i> (Burm.) Merr. (7 accessions each). <i>Vigna trilobata</i> (L.) Verdc. (10); <i>Vigna bourneae</i> Gamble (11); <i>Vigna angularis</i> var. <i>nipponensis</i> (Ohwi) Ohwi and H. Ohashi (13); <i>Vigna mungo</i> var. <i>silvestris</i> Lukoki, Maréchal & Otoul (16); <i>Vigna vexillata</i> (L.) A.Rich. (18); <i>Vigna dalzelliana</i> (Kuntze) Verdc. (34); <i>Vigna radiata</i> var. <i>sublobata</i> (Roxb.) Verdc. (45); <i>Vigna stipulacea</i> (Lam.) Kuntze (99)

In context to the passport data out of a total 66,283 accessions, 55,757 are indigenous collections and 10,526 are exotic collections. In the indigenous collection, 28% (3,368) accessions belong to the state of Andhra Pradesh. While from the state of Pondicherry, Tripura, Mizoram and Andaman and the Nicobar Islands, less

than 100 accessions of grain legumes are conserved. Out of 95 accessions from Andaman and Nicobar Island, 71 accessions are of genus *Vigna* including economically important species like., *Vigna marina* (salt tolerant); tuber cowpea, *Vigna Vexillata*; *Vigna trilobata* (drought tolerant). All the 15 accessions from Pondicherry belong

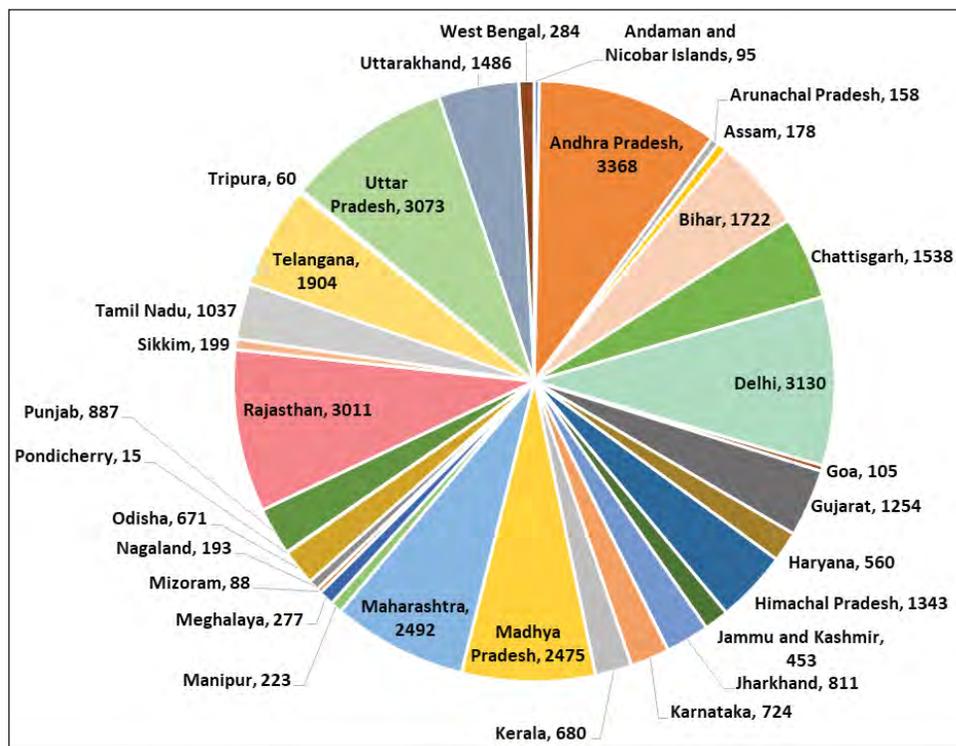


Fig. 2. State-wise collection of conserved grain legumes

to the *Lablab purpureus* (L.) Sweet. Figure 2 represents the state-wise collection of conserved germplasm of grain legumes.

To cope with climatic changes, the breeding programmes are in utmost need of diverse genes to develop varieties with better performance over a large range of environmental conditions. On the other side plant genetic resources are under threat of loss due to various reasons including, genetic erosion, change in land use patterns and climate change. Hence, collection and conservation of plant diversity is needed.

References

- Gore PG, V Gupta, R Singh, K Tripathi, R Kumar, G Kumari, L Madhavan, HK Dikshit, K Venkateswaran, A Pandey, N Singh, KV Bhat, RM Nair and A Pratap (2022) Insights into the genetic diversity of an underutilized Indian legume, *Vigna stipulacea* (Lam.) Kuntz., using morphological traits and microsatellite markers. *PLoS One*, **17**(1): 0262634.
- Hajjar R and T Hodgkin (2007) The use of wild relatives in crop improvement: A survey of developments over the last 20 years. *Euphytica* **156**: 1-13.
- Lewis G, B Schrire, B Mackind and M Lock (2005) Legumes of the World. Royal Botanic Gardens, Kew, UK.
- Naidoo R, A Balmford, R Costanza, B Fisher, RE Green, B Lehner, TR Malcolm and TH Ricketts (2008) Global mapping of ecosystem services and conservation priorities. *Proc. Natl. Acad. Sci.* **105**: 9495-9500.
- Singh M, S Kumar, AK Basandrai, D Basandrai, N Malhotra, DR Saxena, *et al.* (2020) Evaluation and identification of wild lentil accessions for enhancing genetic gains of cultivated varieties. *PLoS One* **15**(3): e0229554.
- Tripathi K, PG Gore, A Pandey, ER Nayar, C Gayacharan, RK Pamarthi, R Bhardwaj and A Kumar (2021) Morphological and nutritional assessment of *Vigna vexillata* (L.) A. Rich.: a potential tuberous legume of India. *Genet. Resour. Crop Evol.* **68**, 397-408.
- Vavilov NI (1992) Origin and Geography of Cultivated Plants. Cambridge University Press.

Genomic Tools in Plant Genetic Resource Management

Dhammaprakash P Wankhede, Rakesh Singh and S Rajkumar*

ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

Significant progress has been made in conservation of plant germplasm in last few decades. However, the utilization of genetic resources has remained poor. The reason for the under utilization of plant genetic resources (PGR) are non-availability of structured information and resources from gene banks. This was due to lack of characterization and evaluation data, and under usage of genomic tools in characterization. With the evolution of genomic tools, which get cheaper with discovery of newer technologies, it is pertinent to use these tools for management and utilization of PGR from vast germplasm conserved in gene banks throughout world. In this context the present article describes the tools that can be used in gene bank management and the way to utilize the germplasm with the examples from the Indian National Gene Bank.

Key Words: Core sets, DNA bar coding, Genetic erosion, Genotyping, Phenotyping, Reference sets

Introduction

Plant genetic resources (PGR) represent natural variation in crops and their wild relatives, which support food security, especially under climate change and nutritional challenges. Significant progress has been made in conservation of plant germplasm in last few decades. However, the utilization of genetic resources has remained poor. The reason for the underutilization of PGR are non-availability of structured information and resources from gene banks. This was due to lack of characterization and evaluation data, and under usage of genomic tools in characterization. With the evolution of genomic tools, which get cheaper with discovery of newer technologies, it is pertinent to use these tools for management and utilization of PGR from vast germplasm conserved in gene banks throughout world. In this context the present article describes the tools that can be used in gene bank management and the way to utilize the germplasm with the examples from the Indian National Gene Bank.

Species Integrity of Seed Gene Bank

Plant germplasm conserved in gene bank is important source of material for studies on crop domestication, evolution and for crop improvement. The identity of each accession is important as it affects all the downstream process in crop improvement and related studies. This process is an important aspect in gene bank management. Most of the germplasm in gene bank are cultivated

and readily identifiable, but occasional problem exists in crop wild relatives, especially vegetable crops. Most of the vegetable crops come under the family Cucurbitaceae where morpho-taxonomy is quite difficult. DNA barcoding is set of coding regions of nuclear and chloroplast DNA, and is useful in systematics study in plants. These loci are universally accepted and used in identification of plant species. In a study by the authors, barcoding loci useful in delineation of species complex in wild relatives of vegetable crops were identified and effectively used in identification of germplasm with unknown species status and problematic species complex. Among the universally accepted barcoding loci, *rbcL* and *trnH-psbA* were more useful in delimiting *Luffa* spp. including *L. acutangula* complex and status of *L. tuberosa*. In vegetable *Amaranthus* spp. nuclear ITS sequence was more appropriate in differentiating vegetable *Amaranthus* species along with delineation of species complexes (*blitum* and *tricolor*). In *Trichosanthes cucumerina*, ITS combined with *rbcL* were effective in delineating *T. cucumerina* species complex. The delineation of taxonomic complex existing within species of *Cucumis melo* were carried out with combine ITS and *rbcL* and they are able to distinguish different taxonomic varieties under the species.

Estimation of Genetic Erosion

In the agricultural sphere, there is ongoing concern and attention to genetic erosion at all levels, including

*Author for Correspondence: Email-S.Rajkumar@icar.gov.in

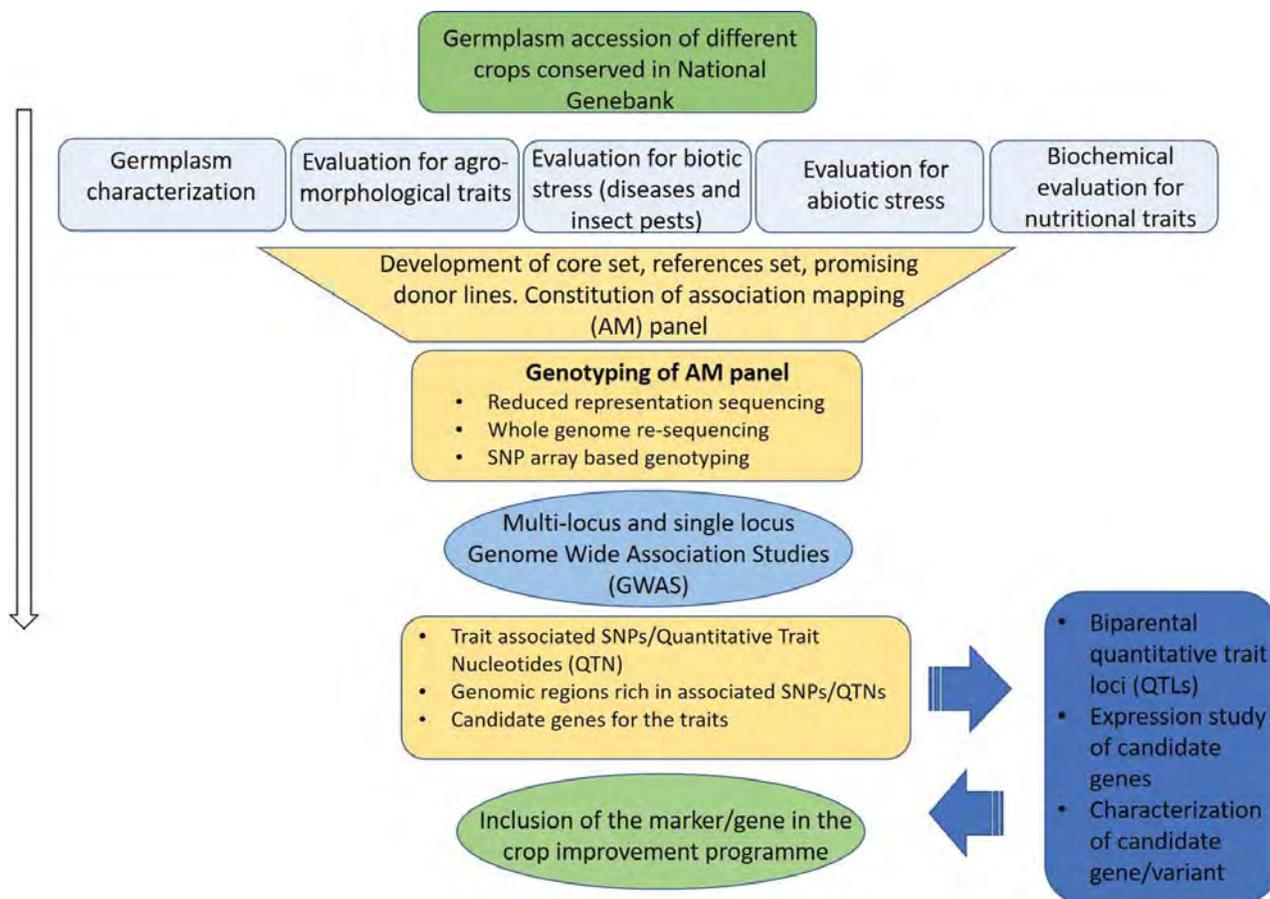


Fig. 1. Overview of approach for utilization of Genebank accessions using Genome Wide Association Studies (GWAS) towards identification marker/gene-trait association for crop improvement

the Food and Agriculture Organization of the United Nations (FAO). It is important to note that along with the loss of diversity at the species, varietal and allelic levels, genetic erosion can also occur at the level of germplasm collections and gene banks due to improper management and inadequate regeneration procedures. A large amount of genetic diversity has and is being collected and stored in gene banks, in which every sample (usually of seed) is kept in airtight containers at -10 to -200 °C and 5 to 7% humidity for 50 to 100 years (Damania, 2008). From time-to-time, a given amount of material is taken out of the gene bank, planted in the field and ‘rejuvenated’: the fresh seed is then stored again. Worldwide, 1308 gene banks are registered and conserve a total of 6.1 million accessions, including major crops, minor or neglected crop species as well as trees and wild plants. Of the 30 main crops, more than 3.6 million accessions are conserved *ex situ*. On one hand these collections serve a very important

purpose - avoiding the loss of individuals and species, and of the genes they carry, which may be unique. On the other hand, by ‘freezing’ seeds they also ‘freeze’ evolution at the time of the collection. Therefore, many advocate that together with the *ex situ* conservation in gene banks, diversity should also be conserved in its original locations (*in situ*), where the plant populations can continue to evolve.

Genetic erosion as a reduction in evenness originates from the diversity indices used in vegetation ecology and population genetics, such as Shannon’s index or Nei’s gene diversity index. Diversity is measured using the frequencies of alleles within a group of genotypes or using the production areas of landraces, cultivars or crop species in a region. Diversity levels are lowered due to increasing dominance of a single or small number of crop species, genotypes or alleles, even though alleles or varieties are not necessarily lost. Using evenness, rare varieties or rare alleles contribute little to the diversity.

The risks of losing alleles or varieties are higher when distributions are very skewed. Using evenness as a measure for genetic erosion offers the opportunity to take action before a reduced diversity results in an absolute loss and reduced richness. Furthermore, it is not as sensitive to the sampling procedure as compared with the previous measures. Considerable overlap between these three views on genetic erosion exists, and most studies use a combination of the different approaches.

A case study was conducted to identify and characterize changes in long-term conserved samples of safflower genetic resources in National Genebank (NGB) and to estimate the risk of genetic erosion during the conservation process. Total four accessions (50 individuals each) were selected based on the availability of historic material in the gene bank and deposition of regenerated material after constant cycle of regeneration. For estimating variation in allelic frequency, genomic SSR markers were used. The markers with high polymorphic information content (PIC) content from the earlier studies were used to generate allelic diversity in both regimes of the accessions. The results showed changes mainly due to some highly significant differences in allele frequencies, whereas the majority of alleles occur in similar frequencies. This implies that either regeneration protocols should be improved or the composition of the collection should be changed in the gene bank.

Molecular Core Development

A large number of genetic materials have been conserved in gene banks, but their use is limited due to an unmanageable number of accessions and the continuous expansion of accession numbers. Core germplasm development has been proposed for better management and use of collections available in gene banks. This requires the development of a core set of accessions to more precisely characterize, explore, and conserve gene bank resources, monitor the genetic drift during preservation, and identify gaps in genetic diversity. Genetic diversity and population structure knowledge form the backbone in building core sets adequately representing variations found in the whole collection, and thereby making the collection small and condensed (Yan *et al.*, 2007; Agrama *et al.*, 2009; El Bakkali *et al.*, 2013).

India's east coast rice collections were characterized using SNP markers. The genetic diversity and population

structure were studied, and core and mini core collections with maximum diversity and minimum redundancy were developed. A total of 2,242 east coast rice accessions from three different states of India, i.e., Andhra Pradesh, Orissa, and Tamil Nadu, have been characterized, and a wide range of gene diversity and PIC was observed. A phylogenetic analysis of the total east coast rice collection revealed three groups, and a population structure analysis revealed four populations. The 36-SNP assay used in this study was validated by comparing the genetic diversity parameters (gene diversity, PIC, major allele frequency, and heterozygosity) across two different rice collections, i.e., east coastal rice and northeast rice collection, and it was observed these markers were sufficient to decipher all genetic parameters very efficiently; hence, they can be effectively utilized for core development and diversity study of different rice genotypes (Choudhury *et al.* 2021).

Identification of Germplasms with Superior Trait Value

The National Gene Bank at ICAR-NBPGR facilitates the conservation of genetic diversity by harboring the diverse plant genetic resources important for food and agriculture. These genetic resources constitute vast variability for several agronomically and economically important traits in respective crop plants. The conserved diversity and variability can cater the present requirements and also provide insurance for the future adverse conditions including biotic and abiotic stress (Paroda and Arora, 1991, Yadav *et al.*, 2018). In this perspective, it is crucial to pinpoint the desired trait specific accessions by the exercise of thorough characterization and comprehensive evaluation for specific traits. Identification of accessions with desired specific traits can be used as promising donors in breeding program and thereby facilitate enhanced utilization of the conserved germplasm. The NGB accessions of several crops have been evaluated for identification of accessions with desired traits. In wheat, 498 and 868 accessions have been identified potentially resistant for different wheat rusts and spot blotch, respectively from initial 19460 wheat accession (Kumar *et al.*, 2016). The characterization of entire NGB collection of barley (6,778 accessions) has helped identification of trait-specific accessions for agro-morphologically and economically important traits including days to spike emergence, days to maturity, plant height, spike length, number of grains per spike and hundred-grain weight (Kaur *et al.*, 2022). Among pulses, in chickpea, novel promising donors with high disease resistance to

Ascochyta blight (*Ascochyta rabiei*) have been identified by evaluation of chickpea germplasm accessions of NGB (Gayacharan *et al.*, 2020). The characterization of complete lentil germplasm collection of NGB over two years led to identification superior accessions for crucial traits such as early maturity, number of secondary branches, pods per plant and seed weight and suitability for mechanical harvesting (Tripathi *et al.*, 2022). In linseed germplasm accessions with superior trait values have been identified for flowering, maturity, test seed weight, seed area, capsules numbers per plant and plant height (Kaur *et al.*, 2018; Saroha *et al.*, 2022a). With increase in awareness about healthy diet among general public, a special emphasis should be given to evaluation of germplasm of NGB for various nutritional aspects to identify germplasm for high nutritional values like protein, fibre, important vitamins, mineral, lignans and with healthy fatty acid ratio and deficient in anti-nutritional factors. Recently, nutri-dense accessions have been identified in cowpea with high protein, total soluble sugar, amylose, and total dietary fibers from by nutritional profiling of 120 biochemically diverse cowpea accessions (Padhi *et al.*, 2022).

In recent years, under the mission projects funded by Department of Biotechnology at institutes of special expertise of ICAR, DBT and SAUs, the comprehensive phenotyping, genotyping and whole genome sequencing have been undertaken in important cereals, pulses and oilseeds. In these projects either complete germplasm accessions of respective crops or substantial number of accessions of crops like including rice, wheat, chickpea, minor pulses (cowpea, greengram, blackgram, mothbean and horse gram), sesame, safflower, linseed and niger are being characterized. These characterizations and evaluation under different biotic and abiotic stress condition and nutritional profiling are expected to unravel the diversity and variability for different traits in these crops and help development of core sets, reference sets for agro-morphological and nutritionally important traits. Further, such exercise would lead to identification of several trait specific accessions which can be utilized in breeding programme for crop improvement.

Employing Genome Wide Association Studies for Accelerated Utilization of Genebank Germplasm

Germplasm collection of Genebanks constitute genetic resources from diverse geographical areas and distinct populations which often show extensive phenotypic

variation for several traits. Such a collection of germplasm accessions is the treasure trove to understand the underlying genetic architecture for the traits in question. Plant scientists have traditionally used the biparental mapping for identification of quantitative (QTL)/genomic regions associated with the desired complex traits. However, this approach assays genetic variation limited to two parents and fewer recombination events in a biparental population. Genome-wide association study (GWAS) on the other hand assays a wide swathe of existing natural variation through population-scale samples and takes into account the historic recombination events across lineages, enabling a finer resolution of QTL (Burghardt *et al.*, 2017). Exploitation of such natural variation is more pertinent in the context of changing climatic conditions and to offer environmentally sustainable crop production.

In order to undertake a GWAS study, a precise phenotyping of large number of accessions for the desired traits is crucial. In case of field phenotyping, multi-location-season phenotyping would be beneficial to reduce the environmental error. Next, to have the phenotype-genotype association, it is important to perform genotyping of the same set of accessions at substantial genome coverage. Different genotyping approaches can be employed as per the crop species, available genomic resources and economics. More the available variants, higher are the chances to identify causative variants, therefore the best approach would be to have the complete genome sequencing of the accessions in the association panel (Gua *et al.*, 2019). Besides the whole genome sequencing, array-based SNP genotyping and reduced representation sequencing approaches have also been employed to get significant markers associated with the trait (Saroha *et al.*, 2022b, Vikas *et al.*, 2022).

Many researchers have preferred to constitute a subset of total germplasm collection (such as core set) as association mapping panel and identified significant QTLs for the complex traits (Soto-Cerda *et al.*, 2021). Once phenotype and genotyping of an AM panel is achieved, the next crucial step is application of statistical methods to identify marker trait association. There are several single locus models for example MLM, EMMAX, GEMMA and ECMLM are available for genetic dissection of traits. However, using these models it is essential to control the false positive rates by applying stringent corrections (Bonferroni corrections) which often result in

exclusion of important loci (Zhang *et al.*, 2019). Multi-locus methods have advantage over the single locus methods as it simultaneously tests multiple markers and thereby increases the statistical power while reducing type 1 error (Zhang *et al.* 2019). Once the statistically significant marker traits association is established, candidate genes for the traits can be selected either by direct identification of causative variant in the candidate gene or on the basis of the functional relevance of genes in the strong linkage disequilibrium (Burghardt *et al.*, 2017). Gene annotations of candidate genes, KEGG analysis and biological relevance have to be established to take the call for candidate genes. Further, validation of the candidate genes can be undertaken such as gene expression studies, experimental validation using RNAi, and genome editing methods including CRISPR/Cas9. Having established the association, the marker/gene can be used in breeding programmes. The illustration of application of GWAS in utilization of genetic resources of NGB is shown in Fig. 1. The similar approaches have been used by several researchers to identify QTLs/QTNs in different crops for agro-morphological traits as well as for biotic and abiotic stress (Pradhan *et al.*, 2020; Kumar *et al.*, 2020; Chaurasia *et al.*, 2021; Vikas *et al.*, 2022; Saroha *et al.*, 2022a). The GWAS approach can accelerate the pace of identification of gene/marker-trait association, genetic dissection of important traits and thereby utilization of germplasm collection of NGB in breeding programmes for varietal development.

References

- Agrama HA, Yan W, Lee F, Fjellstrom R, Chen M-H, Jia M, et al. (2009). Genetic Assessment of a Mini-Core Subset Developed from the USDA Rice Genebank. *Crop Sci.* **49**: 1336–1346. 10.2135/cropsci2008.06.0551
- Burghardt, L.T., Young, N.D., and Tiffin, P. 2017. A guide to genome-wide association mapping in plants. *Curr. Protoc. Plant Biol.* 2:22-38. doi: 10.1002/cppb.20041
- Chaurasia, S., Singh, A. K., Kumar, A., Songachan, L. S., Yadav, M. C., Kumar, S., et al. (2021). Genome-wide Association Mapping Reveals Key Genomic Regions for Physiological and Yield-Related Traits under Salinity Stress in Wheat (*Triticum aestivum* L.). *Genomics* 113 (5), 3198–3215. doi:10.1016/j.ygeno.2021.07.014
- Choudhury DR, Kumar R, S VD, Singh K, Singh NK, Singh R. Identification of a Diverse Core Set Panel of Rice From the East Coast Region of India Using SNP Markers. *Front Genet.* 2021 Nov 25;12:726152.
- Damania AB. 2008. History, Achievements, and Current Status of Genetic Resources Conservation. *Agron. J.* **100**: 9–21
- El Bakkali A., Haouane H., Moukhli A., Costes E., van Damme P., Khadari B. (2013). Construction of Core Collections Suitable for Association Mapping to Optimize Use of Mediterranean Olive (*Olea Europaea* L.) Genetic Resources. *PLoS One* 8, e61265. 10.1371/journal.pone.0061265
- Gayacharan, Rani U, Singh S, Basandrai AK, Rathee VK, Tripathi K, et al. (2020) Identification of novel resistant sources for ascochyta blight (*Ascochyta rabiei*) in chickpea. *PLoS ONE* **15**(10): e0240589. <https://doi.org/10.1371/journal.pone.0240589>
- Guo, D., Jiang, H., Yan, W., Yang, L., Ye, J., Wang, Y., Yan, Q., Chen, J., Gao, Y., Duan, L., et al. (2019). Resequencing 200 flax cultivated accessions identifies candidate genes related to seed size and weight and reveals signatures of artificial selection. *Front. Plant Sci.*, 10, 1682.
- Kaur V, Aravind J, Manju, Jacob SR, Kumari J, Panwar BS, Pal N, Rana JC, Pandey A and Kumar A (2022) Phenotypic Characterization, Genetic Diversity Assessment in 6,778 Accessions of Barley (*Hordeum vulgare* L. ssp. vulgare) Germplasm Conserved in National Genebank of India and Development of a Core Set. *Front. Plant Sci.* 13: 771920. doi: 10.3389/fpls.2022.771920
- Kaur V, S. Kumar, R. Yadav, D.P. Wankhede, J. Aravind, J. Radhamani, J.C. Rana and A. Kumar (2018) Analysis of genetic diversity in Indian and exotic linseed germplasm and identification of trait specific superior accessions. *J. environ. boil.* **39-5**: 702-709. DOI : <http://doi.org/10.22438/jeb/39/5/MRN-849>.
- Kumar S, Archak S, Tyagi RK, Kumar J, VK V, Jacob SR, et al. (2016) Evaluation of 19,460 Wheat Accessions Conserved in the Indian National Genebank to Identify New Sources of Resistance to Rust and Spot Blotch Diseases. *PLoS ONE* **11**(12): e0167702. doi:10.1371/journal.pone.0167702
- Padhi SR, Bartwal A, John R, Tripathi K, Gupta K, Wankhede DP, Mishra GP, Kumar S, Archak S and Bhardwaj R (2022) Evaluation and Multivariate Analysis of Cowpea [*Vigna unguiculata* (L.) Walp] Germplasm for Selected Nutrients—Mining for Nutri-Dense Accessions. *Front. Sustain. Food Syst.* 6:888041. doi: 10.3389/fsufs.2022.888041
- Paroda RS, Arora RK (1991) Plant genetic resources: general perspective (English) In: Plant genetic resources: conservation and management. Concepts and approaches/International Board for Plant Genetic Resources, New Delhi (India). Regional Office for South and Southeast Asia, 1991, 1-23
- Pradhan AK, Kumar S, Singh AK, Budhlakoti N, Mishra DC, Chauhan D, Mittal S, Grover M, Kumar S, Gangwar OP, Kumar S, Gupta A, Bhardwaj SC, Rai A and Singh K (2020) Identification of QTLs/Defense Genes Effective at Seedling Stage Against Prevailing Races of Wheat Stripe Rust in India. *Front. Genet.* 11:572975. doi: 10.3389/fgene.2020.572975
- Saroha A, Pal D, Gomashe SS, Akash, Kaur V, Ujjainwal S, Rajkumar S, Aravind J, Radhamani J, Kumar R, Chand D, Sengupta A and Wankhede DP (2022a) Identification of QTNs Associated with Flowering Time, Maturity, and Plant Height Traits in *Linum usitatissimum* L. Using Genome-

- Wide Association Study. *Front. Genet.* 13:811924. <https://doi.org/10.3389/fgene.2022.811924>.
- Sarooha, A., Pal, D., Kaur, V. Kumar S, Bartwal A, J. Aravind, J. Radhamani, S. Rajkumar, Kumar R, Gomashe SS, Sengupta A, Wankhede DP* (2022b) Agro-morphological variability and genetic diversity in linseed (*Linum usitatissimum* L.) germplasm accessions with emphasis on flowering and maturity time. *Genet Resour Crop Evo.* <https://doi.org/10.1007/s10722-021-01231-3>
- Soto-Cerda, B. J., Aravena, G., and Cloutier, S. (2021). Genetic Dissection of Flowering Time in Flax (*Linum usitatissimum* L.) through Single- and Multi-Locus Genome-wide Association Studies. *Mol. Genet. Genomics* **296**(4): 877–891. doi:10.1007/s00438-021-01785-y
- Tripathi K, Kumari J, Gore PG, Mishra DC, Singh AK, Mishra GP, C G, Dikshit HK, Singh N, Semwal DP, Mehra R, Bhardwaj R, Bansal R, Rana JC, Kumar A, Gupta V, Singh K and Sarker A (2022) Agro-Morphological Characterization of Lentil Germplasm of Indian National Genebank and Development of a Core Set for Efficient Utilization in Lentil Improvement Programs. *Front. Plant Sci.* 12:751429. doi: 10.3389/fpls.2021.751429
- Vikas, V.K., Pradhan, A.K., Budhlakoti, N. *et al.* Multi-locus genome-wide association studies (ML-GWAS) reveal novel genomic regions associated with seedling and adult plant stage leaf rust resistance in bread wheat (*Triticum aestivum* L.). *Heredity* **128**: 434–449 (2022). <https://doi.org/10.1038/s41437-022-00525-1>
- Yadav B, Kaur V, Narayan OP, Yadav SK, Kumar A and Wankhede DP (2022) Integrated omics approaches for flax improvement under abiotic and biotic stress: Current status and future prospects. *Front. Plant Sci.* 13:931275. <https://doi.org/10.3389/fpls.2022.931275>.
- Yadav S, Chet Ram, and Wankhede DP (2018) Biodiversity and Conservation of Plant Genetic Resources (*in-Situ* and *Ex-Situ*). In: Advanced Molecular Plant Breeding Meeting the Challenge of Food Security Eds. D. N. Bharadwaj. Apple Academic Press.
- Yan W., Rutger J. N., Bryant R. J., Bockelman H. E., Fjellstrom R. G., Chen M.-H., et al. (2007). Development and Evaluation of a Core Subset of the USDA rice Germplasm Collection. *Crop Sci.* 47, 869–876. 10.2135/cropsci2006.07.0444
- Zhang Y-M, Jia Z and Dunwell JM (2019) Editorial: The Applications of New Multi-Locus GWAS Methodologies in the Genetic Dissection of Complex Traits. *Front. Plant Sci.* **10**: 100. doi: 10.3389/fpls.2019.00100

Role of Bioinformatics in the Development of Plant Genetic Resources

Tanwy Dasmandal, Dwijesh Chandra Mishra* and Anil Rai

ICAR-Indian Agricultural Statistics Research Institute, Library Avenue, Pusa Campus, New Delhi-110012, India

Bioinformatics plays a significant role in the development of many fields of biological science and plant genetic resources (PGR) is one of them. With the advent of high throughput sequencing technology, bioinformatics continues to make considerable progress in biology by providing scientists with access to the genomic information. There are many areas of plant genetic resources such as development of core set, trait associated gene discovery, genetic diversity analysis, Genome Wide Association Studies (GWAS), phylogenetic and evolutionary analysis, database development and its management etc. where bioinformatics plays important roles. Main role of bioinformatics is to provide computational algorithm and software tools to accelerate the research of PGR. Bioinformatics is a new paradigm in the genomic research, which provides PGR research a great thrust. However, there are many areas such as pan genomics, multi locus GWAS, Genomic Selection with epistasis effects where bioinformatics can play better role.

Key Words: Bioinformatics, High-throughput Technology, PGR, GWAS, Core Set, Phylogenetic Analysis

Introduction

Plant genetic resources can be defined as all materials that are available for improvement of a cultivated plant species (Becker, 1993). A more effective use of plant genetic diversity is required to address the issues of development, food security, and poverty alleviation, according to the FAO's Global Plan of Action for the Conservation and Sustainable Utilization of Plant Genetic Resources (FAO, 1996b). For this purpose, extensive *ex situ* and *in situ* PGR conservation must be ensured, as well as evaluation of conserved accessions and their use by plant breeders needs to be facilitated. Through genetic advancement and the promotion of less common, neglected, or underutilized crop species, the goal should be to promote interspecific variety in agriculture as well as intraspecific variation within a crop. Modern technologies have paved the path towards fulfilment of these aims through the advanced sequencing technologies and the efficient amalgamation of information technologies with biological science or 'Bioinformatics'. With the generation of huge amount of biological data, bioinformatics has become almost an indispensable part for imparting meaning to the raw data through their analysis and visualization and also for easy maintenance and retrieval of the biological data. In the field of plant genetic resources, bioinformatics

has found its role in several areas like in developing germplasm core collection, gene discovery, genomic characterization, database creation and management and so on.

Role in Developing Germplasm Core Collection

Capturing haplotypes: Single nucleotide polymorphism (SNP) data sets from across the genome have enormous potential to enhance *ex situ* conservation. However, there are two issues that have been noticed regarding their use in the production of core collections. Firstly, due to the huge number of SNPs, it may be challenging to assemble the collections that will maximize diversity. To address this issue bioinformatics plays its role in developing computer program (like M+) for identifying optimized core collections from arbitrarily large genotypic data sets. MSTRAT is a popular program for producing genetically variable core collections.

Secondly, it is uncertain how the genome's natural partitioning into linked regions or haplotype blocks would affect the diversification and collection optimization. Large samples are necessary to determine haplotype block structure, and the process is methodologically complicated. However, bioinformatics can be used for simulating the basic structure of haplotype blocks using program like HAPLOTYPISTA.

*Author for Correspondence: Email-dwij.mishra@gmail.com

Genomics-based gene discovery: One of the main goals of germplasm research is gene discovery. In germplasm research using genomics, there are four approaches to finding new genes: map-based, association-based, allele mining-based, and comparative genomics-based. Among these methods bioinformatics has been extensively used for association based gene discovery using germplasm collection through GWAS analysis, and for comparative genomics based gene discovery using orthologous gene strategy where gene function and sequence have already been determined in a model (or other) species.

Graphical Tools for Germplasm Analysis

Bioinformatics has also been extensively used in developing graphical tools for germplasm analysis like GENE-MINE software. These tools have found applications in various fields of PGR studies such as-

- a. a geographical tool that could show the origin of accessions and the distribution of genetic diversity;
- b. a haplotype tool showing genotype information for accessions;
- c. tool for generating graphs that might display phylogenetic trees or networks (graphs containing closed loops, which can be used to represent genetic exchange between organisms) as well as pedigrees;
- d. tool that produces scatter plots of genetic marker distribution on relevant linkage maps, such as principal component analysis and diversity distances between pairs of accessions

Extraction of Functional Genetic Diversity from Heterogeneous Germplasm Collections for Crop Improvement

Large collection sizes, uneven trait characterization, and unpredictable distribution of allelic diversity across diverse accessions restrict efficient use of genetic variation in plant germplasm collections. Conventional and precision breeding might be streamlined by distributing compact subsets of the whole collection that include the largest amount of allelic variation at functional loci of interest.

In general, three bioinformatic approaches come into play to extract functional genetic diversity.

First, in a “candidate gene” approach, subsets that maximized haplotypic diversity are assembled.

Secondly, to find regulatory loci and assembled subsets representing genome-wide regulatory gene

diversity, a general source of phenotypic variation, one can do a keyword search against the Gene Ontology.

Thirdly, machine-learning approach can be developed to rank semantic similarity between Gene Ontology term definitions and the textual content of scientific publications on crop adaptation to stress, a complex breeding objective.

Role in Germplasm Characterization

Trait Mapping using GWAS

For germplasm managers, fundamental researchers, and plant breeders, the capacity to precisely detect and analyze genome-wide genetic variation or specific molecular variations through generations of individuals offers a potent tool. Thanks to the development of NGS, GWAS is presently a useful method to investigate allelic variation in a wider context for extensive phenotypic diversity and improved resolution of QTL mapping. Using GWAS, many research projects have been done to investigate the association between genetic variation and valuable plant traits. At the current stage, several bioinformatics approaches have been introduced as GWAS acceleration tools. Following are some examples: Heap which is a SNPs detection tool for NGS data with special reference to GWAS and BioGPU, a high-performance computing tool for GWAS. But, focusing on one main SNP that correlates with a specific phenotype as normal GWAS output may miss the key genetic variants with particular environment response in the context of complex traits. For this issue, bioinformatics approach is again a current solution. Generalize multifactor dimensionality reduction (GMDR) algorithm on a computing system with graphics processing units (GPUs) is one in some available methods at the moment that can screen potential candidate variants and then use the mixed liner model to detect the epistatic and gene-environment interactions.

Characterization on Climate Change Adaptation

Studying the transcriptome of populations using bioinformatic approaches growing along an environmental gradient may also reveal changes in gene expression of the same set of genes, thus potentially shedding light on the genetics of adaptation. Genomic information emanating from studies on climate change adaptation will support decision-making on what genetic resources to conserve in a gene bank in future. Genomics will also facilitate identification of novel alleles emerging because of climate change adaptation. These novel alleles, which

give plants the adaptive capacity, should be prioritized for conservation, as they are important in developing climate resilient crops.

Phylogenomics and Evolutionary Analysis

There are two important goals in phylogenomic research to accomplish. First is to discover the evolutionary patterns among plant species using nuclear genomic information. Second is to derive new hypothesis for the unknown function of plant genes associated to major divergence events in the evolution of plant species [95]. Bioinformatics have been extensively explored to develop methods and tools for performing plant phylogenomics like ExaML (Exascale Maximum Likelihood), MEGA, PHYLIP and so on.

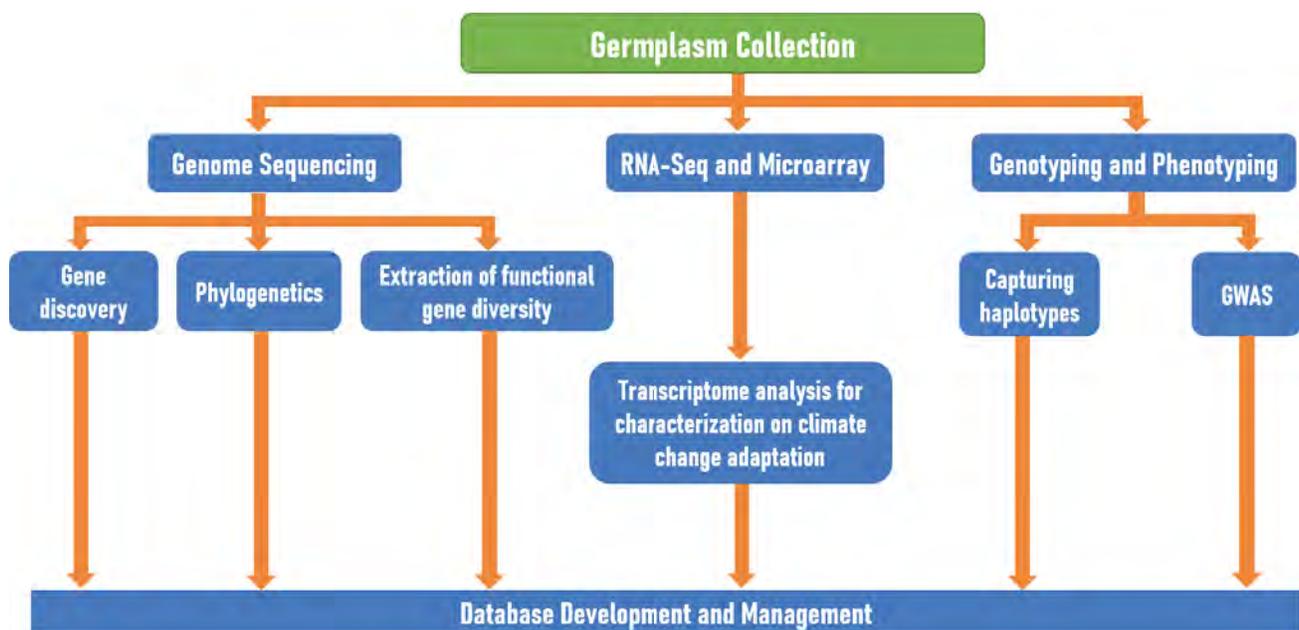
Role in Database Development and Management

To efficiently store and manage data, the use of state-of-the-art database management back-end infrastructure is indispensable. In this context well aligned binary data storage, Relational Database Management System (RDBMS), homogeneous designed data schemata, customized data import and user interfaces must be combined into an all-in-one back-end. Besides these back-end services, efficient data-sharing and data-access is a further challenging task for scientific data engineering. Data re-use and exchange is a key component to gain scientific knowledge in natural sciences [9] and particularly in plant science.

As for example, there are several database systems designed to manage this information for crop improvement; such as the integrated breeding platform (<https://www.integratedbreeding.net/>) and the Triticeae toolbox (<https://triticeaetoolbox.org>). GnpIS-Asso is another such example which is a generic database for managing and exploiting plant genetic association studies.

Future Prospects

- Pangenomics is the new age concept which needs to be explored for germplasm characterization. The pangenome refers to a collection of genomic sequence found in the entire species or population rather than in a single individual. So, instead of sequencing and analysing some selected germplasm, every germplasm in the collection is needed to be analysed and explored.
- Genomic selection is an area of bioinformatics which has been merely exploited in crop science. Genomic selection is an advance form of marker assisted selection which has the potential to reduce the breeding cycle and thus increasing the genetic gain in crops. This technique can extensively be used for crop improvement programs.
- Till date GWAS is limited to single locus study which can explain only a little part of the genetic variance associated with the phenotype. Thus, GWAS studies needs to be extended to multi locus studies which



Role of Bioinformatics in PGR Studies

can capture majority of the genetic variances and can also detect the epistatic interactions as well.

- With the changing time, the demand of agriculture is constantly changing. Unlike previous demands of increasing yields and other quantitative traits in crops, recent era focuses more on qualitative enrichments of crops like nutrient content, aroma etc. And to keep pace with these changing demands bioinformatics has to be accepted as an indispensable part of crop improvement. Bioinformatics analysis of qualitative traits need to be carried out on a large scale on all major as well as minor crop species.
- Role of Bioinformatics in PGR has been largely applied in areas of genomics so far with very little attention paid to other areas of omics like transcriptomics, proteomics and metabolomics

which can give better insights of crop genetics and breeding.

References

- Jia, J., Li, H., Zhang, X., Li, Z., & Qiu, L. (2017). Genomics-based plant germplasm research (GPGR). *Crop J.* **5(2)**: 166-174.
- Reeves, P.A., Tetreault, H.M., & Richards, C.M. (2020). Bioinformatic extraction of functional genetic diversity from heterogeneous germplasm collections for crop improvement. *Agronomy*, **10(4)**: 593.
- Davenport, G., Ellis, N., Ambrose, M., & Dicks, J. (2004). Using bioinformatics to analyse germplasm collections. *Euphytica*, **137(1)**: 39-54.
- Reeves, P.A., & Richards, C.M. (2017). Capturing haplotypes in germplasm core collections using bioinformatics. *Genet. Resour. Crop Evol.* **64(8)**: 1821-1828.
- Ong, Q., Nguyen, P., Phuong Thao, N., & Le, L. (2016). Bioinformatics approach in plant genomic research. *Current genomics*, **17(4)**: 368-378.

Genomic Resource Generation in Medicinal and Aromatic Plants

Rakesh Singh^{1,*} and Ramesh Kumar¹

¹Division of Genomic Resources, ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012

Plant genomic resources are genetic material of actual or potential value which can be utilized for the improvement of specific traits in Agri-horticultural crops. The generation of genomic resource in medicinal plants is important because they contain bioactive compounds or secondary metabolites important for human health. The demand for these compounds is increasing due to their application in herbal medicine. The recent “omics” techniques have made generation of genomic resources much efficient and cost effective. The improvement in sequencing technology from 2nd generation (NGS) to 3rd generation has reduced sequencing cost and thus brought many more crop genomes within range of analysis. The Next Generation Sequencing (NGS) based whole genome and transcriptome sequencing in medicinal and aromatic plant has played a vital role in generating genomic resources for effective conservation, crop improvement and better understanding about secondary metabolite biosynthesis in medicinal and aromatic plants. In present review, the progress of generating genomic resources such SSR resources, EST-SSR resources, transcription factors, transcriptome analysis, and whole genome sequence analysis in selected medicinal and aromatic plants has been updated, which may be further utilize in medicinal and aromatic plant improvement programs.

Key Words: Aromatic plants, Genomic resource, Medicinal plants, Molecular markers
Transcriptome, Whole genome sequencing

Introduction

Medicinal and aromatic plants are very important because they are rich sources of secondary metabolites or bioactive compounds required for production of herbal medicines. The affordability, availability, compatibility, and acceptability of medicinal plants have made them an important element in the primary health care. Over 70% of the population of developing countries relies upon medicinal plants for their treatment and primary care (Jeelani *et al.*, 2018). Medicinal plants have been used for centuries to treat and prevent different diseases. Different secondary metabolites or bioactive compounds derived from the medicinal plants used for producing medicines due to diverse medicinal properties such as anti-inflammatory, immunomodulatory, anticancer, cardiovascular, antimalarial, and antimicrobial.

The present review focuses on four important medicinal and aromatic plants, *Tinospora cardifolia* (Giloe), *Andrographis paniculata* (Kalmegh), *Vetiveria zizanioides* (Vetiver grass), and *Bunium persicum* (Kala jeera). The *Tinospora cordifolia* is a deciduous shrub, belongs to Menispermaceae family (Spandana *et al.*, 2013). In the Ayurveda, this plant is recorded as having bitter, pungent, and astringent tastes (Raghu

et al., 2006). *T. cordifolia* has been reported to have various important medicinal properties *viz.*, anti-oxidant, anti-hyperglycaemic, anti-stress agent, anti-carcinogenic, anti-spasmodic, anti-allergic, anti-leprotic, immunomodulator, anti-microbial (Jeyachandran *et al.*, 2003; Kalikar *et al.*, 2008; Khan *et al.*, 2020; Singh *et al.*, 2003; Asthana *et al.*, 2001; Desai *et al.*, 2002; Rajalakshmi *et al.*, 2009; Ahmad *et al.*, 2015). The *Andrographis paniculata*, belongs to Acanthaceae family, and commonly known as chireta (Chandrasekaran *et al.*, 2009). The plant contains a diterpenoid andrographolide which is bitter in taste, and responsible for the therapeutic interest of the plant. The several pharmacological activities of the plant has been reported such as cytotoxicity, antioxidant, antimicrobial, anti-inflammatory, immune-stimulant, antidiabetic, anti-infective, anti-angiogenic, hepato-renal protective, and insecticidal activities (Okhuarobo *et al.*, 2014). *Vetiveria zizanioides* L. Nash, which is a perennial grass, commonly referred as Khus, and belongs to the Poaceae family. The roots of the plant produce a fragrant and volatile oil content that is in high demand in the perfumery, and cosmetic industries (Sethi *et al.*, 1968). *Bunium persicum* (Boiss.) Fedtsch., commonly known as Kala

*Author for Correspondence: Email-rakesh.singh2@icar.gov.in

jeera, is an important aromatic and medicinal plant from Apiaceae family, grows mainly in cold temperate regions of Central Asia and Northern India. Due to the high amount of aroma and essential oil present in the plant, *Kala jeera* is industrially important.

The advancements in genomic technologies have made generation of genomic resources in medicinal and aromatic plants easy and also to improve the desired traits or secondary metabolites production. Genomic resources such as genomic SSR (Simple sequences Repeats), ESTs (Expressed sequence tags), transcription factors and small RNA etc has been generated using technologies such as transcriptome and whole genome sequencing in some medicinal plants (Singh *et al.*, 2014; Singh *et al.*, 2016; Sun *et al.*, 2019; Kumar *et al.*, 2020; Bansal *et al.*, 2022).

1. Approaches Used to Generate Genomic Resources

Crop improvement goals are shifting toward a trait-oriented approach as agriculture becomes more specialised and location-specific. To achieve these goals, it is crucial to both conserve and make use of the genetic diversity that is already present. Generating genomic resources can significantly improve the use of PGRs (plant genetic resources). Due to omics techniques, the development of genomic resources is now possible in less time and in cost effective manner. Few of the genomic approaches which are being used for the generation of genomic resources in medicinal and aromatic plants (Fig. 1) are discussed below:

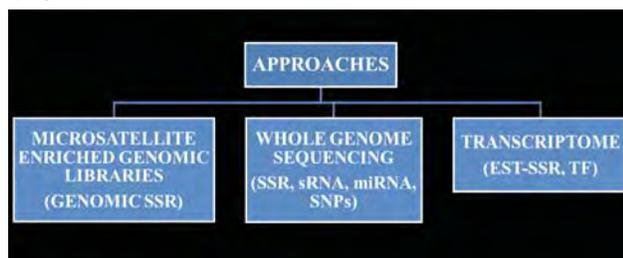


Fig.1. Approaches commonly used for generation of genomic resources in medicinal and aromatic plants.

1.1 Microsatellite Enriched Library

In this approach, the microsatellite containing the DNA region of the genome is hybridized using microsatellite repeat specific probes, the genomic DNA is fragmented/digested by either restriction digestion or sonication (Kandpal *et al.*, 1994; Edwards *et al.*, 1996; Fischer and

Bachmann, 1998). This is relatively simple, robust, low cost, and reproducible in comparison to other methods. The method has been used to generate genomic SSRs in medicinal plants such as *A. paniculata*, and *T. cordifolia* (Kumar *et al.*, 2020, Paliwal *et al.*, 2016).

1.2 Transcriptome Sequencing

RNA sequencing (RNA-seq) based on next-generation sequencing (NGS) platform, enable the simultaneous acquisition of sequences for both gene discovery and transcript identification relevant to biological processes. This approach is appropriate for those organisms for which genomic sequences information's are not available (Ward *et al.*, 2012). In recent years, de novo transcriptome has appeared as a powerful technique to identify genes involved in the biosynthesis of different secondary metabolites of medicinal plants (Huang *et al.*, 2012; Hyun *et al.*, 2012; Singh *et al.*, 2016).

1.3 Whole Genome Sequencing

The ability to sequence an organism's entire genome with new NGS technology at a lower cost and in less time has become one of the key discoveries in the field of "omics," even though "Sanger sequencing" has remained the standard for decoding genomes for several decades. Earlier, even sequencing a small genome would have required a multi-institutional collaborative effort and substantial funding. The advancement of NGS technologies has greatly increased the cost-effectiveness, speed, and efficiency of genome sequencing. The genome sequencing of some medicinal plants such as *A. paniculata*, *Ocimum tenuiflorum*, and *Artemisia annua* is available using NGS platform (Upadhyay *et al.*, 2015; Shen *et al.*, 2018; Sun *et al.*, 2019).

1.4 Genome-wide Association Studies (GWAS)

Genome-wide association studies (GWAS) have become a preferred method due to ongoing advancements in sequencing technologies and concerted community effort, especially when resequencing is carried out after the assembly of the reference genome or when a high-density genotyping array is made available (Michael and Jackson, 2013). This approach has allowed to find the genomic variations linked with either molecular or biochemical phenotype, and traditional agronomic phenotypes. These associations could be used to accelerate the crop improvement programs. The genome wide study has been done in *Matricaria recutita*, a medicinal plant (Otto *et al.*, 2017).

1.5 Small RNA

Small RNA, cis acting regulatory elements and intergenic regions which are part of intron region (non-genic region), also gaining the importance as genomic resources. Small RNAs play an important role in stress management in plants. The small RNAs has been discovered in medicinal plants such as *Panax ginseng*, *Dendrobium huoshanense* (Wu *et al.*, 2012; Wang *et al.*, 2022).

1.6 Single Nucleotide Polymorphism (SNP)

Identification of allele variations in PGRs, which can be obtained by highly reliable DNA-based markers such as SNPs. SNP provides better potentials for studying PGRs management in several ways, including cultivar identification, genetic diversity assessment, genetic map construction, and marker assisted breeding (Ganal *et al.*, 2009). This is because the SNP is more readily available and stable during inheritance than other markers, such as SSRs. The SNPs has been reported in medicinal plants such as *M. recutita*, and *Crepidiastrum denticulatum* (Otto *et al.*, 2017; Dang *et al.*, 2019).

2. Genomic Resources Generated in Medicinal and Aromatic Plants

2.1 SSR Markers Generation through Enriched Genomic Library

SSRs are also known as microsatellites, which are short tandem repeats of nucleotides (1-10) and distributed throughout the genome. Due to codominant in nature, multi allelic, high reproducibility and cross transferability, the SSR markers are one of the choicest marker system for genotyping, population structure assessment, varietal identification, association mapping etc. (Kalia *et al.*, 2011). Paliwal *et al.*; 2016, generated microsatellite markers in *T. cordifolida* with the help of SSR enriched genomic libraries. The genomic libraries of (CT)₁₄, (GT)₁₂, (AC)₁₀, and (AAC)₈ repeats were developed, which were used to generate 90 microsatellite sequences. These g- SSR markers were validated and used for genetic diversity studies in 26 accessions of *T. cordifolia* and one each accession of *T. sinensis* and *T. rumphii*. The markers were found efficient for genetic diversity analysis as well as cross transferability of more than 80% SSR markers was also reported in related species of *Tinospora* (*T. rumphii*, and *T. sinensis*). Kumar *et al.*, 2020, developed SSR markers using SSR genomic libraries enrichment in *A. paniculata* and validated through genetic diversity analysis. Four

types of SSR enriched genomic libraries such as (CT)₁₄, (AG)₁₅, (GT)₁₂, and (AAC)₈ were used to generate 67 genomic SSR markers. The 41 SSR markers were found polymorphic and efficient for genetic diversity analysis. The developed genomic SSR markers could be an important genomic resource for crop improvement programs of *A. paniculata*. Singh *et al.*, 2014, reported genetic diversity and cross genera SSR transferability in *Vetiveria zizanioides* L. Nash by transferring rice hyper variable SSRs markers (HvSSR), out of 120 HvSSR markers studied, 36 showed cross genera transferability. The across genera transferred SSR markers of rice could be an important genomic resource vetiver germplasm improvement programme.

2.2 EST-SSR and Transcription Factor Generation through Transcriptome

In the last ten years, RNA-seq has emerged as the preferred platform for transcriptome analysis and has been widely used to obtain mass sequence data for gene discovery, generation of molecular markers, and transcriptional analysis in a variety of plants. Researchers can analyse functional genes and regulatory mechanisms of medicinal and aromatic plants with the aid of transcriptomics research, which can also help them refine breeding selection and cultivation methods. The transcriptome data can be used to monitor the transcriptional activity of any plant species without reference genome. Singh *et al.*, 2016, generated transcriptome sequence of *T. cordifolia* using 454 GS-FLX pyrosequencing. Identified 4,538 transcripts showing significant similarity with corresponding orthologs were categorized into 58 different transcription factor families. The highest member (457) of basic loop helix (bHLH) transcription family was identified, followed by MYB (295) and NAC (280). Among the assembled transcripts, 5,412 SSR loci consisting of mono- to hexa- nucleotide repeats and also complex motif were identified. A total of 96 EST-SSR were validated and used for genetic diversity analysis among 24 accessions of *T. cordifolia*, which indicated these markers were polymorphic and highly reproducible and can be utilized as important genomic resource.

2.3 Genome Wide SSR Marker Generation through Whole Genome Sequencing

Whole genome sequencing and its de novo assembly could be another approach for the generation of genomic resources in non-model plants. In case of *Bunium*

persicum whole genome sequencing was done using Illumina HiSeq X Ten sequencer. Since no reference genome was available therefore de novo assembly was done. A total of 1,77,029 perfect and 5915 compound SSR motifs were identified in 2,12,585 assembled sequences (Bansal *et al.*, 2022). Total 88 SSR primers were used for their validation and genetic diversity analysis among 25 accessions of *B. persicum*. The genome wide SSRs markers developed in *B. persicum*

will open new avenues for characterizing genotypes and to develop future conservation strategies for *B. persicum*.

The above three approaches have been used by different researchers for the generation of genomic resources in different medicinal and aromatic plants. A comprehensive information about availability of genomic resources in different medicinal and aromatic plants has been summarized in Table 1.

Table 1: The available genomic resources in medicinal and aromatic plants

S. No.	Medicinal plant species	Available genomic resources	References
1	<i>Aconitum carmichaelii</i>	Transcriptome	(Rai <i>et al.</i> , 2017b)
2	<i>Andrographis paniculata</i>	Genome, Transcriptome, g-SSRs (67), EST-SSR (32,341), NAC Transcription factors (2), WRKY Transcription Factor (58)	(Cherukupalli <i>et al.</i> , 2016; Wang <i>et al.</i> , 2017; Kumar <i>et al.</i> , 2020; Zhang <i>et al.</i> , 2021; Kumar <i>et al.</i> , 2022)
3	<i>Artemisia annua</i>	Genome, ESR-SSR (2110), NAC Transcription factor (28)	(Wang <i>et al.</i> 2012; Shen <i>et al.</i> , 2018; Kumar <i>et al.</i> , 2021)
4	<i>Bacopa monnieri</i>	Transcriptome, MYB35	(Jeena <i>et al.</i> , 2017, 2021)
5	<i>Bunium persicum</i>	g-SSRs (177029)	(Bansal <i>et al.</i> , 2022)
6	<i>Bupleurum chinense</i>	g-SSRs (19), EST-SSRs (44)	(Sui <i>et al.</i> , 2009)
7	<i>Camptotheca acuminata</i>	Genome, transcriptome	(Sun <i>et al.</i> , 2011; Zhao <i>et al.</i> 2017)
8	<i>Cannabis sativa</i>	Genome, transcriptome	(Bakel <i>et al.</i> , 2011)
9	<i>Catharanthus roseus</i>	Genome, EST-SSRs (2034), genomic-SSR (314)	(Mishra <i>et al.</i> , 2011; Shokeen <i>et al.</i> , 2011; Kellner <i>et al.</i> , 2015)
10	<i>Chrysanthemum morifolium</i>	EST-SSR (218)	(Feng <i>et al.</i> , 2016)
11	<i>Docynia delavayi</i>	EST-SSR (18)	(Peng <i>et al.</i> 2021)
12	<i>Glycyrrhiza uralensis</i>	Genome, Transcriptome, EST-SSR (7032),	(Liu <i>et al.</i> , 2015; Mochida <i>et al.</i> , 2016)
13	<i>Hippophae rhamnoides</i>	EST-SSR (30)	(Jain <i>et al.</i> , 2010)
14	<i>Lancea tibetica</i>	g-SSR (4441)	(Tian <i>et al.</i> , 2016)
15	<i>Lonicera japonica</i>	Transcriptome	(Rai <i>et al.</i> , 2017a)
16	<i>Nicotiana tabacum</i>	Genome, g-SSRs (1365), EST-SSRs (3521) NAC Transcription factor (280)	(Sierro <i>et al.</i> 2014; Tong <i>et al.</i> , 2012; Kumar <i>et al.</i> , 2021)
17	<i>Ocimum tenuiflorum</i>	Genome, ESR-SSR (471) NAC Transcription factors (110)	(Upadhyay <i>et al.</i> , 2015; Kumar <i>et al.</i> , 2021)
18	<i>Ophiorrhiza pumila</i>	Transcriptome, WRKY transcription factor (46)	(Yamazaki <i>et al.</i> , 2013; Wang <i>et al.</i> , 2022a)
19	<i>Paeonia suffruticosa</i>	EST-SSR (4,373)	(Wu <i>et al.</i> , 2014)
20	<i>Panax ginseng</i>	Genome, Transcriptome	(Li <i>et al.</i> , 2013; Xu <i>et al.</i> , 2017)
21	<i>Panax japonicus</i>	Transcriptome	(Rai <i>et al.</i> , 2016b)
22	<i>Papaver somniferum</i>	Genome, Transcriptome, EST-SSR (14957)	(Desgagné-Penix <i>et al.</i> , 2010; Winzer <i>et al.</i> , 2012; Şelale <i>et al.</i> , 2013; Pei <i>et al.</i> , 2021)
23	<i>Perilla frutescens</i>	Transcriptome	(Fukushima <i>et al.</i> , 2015)
24	<i>Physalis alkekengi</i>	Transcriptome	(Fukushima <i>et al.</i> , 2016)
25	<i>Pueraria lobata</i>	Transcriptome, g-SSR (20)	(Han <i>et al.</i> , 2015; Zhou <i>et al.</i> , 2019)
26	<i>Sarcandra glabra</i>	EST-SSR (25,620), SNP (726,476)	(Xu <i>et al.</i> , 2021)
27	<i>Swertia japonica</i>	Transcriptome	(Rai <i>et al.</i> , 2016a)
28	<i>Tinospora cordifolia</i>	genomic-SSR (90), EST-SSR (25406)	(Paliwal <i>et al.</i> , 2016; Singh <i>et al.</i> , 2016)
29	<i>Trachyspermum ammi</i>	Transcriptome, NAC Transcription factor (68)	(Howyzeh <i>et al.</i> , 2018; Kumar <i>et al.</i> , 2021)
30	<i>Trifolium pratense</i>	Genome, NAC Transcription factor (97)	(Vega <i>et al.</i> 2015; Chao <i>et al.</i> , 2018; Kumar <i>et al.</i> , 2021)
31	<i>Veratrum baillonii</i>	Transcriptome, EST-SSR (40885)	(Wang <i>et al.</i> , 2015)
32	<i>Vetiveria zizanioides</i>	Transcriptome	(Chakrabarty <i>et al.</i> , 2015)
33	<i>Withania somnifera</i>	Transcriptome, EST-SSR (729), AP2/ERF (187)	(Gupta <i>et al.</i> , 2013; Tripathi <i>et al.</i> , 2020)
34	<i>Zingiber officinale</i>	EST-SSR (16,790)	(Vidya <i>et al.</i> , 2021)

2.4 Medicinal Plants Database

Database is a collection of data that is organized for simple access, management, and updating. The genomic resource generated from transcriptome studies were uploaded for public use in the form of user-friendly database. Two medicinal plant genomic resource databases developed by ICAR-National Bureau of

Plant Genetic Resources (NBPGR), New Delhi, one is TinoTranscriptDB and another is ApTransDB. TinoTranscriptDB (<http://www.nbpgr.ernet.in:8080/Tinospora/>) and ApTransDB (<http://www.nbpgr.ernet.in:8080/Andrographis/About.aspx>), are publicly available database of transcripts and SSRs of *T. cordifolia*, and *A. paniculata*, respectively (Fig. 2). Both the database

TinoTranscriptDB
Tinospora cordifolia Transcripts & SSR Database

About Database BLAST Tutorial Download Feedback Links Team Contact Us

Tinospora cordifolia (Willd.) Miers. (Giloee) plant is well known for its anti-spasmodic, anti-inflammatory, antipyretic properties and is being used in the treatment of gout, jaundice and rheumatism. *Tinospora cordifolia* is distributed throughout tropical Indian subcontinent and China. It is a shrub with large, glabrous, deciduous structure belongs to the family Menispermaceae.

The public databases and literature survey revealed very limited information with regard to genomic resources essential for judicious exploitation of *Tinospora cordifolia*. To this end, we have generated its entire transcript based database incorporating information of gene sequences and gene-based SSR (EST-SSR) or microsatellite markers. SSR marker is very useful tool because of its co-dominant inheritance, multi-allelic variation and high reproducibility. Since EST-SSRs markers, designed in this database is derived from transcript sequences, it can have various applications in the field of genomics as well as understanding functional diversity of allelic variants, if any. Moreover, it is equally useful for the generation of high density marker based genetic maps for gene/QTL discovery, phylogenetic studies and comparative genomics.

Copyright © Reg. No. SW-15428/2022 ICAR-National Bureau of Plant Genetic Resources, New Delhi, India
This work is supported by Indian Council for Agricultural Research, Government of India, New Delhi

ApTransDB
Andrographis Transcripts & SSR Database

About Database Tutorial Downloads Feedback Links Team Contact Us

Andrographis paniculata is a herb well known in Asia and belongs to the family *Acanthaceae*. It is native to Asia and distributed throughout India, China, Sri Lanka and Taiwan. *Andrographis paniculata* is a potential medicinal plant of immense economic importance. Leaves of *A. paniculata* contains active constituents like andrographolide, homoandrographolide, andrographestrol and andrographonone. *A. paniculata*, also known as *Kalmegh* in India, is mentioned in ancient Indian medicine (Ayurvedic) literature as a constituent of ayurvedic formulation used for the treatment of liver disorder, and neoplasm. The plant has also hepatoprotective, antioxidant, and hypoglycemic activities and is being used in the management of immune system and neoplastic diseases.

Due to limited information with regard to genomic resources, essential for exploitation of *Andrographis paniculata*, we have generated a database from its entire transcriptome including information relevant to gene sequences and gene-based SSR (EST-SSR) or microsatellite markers. SSR marker is an indispensable tool for molecular mapping, diversity analysis and phylogenetic studies. Moreover, since SSR were designed from transcripts sequences, it is equally useful for comparative genomics including candidate gene determination. Besides molecular markers, the sequences of transcripts will serves as an important resource for the discovery of biosynthetic genes and other regulons associated with its active constituents.

Copyright © ICAR- National Bureau of Plant Genetic Resources, New Delhi, India
This work is supported by Indian Council for Agricultural Research, Government of India, New Delhi

Fig. 2. Database of microsatellite markers generated from transcriptome *Tinospora cordifolia* (Tino TranscriptDB) and *Andrographis paniculata* (Andro TranscriptDB)

Table 2. Genomic resource databases of medicinal and aromatic plants.

S.No.	Name of database	Description	URL
1	TinoTranscriptDB	Transcripts and SSR database of <i>Tinospora cordifolia</i>	http://www.nbprg.ernet.in:8080/Tinospora/
2	ApTransDB	Transcripts and SSR database of <i>Andrographis paniculata</i>	http://www.nbprg.ernet.in:8080/Andrographis/About.aspx
3	croFGD	Catharanthus roseus functional genomics database	http://bioinformatics.cau.edu.cn/croFGD/
4	TCMPG	Integrative database for traditional Chinese medicine plant genome	http://cbcb.cdutem.edu.cn/TCMPG/
5	MPGR	Medicinal plant genomic resources database	http://mpgr.uga.edu/

provides the information of SSR, (EST Expressed sequence tags)-SSR, transcription factor categories, and GO categories, and gene sequences. The genomic information provided can be further utilized to discover the candidate genes related to secondary metabolite biosynthesis through comparative genomics. The different public databases available in case of medicinal and aromatic plants are given in Table 2.

3. Conclusions

Genomic resources such as molecular markers, genes, and transcription factors related to the biosynthesis of bioactive compounds or secondary metabolites are important tools that can be utilized for increasing the production of these compounds. Since very limited genomic resources has been generated in case of medicinal plants, therefore there is need to develop more resources so that, obstacles for crop improvement programs of medicinal and aromatic plants can be addressed effectively.

Acknowledgements

The authors express their gratitude to the Director, ICAR-NBPGR New Delhi for providing facilities.

References

- Ahmad R, AN Srivastava, M A Khan, and A Khan (2015) Evaluation of in vitro anticancer activity of stem of *Tinospora cordifolia* against human breast cancer and Vero cell lines. *J. Med. Plants Stud.* **3**:33–37.
- Asthana JG, S Jain, Ashutosh Mishra, and MS Vijay Kanth, (2001). Evaluation of antileprosy herbal drug combinations and their combinations with dapsone. *Indian Drugs* **38**:82–86.
- Bakel V, H Van Bakel, JM Stout, AG Cote, CM Tallon, AG Sharpe, TR Hughes, JE Page (2011) The draft genome and transcriptome of *Cannabis sativa*. *Genome Biol.* **12**:1–18.
- Bansal S, A Kumar, AA Lone, MH Khan, EV Malhotra, and R Singh (2022) Development of novel genome-wide simple sequence repeats (SSR) markers in *Bunium persicum*. *Ind. Crops Prod.* **178**:114625.
- Chakrabarty D, PS Chauhan, AS Chauhan, Y Indoliya, UCLavania, CS Nautiyal (2015) De novo assembly and characterization

of root transcriptome in two distinct morphotypes of vetiver, *Chrysopogon zizanioides* (L.) Roberty. *Sci. Rep.* **5**(1):1–3.

- Chandrasekaran CV, P Thiyagarajan, K Sundarajan, KS Goudar, M Deepak, B Murali, et al. (2009) Evaluation of the genotoxic potential and acute oral toxicity of standardized extract of *Andrographis paniculata* (KalmCold™). *Food Chem. Toxicol.* **47**:1892–1902.
- Chao Y, L Xie, J Yuan, T Guo, Y Li, F Liu, et al. (2018). Transcriptome analysis of leaf senescence in red clover (*Trifolium pratense* L.). *Physiol. Mol. Biol. Plants* **24**:753–765.
- Cherukupalli N, M Divate, SR Mittapelli, VR Khareedu, DR Vudem (2016) De novo assembly of leaf transcriptome in the medicinal plant *Andrographis paniculata*. *Front. Plant Sci.* **7**:1203.
- Dang H, K Do, J Jung, J Hyun, S Jeong, Y Chaejin, et al. (2019) The newly developed single nucleotide polymorphism (SNP) markers for a potentially medicinal plant, *Crepidiastrum denticulatum* (Asteraceae), inferred from complete chloroplast genome data. *Mol. Biol. Rep.* **46**:3287–3297.
- Desai VR, JP Kamat, KB Sainis (2002) An immunomodulator from *Tinospora cordifolia* with antioxidant activity in cell-free systems. *Proc. Indian Acad. Sci. Chem. Sci.* **114**:713–719.
- Desgagné-Penix I, MF Khan, DC Schriemer, D Cram, J Nowak, PJ Facchini (2010) Integration of deep transcriptome and proteome analyses reveals the components of alkaloid metabolism in opium poppy cell cultures. *BMC Plant Biol.* **10**:252.
- Edwards KJ, JHA Barker, A Daly, C Jones, A Karp (1996) Microsatellite libraries enriched for several microsatellite sequences in plants. *Biotechniques* **20**:758–760.
- Feng S, R He, J Lu, M Jiang, X Shen, Y Jiang et al. (2016). Development of SSR markers and assessment of genetic diversity in medicinal *Chrysanthemum morifolium* cultivars. *Front. Genet.* **7**: 113.
- Fischer D, and K Bachmann (1998) Microsatellite enrichment in organisms with large genomes (*Allium cepa* L.). *Biotechniques* **24**:796–802.
- Fukushima A, M Nakamura, H Suzuki, K Saito, and M Yamazaki (2015) High-Throughput Sequencing and De Novo Assembly of Red and Green Forms of the *Perilla frutescens* var. *crispa* Transcriptome. *PLoS One* **10**:e0129154.
- Fukushima A, M Nakamura, H Suzuki, M Yamazaki, E Knoch, T Mori, N Umemoto, M Morita G Hirai, M Sodeoka, K Saito (2016). Comparative characterization of the leaf tissue

- of *Physalis alkekengi* and *physalis peruviana* using RNA-seq and metabolite profiling. *Front. Plant Sci.* **7**:1883.
- Ganal MW, T Altmann, and MS Röder (2009) SNP identification in crop plants. *Curr. Opin. Plant Biol.* **12**: 211–217.
- Gupta P, R Goel, S Pathak, A Srivastava, SP Singh, RS Sangwan, MH Asif, PK Trivedi (2013) De Novo Assembly, Functional Annotation and Comparative Analysis of *Withania somnifera* Leaf and Root Transcriptomes to Identify Putative Genes Involved in the Withanolides Biosynthesis. *PLoS One* **8(5)**:e62714.
- Han R, H Takahashi, M Nakamura, N Yoshimoto, H Suzuki, D Shibata, M Yamazaki, K Saito (2015) Transcriptomic landscape of *Pueraria lobata* demonstrates potential for phytochemical study. *Front. Plant Sci.* **6**:1–10.
- Howyze MS, S Ahmad, SA Noori, S and JV Shariati, M Amiripour (2018) Comparative transcriptome analysis to identify putative genes involved in thymol biosynthesis pathway in medicinal plant *Trachyspermum ammi* L. *Sci. Rep.* **8(1)**:1–9.
- Huang HH, LL Xu, ZK Tong, EP Lin, QP Liu, LJ Cheng, et al. (2012) De novo characterization of the Chinese fir (*Cunninghamia lanceolata*) transcriptome and analysis of candidate genes involved in cellulose and lignin biosynthesis. *BMC Genomics* **13**.
- Hyun TK, Y Rim, HJ Jang, CH Kim, J Park, R Kumar, et al. (2012) De novo transcriptome sequencing of *Momordica cochinchinensis* to identify genes involved in the carotenoid biosynthesis. *Plant Mol. Biol.* **79**: 413–427.
- Jain A, R Ghangal, A Grover, S Raghuvanshi, and PC Sharma (2010) Development of EST-based new SSR markers in seabuckthorn. *Physiol. Mol. Biol. Plants* **16**:375–378.
- Jeelani SM, GA Rather, A Sharma, and SK Lattoo (2018) In perspective: Potential medicinal plant resources of Kashmir Himalayas, their domestication and cultivation for commercial exploitation. *J. Appl. Res. Med. Aromat. Plants*, **8**: 10–25.
- Jeena GS, S Fatima, P Tripathi, S Upadhyay, and RK Shukla (2017) Comparative transcriptome analysis of shoot and root tissue of *Bacopa monnieri* identifies potential genes related to triterpenoid saponin biosynthesis. *BMC Genomics* **18**: 1–15.
- Jeena GS, S Kumar, and RK Shukla (2021) Characterization of MYB35 regulated methyl jasmonate and wound responsive Geraniol 10-hydroxylase-1 gene from *Bacopa monnieri*. *Planta* **253**:1–13.
- Jeyachandran R, TF Xavier, and SP Anand (2003) Antibacterial activity of stem extracts of *Tinospora cordifolia* (Willd) Hook. f & Thomson. *Anc. Sci. Life*, **23(1)**:40.
- Kalia RK, MK Rai, S Kalia, R Singh, and AK Dhawan (2011) Microsatellite markers: An overview of the recent progress in plants. *Euphytica* **177**:309–334.
- Kaliker MV, VR Thawani, UK Varadpande, SD Sontakke, RP Singh, and RK Khiyani, (2008) Immunomodulatory effect of *Tinospora cordifolia* extract in human immuno-deficiency virus positive patients. *Indian J. Pharmacol.* **40(3)**:107.
- Kandpal RP, G Kandpal, and SM Weissman (1994) Construction of libraries enriched for sequence repeats and jumping clones, and hybridization selection for region-specific markers. *Proceedings of the National Academy of Sciences*, **91(1)**:88–92.
- Kellner F, J Kim, BJ Clavijo, JP Hamilton, KL Childs, B Vaillancourt et al. (2015) Genome-guided investigation of plant natural product biosynthesis. *Plant J.* **82**:680–692.
- Khan T A, AH Ipshita, RM Mazumdar, ATM Abdullah, GMR Islam, and MM Rahman (2020) Bioactive polyphenol profiling and in-vitro antioxidant activity of *Tinospora cordifolia* Miers ex Hook F and Thoms: A potential ingredient for functional food development. *Bangladesh j. sci. ind. res.*, **55(1)**:23–34.
- Kumar R, C Kumar, R Paliwal, DR Choudhury, I Singh, A Kumar, et al. (2020). Development of novel genomic simple sequence repeat (g-SSR) markers and their validation for genetic diversity analyses in kalmegh [*Andrographis paniculata* (burm. f.) nees]. *Plants* **9**: 1–15.
- Kumar R, C Kumar, R Jain, A Maurya, A Kumar, A Kumari, and R Singh (2022) Molecular cloning and In-silico characterization of NAC86 of Kalmegh (*Andrographis paniculata*). *Indian J. Horti.* 2022;**79(1)**:9–14.
- Kumar R, S Das, M Mishra, DR Choudhury, K Sharma, A Kumari, R Singh (2021) Emerging roles of NAC transcription factor in medicinal plants: progress and prospects. *3 Biotech* **10**:1–4.
- Li C, Y Zhu, X Guo, C Sun, H Luo, J Song, Y Li, L Wang, J Qian, S Chen (2013) Transcriptome analysis reveals ginsenosides biosynthetic genes, microRNAs and simple sequence repeats in *Panax ginseng* C. A. Meyer. *BMC Genomics* **14**:1–11.
- Liu Y, P Zhang, M Song, J Hou, M Qing, W Wang, C Liu (2015) Transcriptome Analysis and Development of SSR Molecular Markers in *Glycyrrhiza uralensis* Fisch. *PLoS One* **10(11)**: e0143017.
- Michael TP, and S Jackson (2013) The First 50 Plant Genomes. *Plant Genome* **6(2)**.
- Mishra RK, BH Gangadhar, JW Yu, DH Kim, SW Park (2011) Development and characterization of EST based SSR markers in Madagascar periwinkle (*Catharanthus roseus*) and their transferability in other medicinal plants. *Plant Omics*. **4(3)**:154–161.
- Mochida K, T Sakurai, H Seki, T Yoshida, K Takahagi, S Sawai, S et al. (2016) Draft genome assembly and annotation of *Glycyrrhiza uralensis*, a medicinal legume. *The plant journal* **89(2)**:181–194.
- Okhuarobo A, JE Falodun, O Erharuyi, V Imieje, A Falodun, P Langer (2014) Harnessing the medicinal properties of *Andrographis paniculata* for diseases and beyond: a review of its phytochemistry and pharmacology. *Asian Pacific J. Trop. Dis* **4(3)**:213–222.
- Otto LG, P Mondal, J Brassac, S Preiss, J Degenhardt, S He, JC Reif, TF Sharbel (2017) Use of genotyping-by-sequencing to determine the genetic structure in the medicinal plant chamomile, and to identify flowering time and alpha-bisabolol associated SNP-loci by genome-wide association mapping. *BMC genomics*, **18(1)**:1–18.
- Paliwal R, R Kumar, DR Choudhury, AK Singh, S Kumar, A Kumar, KC Bhatt, R Singh, AK Mahato, NK Singh, R

- Singh (2016) Development of genomic simple sequence repeats (g-SSR) markers in *Tinospora cordifolia* and their application in diversity analyses. *Plant Gene* **5**:118–125.
- Pei L, B Wang, J Ye, X Hu, L Fu, K Li, Z Ni, Z Wang, Y Wei, L Shi, Y Zhang (2021) Genome and transcriptome of *Papaver somniferum* Chinese landrace CHM indicates that massive genome expansion contributes to high benzyloisoquinoline alkaloid biosynthesis. *Hortic. Res.* **8**.
- Peng J, C Shi, D Wang, S Li, X Zhao, A Duan, N Cai, C He (2021) Genetic diversity and population structure of the medicinal plant *Docynia delavayi* (Franch.) Schneid revealed by transcriptome-based SSR markers. *J. Appl. Res. Med. Aromat. Plants* **21**:100294.
- Raghu AV, SP Geetha, G Martin, I Balachandran, PN Ravindran (2006) Direct shoot organogenesis from leaf explants of *Embelia ribes* Burm. f.: a vulnerable medicinal plant. *J. For. Res.*, **11**(1):57–60.
- Rajalakshmi M, J Eliza, CE Priya, A Nirmala, and P Daisy (2009) Anti-diabetic properties of *Tinospora cordifolia* stem extracts on streptozotocin-induced diabetic rats. *Afr. J. Pharm. Pharmacol.* **3**(5):171–180.
- Rai A, H Kamochi, H Suzuki, M Nakamura, H Takahashi, T Hatada, K Saito, M Yamazaki (2017a). De novo transcriptome assembly and characterization of nine tissues of *Lonicera japonica* to identify potential candidate genes involved in chlorogenic acid, luteolosides, and secoiridoid biosynthesis pathways. *J. Nat. Med.* **71**(1):1–5.
- Rai A, M Nakamura, H Takahashi, H Suzuki, K Saito, M Yamazaki (2016a) High-throughput sequencing and de novo transcriptome assembly of *Swertia japonica* to identify genes involved in the biosynthesis of therapeutic metabolites. *Plant Cell Rep.* **35**(10):2091–2111.
- Rai A, M Yamazaki, H Takahashi, M Nakamura, M Kojoma, H Suzuki, K Saito (2016b). RNA-seq transcriptome analysis of *Panax japonicus*, and its comparison with other Panax species to identify potential genes involved in the saponins biosynthesis. *Front. Plant Sci.* **7**:481
- Rai M, A Rai, N Kawano, K Yoshimatsu, H Takahashi, H Suzuki, N Kawahara, K Saito, M Yamazaki (2017b). De Novo RNA Sequencing and Expression Analysis of *Aconitum carmichaelii* to Analyze Key Genes Involved in the Biosynthesis of Diterpene Alkaloids. *Molecules* **22**(12):2155.
- Şelale H, I Çelik, V Gültekin, J Allmer, S Doğanlar, A Fray (2013) Development of EST-SSR markers for diversity and breeding studies in opium poppy. *Plant Breed.* **132**(3):344–351.
- Sethi KL, ML Maheswari, VK Srivastava, R Gupta (1968) Natural variability in *Vetiveria zizanioides* collection from Bharatput part-I. *Indian Perfumer*, **30**(2): 377–380.
- Shen Q, L Zhang, Z Liao, S Wang, T Yan, PU Shi, M Liu, X Fu, Q Pan, Y Wang, Z Lv (2018). The Genome of *Artemisia annua* Provides Insight into the Evolution of Asteraceae Family and Artemisinin Biosynthesis. *Mol. Plant* **11**(6):776–88.
- Shokeen B, S Choudhary, NK Sethy, S Bhatia (2011) Development of SSR and gene-targeted markers for construction of a framework linkage map of *Catharanthus roseus*. *Ann. Bot.* **108**(2):321–36.
- Sierro N, JN Battey, S Ouadi, N Bakaher, L Bovet, A Willig, S Goepfert, MC Peitsch, NV Ivanov (2014) The tobacco genome sequence and its comparison with those of tomato and potato. *Nat. Commun.* **5**(1):1–9.
- Singh R, R Kumar, AK Mahato, R Paliwal, AK Singh, S Kumar, SS Marla, A Kumar, NK Singh (2016) De novo transcriptome sequencing facilitates genomic resource generation in *Tinospora cordifolia*. *Funct. Integr. Genomics* **16**(5), 581–591.
- Singh R, D Narzary, J Bhardwaj, AK Singh, S Kumar, A Kumar (2014). Molecular diversity and SSR transferability studies in Vetiver grass (*Vetiveria zizanioides* L. Nash). *Ind. Crops Prod.* **53**:187–198.
- Singh SS, SC Pandey, S Srivastava, VS Gupta, B Patro (2003). Chemistry and medicinal properties of *Tinospora cordifolia* (Guduchi). *Indian J. Pharmacol.* **35**(2):83.
- Spandana U, SL Ali, T Nirmala, M Santhi, SS Babu (2013) A review on *Tinospora cordifolia*. *Int. J. Curr. Pharm. Rev. Res.* **4**(2):61-8.
- Sui C, J Wei, S Chen, H Chen, C Yang (2009) Development of genomic SSR and potential EST-SSR markers in *Bupleurum chinense* DC. *African Journal of Biotechnology.* 2009;**8**(22): 6233–6240.
- Sun W, L Leng, Q Yin, M Xu, M Huang, Z Xu, Y Zhang, H Yao, C Wang, C Xiong, S Chen (2019) The genome of the medicinal plant *Andrographis paniculata* provides insight into the biosynthesis of the bioactive diterpenoid neoandrographolide. *Plant J.* **97**(5):841–857.
- Sun Y, H Luo, Y Li, C Sun, J Song, Y Niu, Y Zhu, L Dong, A Lv (2011). Pyrosequencing of the *Camptotheca acuminata* transcriptome reveals putative genes involved in camptothecin biosynthesis and transport. *BMC Genomics* **12**(1):1–11.
- Tian Z, F Zhang, H Liu, Q Gao, S Chen (2016) Development of SSR Markers for a Tibetan Medicinal Plant, *Lancea tibetica* (Phrymaceae), Based on RAD Sequencing. *Appl. Plant Sci.* **4**(11):1600076.
- Tong Z, Z Yang, X Chen, F Jiao, X Li, X Wu, Y Gao, B Xiao, W Wu (2012) Large-scale development of microsatellite markers in *Nicotiana tabacum* and construction of a genetic map of flue-cured tobacco. *Plant Breed.*, **131**(5):674–80.
- Tripathi S, Y Srivastava, RS Sangwan, NS Sangwan (2020) In silico mining and functional analysis of AP2/ERF gene in *Withania somnifera*. *Sci. Rep.* **10**(1):1–2.
- Upadhyay AK, AR Chacko, A Gandhimathi, P Ghosh, K Harini, AP Joseph, AG Joshi, SD Karpe, S Kaushik, N Kuravadi, CS Lingu (2015) Genome sequencing of herb Tulsi (*Ocimum tenuiflorum*) unravels key genes behind its strong medicinal properties. *BMC Plant Biol.* **15**(1):1–20.
- Vega JJ, S Ayling, M Hegarty, D Kudrna, JL Goicoechea, Å Ergon, OA Rognli, C Jones, M Swain, R Geurts, C Lang (2015) Red clover (*Trifolium pratense* L.) draft genome provides a platform for trait improvement. *Sci. Rep.* **5**(1):1–0.
- Vidya V, D Prasath, M Snigdha, R Gobu, C Sona, CS Maiti (2021) Development of EST-SSR markers based on transcriptome and its validation in ginger (*Zingiber officinale* Rosc.). *PLoS ONE* **16**(10):e0259146.

- Wang J, MD Qi, J Guo, Y Shen, HX Lin, LQ Huang (2017) Cloning, subcellular localization, and heterologous expression of ApNAC1 gene from *Andrographis paniculata*. *China J. of Chinese Mat. Med.* **42(5)**:890-5.
- Wang L, Z Wang, J Chen, C Liu, W Zhu, L Wang, L Meng (2015). De novo transcriptome assembly and development of novel microsatellite markers for the traditional Chinese medicinal herb, *Veratrum baillonii* Franch (Gentianaceae). *Evol. Bioinforma.* **11**:39–45.
- Wang Y, H Cai, W Jia (2012) u Analysis of SSR information in EST resource of sweet wormwood (*Artemisia annua*) and development of EST-SSR marker. *China J. of Chinese Mat. Med.* **37(5)**:570–4
- Wang C, X Hao, Y Wang, I Maoz, W Zhou, Z Zhou, G Kai (2022a) Identification of WRKY transcription factors involved in the regulation of anti-cancer drug camptothecin biosynthesis in *Ophiorrhiza pumila*. *Hortic. Res.* 2022.
- Wang Y, J Dai, R Chen, C Song, P Wei, Y Cai, Y Wang, B Han (2022b) miRNA-Based Drought Regulation in the Important Medicinal Plant *Dendrobium huoshanense*. *J. Plant Growth Regul.* **41(3)**:1099–108.
- Ward JA, L Ponnala, CA Weber (2012) Strategies for transcriptome analysis in nonmodel plants. *Am. J. Bot.* **99(2)**:267–76.
- Winzer T, V Gazda, Z, He F Kaminski, M Kern, TR Larson, Y Li, F Meade, R Teodor, FE Vaistij, C Walker (2012) A *papaver somniferum* 10-gene cluster for synthesis of the anticancer alkaloid noscapine. *Science* **336(6089)**:1704–8.
- Wu B, M Wang, Y Ma, L Yuan, S Lu (2012) High-throughput sequencing and characterization of the small RNA transcriptome reveal features of novel and conserved microRNAs in *Panax ginseng*. *Plos one* **7(9)**: e44385.
- Wu J, C Cai, F Cheng, H Cui, H Zhou (2014) Characterisation and development of EST-SSR markers in tree peony using transcriptome sequences. *Mol. Breed.* **34(4)**, 1853–1866.
- Xu J, Y Chu, B Liao, S Xiao, Q Yin, R Bai, H Su, L Dong, X Li, J Qian, J Zhang (2017). *Panax ginseng* genome examination for ginsenoside biosynthesis. *Gigascience* **6(11)**, 1–15.
- Xu Y, S Tian, R Li, X Huang, F Li, F Ge, W Huang, Y Zhou (2021) Transcriptome Characterization and Identification of Molecular Markers (SNP, SSR, and Indels) in the Medicinal Plant *Sarcandra glabra* spp. *Biomed Res. Int.* **7**:2021.
- Yamazaki M, K Mochida, T Asano, R Nakabayashi, M Chiba, N Udomson, Y Yamazaki, DB Goodenowe, U Sankawa, T Yoshida, A Toyoda (2013) Coupling Deep Transcriptome Analysis with Untargeted Metabolic Profiling in *Ophiorrhiza pumila* to Further the Understanding of the Biosynthesis of the Anti-Cancer Alkaloid Camptothecin and Anthraquinones. *Plant Cell Physiol.* **54(5)**:686–96.
- Zhang R, Z Chen, L Zhang, W Yao, Z Xu, B Liao, Y Mi, H Gao, C Jiang, L Duan, A Ji (2021) Genomic characterization of WRKY transcription factors related to andrographolide biosynthesis in *Andrographis paniculata*. *Front. Genet.* **11**:601689.
- Zhao D, JP Hamilton, GM Pham, E Crisovan, K Wiegert-Rininger, B Vaillancourt, D DellaPenna, and CR Buell (2017) De novo genome assembly of *Camptotheca acuminata*, a natural source of the anti-cancer compound camptothecin. *GigaScience.* **6(9)**:gix065.
- Zhou RR, JH Zhou, TG Nan, C Jiang, HY Duan, YP Zhao, LQ Huang, Y Yuan (2019) Analysis of genomic SSRs in *Pueraria lobata* and *P. thomsonii* and establishment of DNA identity card for different germplasms of *P. thomsonii* of Jiangxi province. *J. Chinese Mater. Medica* **44(17)**:3615–21.

Policies Impacting Access to Plant Genetic Resources in Last Four Decades

Pratibha Brahma^{1*}, Vandana Tyagi¹, Pragya¹ and Anuradha Agrawal²

¹ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

²Indian Council of Agricultural Research, Krishi Anusandhan Bhavan II, New Delhi-110012, India

A paradigm shift occurred in the international policy environment with respect to biological diversity from “heritage of mankind” to “sovereign rights of a nation”, primarily triggered by the Convention on Biological Diversity (CBD) in 1992. To address the issue of food security, in terms of access to plant genetic resources for food and agriculture (PGRFA) as well as for realization of farmer’s rights, the International Treaty for Plant Genetic Resources (ITPGRFA) was adopted 2001. In 2010 a Nagoya Protocol on Access and Benefit Sharing (NP-ABS) was developed under CBD to facilitate exchange of biological resources bilaterally between countries, on mutually agreed terms. Both the Treaty and CBD offer regulatory mechanisms for exchange of germplasm and fair and equitable share of benefits and are implemented through national policies in mutually supportive manner. However, practically they have impacted germplasm flow. In the world of plant breeding and crop improvement, these regulations are seen to bring a slow-down in the international exchange of germplasm hampering scientific progress.

Introduction

Agro-biodiversity is crucial for food and nutritional security of the world. This component of biodiversity is dependent on the availability of basic raw material in the form of germplasm for improvement of crops, livestock, and other components used as food fibre and feed. Agro-biodiversity needs to be sustained by humans for current and future generations, as opposed to biodiversity existing in natural ecosystems, which is self-sustained. The germplasm and varieties of various crops used for food and animal feed have been developed over centuries mainly through human interventions. Most of the cultivated or domesticated species are not found in the wild habitats as such. The use of plant genetic resources for food agriculture (PGRFA) is essential for crop improvement programs and the interdependence for these resources among countries requires facilitated exchange. Therefore, it is important to develop mechanisms for facilitated access among countries.

The germplasm exchange mechanisms have rapidly changed during the last four decades, due to the trends of globalization and privatization. A major paradigm shift was witnessed in the international policy environment from “heritage of mankind” to “sovereign rights of a nation”, primarily triggered by the Convention of Biological Diversity (CBD), which came into force

in 1993, adopted during the Rio Earth Summit of the United Nations. It was the first legally binding institutional mechanism, providing for conservation and sustainable use of all biological diversity and with provisions of equitable sharing of benefits arising out of the use of biodiversity. The CBD reaffirmed national sovereignty over genetic resources and stressed that the authority to determine access to genetic resources rests with the national governments and is subject to national legislations (<http://www.biodiv.org/doc/legal/cbd-en.pdf> full text of the Convention). It provides for a bilateral approach to access/exchange between countries on prior informed consent (PIC) and mutually agreed terms (MAT).

The CBD was meant to put in place regulations regarding access to germplasm, and raised the required level of negotiations. The Convention provided a framework of regulations for access to genetic resources and transfer of relevant technologies. However, it does not offer special treatment to PGRFA which are of different nature and crucial for food security and sustainable agriculture. The Food and Agriculture Organization of the United Nation (FAO) simultaneously adopted a resolution for revision of non-binding International Undertaking on Plant Genetic Resources (IUPGR) on PGRFA which aimed at systematic PGR management, international co-operation and PGR exchange. The

*Author for Correspondence: Email-Pratibha.brahmi@icar.gov.in

Commission on Genetic Resources for Food and Agriculture (CGRFA) addressed the outstanding issues of access to PGRFA and realization of farmer's rights and adopted the International Treaty for Plant Genetic Resources (ITPGRFA) in the 31st Conference of the FAO in November 2001. The ITPGRFA entered into force in 2004 (<http://www.fao.org/ag/cgrfa/itpgr.htm>).

During the negotiations of the Treaty, it was realized that not only plant breeders but also farmers must have rights over their landraces and varieties (Paroda, 2013). The Treaty thus provided for facilitated access to PGRFA and established the Multilateral System (MLS) of Access and Benefit Sharing (ABS). The objectives of the Treaty and CBD are identical, though the access and benefit sharing mechanisms are dealt differently. The Treaty has created a MLS of ABS while CBD and Nagoya Protocol on Access and Benefit Sharing (NP-ABS) creates mechanism for bilateral arrangements (Halewood, 2015). Both the Treaty and CBD are however meant to be implemented through national policies in mutually supportive manner. With the advent of CBD continued free exchange of germplasm was the main concern but more importantly, the most critical issue is how the farmers who are the true discoverers, conservers, producers and breeders of these invaluable resources can be benefitted from these resources. The Treaty and the NP-ABS though ratified by many countries, has halted the use of genetic resources to a great extent (Paroda, 2018).

PGR Governance Status

Global recognition of the policy significance of interdependence on PGRFA arguably reached its zenith in 2001 when 'interdependence' was explicitly included in Article 11 of the ITPGRFA as one of two criteria - the other being relevance for food security- for including crops or forages in the MLS. Through the MLS, ITPGRFA parties have agreed to create a global, virtual pool of genetic resources for 64 crops (these are listed in the Treaty's Annex 1). In addition to conservation, this germplasm is intended to be utilized for the purposes of training, breeding and research for food and agriculture. Member states agree to provide facilitated access to one another (including natural and legal persons within their borders) on the understanding that monetary benefits will be shared if the recipients incorporate materials in new, commercialized PGRFA products that are not available to others for research,

training or breeding (Dua *et al.*, 2004). Issues were raised on the ownership of PGR and benefit sharing, as the vast collections of germplasm conserved in various gene banks in CGIAR system were collected from gene rich/economically developing nations and stored away from the place of collection (Evenson 1999, Hamilton *et al.*, 2005).

India is signatory to legally binding instruments like CBD, ITPGRFA, World Trade Organization's agreement on Trade Related aspects of Intellectual Property Rights (WTO-TRIPS), and it has also endorsed the Global Plan of Action (1996). TRIPS and CBD compliant legislation/ acts/ policies have been enacted in the country during the past 20 years in terms of providing access to PGR within the country and sharing of benefits arising out of their use. These include the Patents (Amendment) Act 2005, 'Biological Diversity Act 2002', 'Protection of Plant Varieties and Farmers' Rights Act, 2001' and Geographical Indications of Goods (Registration and Protection) Act 1999. The access mechanisms of germplasm under these regimes are depicted in Fig 1.

For complying with the provisions of CBD, Government of India enacted the legislation called Biological Diversity Act (BDA), 2002 and also notified the Biological Diversity Rules, 2004 (<http://www.nbaindia.org>). The objectives of the BDA are to provide for conservation of biological diversity, sustainable use of its components and fair and equitable sharing of benefits arising out of the use of biological resources, knowledge and for related matters, giving effect to the CBD. For the purpose of the Act, a National Biodiversity Authority has been established and which regulates conservation and access to biological diversity for sustainable utilization and equitable sharing of benefits arising out of the utilization of biological resources. India being signatory to the legally binding Treaty has an obligation as a Contracting Party, however as per BDA prior approval of NBA is required for accessing biological resources. Thus for harmonizing provisions of BDA and ITPGRFA, Government of India issued a Gazette Notification in 2014 for implementation of ITPGRFA and provided exemption under Section 40 of BDA, 2002 (to exempt certain biological resources normally traded as commodities) for facilitating export of germplasm through ICAR-NBPGR for Annex 1 crops of Treaty.

India has also ratified Nagoya Protocol (NP) on ABS and is one of the countries already having domestic

measures in place on ABS. NP recalls and recognises the MLS and ABS established under the ITPGRFA in the context of poverty alleviation and climate change and acknowledges the fundamental role of Treaty and FAO CGRFA in harmony with the Convention. The protocol provides for a strong basis for greater legal certainty and transparency for both providers and users of genetic resources. Gazette notification to this effect was issued in 2014 and guidelines notified.

Access to Indian Biological Resources by Nationals of Other Countries

As per the BDA, 2002, no person from outside India or a body corporate, association, organization incorporated or registered in India having non-Indian participation in its share capital or management, can access any biological resources or knowledge associated, for research, commercial utilization, bio-prospecting or bio-utilization, without proper approval of NBA. Also, no person can apply for any intellectual property right in or outside India for any invention based on any research or information on a biological resource obtained from India, without obtaining approval from NBA. Collaborative research projects involving transfer or exchange of biological resources or information

between Governments sponsored institutions of India, and such institutions in other countries, are however exempted, if they conform to the policy guidelines approved by the Central Government (Box 1). The role of national research organizations mandated with agricultural research in development of new varieties is well-established. In addition, there are number of State Agricultural Universities and private sector seed companies engaged in crop improvement and varietal development programs. The public and private sectors are, therefore, supplementary to each other to fulfil the demand of the Indian farmers in terms of new crop varieties/hybrids, quality seed production and their distribution. At national level, access to biological resources from India is regulated by BDA

Box 1. Exemption Provided Under the Biological Diversity Act, 2002

Section 40	Annex 1 crops of ITPGRFA
Section 5	Collaborative research projects conforming to policy guidelines issued by Central Government and approved by Central Government
Section 7	Local people and communities of the area, growers and cultivators of biodiversity, and vaidas and hakims practicing indigenous medicine
Section 40	NTCs (Normally traded commodities) (for material used only as commodity by traders)

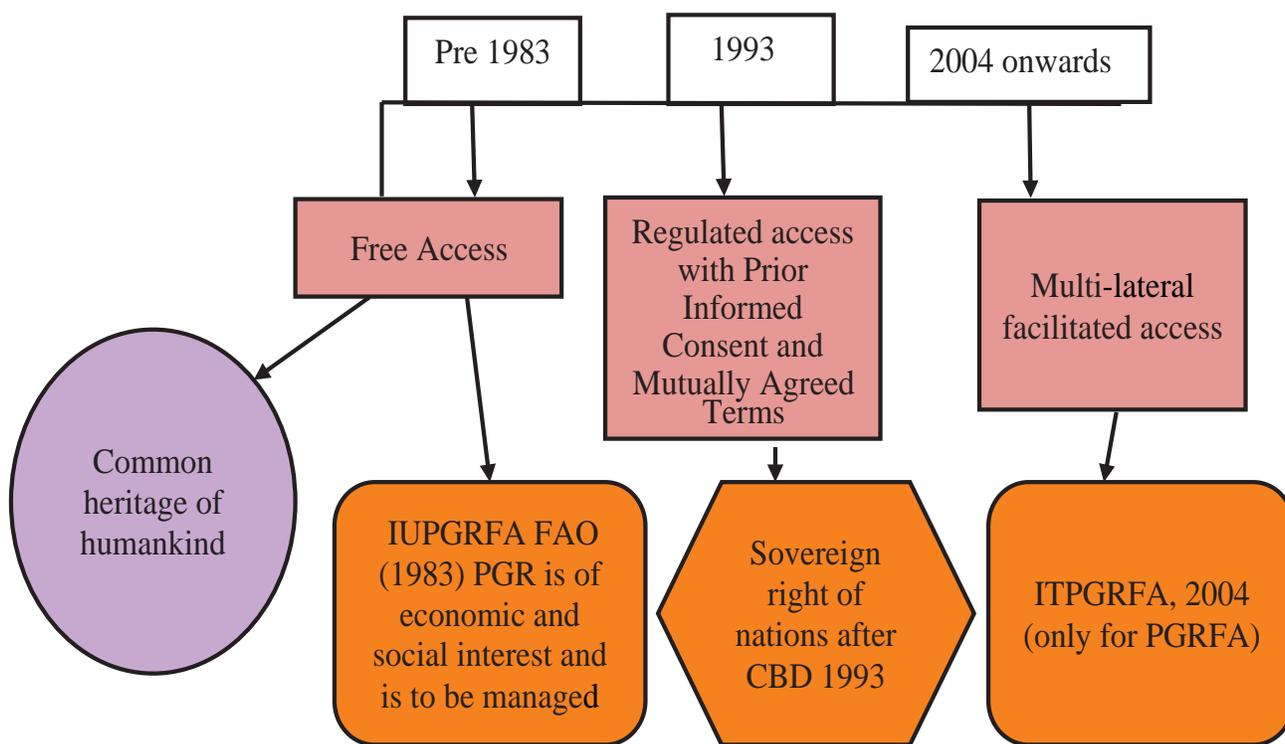


Fig. 1. Access to Genetic Resources under different Policy Regimes

Impact of CBD and Treaty on Germplasm Flow in India

Under the free access before CBD, need-based import and export of research material was facilitated by NBPGR with delegation of the responsibility to Director, ICAR-NBPGR by the Department of Agricultural Research and Education (DARE), Ministry of Agriculture. During that period, India benefitted with introduction of many new crops (e.g. sunflower, soybean, kiwi, dragon fruit, cashew etc.). A detailed account of introduction of fruit crops in and from India has been published recently (Ranjan *et al.*, 2022). However, during the implementation of the BDA 2002 and subsequent notifications to implement the provisions of the Act, many stakeholders expressed their concerns hindering the “business as usual” with respect to genetic resources use.

To address this concern, Tyagi *et al.* (2006) analysed the number of germplasm accessions introduced from CGIAR Centres and NGBs during pre-CBD period (1988-1992) and post-CBD (1997-2001) period. There was an overall decline of 14.5% in introduction the post-CBD era. The results showed that requests for germplasm import from other countries for public-funded research were drastically reduced from 80-90% to only 20-22%. This decline was confined to supplies from NGBs of various countries, indicating cautious approach adopted by different countries in sharing their germplasm, post-CBD. Interestingly, 81% of germplasm introduced from NGBs did not originate or they were not the centres of diversity for that particular biological resource (Tyagi

Category	Pre regulations (1985-1994)	Post regulations (2010-19)	% increase/decrease
ICAR	55	20	35↓
CG Centers	10.9	10.8	0.1↓
Private	4.5	60.8	56.3↑
Universities	17	7	10↓
Others	12.6	1.4	11.2↓

Fig. 2. Share of plant germplasm accessions imported in India, by categories of users

et al., 2006). It indicates that these importing countries might have realized the importance of PGR much earlier and assembled vast collections. Similarly export of germplasm is also reduced to a great extent.

Another analysis for the period 1985-1994 before and 2010-2019 after regulations entered into force has been done by the authors to study the impact on the volume of exchange of germplasm. The years 1995-2009 are not taken into account as this period was considered as transition phase. The results showed that the number of germplasm accessions imported from CG Centres more or less remain unaffected. However, a 35% reduction from NGBs in other countries occurred in germplasm requested by ICAR Institutes. In total, a decline of 56.3% was seen in total imports if, private seed companies are not taken into account (Fig. 2). Private seed companies import mainly from their counterparts/ sister companies. With reference to export of PGR, a sharp decline was observed in number of samples exported, slight increase is observed in year 2018 as the export was undertaken under a collaborative project on ‘Stress tolerant orphan

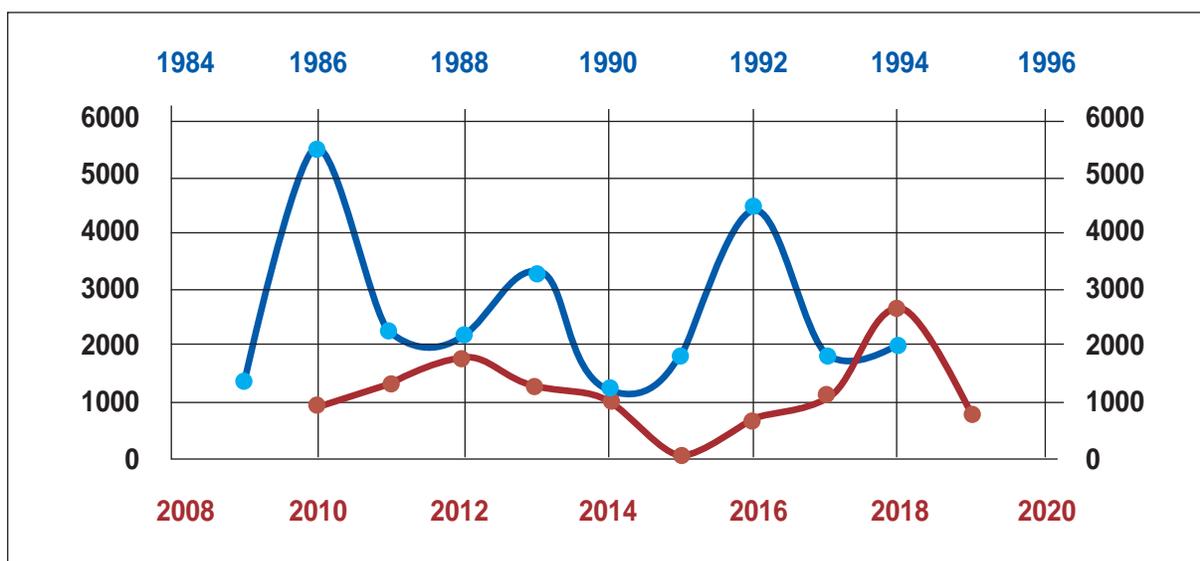


Fig. 3. Number of samples exported

legumes across Sub-Saharan Africa and India' (Fig 3.). This decline in number of import of germplasm is likely to have a long-term effect on breeding in public sector organization especially in crops like pulses, vegetables and other potential crops.

For exchange of components of agrobiodiversity, there is a need for closer look at the provisions of the BDA 2002, with respect to regulation for access to germplasm for research (including crop improvement), as well as some definitions. It is necessary to underline the intent and context of these provisions especially with reference to food and agriculture. For exchange of Genetic Resources for Food and Agriculture (GRFA) including plants, animal, fish, microbes and insects, ICAR had designated its five bureaux dealing with GRFA as 'single window' system for import and export of these components as per extant legislation and international regulations (Office order F. No. 8 (2) / 2011/ Cord. (Tech.) dated 19.10. 2012). Section 16 of BDA, 2002 provides for delegation of power and functions of the Act by National Biodiversity Authority (NBA). Under these special powers of NBA, DARE may be delegated to deal with agrobiodiversity issues including sharing of components of agrobiodiversity including their commercial utilization through its National Agricultural Research System (NARS). India's Biological Diversity Act 2002 is under revision and above suggestions have been provided by the PGR community on the proposed amendments.

Way Forward

The challenge in the coming decades will be for countries to ensure the access, sustainable use and transfer of PGRFA for food/livelihood security and conservation, while ensuring the fair and equitable sharing of benefits from their use with the providers and knowledge holders. Many countries continue to evolve their institutional arrangements in response to changing circumstances, new information and experience. A survey of ABS country measures accommodating the distinctive features of PGRFA was undertaken by the CGRFA which found that there are significant gaps in research about the positive or negative effects of the regulatory measures in practice on various stakeholders and the conservation and sustainable use of PGRFA.

Thus, policy change must be aimed to enhance efficient flow of germplasm around the world. Since the national policy affect germplasm exchange for

PGRFA and owing to the special features of these resources, they needs to be dealt separately from policies governing access to non-food and non-feed biological resources. The overall objectives of the CBD being conservation of total biodiversity, the use of cultivated agrobiodiversity has little impact on the environment and naturally occurring biodiversity which is self-sustaining. Although, anthropogenic pressures are affecting our climate and environment, cultivated diversity is generated continuously and it can be conserved only through utilization. Thus the regulations for general biodiversity need not apply to components of agrobiodiversity.

Acknowledgement

Dr RS Paroda, a global leader in the area of plant genetic resources mangement has been closely associated with the changes in the policy domain with respect to PGR exchange and ABS mentioned in this article. The authors thank him for his continued guidance and congratulate him on his 80th birthday (August 28, 2022), wishing him many more years of productive and happy life ahead.

References

- Dua RP, P Brahma and BS Dhillon (2004) International Treaty on Plant Genetic Resources for Food and Agriculture: An assessment. *Indian J. Plant. Genet. Resour.* **17**: 48-32.
- Evenson RE (1999) Intellectual property rights, access to plant germplasm, and crop production scenarios in 2020. *Crop Sci.* 1630-1635
- Halewood M, P Brahma, PN Mathur and KC Bansal (eds) (2013) *A Road Map for Implementing the Multilateral System of Access and Benefit Sharing in India.* Bioversity International, Rome, ICAR and NBPGR, New Delhi, 126 p.
- Hamilton S, Ruraidh and E Javier (2005) Intellectual property rights and germplasm exchange: the new rules in planning rice breeding programmes for IMPACT.
- Paroda RS (2013) Implementing the International Treaty to address current concerns about managing our plant genetic resources. In M Halewood, P Brahma P, PN Mathur and KC Bansal KC (eds) *A Road Map for Implementing the Multilateral System of Access and Benefit Sharing in India.* Bioversity International, Rome, ICAR and NBPGR, New Delhi, pp 4-9.
- Paroda RS (2018) Agrobiodiversity: dynamic change management. In: *Reorienting Indian Agriculture: Challenges and Opportunities.* CAB International, Wallingford UK, pp. 157-164.
- Ranjan P, Brahma P and Tyagi V *et al.* (2022) Global interdependence for fruit genetic resources: status and challenges in India. *Food Sec.* <https://doi.org/10.1007/s12571-021-01249-6>
- Tyagi V, Singh AK, Chand D, N Verma, Singh RV and Dhillon BS (2006) Plant Introduction in India during Pre- and Post-CBD periods – An analysis *Indian J. Plant Genet. Resour.* **19**(3): 436-441.

PGR Informatics: Sustaining PGR Management and Supporting PGR Policy

Sunil Archak^{1*}, Kuldeep Singh² and RC Agrawal³

¹ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012, India

²Head Genebank, International Crops Research Institute for the Semi-Arid Tropics, Hyderabad-500030, Telangana, India

³Education Division, Indian Council for Agricultural Research, Pusa Campus, New Delhi-110012, India

Introduction

Researchers and policy makers recommend increased use of plant genetic resources (PGR) available as *ex situ* collections in genebanks as part of the strategy to fight food and nutritional insecurity as well as to mitigate the impact of climate change on crop husbandry. However, in the absence of a robust information management system, genebanks will have problems managing their collections and users face a daunting task of choosing appropriate accessions for research. Enhanced use of PGR can only be facilitated by enhanced access to information. An organized digital information system provides fair and just opportunity for all to access. PGR Informatics is the management (creation, storage, retrieval and presentation) and analyses (discovery, exploration and extraction) of diverse information (facts, figures, images, statistics, knowledge and news) related to plant genetic resources.

PGR documentation assists in recording observations and data and to convert them into useful information. PGR informatics is expected to facilitate creation of knowledge in an interactive manner based on awareness and perceptions of the users. Ultimate aim is to achieve wisdom by integrating traditional knowledge and synthesized digital knowledge.

PGR Informatics deals with (1) Accession Level Data (Passport. Data), describing the identity and origin of the material recorded at the stage of augmentation (collecting/import); (2) Trait Level Data (phenotypic data), describing the phenotypic traits of the material observed in field/lab generated by systematic characterization and evaluation experiments; (3) Genotypic Data (Omics data), describing information about the DNA/RNA/protein sequence, metabolomic products, often in association with the results of high-throughput phenotypic data. Other significant datasets include (i) Quarantine data

(pest interception, salvage); (ii) genebank data (storage, seed quality); (iii) Germplasm Distribution (international and domestic supply); etc.

Seminal Changes

The “PGR information” comprises of accession level information (static) and trait information (evolving). PGR informatics is essential to facilitate efficient PGR management by genebanks (setting priorities, planning activities and managing resources), to enhance the utilization by breeders and researchers (genotype selection and indenting; genotype Registration), and to assist policy makers and funding agencies (allocation of funds; political approval of programs; compliance with obligations of global developments including CBD and ITPGRFA). Therefore, information must be up to date, accurate, reliable, systematically stored and easily accessible.

PGR Informatics has come into the limelight since a decade because of (i) Increased awareness about PGR; (ii) Various international agreements coming into force; (iii) Increased availability of information in text, images, maps, videos, etc. (iv) Accessibility to technologies to record, link and archive such diverse types of information; and (v) Ever-increasing power of computers and internet to facilitate access and retrieval.

There have been seminal changes in (i) Technology (from sequential processing to random access) enabling use of computer technologies by non-specialists; (ii) Policy (from open access to regulated access) forcing national genebanks develop own information systems rather than adopt a common global system; and (iii) Objective of genebanking (from conservation to utilization) requiring information systems to expand and build databases around traits and develop relevant online applications.

*Author for Correspondence: Email-sunil.archak@icar.gov.in, sunil.archak@gmail.com

Status of PGR Informatics

Globally, all the major genebanks (CGIAR or national) maintain information in on or the other form. Recently, all the CG genebanks have moved to GRIN Global system and their data are available on Genesys-PGR (1). Researchers can access PGR information from various sources that maintain primarily accession level data (Fig 1). Efforts are on to provide access to trait data for facilitating use. Additionally, novel initiatives aim to link accessions with the composition and the function of food (2) or with genomic and phenomic information (3).

PGR Informatics in India: At present NBPGR is maintaining accession level data in Passport Database (>600K entries) and Exchange Database (>950K), Conservation Database (>400K) and Characterization Database (>200K). The databases are accessible through

various online open access applications including PGR Portal (pgrportal.nbpgr.ernet.in), Import Permit and EC Data Search (exchange.nbpgr.ernet.in), Genebank Dashboard (http://genebank.nbpgr.ernet.in/), National Herbarium of Crop Plants (pgrinformatics.nbpgr.ernet.in/nhcp), Germplasm Registration Information System (www.nbpgr.ernet.in:8080/registration), PGR Map (pgrinformatics.nbpgr.ernet.in/pgrmap), and ipPGR (pgrinformatics.nbpgr.ernet.in/ip-pgr). The applications are widely used by PGR researchers indicated by over 70K page views annually. Links to other PGR informatics applications are available on NBPGR server (4).

PGR Informatics Aiding PGR Policy Implementation

PGR Informatics also serves the major objectives of the Convention on Biological Diversity, rolling Global Plan of Action and International Treaty on Plant Genetic

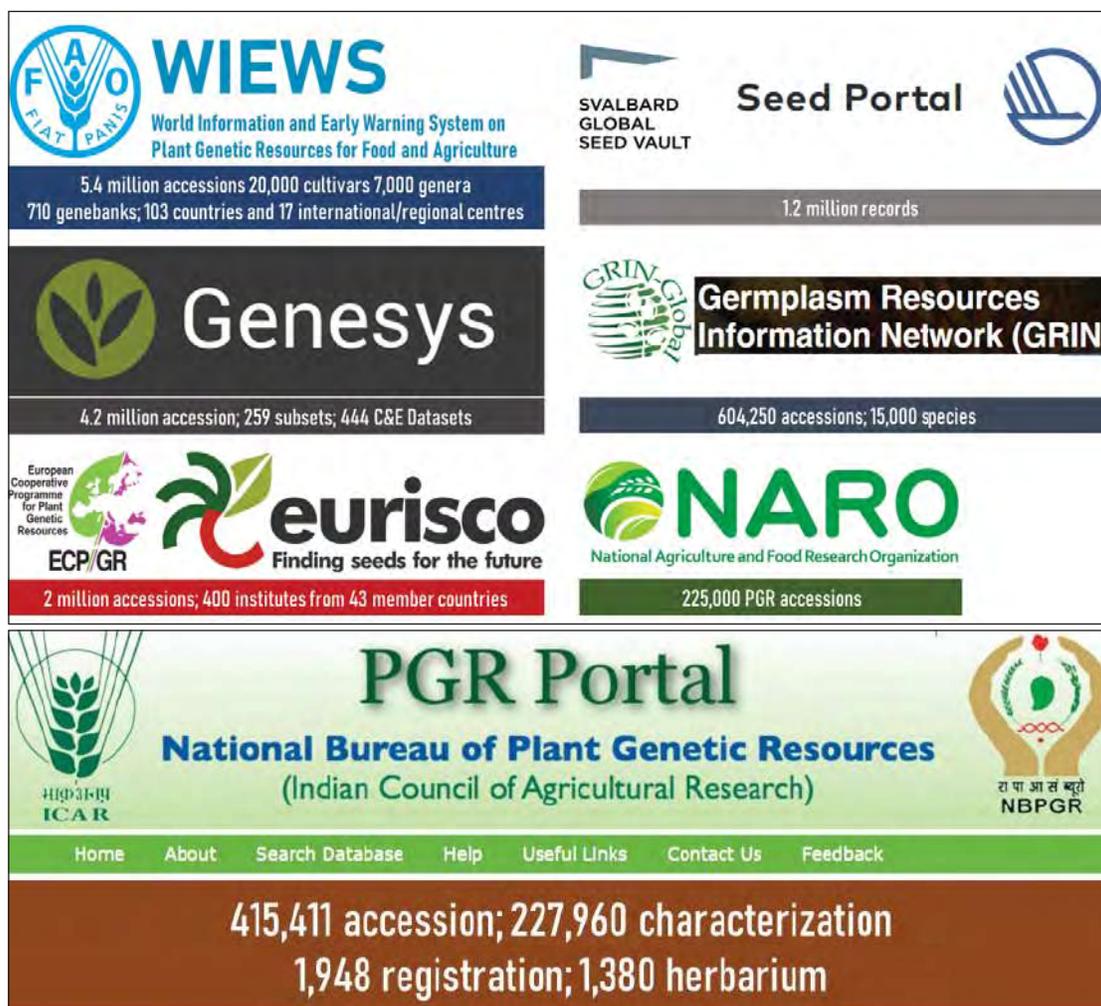


Fig. 1. Major web portals of plant genetic resources information

Resources for Food and Agriculture (Fig 2). For instance, the Global Information System for PGRFA aims to integrate and augment existing systems to create the global entry point to information and knowledge (5).

This service assigns Digital Object Identifiers (DOIs) to plant genetic resources for food and agriculture for reference in third party systems and scientific literature to facilitate tracking eventually benefit sharing.

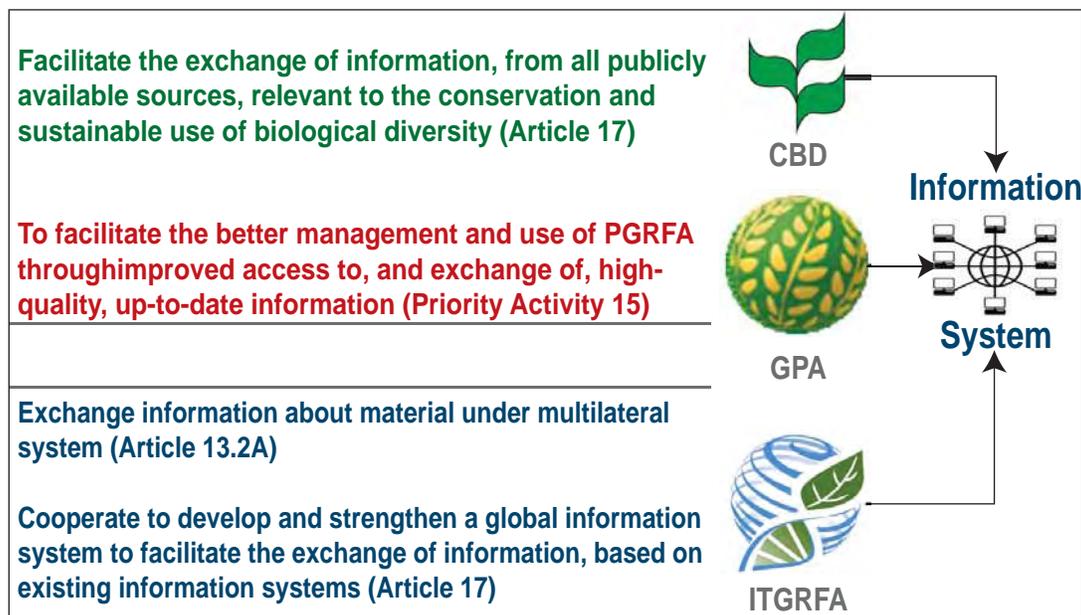


Fig. 2. Interface between PGR Informatics and PGR policy

	<p>Data quality</p> <p>Trait specific evaluation data</p> <p>Genomic data</p> <p>GIS and AI ready</p>
	<p>Information services in local languages</p> <p>Accessibility & Inter-operability</p> <p>Dynamic linking and updating</p> <p>Efficient data delivery</p>
	<p>Harmonization</p> <p>Dedicated funding</p> <p>Institutional mechanism for sustainability</p> <p>Capacity building</p>

Fig. 3. Challenges for PGR Informatics in India

Challenges and Action Points

It is imperative, for achieving effective management and utilization of plant genetic resources, to maintain and expand of PGR Informatics in India. Success depends upon the action aimed at addressing the technical, financial and policy challenges periodically (Fig. 3).

References

- <https://www.genesys-pgr.org>
- <https://foodperiodictable.org>
- <https://divseekintl.org>
- www.nbprg.ernet.in/PGR_Databases.aspx
- <https://ssl.fao.org/glis/>

ARTICLES ON
ANIMAL GENETIC RESOURCES



ARTICLES ON ANIMAL GENETIC RESOURCES

Title	Page No.
Animal Genetic Resources (AnGR) Diversity in India	223
Cattle and Buffalo Genetic Resources of India	229
Goat and Sheep Genetic Resources of India	237
Poultry Genetic Resources of India and its Role in Rural Poultry Production	242
Conservation for Effective Management of AnGR in India	247
Valorization of Indigenous Livestock and Poultry – An Approach towards their Conservation	251
Uniqueness of Animal Genetic Resources Adapted to High Altitude Environment of Leh-Ladakh	258
Genetic Uniqueness for Immune Response Traits in Native Animal Genetic Resources	264
A1/A2 Milk Research in Indian Cattle	269
Breed Registration and the Gazette Notification: A Unique National Framework for Protecting Native Animal Germplasm	279

Animal Genetic Resources (AnGR) Diversity in India

BP Mishra* and SK Niranjan

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

The history of farm Animal Genetic Resources (AnGR) is about 12 to 14 thousand years old; started with the domestication of the animals for companion, food, fur and hide purposes. Among 40 non-carnivores livestock species domesticated by human being, till date, only cattle, sheep, goat, pig and chicken are highly prevalent and known as 'Big five'. Other species like buffalo, dromedaries and Bactrian camel, llama, alpaca, yak, and mithun are region specific. At present, about one-fifth of the global food is of animal origin in the world. A vast genetic diversity of AnGR has been generated during hundreds of years through culmination of various evolutionary forces, including intensive selection and preferred mating by human for various utilization purposes. Initial domestication modulated the wild ancestors to adapt to human habitation and making themselves more useful for the human being. Migration of animals by humans across the continents further disseminated the germplasm, initiating the diversification of the farm animals across the globe. However, artificial selection for different needs and likings by the human is the chief element for development of large-scale genetic diversity, in form of distinct breeds and populations. These breeds as well as specialized populations have been evolved for a specific need or utilization pattern like milk, draft, meat, fibre, carting, riding etc. Today, the world possesses more than eight thousand livestock and poultry breeds, evolved from only few founder populations and catering various needs of human society around the world. As per Global data bank on Animal Genetic Resources of FAO, a total of 8774 breeds of 38 species were reported, worldwide (data from 182 countries) in 2014. Among these, 7718 are local breeds (in one country) and 510 are regional transboundary breeds (in one region) and 546 are international transboundary breeds (in more than one region).

Breed Evolution and Diversity in India

India possesses 11 farm livestock species – cattle, buffalo, sheep, goat, pig, horse, donkey, camel, yak, mithun and rabbit and few domesticated poultry species – chicken, geese, duck, Guianese fowl, Japanese quails etc. In India, there are 536.82 million livestock and 851.12 million poultry (Livestock Census, 2019). The species wise proportion of total livestock is 36.04% cattle, 20.47% buffalo, 27.74% goat, 13.83% sheep, 1.69% pig and rest 0.23% represented by mithun, yak, horses and ponies, mule, donkey and camel.

In fact, Indian subcontinent has witnessed the earliest domestication of many farm species including Indicine cattle, buffalo, goat and poultry. In Indus civilization, cattle husbandry was main occupation playing important role, both in nutritional as well as economic, in their society. Selection of animals for specific work, based on specific traits, has laid foundation of breed formation in Ancient India. In an ancient literature of Parini's "Astadhyayi" first time described four types of cattle-Halika, Sakat, Rathya and Sairika based upon their work type. Different use of cattle during Arya era laid

the initial foundation for the breed development. Some best cattle breeds like Ongole and Gir, probably most ancient breeds of any farm animal across the world. Some of the breeds like Devni, Amrit Mahal, Gao Lao have been evolved about three centuries back.

Currently, in India, there are 199 registered indigenous breeds of livestock and poultry, which include 50 of cattle, 19 of buffalo, 34 of goat, 44 of sheep, 7 of horses and ponies, 9 of camel, 10 of pig, 3 of donkey and one of yak in livestock and 19 of chicken, one of geese and two of duck in poultry. Three breeds of indigenous native dog have also been registered recently, the first time in the country. During the last 10 years, more than 100 new populations were identified across the country and 73 breeds of different farm animal species were registered. Breeds of species like pig, donkey, yak, dog and geese were first time registered in the country (Table 1) in the last decade. With a greater pace, 40 new breeds were added in only last 4 years (2017-19 to 2020-21). Importantly, a number of breeds were registered from remote areas like NEH and also for the minor species; which are although less

*Author for Correspondence: Email: director.nbagr@icar.gov.in

Table 1. Species-wise number of breeds

Species	Extant breeds	New breeds	Total breeds
Cattle	30	20	50
Buffalo	10	9	19
Sheep	39	5	44
Goat	21	13	34
Horse	6	1	7
Pig	0	10	10
Camel	8	1	9
Donkey	0	3	3
Yak	0	1	1
Chicken	15	4	19
Duck	0	2	2
Geese	0	1	1
Dog	0	3	3
Total	129	73	202

in population but contributes significantly to the society. A number of new breeds have also been reported from the remote regions of the country. Like North-Eastern states have registered 21 breeds in recent time (Table 2). Many of the new breeds have been added in breed inventory by the states like Gujarat, Odisha and Tamil Nadu, mainly because of active participation of local agencies and NGOs in these states.

Some of the regions, like Rajasthan, Gujarat, North Eastern states are rich in breed diversity, mainly because of their unique production systems. In fact, many of the breeds evolved due to specific production system in country. For example, pastoralism is a unique production

system of rearing, which has led to the development of many livestock breeds. The nomadic herding community from western India, especially Gujarat and Rajasthan, is responsible for the development of many famous breeds. Gir, Kharai, Nari, Sanchori cows, Banni buffalo, Kachhi, Khadai camel are developed by the pastoral community. More than 60 breeds reared by pastoralists have been recognised and registered till date.

However, considering country's vast geographic and ecological regions, contrasting climatic conditions along with diverse necessity of the farmers, there is still a sizable undefined population, particularly those of cattle and goat. With 10 percent of the global livestock population, the country possesses only 4 percent of the total breeds. There is one breed per 3 million livestock population in India, which is much lower than the world average (one breed per 0.9 million animals), about 4 to 6.5 million for cattle, buffalo and goat, the three most populous species in India. FAO also predicted a greater number of livestock breeds in the country. About 54 percent of total livestock is non-descript, as per recent breed survey (DAHD, 2022). It included 52.0 % of cattle, 45.4% of buffalo, 50.6% of sheep, 63.5% of goat and 56.3% of pig as non-descript. There are a number of states like Madhya Pradesh, Chhattisgarh, Jharkhand, Bihar, Uttar Pradesh, West Bengal, Telangana etc. which are rich in AnGR diversity and have large proportion of non-descript population.

Table 2. State-wise number of breeds

State	New breeds(2010-21)			Total breeds			State	New breeds(2010-21)			Total breeds		
	Pr.	Sec.	Total	Pr.	Sec.	Total		Pr.	Sec.	Total	Pr.	Sec.	Total
Rajasthan	2	1	3	24	5	29	West Bengal	1	0	1	4	1	5
Gujarat	8	1	9	23	4	27	Andaman & Nicobar	2	0	2	3	-	3
Tamil Nadu	8	0	8	21	0	21	Chhattisgarh	2	0	2	3	0	3
Maharashtra	3	0	3	13	1	14	Haryana	1	0	1	3	3	6
Jammu & Kashmir	3	0	3	11	0	11	Nagaland	3	0	3	3	0	3
Karnataka	4	1	5	11	5	16	Sikkim	0	0	0	3	0	3
Odisha	9	0	9	12	2	14	Uttarakhand	3	0	3	3	1	4
Uttar Pradesh	3	1	4	11	3	14	Arunachal Pradesh	1	0	1	2	1	3
Andhra Pradesh	0	1	1	7	2	9	Goa	2	2	4	2	2	4
Assam	5	0	5	7	0	7	Manipur	1	1	2	2	1	3
Himachal Pradesh	2	1	3	7	1	8	Meghalaya	1	1	2	2	1	3
Punjab	2	0	2	6	0	6	Jharkhand	0	1	1	1	1	2
Bihar	3	1	4	5	1	6	Mizoram	1	0	1	1	0	1
Kerala	0	0	0	4	0	4	Telangana	1	0	1	1	0	1
Madhya Pradesh	0	0	0	4	4	8	Tripura	1	0	1	1	1	2

Pr. - Primary home tract; Sec.- Secondary home tract

ICAR-NBAGR: Committed to Preserve Farm Animal Biodiversity of the Country

Established on 21st September 1984, ICAR-National Bureau of Animal Genetic Resources (NBAGR) is working with a mission to protect and conserve indigenous Farm Animal Genetic Resources for sustainable utilization and livelihood security, with many important national and international commitments, to date. The bureau has achieved a number of milestones, including registration and notification of 202 native animal breeds from all parts of the country, during its small stint. This has enabled the recognition of almost 50 percent of native livestock of the country as descript. About 230 breed/populations of indigenous livestock and poultry were characterized and documented by the Bureau. It included, 120 newly identified breeds/populations of indigenous animals across the country. Further, with a target towards zero non-descript AnGR in the country, the bureau has also initiated a country-wide survey in collaboration with State AHDs, SAUs in mission mode. For the long-term conservation program as also included under SDG Indicator 2.5.1, the bureau has cryopreserved the germplasm for native breeds in form of Semen (49 breeds/populations), Somatic cell (24 breeds/populations) and DNA (169 breeds/populations) at National Gene Bank of the Bureau. Further, the bureau has also conserved 17 threatened breeds, in their native tracts through involving livestock keepers and stakeholders under the network program on AnGR. The knowledge about genetic diversity and genomic uniqueness of native breeds has been enriched through genomics research. The molecular genetic work carried out at NBAGR has resulted in some important technologies including SNP chips for almost all livestock species. High-end research at the bureau has also enabled us to identify unique traits like thermotolerance, endurance as well as the uniqueness of the products of native breeds; which would help in the value addition of native breeds. Omics based trait characterization for climatic adaptation, heat/ cold tolerance in native cattle, buffalo and sheep breeds and unique animal products- Milk (Sahiwal), colostrum (cattle/yak) Meat (Kadaknath) are hallmark research initiatives. AGR-IS database is available with all kind of information on native breeds in the country. Bureau is actively involved in creating awareness about the indigenous livestock, their upkeep and conservation through interactions with farmers during their visits to the breeding tracts. Bureau is encouraging the conservation efforts of the farmers and pastoralists through bestowing them Breed Conservation Award and Breed saviour awards. More than 100 farmers/livestock keepers belonging to different states have been awarded for recognizing and conserving native breeds and populations. The Bureau has borne the greatest responsibility towards native animals and their keepers and striving hard to protect the precious animal biodiversity.

Mission towards Zero Non-descript AnGR of India: A new initiative for identifying new breeds

ICAR-NBAGR has taken the initiative to characterize and document all non-descript animal genetic resources of the country. The task has been taken under Mission towards Zero Non-descript Animal Genetic Resources of India, launched by Secretary DARE & DG, ICAR on 11th August, 2021. The Mission would encompass the activities in all states of the country in collaboration of the state Animal Husbandry departments/ SAUs/SVUs/NGOs ICAR-NBAGR. Bureau has initiated documentation of native AnGR in 22 states/UTs. For sensitization of the stakeholders the institute is also organizing State Interface Meets and after the launch of the mission, interface meets with 12 states have been completed. At present, projects on documentation of AnGR are being implemented in 22 states/UTs in collaboration with State Animal Husbandry Departments, KVKs, ICAR institutes and SVUs/SAUs. Bureau's scientists have made the visits in various states after launch of the Mission towards Zero Non-descript AnGR. About 18 new populations of native livestock and poultry have been identified after launch of Mission, which are being characterized in their respective breeding tracts falling in various states.

Breed Registration and Gazette Notification: A Framework for Recognising Native Germplasm

Registration of livestock and poultry genetic resources has been initiated to protect and check the biopiracy of indigenous AnGR. Our country has a mechanism for recognizing the valuable sovereign animal genetic resource with known characteristics in form of a national documentation system. Registration of breeds of native livestock and poultry has greater impact in livestock sector in the country. In the year, 2008, ICAR-National Bureau of Animal Genetic Resources (NBAGR), Karnal was given the temporary authority for the registration of germplasm related to livestock and poultry in the country. First time in the year 2008, all 129 extant breeds of livestock and poultry were registered by the NBAGR. Further 73 breeds were newly added and by 2021, the number reached 202. Further, to provide legal safeguards for germplasm protection, notification of indigenous breeds has started in the year 2019 through publishing Official Gazette by the Government of India. The Government of India has notified all 202 breeds for keeping and rearing for the purposes of animal husbandry, production, breeding, conservation, utilization, consumption, and trade through five Gazette Notifications between 2019-2021.

Registration and, further notification of breeds has certainly broadened its impact through modulating various policies and programmes. Along with preserving the biodiversity as mandate under Delhi Declaration (2016), the stakeholders specially Farmers would be the most benefitted. After breed designation, superior quality germplasm of the breed become available to the livestock owners. Notification provided legal support to Intellectual Property Rights (IPRs) of the registered breeds and for developing mechanism for sharing benefits among the animal keepers.

Risk to AnGR Diversity

History of protecting and utilizing the native germplasm is long but international framework for their sustainable utilization along with preservation of biodiversity could be established in Earth Summit held at Rio de Janeiro in 1992. Convention on Biological Diversity (CBD) of Earth Summit has endorsed sovereignty of any country over its genetic resources, including AnGR. It has necessitated to protect AnGR through developing appropriate policies

in the country. A number of steps, internationally and nationally has been taken since then, specially related to the protection of AnGR biodiversity. To mention, Global Plan of Action for AnGR of FAO (Interlaken Declaration) in 2007, CBD-Nagoya protocol for Access & Benefit Sharing and Indigenous Knowledge in 2010, Aichi Biodiversity Targets of CBD in 2010. Recent UN's Sustainable Development Goals also envisaged management of all genetic resources globally specifically to promote sustainable agriculture and achieving food security. India also has its legal bindings to all these conventions and declarations and striving hard to protect its genetic resources.

At present the loss of indigenous animal biodiversity is well evident, globally. Second Report on State of World's Animal Genetic Resources for Food & Agriculture (2015) of the FAO describes that only 17.7 percent breeds are 'Not at risk', globally. The risk status that the proportion of livestock breeds classified as being at risk of extinction has been increased from 15 percent in 2005 to 17 percent in 2014 in the world. A total of the world's 7.4 percent livestock and poultry breeds have come under 'extinct' and 16.6 percent under 'at risk' category. More than half (58.3 percent) of breeds have been classified as 'unknown' status of risk; further, indicates that the number of breeds at risk is likely to be underestimated, globally.

In India, Breed-wise Livestock Survey 2013 and 2019 conducted by DAHDF, Govt. of India, first time, gave the first indication about the population of the registered livestock breeds in the country. Taking in account the figures in survey, about 14 per cent of the total indigenous livestock breeds are seems to be under threat. The proportion of breeds under threat is much higher in minor species including camel and horses. Recently, Singh and Sharma (2017) assessed the risk status of major livestock species in country using the criteria lay down by FAO (2013).

Drivers of Change

In analysis of country reports by FAO, indiscriminate crossbreeding has been identified as most frequent cause of genetic erosion in the world and reported by maximum countries. Other drivers of change, listed in decreasing importance included – introduction and/or increased use of exotic breeds, lack of or weak AnGR policies, programme and institutions, breeds not profitable/competitive or have poor performance,

Indigenous Breeds at Risk*

Cattle	: <i>Belahi, Pulikulam, Punganur</i>
Buffalo	: <i>Chilika</i>
Goat	: <i>Sumi-Ne, Chegu, Karnah, Teressa</i>
Sheep	: <i>Kachaikatty Black, Tibetan, Nilgiri</i>
Pig	: <i>Agonda Goan, Tenyi Vo</i>
Camel	: <i>Kharai, Marwari, Mewari, Mewati, Malvi, Jalori</i>
Horse	: <i>Zanskari, Kachchi-Sindhi, Bhutia, Spiti, Manipuri</i>

*As per FAO criteria, DAHD-Breed Survey Report 2022

intensification of production or decline of traditional production system and disease management, loss of grazing land, inbreeding or other breeding problems, migration or uptake of alternate employment, change in consumer demand, mechanization, non-appreciation of local breeds, unspecified economic and market factors climate change, globalization and liberal trade, lack of infrastructure for production-processing and marketing and lack of interest among young generation.

The Drivers behind the decrease of most of the indigenous populations are numerous, however, the most evident drivers behind change of AnGR dynamics as well as depleting AnGR diversity are—decreasing utility and increasing pressure for production. Consistently high demand with changing production scenario has derived the preference for highly specialized breeds, which has attributed to the loss of genetic diversity of the livestock in general, and of indigenous breeds in particular. Consistently high demand with changing production scenario has derived the preference for highly specialized breeds, which has attributed to the loss of genetic diversity of the livestock especially indigenous breeds. The introduction of exotic germplasm especially of cattle, chicken, pig and sheep from agriculturally advanced economies has resulted in a serious erosion of highly adapted farm animal breeds. Mechanised agricultural and transport led the down-numbering of the farm animal breeds needed for draught, ploughing and utility as a pack animal. India in its Country Report 2 (2014) has also assessed the impact of drivers of change on AnGR biodiversity in the country in coming years. Economic, livelihood, or lifestyle factors affecting the popularity of livestock keeping were predicted to have a high impact on AnGR management.

Future Prospective and Action Points

- Exploring and characterizing new breeds of indigenous farm animals from all parts of the country is the utmost requirement. Completing documentation of all indigenous AnGR and inventory of breeds of the country should be the target for next five years. Mission towards zero non-descript AnGR would be the most fruitful effort, if implemented country wide and funded adequately. To characterize mixed population by means of molecular tools would be important to upgrade such populations with the closest breeds.
- With each of the registered breed, there should be the society of their breeders which would help to conserve, improve and better utilize the breed in local area. Such societies have been started to be evolved, but in few states like Gujarat, Tamil Nadu and Odisha etc.. Bureau has to have greater involvement in breed society formation.
- Cryopreservation of germplasm in form of semen/somatic cells/embryo should be completed for all registered breeds with sufficient genetic diversity. Cryopreservation at National level with National Gene Bank of advanced and state of the arts facilities. *In situ* conservation of breeds would only be effective through participation of stakeholders and farmers. There is need for true valuation and value addition of the AnGR and their products for long term conservation and sustainable utilization of the native breeds.
- Information System on all aspects of Animal Genetic Resources should be integrated at national and regional levels with active participation of all animal husbandry department and institutions like DAHD, NDDB, ICAR, State AH Deptt., state livestock boards, veterinary universities, NGOs etc.
- Establishment of statutory procedure and authority for protection of AnGR and their stakeholders' benefits. Gazette notification is one of the step forward in this direction. Specific act/regulatory policy for protection of breeds and their keepers knowledge, the benefit sharing as per Nagoyo protocol is highly required in similar line to the PPVFRA for the plant. Such Act would also be helpful to decide the rights and ownership for the synthetic animal varieties/strains used for commercial purposes.

Reference

- Breed Survey (2013) Estimated Livestock Population Breed-wise Based on Breed Survey-2013, Department of Animal Husbandry, Dairying & Fisheries, Ministry of Agriculture & Farmers Welfare, Government of India.
- Breed wise Report of Livestock and Poultry (2022) Based on 20th Livestock Census-2019, Department of Animal Husbandry and Dairying, Ministry of FAHD, Government of India.
- Country Report 2- India (2014) The Second Country Report on State of Worlds Animal Genetic Resources for Food and Agriculture –India, Submitted to FAO
- FAO (2007) Global plan of action and the Interlaken declaration commission on genetic resources for food and agriculture food and agriculture organization of the United Nations Rome.
- ICAR-NBAGR (2016) Guidelines for Management of Animal Genetic Resources of India, National Bureau of Animal Genetic Resources (ICAR), Karnal.
- FAO (2015) The second Report on the State of the World's Animal genetic Resources for Food and agriculture, edited by BD Scherf and D Pilling. FAO, Rome.
- Livestock Census (2019) 20th Livestock Census, Key results (Provisional). Department of Animal Husbandry & Dairying, Ministry of Fisheries, Animal Husbandry & Dairying, Government of India.
- Singh PK and A Sharma (2017) Assessment of degree of endangerment of livestock breeds in India. *Indian J. Anim. Sci.* **87**(3): 316-323.

Cattle and Buffalo Genetic Resources of India

RK Pundir

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

There are 50 registered breed of cattle and 19 breeds of buffalo in the country, these breeds are well adapted to diverse climate of the country and contributing significantly to the livelihood. India possesses 193.4 million cattle and 109.85 million buffalo in the year 2019. The crossbred and buffalo population has increased all the time since the livestock census initiated. Among the cattle and buffalo, 52% cattle and 45.4% buffalo population are non-descript and need to be defined on priority so their development programs can be initiated. India is home of best breeds of buffalo in the world i.e. Murrah and Sahiwal, Gir, Ongole and Tharparkar breeds of cattle. Mehsana and Niliravi are the breeds of buffalo those are equally good in performance in the country. Gir breed of cattle and Murrah breed of buffalo had maximum heads in the country. Breed wise trends during the years 2013-2022, showed that among the 37 cattle breeds included in breed survey/census, 15 are showing increasing trends and 22 showed declining trends. In buffalo, 13 breeds were included in breed survey/census, 4 are showing declining trends and 9 showed increasing trends. For improving the bovines in the country, we need to define non-described bovines on priority and registered unique population as distinct breed, if eligible, accurate breed census/survey, performance recording system at farmers door, ensure availability of superior germplasm, more population to be covered through Artificial Insemination and establishment of breed societies and breed nucleus farms adopting open nucleus breeding system.

Introduction

Indigenous bovines are rich in variability and excellent in desirable attributes like disease resistance, tolerance to hot and humid stresses, adaptability to different environment and production systems and ability to utilize coarse roughages and crop residues. The breeds like Gir, Ongole and Sahiwal were introduced into different countries for their wide adaptation and disease resistance qualities. The indigenous cattle milk carries A2 protein, which is superior to A1 protein that is found in the milk of crossbred cattle. India is home tract of the world best buffalo breed i.e. Murrah, distributed to all parts of the country. India is also home tract of Niliravi and Mehsana buffalo those are equally good in performance as of the Murrah.

India's livestock sector is one of the largest in the world. It has 56.7% of world's buffaloes and 14.7% of cattle population. This sector is also supporting livelihood of more than two-thirds of the rural population. Most of the indigenous breeds of cattle were evolved for draft over the centuries. However, most of the buffalo breeds were evolved for milk. Purebred breeds of cattle and buffalo have been selectively bred over a long period of time to possess a distinctive identity in colour, size,

conformation, and function and have the prepotency to pass these traits in to their progeny.

The world milk production in the year 2020 was 906 million tonnes and India contributed 22% by producing 198.44 million tons. The per capita availability in the country was 406 ml/day against the ICMR recommendations of 280 ml/day. The species wise contribution in milk production in India was indigenous buffalo 34.51%, buffalo non-descript 13.83%, indigenous cows 9.63%, indigenous cows non-descript 10.42%, crossbred cows 27.68%, cows exotic 0.98% and goat 2.95% in the year 2019. It revealed that indigenous bovines are contributing to 68.39% milk production in the country. Being the first ranked in milk production in the world, still milk production productivity (kg) per cow per day is very less i.e. indigenous buffalo 6.43, buffalo non-descript 4.51, indigenous cows 3.90, indigenous cows non-descript 2.57, crossbred cows 8.09, cows exotic 11.88 and goat 0.44. Efforts are needed to improve the milk productivity of different species in the country by defining the bovines (non-descript) of country and initiation of the genetic improvement programs.

In India, as per the 20th Livestock Census (Anonymous, 2019), 36.04% livestock population belong to cattle i.e. Second largest world cattle population.

*Author for Correspondence: Email-Rakesh.Pundir@icar.gov.in

There are 193.4 million cattle in the country comprising 142.1 million indigenous and 51.3 million crossbreds. Among the cattle 26% are exotic/crossbreds, 22% are indigenous and rest 52% still need to be defined. Among the crossbreds 54.9% belong to Jersey or its crossbreds and 43.3% to Holstein Friesian and its crossbreds. Rest 1.8% belongs to other crossbreds. Among the indigenous cattle (142.1 million), 29.5% belongs to pure or grades of indigenous breeds and 70.5% are non-descript. In India, there are 109.8 million buffalo, world buffalo largest population comprising 54.6% pure breeds and their grades and 45.4% need to be defined. The pure breeds are 20.3% and grades of different breeds are 34.3%.

Status of Bovines

Cattle population has increased by 23.9% since the year 1951. The indigenous cattle population has shown a decline of 13.1 million in the year 1951, as there were only indigenous cattle in the country. The first exotic/crossbred cattle population was reported in the year 1982 as 8.8 million which has increased tremendously to 51.3 million in the year 2019. West Bengal is the state which had maximum cattle population as 19.0 million

in the country and shown an increase of 15.18% over the year 2012. Uttar Pradesh was the first state who had maximum cattle population in the year 2012 as 19.6 million but it reported a decline of 3.93% during the years 2012 to 2019 (Anonymous, 2019).

As per the Livestock census, 2019, the indigenous cattle are showing 0.8% increase over the last Livestock Census, 2012. The female cattle population had shown an increased by 18% over the year 2012 in the year 2019. The indigenous/non-descript female cattle population has shown an increase of 10% in the year 2019 over the year 2012. The exotic/crossbred cattle increased by 26.9% during the same period. There a decline of 6% in indigenous cattle population over the 2012, however it is less than 9% during the years 2007-2012. Cattle and buffalo populations and their trends over the years are shown in Figure 1.

The buffalo population in the year 2019 was 109.85 million in the country. It has increased 1.1% over the year 2012. In the year 2019, female population has increased by 8.61%, while males were declined by 42.35% over the same period. The buffalo population had shown an increase over all the time since 1951, a total increase of 66.45 million since the year 1951. The

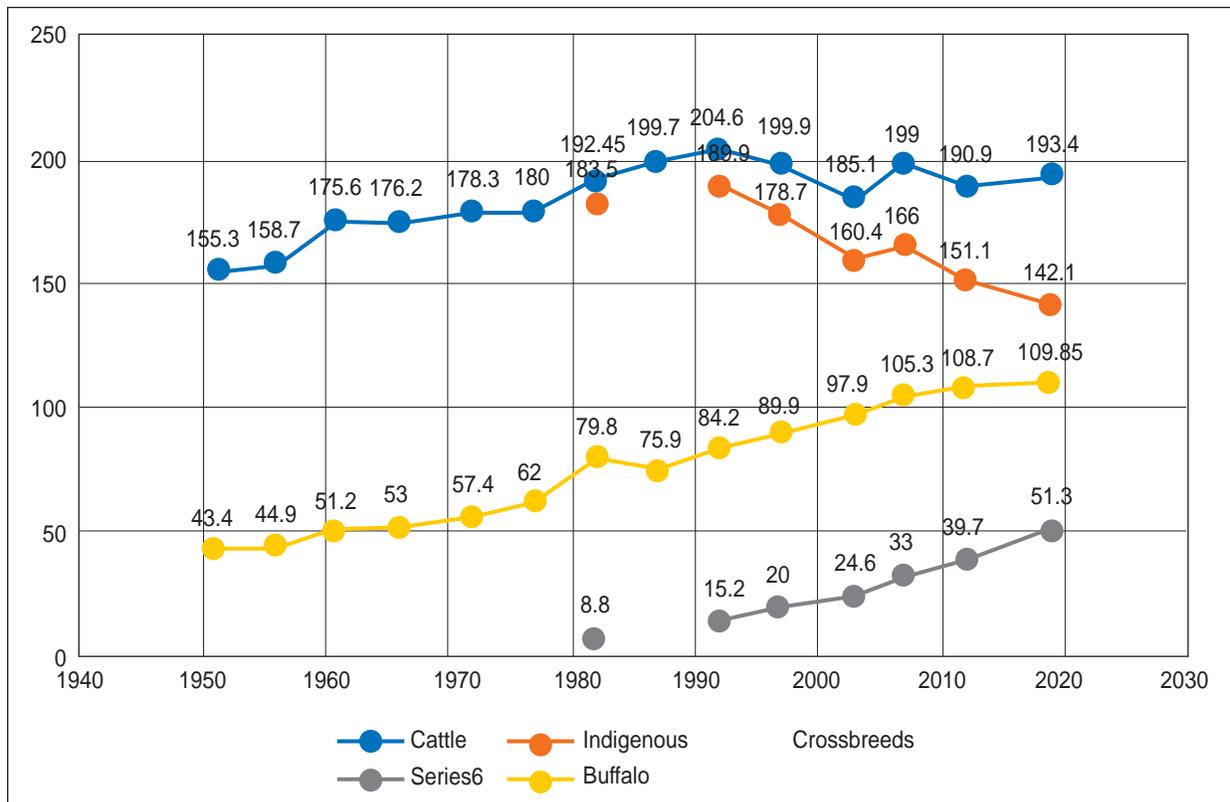


Fig. 1. Cattle and buffalo population (millions) trends

state Uttar Pradesh has maximum buffalo population (33 million) in the country followed by Rajasthan, Gujarat and Madhya Pradesh.

In India, there were 3 Breed Census/Survey Reports available in the country i.e. 2007 (Anonymous, 2007), 2013 (Anonymous, 2013) and 2022 (Anonymous, 2022). In The first Report (2007), there were pure breeds of different breeds counted, while grades of all breeds counted in a single figure, while in rest two reports (2013 and 2022) pure breeds and their grades counted separately.

As per the Breed Survey Report (2022), In India, there were 24.9 million pure breeds, 16.9 million grades of different breeds and 41.8 million non-descript cattle available among the indigenous cattle. There is significant increase in pure breeds cattle as compare to the Breed census Report (2013). The country had maximum number of Lakhimi breed (6.6 million) and contributing 4.8% of the total cattle population including it grades. The Gir breed is also contributing equally 4.8% but in it pure breeds are quite less (2.3 million) as compare to Lakhimi breed. The breeds those are contributing more than 1% population in India are Sahiwal, Bachur, Haryana, Kankrej and Kosali.

There are 22.3 million pure bred buffaloes and 37.6 million grades of different breeds available in the country. Murrah is the buffalo breed which has largest numbers of pure bred (14.2 million) and it grades (32.8 million), contributing 42.8% of the country buffalo population. The breeds those are contributing more than 1% of the country buffalo population are Mehsana, Surti, Jafarabadi and Bhadawari.

Cattle Diversity

There are 50 recognised cattle breeds in India. Cattle breeds, their distribution and population over different years presented in Table 1. There were 30 cattle extant registered breeds in the country by the 2008. The new breed registration process was started in the year 2008 at ICAR-National Bureau of Animal Genetic Resources (NBAGR), Karnal, Haryana. Since the year 2008 to 2022, there were 20 new cattle breeds registered in the country i.e. Motu, Ghumusari, Binjjharpuri, Khariar, Pullikulam, Kosali, Malnad Gidda, Belahi, Gangatiri, Badri, Lakhimi, Ladakhi, Konkan Kapila, Poda Thurupu, Nari, Dagri, Thutho, Shweta Kapila, Himachali Pahari and Purnea.

All the breeds can be classified in to three categories as per their utility i.e. milk (Sahiwal, Tharparkar, Rathi, Red Sindhi and Gir), draft (Amritmahal, Hallikar, Kangayam, Khilar, Ponwar, Kherigarh and Krishna Valley etc.) and dual purpose (Kankrej, Haryana, Gangatiri, Deoni and Dangi etc.). Indigenous Zebu cattle (*Bos indicus*) have some distinctive features like a prominent hump, a long face, upright horns, drooping ears, dewlap and slender legs. The prominent body colour varies from white to gray and black. Zebus experience relatively lower basal metabolic rate and a superior ability for heat dissipation in comparison to their taurine (*Bos taurus*) counterparts. They easily adapt to the extreme tropical heat and acquire resistance to diseases, especially the tick-borne diseases. Zebu cattle, predominantly Ongole, Gir, Sahiwal and Tharparkar, have been imported, improved and propagated/crossed in sub temperate/subtropical regions, such as the southern United States, South America and Australia for developing tropically adapted dairy/beef cattle breeds in these countries.

The best Indian dairy cattle breeds viz. Red Sindhi, Sahiwal, Rathi, Gir and Tharparkar producing more than 2000 kg milk in a lactation of 300 days are mostly available in the north western region of the country. In southern region, most of the breeds are either dual purpose or draft purpose like Hallikar, Kangayam, Khillar and Krishna Valley. Breeds like Red Sindhi, Sahiwal and Tharparkar are available in Pakistan also. India maintains only small populations of these prized breeds and that too largely in a few organized herds. Since these breeds are well adapted to the local Indian environmental conditions, there is a pressing necessity for their protection from dilution through crossbreeding and their conservation and multiplication using latest scientific advancements. Information on Indian cattle genetic resources with regard to their utility, breeding tract and accession number is given in Table 1.

Indian cattle are mostly white/ grey coat color, however almost all the coat colors are available from grey to black. The southern breeds with white/grey coat colour include Amritmahal, Hallikar Pullikulam, Kangayam, Krishna Valley, Ongole and Punganur. Hallikar breed of southern Karnataka also have peculiar grey patches on the face. The grey coat colour in north west region breeds include Gangatiri, Gaolao, Haryana, Kankrej, Kenkatha, Kherigarh, Khillar, Malvi, Mewati, Nagori and Tharparkar. The breeds those had brownish

Table 1. cattle and buffalo breeds, their distribution and population in different year

S. N.	Breed	Population (2007)	Population (2013)	Population (2022)	State(s)	S. N.	Breed	Population (2007)	Population (2013)	Population (2022)	State(s)
Cattle breeds											
1.	Amritmahal	96021	105343	104990	Karnataka	26.	Ladakhi	-	-	-	Leh and Ladakh
2.	Bachaur	454103	741432	3215259	Bihar	27.	Lakhimi	-	-	6648519	Assam
3.	Badri	-	-	741324	Uttarakhand	28.	Malvi	1515753	1158172	595658	Madhya Pradesh
4.	Bargur	20879	14154	42300	Tamil Nadu	29.	Malnad Gidda	1282121	899091	631530	Karnataka
5.	Belahi	-	-	4238	Haryana and Himachal Pradesh	30.	Mewati	75427	14773	9024	Haryana Uttar Pradesh
6.	Binjharपुरी	29749	79428	69406	Odisha	31.	Motu	700908	469320	231954	Odisha
7.	Dangi	303630	119373	139971	Maharashtra Gujarat	32.	Nari	-	-	-	Rajasthan
8.	Dagri	-	-	-	Gujarat	33.	Nagori	837334	373224	210012	Rajasthan
9.	Deoni	165846	151236	183656	Maharashtra Karnataka	34.	Nimari	309237	341828	398341	Madhya Pradesh Maharashtra
10.	Gangatiri	-	-	243153	Uttar Pradesh Bihar	35.	Ongole	257661	115905	303817	Andhra Pradesh
11.	Gaolao	222566	121538	112563	Maharashtra Madhya Pradesh	36.	Poda Thurpu	-	-	-	Telangana
12.	Gir	2126421	1380208	2300090	Gujarat	37.	Ponwar	24072	20067	14480	Uttar Pradesh
13.	Ghumusari	82117	58855	35626	Odisha	38.	Punganur	733	2772	9876	Andhra Pradesh
14.	Hallikar	2191486	1211242	501057	Karnataka	39.	Purnea	-	-	-	Bihar
15.	Hariana	2600111	1639181	1179089	Haryana, Uttar Pradesh, Rajasthan	40.	Pulikulam	-	7352	10495	Tamil Nadu
16.	Himachali Pahari	-	-	-	Haryana, Uttar Pradesh	41.	Rathi	924057	865921	878852	Rajasthan
17.	Kangayam	314817	80260	127577	Tamil Nadu	42.	Red Kandhari	176621	235058	95304	Maharashtra
18.	Kankrej	3884457	1945094	1580802	Gujarat Rajasthan	43.	Red Sindhi	550272	59642	272850	Organized farms only
19.	Kenkatha	179987	393291	76663	Uttar Pradesh Madhya Pradesh	44.	Sahiwal	457177	1092459	1881453	Punjab Rajasthan
20.	Khariar	-	290015	13365	Odisha	45.	Shweta Kapila	-	-	-	Goa
21.	Kherigarh	171414	75116	28433	Uttar Pradesh	46.	Siri	61750	12171	15278	Sikkim West Bengal
22.	Khillar	1419735	1102359	844400	Maharashtra Karnataka	47.	Tharparkar	557621	197291	151056	Rajasthan
23.	Kosali	-	2431859	989803	Chhattisgarh	48.	Thutho	-	-	-	Nagaland
24.	Konkan Kapila	-	-	989803	Chhattisgarh	49.	Umblachery	217960	39050	31195	Tamil Nadu
25.	Krishna Valley	2314	3462	2594	Maharashtra Karnataka	50.	Vechur	160	1075	8963	Kerala
Buffalo breeds											
1.	Bargur	-	-	-	Tamilnadu	11.	Manda	-	-	-	Odisha

S. N.	Breed	Population (2007)	Population (2013)	Population (2022)	State(s)	S. N.	Breed	Population (2007)	Population (2013)	Population (2022)	State(s)
2.	Banni	525099	239572	512851	Gujarat	12.	Marathwadi	181712	278502	217581	Maharashtra
3.	Bhadawari	723516	583599	1065485	Uttar Pradesh, Madhya Pradesh,	13.	Murrah	20488,488	11686198	14246525	All India
4.	Chhattisgarhi	-	-	-	Chhattisgarh	14.	Mehsana	3373006	1676699	3442006	Gujarat
5.	Chilika	1001	2599	11010	Odisha	15.	Nagpuri	139939	73584	104016	Maharashtra
6.	Dharwadi	-	-	-	Karnataka	16.	Nili ravi	591001	129411	108659	Punjab, Haryana,
7.	Gojri	-	-	-	Punjab and Himachal Pradesh	17.	Pandharpuri	272122	287751	376182	Maharashtra
8.	Jaffarabadi	1843727	571077	1113789	Gujarat, Maharashtra,	18.	Surti	2982514	1886280	1122735	Gujarat, Rajasthan,
9.	Kalahandi	146052	115213	18123	Odisha	19.	Toda	50404	3003	14497	Tamil Nadu
10.	Luit (Swamp)	-	-	-	Assam and Manipur						

red coat colour are Gir, Motu, Rathi, Red Kandhari, Red Sindhi and Sahiwal. The Rathi breed had white spots or patches on brownish red coat. Purnea cattle are available in red and white/grey coat colours. Dangi, Ponwar and Siri cattle possess black/white patches on their body. Dangi, Bargur and Nimari had red/white patches on their coat. Calves of Umblachery have red or brown coat colour at birth. Red/brown colour begins to change to grey at the age of 3-4 months and get completely grey colour at 6-8 months of age. Breeds found in hilly tracts like hill cattle of Uttarakhand (Badri) and Himachal Pradesh (Himachali Pahari) and indigenous cattle like Thutho Nagaland, Lakhimi of Assam, Malnad Gidda of Karnataka and Vechur of Kerala are found in multiple coat colours of black, red, brown, white and even fawn/golden (Pundir and Sharma, 2016).

Based on size of the body Vechur, Malnad Gidda, Punganur, Motu, Purnea, Kosali and Hill cattle are known as small type. The breeds Bachaur, Bargur, Binjarpuri, Dangi, Gaolao, Ghumsuri, Tho-Tho, Pullikulam, Kenkatha, Khariar, Krishna Valley, Nimari, Rathi, Siri, Umblachery, and Kherigarh are of medium size. The large size breeds include Ongole, Gir, Hariana, Deoni, Mewati, Nagori, Tharparkar, Kankrej, Malvi, Kangayam, Red Sindhi, Sahiwal, Khillar, Hallikar, Amritmahal, Gangatiri (Pundir and Sharma, 2016).

Based on horn size Kankrej had the thickest horns. Long horns were available in Hallikar, Bargur, Khillar, Pullikulam, Kangayam, Amritmahal, Krishna Valley

breeds. The short type horns breeds are Hariana, Siri, Rathi, Ongole, Bachaur, Nagori, Gaolao, Mewati, Krishna Valley, Ponwar, Binjarpuri and Gangatiri.

The trends in percent in different cattle breeds population during 2013-2022 are presented in Fig. 2 and 3. There are 15 breeds showing increasing trends. The maximum increase was observed in Vechur breed followed by Red Sindhi, Bachur and Punganur. Rathi breed has shown smallest increasing trend as 1.49% over the same period. There are 22 breeds showing declining trends in their populations during the years 2013-2022. The maximum decline was observed in Khariar breed followed by Kenkatha, Red Kandhari and Kherigarh. Amritmahal breed showing almost static population over the same period.

The crossbred/exotic cattle are distributed in larger area of the country. The main breeds in this category include Holstein Friesian and Jersey, however, exotic as well as crossbreds of some other breeds like Brown Swiss have also been reported though comparatively less in number. Some of the synthetic strains/breeds have been developed by using exotic germplasm. These include Frieswal, Karan Swiss, Karan Fries, Sunandini, Vrindavani, Taylor, Jersindh etc.

Buffalo Diversity

Two types of buffaloes are recognized based on their phenotypes and karyotypes i.e. the river/water buffalo (2N=50) found in Indian subcontinent, in Middle-East

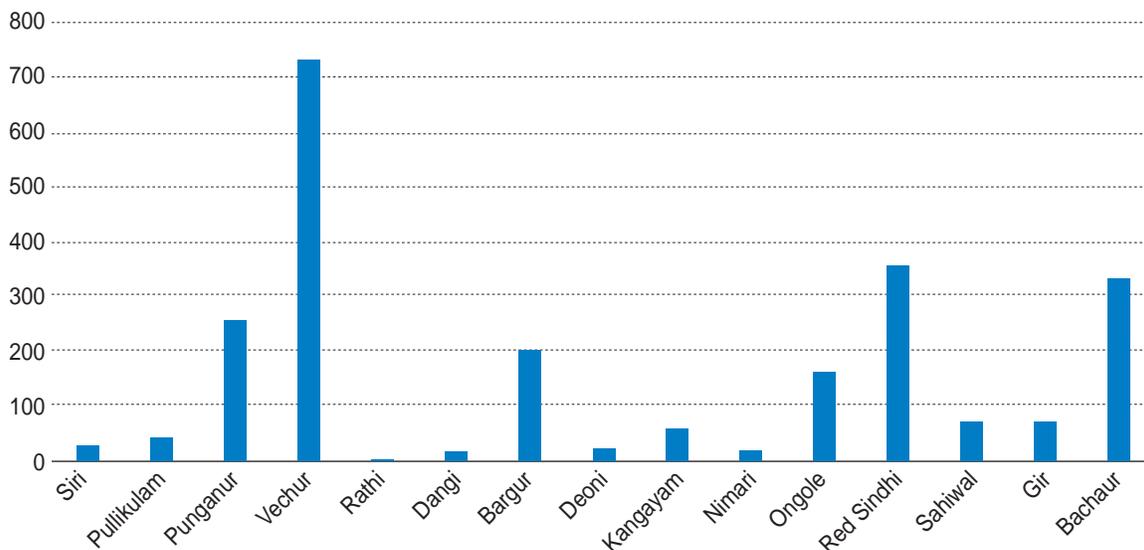


Fig. 2. Cattle breeds showing increasing trends during the years 2013-2022

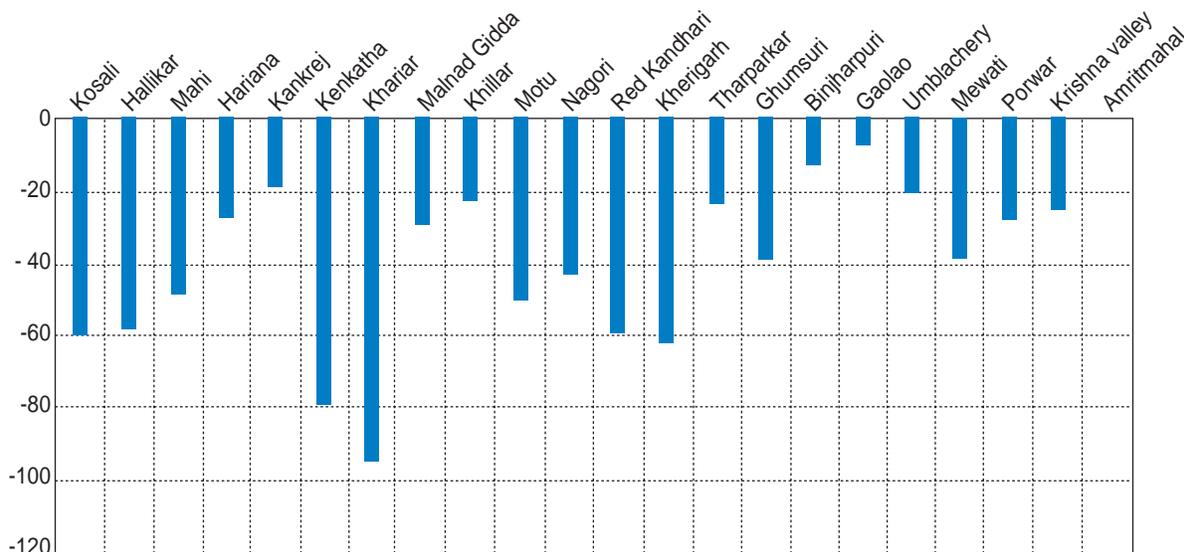


Fig. 3. Cattle breeds showing decreasing trends during the years 2013-2022

and Eastern Europe especially Italy and Bulgaria and the swamp buffalo (2N=48) found in China and South-East Asian countries. These two subspecies were originated and domesticated independently, the river/water buffalo in the Indus valley and /or Euphrates and Tigris valley some 5000 years back, and the swamp buffalo in China where it was domesticated at least 4000 years ago in association with the emergence of rice cultivation. In spite the two subspecies are cytogenetically different but do hybridize among themselves, but the hybrids (2N=49) produced have relatively low fertility or infertile.

Buffalo breeds, their distribution and population over different years are presented in Table-1. There are

19 registered breeds of buffalo in the country and can be classified into six major groups as follows.

- (i) Murrah group comprises Murrah, Nili-Ravi and Gojri which have the home tract in Haryana and Punjab, respectively. Gojri has the ability to graze on hill tops and has characteristic “Pattiah wale seengh”.
- (ii) Gujarat group comprising Jaffarabadi, Surti, Mehsana and Banni from Gujarat.
- (iii) Uttar Pradesh group has Bhadawari.
- (iv) Central India group includes Nagpuri and Pandharpuri breed in Maharashtra, Kalahandi, Manda and Chilika in Odisha, Chhattisgarhi buffalo from Chhattisgarh.

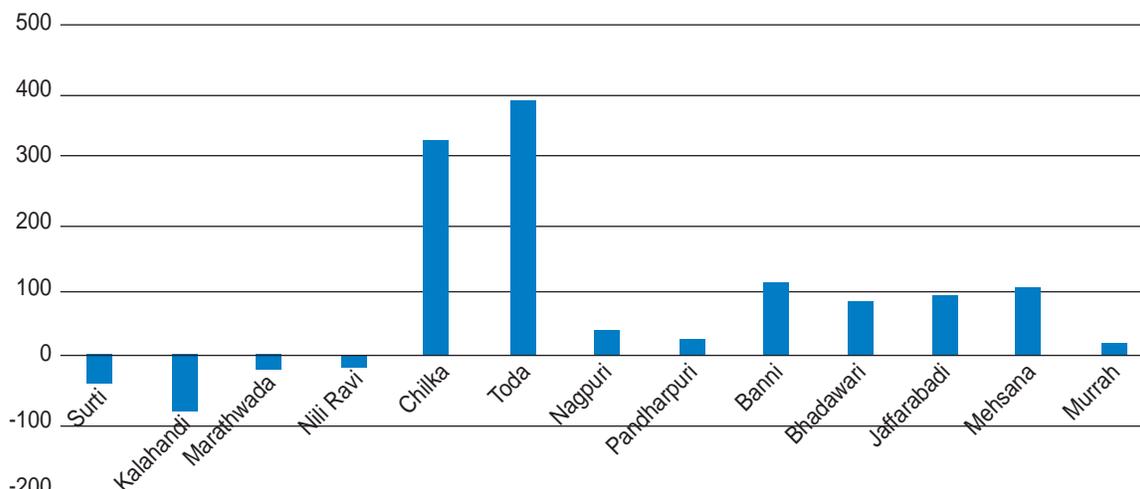


Fig. 4. Buffalo breeds showing trends during the years 2013-2022

- (v) South India group comprises Toda and Bargur buffalo in Tamilnadu, Dharwadi in Karnataka.
- (iv) North-Eastern group includes Luit and Swamp buffaloes mainly available in Assam.

The large size buffaloes include Murrah, Nili-Ravi and Jaffarabadi, medium sized are Mehsana, Marathwada, Nagpuri, Pandharpuri, Bhadawari, Surti and Toda. Swamp buffaloes are available in north-east states of the country. India possesses the best milch buffalo breeds of the world like Murrah, Nili Ravi and Jaffarabadi. Bhadawari breed is famous for high fat percentage in milk. Surti is the smallest size buffalo available in the country. Buffaloes are also used for work and meat production. There are several breeds in India which have regional importance and add to economic value of the farming community viz. Bhadawari and Tarai in Uttar Pradesh, Nagpuri and Pandharpuri in Maharashtra; Parlakhemundi, Manda, Jerangi, Kalahandi, Sambalpur in Orissa and Andhra Pradesh, Toda in Tamil Nadu and South Kanara in Karnataka and Kerala. Mehsana breed has been developed from grading up of Surti buffaloes with Murrah in north Gujarat. Similarly continued grading up of local non-descript buffaloes with Murrah breed in Krishna and Godawari District of Andhra Pradesh has resulted into a strain popularly known as Godavari (Pundir and Sharma, 2016).

All the buffalo breeds can be divided in to two categories based on their coat colour black (Murrah, Mehsana, Banni and Nili-ravi) and grey (Surti, Toda and Chilka). Based on body size they can be further divided in to 3 different categories like large (Jaffarabadi, Murrah

and Banni), medium size (Bhadawari, Marathawada and Pandharpuri) and small (Surti and Chilka). Based on horn patterns buffalo breeds can be divided in to four different categories like short and tightly curved (Murrah, Banni and Mehsana), large but downward (Jaffarabadi, Marathawada and Nagpuri), medium (Bhadawari) and large upward (Toda and Chilka).

Among the 13 breeds of buffalo included breed census, 4 are showing declining trends during the 2013-2022 i.e. Surti, Kalahandi, Marathawadi and Nili-ravi. Maximum declining trends was observed in Kalahandi buffalo. Chilka and Toda breeds has shown more than 300% increase in their population. Murrah breed is showing an increase of 21.91% in their population during the same period.

Strategies for Improving Bovines

For improving of indigenous cattle and buffalo following strategies may be adopted.

- ◆ Characterization of non-descript population and their registration as distinct breed, if eligible.
- ◆ Livestock Census may be conducted breed wise accurately which will help in formulation of appropriate breeding strategies for genetic improvement programs.
- ◆ Establish /strengthening of nucleus breeding farms for all registered breeds in the respective breeding tract to produce superior germplasm for breeding.
- ◆ Performance recording may be imitated in field/ farmer herds.

- ♣ Increasing the AI coverage in field conditions.
- ♣ Genetic improvement through Open Nucleus Breeding System (ONBS) and Progeny Testing of bulls.
- ♣ Multiplication of elite germplasm by different reproductive technologies like cloning, embryo transfer etc.
- ♣ Value addition of different bovine products.
- ♣ Creation of awareness in masses for uniqueness of indigenous bovine germplasm.
- ♣ Efforts should be made for conservation, those breed needed conservation.
- ♣ Establishing *ex situ* and *in situ* conservation units in respective breeding tracts for those breeds needed conservation

- ♣ Somatic cells at National Gene Bank for repository.
- ♣ Establishment of breed society for all registered breed.

References

- Anonymous (2007) Livestock Census, Department of Animal Husbandry and Dairying, MoA, GOI, New Delhi.
- Anonymous (2013) Livestock Census, Department of Animal Husbandry and Dairying, MoA, GOI, New Delhi.
- Anonymous (2019) Basic Animal Husbandry Statistics, Department of Animal Husbandry and Dairying, MoA, GOI, New Delhi.
- Anonymous (2022) Breed Survey Report, Department of Animal Husbandry and Dairying, MoA, GOI, New Delhi.
- Pundir RK and A Sharma (2016) Indian breeds of cattle and buffalo: Huge Sources of Bovine Germplasm. Dairy India, 7th Edition.

Goat and Sheep Genetic Resources of India

Rajeev AK Aggarwal*, **HK Narula**, **SP Dixit**, **Reena Arora**, **Dinesh Kumar Yadav** and **Indrajit Ganguly**

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

Small ruminants (Goat and Sheep) form an integral part of Indian agriculture and widely distributed across all agro-ecological zones. There are 148.88 million goats and 74.26 million sheep in India and are reared mainly by small and marginal farmers. They contribute significantly to rural economy as they can sustain on paltry input resources and harsh environmental conditions. There are 44 and 34 registered breeds of sheep and goat respectively, beside large number of non-descript animals. Due to intermixing among breeds in the field, breed purity is being diluted. Hence there is need to develop suitable breeding policy for genetic improvement and conservation of goat and sheep by different states.

Introduction

Small ruminants are important livestock, which are largely associated with rural people in India and play an important role for keeper's economy. More than 70 percent of landless, marginal and small farmers of rural India rear them. The small ruminants rearing in comparison to other livestock species is highly beneficial for farmers as these species are generally reared on zero or low input system. They are much useful in arid and semi-arid areas where crop cultivation is a risk prone enterprise and play a big role in supporting the livelihood system and nutritional security. India is a rich repository of sheep and goat genetic resources, having 44 and 34 registered breeds of sheep and goat respectively (Fig. 1). These breeds have evolved through natural selection and selective breeding by rearers for adaptation to specific agro-ecological conditions. There is great variation in adaptability, productivity, feed utilization and disease resistance etc. of indigenous sheep and goat breeds of India. There are about 223.14 million goats and sheep (20th Livestock census, 2019) in country and in spite of high slaughter rate and inbuilt prejudice, both goat and sheep populations have been growing for the past few decades. Shrinkage in grazing areas, lack of veterinary health services, conflicts with the agriculturalists, lack of education etc. are the main problems faced by the pastoral communities.

Goat Genetic Resources

Livestock population of the country as per Livestock census, 2019 has been estimated as 536.76 million,

spread over different states and UTs of India (20th Livestock census-2019). Goats contribute 27.73% to the total livestock population and is next to the cattle (36.04%). The population of Indian goat is 148.88 million (Livestock Census, 2019) which is 10.14% higher than population as per Livestock Census, 2012. Among the different states, Rajasthan has highest goat population (20.84 million) followed by West Bengal and Uttar Pradesh, respectively. This increase in goat numbers in the country showed its popularity and usefulness due to their economic significance, low input resources, small generation interval, higher proliferation and adaptability to varied climatic conditions.

India is a good repository of goat genetic resources consisting of 34 recognized breeds (Table 1) and a good number of non-descript population. Out of 34 breeds of goat, 8 breeds viz. Beetal, Gohilwadi, Jakhrana, Jamunapari, Kahmi, Mehsana, Surti and Zalawadi are of dairy type, producing more than 150 kg of milk on an average per lactation. More than 75% of the breeds (26/34) in the country are primarily meat producers as indicated in Table 1. Dressing percentage of different breeds varies from 43 to 58.4%. There are only two breeds viz. Changthangi and Chegu which are pashmina producers. The pashmina produced by Changthangi goat is superior to that of Chegu in terms of fiber diameter and staple length, the important characteristics of Pashmina hair. More than half of the goat breeds in the country are early breeders, demonstrating early sexual maturity and first kidding at an age of less than 18 months.

*Author for Correspondence: Email-Rajeev.Aggarwal@icar.gov.in

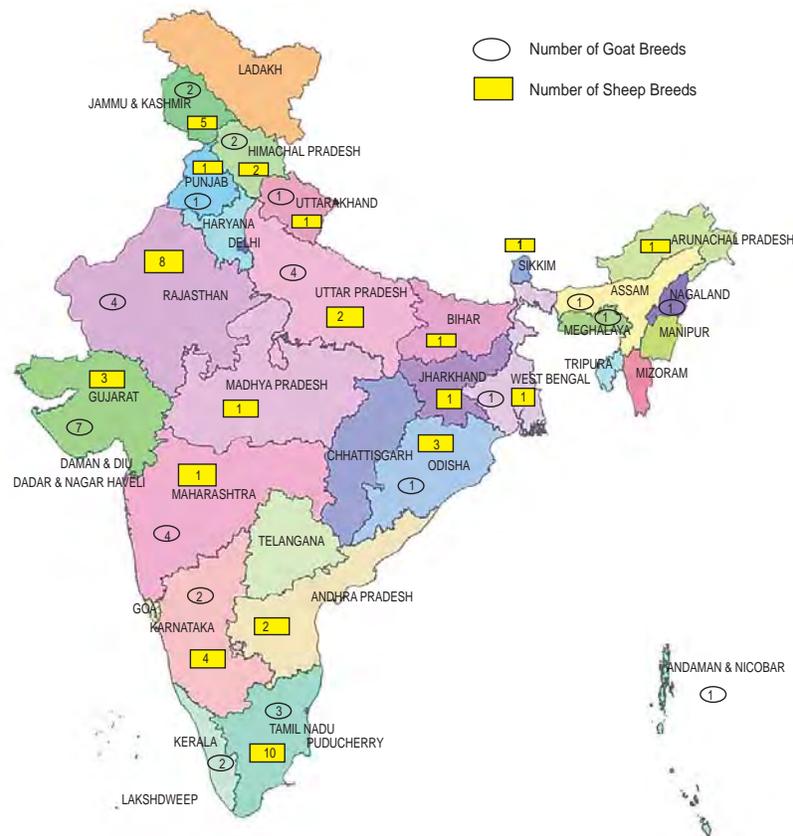


Fig. 1. Geographical Distribution of Goat and Sheep Breeds in India

Reproduced by permission of Surveyor General of India on behalf of Govt. of India under License No. BP15CDLA452. All rights reserved

Sheep Genetic Resources

Sheep is an important livestock species in India and there are about 74.26 million sheep, which contribute greatly to the agrarian economy by providing gainful employment and income to large number of rural poor especially socially backward, marginal and landless labourers. Sheep sector ensure self-employment and family labour under extensive system and act as cushion at the time of drought and famine for keepers. The population of sheep in 2019 increased by 14.1 % over 2012 census which showed the importance of sheep in present scenario of livestock husbandry. India is gifted with vast biodiversity of indigenous sheep that has multi-facet utility for wool, meat, milk, skin, manure and forms a valuable constituent of the rural economy especially in the arid, semi-arid and mountainous areas of the country. Having 44 registered indigenous breeds, sheep contributes around 7-8 % in meat and 100% in wool production. Out of total raw wool produced in the country about 70% is carpet grade, 20% coarse grade and 10% apparel grade. Sheep population is generally

found in the arid and semi-arid areas of western India, deccan plateau and western himalayas. About 60% of sheep in the country are concentrated in six states, namely Andhra Pradesh, Rajasthan, Karnataka, Tamil Nadu, Jammu & Kashmir and Maharashtra.

All 44 distinguished registered sheep breeds are well adapted to specific environment in different agro-climatic region of the country. The classification of sheep has also been based on major products i.e. a) apparel wool, b) carpet wool, c) meat and carpet wool and d) meat types. Sheep breeds based on their geographical presence are also divided into four agro ecological regions.

- (a) **Southern Peninsular Region:** This region is semi-arid in the central peninsula and hot and humid along the coast and comprising highest number of sheep. It comprises the states of Maharashtra, Andhra Pradesh, Karnataka, Tamil Nadu, Kerala and other territories in the central area. This region having coarser wool breed mainly reared for meat production.

- (b) **North Western arid and semi-arid region:** This region has second largest population of sheep and comprises the state of Punjab, Haryana, Rajasthan, Gujarat, plains of Uttar Pradesh and Madhya Pradesh having carpet wool type sheep breeds.
- (c) **North Temperate Region:** This region comprises Jammu and Kashmir, Himachal Pradesh and hilly

regions of Uttaranchal. This region consist mainly wool/ apparel type sheep breeds.

- (d) **Eastern Region:** This region is mostly hot and humid, except for some parts of eastern states, which are sub-temperate and comprises the states of Bihar, Jharkhand, West Bengal, Orissa, Assam, Meghalaya, Arunachal Pradesh, Mizoram, Manipur,

Table 1. Detail of different breeds, distribution, population, utility and accession number of registered breeds of goat

S.No.	Breed	Distribution	Population*	Utility	Accession Number
1.	Attapady	Kerala	31182	Meat	INDIA_GOAT_0900_ATTAPADYBLACK_06001
2.	Barbari	Uttar Pradesh and Rajasthan	4759305	Milk, Meat	INDIA_GOAT_2017_BARBARI_06002
3.	Beetal	Punjab	1234760	Milk, Meat	INDIA_GOAT_1600_BEETAL_06003
4.	Black Bengal	West Bengal	27661976	Meat, Skin	INDIA_GOAT_2100_BLACKBENGAL_06004
5.	Changthangi	Jammu and Kashmir	205940	Hair (Pashmina), Meat	INDIA_GOAT_0700_CHANGTHANGI_06005
6.	Chegu	Himachal Pradesh	2356	Hair (Pashmina), Meat, Pack	INDIA_GOAT_0600_CHEGU_06006
7.	Gaddi	Himachal Pradesh	738425	Hair (Pashmina), Meat	INDIA_GOAT_0600_GADDI_06007
8.	Ganjam	Orissa	211478	Meat, Milk	INDIA_GOAT_1500_GANJAM_06008
9.	Gohilwadi	Gujarat	288453	Meat, Milk	INDIA_GOAT_0400_GOHILWADI_06009
10.	Jakhrana	Rajasthan	655582	Meat, Milk	INDIA_GOAT_1700_JAKHRANA_06010
11.	Jamunapari	Uttar Pradesh	2555965	Meat, Milk	INDIA_GOAT_2000_JAMUNAPARI_06011
12.	KanniAdu	Tamilnadu	1445588	Meat	INDIA_GOAT_1800_KANNIADU_06012
13.	Kutchi	Gujarat	584538	Meat, Milk	INDIA_GOAT_0400_KUTCHI_06013
14.	Malabari	Kerala	1104305	Meat, Milk	INDIA_GOAT_0900_MALABARI_06014
15.	Marwari	Rajasthan	5041776	Meat, Milk	INDIA_GOAT_1700_MARWARI_06015
16.	Mehsana	Gujarat	422509	Meat, Milk	INDIA_GOAT_0400_MEHSANA_06016
17.	Osmanabadi	Maharashtra	3597071	Meat	INDIA_GOAT_1100_OSMANABADI_06017
18.	Sangamneri	Maharashtra	163091	Meat, Milk	INDIA_GOAT_1100_SANGAMNERI_06018
19.	Sirohi	Rajasthan and Gujarat	1952116	Meat, Milk	INDIA_GOAT_1704_SIROHI_06019
20.	Surti	Gujarat	231194	Meat, Milk	INDIA_GOAT_0400_SURTI_06020
21.	Zalawadi	Gujarat	408450	Meat, Milk	INDIA_GOAT_0400_ZALAWADI_06021
22.	Konkan Kanyal	Maharashtra	16892	Meat	INDIA_GOAT_1100_KONKANKANYAL_06022
23.	Berari	Maharashtra	84823	Meat	INDIA_GOAT_1100_BERARI_06023
24.	Pantja	Uttarakhand and Uttar Pradesh	28728	Meat, Milk	INDIA_GOAT_2420_PANTJA_06024
25.	Teressa	Andaman & Nicobar	3362	Meat	INDIA_GOAT_3300_TERESSA_06025
26.	Kodi Adu	Tamil Nadu	399924	Meat, Milk	INDIA_GOAT_1800_KODIADU_06026
27.	Salem Black	Tamil Nadu	491992	Meat, Skin, Manure	INDIA_GOAT_1800_SALEMBLACK_06027
28.	Sumi-Ne	Nagaland	1509	Fibre	INDIA_GOAT_1400_SUMINE_06028
29.	Kahmi	Gujarat	NA	Meat, Milk	INDIA_GOAT_0400_KAHMI_06029
30.	Rohilkhandi	Uttar Pradesh	NA	Meat, Milk	INDIA_GOAT_2000_ROHILKHANDI_06030
31.	Assam Hill	Assam and Meghalaya	NA	Meat	INDIA_GOAT_0213_ASSAMHILL_06031
32.	Bidri	Karnataka	NA	Meat	INDIA_GOAT_0800_BIDRI_06032
33.	Nandidurga	Karnataka	NA	Meat	INDIA_GOAT_0800_NANDIDURGA_06033
34.	Bhakarwali	Jammu and Kashmir	NA	Meat, Milk, Hair	INDIA_GOAT_0700_BHAKARWALI_06034
Indigenous Goats			54323290		
Non descript Goats			94561496		
Total Goats			148884786		

*Indigenous pure plus graded; Source: ICAR-NBAGR and 20th livestock census

Table 2. Detail of different breeds, distribution, population, utility and accession number of registered breeds of sheep

S. No.	Breed	Distribution	Population*	Utility	Accession Number
1	Balangir	Orissa	80435	Meat, Carpet Wool	INDIA_SHEEP_1500_BALANGIR_14033
2.	Bellary	Karnataka	4275218	Wool, Meat	INDIA_SHEEP_0800_BELLARY_14019
3.	Bhakarwal	Jammu and Kashmir	187967	Coarse carpet wool	INDIA_SHEEP_0700_BHAKARWAL_14001
4.	Bonpala	Sikkim	68831	Meat, Wool	INDIA_SHEEP_2200_BONPALA_14034
5.	Changthangi	Jammu and Kashmir	71901	Meat, Wool	INDIA_SHEEP_0700_CHANGTHANGI_14002
6.	Chevaadu	Tamilnadu	67873	Meat, skin, manure, religious, cultural	INDIA_SHEEP_1800_CHEVAADU_14041
7.	Chokla	Rajasthan	382197	Fine carpet quality fleece	INDIA_SHEEP_1700_CHOKLA_14008
8.	Chottanagpuri	Jharkhand	315069	Meat, wool	INDIA_SHEEP_2500_CHOTTANAGPURI_14035
9.	Coimbatore	Tamilnadu	100317	Meat, wool	INDIA_SHEEP_1800_COIMBATORE_14020
10.	Deccani	Andhra Pradesh, Maharashtra	2383932	Meat	INDIA_SHEEP_0111_DECCANI_14021
11.	Gaddi	Himachal Pradesh	666915	Meat, wool	INDIA_SHEEP_0600_GADDI_14003
12.	Ganjam	Orissa	61479	Meat, wool	INDIA_SHEEP_1500_GANJAM_14036
13.	Garole	West Bengal	73582	Meat	INDIA_SHEEP_2100_GAROLE_14039
14.	Gurez	Jammu and Kashmir	25714	Milk	INDIA_SHEEP_0700_GUREZ_14004
15.	Hassan	Karnataka	737401	Meat, wool	INDIA_SHEEP_0800_HASSAN_14022
16.	Jaisalmeri	Rajasthan	680173	Meat, wool	INDIA_SHEEP_1700_JAISALMERI_14009
17.	Jalauni	UP and MP	42931	Meat, wool	INDIA_SHEEP_2010_JALAUNI_14010
18.	Karnah	Jammu and Kashmir	3121	Meat, wool	INDIA_SHEEP_0700_KARNAH_14005
19.	Katchaikatty Black	Tamilnadu	1900	Ram fighting during festivals, hair, meat	INDIA_SHEEP_1800_KATCHAIKATTYBLACK_14040
20.	Kenguri	Karnataka	1286284	Meat	INDIA_SHEEP_0800_KENGURI_14023
21.	Kendrapada	Odisha	10974	Meat, prolificacy	INDIA_SHEEP_1500_KENDRAPADA_14042
22.	Kilakarsal	Tamilnadu	46229	Meat	INDIA_SHEEP_1800_KILAKARSAL_14024
23.	Madras Red	Tamilnadu	297506	Meat	INDIA_SHEEP_1800_MADRASRED_14025
24.	Magra	Rajasthan	131689	Meat, wool	INDIA_SHEEP_1700_MAGRA_14011
25.	Malpura	Rajasthan	209534	Meat, wool	INDIA_SHEEP_1700_MALPURA_14012
26.	Mandya	Karnataka	250038	Meat (Excellent quality)	INDIA_SHEEP_0800_MANDYA_14026
27.	Marwari	Rajasthan and Gujarat	2870057	Meat, wool	INDIA_SHEEP_1704_MARWARI_14013
28.	Mecheri	Tamilnadu	1242742	Skin (finest quality), meat	INDIA_SHEEP_1800_MECHERI_14027
29.	Muzzafarnagri	UP and Uttarakhand	41760	Meat, wool	INDIA_SHEEP_2024_MUZZAFARNAGRI_14014
30.	Nali	Rajasthan	250343	Carpet Wool	INDIA_SHEEP_1700_NALI_14015
31.	Nellore	Andhra Pradesh	14043835	Meat	INDIA_SHEEP_0100_NELLORE_14028
32.	Nilgiri	Tamilnadu	3144	Apparel Wool	INDIA_SHEEP_1800_NILGIRI_14029
33.	Patanwadi	Gujarat	879620	Meat, wool	INDIA_SHEEP_0400_PATANWADI_14016
34.	Poonchi	Jammu and Kashmir	20232	Wool	INDIA_SHEEP_0700_POONCHI_14006
35.	Pugal	Rajasthan	170450	Meat, wool	INDIA_SHEEP_1700_PUGAL_14017
36.	Ramnad White	Tamilnadu	396856	Meat	INDIA_SHEEP_1800_RAMNADWHITE_14030
37.	Rampur Bushair	Himachal Pradesh	18239	Pelt, Meat, Wool	INDIA_SHEEP_0600_RAMPURBUSHAIR_14007
38.	Shahbadi	Bihar	60295	Meat, wool	INDIA_SHEEP_0300_SHAHBADI_14037
39.	Sonadi	Rajasthan	39373	Meat, wool	INDIA_SHEEP_1700_SONADI_14018
40.	Tibetan	Arunachal Pradesh	318	Meat, wool	INDIA_SHEEP_2300_TIBETAN_14038
41.	Tiruchi Black	Tamilnadu	21605	Meat	INDIA_SHEEP_1800_TIRUCHIBLACK_14031
42.	Vembur	Tamilnadu	102134	Meat	INDIA_SHEEP_1800_VEMBUR_14032
43.	Panchali	Gujarat	NA	Milk, meat, manure, wool	INDIA_SHEEP_0400_PANCHALI_14043
44.	Kajali	Punjab	NA	Wool, Meat	INDIA_SHEEP_1600_KAJALI_14044
Indigenous Sheep			32620213		
Non descript Sheep			37552269		
Total Sheep			70172482		

*Indigenous pure plus graded; Source: ICAR-NBAGR and 20th livestock census

Tripura, Nagaland and Sikkim. The breeds of this region are mainly of meat/ wool type.

Future Prospective and Action Points

Small ruminants rearing contributes significantly to the sustenance of small and marginal landholders, and landless rural people in India. They also play a significant role in ensuring food and nutritional security for the families of millions of resource poor rural households. In spite of having rich repository of sheep and goat breeds there is substantial non-descript population of these species, which necessitates undertaking characterization of these animals on fast track. Breeding policy for multiplication and conservation of small ruminants by different states should also be framed and conservation of indigenous breeds should be promoted. The issues

like decreasing grazing resources, lack of veterinary health services, conflicts with the agriculturalists, organized marketing system, availability of superior breeding bucks/rams etc. need to be suitably addressed involving stakeholders to popularize and improve goat and sheep in field.

References

- BAHS (2020) Basic Animal Husbandry Statistics. Government of India. Ministry of Fisheries, Animal Husbandry and Dairying. Department of Animal Husbandry and Dairying, Krishi Bhawan, New Delhi.
- ICAR-NBAGR Animal Genetic Resources of India (AGRI-IS). ICAR-National Bureau of Animal Genetic Resources.
- CSWRI-50 Years of Research Contribution (2012) CSWRI, Avikanagar (Edited by AK Shinde, CP Swarankar and LLL Prince)

Poultry Genetic Resources of India and its Role in Rural Poultry Production

AK Mishra

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

Poultry is one of the fastest growing segments of Indian agriculture. Scientific, technological advancements and introduction of high yielding germplasm have led to great expansion of poultry industry in last few decades. However, now a day's indigenous breeds are gaining importance due to increasing demand for their quality products. The peoples are ready to pay higher cost to egg and meat of indigenous breeds due to their quality and taste. Native breeds are acceptable by the local people since it suits their socio-cultural way of life. The traditional system of poultry production is still prevalent in the rural areas and is the backbone of rural poultry production. Presently the poultry farming have two distinct facets- the commercial – high input intensive production and extensive low-input rural/backyard poultry production. Each one is equally important, but has widely diverse input needs, especially the type of germplasm. Chicken and ducks are two major avian species in India used for production of eggs and meat both under un-organized and organized sector. The wide variation of agro-climatic conditions has facilitated the evolution and development of many poultry breeds out of which 19 chickens, 2 duck and 1 of geese have been registered.

Introduction

Traditional poultry farming is as old as its civilization and it occupies a pivotal position in India due to its huge potential to bring about rapid economic growth with low investment. Presently poultry sector is recognized as an organized and science based industry with tremendous potential to fight poverty, unemployment and malnutrition. The indigenous breeds are well known for their adaptability to local climatic geographical conditions and resistance to tropical diseases and considered as the gold mines of genomes and major genes but popularity of native breeds is decreasing day by day for being low egg producers, slow growers and smaller egg size etc. They are, however, good brooders and efficient foragers. The purity of native breeds has facing the problems of extinction due to large scale introduction of exotic breeds and exotic inheritance based rural poultry varieties and strains under various rural development programmes. However, rearing of indigenous breeds in backyard poultry production system is a conventional activity of a large number of the landless and small farmers. India and the neighboring countries are considered to be the original home of the well-known Red Jungle Fowl (*Gallus gallus*) from which the present day domestic birds have descended.

Chicken Genetic Resources

There are 19 registered breeds of chicken distributed in different parts of the country. The indigenous birds

have certain characteristics in common such as small compact body and body weight of about 1-1.5 kg. They vary greatly in their plumage pattern, comb type and body confirmation. The breast bone is low and well covered with feathers which lie close to the body. They generally lay brown shelled small eggs. The hens are broody. The birds are reared in free range backyard system. Scavenging with supplementation of kitchen waste is the most common feeding system. List of registered breeds and their details are given in Table 1. The indigenous breeds contribute to only 6.5% of total fowl population (807.85 m) of the country. As Breed Survey Report of 2019, the total populations of different breeds varied from 26615 (Nicobari) to Aseel (33680583). However, population of Miri, Daothigir, Kadaknath, Punjab Brown, Ankaleswar, Chittagong and Busra breeds was also above 10 lakhs. Three breeds namely Hansli, Kalasthi and Nicobari had population size less than 1 lakh.

Duck Genetic Resources

In India Duck farming is mainly concentrated in eastern and southern states. Coastal areas of Andhra Pradesh, Tamil Nadu, Kerala, West Bengal, Orissa, and certain parts of Assam and Jharkhand constitute the main habitat of ducks. Traditional duck rearing is mostly in the hands of poor farmers. Ducklings at the age of 6 weeks are allowed to forage in harvested paddy fields, wet land, back waters and irrigation channels. In south, some of

*Author for Correspondence: Email-anil.mishra2@icar.gov.in; anilmishra65@gmail.com

Table 1. Registered chicken breeds of India and their population

Breed / population	Place of origin and important characteristics
Aseel 33680583	Home tract is Khammam district of Andhra Pradesh, Koraput & Malkangiri of Odisha and Bastar and Dantiwara of Chattishgarh. It is game bird, known for its high stamina, majestic gait and dogged fighting qualities and large Body size. It is biggest in size among native breeds. The plumage colour are mostly Red (or Brown) and black. It has various (9) varieties having different plumage colours. The most popular varieties of Aseel are Peela, Yakub (black & red), Nurie, Kagar, Chitta, Jawa, Sabja, Teekar and Reza. The Aseel means "ASLI". Annual egg production ranges from 65 to 70 with average egg Weight of 41 g. The adult body weight ranges from 2.59 to 4.0 Kg.
Kadakhnath 2037475	Place of origin is Dhar and Jabua of MP, known as Kalamasi and characterized by presence of black pigments on external and internal surfaces. The black colour of muscles and tissues is due to deposition of melanin pigment which causes increase in protein and decrease of fat and muscle fibre. The skin, beak, shanks, toes and soles of feet are slate-like in colour. There are 3 varieties of Kadakhnath; Jet Black (black in colour), Pencilled (black with white feathers in neck) and Golden (basically black with golden feathers on head & neck). Egg production ranges from 85–90 per annum with average egg weight of 40 g.
Chittagong 1241000	The home tract is Meghalaya and Tripura. Presently available in parts of Bangladesh and also known as Malay. Large, heavy bird, strong hardly and quarrelsome temperament.
Ankaleshwar 1322006	Distributed in Bharuch and Narmada districts of Gujarat. The breed might have been named after the name of Ankleshwar in Bharuch district. Plumage colour ranges widely; a combination of white and light grey to brown. The golden colour are most prevalent. Annual egg production is approx. 80.
Ghagus 760964	Found in Kolar and adjoining parts of Bangalore (Karnataka) and Chittoor and Anantpur districts of AP. The birds might have derived its name by a peculiar sound. The predominant plumage colour is brown followed by black cocks have shining bluish black feathers on breast, tail and thighs. Neck is covered with golden feathers. Egg production ranges from 45–60 per annum with average egg weight of 42.25 g.
Busra 1218724	Reared by tribal people living all along on either side of boundary line of Maharashtra and Gujarat. Busra nomenclature might have come from Busrawal – a village in Sakri taluk of Dhule district or from name of a tree "Busrawal (Marathi)/Bahawa (Advasi dialect). Birds are small in size and have wide variation in body colour. Plumage is mostly white mixed with black feathers on neck, back, tail and reddish brown feathers on shoulders and wings. Annual egg production is ranges from 40 to 55.
Daothigir 3696257	Found in Bodoland region of Assam. It derives its name from the name of plant "Thigir"(Dillenia indica). This plant bears flowers of different colours similar to the plumage colour of these birds. In Bodo language "Dao" means bird and hence birds are known as Daothigir. Birds are small sized, compact but heavy and have long legs. Plumage is mostly black interspersed with white feathers. Egg production ranges from 60 to 70 per annum with average egg weight of 44 g.
Danki 168332	Distributed in Vizianagaram and adjoining parts of Srikakulam and Visakhapatnam districts of AP. Birds are mainly used for game (fighting) purpose during Sankrati. The predominant plumage colour is brown followed by black. Local people call these birds by different names on the basis of plumage colour: Khaki or sanwala (Black); dega (red); parla (brick color), Satua (white) and pingle (spotted). This is fairly heavy breed, glossy and lustrous plumage, single comb, long necks and legs. Egg production ranges from 25-35 per annum with average egg weight of 46 g.
Kalasthi 49736	Distributed in Chittoor and adjoining parts of Nellore and Cuddapah districts of Andhra Pradesh. Breed might have been named after the name of the area i.e. Sri Kalasthi in Chittoor district. The predominant plumage colour is bluish black followed by brown. Egg production ranges from 30–40 2 per annum with average egg weight of 43 g.
Harringhata Black 455760	Distributed in Nadia and North 24 Pargana districts of West Bengal. Plumage colour is Black; some cocks have brown feathers on neck and wings. Egg production ranges from 25-90 per annum with average egg weight of 36.53 g.
Kashmir Favorolla 597626	Locally known as "Kashir kukkar. Available in Anantnag, Baramullah, Budgam, Kupwada, Srinagar and Pulwama districts of J&K. The birds with plumage of all shades varying from Jet black, dark brown and golden to pure white are available. Egg production ranges from 2060-85 per annum with average egg weight of 45.76 g.
Miri 5411093	Home tract is Dhimaji, North Lakhimpur, Sibsagar and adjoin districts of Dibrugarh and Majhauil island in upper Assam. The name is derived after the name of tribe rearing them (Miri or Misisng). The local name is "Porog". The birds have no standard plumage colour and majority of the birds are white followed by brown and black. Egg production ranges from 50-65 per annum with average egg weight of 42.06 g.
Punjab Brown 1365800	Native tract is almost all districts of Punjab and Haryana. Plumage colour is mostly brown. Males usually have black spots/stripes on neck, wings and tail. Egg production ranges from 60 -80 per annum.
Tellichery 148705	The breed derives its name from the name of a place "Tellichery "in Kannur district of Kerala. The birds are distributed in Calicut district and surrounding areas in Kannur and Malappuram districts of Kerala and Mahe of Pandicherry. These birds are fast movers and are not easy prey for the predators. Birds are also thought to have some medicinal value. Plumage colour is black with shining bluish tinge on hackle, back and tail feathers. Few have golden mixed with bluish feathers on neck. Typical birds have blackish red comb. Egg production ranges from 60-80 per annum with average egg weight of 46 g.

Breed / population	Place of origin and important characteristics
Nicobari 26615	Originated in Nicobar group of Islands. They are small size birds with short legs, compact and round body. It is a brownish matty coloured hardy bird. It produces highest number of eggs under free-range condition with supplementary feeding among all the indigenous breeds of India. The average annual egg production varies from 110 to 235 with average egg weight of 49.06 g.
Mewari 114624	Distributed in Ajmer, Sirohi, Jaipur, Udaipur, Bhilwara, Dungarpur, Banswara, Rajsamand, Chittorgarh and Pratapgarh districts of Rajasthan. The plumage colour in females - Varied from light to dark brown and grey with pencil lining and in males- Bright gold and bronze feathers forming a "Shawl" or Cape over the back of the bird from neck to lower back. Egg production ranges from 37-52 per annum with average egg weight of 52.86 g.
Kaunayen 114828	Breeding tract consists of Thoubal, Imphal West, Imphal East and Bishnupur districts in the valley region. The predominant plumage color is black followed by brown (or red) with or without patches. Neck, breast and thighs are generally bare, hard and rose red colored in fighting cocks. Used for cock fighting. Egg production ranges from 25-40 per annum.
Hansli 91385	Reared in Mayurbhanj and Keonjhar districts of Odisha for game and meat purpose. Plumage is predominantly black. Males have golden yellow or red hackle and saddle feathers. Egg production ranges from 39 - 96 per annum with average egg weight of 46.5 g.
Uttara Not included in survey	Distributed in Kumaon region of Uttarakhand. Its plumage is black in colour. These birds have feathered shank which is not present in any other native breeds. About 18% of birds have bunch of feathers on head (crest/crown). The birds are more noisy and flighty. Annual egg production ranges from 125 to 160 with average egg weight of 51.33 g.

the duck farmers move with their flocks to different places in search of food for ducks depending upon the availability of harvested field. This system of rearing in the rural areas is mostly extensive. India has only two registered breed of duck namely Pati and Maithili.

Pati duck: Pati ducks are reared in backyard production system in rural areas of Assam. Plumage is dark brown in drakes with grayish black head; tail with black and white feathers. Ducks are solid brown. A white ring may or may not be present at neck in both sexes. The bill, shank and feet are predominantly yellow. Pati ducks are used for meat, egg and ritual sacrifices. The average body weight is 1.58 kg. The estimated population of Pati duck is 18.21 lakhs.

Maithili duck: Maithili ducks are distributed in Motihari, Sitamarhi, Madhubani, Araria, Kishanganj and Katihar districts of Bihar. These ducks have uniform light/dark brown feathers throughout the body. Circular spots on the feathers (Mosaic pattern) in ducks and dark brown to ash colour in drakes. Head is bright black to greenish black in drakes and brown in ducks. Body carriage is slightly upright and bill shape is horizontal. Average age at first egg is 191.12 days (range 159-223). Average annual egg production is 54.6 (range 33-71). Average egg weight is 49.53g. Body weight at 6 month of age is 1.18kg (range 1.12-1.24). Population size is approximately 46,000.

Geese Genetic Resources

Kashmir Anz is the only breed of geese being registered by the National Bureau of Animal Genetic Resources, Karnal. These cinnamon or white colored birds are reared

for meat, eggs, feathers, or as a hobby in areas located around the water bodies (lakes and wet lands) across the Jammu and Kashmir valley but are more abundant in districts of Srinagar, Bandipora, Ganderbal, Budgam and Baramulla. These geese are mostly found in and around the water bodies (Lakes and Wetlands) across the Valley but are more abundant in districts of Srinagar, Bandipora, Ganderbal, Budgam and Baramulla. Name Kashmir Anz is derived from two words- Kashmir representing the breeding tract of the breed and Anz meaning Geese in local language. Kashmir Anz geese are hardy, disease resistant and good foragers, requiring minimum inputs for rearing and management.

Indigenous Poultry and Rural Empowerment

Backyard production system has over the years contributed to a great extent to the agrarian economy of the country. For rural poultry production Indigenous breeds are well suited as these are well known for their tropical adaptability and disease resistance. In India two types of production system of poultry production exists:

- Rural/Backyard poultry production system
- Commercial poultry production

Eggs and chicken meat are produced under this system at low cost by scavenging birds in the backyard and without feed supplementation with commercial rations. It provides livelihood security to the underprivileged family in addition to securing the availability of protein rich food at cheaper cost. It is an old age practice in rural India and available in three forms (Singh, 2007).

- (a) Traditional scavenging system
- (b) Semi-scavenging system
- (c) Small-scale intensive system

The choice of system is largely determined by the availability of resources and inputs, which depends on the keeper's socio-economic circumstances. Requirements of the different systems are quite different from each other including the type of germplasm to be used (Singh, 2007).

Traditional backyard production system: This is one of the most sustainable production systems with hardly any dependence on external sources (including chicks). This system is cost effective and satisfies consumer preferences for eggs and meat of indigenous/local birds and cultural linkages. Traditional backyard system exists in two forms:

- **Un-improved backyard system:** Includes use of native birds, scavenging, no regular water or feed, little or poor night shelter, no vaccination and medication.
- **Improved backyard system:** Includes use of improved germplasm, scavenging, regular water, supplementary feeding, improved shelter, care of chicks in the early age, vaccination against prevalent diseases and deworming.

The birds utilized in the system are although poor in egg and meat production but are hardy, adaptable to tropical climate, have better disease resistance, scavenging for their own feed, can run and fly to escape predators and are capable of reproducing to supply replacement stock for the household flock. Number of birds to be reared in this system usually should not be more than 25.

Semi-scavenging system: This system is used for small flocks under partially controlled management and where the scavenged feed account for a substantial part of the total feed consumed. Birds are kept most of the times under confinement with supplemented diet. They are allowed to scavenge in a day partly and are managed for predator's protection. Number of birds to be reared under this system varies from 50–200.

Small-scale intensive system: This system consists of small to medium sized flocks kept by a family as a means of acquiring assets to defeat poverty. They represent quite a large proportion of the assets and income of

their owners, and are often financed from loans, but it is highly risky and dependent on good marketing facilities.

Suitability of Indigenous Chicken for Rural Poultry

The native breeds are good foragers, efficient mothers, require less cost and special care to grow and thus have characteristics essential for raising poultry under village conditions. Indigenous breeds enjoy the advantage of superiority over exotic breeds/strains due to following characteristics:

- Preference for coloured bird by the peoples.
- Capability of self-defence from predators due to its alertness, light body weight, longer shank length and aggressiveness.
- Thrive well under adverse environments like poor housing, management and feeding.
- Comparatively disease resistant to protozoon & ecto-parasites and have better immunocompetence.
- Possesses characteristics essential of scavenging chicken.
- Have better adaptability to extreme climatic conditions prevailing in the country.
- Comparatively hardier and require less health care than exotic birds.
- Having broodiness or self propagation.

Large scale introduction of exotic breeds/strains of poultry under various rural development programmes resulted in extinction of some of breeds and few are facing the danger of extinction. Hence, conservation of native breeds is of prime importance. High yielding germplasm have contributed significantly in the progress of commercial poultry production but most of the high yielding exotic breeds have temperate origin. India being a tropical country having vast hot and humid area around coastal regions is not suitable for full expression of its production potential. In the peak summer and humid months, high yielding stocks suffer from tropical stress manifested as low egg production and reduction in the feed efficiency.

Future Perspectives

- Besides, the huge growth, the Indian poultry sector has been chicken dominated which in turns ignoring the potentials of others diversified poultry species. Hence, policies for improving the other poultry

species viz. duck, geese, guinea fowl, quails etc. needs intensification.

- Conservation is a very costly affair, which needs proper planning, source of regular financing and follow-up the action plan. National institution, state Governments, State Agricultural Universities and commercial entrepreneurs should be included in the conservation work.
- Avian Genetic Resource Task Force should be established to plan the conservation strategies and its follow-up for recognised native breeds. Improvement and utilization of the native breeds are the only tools for its sustainability in the traditional production system.
- A net work programme having a research centre in each state would be a good proposition as the particular breed has acquired adaptability for the particular environment. These centers would conserve, evaluate, and improve the productivity of indigenous breed and provide improved native parent stocks to poultry farmers, Government poultry unit / private hatchery etc. and thus will helps the sustainability of the breed in their ecologies.
- All the States are requested to consider having a breeding policy for indigenous poultry and evolve measures to conserve native breeds.

References

- Mohapatra SC and SC Misra (2008) Poultry production in India: Threats and Opportunities, *In: proceedings of silver Jubilee Annual Conference and national Symposium of IPSACON-2008*, held at AAU Anand from December 10-12 2008, pp: 3-8.
- Singh RV and D Sharma (2005) Poultry Genetic Resources, Lecture given in “Short course on Advances in Quantitative and Molecular techniques for optimizing Poultry production, from 21 Feb. to 2 March 2005, held at CARI, Izatnagar sponsored by ICAR, New Delhi, pp: 2-5
- Singh DP (2007) Selection of suitable germplasm for rural poultry production. *In: Souvenir, National Symposium on “Recent trends in policy initiatives and technological interventions for rural prosperity in small holder livestock production systems”* organized by ISAPM at Sri Venkateshwara Veterinary University, Tirupati during 20-22, June, 2007. Pp. 110-114.
- Singh, DP, TS Johri, UB Singh, D Singh and R Narayan (2003) Tested model for sustainable organic chicken production. *In proceedings of International Seminar on ‘Downsizing Technology for Rural Development’, RRL, Bhubaneswar, India, October 7-9, 2003, Vol.II : 665-71.*
- Vij PK (2019) Indigenous Poultry Genetic Resources and its Role in Rural Empowerment, *In Management of Indigenous Animal Genetic Resources under field condition for Rural Empowerment & Livelihood Security*, KN Raja, AK Mishra and RK Vijn (Eds), published by ICAR-NBAGR, Karnal, 226 pp.
- Vision 2050. ICAR-Central Avian Research Institute, Izatnagar.

Conservation for Effective Management of AnGR in India

MS Tantia*, RAK Aggarwal and Rekha Sharma

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

Livestock sector forms an important livelihood activity for most of the farmers, supporting agriculture in the form of critical inputs, contributing to the health and nutrition of the household, supplementing incomes, offering employment opportunities, and finally being a dependable “bank on hooves” in times of need. India has vast Animal Genetic Resources (AnGR), which play a vital role in improving the socio-economic conditions of rural masses. AnGR is also of significant social and cultural importance, supplementing family incomes and generating gainful employment in the rural sector, particularly among the landless, small, marginal farmers and women. Global demand for livestock products is expected to double by 2050, mainly due to improvement in the worldwide standard of living. Meanwhile, extensive livestock production systems are being fast replaced with intensive and industrial production system due to change in land use, higher demand of livestock product and more and more keepers shifting to high producing improved breeds. The loss of a breed means the loss of a livelihood strategy and loss of indigenous knowledge. It also emphasizes the need for the active involvement of indigenous communities and the role of local knowledge and institutions in conservation.

Introduction

There are about 302.79 million bovines, 74.26 million sheep, 148.88 million goats and about 9.06 million pigs as per 20th Livestock Census in the country. Horses and Ponies, Mules, Donkeys and Camel population decreased by 45.6, 57.1, 61.23 and 37.1 percent, respectively, over previous Census. AnGR provide a range of products and services to their keepers and to the wider society – including food, fibre, transport and fuel. Most of AnGR are reared in extensive production system which contributes directly to food and livelihood security because they produce more valuable nutrients for humans, such as proteins, than they consume. AnGR rearing can contribute to farmer’s risk management of natural calamities in many ways. They are mobile, which increases survivability and may also be relatively omnivorous, and thereby able to survive dramatic effects on specific feed resources. Indigenous AnGR keepers require very little resources as they are mostly very well adapted to the local environments and can survive on meager feed resources without any housing needs, Indigenous AnGR keepers are able to earn their livelihood by selling animal products like milk, egg, wool, etc. and also by selling the animals for meat purposes.

There are 202 registered breeds comprising 50 cattle, 19 buffalo, 44 sheep, 34 goat, 7 horse, 9 camel, 3 donkey, 10 pig, 19 chicken, 3 dog, 2 duck and one each of Yak and Geese. All these breeds have been developed

over centuries by natural and manmade selection to meet the specific needs as per production systems and native environments. The defined breeds have become prominent during last two decades with various activities and effective awareness generation by ICAR-NBAGR. More and more populations in the country are now being characterized and registered as breeds. Proportion of non-descript animals in each species has declined significantly and purity within breeds is also increasing. Various State Animal Husbandry Departments (AHD) which are custodian of AnGR, are making effective strategies for conservation and improvement of their resources.

Attributes of indigenous animal genetic resources can be summarized as:

1. Large number of breeds in each farm animal species adapted to the specific agro-climatic conditions.
2. Diversified draft, milch and dual purpose cattle breeds. The draft breeds can significantly contribute in agricultural operations to save fossil fuels.
3. Adaptability of germplasm to diverse changing climatic conditions of hot arid, humid tropical and temperate climates and better resistance to parasites and diseases.
4. Capability to survive and produce on coarse and poor quality feed and fodder resources (low input).

*Author for Correspondence: Email-Madhu.Tantia@icar.gov.in

5. Availability of best breeds of buffaloes, a multipurpose farm animal species.
6. Large amount of ITK available with the livestock keepers for management of AnGR.
7. Seasonal migration of nomadic pastoralists help overcome adverse conditions especially during winter and rainy seasons which enable them to sustain the breed population maintained by them.

Conservation aims to maintain the option value of genetic diversity, it is therefore a priority of the Global Plan of Action for Animal Genetic Resources. Conservation measures for threatened breeds have already been established in some countries. Most current conservation programmes are based in developed countries with strong collaboration between gene banks and animal breeding organizations. In developing countries, the focus is typically on *in vivo* conservation. Realization that AnGR are at risk of being lost has alarmed the researchers and planners which stimulated National livestock conservation efforts. The need for conservation is based on economic, cultural, and ecological values; unique biological characteristics; shifts in market demand; and research needs. Livestock breeds are not biological taxa but rather represent the outcome of social processes and are therefore unlikely to survive outside the social contexts and production systems that formed them. Conservation of ecosystem/production system guarantees breed survival.

Traditional pastoralists have often tended to foster biodiversity, in both plants and animals. Many pastoral societies have developed elaborate systems that result in the preservation of genetic resources. Pastoralists have deliberately developed livestock to meet different needs and conditions.

In the changing socio-economic scenario, it is becoming increasingly difficult to save local breeds specially those which are low producing. Major reasons are increased pressure on land to meet food requirement, the need for higher income per unit land/input to sustain better quality of life, shift of livestock activity from subsistence occupation to commercial entrepreneurship etc. The changing scenario calls for breeds with high performance to have higher profits. Sustaining low producing local breeds with poor economic viability is therefore really a challenge. Use of few specialized breeds with optimized specific production traits has lead to narrowing of the genetic base, as native breeds

and species are neglected in response to market forces. Declining livestock diversity may have adverse affect on our capacity to mitigate the enormous challenges posed by climate change and emerging diseases. We may need to rely back on the adaptability and potential of indigenous animal genetic resources to face an uncertain future.

Commercial breeds of livestock possess greater genetic variability than most crop varieties do. This diversity allows intensification of selection within breeds to be a fruitful approach for improving livestock productivity. However, if continued emphasis on breed replacement and increasing selection intensity (e.g. for greater productivity) take place at the expense of maintenance of genetic diversity, including the advantages of disease resistance and environmental adaptation, there may be significant long-term costs. As an example, Holstein cattle have become the pre-eminent dairy breed world-wide and have enjoyed sustained improvements in milk production potential, but only at the cost of declining genetic diversity within the breed. These losses weaken the potential of breeding programs that could improve hardiness of livestock.

Indigenous breeds are considered hardy and well adapted to the environment. The hardiness of the indigenous breeds is believed to have resulted from natural selection under the management practices of the native breeders/herders and from the adverse feed conditions. Indigenous breeds show a high level of fertility and reproduction. *In situ* management of animal genetic resources can only be successfully accomplished through breeder actions.

Drivers of Genetic Erosion

Three factors are considered as being largely responsible for the declining genetic diversity of livestock:

- Destruction of the native habitats of livestock breeds;
- The development of genetically uniform livestock breeds;
- Farmer and/or consumer preferences for certain varieties and breeds (and changes in these consumer preferences over time);
- Climate change.

Among these, commercial interests are considered as the most important pressure on livestock diversity. Important factors in determining the direction and nature of change include: growth performance (productivity),

pest and disease resistance, ease of handling, adaptation to current levels of technology, and to a relatively minor extent consumer choice.

The characteristics of the indigenous breeds (low growth rate, lower level of production) imply that the potential for altering gross income is lower than more prevalent breeds under current marketing conditions. However, adaptation to the environment and reproductive performance may alter this situation. Short-term ownership negatively affects breed conservation by creating an unstable situation for maintaining or increasing animal numbers. However, it is doubtful that any effective selection will be implemented; therefore, the population may behave as if it is a randomly mated population, with minimal loss of alleles due to selection. With the relatively small total population size and small individual flock sizes, genetic drift is an important factor affecting within-breed genetic diversity. With the small flock/herd sizes, one should expect random gene frequency changes that are cumulative over generations.

Conservation infrastructure consists of a set of actions taken by the public sector for the public good. These actions include development of cryopreserved germplasm reserves that can be used to regenerate the breed, reduce inbreeding levels, and use molecular genetic tools to evaluate genetic diversity and/or genes of interest. A sufficient quantity of semen and, potentially, embryos should be collected to regenerate the breed if necessary and to relieve potentially high levels of inbreeding.

Breeds which are Facing Wxtinction

- Most of the draft cattle breeds like Krishna Valley, Nagori, Khilar, Bargur, Amritmahal, Punganur, Ponwar, etc.
 - Many of the buffalo breeds like Bhadawari, Toda, Surti are facing threat as Murrah is being used as improver breed throughout the country due to increased demand of liquid milk.
 - Due to very little value for the wool from the Indian breeds and scarce grazing resources most of sheep breeds are losing ground. Sheep are being maintained as meat animal but has to compete with goat which are more prolific and have an advantage over sheep for value of meat in large part of country.
 - Most of the native breeds of chicken face extinction due to over emphasis on commercial chicken farming.
- The pack animal species like camel, equines, donkey, Yak etc. face threatened due to their very limited utility and changing production systems.

Conservation Methods

Conservation methods can be broadly categorized as *in situ* and *ex situ*. *In situ* conservation means that animals are kept within their production system. Important factor of *in situ* conservation cum genetic improvement is that selection should be carried out for its traditionally valued characteristics and in the environment to which it is adapted. The herds must be managed within the natural environment for that breed and need to be exposed to conditions prevalent in the field.

Ex situ conservation applies to situation where animals are kept outside their area of origin (herds kept in experimental farms, farm parks, within protected areas or in zoos) or more often, when genetic material is conserved and stored in gene banks in the form of semen, ova, embryo or DNA. Conservation through any of these methods has its own merits and demerits.

1. **Organized flocks/herds:** Maintenance of small population at a place away from the main breeding tract of the breed is the *ex situ* conservation of the live animals. This may be in the form of organized herd maintained in a research institution, bull mother farm, state owned livestock farm, zoo or breed park. This population can be used in regeneration of endangered breed and new breed development.
2. **Cryopreservation of embryos:** This is ideal for breed improvement, conservation and revival of lost breed. Its main importance is due to its diploid nature and containing all genes. However conserving embryos finds limited use, as its production and transfer require highly skilled manpower and large resources.
3. **Somatic cell banking:** Somatic cells can be used as genetic material to restore precious germplasm if the need so arises thus Fibroblast bank offers a practical approach to preserving the germplasm. However, cryobanking of somatic cells has been an underutilized tool in the national livestock conservation program. Skin fibroblast cells are selected for long-term preservation due to their easy accessibility, non-invasiveness, and any limitations on the sex or age of the animal.

4. **Epididymal sperms banking:** Epididymal spermatozoa particularly caudal spermatozoa are mature and have full competence to undergo normal fertilization and cause fetal development. *In vitro* fertilization (IVF) experiments have revealed that epididymal semen possesses binding sites for important zona pellucida proteins. Collection of cauda epididymal semen from slaughtered animals would be a rapid and cheap alternative of sperms conservation as it would obviate the requirement of time consuming and extensive training of males for semen donation.
5. **Cryopreservation of embryonic stem cell lines:** This can be excellent biological material for producing live animals and producing genetically modified animals. This also finds usage in gene and cell therapies, and for producing vital therapeutic proteins.
6. **Cryopreservation of spermatogonial stem cell lines:** Transplantation of spermatogonial stem cells (SSC) from a donor testis into the seminiferous tubules of a recipient testis results in donor-derived spermatogenesis. SSCs transplantation has been demonstrated in goats, dog, cow, pig, baboon and bovine spermatogonial stem cells shown to be capable of colonizing recipient mouse seminiferous tubules. An *in vitro* system that supports the proliferation and maintenance of SSC could be used to preserve and expand SSC numbers as well as aid in genetic modification.
7. **Storage of DNA:** Cryogenic storage of DNA has several advantages over the live germplasm as it is very easy to obtain, store, transport at low cost with no chance of disease transfer. DNA may find use in gene conservation through their introgression by transgenesis or knock out technology, and can help in recreation of lost breeds by cross checking of different populations or genetic material used.
8. **Frozen Semen:** This is an ideal and most common for genetic resource utilization activities, providing sample half of the genetic material of preserved

breeds in a form that permits convenient introgression into recipient population. Availability of established semen freezing technology and presence of semen freezing infrastructure across the country makes it method of choice for conserving indigenous livestock biodiversity. ICAR- National Bureau of Animal Genetic Resources (NBAGR) is playing a pivotal role in *ex situ* conservation through semen, somatic cell and DNA cryostorage of indigenous livestock for posterity by establishing a National Gene Bank at Karnal.

Future Prospective and Action Points

Genetic diversity defines not only animal breeds' production and functional traits, but also the ability to adapt to different environments, including food and water availability, climate, pests and diseases.

Diverse animal genetic resources are a key to economic development. Many local livestock breeds continue to represent the lifeline of rural populations. The diversity of these resources makes possible human livelihoods in some of the most inhospitable areas where crop production cannot be exclusively depended upon. While they may not be able to compete with "improved breeds" in milk and meat yields, they fulfill a much wider range of functions and provide a larger range of products. Being able to thrive even with low fodder inputs, their maintenance is ecologically more sustainable, especially in marginal environments. Requiring lower levels of health care and management, they commonly entail a lower workload in comparison with exotic breeds. As is becoming increasingly clear, they often have scope for specialty products and can be essential to preserve habitats and cultures.

Given the above conditions, there are two areas in which to base conservation efforts. These consist of developing a conservation infrastructure (a public service) and breeder actions (a private-sector activity).

Nongovernmental organizations have to play a key role in the conservation of indigenous breeds, and their engagement is likely to continue by assisting breeders with technology transfer.

Valorization of Indigenous Livestock and Poultry – An Approach towards their Conservation

Rekha Sharma*, Reena Arora* and Sonika Ahlawat

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

Indigenous livestock species have co-existed with humans as important contributors to food, income, socio-economic and cultural status for centuries. The livestock scenario in present times has changed from a resource-driven activity based on local conditions and environments to one driven by demand. Consequently, specialized traits of low-producing indigenous livestock breeds are ignored in emerging, high-input-based farming systems. This has led to a progressive replacement of traditional multipurpose breeds with high-yielding ones and more profit-oriented farming. Since indigenous livestock cannot compete in production with industrial livestock systems; it is reasonable to focus on their unique qualitative aspects as a means of conservation and source of livelihood. Fortunately, many local breeds and species have a large but often unrecognized potential to produce items that customers appreciate and demand. Many local breeds are bestowed with unique qualities like colored wool, disease resistance, patterned hides, super-fine fiber, especially palatable meat, or milk with therapeutic or health benefits. Local breeds can produce unique products that can generate significant levels of demand and can help rescue a threatened breed from further decline or extinction.

Introduction

Rearing indigenous livestock breeds in today's economy is usually less lucrative than keeping high-yielding commercial breeds. Characterization of indigenous germplasm, including their product quality traits, needs immediate attention as it could strengthen the position of local breeds in the current consumer scenario. The value of local breeds as sources of culinary delicacies is well established in parts of the developed world. In Europe, many special kinds of cheese and meats are associated with particular breeds. Products of local breeds are often processed in traditional ways. The need to conserve milk or meat without refrigeration has led to the development of unique sausages and cheese. In Germany, meat from breeds such as the Heidschnucke sheep and Schwäbisch-Hallische pig fetches a premium price in gourmet restaurants. In North America, too, there are efforts to market the meat and wool from heritage breeds. In these countries, the marketing of cheeses, sausages, wool, and other specialty products has contributed to the conservation of indigenous breeds, enhanced regional identities, and stimulated rural economies. In developing countries, however, examples of this approach are rare.

It is imperative to identify and document traditional livestock, their products, and their processing methods. The genetic valuation of these breeds is also warranted

so that biomarkers related to specific traits can be used for breed improvement and viable conservation. Further, analysis of the special properties of the products in terms of sensory qualities, nutritional value, and medicinal effects need to be evaluated. Such an approach will result in:

- Inventory of existing traditional products and processing methods from the selected breeds
- This can further lead to better understanding of the technological requirements for producing products tailored to urban consumer preferences
- Awareness among communities, policy makers and private enterprise regarding economic opportunities inherent in local breeds
- Insights into the potential and promise as well as criteria for a special label/brand for products

The idea behind creating niche markets is to raise the value of local products and support sustainable, regional, small-scale production systems. Realizing the opportune time to break new ground, the science and technology ministry (DST) of India has initiated funding research projects that seek to pinpoint bioactive ingredients in dung, milk, and urine of “pure indigenous cows” for use in medicinal, nutritional, agricultural and household products. The program is titled “Scientific utilization

*Authors for Correspondence: Email-Rekha.Sharma1@icar.gov.in; Reena.Arora@icar.gov.in

through research augmentation-Prime products from indigenous cows (Sutra-Pic). Recent scientific efforts in underpinning the quality attributes of the Indian livestock that can be exploited for augmenting their value and promoting sustainable utilization are compiled below:

Potential Dairy and Meat Producing Indian Livestock and Poultry

Superior Milk of Indigenous Cow

Consumer perception of Indian or desi cow milk is associated with the assumption that it differs from the milk of taurine cattle and their crossbreds. Numerous studies from different parts of the world have compared the milk composition of their indigenous and universally used breeds including their crossbreds. There is a dearth of knowledge in this regard from Asia including India. Milk of indigenous cattle maintained only on grazing had a more favorable nutrient profile than the cows maintained under an intensive system of management (Sharma *et al.*, 2018; 2018a). The saturated fatty acids (SFA) proportion was lower (61.40%) mainly due to the medium-chain FA group (15.1%). The milk was enriched in total n-3 fatty acids and total unsaturated fatty acids (UFA, 38.6%) including MUFA (31.4%) and PUFA (7.2%). The n-6/n-3 ratio (2.7) and Atherogenic index (33.9) were significantly lower in grazing milk. Concentrations of minerals (Zn, Fe, P, Cu) and all the vitamins except vitamin B5 were significantly higher ($P < 0.05$) than in the cattle under intensive management. Vitamin A, C, E, and β -carotene were more than double. Being antioxidants, these are nutritionally desirable to reduce oxidative stress. Moreover, the yellow tint of cheese and butter caused by carotenoids of milk fat is an attractive attribute for consumers. It is considered to be an indicator that the product has originated from cows thriving on pasture diets. Higher milk productivity was never the reason why Indian zebu cows (*Bos indicus*) were revered in India as it is the quality of milk that has been the talk of folklore. It is possible, that almost total pasture feeding in ancient times was responsible for the proclaimed health-promoting qualities of the milk. Organic milk production with particular emphasis on grazing is gaining momentum throughout the world which suggests a promising future for indigenous cows maintained on grazing.

Bhadawari Buffalo – Milk with the Highest fat Content

Bhadawari breed is famous for the high-fat content in

their milk. Bhadawari buffaloes are found in the ravines of Yamuna, Chambal, and Utangan rivers spread over Uttar Pradesh and Madhya Pradesh. Though the total lactation yield is lower, fat content in the milk has been recorded as high as 13%. The average milk yield of the breed is 1294 Kg per lactation with an average fat % of 7.88 (Yield ranging between 540-1400 Kg per lactation and fat % from 6 to 12.8).

Chilika Buffalo – Curd with a Longer Shelf Life

Chilika buffalo are unique as they are found only around Chilika Lake, in the Khordha, Puri, and Ganjam districts of Odisha, and feed on seaweed in neck-deep, brackish waters of Chilika Lake. Perhaps because of the high salt content in the animal's diet, the milk is not only very tasty but can also be stored without refrigeration for days. Both the milk and other products such as curd made from milk are attracting attention. It is widely observed and reported that Chilika curd has a longer shelf life and has been scientifically established. A total of 64 microbial isolates were isolated from curd and milk samples collected from Chilika (Nanda *et al.*, 2013). Surprisingly, 8 Lactobacillus isolates out of the above were found to show anti-fungal effects against the test organism *Candida parapsilosis* (NCDC 279), which can be correlated with the preservation of curd from fungal spoilage and enhancing its shelf life. Such isolates can be used as a starter for making curd with better shelf life. Products with extended shelf life can be beneficial for a country like India where major population reside in villages that can't afford refrigeration facilities and there is no need for a cold chain during transportation.

Buffalo Milk as a Source of Bio-defensive Milk Peptides

Buffaloes are considered more resistant to diseases than cattle (Pal and Chakravarty, 2020), although, little scientific evidence supports this assumption. However, a study was conducted to compare the transcriptomic profiles of milk somatic cells of Sahiwal cattle and Murrah buffaloes (Ahlawat *et al.*, 2021). A noticeable enrichment of innate and adaptive immune response genes and various host defense peptides such as lysozyme, defensin β and granzymes was reported in buffaloes. The expression of LYZ1 (lysozyme C) was observed to be seven times higher in buffalo milk in comparison to cattle. Lysozyme is a bacteriolytic enzyme found in body fluids such as tears, saliva, and milk and therefore forms an important component of mucosal immunity in

mammals (Masschalck *et al.*, 2001). β defensin is known to function as a natural antibiotic (Meade and O'Farrelly, 2019) and granzymes are released by CD8 T cells and natural killer (NK) cells that trigger programmed cell death of infected or abnormal target cells. Based on these observations, buffalo milk can be exploited in the future for various bio-defensive milk peptides and hence, offers novel paradigms for the development of effective and novel therapies.

Goat Milk – Therapeutic Potential

The vitamin and mineral contents of goat and cow milk are fairly similar, though goat's milk contains more calcium, vitamin B6, vitamin A, potassium, niacin, copper, and the antioxidant selenium. Goat milk contains 25% more vitamin B6, 47% more vitamin A and 13% more calcium than cow's milk. Goat milk is an excellent source of calcium, phosphorus, and potassium. It is also a good source of magnesium, sodium, and iron. Goat milk offers a wide variety of health benefits such as better digestibility, more alkalinity, less α 1 casein than cow's milk and is, therefore, less allergenic. It is also useful in the treatment of ulcers due to its more effective acid buffering capacity. Unlike cow milk, which is slightly acidic, goat milk is alkaline, which is very useful for people with acidity problems. It is more digestible because of its small sized globules, uniform protein, fat distribution, and less lactose.

Kadaknath Chicken Black Meat – Better Functional Attributes

The autochthonous Kadaknath is the only black meat chicken breed among the 19 chicken breeds registered in India (<https://nbagr.icar.gov.in/en/home/>). It is the only indigenous animal genetic resource in India to get the Geographical Indication (GI) tag for the protein-rich and black-colored meat in 2018 (https://ipindia.gov.in/writereaddata/Portal/IPOJournal/1_2598_1/Journal_104.pdf). The tribal community of Bhil and Bhilala are the primary custodians of this unique backyard poultry. The peculiarity of this breed is that the entire bird and its internal organs are black due to the deposition of melanin pigment, a genetic condition called “Fibromelanosis” and is supposed to have aphrodisiac and medicinal properties. But, there is a paucity of literature that can endorse the claims of nutritional and medicinal properties of Kadaknath meat. A recent study reported that the Kadaknath meat is an enriched source of functional biomolecules (carnosine, anserine, creatine). Its breast

meat carnosine content was more than double of the Cobb broiler (Sharma *et al.*, 2022). The genetic background may be a key determinant as a significant abundance of CARNS1 and SLC36A1 expression was identified in the Kadaknath breast. The superior functional property of Kadaknath meat was established by the antioxidant capacity (Sehrawat *et al.*, 2021) and a stronger ability to inhibit the formation of advanced glycation end products (AGEs). The identification of fairly unknown nutritional and functional advantages of Kadaknath meat could potentially change the paradigm with its meat consumption. It will help in developing a brand name for Kadaknath products that will propel an increase in its market share and ultimately conservation of this unique but endangered poultry germplasm.

Barbari Goat Meat – A Blend of Taste and Nutrition

Meat from livestock is an ideal source of protein and nutrients such as iodine, vitamin B12, zinc, and iron. Goat meat has less fat and cholesterol as well as lower saturated fatty acids content than chicken, lamb, beef, and pork (Ivanovic *et al.*, 2016), which will provide an edge in marketing as a health food for cardiovascular diseases. Barbari is a dual-purpose breed used for both meat and milk and is considered the best meat breed (Paramasivam *et al.*, 2002; Mandal *et al.*, 2016). Among Indian goat breeds, lower cholesterol levels have been reported in Barbari muscles (Das and Rajkumar, 2010). Therefore goat meat or chevon from such breeds as Barbari presents a healthier option as compared to chevon from other breeds. Chevon from Barbari goat is considered of good quality and this breed is preferred for commercial goat farming (Das and Rajkumar, 2010; Umaraw *et al.*, 2017). Functional genomics analysis of Barbari muscles has also identified significant genes and pathways associated with triacylglycerol biosynthesis and lipid metabolism (Kumar *et al.*, 2021), which lends support to the better organoleptic properties of its meat.

Bandur Sheep Mutton – A Prized Delicacy

Mutton from the Bandur sheep breed of Karnataka is favored by consumers for its unique flavor and fetches a higher price than other local breeds. Research revealed a higher back fat thickness in Bandur animals as compared to local sheep. Tenderness of different muscles (longissimus dorsi, brachicephalicus, bicep, semimembranosus, psoas major, semitendinosus, and tricep) was also greater in Bandur sheep. The amino acid and fatty acid analysis revealed a significantly higher

level of oleic acid and histidine in Bandur (Kumar *et al.*, 2018; Arora *et al.*, 2019). Higher content of oleic acid has been positively correlated with overall palatability while histidine is an essential amino acid required in the diet. The discovery of higher fat content, tenderness of muscles, oleic acid and histidine will add value to the mutton from Bandur sheep. Genes associated with muscle tenderness viz., HSPB1, DNAJB5, HSPA6, were also overexpressed in Bandur sheep. The highly connected genes identified by transcriptomics (CNOT2, CNOT6, HSPB1, HSPA6, MAP3K14, and PPARD) and miRNA form potential biomarkers for unique muscle traits of Bandur sheep (Kaur *et al.*, 2020; 2020a; Arora *et al.*, 2021). The phenotypic and genetic evaluation of mutton quality characteristics of Bandur sheep has presented the scientific basis of superior quality/taste of Bandur sheep mutton that will augment sustainable conservation of this breed and its recognition as a Geographical Indicator.

Potential Value of Wool and Fibre from Indian Breeds

Changthangi Goat – The Source of Soft Gold (Pashmina Fibre)

Pashmina is well known for its fineness, warmth and softness. It is luxurious, softer and warmer than superfine merino wool. The word pashmina originated from the word 'pashm' which means 'soft gold' in the local language. The fiber is mostly used for preparing shawls with designs by the artisans of Jammu and Kashmir and Himachal. In India, two pashmina-producing goat breeds are found i.e. Changthangi and Chegu. Changthangi breed is domesticated in the Ladakh region of Jammu and Kashmir and Chegu in Lahul and Spiti and Kinnaur region of Himachal Pradesh. The physic-mechanical assessment of pashmina fibre has revealed optimal values of resiliency and compression for fine wool, thereby affirming the superiority of Pashmina in terms of softness and fullness, over other wool fibres (Bumla *et al.*, 2012). Recent studies based on skin transcriptome have delineated the genes and pathways relevant to Pashmina production (Ahlawat *et al.*, 2020; Bhat *et al.*, 2021). Comparison of the skin transcriptome of Changthangi with meat-type goat identified a higher expression of genes associated with fine fibre in Changthangi goat. Among all shades, the white Pashmina fetches the highest price. The evaluation of differentially expressed genes in black, white, and brown skin samples of Changthangi goats revealed enrichment of ASIP gene in white skin

in comparison to brown and black skin samples (Bhat *et al.*, 2019). There have been meager systemic/scientific approaches for qualitative and quantitative improvement in Pashmina production and productivity. The pashmina production system is mostly open range type and depends on pastures.

Magra Sheep – Lustrous Wool

Magra sheep, formerly known as the Bikaneri breed is an important breed of Rajasthan. Magra sheep have been identified to be suitable for the desert tracts as the breed can sustain if watered only twice a week without any adverse effects on body weight. This unique breed is the only lustrous carpet-wool-producing breed in India (Dass *et al.*, 2003). This breed can be used to improve other wool-type Indian sheep (Mehta *et al.*, 2004)

Prospective Value of Non-conventional Livestock Species and their Products

Equid Milk

Milk from nontraditional animal species (donkey, camel) is recently gaining momentum mainly because they are considered suitable to supplement the needs of special population groups (i.e., infants, elderly). Research on their milk has dramatically increased over the past few years to decipher their unique functional properties, namely, antimicrobial activity, immunomodulation, and hypoallergenicity, especially highlighting their effect on human health and the potential utilization of medicinal, nutritious and cosmetic properties.

Donkey Products

The donkey (*Equus asinus*) is one of the most valuable domestic animals for the economy in many developing countries. Donkey meat is a healthy food with high protein but low fat and cholesterol contents while donkey milk composition is similar to human milk and more suitable for infants compared with those of other mammals. It is drunk fresh or in powdered form. It is considered a good alternative for consumers with dairy allergies such as cow's milk protein allergy. The low allergenicity is mainly attributed to the low casein content, which is very close to that in the human milk. In particular α s1- and β -caseins in different phosphorylated forms are present. The sale of donkey's milk is practiced now as villagers of Karnataka and Tamil Nadu sell donkey milk at a high price in Bengaluru. India has lost more than half of its donkeys with a massive reduction of >60% in its population since 2012. ICAR-National Research Centre

on Equines (NRCE) has been entrusted with the job to explore possibilities of promoting donkey milk in the country that can lead to their conservation.

Camel Milk

The dromedary camel is an important component of the dry land and desert ecosystem. It is not only an important means of transport but also serves as a source of milk for camel-rearing societies. Camel milk is considered white gold and a powerhouse of good health for its rich nutrients and immunity-building properties. It is reputed to be anti-infectious, anti-cancerous, and anti-diabetic. Covid-19 has put greater focus on immunity boosters and increased interest in camel milk products due to its high vitamin C, many minerals, and immunoglobulins. It may act as the main criterion for the sustenance of indigenous camels in the present era of diminishing draught utility. Camel milk is also used in Kazakhstan as an adjunct to chemotherapy for some forms of cancers. It is claimed to have a remedial effect for at least 13 different kinds of diseases and also to be an aphrodisiac by the Somali people. The current scientific evidence for the therapeutic actions of camel milk continues to unfold, and efforts are underway to precisely identify the therapeutic constituents. The establishment of such components in camel milk may act as the main criterion for the sustenance of indigenous camel breeds in the present era of its diminishing draught utility. Camel milk has been reported to be used for treatment of various diseases including tuberculosis. Camel milk production has become a booming trade in Middle Eastern countries. As camel milk has gained popularity globally, India is now warming up to the idea of camel milk production and consumption. Camel milk from Kutch region of Gujarat is already available in the retail shelves of major metros and cities across the country. Camel herders are more willing to keep an increasing number of animals as they benefit from the extra income gained from selling camel milk. This can help revive the continuously declining numbers of indigenous camels and the livelihoods of their herders. The claimed therapeutic actions of milk have recently been the subject of numerous studies.

Sheep Milk

Sheep milk is considered an excellent source of nourishment for humans. The amount of fat, calorie, essential vitamins and minerals are nearly twice that found in cow milk. A sufficient quantity of calcium and

magnesium make it a perfect food for aged people. It also contains the highest amount of casein and whey protein when compared to milk from other ruminants. Favorable fatty acids are abundant like conjugated linoleic acid (CLA), and unsaturated fatty acid in sheep milk. As a result, sheep milk finds use in cosmetic skincare products like creams, soaps, and lotions. Indian sheep are low producers and sheep milk has never held prominence in the Indian dairy industry, while sheep breeds from Europe and the Mediterranean region are major dairy breeds. Some of the prominent dairy sheep breeds are Eastern Friesian (Germany), Lacaune (France), and Awassi (Middle East). The famous classical cheeses made from sheep milk are Roquefort, Feta, Manchego, and Pecorino. The Patanwadi sheep holds potential as a dairy breed as it is considered a higher milk yielder among Indian sheep, with an average daily milk yield of 800g and a lactation period of 90 days (Mohapatra *et al.*, 2020).

Yak Churpi Cheese

Churpi is a traditional cottage cheese made from yak milk, found in different hilly regions of India including Sikkim, Darjeeling, and Ladakh. Yeast, mold, lactic acid bacteria, and Bifidobacterium sp. are the major participating microbes in Ladakhi churpi. Microbial interaction during the fermentation of milk makes it more nutritious. It is enriched in riboflavin, thiamine, and vitamin C. It also contains a very good amount of protein (60–63%) and carbohydrates (23–24%); and a low amount of fat (7–8%) (Panda *et al.*, 2016). Commercialization based on exploiting the unique properties of Churpi can add commercial value to the declining indigenous yak and cattle (Siri) populations of the Himalayan region.

Future Prospective and Action Points

1. Spinning a value chain from native livestock produce is gaining momentum across the globe. The idea behind creating niche markets is to raise the value of local products and support sustainable, regional, small-scale production systems. India is bestowed with large livestock biodiversity that offers a great opportunity to produce a variety of convenient value-added livestock products as well to improve their income from the livestock sector.
2. Value can be added to products from traditional farming systems through the identification of quality

attributes. Increasing consumer demands for superior quality food and regional products point toward potential markets for such products.

3. Efforts need to be directed towards value addition to livestock by processed products. Goats, sheep, and equine milk can be processed into yogurt, cheese, skincare, or utility products. Their hair and wool can be utilized in making traditional clothes and carpets. Dung can be a source of income in the present time of upcoming organic farming. Demand for processed cow urine and ghee is also anticipated for use in traditional medicine.
4. A great scope exists in India for the processing of milk/meat to value-added products such as ready-to-eat/ready-to-cook products for the growing population with fast urbanization and change in lifestyle. Livestock farming can become more remunerative by changing the place, time, and form of its products as per the market requirement with an innovative mindset after a watchful study of consumer preferences. Farmers need to grab these prospects of enhancing their income from the identical amount of livestock that they are having.
5. State government departments can guide the farmers for skill up-gradation training conducted by them from time to time for preparation of value-added processed livestock products, in labeling and packaging of the product, and in extending market support as well as an incentive to communities for the sale of value-added material.
6. By concerted efforts, it may be possible to find a match between the qualities of the local breeds, the features of a particular product, and the demands of a specific market. Making this match will help conserve the breed as well as provide a livelihood for people involved in the value chain. This conservation strategy could establish a new place for indigenous breeds in modern agriculture.

References

- Ahlawat S, R Arora, R Sharma, U Sharma, M Kaur, A Kumar, KV Singh, MK Singh and RK Vijn (2020) Skin transcriptome profiling of Changthangi goats highlights the relevance of genes involved in Pashmina production. *Sci. Rep.* **10**(1): 1-10. doi:10.1038/s41598-02063023-6. PMID: 32269277; PMCID: PMC7142143.
- Ahlawat S, R Arora, U Sharma, A Sharma, Y Girdhar, R Sharma, A Kumar and RK Vijn (2021) Comparative gene expression profiling of milk somatic cells of Sahiwal cattle and Murrah buffaloes. *Gene.* **764**: 145101. doi:10.1016/j.gene.2020.145101. Epub 2020 Aug 30. PMID: 32877747.
- Arora R, NK Siddaraju, SS Manjunatha, S Sudarshan, NF Mohamed, A Kumar, P Chhabra, M Kaur, RM Sreesujatha, S Ahlawat and RK Vijn (2021) Muscle transcriptome provides the first insight into the dynamics of gene expression with progression of age in sheep. *Sci. Rep.* **11**(1): 1-11. https://doi.org/10.1038/s41598-021-01848-5.
- Arora R, NK Siddaraju, S Sudarshan, MN Fairoze, M Kaur, A Sharma, Y Girdhar, RM Sreesujatha, SK Devatkal, S Ahlawat, RK Vijn and SS Manjunatha (2019) Transcriptome profiling of longissimus thoracis muscles identifies highly connected differentially expressed genes in meat type sheep of India. *PLoS One.* **14**(6): e0217461. doi:10.1371/journal.pone.0217461.
- Bhat B, A Singh, Z Iqbal, JK Kaushik, AR Rao, SM Ahmad, H Bhat, A Ayaz, FD Sheikh, S Kalra, S Shanaz, MS Mir, PK Agarwal, T Mohapatra and NA Ganai (2019) Comparative transcriptome analysis reveals the genetic basis of coat color variation in Pashmina goat. *Sci. Rep.* **9**(1): 1-9. doi:10.1038/s41598-019-42676-y. PMID: 31015528; PMCID: PMC6478727.
- Bhat B, M Yaseen, A Singh, SM Ahmad and NA Ganai (2021) Identification of potential key genes and pathways associated with the Pashmina fiber initiation using RNA-Seq and integrated bioinformatics analysis. *Sci. Rep.* **11**(1): 1-9. doi:10.1038/s41598-021-81471-6. PMID: 33469142; PMCID: PMC7815713.
- Bumla NA, A Maria, JS Sasanand AM Khateeb (2012) Quality of Indian pashmina fibre in terms of its physico-mechanical properties. *Wayamba J. Anim. Sci.* 459–462. http://www.wayambajournal.com/documents/1348636207.pdf.
- Das AK and V Rajkumar (2010) Comparative study on carcass characteristics and meat quality of three Indian goat breeds. *Indian J. Anim. Sci.* **80**: 1014–1018.
- Dass G, VK Singh, SK Chopra and M Ayub (2003) Wool production and quality of Magra sheep under hot arid zone of Rajasthan. *The Indian J. Small Ruminants.* **9**(1): 10-12.
- Ivanovic S, I Pavlovic and B Pisinov (2016) The Quality of Goat Meat and It's Impact on Human Health. *Biotech. Anim. Husb.* **32**(2): 111–122. doi:10.2298/bah1602111i.
- Kaur M, A Kumar, NK Siddaraju, MN Fairoze, S Ahlawat, RK Vijn, A Yadav and Arora R (2020) Exploring the skeletal muscle miRNAome of Bandur sheep using RNA sequencing. *Indian J. Anim. Sci.* **90**(8): 118–122.
- Kaur M, A Kumar, NK Siddaraju, MN Fairoze, P Chhabra, S Ahlawat, RK Vijn, AYadav and R Arora (2020a) Differential expression of miRNAs in skeletal muscles of Indian sheep with diverse carcass and muscle traits. *Sci. Rep.* **10**(1): 1-11. 16332 doi:10.1038/s41598020-73071-7.
- Kumar SN, MR Jayashankar, N Ramakrishnappa, W Ruban and RM Sreesujatha (2018) Carcass and meat quality characteristics of Bandur ram lambs. *Indian J. Anim. Res.* **52**(5): 774779. DOI:10.18805/ijar.B-3261.
- Kumar A, M Kaur, S Ahlawat, U Sharma, MK Singh, KV Singh, P Chhabra, RK Vijn, A Yadav and R Arora (2021)

- Transcriptomic diversity in longissimusthoracis muscles of Barbari and Changthangi goat breeds of India. *Genomics* **113**(4): 1639-1646. doi:10.1016/j.ygeno.2021.04.019.
- Mandal A, R Behera, S Rai, M Karunakaran and TK Dutta (2016) Performance evaluation of Barbari goats in semi-arid region of India: A review. *Research and Reviews: J. Dairy Sci. Tech.* **5**: 25–29.
- Masschalck B, R Van Houdt, EG Van Haver and CW Michiels (2001) Inactivation of gramnegative bacteria by lysozyme, denatured lysozyme, and lysozyme-derived peptides under high hydrostatic pressure. *Appl. Environ. Microbiol.* **67**(1): 339–344.
- Meade KG and C O’Farrelly (2019) β -Defensins: Farming the microbiome for homeostasis and health. *Front. Immunol.* **9**: 3072. <https://doi.org/10.3389/fimmu.2018.03072>.
- Mehta SC, SK Chopra, VK Singh, M Ayub and V Mahotra (2004) Production and quality of wool in Magra breed of sheep. *Indian J. Animal Sci.* **74**(7): 792-794.
- Mohapatra A, K De, V Prakash, D Kumar and SMK Naqvi (2020) Effect of different milking methods on milk yield and growth of lambs. *The Indian J. of Small Ruminants.* **26**(1): 62-66. doi:10.5958/0973-9718.2020.00005.7.
- Nanda DK, R Singh, SK Tomar, SK Dash, S Jayakumar, DK Arora, R Chaudhary and D Kumar (2013) Indian Chilika curd – A potential dairy product for Geographical Indication registration. *Indian J. Tradit. Knowl.* **12**(4): 707-713.
- Pal A and AK Chakravarty (2020) Disease resistance for different livestock species. *Genetics and Breeding for Disease Resistance of Livestock.* 271-296. <https://doi.org/10.1016/B978-0-12-816406-8.00019-X>.
- Panda A, K Ghosh, M Ray, SK Nandi, S Parua, D Bera, SN Singh, SK Dwivedi and KC Mondal (2016) Ethnic preparation and quality assessment of Chhurpi, a home-made cheese of Ladakh, India. *J. Ethnic Foods* **3**(4): 257-262. doi:10.1016/j.jef.2016.12.004.
- Paramasivam A, S Arunachalam, T Sivakumar and V Ramesh (2002) Growth performance and carcass traits of Barbari goats under different system of management. *FAO. Indian J. Animal Sci.* **72**(11): 1016–1018.
- Sehrawat R, R Sharma, S Ahlawat, V Sharma, MS Thakur, M Kaur and MS Tantia (2021) First report on better functional property of black chicken meat from India. *Indian J. Anim. Res.* **55**(6): 723-733.
- Sharma R, S Ahlawat, RAK Aggarwal, A Dua, V Sharma and MS Tantia (2018) Comparative milk metabolite profiling for exploring superiority of indigenous Indian cow milk over exotic and crossbred counterparts. *J. Food Sci. Technol.* **55**(10): 4232-4243. doi:10.1007/s13197-018-3360-2.
- Sharma R, S Ahlawat, H Sharma, RAK Aggarwal, V Sharma and MS Tantia (2018a) Variable sialic acid content in milk of Indian cattle and buffalo across different stages of lactation. *J. Dairy Res.* **86**(1): 98-101.
- Sharma R, R Sehrawat, S Ahlawat, V Sharma, A Parmar, MS Thakur, AK Mishra and MS Tantia (2022) An attempt to valorize the only black meat chicken breed of India by delineating superior functional attributes of its meat. *Sci. Rep.* **12**(1): 1-12. <https://doi.org/10.1038/s41598-022-07575-9>.
- Umaraw P, AK Verma and P Kumar (2017) Barbari goats: current status. In *Sustainable Goat Production in Adverse Environments, Volume II*, pp 29–40. https://doi.org/10.1007/9783-319-71294-9_3.

Uniqueness of Animal Genetic Resources Adapted to High Altitude Environment of Leh-Ladakh

Manishi Mukesh^{1*}, Saket Niranjana¹, Monika Sodhi¹, RS Kataria¹, Rahul Behl¹, Mohammad Iqbal², Vijay K Bharti³, Tsewang Dorjay³ and BP Mishra¹

¹ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

²Animal Husbandry Department, Leh-194101, India

³DRDO-Defense Institute of High-Altitude Research, Leh-194101, India

In difficult terrain of Ladakh region, where land resources are meager, animal wealth plays an important role in the life of the local people. Common native livestock genetic resources in Ladakh include yak, cattle, dzomo, dzo, goat, sheep, donkeys, horses, double hump camel, *etc.* For adaptability at cold as well as hypobaric hypoxic condition of Leh-Ladakh, the native animal genetic resources have developed certain physical and physiological adaptations. The physical changes include small statured body, large hair fur, and thick skin while physiological changes include unique circulatory adaptability with enlarged lungs, heart and larger concentration of red blood cells to accommodate the hypobaric-hypoxic environment. These unique traits make these livestock thrive and perform at under low oxygen and temperature; high ultraviolet exposure, and scarce nutrition in lean period.

Introduction

Ladakh, land of many high passes, (La means passes and Dhak means many) is one of the world's highest inhabited regions surrounded by snow-capped Himalayan, Zaskar and Karakoram ranges. This is a newly formed union territory in the northern most region of India bordering China and Pakistan. It lies on the rain shadow side of the Himalayan, and combines the condition of both arctic and desert climate and hence often called "COLD DESERT". This cold-arid desert at over 3000-meter mean sea level has very harsh climate characterized by extreme temperature (-40°C in winter and 35°C in summer); low humidity (25-40%), precipitation (80-300 mm) and oxygen level (nearly 60-70% of the oxygen concentration at sea level); high UV radiations and wind erosion. As per 20th livestock census and departmental census of UT Ladakh (2021), Ladakh has 0.842 lakh cattle, 0.203 lakh yak, 2.44 lakh sheep, 3.33 lakh goat, 0.038 lakh horse, 0.066 lakh donkey and 0.029 double hump camel (Fig. 1). Ladakh is one of the most sparsely populated regions and has huge barren lands without vegetation and assured water supply. The soil of the area is sandy loam and slit clay. The cropping season is from April to October with possibility of only single crop annually. The major field crops in the area are

barley, wheat, pulses, oil seed, peas and millets while horticulture crops are apricot, apple, walnut, pear and peach. The irrigation is mainly through channels from the glacier.

In past decade, ICAR-NBAGR has accomplished comprehensive characterization of several livestock species of Ladakh such as Ladakhi cattle, Ladakhi yak, Ladakhi donkey and Zaskar ponies in their breeding tract. The information gathered during several years of research has not only helped to understand the uniqueness of these well adapted livestock species but have also helped the policy makers, stake holders to realize the potential of these livestock breeds. In this article, summarized information collected during years of field survey and research by our team along with available information on livestock species of Ladakh is presented.

Ladakhi Cattle: Native Cattle of Leh-Ladakh

The total cattle population in Leh & Ladakh region is around 84,201 out of which, 45,432 are indigenous and 38,778 are cross-bred cattle. The preference for native cattle over crossbred cattle is more in Leh than in Kargil district. The local cattle from Leh and Ladakh region, is a unique germplasm having excellent adaptation potential

*Author for Correspondence: Email-Manishi.Mukesh@icar.gov.in

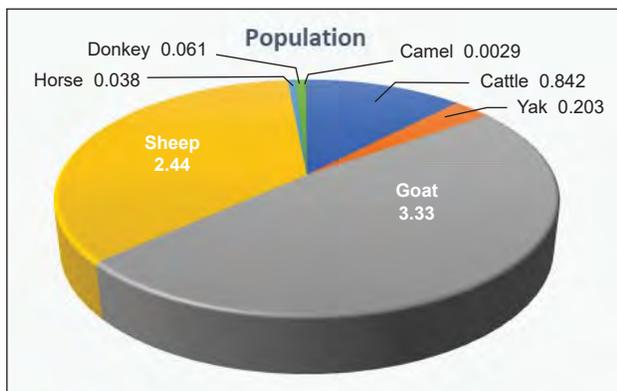


Fig. 1. Population size of different livestock species of Leh-Ladakh region

to high altitude hypobaric stress. Most of the households in villages of Ladakh, irrespective of land holdings, maintain the Ladakhi cows as an alternate source of income or livelihood. The typical native Ladakhi cow and bull are depicted in Fig. 2.

In spite of harsh conditions and meagre feed resources, these cows provide 2.5-4.5 kg of milk and are good source of milk, butter and churpi (local home-made product from curdled milk with long shelf life). They serve as an important animal protein source for local people, particularly during lean winter period. The milk of Ladakhi cow is consumed as such or converted into products like ghee, butter or churpi for personal as well as commercial use. The Ladakhi cattle is a short stature animal with body length of 88.48 ± 0.56 , and height at wither of 91.27 ± 0.41 . The morphometric data along with microsatellite and SNP marker data showed genetic distinctness of native cattle from other native

cattle breeds of India. Considering its uniqueness, the native cattle populations of Ladakh was recognized in 2018 as 42 cattle breed of India, and named as “Ladakhi cattle”.

Ladakhi Yak – A Multi-utility Species of Ladakh

The yak (*Bos grunniens*), a large ruminant of the bovidae family is genetically adapted through natural selection over millennia and presents the best example of high-altitude adaptation. Yak defends cold by conserving rather than generating the heat, through its compact body structure, thick outer long hairs and a fine down undercoat in winter. In addition, their physiological and anatomical traits such as large lungs and a large heart with thin-walled pulmonary arteries lack of pulmonary vasoconstriction and high energy metabolism enables them to survive easily at high altitude hypoxic condition. The Ladakh has sizeable population of yak and is mostly reared by nomads for multi-faceted utility. These yaks are reared in alpine region over 4000 m mean sea levels. It provides milk, meat, fibre, manure, draft power, and transportation in highly challenging areas of high altitude, where other kind of species might not survive. The typical Ladakhi yak male and female are shown in Fig. 3.

The churpi and butter made up of yak milk is quite popular amongst nomads and local Ladakhi people. Apart from source of quality milk, yak is also reared for hair fiber. Additionally, its utility as a pack animal for snow-bound areas has also been widely recognized. Apart from Ladakh, the yak population in India is also present in Sikkim, Arunachal Pradesh and Himachal Pradesh. In addition to pure yak, yak-cattle hybrids



Fig. 2. Typical Ladhaki Cow and Typical Ladhaki Bull



Fig. 3. Typical Ladakhi yak male and Typical Ladakhi yak female

are generally reared at mid region of Ladakh (3500 to 4000 m msl). Most of the hybrids are produced as yak as male and native cow (Ladakhi cattle) as a female parent. Cross hybridization of yak and cattle is an age-old practice being used by yak herders. They are preferred due to higher milk production, their better adaptability to warm climate of lower region, without any need to shift to higher altitude during the summer.

Zanskari Ponies—Native Horse of Zanskar Region

Ladakh is also endowed with local equine population called Zanskari ponies due to their major distribution in the Zanskar valley of Ladakh. The body height ranges between 132–147 cm. These ponies look like a small horse with stocky body, exhibit thicker manes, shorter legs, wider barrels heavier hair coat, long tail and heavier bones. The predominant colors are grey, brown and bay. The body confirmation of ponies is most suitable for balancing while riding and load carrying. These ponies have proved their stamina as hardy pack animals with minimum maintenance, during operation Vijay and are called as ‘trucks of the mountains. Hence, due to their suitability as pack animals, these ponies are valuable for deployment as all-terrain vehicles in Ladakh-sector and other high mountainous areas. A typical animal of Zanskari pony is shown in Fig. 4.



Fig. 4. Typical Zanskari pony

Ladakhi Donkey—A Neglected Species of Ladakh

The Ladakhi donkey is reared as a pack animal for transportation of various materials by Buddhist

communities—Tsering, Tashi, Sonam, Namgyal, Tundup, Dorje. These animals are also being for food-logistics supply by Indian Army manure and to some extent for ploughing of fields during cultivation season. Typical male and female animal of Ladakhi donkey is shown in Fig. 5. The coat colour of Ladakhi donkeys varies from light brown to dark brown/black, though majority of animals are dark brown or black. Many animals have white markings around muzzle and eyes and few animals have white leg markings and zebra markings. The mean height at withers is 94.27 ± 3.75 and 93.85 ± 3.8 cm in male and female animals, respectively. The body length in the male and female animals is 95.53 ± 5.66 and 97.46 ± 7.37 cm, respectively. These donkeys are able to carry upto 50 kg of load in rugged terrain and can travel about 20 Km in 8-10 hours.



Fig. 5. Typical Ladakhi donkey-male



Typical Ladakhi donkey-female

Changthangi Goat – World Famous Pashmina Goat

The Changthangi goat is another important livestock species that is native to high altitude of Leh-Ladakh. This is also being referred as Changpa goat as these are reared by Changpa nomads of Changthang region of Ladakh. These goats survive by grazing the local grasses in vast natural grassland of Changthang area. The Changthangi goat is a medium sized goat with 50 cm height at the withers, and a body weight of about 29 kg. Due to extreme cold environmental conditions prevalent in its ecological habitat, these goats possess

double hair coat; outer coat of long course hair and inner coat of shorter fine hair. The inner fine hair is used to produce world’s finest pashmina fibre and most costly animal fiber with an average fineness of 11–13 micron. The colour of the hair is generally white but other variants such as black or brown also exist in some animals. These goats play major role in sustaining the economy of local nomadic people as these provides fine quality wool to make famous pashmina shawls which is of high demand both in national as well as international market. These goats are also used as a source of meat, skin and manure. A typical moving herd of Changthangi goat is shown in Fig. 7.



Fig. 6. Typical herd of Changthangi goat in Changthang area of Ladakh

Changthangi Sheep–Dual Purpose Breed

The Changthangi sheep is also a very important livestock species found in Changthang area of Ladakh. Locally, this is also known as Changluk sheep (Chang-Northern, Thang Plateau/Pfain, Luk-Sheep). Similar to Changthangi goats, these sheep are also reared by local nomads of Changpa tribe. This is a dual-purpose breed mainly reared for mutton, and wool production. The wool of Changthangi sheep is mainly used locally by the changpa nomads for preparing many types of items such as rugs, socks, sweaters, quilts, mattresses etc. Changthangi sheep is a double coated animal with a marked difference in diameter and surface characteristics of primary and secondary fibres. The average fibre diameter, medullation percentage, staple length, number of crimps, fibre length, scouring yield and burr content were $31.19 \pm 0.71 \mu\text{m}$, $11.37 \pm 0.97 \%$, $11.34 \pm 0.55 \text{ cm}$, 4.13 ± 0.36 , $134.58 \pm 3.56 \text{ mm}$, $81.06 \pm 0.94 \%$ and $3.19 \pm 0.65 \%$, respectively. Changthangi wool is a medium type wool suitable for making finer textured fabrics. Further, the undercoat having finer diameter can be utilized for making luxurious fabrics next only to pashmina.

Double Hump Camel

Double hump camel (*Camelus bactrianus*) is another important and unique livestock species which is very well adapted to high altitude environments. The double hump camels are even-toed ungulate animal with a unique characteristic like two humps at its back and

has long woolly hairs on its body. The body size and shorter in height as compare to single hump camel. These animals are only living remnants of India's connection with famous silk route. In India, Bactrian camels first appeared in Ladakh in 1870, were brought by travelers and traders from Yarkand in Xinjiang province of modern-day China. These double hump camels thrive in the high-altitude cold desert ($+30^\circ\text{C}$ to -40°C) region of Nubra Valley, Partapur, Leh-Ladakh and survive hypoxic and hypobaric condition. As per the estimate in 2020, there are about 298 double camels in Ladakh. The photo of typical double hump camel in nubra valley is shown in Fig. 7. The total lifespan of Bactrian camel is between 20-38 years. An adult Bactrian camel stands up to 63-69 inches with the body length of 52-71 inches. These camels are known for their ability to endure long periods of travel without water and food for a week, even in harsh desert conditions. These camels have the ability to carry weight up to 200 to 250 kgs on the hilly rough terrain. With the development of tourism industry in Leh-Ladakh, this camel has become a source of tourist attraction in the region especially in Nubra valley.

Conclusion

It is expected that the Ladakhi cattle, yak, donkey, pashmina goat, changthangi sheep, double hump camel and zanskari ponies, the multifunctional local breeds well adapted to high altitude will continue to play a role in the livelihoods of poor people and marginal areas of Leh-Ladakh. As a part of mission towards zero non-descript,

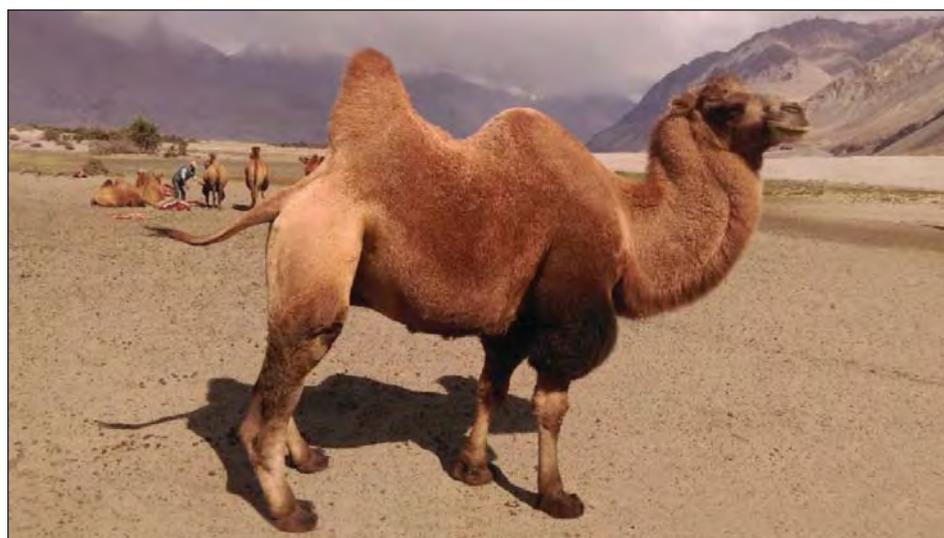


Fig. 7. Typical double hump camel

ICAR-NBAGR efforts are underway to characterize non-descript populations of Ladakh such as Changkhi dog, Malluck sheep and Malra goat populations. The cataloguing, documentation and characterization of remaining non-descript populations will have wider impact on the local tribal communities of Ladakh whose livelihood is dependent on these genetic resources.

References

- Bumla, Nazir A., A Maria, Jasvinder S Sasan, and Adil M Khateeb (2012). Quality of Indian pashmina fibre in terms of its physico-mechanical properties. *Wayamba J. Anim. Sci.*: 459-462.
- Ganai, TAS, SS Misra, and Feroz Din Sheikh (2011). Characterization and evaluation of Pashmina producing Changthangi goat of Ladakh. *Indian J. Anim. Sci.*, 81, 6: 592-599.
- Malik, Asma Altaf, Hilal Musadiq Khan, Asif H Sofi, Masood Saleem Mir, Javid Farooq, Feroz D Sheikh, Abdul Qayoom Mir and Muzamil Abdullah (2021). Wool characteristics of Changthangi sheep. *Small Rumin. Res.*, 199: 106364.
- Misra, RK, B Singh and VK Jain (1998). Breed characteristics of Changthangi pashmina goat. *Small Rumin. Res.* 27, 2: 97-102.
- Monika Sodhi, RS Kataria, Saket K Niranjana, Parvesh K, Preeti Verma, Shelesh K Swami, Ankita Sharma, Vijay K Bharti, Bhuvnesh Kumar, M Iqbal, Stanzen Rabgais, Prabhat Kumar, Arup Giri, Sahil Kalia, Deepak Gagoi, Prabhu Prasad Sarangi and Manishi Mukesh (2018). Sequence characterization and genotyping of allelic variants of beta casein gene establishes native cattle of Ladakh to be a natural resource for A2 milk. *Def. Life Sci. J.*, 3: 177-181.
- Preeti Verma, Ankita Sharma, Monika Sodhi, Kiran Thakur, Vijay K Bharti, Prabhat Kumar, Arup Giri, Sahil Kalia, Shelesh Kumar Swami, Manishi Mukesh (2018b). Overexpression of genes associated with hypoxia in cattle adapted to Trans Himalayan region of Ladakh. *Cell Biol. Int.*, 2018, 42 (9): 1141-1148.
- Preeti Verma, Ankita Sharma, Monika Sodhi, Monika Thakur, Ranjit S. Kataria, Saket K. Niranjana, Vijay K Bharti, Prabhat Kumar, Arup Giri, Sahil Kalia, & Manishi Mukesh (2018a). Transcriptome analysis of circulating PBMCs to understand mechanism of high-altitude adaptation in native cattle of Ladakh region. *Sci. Rep.* 16; 8(1): 7681.
- Saket Kumar Niranjana, Ranjit S Kataria, Monika Sodhi, Vijay K Bharti, Bhuvnesh Kumar, Ajay Garg, MC Yadav, Ankita Sharma, Prince Vivek, Arup Giri and Manishi Mukesh. Evaluation of physiological parameters in response to endurance exercise of Zanskar ponies adapted to high altitude of Ladakh region. *Def. Life Sci. J.*, 3(2): 172-176.

Genetic Uniqueness for Immune Response Traits in Native Animal Genetic Resources

RS Kataria*, Ankita Gurao, Ravinder Singh, SK Niranjan and BP Mishra

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

Breeding of animals for disease resistance is an important intervention in the era of emerging and re-emerging pathogens in livestock and poultry. Livestock and poultry species of India are considered as an important genetic resource for climate resilience as well as relative less susceptibility to tropical diseases. Indian livestock populations with a broad genetic base, due to availability of diverse breeds and populations, have advantage over the exotic counterpart. Introduction of cross breeding in India, cattle in particular, with exotic germplasm, has further helped in identification of several genes and pathways involved in gaining better immunity against the prevalent diseases. Unique alleles have been identified in the major histocompatibility region and other immune response genes, including toll-like receptors, having important role in disease resistance. Major impediment in associating genetic polymorphism with the disease resistance however, is the non-availability of standardized phenotypic data. Though attention is being diverted towards understanding the genetic basis of disease resistance in native livestock and poultry, there is a need to utilize the latest genome based technologies for fully understanding as well as exploiting the genetic potential of indigenous breeds.

Introduction

The biotic stress which includes pathogenic insults is the prime selecting factor for the evolution in all vertebrates. The pathogens, invading glands, mucosal surfaces of digestive tract, reproductive tract etc. are sensed first by the physical or chemical barriers of the innate immune system. After the failure of these barriers, the system reacts by initiating a cascade of immune cell-mediated responses. The innate immunity is functionalized by several preexisting defenses such as proteins and peptides secreted by the resident macrophages, mobile neutrophils etc., while the adaptive immune response functions in a more sophisticated way, and enables the living system to create a record of past intruders/foreign particles/antigens. The livestock that have adapted and evolved close to humans have influenced greatly the nutritional status, economic affairs, and social life of their human counterparts. Health status as well as ability of the animals to recover from the infectious diseases determine not only the production/economic aspect, but also poses threats to zoonosis and major outbreaks in the human population as evident from anthropological history. Recent COVID outbreak is a good example of SARS-CoV2 crossing the human-animal barrier and causing havoc.

Livestock sector plays a significant role in India's rural livelihood as it employs a major section of the

labour force and also provides a large share of draft power, being used for the cultivation of crops. India's livestock sector is one of the largest in the world with 535.8 million population, registering growth of 4.6% from the last census of 2012 to 2019. Contribution of livestock sector to the national economy in terms of Gross Domestic Product (GDP) is 4.1% and 25.6% of total agricultural GDP. India is one of the few countries in the world, where most of the varied natural agro-climatic conditions are available within the same region, ranging from the arid/semiarid Thar Desert, tropical and costal region, arid cold region of Ladakh. Indian livestock landraces/breeds are well adapted to the diverse geo-climatic conditions of the country. As a result, these breeds can survive the rugged gradient of temperature, humidity, hypoxic and subzero temperature, landscapes etc. without much management interventions, under an extensive system of rearing.

The immune system's function is influenced by multiple factors such as genetic make-up, environmental influences, behavioral patterns, dietary factors, etc. Those animals/livestock, who cannot control the environmental influences are forced to sustain the stresses and eventually shape their genetic makeup and these beneficial traits get fixed in the long run by the virtue of adaptive evolution. Consequently, the local landraces of livestock that have evolved in the native environment, are more resistant to

*Author for Correspondence: *Email-Ranjit.Kataria@icar.gov.in

pathogen attacks than their exotic counterparts. Several innate and adaptive immune genes have been implicated, when analyzing variations in cohorts of disease-resistant or susceptible production animals. Since the race between the immune system and the ever-evolving pathogenic forces is very dynamic, a thorough understanding of livestock immunology can help in building the strategies to overcome the potential outbreaks. In this aspect, the local animal genetic resources are major germplasm carrying the plethora of beneficiary genetic and physical traits needed to combat the locally prevalent diseases.

All the livestock species in India are very well adapted to diverse the agro-climatic conditions and are considered to be better equipped to fight the tropically prevalent infectious diseases in the region, than the exotic breeds and presence of unique genetic attributes could play a major role in that. Genetic selection to increase resistance against infectious diseases, will prove to be a low cost and sustainable practice. Besides, all the above properties, there are some of the common qualities in our pure breeds, cattle in particular. Their skin is having sweat and sebaceous glands, which secrete an oily fluid, acting as repellent to the insects and mosquitoes. It also decreases the rate of most of vector borne infections. In case of chlamydial abortion, Johne's disease, Babesiosis, Tuberculosis, Mycoplasmosis, Leptospirosis, Brucellosis and diarrhea, our native breeds show better resistance. The globulins levels in the blood of pure Indian breeds are high, while creatinine level is less than that of the crossbreds or exotic cattle. Indicus cattle are also considered relatively resistant to mastitis and ticks as well as tick borne protozoan diseases such as Babesiosis and Theileriosis, exotic and crossbred being susceptible to. Studies have shown Holstein and Jersey crossbreds of India having a higher risk (94.54%) of mastitis than the local cattle breeds (31.25%). Similarly local indigenous chicken are reported to be more resistant to *Pasteurella multocida* infection than exotic.

Among the various immune response genes, keeping in mind the disease resistance related traits, major histocompatibility complex (MHC) genes play a major role. The bovine major histocompatibility region, BoLA contains approximately 200 genes, most of which are immune-related. Class II bovine MHC genes polymorphism are considered to be most likely influencing the immune traits, among which the BoLA-DRB3 has shown more influence on ability to evoke the immune response. Selection, genetic drift, and population bottlenecks have significant effect on

the allelic diversity of MHC class I and class II loci in a population. Genetic diversity in MHC genes is also considered as an important marker for the assessment of survival and risk status of animal species. Comparative analysis of indicus and exotic as well as crossbred cattle has shown extensive allelic diversity as well as specific alleles at the DRB3 and DQA loci of MHC region, associated with resistant to sub-clinical and clinical mastitis in indicus cattle breeds of India. A high level of duplication and allelic diversity has been reported for BoLA-DQA in Mewati, Konkani, Tharparkar, Ladakhi, Malnad Gidda, Ongole, Gir, and Sahiwal cattle breeds (Kumari *et al.*, 2021). Similarly, in buffalo extensive duplication and variability have been reported for DQA alleles (Mishra *et al.*, 2020).

Another study conducted on clinical mastitis incidents in indicus and crossbred cattle, clearly indicated that glutamine and glutamic acid at positions 62 and 66 playing an important role in resistance to clinical mastitis. While, arginine at 62 position and deletions of amino acid at positions 58 and 59 make the animals more susceptible to mastitis. Expression of BoLA-DQA, DQB and DRB3 gene of healthy and mastitis affected indicus, exotic and crossbred animals has been studied and there was significant higher expression of the BoLA-DQA gene in healthy indicus cattle compared to mastitis affected indicus cattle and both healthy and mastitis affected exotic and crossbred cattle. Whereas the other studied genes BoLA-DQB and DRB3 presented no significant differences in expression in healthy and mastitis affected indicus, exotic and crossbred cattle. This indicates that duplication of gene and polymorphism probably playing more significant role in differential immune response among indicus and crossbred/exotic cattle at MHC locus. A study on response to vaccination against PPR and MHC locus diversity in Sirohi goat has revealed rich diversity of major histocompatibility complex (MHC) region in Indian goat.

Pattern Recognition Receptors Diversity

Nucleotide-binding oligomerization domain-containing protein 2 (*NOD2*) and toll-like receptors (*TLR*) are the foremost receptors to identify the pathogen-associated molecular patterns (*PAMPs*). Toll-like receptors are transmembrane proteins with an extra-cellular domain consisting of leucine-rich repeats (LRRs) domain and cytoplasmic (TIR) domain. A total of 13 TLRs have been identified in mammals. In Indian buffalo, all the ten TLR genes reported in bovines, have been

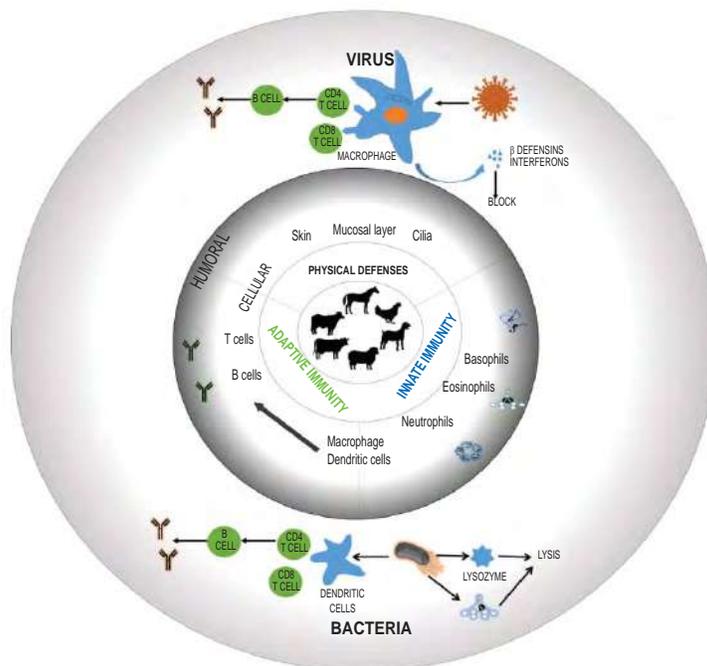


Fig. 1. The immune cell components and host-pathogen interaction in livestock

characterized, documenting the complete gene sequence based structural variability across bovines (Dubey *et al.*, 2013). The studies have shown the differential evolutionary lineages of buffalo TLRs compared to cattle and other ruminant species. The non-synonymous SNPs in other receptor gene, *NOD2* have been associated with incidences of mastitis in buffaloes (Dhiman *et al.*, 2017). The *NOD2* works in synergy with other PRRs, *TLR2* in a dose-dependent manner, and also cross talks of *NOD2* and *TLR2* leads to the activation of *NOD2-TLR2* specific gene panel, otherwise, they both have different signaling cascades.

Other Immune Response Genes

Several candidate genes have been identified with genetic variability having significant association with disease resistance in Indian livestock breeds. *NRAMP1* (natural resistance-associated macrophage proteins), is a divalent iron transporter, pumping iron out of the phagosome, which is considered as an important defense against mycobacterial infections in particular. The polymorphism within the gene has also been linked to other intra-cellular pathogens like *Brucella* and *Salmonella* infections. The cases of bovine tuberculosis (bTB) are reported higher in cattle than the buffaloes, particularly, the prevalence of bTB is more in cross-bred than the indigenous cattle of India. The beta-defensins have been widely associated with incidences of mastitis. Particularly, the *LAP* and

BNBD4 expression is reported to be upregulated in PBMCs of mastitis-affected cattle and buffalo (Gurao *et al.*, 2018). Polymorphism and unique alleles within several other candidate genes including cytokines have been identified in indigenous livestock breeds, which need to be further investigated. Targeted sequencing of exonic regions of *CHGA*, *CHGB*, *CHGC*, *NRAMP1*, *NRAMP2*, *DEFB1* and *TAP* genes in Gir cattle and Murrah buffalo revealed 54 and 224 SNPs, respectively. Polymorphism identification in innate immune genes would advance our knowledge about the role of these genes in elucidating the disease resistance/susceptibility in Indian breeds.

Whole Genome Based Studies

Molecular markers are more important in breeding of animals through selection for traits like disease resistance, with low to moderate heritability and for which measurement of phenotype is difficult. Advanced technologies, including whole genome sequencing, transcriptome and proteome analysis will help in dissecting the genomic variation and developing molecular markers useful for selection of livestock and poultry resistant to diseases. Several breeds/populations still need to be explored genetically to answer the question of being resistant to many tropical diseases. Transcriptome analysis of Foot and Mouth Disease (FMDV) serotype O/IND/R2/75 infected two indigenous

cattle breeds, Malnad Gidda, and Hallikar, and an exotic breed Holstein Frisian, identified MHC class II, *IFN- α* , *IFN- γ* , *IFN- λ 3*, and *RSAD2* ('viperin) transcripts' to be upregulated in the indigenous cattle than in the exotic cattle breed. This indicates that the indigenous cattle breeds could neutralize the FMDV better than the exotic breeds (Saravanan *et al.*, 2021).

A whole-genome sequence based comparative analysis of Indian cattle breeds, Kangayam, Tharparkar, Sahiwal, Red Sindhi, and Hariana, identified the genetic variants, within the genes and pathways involved in the innate immune response (Vijayakumar *et al.*, 2022). Additionally, the workers suggested non-synonymous polymorphism in antiviral effectors genes such as *MX1*, *OASL*, *IFITM5*, *IFITM3*, *IFIT5*, *TGFB1*, *GBP1*, *GBP7* and *RSAD2* in the same panel of indigenous cattle breeds, should be considered for association between BVDV persistence and antiviral activity. Major variants identified were within toll-like receptors, retinoic acid-inducible gene I-like receptors, NOD-like receptors, Jak-

STAT signaling pathways, and several non-synonymous variants in the candidate immune response genes. These pathways and genes directly or indirectly contribute to the disease resistance of *Bos indicus* cattle breeds. Genome wide analysis in indicus Sahiwal cows could identify important candidate genes for clinical mastitis, which included *DNAJB9*, *ELMO1*, *ARHGAP26*, *NR3C1*, *CACNB2*, *RAB4A*, *GRB2*, *NUP85*, *SUMO2*, *RBPJ*, and *RAB33B* genes. The findings helped in elucidation of the genetic architecture of the disease in *Bos indicus*, and potential regions for the fine mapping and future exploration.

Future Prospectives

Although the work carried out so far has clearly indicated that the indigenous livestock species carry significant genetic variation, making them resistant to many tropical diseases, exotic germplasm is susceptible to. Recently developed next generation sequencing tools, have been helping in delineating the molecular basis

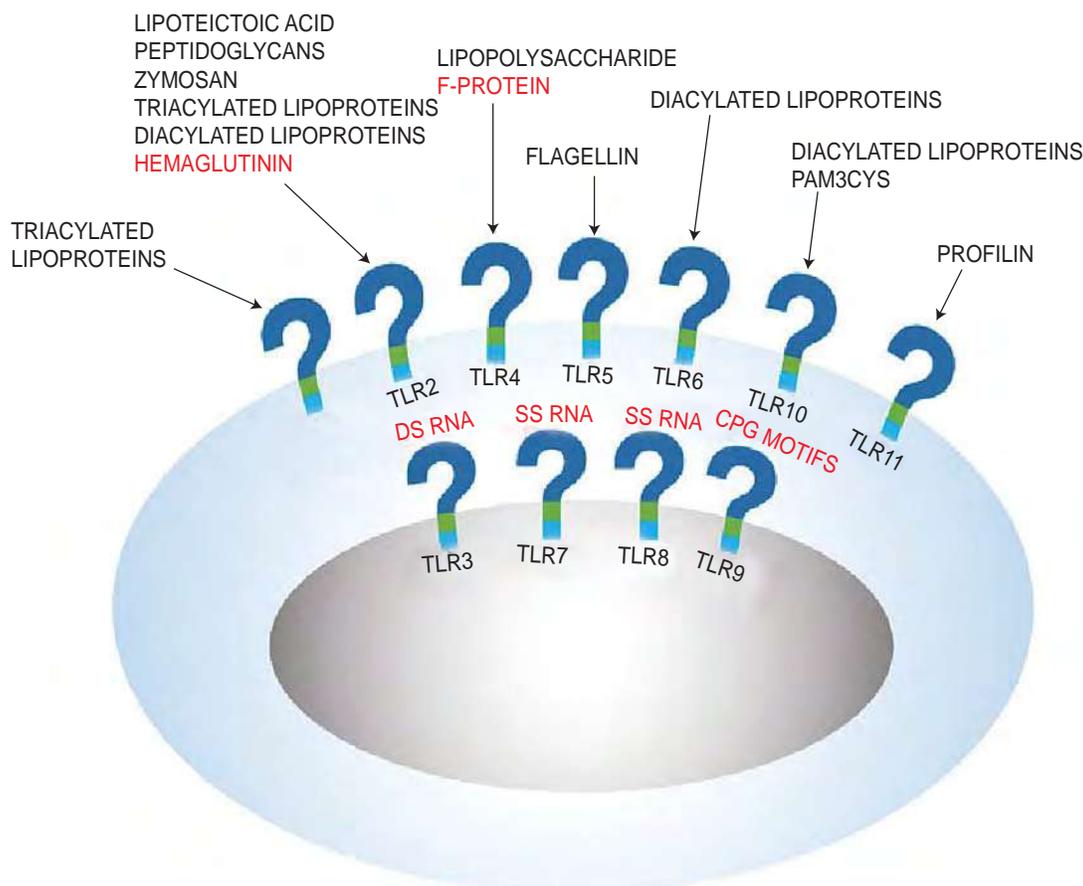


Fig. 2. Cellular localization and the ligand for various Toll-like receptors

of such privilege, original native livestock species are endowed with. Genome sequencing supplemented with transcriptome, proteome and epigenome exploration will pave the new ways for deep understanding as well as exploitation of native germplasm for breeding animals more resistant to diseases. Prediction of genomic breeding value through identification of SNPs in native germplasm will fasten the genetic gains against disease resistance traits. However the availability of quality phenotypic data on the disease resistance traits is the major issue, which needs to be resolved for faster implementation of genetic selection of animals more resilient to tropical diseases.

References

- Dhiman A, SK Mishra, PK Dubey, S Goyal, M Sehgal, SK Niranjana, M Sodhi, BP Mishra and RS Kataria (2017) Identification of genetic variation in NOD-like receptor 2 gene and influence of polymorphism on gene structure and function in buffalo (*Bubalus bubalis*). *Research in Veterinary Science* **115**: 43-50.
- Dubey PK, S Goyal, P Kathiravan, BP Mishra, SK Gahlawat and RS Kataria (2013) Sequence characterization of river buffalo Toll-like receptor genes 1–10 reveals distinct relationship with cattle and sheep. *Int. J. Immunogenet.* **40**: 140-148.
- Gurao A, RS Kataria, R Singh, RD Kumar, SK Mishra and SK Kashyap (2018) Association analysis of differential expression of beta defensins with mastitis in indicine dairy cattle. *Ind. J. Dairy Sci.* **71**: 534-537.
- Kumari N, SK Mishra, S Saini, A Kumar, S Loat, N Dhilora, SK Niranjana, M Sodhi and RS Kataria (2021) Identification of novel allelic patterns and evolutionary lineage of BoLA MHC class II DQA locus in indicine and taurine cattle. *Anim. Biotechnol.* **4**: 1-7.
- Mishra SK, SK Niranjana, R Singh, P Kumar, SL Kumar, B Banerjee and RS Kataria (2020) Diversity analysis at MHC class II DQA locus in buffalo (*Bubalus bubalis*) indicates extensive duplication and trans-species evolution. *Genomics* **112**: 4417-4426.
- Saravanan S, N Guleria, HB Ranjitha, BP Sreenivasa, M Hosamani, C Prieto, V Umapathi, HK Santosh, S Behera, VV Dhanesh and GS Krishna (2021) Induction of antiviral and cell mediated immune responses significantly reduce viral load in an acute foot-and-mouth disease virus infection in cattle. *Genomics* **113**: 4254-4266.
- Vijayakumar P, A Singaravadivelan, A Mishra, K Jagadeesan, S Bakayaraj, R Suresh and T Sivakumar (2022) Whole-genome comparative analysis reveals genetic mechanisms of disease resistance and heat tolerance of tropical *Bos indicus* cattle breeds. *Genome* **202**(64): 241-254.

A1/A2 Milk Research in Indian Cattle

Monika Sodhi*, M Mukesh, RS Kataria, SK Niranjana and BP Mishra

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

Bovine beta casein A1 and A2 are the most common variants in cattle breeds. Because of the bioactive peptide beta casomorphin-7 (BCM7) produced by raw or processed A1-milk, which has high affinity for opioid receptors and can exert regulatory activities, A1 variant is considered a risk factor for many human diseases, but not by A2-milk, during digestion. The frequency of A1/A2 allele of beta casein is breed dependent and can be very easily checked with simple PCR-RFLP technique. The aim of this article was to review the different studies and assess the status of A1/A2 in Indian native cattle breeds as well as crossbred/taurine populations and breeding bulls being used at different AI centres in India. Analysis of frequency pattern clearly indicated that all Indian native cattle breeds have high frequency of A2 allele and homozygous A2A2 genotype and hence are very good resource for A2 milk. Crossbred, taurine cattle populations and breeding bulls being used at different AI centres have a higher frequency of A2 allele and A1A2 genotype. The percentage of animals with homozygous A2A2 genotype is low. This suggests the need for screening of crossbred/exotic bulls being used in A1 and modify the existing breeding policy so as to drift the herds towards A2.

Introduction

Milk from a variety of livestock species and cow in particular has been included in the diet for infants, children and adults worldwide since it is the most common source of animal proteins and microelements known to be essential for human nutrition. Worldwide, the different sources of milk are cow, buffalo, goat, sheep and camel contributing 85%, 11%, 2%, 1.4% and 0.2% respectively to the total world milk production. With a herd capacity of 264 million cows contribute highest to the total milk production that is 600 million tons every year. Cow milk contains various components like lipids, proteins, amino acids, vitamins, minerals, immunoglobulins, hormones, growth factors, cytokines, nucleotides, peptides, polyamines, enzymes and other bioactive peptides with physiological functionality. It also provides a high quality source of energy, proteins and selected micronutrients such as calcium, magnesium, zinc and phosphorus to most human population. Cow milk generally contains about 3.5 % protein, of which approximately 80 % are caseins and 20 % are whey proteins. More than 95% of the cow milk proteins are constituted by caseins ($\alpha S1$ -, $\alpha S2$ -, β -, k -CN) and whey (α -LA and β -LG) genes. Milk and milk products are considered as functional foods as digestion of different components of cow milk specially proteins lead to the formation of biologically active molecules that can have a direct and significant effect on health (Marshall 2004)

including digestive functions, metabolic responses to absorbed nutrients, growth and development of organs or different diseases.

Beta Casein Variants

Among the caseins, beta casein, the second most abundant protein with an excellent nutritional balance of amino acids is a rich source of such bioactive peptides. It holds special significance as the bioactive peptides may exert regulatory activities in the human beyond nutrition and also act as promoters of different physiological functions (Kostyra *et al.*, 2004; Silva and Malcata, 2005). The bovine beta casein (β -CN) gene spanning over a region of 8.5 kb, has nine exons and different mutations have led to formation of 15 genetic variants (A1, A2, A3, B, C, D, E, F, G, H1, H2, I, J, K and L). These variants hold importance as digestion of β -casein with different variants not only result in generation of different group of active peptides (Nguyena *et al.*, 2015), but also has an influence on milk protein composition and milk-production traits according to its genetic polymorphism (Visker *et al.*, 2011).

Among these variants, A1 and A2 are the most common genetic variants, with A2 being the ancestral allele/variant of the gene. These variants differ at amino acid position 67 corresponding to single nucleotide difference cytosine to adenine. β -casein allele A2 has cytosine at position 8101, codon CCT corresponding

*Author for Correspondence: Email-Monika.Sodhi@icar.gov.in

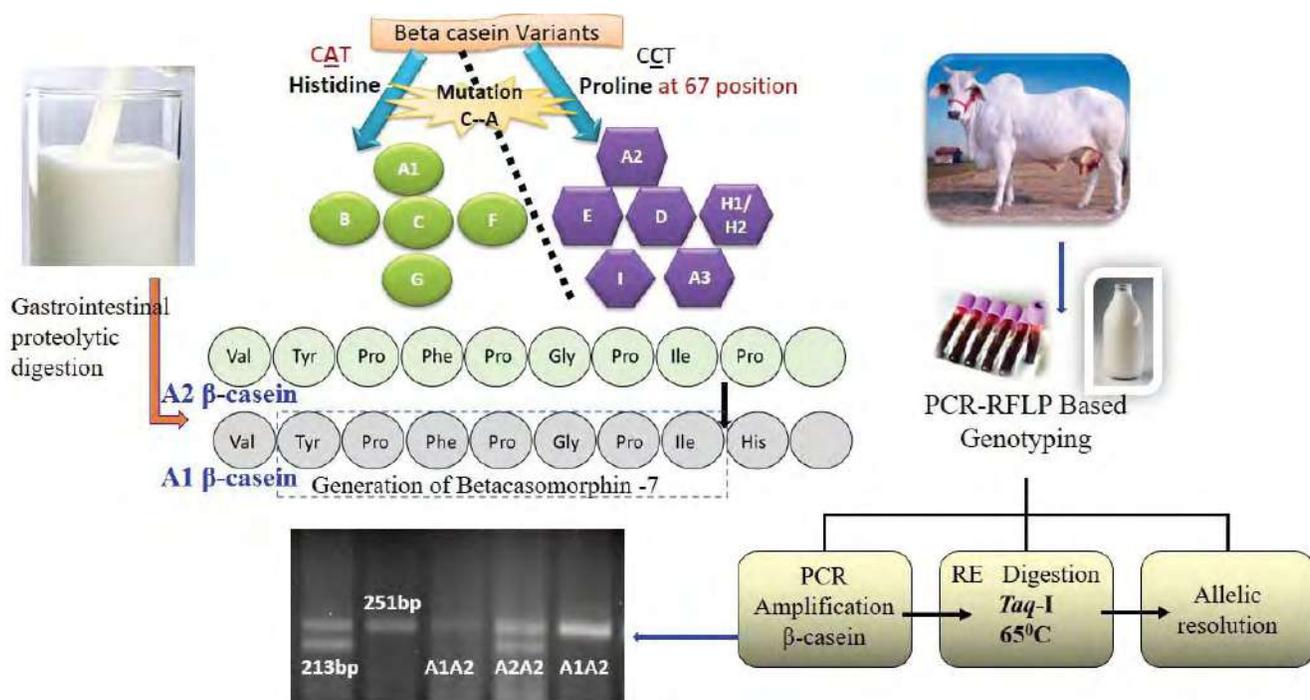
to amino acid proline at position 67. In variant A1, conversion of cytosine to adenine leading to codon change from CCT to CAT replaces proline with histidine at position 67. Based on this A1/A2 variation, milk is generally referred as A1 or A2 milk. On the basis of common variation of Pro67His change, the variants, A1, B, C, F or G having a common amino acid histidine (His) at position 67 (–Tyr60–Pro61–Phe62–Pro63–Gly64–Pro65–Ile66–His67–), but variations at other positions of amino acids are called A1 type. Similarly, variants A2, A3, D, H1, H2 and I alleles having a common amino acid proline (Pro) at position 67 (–Tyr60–Pro61–Phe62–Pro63–Gly64–Pro65–Ile66–Pro67–), but variations at other positions are categorized as A2 type.

Bioactive Peptides from Beta Casein A1/A2 Variants

A1 and A2 variants of bovine β -casein, generally called A1/A2 milk differ at amino acid position 67 with histidine in A1 variant/A1 milk and proline in A2 variant/milk. This polymorphism leads to key conformational changes in the secondary structure of expressed β -casein protein. Because of the structural differences, the bioactive peptides generated upon digestion of A1/A2 beta casein are different. Gastrointestinal proteolytic digestion (leucine aminopeptidase, elastase and carboxypeptidase

Y) of A1 β -casein (raw/processed milk) releases a 7 amino acid bioactive peptide ‘opioid’ called beta-casomorphin 7 (BCM-7) (Fig 1) in small intestine, while proline in A2 β -casein at 67 position prevents the split at this particular site and generates nine amino acid peptide BCM-9 (Kostyra *et al.*, 2004; De Noni 2008). Release of BCM-7 from A2 β -casein is minimal under normal gut conditions. In hydrolysed milk with A1 beta-casein variant, BCM-7 level is 4-fold higher than in A2 milk. Further, BCM-7 is released not only from milk but also from almost all milk product including yoghurt and cheese, infant formulas

Beta-casomorphin suggest the peptide from beta casein with opiate properties similar to morphine, that includes affinity to opioid receptors, especially the MOP (μ -opioid receptor). The beta-casomorphine-7 has been identified as the “atypical” opioid peptide and exerts its influence on nervous, digestive, and immune functions via the MOR. In addition to BCM7/9, digestion of beta casein results in the release of other encrypted casomorphins including the non-opioid peptides (BCM5, BCM11, Immunopeptides). The BCM-9 is also an opioid agonist but with lesser affinity for μ -opioid receptor. The BCM-5, which is the more potent than BCM-7 and BCM-9,



Different genotypes for A1/A2 allele of β -casein

Fig. 1. Different variants of Beta-casein, their digestion and genotyping protocol

is primarily released from further proteolytic digestion of BCM-7 and BCM-9 by brush border peptidases. Various epidemiological, biochemical data or animal trials suggest the association of A1 milk (cow milk with A1 β -casein) as a risk factor for digestive discomforts, type I diabetes, coronary heart disease, arteriosclerosis, sudden infant death syndrome etc. (Kuellenberg *et al.*, 2022; Sodhi *et al.*, 2022; Yadav *et al.*, 2020; Brooke-Taylor *et al.*, 2017; Laugesen and Elliott, 2003; Tailford *et al.*, 2003)

Status of β -casein Variants across Taurine Breeds

Although a clear link between A1 β -casein and a disease state has not yet been confirmed, the importance of monitoring the status of A1/A2 alleles in dairy animals

as a precautionary measure has been realized. The distinguishing amino-acid sequence that characterizes A1 beta-casein is essentially unique to some European cattle breeds. Asian and African cattle; goats, sheep, yak and camel; all produce A2 milk. Human beta-casein is also of the A2 type as defined by the relevant amino-acid sequence (proline) fixation of A2 allele in buffalo breeds have also been reported in different studies (Mishra *et al.*, 2009; Ramesha *et al.*, 2016). Animals belonging to Genus Bos, which includes *Bos Indicus* (Indian humped Zebu), *Bos taurus* (Exotic) and Yak were initially of A2A2 type only but later due to point mutation A2 allele was replaced with A1 some 5000-10,000 years ago and some animals got A1A2 or A1A1 genotype (Ng-Kwai

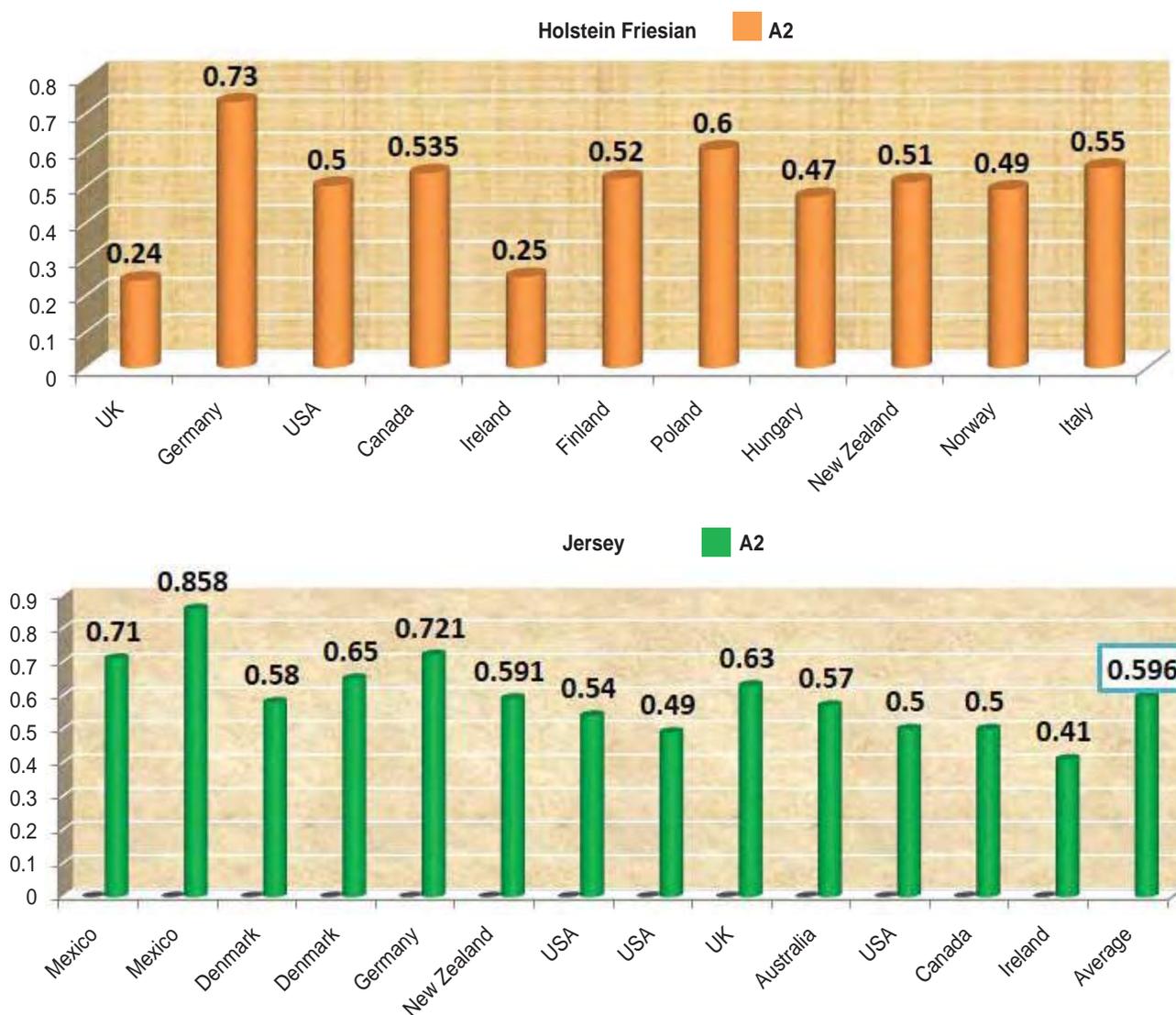


Fig. 2. Status of A1/A2 variant in *Bos taurus* breeds

and Grosclaude, 2002). With the advent of selective breeding for high production, better fertility and protein quality, unconsciously, genetically superior bulls carrying A1 allele were used in breeding programs. Hence, A1 variant became prominent in European cattle some thousands of years ago while making selection for high milk production. The frequency of A1 beta-casein with A1 variant varies breed by breed. A1 β -casein is a major variant of β -casein in the milk of the common dairy cows of north European origin: Friesian, Ayrshire, British Shorthorn, and Holstein. Also, the frequency of A1 has increased over the last century as the *Holstein Friesian (HF)* has become predominant in many countries for its high milk production character. Artificial insemination of large number of cows with semen of *HF* bulls multiplied A1 gene frequency over time. Several reports indicate that A1/A2 frequency is breed as well as area specific. Frequency of A1 allele of beta casein is high in Holstein Friesian (HF) breed in North America but low in German HF. Overall, across countries frequency of A1 allele in HF ranges between (40–65%). Compared to HF, frequency of A2 allele is higher in Jersey. Breeds like Guernsey, Kerry, Channel Island cows; Southern French breeds; Charolais and Limousin also have higher frequency of A2 allele compared to HF.

Indian Native Cattle Breeds: A Resource for A2 Allele

Cattle genetic resources are the backbone of the farmer's economy since centuries producing milk as well as draught power. With 190.90 million cattle that is around 12.5% of world cattle population and producing 165.4 million tons of milk, India ranks first in the total milk produced annually (Livestock animal-husbandry-statistics, 2018). India is home to some of the best zebu (*Bos indicus*) breeds represented by 50 well-defined breeds. Besides these, many lesser-known cattle populations are also spread out in the length and breadth of the country each having its own special features contributing towards milk pool. The status of A1/A2 alleles of β -casein gene was delineated systematically first time at ICAR-NBAGR, Karnal in 2009, under the DBT funded project and later in National fund project. In a comprehensive analysis, nearly 4000 animals representing 27 breeds of Indian cattle and 231 animals from eight buffalo breeds have been genotyped to understand the distribution of A1/A2 variants. Blood samples were collected from the random, true to the breed

animals by visiting the specific breeding tract of the breed. DNA was isolated and PCR-RFLP genotyping protocol was followed to ascertain the allelic status. The process included amplification of 251bp region of β -casein exon 7 harbouring the mutant site using specific primer pair restriction digestion of the PCR products with *Taq-I* restriction enzyme followed by visualization on 3% Ethidium bromide stained agarose gel. Three restriction fragments corresponding to different genotypes: A1A2 (251 & 213bp); A2A2 (251bp) and A1A1 (213bp) were observed for the analysed samples (Fig. 1).

The data on 15 Indian native cattle breeds (Mishra et al., 2009) from different agroclimatic regions and of different utility (milch, dual and draft purpose) revealed that majority of the Indian native cattle (97.4%) have A2A2, the desirable genotype, followed by heterozygous A1A2 (2.6%) genotype. None of the animal showed homozygous A1A1 genotype. Out of 861 animals screened, only few animals of Malnad Gidda and Kherigarh cattle showed heterozygous A1A2 genotype with frequency of 0.191 and 0.218, respectively. The study was further extended to 3400 animals representing 27 Indian native cattle, crossbred (Karan Fries and Frieswal) and exotic (Holstein Friesian and Jersey) cattle (Mukesh et al., 2022). Across the Indian native cattle, highest frequency was observed for A2A2 genotype (0.905), followed by heterozygous A1A2 (0.091) and homozygous A1A1 (0.004) genotype. The overall frequency of favourable A2 allele across all the 2500 animals genotyped was 0.95 and its distribution across the different utility types was 1.0, 0.95 and 0.92 for milch, dual and draft purpose breeds, respectively. As observed by Mishra et al. (2009), all the animals belonging to dairy/milch breeds (Gir, Tharparkar, Rathi, Red Sindhi, and Sahiwal) showed only homozygous A2A2 genotype that is fixation of A2 allele. The other cattle breeds showing complete absence of A1A1/A1A2 genotype were Belahi, Konkan Kapila, Kangyam, Nimari, Red Kandhari, Malvi, Amritmahal, Kankrej, Hariana and Mewati (Table 1). The percent of heterozygous genotype was higher in draft purpose breeds compared to dual purpose breeds. Across the different climatic regions, breeds from arid and semiarid region showed higher frequency of A2 allele that is 0.98 and 1.00, respectively. Frequency of A2 allele in breeds from humid subtropical region mainly consisting of mainly draft and dual-purpose breeds was 0.90.

Table 1: Frequency profile of A1/A2 allele of beta casein and corresponding genotypes across Indian native, crossbred and taurine cattle in India

S No	Breed/ Population	Allelic Frequency		Genotypic Frequency			Sample No	References
		A1	A2	A1A1	A1A2	A2A2		
1	Gir	0.000	1.000	0.000	0.000	1.000		Mukesh <i>et al.</i> , 2022
		0.146	0.854	-	-	0.833	24	Patel <i>et al.</i> , 2020
		0.172	0.828	0.006	0.331	0.663	172	Patel <i>et al.</i> , 2019
		0.000	1.000	0.000	0.000	1.000	45	Mishra <i>et al.</i> , 2009
	Gir Bulls	0.222	0.778	0.037	0.370	0.593	54	Patel <i>et al.</i> , 2019
2	Rathi	0.000	1.000	0.000	0.000	1.000		Mukesh <i>et al.</i> , 2022
		0.000	1.000	0.000	0.000	1.000	30	Saran <i>et al.</i> , 2019
		0.000	1.000	0.000	0.000	1.000	46	Mishra <i>et al.</i> , 2009
3	Red Sindhi	0.00	1.00	0.00	0.00	1.00		Mukesh <i>et al.</i> , 2022
		0.000	1.000	0.000	0.000	1.000	33	Mishra <i>et al.</i> , 2009
4	Sahiwal	0.00	1.00	0.00	0.00	1.00		Mukesh <i>et al.</i> , 2022
		0.15	0.85	0.000	0.30	0.70		Pandey <i>et al.</i> , 2019
		0.067	0.933	0.005	0.124	0.871	26	Saran <i>et al.</i> , 2019
		0.37	0.63	0	0.75	0.25	12	Srinivas <i>et al.</i> , 2019
		0.06	0.94	0	0.13	0.87	306	Kumar <i>et al.</i> , 2018
		0.00	1	0	0	1.00	15	Haq <i>et al.</i> , 2012
5	Tharparkar	0.000	1.000	0.000	0.000	1.000		Mukesh <i>et al.</i> , 2022
		0.000	1.000	0.000	0.000	1.000	30	Saran <i>et al.</i> , 2019
		0.000	1.000	0.000	0.000	1.000	14	Haq <i>et al.</i> , 2012
		0.000	1.000	0.000	0.000	1.000	44	Mishra <i>et al.</i> , 2009
		0.000	1.000	0.000	0.000	1.000	47	Mishra <i>et al.</i> , 2009
6	Badri	0.09	0.91	-	-	-		Mukesh <i>et al.</i> , 2022
		0.12	0.88	0	0.24	0.76	90	Hussain <i>et al.</i> , 2019
7	Belahi	0.000	1.000	0.000	0.000	1.000		Mukesh <i>et al.</i> , 2022
8	Deoni	0.08	0.92	-	-	-		Mukesh <i>et al.</i> , 2022
		0.29	0.71	0	0.58	0.42	12	Srinivas <i>et al.</i> , 2019
		0.000	1.000	0.000	0.000	1.000	40	Ramesha <i>et al.</i> , 2016
9	Gaolao	0.08	0.92	-	-	-		Mukesh <i>et al.</i> , 2022
10	Hariana	0.000	1.000	0.000	0.000	1.000		Mukesh <i>et al.</i> , 2022
		0.000	1.000	0.000	0.000	1.000	48	Mishra <i>et al.</i> , 2009
11	Kankrej	0.000	1.000	0.000	0.000	1.000		Mukesh <i>et al.</i> , 2022
		0.083	0.917	-	-	-	24	Patel <i>et al.</i> , 2020
		0.107	0.893	0.000	0.214	0.786	28	Patel <i>et al.</i> , 2019
		0.000	1.000	0.000	0.000	1.000	30	Saran <i>et al.</i> , 2019
		0.03	0.97					Rangel <i>et al.</i> , 2017
	0.000	1.000	0.000	0.000	1.000	32	Mishra <i>et al.</i> , 2009	
Kankrej Bulls	0.067	0.933	0.000	0.154	0.867	60	Patel <i>et al.</i> , 2019	
12	Konkan Kapila	0.00	1.00	0.00	0.00	1.00		Mukesh <i>et al.</i> , 2022
13	Ladakhi	0.08	0.92	-	-	-		Mukesh <i>et al.</i> , 2022
		0.100	0.900	0.000	0.210	0.790	82	Sodhi <i>et al.</i> , 2018
14	Malnad Gidda	0.07	0.93	-	-	-		Mukesh <i>et al.</i> , 2022
		0.000	1.000	0.000	0.000	1.000	6	Sridharan <i>et al.</i> , 2022
		0.20	0.80	0	0.40	0.60	10	Srinivas <i>et al.</i> , 2019
		0.014	0.986	0.000	0.029	0.971	104	Ramesha <i>et al.</i> , 2016
		0.096	0.904	0	0.191	0.809		Malarmathi <i>et al.</i> , 2014
		0.21	0.79	0.13	0.18	0.69		Navyashree, 2014
0.096	0.904	0	0.191	0.809	47	Mishra <i>et al.</i> , 2009		

S No	Breed/ Population	Allelic Frequency		Genotypic Frequency			Sample No	References
		A1	A2	A1A1	A1A2	A2A2		
15	Mewati	0.000	1.000	0.000	0.000	1.000		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000	40	Mishra et al., 2009
16	Ongole	0.020	0.980	0.000	0.019	0.981		Mukesh et al., 2022
		0.040	0.96	0	0.080	0.92	12	Srinivas et al., 2019
		0.06	0.94	0	0.11	0.89	38	Ganguly et al., 2013
Draft Cattle Breeds								
17	Amritmahal	0.000	1.000	0.000	0.000	1.000		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000	5	Sridharan et al., 2022
		0.000	1.000	0.000	0.000	1.000	50	Inamdar et al., 2019
		0.000	1.000	0.000	0.000	1.000	37	Mishra et al., 2009
18	Bargur	0.063	0.937	0.00	0.125	0.875	48	Raja et al., 2021
19	Dangi	0.14	0.86	-	-	-		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000	31	Jawane et al., 2018
20	Kangayam	0.000	1.000	0.000	0.000	1.000		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000		Kathiravan et al., 2021
		0.000	1.000	0.000	0.000	1.000	22	Malamathi et al., 2014
		0.000	1.000	0.000	0.000	1.000	48	Mishra et al., 2009
21	Kherigarh	0.00	0.89	-	-	-		Mukesh et al., 2022
		0.109	0.891	0	0.218	0.783	23	Mishra et al., 2009
22	Khillar	0.14	0.86	-	-	-		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000	12	Ramesha et al., 2016
23	Malvi	0.00	1.00	0.00	0.00	1.00		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000	44	Mishra et al., 2009
24	Nagori	0.06	0.94	-	-	-		Mukesh et al., 2022
25	Nimari	0.00	1.00	0.00	0.00	1.00		Mukesh et al., 2022
		0.000	1.000	0.000	0.000	1.000		Mishra et al., 2009
26	Punganur	0.000	1.000	0.000	0.000	1.000	3	Sridharan et al., 2022
		0.080	0.92	0	0.17	0.83	12	Srinivas et al., 2019
27	Ponwar	0.13	0.87	-	-	-		Mukesh et al., 2022
28	Red Kandhari	0.00	1.00	0.00	0.00	1.00		Mukesh et al., 2022
			1.000	0.000	0.000	1.000	39	Mishra et al., 2009
		0.000						
29	Umblachery	0.20	0.80	-	-	-		Mukesh et al., 2022
		0.02	0.98	0.000	0.050	0.950	42	Raja et al., 2021
30	Vechur	0.20	0.80	-	-	-		Muhammed and Stephen, 2012
	Kasargod local	0.39	0.61	-	-	-		
		0.042	0.958	0.000	0.083	0.917	48	Ramesha et al., 2016
Crossbred Cattle								
31	Karan fries	0.29	0.71	0.09	0.40	0.51	460	Mukesh et al., 2022
		0.169	0.831	0.000	0.338	0.662	59	Ramesha et al., 2016
		17.5	82.5	0.08	0.19	0.73	73	Jaiswal K et al., 2013
		0.208	0.792	0.125	0.166	0.709	24	Haq et al., 2012
32	Frieswal							
	Heifers	0.32	0.67	0.12	0.40	0.48	124	Ganguly et al., 2013
	Bulls	0.44	0.56	0.23	0.42	0.35	48	
	Mean	0.37	0.63	0.17	0.39	0.44	100	
33	Vrindvani	0.35	0.65	0.11	0.47	0.42	354	Kumar et al., 2018

S No	Breed/ Population	Allelic Frequency		Genotypic Frequency			Sample No	References
		A1	A2	A1A1	A1A2	A2A2		
34	Hardhenu	0.66	0.34	0.32	0.68	0.00	50	Ramkaran <i>et al.</i> , 2017
35	HF cross	0.64	0.36	0.29	0.71	0.00	14	Kathiravan <i>et al.</i> , 2021
		0.479	0.521	-	-	-	24	Patel <i>et al.</i> , 2020
		0.375	0.625	0.091	0.567	0.342	263	Patel <i>et al.</i> , 2019
		0.50	0.50	0	1.0	0	12	Srinivas <i>et al.</i> , 2019
		0.638	0.362	0.28	0.72	0.00	47	Shende <i>et al.</i> , 2017
		0.294	0.706	0.000	0.588	0.412	17	Ramesha <i>et al.</i> , 2016
	Rathi X HF	0.000	1.000	0.000	0.000	1.000	10	Saran <i>et al.</i> , 2019
	Sahiwal X HF	0.32	0.68	0.00	0.64	0.36	50	Pandey <i>et al.</i> , 2019
	Dangi X HF							
	HF 75 %	0.13	0.86	0.06	0.13	0.81	15	Jawane <i>et al.</i> , 2018
	HF 62.5 %	0.03	0.97	0.000	0.06	0.94	17	
	Overall	0.08	0.92	0.03	0.09	0.88	32	
	Kangayem X HF	0.405	0.595	0.17	0.46	0.37	63	Malamathi <i>et al.</i> , 2014
36	Jersey Cross	0.69	0.31	0.380	0.620	0.000	29	Kathiravan <i>et al.</i> , 2021
37	Breeding Bulls							Sodhi <i>et al.</i> , 2012
	Holstein	0.441	0.559	0.216	0.451	0.333	51	
	Jersey	0.325	0.675	0.025	0.600	0.375	40	
	Crossbred	0.298	0.702	0.101	0.393	0.506	89	
	Mean	0.355	0.645	0.114	0.481	0.405		
Taurine breeds								
38	Holstein Friesian							
		0.500	0.500	0.000	1.000	0.000	2	Kathiravan <i>et al.</i> , 2021
		0.565	0.435				23	Patel <i>et al.</i> , 2020
		0.169	0.831	0.000	0.338	0.662	59	Ramesha <i>et al.</i> , 2016
39	Jersey	0.077						Ramesha <i>et al.</i> , 2016
		0.25	0.75	0.000	0.50	0.50		Kathiravan <i>et al.</i> , 2021
40	Karan Swiss	0.107	0.893	0.00	0.214	0.786	14	Haq <i>et al.</i> , 2012

In line with data generated at ICAR-NBAGR, other researchers have also reported the complete absence of A1 allele in Kangayam (Malarmathi *et al.*, 2014, Kathiravan *et al.*, 2021), Amritmahal (Inamdar *et al.*, 2019); Gir (Paradkar *et al.*, 2021); Malvi & Nimari (Pandey *et al.*, 2021) cattle; and predominance (≥ 0.90) of the A2 allele in Indian native cattle breeds including Kankrej-0.97; Malnad Gidda-0.98; Ongole-0.94; Umblachery-0.98; Kasargod-0.958; Bargur-0.937, Umblachery-0.98 and Ladakhi-0.90 (Table 1). The variations in the frequency of A2 allele was mainly due to the small sample size or collection of samples from the organized herds/farms, for instance frequency of A1 allele in Sahiwal cattle maintained in organized farm was 0.15 (Pandey *et al.*, 2021). Complete absence of A1 allele has also been reported for Deoni, Khillar (Ramesha *et al.*, 2016) and

Dangi (Jawane *et al.*, 2018) cattle. Overall, the findings indicate the preponderance of A2 β -casein variant and also pointed towards the indicine origin of A2 allele.

Status of β -casein Variants in Crossbred Cattle

India continues to be the largest milk producing country in the world. Population statistics indicate approximately 51% of the Indian cattle population consist of registered/recognized indicus breeds, crossbred cattle constitute only 13%, while others are nondescript cattle. However, in terms of milk production, crossbred cattle and Indicus breeds contribute 24.4 and 20.7 percent respectively to the national milk pool. Considering the contribution of crossbred cattle towards milk produced, it is important to genotype the crossbred population in the country for the status of A1/A2 allele of beta casein. Karan Fries, one

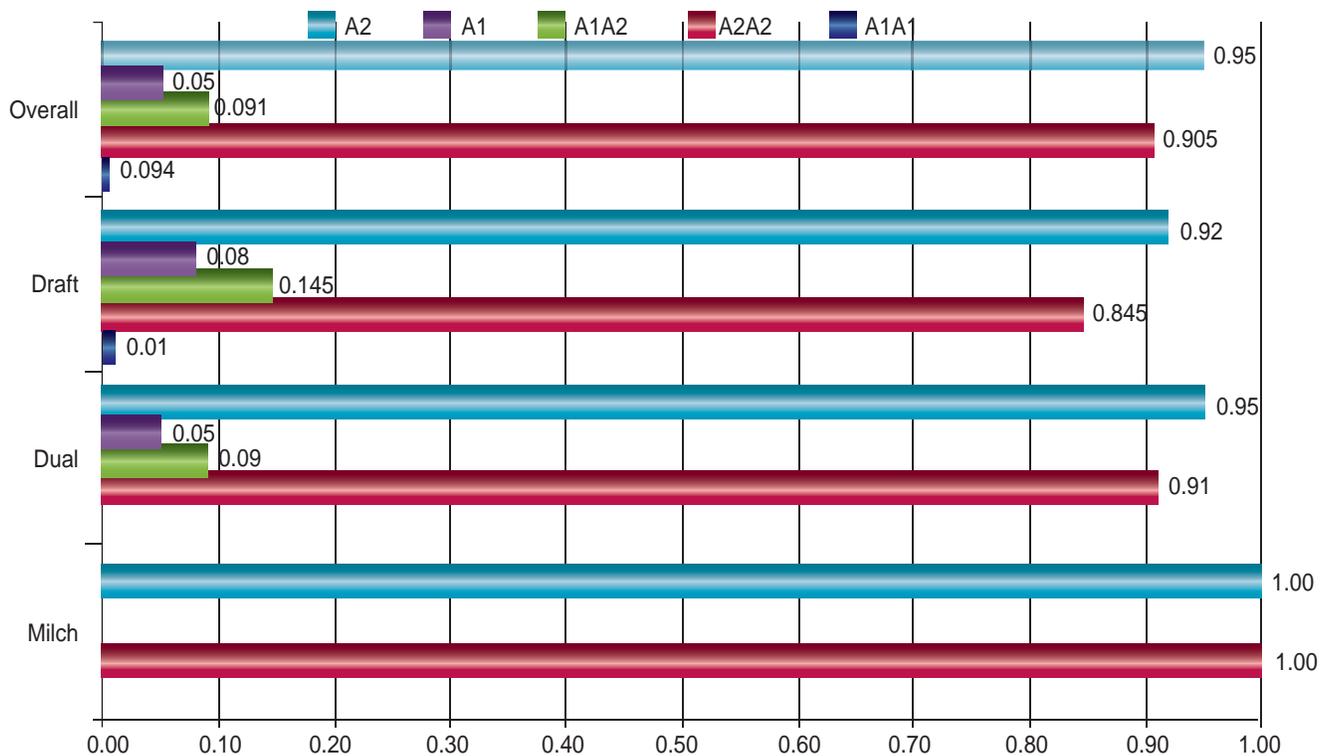


Fig. 3. Overall and utility wise status of β -casein A1/A2 alleles and genotypes in Indian cattle

of the crossbred populations developed using Holstein Friesian (exotic breed) and Tharparkar (indigenous breed) with the aim to enhance milk production coupled with high disease resistance and adaptability to local conditions; Frieswal (cross of Holstein Friesian and Sahiwal); Hardhenu and other crossbred cattle available across the country have been genotyped by different researchers (Table 1). Genotyping of 686 crossbred animals (Karan Fries and Frieswal) by Mukesh *et al.* (2022) revealed the predominance of A2 allele with a frequency of 0.71. Though the observed frequency of A1 allele (0.29) was much higher compared to Indian native cattle, the contribution towards A1 allele was majorly from the heterozygous A1A2 genotype rather than homozygous A1A1. Frequencies of the three genotypes A1A1, A1A2 and A2A2 was 0.09, 0.40 and 0.51 respectively. Similar allelic and genotypic profile (A1-0.32; A2-0.67; A1A1-0.12; A1A2-0.40; A2A2-0.48) with high frequency of A1A2 genotype was observed by Ganguly *et al.* (2019) while genotyping 124 Frieswal heifers. Frequency for A2 allele in HF crossbred cow was observed to be 0.595 (Malarmathi *et al.*, 2014) while in Vrindavani (crossbred cattle) frequency of 0.65 has been recorded for A2 alleles. Most of the studies pointed

towards higher frequency of A2 allele even in Indian crossbred populations except for HF crossbred (A2-0.31) as well as Jersey crossbred (A2-0.36) in Tamil Nadu (Kathiravan *et al.*, 2021), Hardhenu crossbred cattle of Haryana region (0.34: Ramkaran *et al.*, 2017), and HF crossbred of Maharashtra (0.36: Shende *et al.*, 2017).

Status of A1/A2 β -casein Variants in Breeding Bulls

In India, Holstein Friesian (HF) and Jersey cattle have been extensively used since 1960 for crossbreeding and genetic improvement programme. Considering the widespread use of taurine germplasm in our country's cross-breeding program and fact that these cattle could be the potential source for undesirable A1 allele, it is essential to ensure the status of A1/A2 alleles in breeding bulls being used at different AI centres. Frequency profiling of A1/A allele and corresponding genotypes in crossbred, taurine (Holstein Friesian and Jersey) and Indicus bulls being used in different AI centres was reported by Sodhi *et al.* (2012). As per the report, frequency of A2 allele was 0.88 in Indicus bulls, 0.702 in cross-bred bulls, while in Jersey and Holstein Friesian bulls, it was 0.675 and 0.559 respectively. Among the frequencies for three genotypes across taurine and crossbred bulls, least observed frequency was of

homozygous A1A1 genotype (0.114) followed by A2A2 (0.405) while maximum frequency of 0.481 was observed for heterozygous A1A2 genotype. Congruently, the mean frequencies of A1 and A2 alleles was 0.355 and 0.645 respectively (Fig. 3). Similar values of highest mean frequency of A1A2 genotype (0.475) and least mean frequency for A1A1 genotype (0.095) for semen samples from different breeding bulls have also been reported by Mukesh *et al.* (2022). Across different categories of breeding bulls, the frequency of A1A2 genotype was higher in Jersey (0.64) and HF bulls (0.449) in comparison to crossbred (0.39) and indigenous bulls (0.24) while trend of frequency of A2 allele was highest in indicus (0.88) followed by crossbred (0.70), Jersey (0.64) and HF bulls (0.56).

Kathiravan *et al.* (2021) also reported higher percentage of heterozygous genotypes for Jersey (0.50), HF (1.0), Jersey crossbred (0.62) and HF crossbred (0.71) bulls. Though, the data indicated predominance of the

desirable A2 allele across all studied breeding bulls, still there is a need for careful screening of sire lines being used in the breeding programmes as 65-70% of exotic (JSC and HF) or crossbred bulls being currently used in AI program in India are source of A1 allele.

The presence of large crossbred cattle populations and higher proportions of the A1 allele in exotic animals necessitates careful screening of animals and fine-tuning of existing breeding programmes. Such an approach would be an effective measure to prevent the dissemination of the undesirable A1 allele in our existing A2-predominant indigenous cattle populations. Initially, information regarding the screening of bulls for the A1A2 genotype in government semen stations was not available. But in recent years, many of the AI centres have availed the genotype testing facility at ICAR-NBAGR and got the bulls (semen straws) certified as A2 bulls. Specific herds can be converted to A2 type by selective breeding within 4 years using

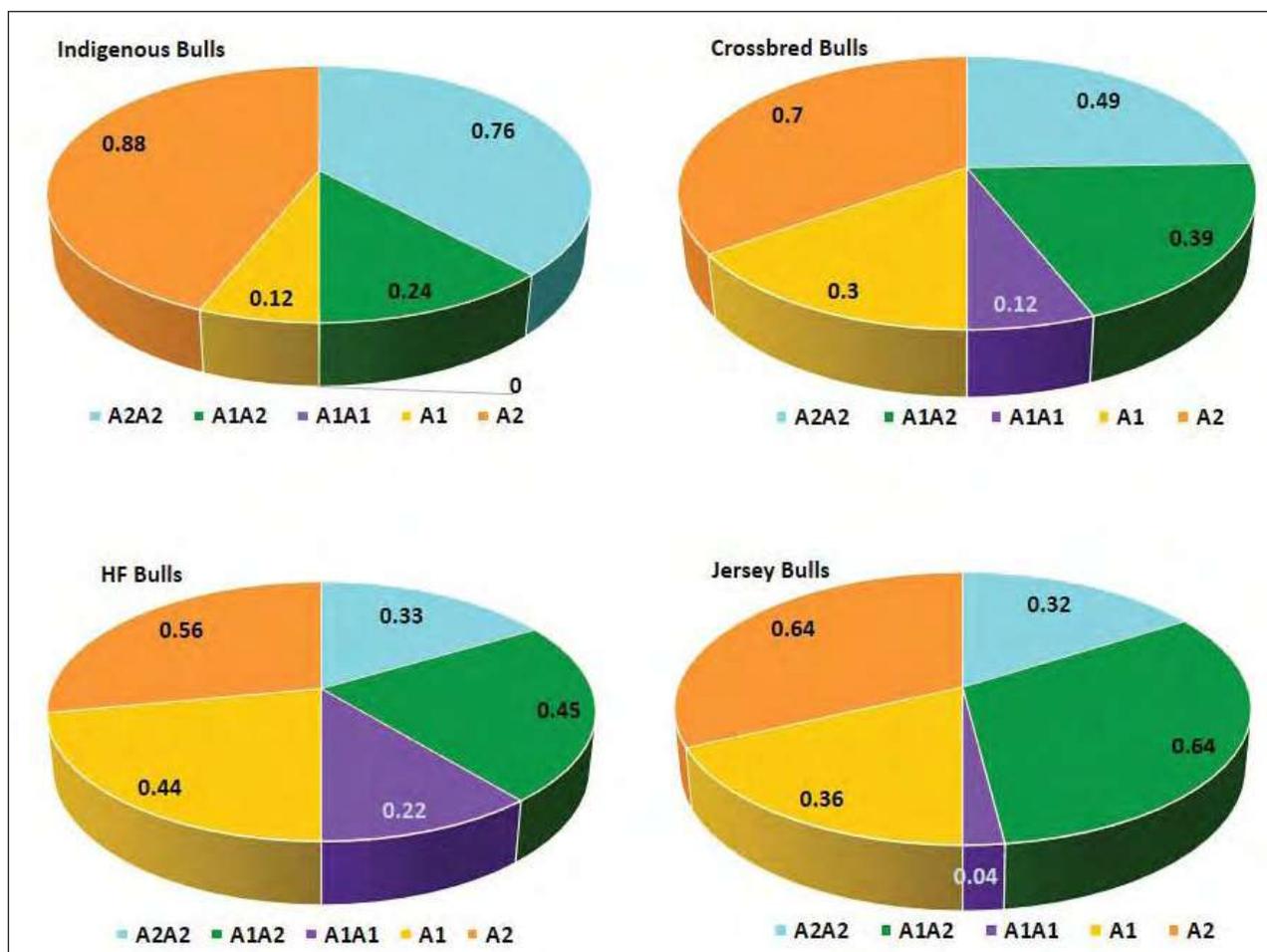


Fig. 4. Overall Status of A1/A2 variants in breeding bulls

intensive methods of animal selection that incorporate the use of certified A2 semen to eliminate all A1 beta-casein from the milk (Pal *et al.*, 2015).

Conclusion

The A1/A2 hypothesis is both intriguing and potentially very important for public health, if it is proved correct. The BCM-7 released from A1 type has been the central theme for hypothesis, whereas the BCM-9 released from A2 type milk need be investigated for the claimed health ailments. The epidemiological data and animal trials point towards potential health hazards of consuming A1 type milk or derived BCM-7 while A2 milk is considered safe for drinking. Work is in progress in different labs across globe to authenticate the fact. Till that time, it is essentially important to monitoring the status of A1/A2 alleles in dairy animals as a precautionary measure. The studies conducted on Indian native cattle strongly demonstrate that Indian cattle naturally harbor the preferred A2 allele and thus are a good resource for A2 milk. Further, though, the frequency of A1 allele is high in crossbred cattle and breeding bulls, its A1A2 genotype mainly contributing for this high A1 allele frequency. The overall frequency of homozygous A1A1 genotype is very low across all the categories of animals that is Indian breeds, taurine or crossbred populations, as well as breeding bulls. Hence, allelic profiling for A1/A2 beta casein in breeding bulls and bringing in changes in the breeding program accordingly can very easily help to drift the herds towards A2 and minimize the risk of disseminating the A1 allele in Indian cattle.

Suggested Readings

- Brooke-Taylor S, K Dwyer, K Woodford and N Kost (2017) Systematic review of the gastrointestinal effects of A1 compared with A2 β -Casein. *Adv Nutr Bethesda Md.* **8**: 739–748. doi: 10.3945/an.116.013953.
- De Noni I (2008) Release of β -casomorphins 5 and 7 during simulated gastro-intestinal digestion of bovine β -casein variants and milk-based infant formulas. *Food Chem.* **110**: 897–903. doi: 10.1016/j.foodchem.2008.02.077.
- Kaminski S, A Cieslinska and E Kostyra (2007) Polymorphism of bovine β -casein and its potential effect on human health. *J. Appl. Genet.* **48**: 189–198. doi: 10.1007/BF03195213.
- Kuellenberg de Gaudry D, S Lohner, K Bischoff, C Schmucker, S Hoerrlein, C Roeger, L Schwingshackl and JJ Meerpohl (2022) A1- and A2 beta-casein on health-related outcomes: a scoping review of animal studies. *Eur J Nutr.* **61**(1): 1-21. doi: 10.1007/s00394-021-02551-x. Epub 2021 Jun 1. PMID: 34075432; PMCID: PMC8783860.
- Kullenberg de Gaudry D, S Lohner, C Schmucker *et al.* (2019) Milk A1 beta-casein and health-related outcomes in humans: a systematic review. *Nutr Rev.* doi: 10.1093/nutrit/nuy063.
- Mencarini IR, KB Woodford and KM Old (2013) Comparing herd selection strategies for A2 beta-casein. *Proc. N. Z. Soc. Anim. Prod.* **73**: 149–154.
- Mishra BP, M Mukesh, B Prakash, M Sodhi, R Kapila, A Kishore, RS Kataria, BK Joshi, V Bhasin, TJ Rasool and KM Bujarbaruah (2009) Status of milk protein, b-casein variants among Indian milch animals. *Indian J Anim Sci* **79**: 722–725
- Mukesh M, S Swami, G Bhakhri, Vipul Chaudhary, V Sharma, N Goyal, P Vivek, V Dalal, AK Mohanty, RS Kataria, P Kumari, SK Niranjana and M Sodhi (2022) Demographic pattern of A1/A2 beta casein variants indicates conservation of A2 type haplotype across native cattle breeds (*Bos indicus*) of India. *Biotech* **12**(3);, 167. <https://doi.org/10.1007/s13205-022-03232-0>
- Ng-Kwai-Hang KF and F Grosclaude (2002) Genetic polymorphism of milk proteins. In: Fox PF, PLH McSweeney (eds). *Advanced Dairy Chemistry: Volume 1: Proteins, Parts A & B.* Kluwer Academic/Plenum Publishers; New York, NY, USA. pp. 739–816.
- Pal S, K Woodford, S Kukuljan and S Ho. Milk Intolerance, Beta-Casein and Lactose. *Nutrients.* 2015 Aug 31; **7**(9): 7285-97. doi: 10.3390/nu7095339.
- Phelan, Martha *et al.* (2009) “Casein-derived bioactive peptides: biological effects, industrial uses, safety aspects and regulatory status.” *International Dairy Journal* **19**: 643-654.
- Sodhi M, M Mukesh, BP Mishra, A Kishore, B Prakash, R Kapil, K Khate, RS Kataria and BK Joshi (2012b) Screening of taurine and crossbred breeding bulls for A1/A2 variants of β -casein gene. *Indian J Anim Sci* **82**: 2–9
- Sodhi M, M Mukesh, V Sharma, RS Kataria and R Sobti (2022) Harnessing potential of A2 milk in India: an overview. Book chapter in *Advances in Animal Experimentation and Modeling* DOI:10.1016/b978-0-323-90583-1.00016-7. Corpus ID: 245063397
- Yadav S, NDS Yadav, A Gheware *et al.* (2020) Oral Feeding of Cow Milk Containing A1 Variant of β Casein Induces Pulmonary Inflammation in Male Balb/c Mice. *Sci Rep* **10**: 8053. <https://doi.org/10.1038/s41598-020-64997-z>

Breed Registration and the Gazette Notification: A Unique National Framework for Protecting Native Animal Germplasm

SK Niranjan*, RK Pundir and BP Mishra

ICAR-National Bureau of Animal Genetic Resources, Karnal-132001, Haryana, India

The FAO's Global Plan of Action for AnGR (Interlaken Declaration), 2007 is global framework for the protection of Animal Genetic Resources (AnGR) diversity, which laid down four strategic priority areas including characterization, inventory of native AnGR for adoption by all countries. India possesses a wide range of AnGR diversity, distributed over diverse geographical and ecological regions, and bears the proactive responsibility to document its own native AnGR on priority. Registration of animal breeds is a step towards inventoring and documenting precious resources at national level. In India, registration has been initiated by the Indian Council of Agricultural Research (ICAR) in the year 2008. Further to provide statutory recognition to the breeds, the registered breeds are the Gazette notified by the Govt. of India. At present, the number of registered and notified breeds has reached 202 belonging to 13 farm animal species in the country. Registration of animal breeds has shown a greater impact on socio-economic arena, including initiation of breed-based livestock census to formulating breeding policies and development programs for registered breeds in the country. The national framework for registration of native AnGR is unique in the world and may be a model to other countries, specifically African and South Asian countries for claiming sovereignty over and protecting their own native germplasm.

Introduction

Protection of biodiversity has widely been perceived as global necessity. Year 1992 may be a benchmark timepoint, when The Convention on Biological Diversity (CBD) was held during Earth Summit held at Rio de Janeiro, envisaging protection, preservation and sustainable utilization of biodiversity. It necessitated to protect all bioresources including Animal Genetic Resources (AnGR) through taking appropriate measures and developing policies in that direction by all member-countries. India as a signatory of the CBD, also promulgated legislation for protection of biodiversity in form of The Biological Diversity Act in 2002. Protection of AnGR *per se* got the momentum after developing The Global Plan of Action for AnGR by the Food & Agriculture Organization (FAO) as Interlaken Declaration in 2007 and subsequently CBD-Nagoya protocol for Access & Benefit Sharing and Indigenous Knowledge in 2010. Further, various targets specially CBD's Aichi Biodiversity Targets (2010) and the UN's Sustainable Development Goals (SDGs) have further crusaded the global efforts for the protection of AnGR diversity. SDG 2 (Zero Hunger) envisioned the proper management of all kinds of genetic resources to promote sustainable agriculture and achieving food security and oriented towards the preservation of farm

animal biodiversity. 'Delhi Declaration' in 2016 is another milestone framework for protection of valuable germplasm globally.

India possesses a huge AnGR diversity distributed over a large range of geographical, ecological and climatic regions. A range of farm animal species like zebu cattle, riverine buffalo, swamp buffalo, sheep, goat, pig, horse, donkey, camel, yak mithun, chicken, duck and geese are natively inhabited in the region for centuries. In any given geographical regions in the world, none of country is that much rich in farm animal diversity. These species are further diversified in form of unique populations called breeds specific utility and unique attributes of adaptation. The animal breeds have been evolved through of various evolutionary processes including man-made selection for different needs. In fact, natural adaptation to specific climate and habitat has been confounded with human derived artificial selection and breeding in breed formation. It is widely assumed that a large number of the livestock and poultry breeds have been developed during last few centuries in the country. Further recent introgression of exotic grmplasm also generated new populations in form of synthetic breeds and variants with improved production traits. Today, treasure in form of hundreds of animal breeds - adapted to local climate and production system and highly resilient and resulting into

*Author for Correspondence: Email-saket.niranjan@icar.gov.in

a unique gene pool of allelic combinations - has been accumulated over the centuries. Creating inventory of animal breeds, with their unique attributes is important to recognise their characteristics and uniqueness. In fact, the characterization and inventory have been recognized as the foundation of proper management of AnGR.

Registration of Animal Breeds

‘Animal breed’ is considered as a referral point for the farm animal diversity, and as per FAO, it can be defined as “sub-specific group of domestic livestock with definable and identifiable external characteristics that enable it to be separated by visual appraisal from other similarly defined groups within same species. Moreover, breed is a group for which geographical and/or cultural separation from phenotypically similar groups has led to acceptance of its separate identity. Each breed serves a specific purpose to the food and agriculture for the mankind within a well-defined geographical region.

Registration of native breeds of livestock and poultry has been envisioned to protect and check the bio piracy of indigenous AnGR and our country has developed the mechanism for recognising breeds with known characteristics in form of authentic national documentation system. Indian Council of Agricultural Research (ICAR) initiated “Registration of Animal Germplasm” specifically indigenous livestock and poultry breeds in the country in the year 2007. In the year, 2008, ICAR-National Bureau of Animal Genetic Resources (NBAGR), Karnal was given the temporary authority for the registration of germplasm related to livestock and poultry in the country. Subsequently, in 2008, ICAR constituted a Breed Registration Committee (BRC) under the chairmanship of Deputy Director General (Animal Science), ICAR for the registration of new breeds. This mechanism is the sole recognised process for registration of “Animal Genetic Resources” material at national level.

Breed Registration Process

The registration of Indian livestock and poultry germplasm revolves around the concept of a breed. Breeds of domesticated animals, which are unique, stable and uniform, and have potential attributes of academic, scientific, or commercial value can be registered. Any livestock population which has been characterized must be documented and inventoried, if having the breed characters. First, all of the information recorded during phenotypic characterization, should be

formatted in a shape of breed descriptor. Such a physical characterization along with management practices can be published in different scientific journals. After phenotypic characterization, it should be clear that if the population is found distinct, then only it should to be registered as breed.

First, all of the information recorded during phenotypic characterization should be shaped as a breed descriptor. Such a physical characterization along with management practices can be published in different scientific journals. The registration involves a process for screening of the applications submitted for registration as per Guidelines developed for this purpose. The application can be submitted by any citizen of India /breed society/NGO/Govt. Agency. The application must be accompanied by a complete description of the breed using standard descriptors. All claims concerning the material submitted for registration should accompany scientific evidence for uniqueness, reproducibility, and value. The population, for consideration of registration should have at least 1000 animals. The breed should complete a minimum of 10 generations. A detailed history of the breed, Difference, distinction, and details that are specific for that breed should also be provided. Representative photographs, a list of the registered animals of the breed, letters explaining certain questions about the breed from at least three different breeders/owners of

Breed registration criteria

- Unique, stable and uniform population with potential attributes of academic, scientific or commercial value
- Population: at least 1000 animals and also maintained in field conditions for minimum of 10 generations
- Breed application, with standard descriptors
- Accompanying with scientific evidence
- Signed by the applicant
- countersigned by Director, AHD of concerned state with rubber seal
- A detailed history of the breed
- List the difference, comparison to other breeds
- Photographs of the breed
- Letters from at least three different breeders/ owners of the breed.

the breed should also be submitted. After registration, NBAGR provides the unique Accession number to each breed after registration. The newly registered breeds are also notified through Official Gazette published by the Government of India. Detailed guidelines, descriptors and application form for registration of new breeds can be accessed at www.nbagr.res.in/guidelines.html.

Present Status of Breed Registration

Initially, the known breeds of livestock and poultry were registered as extant breeds, and simultaneously process for registration of new breeds was initiated. First time in the year 2008, all 129 extant breeds of livestock and poultry were registered by the ICAR-NBAGR. Further 73 new breeds were added and by 2021, the number reached 202; which included 177 breeds of livestock and 22 of poultry and 3 of dog. The breeds include 50 of cattle, 19 of buffalo, 34 of goat, 44 of sheep, 7 of horses and ponies, 9 of camel, 10 of pig, 3 of donkey and one of yak in livestock and 19 of chicken, one of geese and two of duck in poultry and three for dog species. During the last 10 years, more than 100 new populations were identified across the country and 73 breeds of different farm animal species were registered.

The Gazette Notification of Registered Breeds

After realizing the need to protect the valuable agricultural genetic resource biodiversity specific to indigenous farm animal germplasm in the country and claiming its sovereignty over the germplasm, the Gazette notification for the livestock and poultry breeds was initiated by the Government of India in October, 2019. To provide legal safeguards for germplasm protection, notification of indigenous breeds being registered by ICAR has started in the year 2019 through publishing the Official Gazette by the Government of India. All registered breeds upto that year (total 184 breeds) were first time notified by the Government of India to provide statutory recognition of and claiming sovereignty over the native germplasm [Gazette Notification: Ministry of Agriculture and Farmers' Welfare, No. 3364 (S.O. 3699(E)) (October 14, 2019)] and further all newly registered breeds (18 breeds) in subsequent years were notified through three more Gazette notifications. These breeds got the statutory recognition; and shall be the notified breeds for the whole of India for purposes of animal husbandry, production, breeding, conservation, utilization, consumption and trade from the date of publication of the notification in the official Gazette of Govt. of India. The notified breeds of

the specified States received the statutory recognition; and were recognised as notified breeds for the whole of India for keeping and rearing for various purposes as mentioned in the notification.

Impact of Breed Registration

Registration of breeds of native livestock and poultry has a broader impact through modulating various policies and programs and affecting all stakeholders of the livestock sector in the country. Most importantly, it enabled to conduct breed wise livestock census by the Govt. of India which could ensure the precise population of each of the breed in the country. This has come to be much useful for suitable policy formulation for their conservation and development if the breed is endangered. A total of 73 new breeds were added after the initiation of the registration process, which has put about 25 million livestock and poultry into the descript category in the country. In the 19th Livestock Census (2012), a total of 143 breeds were included in breed wise survey. Further, 20th Livestock Census was carried out on 184 registered breeds in the country. Based on breed wise census, it is imperative to define the risk status of the breeds and as per preliminary data about 24 breeds are a risk in the country. Breeding policies have been formulated for many newly registered breeds in many states with recent example of Haryana, Odisha, Nagaland, Ladakh (UT). At national level, National Kamdhenu Breeding Centre, An initiative by Govt. of India has covered all NBAGR registered breeds of cattle (41) and buffalo (13) for conservation, promotion, and development. Farmers are best benefitted by the registration process. After breed recognition, superior quality germplasm of the breed becomes available to the livestock owners. Various governmental facilities for animal-based farming, small to large /commercial scale become available to the farmers when the animal farming is based on the registered breeds. Breed societies have been developed for various native livestock breeds in their native tract by the local livestock keepers, which ensured improvement of germplasm, strengthening of the production system and marketing of the products and benefit-sharing among the society. As an example, benefits of Ongole embryo germplasm export were disseminated among the society of the breed.

The Gazette Notification also provided legal support to germplasm protection of the registered breeds and for developing mechanisms for sharing benefits among the

animal keepers. In an era of globalization, there remains a constant threat of germplasm piracy as well as forged patenting and IP rights to the inherent characteristics. Registration and notification would keep the proper check for such spurious activities related to precious native germplasm. Notification of breeding areas of registered breeds also enables to develop suitable breeding strategies for the genetic improvement and development programme of low productive native animals. As defined in Livestock Breeding Policy, such breeds can be improved by their own germplasm through selective breeding and no other germplasm can be introduced in the notified breeding tract of registered breeds. In this way, registration of native breeds would be helpful in not only the protection of our germplasm but also their improvement as well as avoiding genetic dilution. Breeding policies for many newly registered breeds are being developed by the respective state departments.

Nevertheless, the framework of breed registration and the gazette notification has certainly broadened its impact through modulating various policies and programmes. Along with preserving the biodiversity as mandated, the stakeholders specially Farmers would be the best benefitted. After breed designation, superior

quality germplasm of the breed become available to the livestock owners. This *sui generis* system would also provide the legal support to Intellectual Property Rights of the registered breeds and for developing mechanism for sharing benefits among the animal keepers.

Future Action Points

- New breeds of indigenous livestock and poultry with unique characters may be explored and registered specifically from remote and inaccessible regions of the country.
- Synthetic populations of livestock species evolved by different organizations in India may also be registered.
- A legislation may be promulgated for animal breed protection and farmers rights in this direction.

References

- FAO (2007) Global plan of action and the Interlaken declaration commission on genetic resources for food and agriculture food and agriculture organization of the United Nations Rome.
- ICAR-NBAGR (2016). Guidelines for Management of Animal Genetic Resources of India, National Bureau of Animal Genetic Resources (ICAR), Karnal, http://www.icar.nbagr.gov.in/guidelines_management_AnGR.pdf.

ARTICLES ON
FISH GENETIC RESOURCES



ARTICLES ON FISH GENETIC RESOURCES

Title	Page No.
FAO Resources for Strategic Planning	285
Informatics as Future for Fish Genetic Resources Management	289
The New Finfish Genetic Resource Discoveries from Indian Waters – Need for Enhancing Taxonomy Capacity	293
A Review on Ichthyofaunal Diversity and Distribution of North-East India	298
Diversity of Fish Genetic Resources below the Species Level, Characterization and Applications in Resource Management	301
Genomic Research in Fishery Sector: Current Status and Future Prospects	305
Reservoirs Fisheries Development and its Trade-offs with Conservation of Natural Fish Genetic Resources	308
Geo-Spatial Tools for Science-Based Management of Inland Aquatic Habitats and Conservation of Fish Genetic Resources	312
Mariculture Development in India: Status and Way Forward	317
Fishing Technologies for Conservation of Marine Fish Genetic Resources	322
Boosting Widespread Adoption of Sustainable Agriculture – New Metrics and the Role of Science	325

FAO Resources for Strategic Planning

Graham Mair* and Daniela Lucente

Food and Agriculture Organization of the United Nations, Viale delle Terme Di Caracalla, Rome, Italy

Effective management of aquatic biodiversity used in aquaculture will play a critical role in the future development and sustainability of the sector. In this context, a global assessment, conducted by FAO, on the status of management of aquatic genetic resources for food and agriculture (AqGR) identified a number of significant challenges and needs. In response to this report, FAO and partners have been developing a number of key resources to facilitate countries to better manage their AqGR. This paper describes these resources, which include, *inter alia*: a framework of essential criteria; AquaGRIS – a global information system; and a *Global Plan of Action for the Conservation, Sustainable Use and Development of Aquatic Genetic Resources for Food and Agriculture*. These resources can be applied at a range of different levels by diverse aquaculture stakeholders such as policy makers, resource managers, producers and researchers, and can be integrated and used together in the development of strategic approaches for the management of aquatic biodiversity at national, regional and even global scales.

Introduction

It is well accepted that biodiversity underpins sustainable growth in food production and must be effectively managed for future food security. This is every bit as important for aquatic as it is for terrestrial biodiversity. FAO is committed to support countries to move towards more responsible management of biodiversity and to collaborate with partners (academia, research institutes, governmental and non-governmental organizations) to achieve this goal.

Aquatic genetic resources or food and agriculture (AqGR), are represented by DNA, genes, chromosomes, tissues, gametes, embryos and other early life history stages, individuals, strains, stocks and communities of organisms of actual or potential value for food and agriculture. *The State of the World's Aquatic Genetic Resources for food and agriculture* (FAO, 2019a), the first global report on the status of AqGR, illustrated some key points of differentiation for these resources compared to terrestrial genetic resources. One of the major points is that most aquaculture species have been domesticated for a much shorter period than terrestrial genetic resources and thus retain higher levels of genetic diversity than their terrestrial counterparts. With our present day understanding of the importance of genetic

diversity and the plethora of tools available to us to characterize and manage such diversity, we have a window of opportunity to act to secure the future of AqGR, but real action is required in the near future to ensure we do not squander this opportunity. Paramount among the identified needs and challenges are:

1. **The lack of information on the status of AqGR:** Most countries know which species are cultured but generally there is little information available on genetic resources below the level of species (i.e. farmed types such as strains or varieties).¹ Lack of reliable information constrains policy makers and resource managers to develop effective management strategies for AqGR and often means that producers have confusing and unreliable information about the diversity and characteristics of the genetic resources available for culture.
2. **Some genetic resources are at risk but conservation needs are often undetermined:** Without information on both cultured species and their wild relatives² there is no information on the status of threat to these genetic resources. Creating a knowledge base on cultured and wild relative resources enables identification of resources that may need to be conserved locally, either *in situ* and/or *ex situ*.

*Author for Correspondence: Email-graham.mair@fao.org

1. "Farmed types" refers to a farmed aquatic organism that could be a strain, variety, hybrid, triploid, monosex group, or other genetically altered form or wild type and is considered a generic descriptor of AqGR below the level of the species (Mair and Lucente, 2020).
2. Wild relative refers to an organism of the same species as a farmed organism (conspecific) found and established in the wild, i.e. not in aquaculture facilities.

Even species that are cultured extensively such as common carp and *Pangasius catfish* can be under threat in the wild.

3. **Unsustainable use of AqGR:** The majority of cultured species have no systematic genetic improvement programme in place. Among the majority of farmed types in aquaculture that are not the result of planned selective breeding, poor genetic management is commonplace. Deficiencies in genetic management include inadequate diversity of founder stocks, maintenance of low effective population size, breeding of close relatives, and uncontrolled hybridisation. Such practices can lead to loss of genetic variation, genetic drift, inbreeding and unplanned hybrid introgression which in turn have negative consequences for aquaculture such as loss of adaptive potential, declining performance due to inbreeding depression, reduced response to selection (when applied) and loss of specific characteristics of species. These effects are cumulative and, once in place, are difficult to reverse.
4. **The slow uptake of genetic improvement:** It is commonly understood that selective breeding should be at the core of most attempts to develop farmed types for aquaculture and the benefits and economic returns from selection programmes are well proven. However, the uptake of selective breeding is slow and it is considered likely that no more than 15 percent of global aquaculture production results from well-managed breeding programmes. Selective breeding can improve commercially important traits for aquatic species by 10-13 percent per generation and thus there is a very significant opportunity to generate cost-effective increases in aquaculture efficiency through accelerated application of appropriate genetic improvement. Therefore, it is important to identify those aquaculture species and sectors that would benefit most from the application of genetic improvement and to identify the most appropriate method of genetic improvement based on the analysis of their likely risks and benefits.

A Framework of Essential Criteria

Following stakeholder consultation and validation, FAO has developed a framework of minimum requirements in the management of AqGR (Framework) that can assist countries to establish conditions necessary to sustainably manage their AqGR (FAO, 2018). It represents a needs

assessment document that calls for national dialogue to develop an implementation strategy, and a review or revision of national policy and practice, and furthermore to identify national gaps and needs for a sustainable management of the AqGR. The Framework provides guidance on a set of essential requirements that would need to be met to facilitate effective management of AqGR and these fall under five distinct components: information and databases; governance, policy and planning; infrastructure and equipment; capacity building and training; and enabling the private sector. In any specific country, stakeholders can map current policies, practices, infrastructure and resources onto the elements of the Framework and assess which requirements would need to be created or better developed in a particular area in order to enhance national capacity for effective management of AqGR.

AquaGRIS – A Global Information System for AqGR

The global assessment (FAO, 2019a) identified the lack of information on AqGR as a principal constraint to building effective management strategies. FAO is addressing this through the development of the Aquatic Genetic Resources Information System – AquaGRIS. The database of AquaGRIS will contain data on all cultured species, collected primarily through a purpose designed questionnaire and related to primary and secondary farmed types (Mair and Lucente, 2020). It also draws on some data from the country reports prepared for the global assessment mentioned earlier and production data from the FAO's aquaculture and fisheries database available through *FishstatJ* (FAO, 2019b). A range of different reports can be generated for different aspects of AqGR management, including species and country fact sheets.

AquaGRIS has application for a range of different stakeholders. Policy makers and resource managers can use the information as a basis for policy and strategy development and to monitor their implementation. Producers can find out what resources are available to them, their properties and even where to find them. Academics and breeders can use AquaGRIS to inform and prioritize their research and development and to communicate information about newly developed farmed types entering commercial production.

The current version of AquaGRIS is a prototype and, at the time of writing, contains detailed and searchable datasets on a few species such as common carp, Atlantic salmon, whiteleg shrimp, milkfish, *Kappaphycus* and *Artemia*, covering AqGR in over 40 countries. Work is on-going to develop a fully functional version of AquaGRIS that will include an improved user interface for data entry and validation expanded to collect data related to wild stocks of cultured species and also data that can be used to generate indicators for progress against the Global Plan of Action (see below). In the longer term it is hoped that AquaGRIS can be used to generate indicators of progress against SDG targets (e.g. SDG 2.5) and the targets and indicators of the CBD's Post-2020 Biodiversity Framework.

A Global Plan of Action

The global assessment identified over 40 needs and challenges. At the request of the Commission on Genetic Resources for Food and Agriculture, and following regional consultations, FAO developed a comprehensive Global Plan of Action (FAO, 2022) in response to these needs and challenges. The Global Plan of Action (GPA) is an internationally agreed framework aiming to enhance the management of AqGR to optimize their contribution to the promotion of food security and sustainable development, and alleviation of poverty. The GPA is a rolling plan, with a time horizon of ten years and is voluntary and non-binding. The GPA covers 21 strategic priorities under four priority areas and includes nearly 100 possible actions. It is understood that the priorities and important actions will differ from country to country depending on the status of countries' AqGR. The GPA can thus be considered as a menu of priorities and actions from which individual countries can identify key steps to take in relation to the status of their own genetic resources and the priorities for furthering conservation, sustainable use and development. The four priority areas which are tailored to the characteristics of AqGR are: inventory, characterization and monitoring of AqGR; conservation and sustainable use of AqGR; development of AqGR for aquaculture; and policies, institutions and capacity building.

Other Resources under Development

In addition to the above-mentioned core resources, FAO is developing other initiatives to support communication and awareness raising on the importance of effective

management of AqGR. During the implementation of the global assessment and the development of AquaGRIS it was recognized that the lack of harmonization and standardization of the use of terminology to describe AqGR was a significant constraint to raising awareness and understanding of the status of these resources. The effective use of AquaGRIS requires application of standard terminology based on a classification of farmed types that FAO has developed with a group of experts. To support AquaGRIS users and stakeholders in adopting and regularly using this terminology, FAO is also developing a thesaurus of terminology related to genetics in aquaculture that will be made available in future through the AquaGRIS interface and other media.

In response to the identified need for building awareness and capacity in relation to genetic management and improvement in aquaculture, FAO, in collaboration with the Norwegian Institute of Food, Fisheries and Aquaculture Research (Nofima), a leader and global pioneer in the application of genetics in aquaculture, have developed an online training programme on management and development of AqGR. The 18 modules for this basic course, including over 6 hours of video lectures (in short segments averaging 13 minutes) by some world leading experts, is targeting a range of stakeholders from students and industry representatives and resource managers, and cover a range of issues with a focus on genetic management and improvement in aquaculture. The content of the course is finalized at the time of writing and the course will be made available on line following the development of an appropriate delivery platform. FAO is also developing a number of case studies on genetic management in aquaculture and also guidelines for *ex situ in vitro* gene banking in aquaculture and guidelines for genetic management for stocking of captive reared farmed types into the environment (e.g. for conservation and fisheries enhancement).

Conclusions

The resources outlined above can be integrated and used together, through a stakeholder driven strategic planning process, to develop a national plan for advancing the management of AqGR as outlined in the figure below. The Framework can be used to conduct a form of broad audit of national or regional capacity across its five components, and to identify potential gaps that may necessitate capacity building. AquaGRIS can be used to construct an inventory of genetic resources in the



FAO resources for sustainable management of aquatic genetic resources in aquaculture

country that can inform development of priorities within a strategy. Based on the results from the Framework analysis and the national inventory of AqGR, priorities for a country can be identified from the elements of the GPA. Lastly, the other resources can be used to raise awareness of the importance of AqGR and potentially to build capacity in some areas.

The work of FAO in recent years has generated a comprehensive and holistic understanding of the global status of AqGR, identified some important needs and challenges, and generated some useful tools in response to these needs and challenges. However, these resources are yet to be widely adopted and utilized by countries. It is hoped that the growing awareness of the importance of biodiversity will be a catalyst for real actions on the ground in the countries and that countries and regions will begin to realize the present opportunity to secure the future supply of aquatic food through enhanced and effective management of AqGR.

References

- FAO (2018) Aquaculture development. Development of aquatic genetic resources: A framework of essential criteria. FAO Technical Guidelines for Responsible Fisheries 5 Suppl. 9. Rome. 88 pp. www.fao.org/3/ca2296en/ca2296en.pdf
- FAO (2019a) The State of the World's Aquatic Genetic Resources for Food and Agriculture. FAO Commission on Genetic Resources for Food and Agriculture Assessments. Rome. 291 pp. www.fao.org/3/ca5256en/ca5256en.pdf
- FAO (2019b) Fisheries and Aquaculture Department, Statistics and Information Service. FishStatJ: Universal software for fishery statistical time series. Copyright 2019. <https://www.fao.org/fishery/aquagris/home>
- FAO (2022) Global Plan of Action for the Conservation, Sustainable Use and Development of Aquatic Genetic Resources for Food and Agriculture. Commission on Genetic Resources for Food and Agriculture. Rome. <https://doi.org/10.4060/cb9905en>
- Mair GC and D Lucente (2020) What are "Farmed Types" in aquaculture and why do they matter? FAO Aquaculture Newsletter No. 61: 40–42. www.fao.org/3/ca8302en/ca8302en.pdf#page=40

Informatics as Future for Fish Genetic Resources Management

Simon Wilkinson

Network of Aquaculture Centres in Asia-Pacific, Bangkok, Thailand

Informatics is the application of data science to extract and communicate actionable knowledge in the context of a specialised domain. Informatics will play an increasingly important role in management of genetic resources for aquaculture as the industry pursues automation and optimisation of processes in pursuit of intensification. This will likely parallel the adoption of industrial control systems in technology-oriented “smart farms”, which will drive instrumentation of farming systems. Aquaculture has historically under-invested in genetic resource management and all aspects of information technology, including informatics, but both have potential to generate substantial benefits. Data capture and computational resources for informatics are no longer limiting. They are cheap, easy to obtain and manage, due to advances in microelectronics, software virtualisation and cloud computing. Human resources are now the main bottleneck in application of informatics in aquatic genetic resources management. There is a need for the aquaculture sector to invest in developing its own professional capacity in informatics, as it is unlikely to be able to recruit personnel from more industrialised, higher-paying sectors

Introduction

Informatics has its foundations in data science, collecting, organising, curating and analysing information to extract actionable knowledge that can be used to inform decision making and address specific problems (Fridsma 2018). Informatics is also concerned with the design of information systems and tools to interpret, visualise and communicate knowledge from data. Informatics is distinguished from ‘pure’ data science by the requirement to have a degree of specialist knowledge in a domain to understand the issues relevant to that domain and to devise appropriate collection, analyses and applications. Hence there are many specialisations within informatics, such as veterinary informatics, breeding informatics and so on.

We presently stand at the beginning of a revolution in computational informatics. This revolution is being driven by colossal leaps in automated data capture and storage; the availability of massive and affordable cloud-based computational processing on demand; and near-ubiquitous electronic network communications. Collectively, these allow informatics to operate on a scale that was unimaginable, just a few years ago. We have never had so much information. Our problem, now, lies in how to generate knowledge from it. This in turn requires public and private investment in both people and computing infrastructure.

Informatic Opportunities in Fish Genetic Resources Management

Informatics can play a valuable role in nearly every aspect of aquaculture and will increasingly do so as the industry pursues automation and optimisation of processes in pursuit of intensification. In the context of genetic resources management (setting aside farm management), informatics can play a role in many ways.

In aquaculture, non-exhaustive examples include:

- Analysis of molecular genetic sequences, markers, genotypes and phenotypes, production traits, pedigrees and similar information in broodstock holdings that is of interest to farm productivity and/or conservation genetics. In aquaculture, such information can inform selective breeding programmes that improve growth rates or favourable production characteristics of farmed animals by selecting appropriate breeders.
- Quantifying the performance of animals with different genotypes, phenotypes and traits, in the context of particular farm conditions or management regimes. For example, growth and survival at different temperatures, salinities, resistance to specific diseases or performance on certain nutritional regimens.

*Author for Correspondence: Email-simon@enaca.org

With regards to management of wild genetic resources, recording and curating data such as:

- Fisheries catch and effort.
- Distribution and habitat records (particularly for threatened species).
- Environmental monitoring and management data.
- General taxonomy and biology data.

These are a range of very different problems with very different sources of data that are likely to be carried out by informatics specialists with a diverse range of experience, many of whom will use other professional titles.

One area of emerging interest is the application of on-farm informatics. This will likely parallel the adoption of industrial control systems in technology-oriented “smart farms”. While it already occurs to a degree through assessment of common farm parameters such as survival, feed conversion ratio and similar, advances in microelectronics, network communications and sensor technologies will increasingly drive the use of automation and industrial control systems in farm management. The instrumentation associated with these developments will permit real time monitoring and capture of detailed data from the farm environment, facilitating more complex analyses and optimization of farming systems and practices, and their interaction with genetic variation of stock.

Aquatic Genetic Resources Lagging Behind

As a relatively new industry, aquaculture lags well behind terrestrial agriculture in most respects, such as in the adoption of traditional/standard practices such as selective breeding and in more recent innovations such as automation.

In terrestrial agriculture, selective breeding is taken for granted. Everything you eat is an improved variety. Science-based selection has been underway for many decades, and informal selection for desirable characteristics has been underway for centuries or millennia.

More recently, in some industries, such as dairy cattle, commercial genomics services are available to evaluate individual breeders or even embryos against desirable traits including yield, fertility, survival, heat tolerance, feeding efficiency and others, enabling farmers to select high quality genetic material that best suits their business needs and priorities.

By contrast, aquaculture has under-invested in genetic resource management, and very few improved varieties are available in aquaculture. Genetic mismanagement and inbreeding are rampant. In many cases we are farming degraded genetic resources that have lower performance than unimproved, wild-type animals. Wild-sourced seed are still commonly used in some sectors of aquaculture.

Aquaculture has also under-invested in all aspects of information technology, including informatics. The reasons for this are not clear, but as an emerging agricultural sector it is perhaps logical that most effort should be put into solving the biological problems first. Secondly, with little to no automation and the small-scale nature of most farms, the field has attracted few information technology specialists, and is dominated by biology graduates.

But there is some good news. Advances in IT are making informatics more accessible

In the recent past, setting up infrastructure for computational informatics required the purchase or lease of a server, physical or virtual, its co-location in a commercial data centre or on premises with an expensive lease line connection to the internet, and you were limited to whatever hardware you could afford at the time. Data acquisition and entry, particularly from field sources, was largely a manual process.

Recent advances in microelectronics, sensor technologies, network communications and software virtualisation, containerisation and orchestration have radically improved the accessibility of computational informatics by slashing the cost and maintenance requirements, while making the hardware available to anyone via cloud computing.

Networked Sensors and Microcontrollers

From an informatics point of view, the availability of cheap sensors and microcontrollers has opened the possibility of adding industrial control systems to aquaculture production systems. This in turn allows for 24/7 automated capture of data for real-time monitoring or historical analysis of trends.

Common sensors such as temperature, pH, ammonium, turbidity, motion sensors and many more are typically a few dollars (dissolved oxygen is an exception) and can be connected to programmable microcontrollers, which may relay data to a networked collection point, exercise control over hardware such as pumps, motors

and aerators for farm management, or trigger warnings if environmental parameters cross acceptable thresholds.

The recently released Raspberry Pi Pico W microcontroller, for example, costs only \$6, includes WIFI connectivity for easy networking, is programmable in Python or C and has excellent documentation and a community of makers and users from whom advice can be sought.

Cloud-based Server Infrastructure

From a computational informatics perspective, it is no longer necessary to own or maintain server hardware. Cloud-based computing providers allow you to set up, tear down, and change the specifications of servers at a moment's notice.

Opening an account with a cloud-based computing provider just takes a couple of minutes and a credit card. Having done that, you are free to commission *as many servers as you need*, on demand, whether virtualised or physical. You can choose the location of servers from a list of data centres around the globe. You can specify what operating systems you want them to run, the number of CPUs and how much RAM you need for the task at hand. Dedicated GPUs are available to support applications that require high levels of parallel processing.

From specifying requirements to provisioning a new server to spinning it up with a first-class internet connection takes a few minutes. You will typically be billed by the hour at a rate that is astonishingly cheap and would be impossible to achieve purchasing your own equipment, from as little as \$5 per month. Once your task is accomplished, you can decommission the servers and walk away without paying anything more. Or come back later and re-commission servers when you have need. Automatic provisioning and scaling of servers in response to load is possible with many providers.

These are the basics, most cloud-based computing providers offer a full range of services that are beyond the scope of this discussion, but for practical purposes if you are likely to need it, they are likely to have it.

Containerisation

Server resources can be efficiently partitioned still further through containerisation technologies, such as Docker, which enable the creation of very light-weight virtual machines or “containers”. The mechanism is

beyond the scope of this paper, but essentially a Docker container makes use of the Linux kernel already running on the host machine, rather than requiring a full copy of all the files and a separate running instance, so it is extraordinarily efficient. A container's processes are assigned to a separate namespace, so they are logically separated from the host and operate as an independent, virtualised machine.

The strength of Docker and a related tool, Compose, is that an entire network of servers and services can be built from a configuration file, in which you specify the software, data volumes, and network topology you require. Docker and Compose will retrieve the relevant server images, set up containers, virtual network connections and persistent data storage volumes with a single command. Since data volumes are persistent, containers (servers) can be shut down, discarded, rebuilt or swapped out as needed. The container infrastructure is essentially disposable and readily upgradeable, simplifying maintenance greatly.

Orchestration

Having established a fleet of servers and / or containers, maintaining them and enforcing standardised configurations could be difficult. But in a manner similar to Docker as described above, free software tools such as Ansible can be employed to automate and standardise deployments, reducing the task to preparation of a common specification file (‘playbook’ in Ansible terminology), which will implement a prescribed series of actions against designated groups of servers and report their status and any variation from the prescribed configuration.

Such software enables a single administrator to manage potentially hundreds of machines, a task that used to take a significant work force.

We still have a people problem

The above discussion above boils down to this: Data capture and computational resources for informatics are no longer limiting. They are cheap, astonishingly easy to obtain, and much easier to maintain than ever. So why isn't informatics being broadly applied in aquaculture?

The slow uptake of informatics in aquaculture generally, including for management of genetic resources, is primarily due to a lack of personnel with relevant training and expertise. The availability of computer

hardware, software and networks has outstripped the available human resource capacity; it is the people with the knowledge and skills to harness these tools that is lacking.

It is a fact that aquaculture is, relatively speaking, a low paid industry. Most commercial farms simply lack the scale and infrastructure to require and support advanced information technology professionals. Graduates in informatics, particularly those with a genetics background, will certainly look to more technologically advanced industries and the more highly paid jobs available in larger companies for their careers, for the foreseeable future.

We Must Look Forward

What then, to do? In the authors opinion, the foundations of informatics should be taught as a component of aquaculture at tertiary level institutions, building on existing record keeping components. Aquaculture and conservation-oriented research schools should consider establishing related streams in informatics to support the

evolution of aquaculture to a “smart”, technologically based industry, with a view to expanding informatics offerings over time as industry demand grows. It will take time, but there will never be a better time to start than right now.

It is likely that aquaculture will benefit from spill-over of innovation from other sectors and may itself become a source of expertise for aquatic conservation. But to lead the way in solving aquaculture and aquatic conservation problems it would be better for the industry to develop its own native capacity in informatics.

References

- Besson M, H Komen, G Rose and M Vandeputte (2020) The genetic correlation between feed conversion rate and growth rate affects the design of a breeding program for more sustainable fish production. *Genet. Sel. Evol.* **52**(5): DOI: <https://doi.org/10.1186/s12711-020-0524-0>
- Fridsma DB (2018) Data sciences and informatics: what's in a name? *J. Am. Med. Inform. Assoc.* **25**(1): 109. doi: 10.1093/jamia/ocx142
- Smith RD and M Williams (2000) Applications of informatics in veterinary medicine. *Bulletin Medical Library Association* **88**(1): 49-51.

The New Finfish Genetic Resource Discoveries from Indian Waters – Need for Enhancing Taxonomy Capacity

TK Teena Jayakumar¹, KK Bineesh², TT Ajith Kumar¹ and Kuldeep K Lal¹

¹ICAR-National Bureau of Fish Genetic Resources, Lucknow-226002, Uttar Pradesh, India

²Zoological Survey of India, Marine Biology Regional Centre, Chennai-600028, Tamil Nadu, India

India is rich in diversified biological resources and ranks among the top ten species-rich nations with four global biodiversity hotspots. Indian fish fauna is highly diverse and explorations and germplasm inventories of ichthyodiversity are being updated with several new discoveries. However, both geographical and taxonomic knowledge gaps exist, as many regions especially the far-flung and inaccessible pockets are either under- or unexplored, and many taxonomic groups remain poorly studied. During the last two decades, much information on fishes from the Indian regions have been generated and added to the catalogue of Indian fishes due to the advent of integrated taxonomical approach. Increasing number of new species every year, shows that much more of the diversity yet remains to be discovered and documented. Continuous extensive surveys and explorations are warranted in areas such as deep-sea habitats, islands, subterranean regions, caves and hill streams for unravelling the hidden diversity.

Introduction

India has rich and diverse fishery resources, shares four of the mega biodiversity hotspots, ranging from hill streams to deep seas. Of the 36,272 (Fricke *et al.*, 2022) fish species reported from the world, India supports about 3,496 species owing to 9.6% of the total fish diversity (Banerjee *et al.*, 2022). During the last two decades, about 7,957 fish species have been added to the world's ichthyofaunal catalogue, with the annual increase of 397 new species (Fricke *et al.*, 2022). In India during the last 11 years, an average of 24 new species per year were described (Fig. 1). The recent Indian discoveries of new fish species is reported by Jayakumar *et al.*, 2021 and Banerjee *et al.*, 2022. In the Indian context, intensive systematic studies on fishes, often supplemented by molecular-based phylogenetic works, have prompted the revision of several taxa and still many groups of fishes are expanding with newly described species, with a net increase in new fish species. Nevertheless, the increase in species discoveries of each year suggests that there are still many more new taxa to be discovered from the diverse habitats of fresh and marine waters of India.

Freshwater Fish Diversity

Native freshwater fishes of India are recognized under 40 fish families of 12 orders, comprising 858 species belonging to 167 genera (Gopi *et al.*, 2017). Among the Indian freshwater fishes, four families such as

Cyprinidae, Nemacheilidae, Sisoridae and Bagridae are most species-rich with 50 or more species, containing approximately 70% of the total species. The Ganga-Brahmaputra system exhibits among the highest richness of large-bodied freshwater fishes (freshwater mega-fauna) in the world and supports unique and threatened species. The Western Ghats, a global biodiversity hotspot, alone host 288 fish species of which 118 are endemic. The Eastern Himalaya, another biodiversity hotspot, contains exceptional freshwater biodiversity and ecosystems that are of vital importance to local and regional livelihoods. According to the conservation status of freshwater species (IUCN, 2016), 148 species are in threatened category (13 Critically Endangered, 63 Endangered and 72 Vulnerable), while 36 species are Near Threatened, which is likely to slip in to threatened category if proper conservation measures are not taken.

Recent ichthyological explorations on subterranean caves, caverns, lava tubes and phreatic spaces resulted in the discovery of many new species. Habitats, such as deep burrows, ant hill and other similar underground biogenic structures of several burrowing and cryptobiotic animals are encompassed in the subterranean category. These subterranean spaces are poorly explored as regards to biodiversity mapping. The main limitations includes lack of easy access to these underground spaces, prevalence of darkness with high humidity, and the fear of unknown and uncertain things and/or

*Author for Correspondence: Email-teenajayakumar@gmail.com

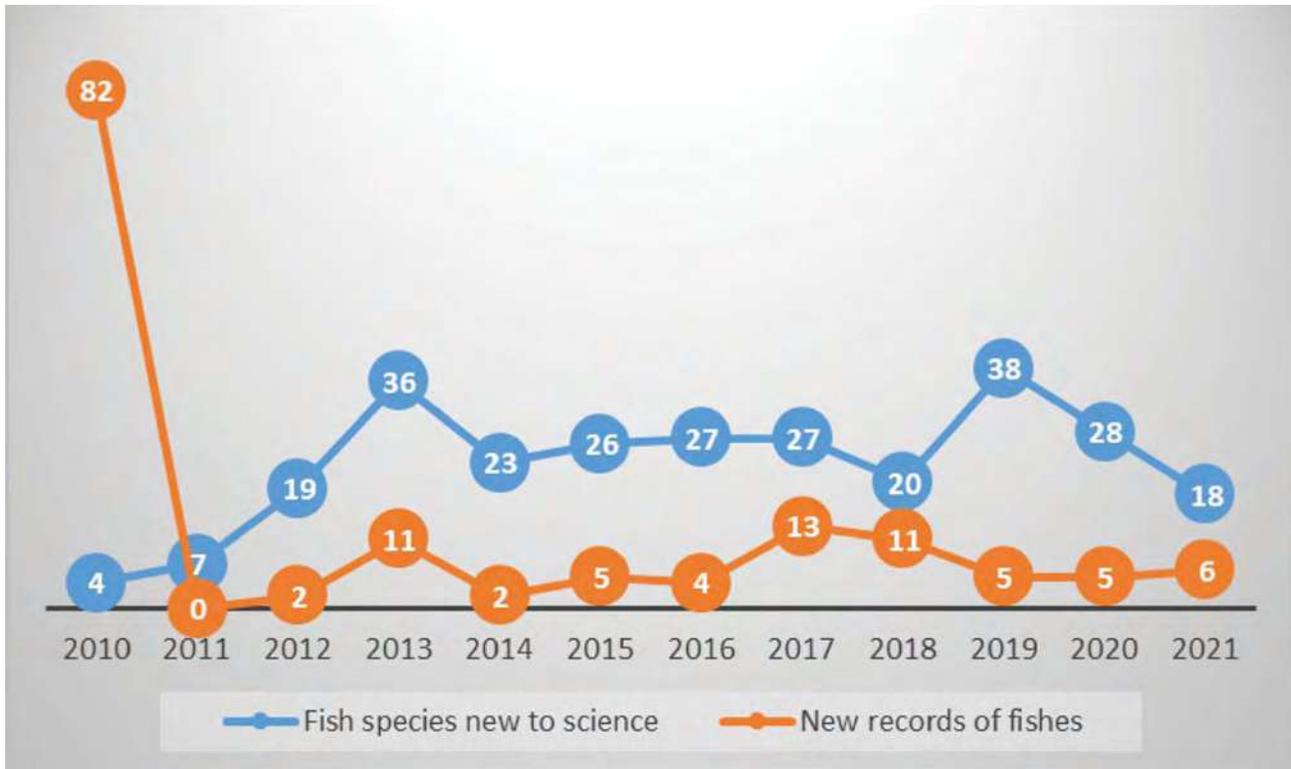


Fig. 1. New species and new records of fishes described in India from 2010-2021
(Source: Animal discoveries 2010-2021, Zoological Survey of India)

events. In India, so far, limited concerted efforts have been made to survey the already known subterranean habitats. Few species described, includes *Nemacheilus evezardi* and *Horaglanis krishnai*. Recently the “living fossil” Gollum snakehead, *Aenigmachanna gollum* was discovered from subterranean waters in the Western Ghats– Sri Lanka, one of the most significant global biodiversity hotspots. This fish was a serendipitous discovery when several individuals were washed out of their aquifer habitat into paddy fields during the 2018 Kerala floods. Western Ghats which has exceptionally high level of biological diversity and endemism is recognized as one of the world’s eight ‘hottest hotspots’ of biological diversity. The exploratory surveys in the area has led to the discovery of new families such as Kryptoglanidae and Aenigmachannidae, new genera such as *Sahyadria*, *Aenigmanachanna*, and *Waikhomia* and various new species in the genera; *Dawkinsia*, *Pethia*, *Pangio*, *Balitora*, *Mystus*, *Badis*, *Amblyceps*, *Channa* and *Pseudolaguvia*.

Similarly, North East India with unique topography, varied watershed pattern and physiography is enriched

with diversified fish fauna. This region is augmented with 422 species belonging to 39 families and 12 orders (Goswami *et al.*, 2012). A recent expedition in caves of Meghalaya, has led to the discovery of world’s largest cave fish, a yet-to-be described species of mahseer which is nearly five times the average length (8.5 cm) of all known subterranean fishes till date. Other discoveries include catfishes, cyprinids and loaches such as *Garra simbalbaraensis* from Simbalbara river Himachal Pradesh, *Glyptothorax gopii*, *Neolissochilus kaladanensis*, *Cabdio crassus* and *Laubuka parafasciata*, from river Kalandan, *Rita bacalu* from river Godavari and *Pangasius silasi* from river Krishna. A new species of blind freshwater eel *Rakthamichtys mumba* was discovered from a 40-foot deep well in Mumbai. Similarly, a new species of swamp eel, *Monopterus rongsaw* was discovered from swampy regions of Meghalaya, which was a serendipitous ‘by-catch’. The Northeast India is far less studied than the Western Ghats. The uniqueness of northeast India and the Eastern Himalayas is becoming apparent with increased research.

Marine Fish Diversity

The geographic territory of India is an integral part of Central Indian Ocean Region, consisting of three distinct marine zones such as Arabian Sea, Bay of Bengal and Indian Ocean. India has vast marine resources in the form of coastline (over 8,000 km), exclusive economic zone (EEZ, 2.02 million km²) and various ecosystems including estuaries, coral reefs, mangroves, lagoons, rocky, marshes and sandy areas. Of the overall fish diversity known from India, the marine fishes make up 75.6 percent, containing 2,443 species belonging to 230 families of 40 orders (Gopi and Mishra, 2015). The order Perciformes is the most species-rich group of Indian marine fishes having a total of 1,367 species accounting for 56.0% of the total marine fish species recorded. Among the fish diversity-rich areas in the marine waters of India, Andaman and Nicobar archipelago exhibits the highest number of species with 1,431, followed by the east coast of India with 1,121 species and the west coast with 1,071 species (Gopi and Mishra, 2015). The coral islands of Lakshadweep exhibit a moderate diversity with 753 species. According to the IUCN (2014), 50 species are threatened (6 of them critically endangered, 7 endangered and 37 vulnerable), while 45 are near-threatened. Despite the large number of fishes listed as critically Endangered and Endangered, only 10 Elasmobranchs, seahorses and pipefishes and one grouper are listed in Schedule I of the Wildlife (Protection) Act, 1972 of the Government of India. Marine fish diversity is in ever-increasing danger with depletion of resources.

Diversity of species associated with coral reefs and associated habitats of India is still unknown. Few recent discoveries of pelagic and reef fishes includes, *Dussumieria modakandai*, *Nemipterus andamanensis*, *Pempheris sarayu*, *Hoplolatilus andamanensis*, *Pteropsaron indicum*, *Stolephorus tamiliensis* and *S. hindustanensis*. Though eels are important components of aquatic ecosystems and have fascinated biologists with mysteries of their ecology for centuries, information concerning species diversity, geographic distribution and biology of eels are highly limited in India. As most of the marine eels do not possess economic value, they have been landed as by-catch in trawl landings and are used as ingredients for poultry feeds leading to the concealment of these groups from taxonomic assessment resulting in biodiversity loss. Recent research on Anguilliformes has resulted in description of few new species like; *Rhynchoconger smithi*, *Cirrhimuraena indica*, *Ophichthus chennaiensis*,

Gymnothorax aurocephalus, *G. odishi*, *Xyrias anjaalai*, *Ariosoma melanospilos*, *A. indicum*, *A. maurostigma* and *A. albimaculatum*.

The fishes dwelling in deep-reef and upper-slope areas are underexplored and new unusual discoveries above the species level can be found. Midwater fishes are pelagic, which spend their entire life off the bottom are often widespread such as lanternfishes (Myctophidae). But only fewer studies are carried out in these groups. Deep-living mid-water species that do not migrate to shallow waters are less known than those that can be caught near the surface at night. More new taxa are expected as deep-ocean trawling continues, but the increase in new taxa is correlated with sampling effort.

The diversity of deep-sea Chondrichthyans species along the Indian coast are poorly known, and considered to be higher than thought earlier. Despite the rich deep-sea chondrichthyans diversity, only two new species of sharks *Mustelus mangloreensis* and *Planonanus indicus* have been described from Indian EEZ in the past decade. The exploratory surveys by FORV *Sagar Sampada* have brought out many rare deep-sea Chondrichthyans that were collected beyond 200-1100 m depth of Indian EEZ during 2008-2019. While extensive explorations have been made, there are still many species known from single type specimens (eg. Broadnose catshark, *Apristurus investigatoris*, Prickly skate, *Fenestra jamamillidensis* and Travancore skate, *Dipturus johannisdavisi*).

Studies on the taxonomy of deep-sea fishes from Indian EEZ have resulted in some distribution extension and description of few species new to science (eg. *Chelidoperca maculicauda*, *Glossanodon macrocephalus*, *Epigonus indicus* and *Eptatretus wadgensis*). There are many taxonomic issues contributing to the confusion in the alpha taxonomy of many families of deep-sea fishes such as Ophidiidae, Chlorophthalmidae, Synodontidae, Myctophidae, Lophiidae, Centrophoridae, Rajidae etc. Despite scarce studies conducted on deep sea resources of this region in the past, a complete documentation is still lacking.

Molecular Approaches in Taxonomy

The identification of fish species is traditionally based on morphological, meristic and anatomical characters. However, the astounding diversity in size, shape and their morphological plasticity makes the fish and their developmental stages difficult to identify with morphological features alone. Molecular studies are

increasing in fish taxonomy for higher resolution and complementing morphological characterization. These studies are useful in discovering new species that, after investigation, may have demonstrable morphological differences. Accurate identification of fishes is very important, especially in the case of morphologically similar or cryptic species, for fisheries management, biodiversity, and population studies. The analysis involving DNA sequences and quantitative morphometric and meristic comparisons have added a new dimension in taxonomic research and also forensic identification.

Major Challenges

Global taxonomy initiative under CBD recognise the lack of taxonomic capacity among nations and considers it important for the effective mainstreaming of aquatic genetic resources. In India, both geographical and taxonomic knowledge gaps exist, as many regions especially the distant and difficult terrains are under- or unexplored, while many taxonomic groups remain poorly studied or have conflicts. Many species might undergo unrecognised extinctions in the absence of targeted efforts. The taxonomic knowledge gaps existing are limitations in achieving the relevant objectives of CBD and policy making. Explorations and germplasm inventories of fish biodiversity in the aquatic water bodies of India are being progressively updated and analysed with several new discoveries. Some of the major reasons for taxonomic impediment are follows;

Inaccessible historic type specimens: Most of the type specimens of fishes are deposited in international museums. The lack of access to the type specimens housed elsewhere is a crucial constraint in carrying out taxonomic works in India. The type specimens' studies aid the differentiation and credible comparison. Though, the online available resources from some museums and original descriptions are useful, this cannot fully replace the information obtained from examining the type specimen.

Lower publication value of taxonomy: The taxonomic journals are mostly niche specific and rate lower on Impact Factors and Citation Indexes. This stifles the original taxonomic research. Moreover, most of the taxonomic information about our biodiversity occurs in the form of scattered and inaccessible publications.

Shortage of trained manpower: The taxonomic crisis is worsened by a sharp decline in expertise and

resources. The number of field taxonomists who is doing survey, studying the natural habitat and identifying and classifying species from the real nature is fast dwindling in the country.

Lack of job opportunities: Taxonomists lack adequate job opportunities. No specific recruitment of field taxonomists is made in the universities and research institutions. Dangerous 'demise' of taxonomy reflects the failure of fellow biologists and policy makers to support taxonomic expertise and resources.

Thrust Areas and Way Forward

There is need for building the institutional and individual taxonomic capacity, through academic curricula combining the morphological and molecular taxonomy tools. The training and education in integrative taxonomy will add to skills, job opportunities and teaching value of such trained personals. A network of taxonomic institutions, individual taxonomists and repositories should be developed in a collaborative manner. Recognition of the institutions as centres of excellence in taxonomic research, repositories under section 39 of biological diversity act 2002 is need of the hour. For accurate generic and species determinations, it is essential to study specimens from across political boundaries and continents and hence, easy facilitations under the BDA 2002 is necessary for exchange of preserved specimens. It is generally accepted among the scientific community that the types are the property of science and should be made available to *bonafide* researchers throughout the world. The efforts to conserve biodiversity will be easier if we know the basic units that are species and their relationships. Taxonomy provides discovery and identification of these units, which is the foundation for all studies on biodiversity. Thus, India, one of the 17 mega-diverse nation in the world, has to overcome taxonomic impediments for the growth of biodiversity science, conservation planning and policy making.

References

- Banerjee D, C Raghunathan, AN Rizvi and D Das D (2022) Animal discoveries 2021: New Species and New Records, Director, Zool. Surv. India, Kolkata, 232pp.
- Dar GH, AAKhuroo, CS Reddy and AH Malik (2012) Impediment to taxonomy and its impact on biodiversity science: an Indian perspective. *Proceedings of the National Academy of Sciences, India Section B: Biol. Sci* **82**(2): pp.235-240.
- Fricke R, WN Eschmeyer and R Van der Laan (eds.) (2022) Eschmeyer's catalog of fishes: genera, species,

- (<http://researcharchive.calacademy.org/research/ichthyology/catalogue/fishcatmain.asp>). Electronic version accessed 13.07.2022.
- Gopi KC and SS Mishra (2015) Diversity of marine fish of India. In: K Venkataraman and C Sivaperuman (eds) *Marine Faunal Diversity in India: Taxonomy Ecology and Conservation*. Academic press, 533 pp.
- Gopi KC, SS Mishra and L Kosygin (2017) Pisces, pp. 527–570. In: K Chandra, KC Gopi, DV Rao, K Valarmathi & JRB Alfred (eds.). *Current Status of Freshwater Faunal Diversity in India*. Director, Zoological Survey India, Kolkata, 624
- Jayakumar TTK, TTK Ajith, M Singh, V Mohindra, RK Singh, R Dayal, JK Jena, Kuldeep K Lal (2021) Integrated taxonomy, conservation and sustainable development: Multiple facets of biodiversity, *Aquaculture Asia* **25**: 1-6
- Trajano E, ME Bichuette and BG Kapoor (eds) 2010. *Biology of subterranean fishes*. CRC Press, 496 pp.
- Umesh CG, KB Sudip, B Dilip, S Konthoujam, S Bishnupriya and C Kimneilam (2012) Fish diversity of North East India, inclusive of the Himalayan and Indo Burma biodiversity hotspots zones: A checklist on their taxonomic status, economic importance, geographical distribution, present status and prevailing threats. *Int. J. Biodivers. Conserv.* **4**(15):, pp.592-613.

A Review on Ichthyofaunal Diversity and Distribution of North-East India

Kangkan Sarma¹, Rajdeep Das¹, Ratul Chandra Bharali² and Dandadhar Sarma¹

¹Department of Zoology, Gauhati University, Guwahati-781014, Assam, India

²Department of Zoology, Udalguri College, Udalguri-784509, BTR, Assam, India

The present communication reviews the ichthyofaunal diversity of the North-east India. Eastern Himalayas are one of the biodiversity hotspots in India and harbour very diverse fish fauna. The biodiversity richness of the north east India has attracted many taxonomists to explore fishes and have reported different views on the number of fish species from this area. This study compiles all the previous studies and found the ichthyodiversity to be a total of 512 fish species from 128 genera, 40 families and 12 orders. But about 16% of fish diversity in North-East India comes under the threatened category for which proper conservation measures are desirable and about 33% of fishes come under the data-deficient (DD) and not-evaluated (NE) categories and their conservation status needs more attention.

Introduction

North East region is hilly land-locked area lying between 21° 57' & 29° 23' and 87° 58' & 97° 09' and physiographically categorized as Eastern Himalayan, Northeast Hills, Brahmaputra and Barak Valley plains. With five important drainage system; the Brahmaputra, the Barak-Surma-Meghna, the Koladyne, the Chindwin and the Karnaphuli forming fresh water ecosystem of the region, comprising of 19,868 km of rivers, tributaries, 14,3338 ha wetlands and lakes, 23,792 ha reservoirs. The hills and undulating valleys of this region imparts are source of torrential hill streams, leading to formation of large rivers and become part of the Ganga-Brahmaputra-Barak-Chindwin-Koladyne-Gomati-Meghna system (Lokeshwor, 2013). The region has a much more sophisticated geomorphic history and pervasive topographic features than the Central Himalayas which facilitated the region's rich biological diversity and ecosystem structure (Vishwanath, 2021).

Endemism is ecological state of a species being unique to a defined geographic location, such as an island, nation, country or other defined zone, or habitat type (Darlington, 1957). The North-eastern region is particularly a hotspot for endemic freshwater biota. The high degree of endemism in this region is thought to have resulted from its long period of isolation and complex evolutionary history, which promoted *in situ* diversification (Darlington, 1957). The Indo-Burma biodiversity hotspot has an amazing freshwater fish fauna, with 1262 recorded species

including 566 endemics accounting for about 10 % of the world total (Tordoff *et al.*, 2012).

Freshwater fishes are one of the most threatened taxonomic groups (Darwall and Vie, 2005) because of high sensitivity to any quantitative and qualitative alterations in their habitat as well as physico-chemical parameters. The North-eastern region of India is considered to be one of the hotspots of freshwater fish biodiversity in the world (Ramanujam *et al.*, 2010). A great number of species have been reported from most of the North-eastern states. Out of a total of 2500 species of fish in India, 930 are confined to freshwaters, belonging to 326 genera, 99 families and 20 orders (Talwar and Jhingran, 1991). North-eastern states of India are rich in fish resources as evident from the reports of Talwar and Jhingran (1991), Acharjee *et al.* (2012) and Vishwanath (2021). Therefore, an attempt was made to note the fish species occurrence, distribution and also to add further information on the fish species diversity of North-Eastern part of India.

Fish Diversity of North-east India and Endemism

The freshwater of the Himalayas and Indo-Burma are highly diverse (Allen *et al.*, 2010). Significant contribution on the systematics and fisheries of the river basin of northeast India includes work of Talwar and Jhingran (1991), Menon (1999), Vishwanath (2021).

Dey (1984) made systematic account of the ichthyofauna of the Northeast India and listed 123 fish species from this region. A total of 177 Cyprinid species

*Author for Correspondence: Email-sarma_dandadhar@yahoo.com

belonging to 2 families, 4 subfamilies and 27 genera are so far reported from the inland waters of India. Out of which 43 species are considered of great economic importance. Menon (1999) listed 446 primary freshwater species under 33 families and 11 orders from the India. Of the primary freshwater species 68% are constituted by the Cyprinoids, 18% by Siluroids while 14% by other groups. Nelson (2006) reported 27,977 fish species under 4,494 genera, 62 orders and 515 families.

Yadava and Chandra, (1994) reported that ichthyofaunal studies of the northeast region of India, which has elements of the Indo-gangetic region and to some extent, elements of the Myanmarese and South-Chinese regions, is scarcely studied. Kottelat & Whitten (1996) also estimated the Brahmaputra-Irrawaddy to contain 200 species of fish.

Sen (2003) reported 291 species belonging to 38 families and 12 orders; of which 35 species were identified as endemic species in different states of northeast India. Vishwanath (2012) reported about 300 fish species under more than 100 genera and 30 families. He listed candidates of aquarium fishes belongs to groups nemacheilides, Cobitids, Psilorhynchids, Sisorid cat fishes, badids, danionins and also the genera of cold-water fishes viz, *Tor*, *Neolissochilus*, *Schizothorax*, *Labeo*, *Cirrhinus*, *Semiplotus*, *Poropuntius Barilius*, *Raiamas*, *Danio*, *Garra*, *Pterocryptis*. Goswami (2012) reported and recorded 442 fish species from North East India, belonging to 133 genera and 38 families. The maximum diversity observed for family Cyprinidae with 154 species.

Acharjee *et al.* (2012) studied on Ichthyofaunal diversity of Dhansiri River, Dimapur, Nagaland, India. Das *et al.*, (2014) studied on fish diversity and drainage analysis of river Siang, East Siang District of Arunachal Pradesh. This study reveals revealed the presence of 82 species of fishes belonging to 8 orders, 24 families and 53 genera. Species representing the order Cypriniformes dominated the ichthyofauna while those representing Tetradontiformes are in less number.

Brahmaputra river system form lucrative fields of Ichthyological importance in northeast India. Several workers have studied the River Brahmaputra and its tributaries in Assam. Baishya *et al.* (2016) recorded 52 small indigenous fish species under 15 families and 33 genera in the upper reach of Brahmaputra.

The most recent work on the compilation of the fish species reported from this region was carried out by Vishwanath (2017) studied the Diversity and conservation status of freshwater fishes of the major rivers of northeast India and reported 318 fish species under 113 genera and 36 families. A total of 229 species are in the Brahmaputra drainage, 103 in the Chindwin, and 27 in the Kaladan. He mentioned 27 species endemic to the Brahmaputra basin as well as 41 species endemic to the Chindwin drainage. Dey *et al.*, 2021 reported a total of 117 fish species from 12 selected sites of the Kameng river basin. Cyprinids represented the dominant group with 45 species. *Garra annandalei*, *G. quadratirostris*, *Neolissochilus hexagonolepis* and *Schizothorax richardsonii* were the most dominant species.

In last five years explosion of literature on taxonomic research of new species discovered has been witnessing from this region. Some of the publication on the description of new species from the rivers of NE region includes, *Schistura rebuw* (Choudhury *et al.*, 2019); *Mustura subhashi* (Choudhury *et al.*, 2021); *M. daral* (Rameshori *et al.*, 2021). Description of so many new species reflects the undiscovered rich biodiversity of the region as many lotic water bodies have not been completely explored because most of the rivers are located in inaccessible mountainous terrain with dense forest cover.

Conclusion

Besides the probable importance of this system, we have found a gap of information regarding the list of fish fauna in the drainage systems of the states of NE, India. In recent years many new species have been described from this region suggesting the importance of water bodies of NE, India. An extensive study on the taxonomy and biology of the freshwater fishes in India has been achieved. The study highlights the need to explore fish fauna of the major river system of North-East India at regular intervals to build a comparable database that might be utilized by the stakeholders for research, planning and management activities. Also, adequate information on the biology and distribution of species for accurate assessment is lacking. Fresh water ecosystems are especially vulnerable to human activities. Effective conservation measures should be taken in order to stop anthropogenic activities as well as habitat degradation by involving local people, making them aware of the

potential ichthyofaunal hub of freshwater fishes in the north-eastern region of India.

References

- Menon AGK (1999) Checklist of the Freshwater fishes of India. Record of Zoological Survey of India, Zoological Survey of India, Govt. of India. pp.366.
- Acharjee BK, M Das, P Borah and J Purkayastha (2012) Ichthyofaunal diversity of Dhansiri river, Dimapur, Nagaland, India. *Check List* **8**(6): 1163-1165.
- Baishya RA, S Basumatary, HK Kalita, B Talukdar, A Dutta and D Sarma (2021) Status and Diversity of Indigenous Ornamental Fishes of the Upper Reaches of River Brahmaputra, Assam. *Journal of the Inland Fisheries Society of India* **47**(2): 70-77.
- Choudhury H, R Das, RC Bharali, K Sarma, LK Tyagi, KK Lal and D Sarma D (2021) Description of a new species of Mustura Kottelat (Teleostei: Nemacheilidae) from the Brahmaputra drainage, India. *J. Fish Biol.* **99**(2): 450-461.
- Choudhury H, A Dey, RC Bharali, D Sarma and W Vishwanath (2019) A new species of stone loach (Teleostei: Nemacheilidae: Schistura) from Arunachal Pradesh, India. *Zootaxa* **4551**(1): 40-52.
- Darlington PJ (1957) *Zoogeography: the geographical distribution of animals* (No. 591.9). Wiley.
- Darwall WRT and JC Vie (2005) Identifying important sites for conservation of freshwater biodiversity: extending the speciesbased approach. *Fish Manag Ecol* **12**: 287-293. <https://doi.org/10.1111/j.1365-2400.2005.0049.x>
- Das BK, P Boruah and D Kar (2017) Ichthyofaunal Diversity of Siang River in Arunachal Pradesh, India. In *Proceedings of the Zoological Society* **70**(1): 52-60.
- Kottelat M and T Whitten (1996) *Freshwater biodiversity in Asia: with special reference to fish* **343**: World Bank Publications.
- Lokeshwor Y (2013) Northeastern India-a natural repository of stone loaches. *Message from the Co-chairs*.
- Sen N (2003) Records of Zoological Survey of India. **101**: 81-99.
- Ramanujam SN, M Manorama and S Dey (2010) Ichthyodiversity of Meghalaya: India. *Electron J. Ichthyol.* **2**: 15-26.
- Rameshori Y, Y Chinglemba, A Darshan and W Vishwanath (2022) *Mustura daral*, a new species of stone loach from Arunachal Pradesh, Northeast India (Teleostei: Nemacheilidae). *Zootaxa*, **5129**(2): 285-294.
- Talwar PK and AG Jhingran (1991) *Inland fishes of India and adjacent countries*. Oxford and IBH Publishing Co. Pvt. Ltd, New Delhi.
- Tordoff AW, MC Baltzer, JR Fellowes, JD Pilgrim and PF Langhammer (2012) Key biodiversity areas in the Indo-Burma hotspot: process, progress and future directions. *J. Threat. Taxa.* 2779-2787.
- Vishwanath W (2017) Diversity and conservation status of freshwater fishes of the major rivers of northeast India. *Aquatic Ecosystem Health & Management*, **20**(1-2): 86-101.
- Vishwanath W (2021) *Freshwater Fishes of the Eastern Himalayas*. Academic Press.
- Vishwanath W, WS Lakra and UK Sarkar (2012) *Fishes of Northeast India*. NBFGR, Lucknow, UP, India.
- Yadava YS and R Chandra (1994) Some threatened carps and catfishes of Brahmaputra river system. In: PV Dehadrai, P Das and SR Verma (eds). *Threatened Fishes of India*. Natcon Publication, Muzaffarnagar 45-55..

Diversity of Fish Genetic Resources below the Species Level, Characterization and Applications in Resource Management

Rajeev K Singh*, PR Divya, Vindhya Mohindra and Kuldeep K Lal

ICAR-National Bureau of Fish Genetic Resources, Lucknow-226002, Uttar Pradesh, India

Intraspecific variation includes the genetic and phenotypic diversity found within and among populations that are often threatened by a reduction in population, local extinctions, climate change, and anthropogenic activities. Documentation of these below-species level variations constitutes the primary step towards conservation. High-resolution genetic markers can potentially be deployed to understand the pattern of genetic diversity at intra and inter-specific levels. The inferences drawn from genetic diversity analysis holds many applications, mainly taxonomy/systematics of species, understanding their genetic connectivity, identifying the breeding populations/parentage assignment, fisheries management, and conservation measures including ranching programs for the purpose of genetic rescue and stock enhancement of wild relatives. These variations form strong foundation for creating base populations in a selective breeding program.

With the adoption of novel high-resolution molecular tools and genome-wide approach, a comprehensive set of applications relevant to fisheries management, aquaculture development, biosecurity, and traceability in the supply chain system, can successfully provide momentum to the sectorial growth. This article highlights the significance and need for intraspecific genetic variabilities; and presents lead research undertaken for registering the genetic stocks and elite germplasm in the country, along with significant breakthroughs globally.

Introduction

Intraspecific variation includes diversity within the species, including from its geographic distribution and encompasses temporal persistence, which results in stability of the populations. As the selection pressure is working on it, in addition to changes on random population sizes and genetic drift, its maintenance is important for the ecosystem's ability to adapt and resilient to climate change, there are consequences of human interventions also, on intraspecific variability (Mimura *et al.*, 2016). Roches *et al.* (2021) suggested the rate of loss of intraspecific variation is greater than the species loss.

Biodiversity is among the most important resources, globally. Several biotic and abiotic factors contribute to the sustenance of biodiversity. The FAO's Commission on Genetic Resources for Food and Agriculture (CGRFA) envisions the conservation of biodiversity for Food and Agriculture and promotes its sustainable usage for food security, human well-being, and overall development. India has set national targets within the framework of the Aichi targets. National Biodiversity Target 17 encourages the maintenance of genetic diversity of cultivated plants, farm livestock, and their wild relatives, including other socioeconomically as well as culturally valuable species.

It also suggests to develop and implement strategies for minimizing genetic erosion and safeguarding their genetic diversity. Similarly, SDG goals 14 highlights to conserve and sustainably use the oceans, seas, and marine resources, and SDG target 14.4 aims to end overfishing, illegal, unregulated, unreported fishing and destructive fishing practices. Genetic stock identification can be a supporting tool in resolving traceability issues, as done in the 'Fish Pop-Trace' Project by the European Union.

The loss of genetic diversity has been recognized as a serious concern among the scientific communities, which is evident from CBD post-2020 draft that includes genetic diversity among the five main goals. The significance of intraspecies diversity was highlighted in a thematic document of FAO (2021), which has given thrust to collating the genetic information, especially below the species level, e.g. stocks and strains. Intraspecific studies advocate the characterization at both levels to form the baseline information. At present, this baseline information is available for the commercially important fish species, while it is still needed for several other potential species.

Documentation of intraspecific variation, at genotype and phenotype levels, forms the primary step towards its conservation. The distribution and pattern of genetic

*Author for Correspondence: E-mail-Rajeev.Singh1@icar.gov.in

variability at intra/inter-specific level are deciphered, through the use of high-resolution genetic markers. Advances in sequencing technologies can potentially provide molecular resources for generating genotypes/haplotypes. The conclusions from genetic diversity analysis are important for multiple applications including conservation and management, understanding genetic connectivity of fish wild stocks/populations, breeding populations, selection programs and also in taxonomy/systematics. The present paper highlights the significance and role of intraspecies genetic variability, in these areas.

Significance of Intraspecific Variation Studies

1. Genetic Stock Identification

In India, fish diversity below the species level has been characterized for several species which are important from a conservation or aquaculture point of view. Genetic variability in natural populations of aquaculture species is documented so as to plan its use in domestication, genetic improvement and conservation of the natural gene pool. Earlier, the Indian Council of Agricultural Research (ICAR) initiated a program of registering the genetic stocks and elite germplasm of potentially cultivable fish species both from natural populations and domesticated sources. This was done to formulate the guidelines related to biodiversity and IPR protection. A total of 71 fish/shellfish species found in Indian waters have been genetically characterised, of which 34 are cultivable species. With an aim to integrate genotype information with biological descriptors, the fish/shellfish species characterised by ICAR-NBFGFR include *Catla catla*, *Cirrhinus mrigala*, *Labeo rohita*, *L. calbasu*, *L. dero*, *L. dyocheilus*, *L. dussumieri*, *Clarias magur*, *Pangasius pangasius*, *Horabagrus brachysoma*, *Gonoproktopterus curmuca*, *Channa marulius*, *Etroplus suratensis*, *Harpadon nehereus*, *Pampus candidus*, *Rachycentron canadum*, *Sillago malabarica*, *Scomberomorus commerson*, *Rastrelliger kanagurta* and invertebrates like *Hippocampus kuda*, *H. trimaculatus*, *Fenneropenaeus indicus*, *Penaeus monodon*, *Panulirus homarus*, *Thenus unimaculatus*, *Parapenaeopsis stylifera* and *Perna viridis*.

2. Resource Management

Validated molecular markers can be suitably applied for tracing the population and this can be a useful approach to curtail illegal, unreported and unregulated (IUU) fishing in the marine environment. 'Fish Pop trace' is an international programme by European Union to address

the problems of IUU fishing in their region. In order to satisfy the increasing number of consumers demanding for edible products of certified origin, the European Commission documented three protection labels including the Protected Geographical Indication (PGI) that indicates a connection with the area, where at least one of the production stages possesses a characteristic feature which links to its geographical origin. In the case of *Sardina pilchardus* (*European pilchard*), the region of the fish origin was identified using mitochondrial markers (Gonzalez *et al.*, 2007). Similarly, genetic tools for geographic traceability of cods and soles are worked out from European countries (Bylemans *et al.*, 2016) and yellowfin tuna (Pecoraro *et al.*, 2018) across its range of natural distribution. The ability to trace the fine scale population structuring depends on marker diversity and species population history. Among the marine species, the genetic divergence varies between species, based on its dispersal capacity, larval distribution, breeding season, behavioural characteristics, etc. In addition, oceanographic discontinuities reduce gene flow and environmental characteristics that determine local adaptation, are sometimes undetectable drivers of population differentiation. Such in-depth genetic studies help to define the conservation and management units and give an indication to the policymakers if they are to be managed as single or multiple populations. However, genetic information has not always been incorporated into fishery management and policy decisions in several countries. At this juncture, a consensus among scientists, governmental organizations, and policymakers are crucial in developing and implementing policies for the sustainable management.

3. Ranching Programs for Genetic Rescue and Stock Enhancement of Wild Relatives

Interspecific variations can deplete in many fish species, due to over exploitation and habitat destruction of the wild fish populations, "Freshwater fish are under ever increasing pressure with one in three threatened with extinction, according to the new report published by 16 global conservation organizations." (WWF, 2021). In this direction, captive breeding of threatened populations and reintroduction can be a means of recovery efforts or genetic rescue. By ensuring that the stocked population is imbued with the genetic diversity from wild population, reintroduction of the stocked fish will likely be more successful and portfolio effects will be possibly preserved.

ICAR-NBFGR with Uttar Pradesh State fisheries Department has carried out ranching five carp species including Indian major and minor in the protected areas of river Ganga and its tributaries. For ranching advanced fingerling are produced from wild originated individual tagged broodstock maintained at Live Fish Germplasm Resource Centre of NBFGR at Lucknow. A total of 375000 advanced fingerlings have been stocked during 2017 to 2020 in the areas of river falling under protected or religious sanctuaries, which have low human interference. Similarly, stock-specific, breeding-assisted river ranching of two food fishes (*Horabagrus brachysoma* and *Labeo dussumieri*) has also been successful in Kerala and the landings of *H. brachysoma* after two years increased from 1.8% to 11% and that of *L. dussumieri* showed an increase from 0.68% to 3.9% of the total landings from the Vembanad Lake and adjacent rivers in the state (Padmakumar *et al.*, 2011). Recently, further the stocking program is in place by Kerala state department with seed produced from Peninsular and Marine Fish Genetic Resources Center of ICAR-NBFGR at Kochi. Over 250000 fingerlings of endemic species, *H. brachysoma* and *L. dussumieri* were ranching by the department for enhancing their natural populations. Nilgiri catfish, *Hemibagrus puctatus* a species endemic to Western Ghats from river Cauveri, considered extinct for over twenty years, could be an example for genetic rescue in future. The broodstock raised through wild collection has been bred in captivity and with propagation technology developed, it will be a candidate for ranching and stocking in the native distribution range.

4. Selective Breeding

Selective breeding has great potential for genetic improvement of economic traits in aquatic species. Compared to livestock, where the bulk of production comes from less than a dozen species, aquaculture circumvents not less than five dozen species to support nutritional security. The selective breeding program has a prerequisite of the creation of a base population, for which the estimates of genetic diversity and genetic parameters are required. The base populations are mostly selected based on the phenotypic and genotypic performance and evaluation. Genotypic performance evaluation is achieved based on the intra-specific variability studies. Selective breeding has been used largely in European aquaculture and several breeding programs majorly, on growth performance, are ongoing.

Of total aquaculture produce in Europe, about 80-83% was obtained from selective breeding (Janssen *et al.*, 2017). This includes six major cultured species, salmon (*Salmo salar*), rainbow trout (*Oncorhynchus mykiss*), gilthead seabream (*Sparus aurata*), European seabass (*Dicentrarchus labrax*), common carp (*Cyprinus carpio*) and turbot (*Scophthalmus maximus*). In rohu carp (*Labeo rohita*), a collaborative program of ICAR-CIFA with the Institute of Aquaculture Research (AKVAFORSK), Norway resulted into a genetic gain of about 17% per generation for growth performance (Jayanti rohu), which is still demonstrating consistent results even after nine generations. The program used base population from five riverine sources for providing broader genetic base to founder individuals. Similarly, in order to develop base population for *Labeo catla*, another major carp, phenotypic and microsatellite marker were used for optimizing the proportions of individuals from each geographic location. The estimates of genetic differentiation and partitioning of genetic variance displayed adequate genetic heterogeneity for selection of populations.

Currently, the black tiger shrimp, *Penaeus monodon*, is being considered for re-introduction into aquaculture, as an alternate species for Indian shrimp farming industry. The Govt. of India has also recognized the need for species diversification for nutritional security and has entrusted RGCA, MPEDA to take lead through PMMSY. In this context, the domestication of *P. monodon* is being carried out at RGCA. The founder populations for developing the SPF domesticated stock were selected on the basis of their diversity profiles, investigated through the molecular markers (Mandal *et al.*, 2012). The population for Andaman and Nicobar Islands was identified as founder populations based on the heterozygosity information.

5. Parentage Assignment

Success of a breeding program largely depends on ability to minimise inbreeding levels, which requires prior knowledge genetic diversity estimates, below the species level. The molecular markers, particularly microsatellites and SNPs, are used to trace inherited alleles in offspring from a pool of brooder parents and thereby, make an effective mode of pedigree management. Recently, molecular panels were deployed to ensure that white-leg shrimp (*Penaeus vannamei*) lines belonged to multiple genetic sources. Similarly, microsatellite panel

was developed for meagre (*Argyrosomus regius*) for parental assignment (Vallecillos et al., 2022). In marine species, parentage assignment was explored, Atlantic cod *Gadus morhua* and sole, *Solea solea* (Bylemans et al., 2016). For a large variety of commercially reared marine species, escapees and deliberate releases have been reported. Such studies are focused mainly on salmonids and several other important species. In such scenarios, developing baseline genetic information becomes pertinent for aquaculture species.

Way Forward

The characterization of diversity below-the-species level of cultivable fish species is required for a variety of applications, as mentioned above. However, characterization of biodiversity should be extended to many commercially/evolutionary important species through integrative novel technologies of phenomics and genomics. It would help in large scale data collection and analysis at both the levels, for better understanding of the complex association between genotypes and phenotypes, as well as hidden/cryptic variability. Furthermore, scientific evidences indicate the need for adopting the holistic approaches incorporating ecosystem services and functions.

Fishes usually adapt to the changing environmental conditions in response to the climate change scenario and these adaptations are reflected at the genomic level also. Identifying evolutionarily significant units (used to delineate fish stocks), determining connectivity among locations (useful for defining marine protected areas), and generating genetic traceability tools (which allow for assigning individuals back to their birth areas in the case of mixed stock fisheries) needs attention in the changing scenario of fisheries environment. Validated molecular markers can be suitably applied for tracing the population and this can be a useful approach to curtail illegal, unreported and unregulated (IUU) fishing in the marine environment. One example is 'Fish Pop trace', an international programme by European Union, to address the problems of IUU fishing in their region,

The advent of newer tools such as environmental DNA (eDNA) monitoring the distribution and relative abundance of aquatic organisms, from microbes to sharks and whales, from water samples has made it more useful. This may be proved to be a fast and cost-efficient method and a prerequisite for ecosystem-based management of fishery resources, for monitoring at all trophic levels.

Acknowledgements

The authors thank Dr. Trilochan Mohapatra, Secretary, DARE and Director General, ICAR; Dr. Joykrushna Jena, DDG (Fisheries Science), ICAR for their support and guidance. Thanks also due to Director, ICAR-NBFGR for providing the facilities for this manuscript.

References

- Bylemans, Jonas; Gregory E Maes, Eveine Diopere, Alessia Cariani, Helen Senn, Martin I. Taylor, et al. (2016) Evaluating genetic traceability methods for captive-bred marine fish and their applications in fisheries management and wildlife forensics. *Aquaculture Environment Interactions*, doi: 10.3354/aei00164
- Gonzalez EG and R Zardoya (2007) Relative role of life-history traits and historical factors in shaping genetic population structure of sardines (*Sardina pilchardus*). *BMC Evolutionary Biology* 7: 197. doi: 10.1186/1471-2148-7-197. PMID: 17953768; PMCID: PMC2204010.
- Janssen, K, H Chavanne, P Berentsen and H Komen (2017) Impact of selective breeding on European aquaculture, *Aquaculture* 472(1): 8-16, <https://doi.org/10.1016/j.aquaculture.2016.03.012>.
- Mandal A, D Rao, D Karuppaiah, A Gopalakrishnan, J Pozhoth, YC Samraj and RW Doyle (2012) Population genetic structure of *Penaeus monodon*, in relation to monsoon current patterns in Southwest, East and Andaman coastal waters of India. *Gene* 10 491(2): 149-57. doi: 10.1016/j.gene.2011.10.002.
- Mimura, M, T Yahara, DP Faith, EI Vázquez-Domínguez, RI Colautti, H Hitoshi Araki, et al. (2016). Understanding and monitoring the consequences of human impacts on intraspecific variation. *Evolutionary Applications*, doi: org/10.1111/eva.12436
- Padmakumar KG, L Bindu, PS Sreerexha, A Gopalakrishnan, VS Basheer, N Joseph, PS Manu and Anuradha Krishnan (2011) Breeding of endemic catfish, *Horabagrus brachysoma* in captive conditions. *Curr Sci* 100(8): 1232-1236.
- Pecoraro C, M Babbucci, R Franch, et al. (2018). The population genomics of yellowfin tuna (*Thunnus albacares*) at global geographic scale challenges current stock delineation. *Scientific Reports* 8, 13890 doi.org/10.1038/s41598-018-32331-3
- Roches DS, LH Pendleton, B Shapiro et al. (2021) Conserving intraspecific variation for nature's contributions to people. *Nature Ecology & Evolution* 5, 574–582. <https://doi.org/10.1038/s41559-021-01403-5>
- Vallecillos A, E María-Dolores, J Villa, FM Rueda, J Carrillo, G Ramis, M Soula, JM Afonso, E Armero (2022) Development of the First Microsatellite Multiplex PCR Panel for Meagre (*Argyrosomus regius*), a Commercial Aquaculture Species Fishes v.7 no.3
- WWF, 2021. *World's Forgotten Fishes: Valuing freshwater fish is critical for people and nature*, WWF. Retrieved: https://policycommons.net/artifacts/1294658/world's_Forgotten_fishes_report_final_1/1897863/ on 25 Jul 2022. CID: 20.500.12592/cgcz.

Genomic Research in Fishery Sector: Current Status and Future Prospects

Murali S Kumar, Ravindra Kumar, Basdeo Kushwaha, Rajeev K Singh, Vindhya Mohindra*

ICAR-National Bureau of Fish Genetic Resources, Lucknow-226002, Uttar Pradesh, India

Large scale efforts on genomics has resulted in greater understanding of the genomic architecture, their evolution and underlying biological mechanisms in fish. Although, significant achievements in fish genome sequencing initiatives have been undertaken globally, further targeted efforts are needed to realize its full potential. It is expected that the knowledge of structure and function of genomes and their interactions with non-genetic components of production systems will help to improve productivity, as well as in conservation and sustainable management of fisheries. In this direction, integrative multi-omics data intensive projects may be carried out in consortium mode research by pooling the expertise from different domains. These efforts will pave way for efficient resource use, to reduce production cost and increase farmers' income.

Introduction

Fish genetic resources (FGR) are vital for their role in food security, nutrition and livelihood generation. As per key UN Goal of 2030 for Sustainable Development, the capture fisheries and aquaculture, which form important components of fish genetic resources, shall have to play an essential role in meeting nutritional security and achieving sustainable development. For this purpose, an urgent need is felt for mainstreaming agrobiodiversity, which includes its characterization, conservation and sustainable utilization. With the limitation in increasing capture fishery production, aquaculture is the next best option for meeting the demand of growing population. The FGR offers its diversity for improving the production of domesticated species and provides new species for diversification of aquaculture. It also supports the country's foreign exchange, including seafood exports as well as ornamental fish trade. Traditionally, the genetic improvements in fisheries were achieved through the selective breeding programmes, however, with the advancements of modern genomic tools, it is now possible to address biological queries or alter phenotypes in the ways not previously imagined. Application of genomics in fisheries has resulted in a significant surge in bio-prospecting of genes and allele mining, whereby several novel genes and alleles have been discovered, which may have potential role in increasing production and planning conservation strategies. This article focusses on some of the strides made in fish genomic research in India along with future prospects.

Genome Sequencing

Whole genome information provides insights into structure and organisation of genes, which gives

understanding into their function and roles in biological systems. The genome re-sequencing offers opportunity to unearth and understand the genomic variations, developing genome-wide markers to estimate genomic variability and for their associations with a particular trait, while comparative genomics helps to infer evolutionary processes and ancestry of the genes among the species. The completion of human genome project, at the turn of the century, followed by advancements in massive parallel high-throughput DNA sequencing technologies and strong computational tools, have transformed the genomics sector in a significant manner. Human genome sequencing initially employed Sanger sequencing, which even though produced high-quality DNA sequences, was time and cost intensive. Second (Next) generation sequencing (NGS) methods in early 2000s, like pyrosequencing (Roche 454), bridge amplification-based sequencing (Illumina) etc., heralded large scale genomics research in fisheries sector. It was followed by Third Generation Sequencing (3GS) technologies, like Pacific Biosciences Sequencing and Oxford Nanopore Technologies that can produce long reads, revolutionised the study of fish genomes at a high resolution. Genome sequencing projects in fish started in the early 2000s, with Japanese pufferfish, *Fugu rubripes*, to be the first to have its whole genome sequenced in 2002. In 2009, Genome 10K Project was launched by a consortium of scientists in Santa Cruz, California to sequence 10,000 vertebrate genomes, including 4000 fish species, with a mission to understand how complex animal life evolved through the changes in DNA. The project aimed to sequence at least one individual from each of the ~66,000 vertebrate species. The publication of zebrafish (*Danio rerio*) genome in 2013 started a new

*Author for Correspondence: E-mail-Mohindra.Vindhya@icar.gov.in

chapter in fish genomics.

In 2020, Fish10K (The 10,000 Fish Genomes) Project was announced, as a sub-project of Earth BioGenome project, to exclusively sequence 10,000 fish species, with an aim to study the evolution and development of vertebrates, based on the big data of fish genomics. In contrast to the draft assemblies generated earlier, the focus is now to use combination of short-reads, long-reads and Hi-C technologies to construct chromosomal-level genome assemblies. Globally, around 867 fish genomes were sequenced and assembled at different levels (<https://www.ncbi.nlm.nih.gov/genome>, accessed on August 22, 2022), which included genome sequences of over 86 aquaculture species. The smallest assembled genome is that of silver spinyfish, *Diretmus argenteus* (302.36 Mb), while the largest one cloudy catshark, *Scyliorhinus torazame* (4470.98 Mb) (<https://www.ncbi.nlm.nih.gov/genome/?term=Scyliorhinus+torazame>).

In India, several fish genomes have been sequenced and assembled under different schemes, includes magur (*Clarias magur*); rohu (*Labeo rohita*) by ICAR-NBFGR and ICAR-CIFA under DBT funding; hilsa shad (*Tenualosa ilisha*), and catla (*Catla catla*) by ICAR-CIFA and ICAR-NBFGR and White Shrimp (*Penaeus indicus*) by ICAR-CIBA under Consortium Research Project (CRP) on Genomics of ICAR led by ICAR-NBFGR; wild variant of zebrafish (*Danio rerio*) by CSIR-IGIB, New Delhi, pabda (*Ompok bimaculatus*) by AIIMS, New Delhi. An oomycete (*Aphanomyces invadens*), a fish pathogen, has also been sequenced at ICAR-NBFGR. Several gene variants and novel genes were identified for the first time in fish in these studies. ICAR-NBFGR has also developed a databases on AqGR such as AqGRISI (Aquatic Genetic Resource Information System of India), FBIS (DNA barcode information), FishMicrosat (microsatellite information), FMiR (mito-genome resource), Fish Karyome (chromosome information) and HRGFish (hypoxia responsive genes) focused on fisheries (Murali *et al.*, 2019). Mitochondrial genome is a valuable resource not only for species identification but also for understanding the evolution and phylogenetic distribution of the species. It has a small size (16-17 kb) and circular structure with maternal inheritance, rapid evolution, limited recombination and variable evolutionary rates across its genes. Globally at present, mitochondrial genomes of over 2900 fish species, representing about 426 families under 67 orders, have been sequenced (<https://mail.nbfgr.res.in/fmir/>).

Transcriptome Sequencing

If genome is said to be the static molecular part, then the transcriptome, consisting all of the mRNA, rRNA, tRNA and ncRNA etc., can be called as the dynamic part. Transcriptome sequencing or RNA-Seq is an important tool, that utilizes deep sequencing technologies and facilitates to understand the functional structure and large-scale expression profiles of the genes. It has the potential to enhance our understanding of the relationships between the genotypes and the phenotypes, in relation to varying biology and environment, which in turn would explain involvement of the genes, through their expression profiles, influencing a particular trait.

Fish-T1K (Transcriptomes of 1,000 fishes) project was launched in 2013, jointly by Beijing Institute of Genomics (BGI) and China National Genebank (CNGB) to generate large-scale transcriptome sequences for 1,000 diverse fish species, for evolution studies in fishes. Over 98,400 RNA-Seq reads data on fish species can be found in NCBI SRA database (<https://www.ncbi.nlm.nih.gov/sra/?term=RNA-Seq+AND+Fishes>, accessed on August 22, 2022). It has resulted in greater understanding of biotic- (bacterial and fungal), abiotic- (hypoxia, ammonia, salinity, temperature etc.) stress tolerance mechanisms and understanding of biological processes, such as growth, reproduction, disease resistance etc. In India, the transcriptome sequencing works have been undertaken for identification of genes controlling important production traits like growth, disease resistance and reproduction; and marker discovery. The species include Indian major carps (*Labeo rohita* and *Catla catla*), hilsa shad (*Tenualosa ilisha*), magur (*Clarias magur*), Nile tilapia (*Oreochromis niloticus*), common carp (*Cyprinus carpio*), Indian white shrimp (*Fenneropenaeus indicus*), spotted snakehead (*Channa punctata*).

Molecular Markers Discovery

Rapidly advancing molecular and computational tools have led to generation of gigantic information and associated population level molecular markers. These provides indispensable insights onto the taxonomic position, phylogeographic positions, conservation management units, population profiles and demographic history of the species of concern. Studies have demonstrated the application of genomics led conservation studies (Garner *et al.*, 2016; Stockwell *et al.*, 2016). Mitochondrial markers and genomic (microsatellite) marker panels have been developed for several freshwater and marine fish species, useful

in stock characterization across their natural range of distribution, which have applications in conservation as well as selective breeding programs. Large-scale Single nucleotide polymorphism (SNP) were recently identified in riverine populations of *L. rohita*, which have the potential to be valuable genomic resource.

Future Prospective and Action Points

Integrative multi-omics technologies need to be carried out for specialised, commercially and evolutionary important freshwater and brackish/marine species, for unique insights into the deeper fundamental biological understanding of their genomic architecture. These omics technologies may include genomics, transcriptomics, proteomics and metabolomics. This will facilitate effective genome mining for bioprospecting of novel genes and alleles from unexplored genetic diversity, for important production traits, stress (biotic and abiotic) tolerance and their biomarkers. The mechanism of osmoregulation in response to long term salinity challenge, the integrated analyses of metabolomics and transcriptomics has been successfully studied (Qin *et al.*, 2022). These technologies also help in overcoming the diseases in aquaculture, by studying gene expression profiles in combination with immune-related biosynthetic pathways during host-pathogen interactions. Innovative multi-omics strategies can also be formulated to discover natural products, especially the marine metabolites and novel molecules with biological activity, which may have the potential for converting them into commercial products.

For application of Integrative multi-omics technologies, consortium mode research would prove to be the pre-eminent way to operate data intensive projects, where the pooling of expertise from different domains can provide paramount output for public goods.

Indian major carps (IMCs) are the backbone of Indian freshwater aquaculture and Ganga basin is the home of their origin. Extensive genomic resources of IMCs need to be generated for different populations to identify selection signatures, to understand genome diversity and variations for the genetic basis for domestication and adaptation to diverse environments. Availability of genomic resources would allow to identify the genomics regions/genes under selection and their link to important production traits. This would lead to the development of tools for genomic selection, to be utilized for future selective breeding programs for increased production.

Genome information forms the basic information for various genome editing technologies, which has potential to transform the aquaculture and trade. Most of the gene editing research is focussed on economic traits, like growth, disease resistance, sterility etc. It looks possible in future, to design a tailor-made fish through genome editing technologies, as per the needs of aquaculture trade or consumer preference, like skin colouration and pigmentation, nutritional profile, meat quality, texture etc.

Acknowledgements

The authors thank Dr Trilochan Mohapatra, Secretary, DARE and Director General, ICAR; Dr Joykrushna Jena, DDG (Fisheries Science), ICAR, New Delhi; and Dr Kuldeep K. Lal, Director, ICAR-NBFGR, Lucknow for their support and guidance.

References

- Garner BA, BK Hand, SJ Amish, L Bernatchez, JT Foster, KM Miller, PA Morin, SR Narum, SJ O'Brien, G Roffler, WD Templin, P Sunnucks, J Strait, KI Warheit, TR Seamons, J Wenburg, J Olsen and G Luikart (2016) Genomics in Conservation: Case Studies and Bridging the Gap between Data and Application. *Trends Ecol. Evol.* **31**(2): 81-83.
- Hallerman E (2021) Genome editing in cultured fishes. *CABI agric. biosci.* **2**(46): <https://doi.org/10.1186/s43170-021-00066-3>
- Lu G and M Luo (2020) Genomes of major fishes in world fisheries and aquaculture: Status, application and perspective. *Aquac. Fish.* **5**(4): 163-173.
- Mohindra V, LM Chowdhury, N Chauhan, RK Maurya and JK Jena (2021). Transcriptome analysis revealed hub genes for muscle growth in Indian major carp, *Catla catla* (Hamilton, 1822) *Genomics* <https://doi.org/10.1016/j.ygeno.2022.110393>.
- Murali S, A Pathak, I Rashid, R Kumar, M Singh and B Kushwaha (2019) Genomic Resources for Fisheries Research. *Fish. Chimes* **39**(5): 34-35.
- Okoli AS, T Blix and AI Myhr *et al.* (2022). Sustainable use of CRISPR/Cas in fish aquaculture: the biosafety perspective. *Transgenic Res.* **31**: 1–21.
- Qin, Hui, Yu Zhaolong, Zhu Zongxian, Lin Yilong, Xia Junhong and Jia Yongxia (2022) The integrated analyses of metabolomics and transcriptomics in gill of GIFT tilapia in response to long term salinity challenge. *Aquac. Fish.* **7**(2): 13-139.
- Roy A, R Basak and U Rai (2017) *De novo* sequencing and comparative analysis of testicular transcriptome from different reproductive phases in freshwater spotted snakehead *Channa punctatus*. *PLoS ONE* **12**(3): e0173178.
- Stockwell BL, WA Larson, RK Waples, *et al.* (2016) The application of genomics to inform conservation of a functionally important reef fish (*Scarus niger*) in the Philippines. *Conserv. Genet.* **17**: 239–249.

Reservoirs Fisheries Development and its Trade-offs with Conservation of Natural Fish Genetic Resources

Vasu Sugunan¹ and VVR Suresh²

¹Assistant Director General (Rtd.), Indian Council of Agricultural Research, New Delhi-110001, India

²Head-in-Charge, Mariculture Division, ICAR-Central Marine Fisheries Research Institute, Kochi-682018, Kerala, India

The reservoirs spread across India are ideal water resources for fish production at very low input costs through environmentally safer and socially equitable manner. Nevertheless, the present fish production from the reservoirs is about one-third of its estimated potential. Enhanced capture fisheries, culture-based fisheries and recently cage culture systems are being implemented to bridge this gap, where stocking or artificial recruitment is the essential management tool. The stocked fishes can, at some stage or other, interfere with the natural ecosystem processes and have trade-offs with conservation of natural fish genetic resources, mainly of the rivers on which the reservoirs are built. Though it is difficult to prescribe a totally safe stocking protocol from a genetic point of view, scientifically sound strategies need to be put in place to minimize the adverse impacts of stocking. This paper focuses on the potential adverse impacts of stocking and suggests means that can help minimize its adverse impacts on the fish genetic resources.

Introduction

Reservoirs are “man-made impoundments created by obstructing the surface flow by erecting a dam of any description across a river, stream or any watercourse”. However, such water bodies, if less than 10 ha in area, have been excluded from the purview of this definition (Sugunan, 1997). Based on the area, the reservoirs are classified as small (<1000 ha), medium (1000 to 5000 ha), and large (>5000 ha) by the Government of India for the purpose of fisheries management (Srivastava *et al.*, 1985; Sugunan, 1995). India has about 20,000 reservoirs, covering 3,460,301 ha area. In terms of number, reservoirs are predominantly distributed in Tamil Nadu, Karnataka, Andhra Pradesh, and Odisha, while by area, the predominance is in Madhya Pradesh, Andhra Pradesh, Karnataka, Tamil Nadu, Uttar Pradesh, Gujarat, and Maharashtra. Besides these, many reservoirs are either under construction or in the project stage. India presently produces 14.15 million tonnes of fish annually and by 2025, the demand for fish is expected to touch 20 million tonnes. The bulk of this additional production needs to emanate from the inland segment as marine fisheries have plateaued already. As no substantial increase in fish production is expected from rivers, estuaries, backwaters, and coastal lagoons in the country due to a number of reasons, the reservoirs dotting

the length and breadth of the country need to be tapped for meeting our fish production targets.

The fish fauna of reservoirs basically represents the species present in the river on which they are constructed. Indian reservoirs support relatively rich fish diversity; harbouring not less than 60 species, of which about 40 species are contributing to commercial fisheries (Sugunan, 1995). The Indo-Gangetic carps generally dominate the commercial fisheries of Indian reservoirs. The Indian major carps, catfishes, feather backs, air-breathing fishes, and minnows are the most widely distributed fishes in Indian reservoirs. The average fish yield from reservoirs has been estimated at 110 kg/ha/year (Sharma and Suresh, 2013), while the category-wise mean fish yield has been estimated at 190 kg/ha/year from small reservoirs, 98 kg/ha/year from medium reservoirs and 34 kg/ha/year from large reservoirs against the potential yields of 500, 250 and 150 kg/ha/year from the respective categories. The present production from all categories of reservoirs is estimated at 386,259 tonnes, which is only about one-third of its potential of 1,022,062 tonnes. To bridge this gap, various forms of enhancements are being implemented in the reservoirs.

Enhancements

Enhancements in general are limited technical interventions in existing aquatic resource systems, which

*Author for Correspondence: Email-vasu.sugunan@gmail.com

can substantially alter the environmental, institutional, and economic attributes of the system (FAO, 1997). The common enhancement practices followed in reservoirs are enhanced capture fisheries and culture-based fisheries. The medium and large reservoirs are utilized as enhanced capture fisheries, while the small reservoirs are utilized as culture-based fisheries. Recently, enclosure culture systems such as cage and pen culture, involving artificial feeding are also being popularized in Indian reservoirs. The main accent of enhanced capture fisheries is augmenting the natural fish stocks and conserving them through encouraging natural recruitment and growth. Stocking is an integral part of enhanced capture fisheries as a measure to build up initial stocks of breeding populations along with periodic re-stocking to compensate for recruitment failure or stock losses. Introductions are resorted to for correcting imbalances in the species spectrum. Stocking and recapturing at regular intervals (often annual) are the basic strategy followed in culture-based fisheries, where the stocked fishes are recaptured before they breed within the water body. Selection of species for stocking, determining the size of seed at stocking, stocking density, growing time, and size at recapture are the key management decisions to be taken for culture-based fisheries. Principles of culture-based fisheries are akin to extensive aquaculture, although the fishes are grown in open water systems without artificial feeding or fertilization of water. Available examples in India suggest that the unit cost of fish production in culture-based fisheries, involving Indian major carps can be as low as Rs. 8.2/kg, with a benefit-cost ratio of 3.67. The benefit-cost ratio involving *Macrobrachium rosenbergii*, in culture-based fisheries, can be as high as 5.3 (Kutty *et al.*, 2008). These are way better than their production through aquaculture.

Stocking – the Key to Reservoir Management

Reservoir, as an ecosystem, includes both natural and man-made features and the fisheries management involves capture fisheries, enhancement, and enclosure aquaculture. Stocking or artificial recruitment is an essential management tool deployed in both enhancement practices and enclosure aquaculture. Obviously, fish stocks inducted into the ecosystem from extraneous sources can cause changes in the recipient system and its fish community (Mbabazi *et al.*, 2004). Reservoirs, being a continuum of the riverine ecosystem, the stocked fish, at some stage or other, will have an unavoidable interface with natural ecosystem structure and processes,

mainly of the rivers. Hence it is difficult to prescribe a totally safe stocking protocol from a genetic point of view. At the same time, being a resource of colossal size, with high untapped production potential and very low cost of production, utilization of this resource cannot be ignored while formulating the development agenda. This paper focuses on the potential adverse impacts of introductions and stocking and suggests means that can minimize (not totally neutralize) the adverse impact of stocking as a management tool on fish genetic stocks.

Species Stocked

Indian major carps (*Labeo catla*, *L. rohita*, *Cirrhinus cirrhosus*) have been the mainstay of stocking in reservoirs in India. They do not generally breed in the reservoirs outside their natural distribution and do not seriously compete with other indigenous fish species (Khan *et al.*, 2015), and are thus ideal candidate species for stocking. Examples of successfully involving shellfish species in culture-based fisheries in India are *M. rosenbergii*, in Malampuzha reservoir in Kerala (Kutty *et al.*, 2008), and in Ukai reservoir, Gujarat without noticeable environmental impacts.

Impact on Native Fish Genetic Resources

Species that got introduced or stocked into a reservoir can lead to a range of impacts on the native species. This may be in the form of competition, predation, parasitism, niche modification, etc. The inducted species establishes itself if it can overcome resistance from the natives and the resultant ecological conditions disturb the indigenous fishes. Stocking for culture-based fisheries in small reservoirs is considered to cause a lesser impact as most of these water bodies dry up during summer or the water level becomes too low allowing complete harvest. Fish stocks are not allowed to breed in the system and the whole process is more akin to culture fisheries. Hence chances of contaminating the wild natural fish stocks are relatively less. The enhanced capture fisheries, however, present a different scenario altogether as the system has to co-exist with natural ecosystem processes, and fisheries management norms are in the lines of capture fisheries.

Thus, stocking for enhanced capture fisheries can lead to a higher adverse genetic impact on the natural stocks. The introduction of common carp in 1957 into the Dal Lake in Kashmir has altered the energy flow of the ecosystem to the disadvantage of native Snow Trouts. In Gobindsagar Reservoir, common carp has adversely

affected the fishery of *C. cirrhosus* and *C. reba*. Silver carp was introduced in India, largely for aquaculture, guided by the hope that being a phytoplankton feeder it can effectively utilize the phytoplankton, a vacant niche in culture systems. The fish accidentally escaped into Gobindsagar Reservoir and established there at the cost of *L. catla*. Silver carp has also affected the population of *L. rohita* and *Osteochilus waandersii*. At present, commercial catches of Gobindsagar reservoir have three exotic species (Common carp, Grass carp, and Silver carp). The story of Tilapia is well known; it has well-established breeding populations in most of the reservoirs in peninsular India. They have brought down the population of *L. kontius* in Vaigai reservoir and *Hypselobarbus dubius* in Amaravati reservoir. There are reports of the predatory African catfish (*Clarias gariepinus*) being established in reservoirs. A sizeable proportion of stocking material used in Dahod reservoir in Madhya Pradesh and Pahuj reservoir in Uttar Pradesh involved *Ctenopharyngodon idella* (25 to 40%) and *Cyprinus carpio* (10%) (Sharma and Suresh, 2013). Stocking of *C. carpio* has been in practice in several other reservoirs too despite it being among the world's worst 100 invasive species (GISD, 2019).

Stocking of Indo-Gangetic carps in peninsular reservoirs in the past did not make a significant adverse impact, mainly since they by and large did not breed in the recipient ecosystem. As the only exception, *L. catla*, stocked in Stanley and Sathanur reservoirs (Cauvery river system), and Ukai reservoir (in Narmada), has been established in the systems and formed a fishery (Sreenivasan 1984; Sugunan, 1995). However, in most reservoirs in the peninsular rivers (Nagarjunasagar, Bhavanisagar, Krishnagiri, Malampuzha and Peechi), the Gangetic carps failed to breed (Sugunan, 2010).

Enclosure Culture

Enclosure aquaculture is fast picking up pace in India (Sugunan, 2015). Nearly 16,000 cages have already been installed across various states in different water bodies and reservoirs. (<https://www.newindianexpress.com/opinions/2022/may/23/cage-culture-for-fisheries-holds-promise-2456939.html>). Species used in these cages are *Pangasianodon hypophthalmus*, mono-sex Tilapia (GIFT variety), *L. rohita* (Jayanti variety), *Puntius brevis*, *L. bata*, *Lates calcarifer*, *C. carpio*, and the freshwater prawn, *M. rosenbergii*. As enclosure culture systems in reservoirs mainly involve exotic species, the risks

involved in terms of genetic and ecological impacts are far higher compared to enhancements. Cage aquaculture is practiced mainly in larger reservoirs where enhanced capture fisheries are followed, hence the chances of the introduced species entering the natural ecosystems and possibilities of their interaction with other biotic communities are higher. Almost all the fish species cultured in cages carry higher risks of adverse impacts on native species. Accidental escape of fishes stocked in the cages to natural waters is not completely avoidable. Cage farming is expected to have other environmental impacts too due to the use of artificial feed, antibiotics, and other irrational practices for gaining revenue, which are potential threats to native fish species.

Cage culture is being pursued aggressively in reservoirs of Andhra Pradesh, Telangana, Karnataka, Bihar, Jharkhand, Orissa, Maharashtra, Chhattisgarh, Tamil Nadu, Rajasthan, and Uttar Pradesh. Cage culture needs to be practiced by following the best management norms to ensure environmental protection and to avoid adverse impact on native species. The selection of water bodies for cage culture and determination of the number of cages to be installed need to be taken after assessing the carrying capacity of the water body in question. Similarly, only mono-sex fish seeds are allowed for stocking in cages, especially when exotic and genetically selected species like GIFT tilapia are involved. All stocking materials used in India are hatchery produced, mostly by private hatcheries without following any certification or guarantee procedures to ensure quality. In most cases, non-targeted or undesired species get mixed up with Indian major carp seeds used for stocking. Many private hatcheries follow mixed spawning of Indian and exotic major carps, which results in hybridization and genetic contamination of seeds that are sold from the hatcheries. Such seeds, if stocked in reservoirs, can play havoc with the genetic integrity of natural riverine fish fauna. All the brood stock and seed in nursery stages are raised on artificial feeds, possibly containing growth and disease resistance enhancing drugs. The environmental and economic consequences of the escape of these stocks into natural waters are yet to be completely ascertained. Though appropriate guidelines have been promulgated by the Government of India for cage culture in reservoirs (DAHDF, 2016), the monitoring and control mechanism for compliance is rather weak.

Suggestions

- Culture-based fisheries should be practised in small shallow reservoirs, preferably where near-total, annual harvesting is possible. Small reservoirs dry up during summer or water level drops drastically allowing near total harvests. This allows an ‘annual stocking-harvesting system’ that minimizes the genetic mixing up of stocked and natural stocks.
- In enhanced capture fisheries, stocking exacerbates the genetic contamination of wild stocks. As far as possible, native species should be selected for stocking.
- The seed for stocking should be produced by breeding wild-caught parents. The farm-produced fish seed with genetic depression (especially the seed produced through mixed spawning) should be strictly avoided.
- Exotic fishes of any kind should be totally banned from stocking in reservoirs, especially in large and medium reservoirs, where enhanced capture fisheries are the option.
- Exotic fish seeds when used in cage culture, should be of mono-sex so that they do not get a chance to breed if they escape from cages.
- Certification and monitoring of hatcheries and implementation of ‘good hatchery practice’ need to be made mandatory.
- Reservoir fisheries in India being a State subject, the State Governments have to play the lead role in improving governance and monitoring the environment by framing and implementing appropriate policies.

Reference

- DAHDF (2016) Guidelines for Cage Culture in Inland Open Water Bodies of India. National Fisheries Development Board, Hyderabad. 14p.
- FAO (1997) Aquaculture development. Technical guidelines for responsible fisheries, No. 6. FAO, Rome.
- GISD (2019) Global Invasive Species Data Base. <http://www.iucngisd.org/gisd/about.php> (Accessed on 5 July 2022)
- Khan MF, P Panikkar and AP Sharma (2015) Modelling the food web for assessment of the impact of stock supplementation in a reservoir ecosystem in India. *Fish. Manag. Ecol.* **22**: 359–370.
- Kutty MN, C Mohanakumaran Nair and KR Salin (2008) Reservoir fisheries of freshwater prawn– success story of an emerging culture-based giant freshwater prawn fishery at Malampuzha dam in Kerala, India. *Aquaculture Asia*, April-June 2008, 40.
- Mbabazi Dismas, Richard Ogutu-Ohwayo, SB Wandera and Y Kiziito (2004) Fish species and trophic diversity of haplochromine cichlids in the Kyoga satellite lakes (Uganda). *Afr. J. Ecol.* **42**(1): 59-68
- Sharma, AP and VR Suresh (2013) Strategies for inland fisheries resources enhancement in India and their impacts: A review. *In: Recent Advances in Culture Based Fisheries in India*. Pp. 22–40. (Eds. VV Sugunan, AP Sharma, and BC Jha). Hindustan Publishing Corporation (India), New Delhi.
- Sreenivasan A (1984) Influence of stocking on fish production in reservoirs of India. *Fishing Chimes* **13**(1): 18-21.
- Srivastava, UK, DK Desai, VK Gupta, SS Rao, GS Gupta, M Raghavachari and S Vatsala (1985) *Inland Fish Marketing in India-Reservoir Fisheries*. Concept Publishing Co. New Delhi.
- Sugunan VV (1995) *Reservoir Fisheries in India*. Fisheries Technical Paper No. 345. FAO, Rome.
- Sugunan VV (1997) Fisheries management of small water bodies in seven countries in Africa, Asia and Latin America. Technical Circular No. 933, FAO, Rome.
- Sugunan VV (2010) Inland fisheries resource enhancement and conservation in India, pp 35-60. *In: Miao Weimin, Sena De Silva and Brian Davy (ed). Inland Fisheries Resource Enhancement and Conservation in Asia*. Food and Agriculture Organization of the United Nations, Regional Office for Asia and the Pacific, Bangkok.
- Sugunan VV (2015) Enclosure aquaculture in inland waters of India. *In: Souvenir of 5th International Symposium on Cage Aquaculture in Asia – CAA5*. Pp. 41–54. (Eds. B Ignatius and J Imelda). Asian Fisheries Society, Malaysia.

Geo-Spatial Tools for Science-Based Management of Inland Aquatic Habitats and Conservation of Fish Genetic Resources

P Krishnan^{1*}, G Kantharajan², Rejani Chandran², A Anand³ and Vindhya Mohindra²

¹Bay of Bengal Programme Inter Governmental Organization (BOBP-IGO), Chennai-600018, Tamil Nadu, India

²ICAR–National Bureau of Fish Genetic Resources (NBFGR), Lucknow-226002, Uttar Pradesh, India

³Regional Remote Sensing Centre, National Remote Sensing Centre (NRSC), ISRO-Department of Space, Nagpur-440033, Maharashtra, India

Inland fisheries resources are vital for the human community and contribute to the development of a particular region. However, inland water bodies are highly dynamic, surrounded by terrestrial ecosystems, and face multiple threats from various anthropogenic stressors. Hence, the conservation and management of aquatic resources are becoming formidable which require the use of geo-spatial technologies for evidence-based scientific management of their habitats and fish genetic resources. The applications of geo-spatial tools are well acknowledged in data-scarce regions and in the field of natural resource management where the decision can be made based on the outputs arising from limited scientific assumptions. The remote sensing (RS) and Geographical Information System (GIS) based habitat characterization of inland water bodies, viz., inventory, monitoring of water spread and quality, habitat risk and suitability mapping, habitat prediction, and connectivity assessment is getting momentum in recent times, and incorporation of field exploration data with landscape information derived from geo-spatial tools enhance our understanding on aquatic habitats. The applications of geo-spatial models enhanced the potential of ecosystem service quantification and conservation planning in inland aquatic habitats. The demonstration of RS and GIS applications at a larger landform scale in inland open water bodies is necessary to illustrate its potential for science-based management of inland aquatic habitats for the conservation of fish genetic resources.

Introduction

Inland fisheries resources are vital for the human community as they not only act as food source and livelihood options but also contribute to the development of the entire region. The dependence of the human community on these resources has gradually increased due to growing demand from exploding populations around the world (Lynch *et al.*, 2016). The mismatch between the demand and supply from the natural inland water bodies led to the over-exploitation of its fishery resources. The inland water bodies are highly dynamic, surrounded by terrestrial ecosystems, and broadly classified into a lotic and lentic system based on the nature of the flow. Hence, the conservation and management of aquatic genetic resources are becoming formidable as they face multiple threats from various anthropogenic sources. The combined effect of these factors leads to the extirpation of certain fish species from a particular range of their distribution or even extinction.

The traditional habitat survey involves the inventory of resources, collection, recording, and analysis of multiple parameters at selected locations to understand the overall ecosystem status and health. Integration

and compilation of such datasets are mandatory for the extraction of information related to the spatio-temporal trend in habitat conditions and its association with other biological components. Further, visualising the scientific evidence at a spatial scale will help in convincing the policymakers for making the effective decision regarding fish genetic resource management.

The geo-spatial technologies play an important role in ecology, biodiversity, and conservation studies at large spatial and/or long-term temporal scales. Remote Sensing (RS) is the process of detecting and monitoring the physical features of an area at a distance and the Geographic Information System (GIS) is a computer system that analyses and visualises information that is attached to a particular location (<https://www.usgs.gov>). GIS effectively addresses problems that are inherent to a spatial analysis by effectively collating, archiving, displaying, and modelling spatio-temporal data. These tools are essential for inventory and mapping of the biological resources and monitoring the ecosystem components and health due to their simplicity and ability to answer user-defined queries (Ingole *et al.*, 2015). The applications of geo-spatial tools are well acknowledged

*Author for Correspondence: Email-krishnanars@yahoo.com

in data-scarce regions where the decision can be made based on the outputs arising from limited scientific assumptions (Anand *et al.*, 2020). The limited data on habitat, fish diversity and distribution information from field surveys and published literature linked with open source landform data such as Digital Elevation Model (DEM) are helpful for characterisation of an aquatic ecosystem from a regional to global scale.

The availability of open-source remote sensing imageries accelerated the applications of geo-spatial tools in environmental management studies and led to a better understanding of the structure and functions of natural resources (Chaminé *et al.*, 2021). With the introduction of high spatio-temporal resolution datasets, the contiguous monitoring of the aquatic ecosystem is possible at various scales. This facilitated the evidence-based scientific management of the ecosystem for its conservation and sustainable utilisation. This paper reviews the potential applications of RS and GIS in the context of habitat characterization of inland aquatic systems for the conservation and management of fish genetic resources.

Habitat Inventory and Monitoring of Water Spread

The inland water bodies are known for their complex geographical settings, varying sizes, and high level of interaction with surrounding terrestrial ecosystems compared to their marine counterparts. Hence, the use of geospatial technologies is necessary for the identification of remotely located inland water bodies (Ingole *et al.*, 2015). Additionally, due to the availability of high spatial resolution imageries, the resource inventory and mapping of small-sized pools and water bodies with an extent <5 ha is achievable (Anand *et al.*, 2020). Surface water monitoring is possible due to the repetitive mapping of these waterbodies at selected time intervals which is also important to understand the ecological and hydrological processes in any water body. Understanding the water spread dynamics in lotic water through RS and GIS helps in identifying the permanent water bodies which act as refugia for fish genetic resources, especially during the non-monsoon months (Kantharajan *et al.*, 2022). The Sentinel-2 imagery (10 m resolution) based water map of Godavari and Pranhita River confluence generated using the 'Normalized Difference Water Index' is shown in Fig. 1. The RS and GIS-based monitoring of the anthropogenic degradation of lentic and lotic water bodies provides meaningful insight for the development of regional or national environmental management plans.

Monitoring of Habitat Quality

The habitat quality of inland waterbodies, *viz.*, wetlands, lakes, and rivers depend on multiple factors which include geographical, hydrological, and socio-economic development (Joshi *et al.*, 2022). For instance, the downstream habitat quality of the riverine ecosystem is linked to its upstream catchments. Further, the degraded habitat conditions at one segment along the river affect the flow of benefits to the downstream. Identification of such strategic locations through manual monitoring is cumbersome and involves engagement of lot of manpower and money. Further, the source identification of such degradation process requires a thorough understanding of the interaction between topography, land use, flow hydrology, and socio-economic settings of the region. The mapping of vegetation in open water bodies through spectral indices is essential to quantify the extent of spread and initiate appropriate measures. Likewise, qualitative assessment of siltation mapped through spectral index, *i.e.*, Normalised Difference Turbidity Index (NDTI) helps in prioritising sub-watershed for land-use management. The NDTI based qualitative assessment performed to assess the temporal variation in turbidity before and after lockdown associated with COVID-19 in river Ganga revealed a drastic reduction post lockdown (Garg *et al.*, 2020).

Mapping of deep pools based on consistent water presence

Deep pools are the unique habitats in the riverine ecosystem which are abode to fish species round the year and act as essential habitats for feeding and breeding activities. Traditionally, these habitats are identified with the help of information obtained from local fishermen and by conducting intensive habitat exploratory surveys. However, through RS and GIS techniques the area which holds water for the selected time period can be mapped through 'Water Presence Frequency' (WPF) approach (Kantharajan *et al.*, 2022).

The monthly water map of a selected eco-region is generated for twelve months using 'Normalized Difference Water Index' which are overlaid to delineate 'deep pools' with 100% WPF. These deep pools are expected to hold water throughout the year and can be monitored efficiently to ensure their habitat provision services to fish genetic resources.

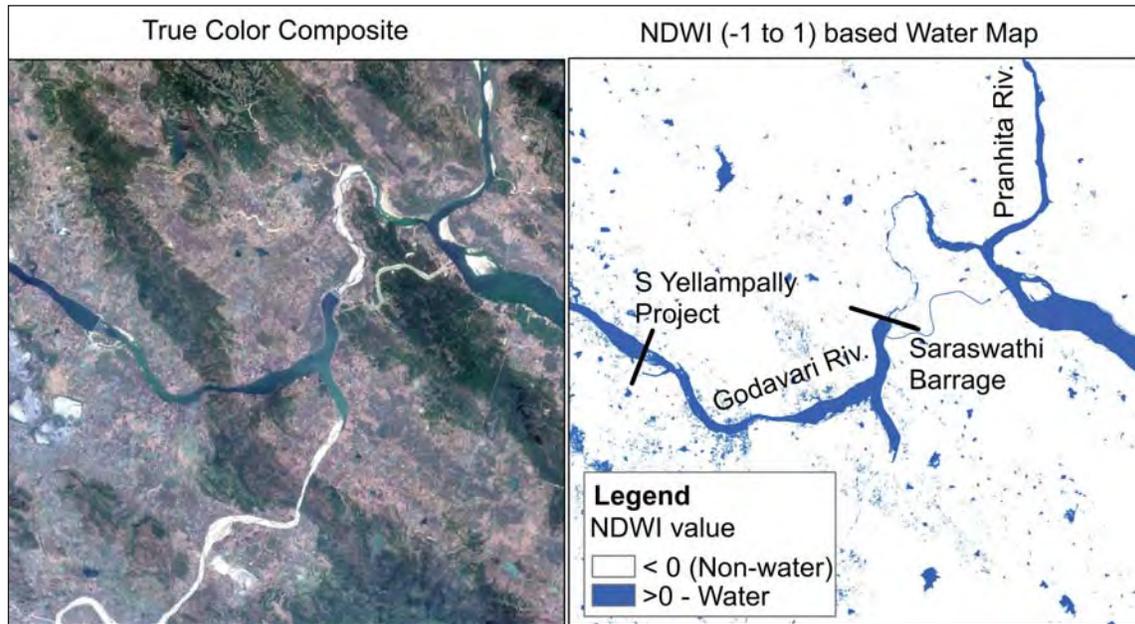


Fig. 1. Sentinel-based NDWI map depicting the water pixels along the Godavari-Pranhita River confluence at Kaleshwaram, Telangana

Further, the RS and GIS tools are also useful in evaluating the impact of invasive species on habitat degradation and the progress of restoration activities for improving the water quality. For instance, Choudhary *et al.* (2021) mapped the deteriorated habitat quality for bird species in Keoladeo National Park, India, a Ramsar Wetland, due to the land cover changes and spread of an invasive plant, *Prosopis juliflora* in the wetland.

Mapping of Habitat Risk

The inland water bodies are threatened by multiple anthropogenic stressors which affect their ecosystem structure and functioning. The quantification of risk arising from such stressors is essential for prioritising areas for restoration by considering their spatio-temporal exposure, intensity, and consequences (Sharp *et al.*, 2020). The RS-based data-driven geo-spatial models are in use for the risk assessment of such habitats by considering various aspects, *viz.*, water flow obstruction, land-use pattern, inflow from agriculture landscape, developmental activities, destruction of riparian vegetation cover, the spread of non-native invasive species, and the extent of fishing.

Mapping of Habitat Suitability

Fish species require optimum environmental conditions for their survival and growth thus, they choose habitats with stable environmental conditions and confine their

distribution to that particular geographical range. Though these factors which govern the suitability of habitats are often considered complex, it broadly covers topography, hydrology, water quality, and riparian features. Apart from this, the co-occurrence of niche species and non-native species also determines the habitat suitability for fish species. Thus, habitat suitability of any open water body including rivers can be mapped spatially considering the optimum physico-chemical habitat parameters required for fish species. Based on the suitability ranking and scores, the selected waterbody can be classified into different zones which enable easy monitoring and management. Further, the simulations of a particular parameter foresee the habitat supporting ability of the aquatic ecosystem in accordance with climate change and ongoing habitat degradation. Further, the species-specific environmental preference datasets coupled with geo-spatial models lead to mapping the specific habitat suitability map.

Vyas *et al.* (2013) performed the physical habitat assessment of river Denwa in Narmada Basin, India and developed a Habitat Suitability Map based on assigned scores to demonstrate its suitability to sustain aquatic biota. The habitat suitability of Pengba fish (*Osteobrama belangeri*) in Loktak lake, India, was evaluated for water quality parameters through geo-spatial models (Khwairakpam *et al.*, 2020).

Spatial Habitat Prediction and Assessment

The fish diversity and abundance data collected through exploratory surveys and field sampling from selected locations reveal partial information on fish distribution patterns at the landscape level. However, bringing this information into GIS platform along with other associated habitat data, *viz.*, topography, climate, flow hydrology, and other environmental factors is helpful in predicting the expected distribution range of the fish species through modelling tools. This is essential for the management of threatened or endangered fish species which are on the verge of extinction to bring back their populations to their original state.

On the other hand, the habitat prediction framework of Meixler and Bain (2012) classifies the river habitat based on stream size, stream quality in terms of land-use pattern, and optimum range of selected water quality parameters which are required for the survival of fish species and accordingly classify the rivers into different categories. Further, the fish species reported in the particular geographical area were classified based on stream size, stream quality, and water quality preferences and tolerance so that to predict the fish diversity in a classified river segment. The validated habitat prediction map generated through this framework revealed the potential of such framework in categorising fish habitat classes and predicting fish distributions at the landscape scale to facilitate better-informed management decisions.

Mapping Spatial Habitat Connectivity

Maintaining the connectivity between and within the inland water bodies is essential to preserve the fish diversity and to ensure the sustainable livelihood of the associated community in the eco-region (Kantharajan *et al.*, 2022). For instance, the water level, nutrient dynamics, and fish diversity of a wetland along the riparian corridor depend on its associated riverine ecosystem to which it is connected (Joshi *et al.*, 2022). Failure to protect this habitat connectivity, as well as the inability of fishes to move freely in them, may have a significant impact on fish populations, which will eventually disturb the socio-economics of dependent local communities.

Fish species, such as Hilsa, migrate from coastal regions to the river streams to complete their life cycle. Apart from this, there are numerous local migratory fish species, *viz.*, Mahseers and Catfishes which migrate from one stream to another stream for several purposes, and

especially for breeding and feeding. The free-swimming larvae originating from spawning habitats must reach productive nurseries to maximize survival. The spatial mapping of fragmentation, and connectivity assessment between habitat patches aid in the development of possible migration maps for fish species conservation.

The spatial data on landscape variables play an important role in 'Landscape Genetics', to derive meaningful conclusions by integrating with the population genetics of a fauna (Storfer *et al.*, 2008). This explores the possible influence of habitat fragmentations on the gene flow of a particular organism, intra- and inter-specific speciation and genetic diversity.

Mapping Ecosystem Services

The concept of ES is essential for justifying the arguments regarding the conservation and restoration of any natural ecosystem. The inland open waterbodies provide multiple ES, grouped under provisioning, supporting, regulation and cultural services. The GIS-enabled models are widely used for ES quantification and help the resource managers to visualise the spatio-temporal pattern and estimate the impact of projected LULC change, climatic conditions, and extent of management measures on the provision of services (Sharp *et al.*, 2020).

The geo-spatial model-based study conducted in the lake Hawassa watershed in Ethiopia revealed that the sediment export to the lake was mainly contributed by the cultivable lands while vegetation cover in the watershed retains sediment thus, maintaining the water quality in the lake (Degife *et al.*, 2021). The sediment retention potential of riparian vegetation shall be converted into monetary value to fix a 'payment for ecosystem services' to encourage farmers to carry out Best Management Practices in their cultivable lands.

Conservation and Restoration Planning

The purpose of designating 'Protected Areas (PAs)' is to conserve representative species, and habitats of importance from threat factors to ensure their persistence (Margules and Pressey, 2000). This process should be performed holistically coupled with habitat and anthropogenic factors. For instance, prioritisation of wetlands for implementing management measures requires integration of geospatial data from various sources including hydrology, topography, diversity and abundance of flora and fauna, surface water coverage, farming period, existing management factors and level of protection and anthropogenic stressors. Spatial datasets

on these factors can be overlaid on the GIS platform for prioritisation and the weightage of each factor can be decided based on its importance as per the policymaker's opinion and expert consultation. Further, RS and GIS offer scope for monitoring and evaluating the progress of conservation targets set for a particular species/habitat and track how these actions progress over time.

Another important role for GIS in fisheries resource management is involving the public in decision-making. Participatory GIS (P-GIS) aims to involve the public to generate and convert cognitive spatial knowledge into cartographic and descriptive information for spatially informed management decisions (Herlihy and Knapp, 2003). For instance, the capture of local and traditional knowledge on fish migration path, and behaviour and ecosystem services through P-GIS help in designating conservation measures.

Conclusion

The geographical setting and increased dependence of the human community on freshwater resources make them highly vulnerable and also disturb the equilibrium. Hence, spatial knowledge on their habitat characteristics, inherited biodiversity, and associated risk factors are immensely required for their conservation and management. The geo-spatial tools help in the analysis and visualisation of datasets which are essential for evidence-based resource management. Incorporation of field exploration data with landscape information derived from remotely sensed imageries in the GIS platform enhances our understanding of aquatic habitats. Further, the demonstration of RS and GIS at a larger landform scale is necessary to illustrate its potential for science-based management of inland aquatic habitats for the conservation of fish genetic resources.

References

- Anand A, P Krishnan, G Kantharajan, A Suryavanshi, P Kawishwar, U Raj, Ch Srinivasa Rao, SB Choudhury, C Manjulatha and DE Babu (2020) Assessing the water spread area available for fish culture and fish production potential in inland lentic waterbodies using remote sensing: A case study from Chhattisgarh State, India. *Remote Sens. Appl.: Soc. Environ.* **17**: 100273.
- Chaminé HI, AJ Pereira, AC Teodoro and J Teixeira (2021) Remote sensing and GIS applications in earth and environmental systems sciences. *SN Appl. Sci.* **3**: 1-3.
- Choudhary A, K Deval and PK Joshi (2021) Study of habitat quality assessment using geospatial techniques in Keoladeo National Park, India. *Environ. Sci. Pollut. Res.* **28**: 14105-14114.
- Degife A, H Worku and S Gizaw (2021) Environmental implications of soil erosion and sediment yield in Lake Hawassa watershed, south-central Ethiopia. *Environ. Syst. Res.* **10**: 1-24.
- Garg V, SP Aggarwal and P Chauhan (2020) Changes in turbidity along Ganga River using Sentinel-2 satellite data during lockdown associated with COVID-19. *Geomatics, Nat. Hazards Risk.* **11**: 1175-1195.
- Herlihy PH and G Knapp (2003) Maps of, by, and for the Peoples of Latin America. *Hum. Organ.* **62**: 303-314.
- Ingole NA, RN Ram, R Ranjan and AK Shankhwar (2015) Advance application of geospatial technology for fisheries perspective in Tarai region of Himalayan state of Uttarakhand. *Sustain. Water Resour. Manag.* **1**: 181-187.
- Joshi KD, AK Pathak, S Kumar, R Dayal, AK Singh, R Kumar, G Kantharajan, V Mohindra and KK Lal (2022) Fish community diversity assessment of protected Saraiyaman wetland in the Ganga River basin, India. *Community Ecol.* **23**: 63-73.
- Kantharajan G, A Anand, P Krishnan, RK Singh, K Kumar, AK Yadav, V Mohindra, SP Shukla and KK Lal (2022) Applications of Sentinel-2 satellite data for spatio-temporal mapping of deep pools for monitoring the riverine connectivity and assessment of ecological dynamics: a case from Godavari, a tropical river in India (2016–2021). *Environ. Monit. Assess.* **194**: 1-21.
- Khwairakpam E, R Khosa, A Gosain and A Nema (2020) Habitat suitability analysis of Pengba fish in Loktak Lake and its river basin. *Ecohydrol.* **13**: e2164.
- Lynch AJ, SJ Cooke, AM Deines, SD Bower, DB Bunnell, IG Cowx and TDJ Beard (2016) The social, economic, and environmental importance of inland fish and fisheries. *Environ. Rev.* **24**: 115-121.
- Margules CR, RL Pressey (2000) Systematic conservation planning. *Nature* **405**: 243–253.
- Meixler MS and MB Bain (2012) A GIS framework for fish habitat prediction at the river basin scale. *International Journal of Ecology* 146073.
- Sharp R, J Douglass, S Wolny, K Arkema and K Wyatt (2020) InVEST 3.10.2. post28+ug.ga4e401c.d20220324 User's Guide. The Natural Capital Project, Stanford University, University of Minnesota, The Nature Conservancy, and World Wildlife Fund.
- Storfer A, MA Murphy, JS Evans, CS Goldberg, S Robinson, SF Spear and LP Waits (2007) Putting the 'landscape' in landscape genetics. *Heredity* **98**: 128-142.
- Vyas V, A Kumar, V Parashar and S Tomar (2013) Physical habitat assessment of River Denwa using GIS techniques. *J. Indian Soc. Remote Sens.* **41**: 127-139.

Mariculture Development in India: Status and Way Forward

A Gopalakrishnan*, Bobby Ignatius and VVR Suresh

ICAR-Central Marine Fisheries Research Institute, Kochi- 682018, Kerala, India

Mariculture has immense potential in India in the context of fast-growing demand for seafood, which cannot be met by capture fisheries sector alone. Mariculture can also play an important role in increasing fish production from the coastal and offshore waters by involving local fishers and entrepreneurs to take up the activity. The pioneering attempts for mariculture development in the country by ICAR-Central Marine Fisheries Research Institute (CMFRI) on seed production and farming of finfishes such as cobia, silver pompano, Indian pompano, sea bass, groupers, snappers, breams and ornamental fishes, shell fishes such as molluscs, marine pearl production and seaweed farming, sea-cage farming, Integrated Multitrophic Aquaculture (IMTA), Recirculating Aquaculture System (RAS) and aquatic animal health management are highlighted and suggested strategies for way forward.

Introduction

Globally, aquaculture has emerged as a fastest growing food production sector with an annual growth rate of >6% in the last two decades. Mariculture is the fastest growing subsector of aquaculture and has very high growth potential. In 2020, mariculture contributed around 33.0 million tonnes of food fish, which formed about 27% of the global food fish aquaculture production. The total mariculture production including seaweeds was 68.1 million tonnes, which constituted 55.6% of the total world aquaculture production during 2020. Mariculture has immense potential in India in the context of fast-growing demand for seafood, which cannot be met by capture fisheries sector alone. The National Policy on Marine Fisheries (NPMF, 2017) clearly states that mariculture can play an important role in increasing fish production from the coastal waters and the government will address the Institutional and commercial needs of this emerging sector, which will include leasing rights policies, spatial planning, technological inputs such as husbandry, seed, feed and health management, environmental and social impacts as well as capacity building of local fishers and entrepreneurs to take up mariculture. The pioneering attempts for mariculture development in the country was initiated by ICAR-Central Marine Fisheries Research Institute (CMFRI) in the 1970s in Mandapam and Tuticorin with seaweed and bivalve culture, followed by attempts in induced maturation and breeding of Indian white shrimp in Njarakkal, Kerala and promoting semi-intensive shrimp farming in Kerala. Though India has a projected mariculture production potential of 4

to 8 million tonnes annually, the current mariculture production is less than 0.1 million tonne. However, the lessons learned from the successful expansion of inland and brackishwater aquaculture in India could be capitalized to boost mariculture production in a phased manner. This involves making the best use of available technologies such as seed production and farming of finfishes such as cobia, silver pompano, Indian pompano, sea bass, groupers, snappers, breams and ornamental fishes, shell fishes such as mussels, oysters, clams, green tiger shrimp and blue swimmer crab. Technologies for marine pearl production and seaweed farming are also available in the country.

Recent Developments

In recent times, the Government of India has taken several measures to streamline mariculture development in the country. Recognizing the prospects offered by the sector, the National Fisheries Development Board (NFDB) had constituted a committee with Director, CMFRI as the Chairman to prepare the Draft National Mariculture Policy (NMP). The Policy addresses various aspects such as mariculture area development, leasing and licensing, mariculture systems and species, environmental sustainability, seed and feed development, health management, certification, insurance, market support as well as institutional and legal framework. The Committee submitted its final report in November, 2019 and is presently under the consideration of the Ministry of Fisheries, Animal Husbandry and Dairying.

*Author for Correspondence: *Email- agopalkochi@gmail.com

Another major initiative to boost mariculture production is the establishment of National Brood-bank Facility for cobia and silver pompano by CMFRI. It is aimed to enhance the availability of quality seeds of cobia and pompano which are the most suitable species for sea-cage farming in Indian waters. CMFRI has established brood bank facilities at its Centres in Mandapam and Vizhinjam with a targeted production of 48 million silver pompano larvae and 30 million cobia larvae per year. The institute has also signed Memorandum of Understandings (MoUs) with seven Private/ State run facilities for technology transfer and seed production of these species. The All India Network Project on Mariculture [AINP (M)] is another major ongoing initiative to address technological constraints in mariculture. It is being headed by CMFRI and has five collaborating partners across the coastline. For development of seed production and farming technologies, ICAR-CMFRI has prioritized 76 species, which include marine finfishes (food and ornamental), shellfishes and invertebrates. Research efforts has so far yielded fully mature technologies for round the year seed production of six marine finfishes (cobia, silver pompano, Indian pompano, orange spotted grouper, pink ear sea bream and John's snapper) and 27 species of expensive marine ornamental species including five cross breeds.

Sea Cage Farming

Sea cage culture has made a beginning for intensive production of commercial finfish in coastal areas in the country. Since inception in 2007, many innovations on designing and fabrication of cages and mooring systems were made. Subsequently, demonstrations of cage farming were undertaken under a participatory mode with the help of local fishermen co-operatives and entrepreneurs. ICAR-CMFRI has developed and standardized guidelines and practises including Good sea cage farming practices for different regions in the country. Two different versions of indigenously fabricated 6 m diameter cages (GI and HDPE) developed by ICAR-CMFRI are presently being adopted by fishers. On an average, 2-3 tonnes of fish can be produced in a 6-meter diameter cage per cycle. The net economic return per crop (8 months/year) ranges from Rs. 1.5-2.5 lakh depending on the species grown. A major scheme on sea cage farming was operational with NFDB support in Maharashtra, Tamil Nadu, Kerala and Karnataka under the Blue Revolution Scheme of the Union Government

during 2017-2020 and under Pradhan Mantri Matsya Sampada Yojana (PMMSY) along Bahabalpur coast, Odisha during 2020 - 2022.

Suitable sites are to be demarcated for different mariculture activities such as cage farming, bivalve farming, pen culture, seaweed culture, hatcheries and nurseries based on scientific criteria and considering the socio-cultural attributes and other logistics. The potential zones for mariculture development should be identified based on the criteria developed through scientific evaluation of environmental parameters suitable for the type of farming, negligible impact on environment, avoidance of conflict with other users, protecting livelihoods of local fishing communities and ensuring their access to fishing grounds. Satellite remote sensing data and GIS can be employed to provide essential tools to support. Marine Spatial Planning (MSP) can be employed for data management, analysis, modelling and decision-making taking cognizance of CRZ zoning. The institute has identified and geo-referenced potential cage farming sites along the Indian coastline within 10 km (146 sites; 47384 ha) with a production potential of 2.13 million tonnes/year [based on the estimate that 15 cages (6 M dia.) can be accommodated in 1 ha, multiplied by the production figure of 3 tonnes/cage/year in an area of 47384 ha]. A planned massive programme to commercialize sea-cage farming will go a long way in realising the Blue Economy potential in India.

Integrated Multitrophic Aquaculture (IMTA)

The idea of bio-mitigation of the environment pollution along with increased biomass production integrating commercially important species of different trophic levels is emerging as an innovation in aquaculture. IMTA is the practice which combines appropriate proportions of the cultivation of fed aquaculture species (*e.g.* finfish/shrimp) with organic extractive aquaculture species (*e.g.* shell fish/herbivorous fish) and inorganic extractive aquaculture species (*e.g.* seaweed) to create balanced systems for environmental stability (bio-mitigation) economic stability (product diversification and risk reduction) and social acceptability (better management practices). This concept is being adopted in cage aquaculture wherein appropriate proportions of finfishes/shrimp with shell fish/herbivorous fish are integrated with sea weed farming. This system can mitigate the potential negative externalities of sea cage farming with simultaneous enhancement in seaweed yield. This

technique (16 rafts of 12ft×12ft size installed around a 6m dia. cage) has proven to enhance sea weed yield by about 122% in one cycle (45 days) of seaweed farming (additional 176 kg per cycle/raft of 12ft×12 ft size) with commensurate income enhancement. The technology is currently adopted by more than 150 farmers in Palk Bay region with the support of CMFRI.

On a global basis the search for additional areas to expand aquaculture to satisfy the growing local and export markets are necessitating an expansion of its activities farther off the coast. Mariculture is considered “offshore” when it is located > 2 km or out of sight from the coast, in water depths > 50 m, with wave heights of 5 m or more, ocean swells, variable winds and strong ocean currents. The economic, technological and sustainability issues of developing offshore mariculture in India needs to be explored.

Recirculating Aquaculture System (RAS)

Recirculating aquaculture systems (RAS) are onshore systems, in which fish can be grown at high density under controlled environmental conditions. Recirculation systems use land-based units to pump water in a closed loop through fish rearing tanks and consist of a series of sub-systems for regular water treatment process. These facilities can be used for both seed production and grow out of fishes in a strictly controlled environment even for exotic temperate species such as Atlantic salmon. CMFRI has successfully developed and adopted cost effective RAS facility for year-round maturation of broodstock and seed production and for nursery rearing of seven species of marine finfishes. Such RAS facility has been popularized among stakeholders in different coastal States.

Culture of Seaweeds

Seaweed farming offers immense scope as a livelihood opportunity and for developing a large number of by-products with several applications. Seaweed farming has the advantage of low capital input, as it is a primary producer requiring no inputs. Sea weed farming has picked up as an economically viable farming practice over the past decade on the shores of Palk Bay, Tamil Nadu in India. A floating system of 3.7×3.7 m rafts with a 45-day farming cycle for a total of 270 production days per year is being practiced by the self-help groups (SHG) in the region. Considered as one among the most environmentally benign activity, it has considerable potential to augment the livelihoods of coastal dwellers

in the country. A recent feasibility study conducted in the sea water inundated areas in South Andamans has revealed that these regions have huge potential for the enterprise. Similar studies need to be conducted in other suitable areas for enabling further spread of this promising livelihood activity. CMFRI has identified and geo-referenced potential seaweed farming sites along the Indian coastline within 10 kms (342 sites; 24167 Ha) with a production potential of 9.7 million tonnes (wet weight)/year [based on the estimate that 400 rafts (12ft×12ft) can be accommodated in 1 ha, multiplied by the production figure of 1 tonnes wet weight/raft/year in an area of 24167 ha]. Additionally, in future years, seaweed farming can earn carbon credits to the farmers. Farmed and wild collected seaweeds do not meet the current industrial demand for raw material. Large scale production of seaweed planting material through micropropagation and development of processing and marketing are highly essential for future expansion of seaweed farming in India.

Bivalve Farming

Mussel and oyster culture have gradually spread across the backwater belts of Kerala, Karnataka, Goa and Maharashtra owing to their high profitability. A number of methods such as stake culture, on-bottom culture, long-line culture, raft culture, rack culture, etc. are followed for mussel and oyster farming. Over 2000 farmers are practicing rack culture of green mussel in North Kerala, especially in Padanna estuary areas in Kasaragod, which is contributing to three fourth of green mussel production in India. CMFRI has established commercial farming of green mussels and edible oysters in coastal areas along the west coast of India with an annual production of over 10,000 tonnes benefitting nearly 6000 women self-help groups (production cost of Rs 90/kg versus with a farm-gate price of Rs 200/kg for green mussel; production cost of Rs 5/- per oyster versus with a farm-gate price of Rs 15/oyster). Though technology for mussel and oyster farming is fairly well available, what is lacking is adequate marketing and processing infrastructure. More number of entrepreneurs may be encouraged to take up these ventures by providing technological, financial, marketing as well as logistical support. Currently Indian bivalves are not exported to Europe, as the produce does not meet the monitoring protocols set by the EU. The focus here has to be on classification of bivalve growing water bodies following the regulations of the European Union (EU Directive 2006/113/EC). It is also necessary to

make a prospective (5 years) plan to improve hygiene in farming areas using international guidelines as a criterion and to conduct awareness campaigns for improving bivalve consumption in India. Although ICAR-CMFRI has developed the technology for cultured spherical and *Mabe* pearl production and pearl oyster seed production (hatchery technology), the high investment costs for these has been the limiting factor in the commercialization of these technologies. The gestation period (2½ to 3 years) in the attainment of appropriate size of the pearl oysters (suitable for surgery) is the major impediment in the commercialization process. Selective breeding of native stocks for faster growth and larger size and or production of triploid oysters – which can hold larger nucleus are the options.

Seed and Feed

To meet the additional requirement for seed of cultivable species in future, innovative measures need to be taken for establishing hatcheries, seed banks, rearing units and SPF/SPR/genetically improved brood banks. A system of seed certification can be developed by agencies concerned in order to ensure supply of quality seed. Financial and technical backstopping needs to be provided to establish hatcheries. Establishment of a few marine finfish brood banks is needed to provide fertilized eggs/newly hatched larvae to the hatcheries where further rearing and seed production can be carried out. Centres for the supply of fresh stock of fragments and import of germplasm of seaweeds after necessary quarantine also need to be set up.

The fish meal is the major protein component and determines the cost effectiveness of the feed. The increasing demand for the fish meal in poultry and shrimp farming sector results in steady increase of feed prices threatening economic viability of mariculture operations. Besides, global concern on use of fish meal produced from low value fishes which are consumed by people also force us to look for an alternative. Hence, replacement of fish meal with other protein sources of plant and animal origin needs to be explored without compromising the nutritional requirements of fishes grown in mariculture system. To ensure alternate protein sources for fish feed which is more sustainable, efforts are being made to develop feeds based on insect meal. Ongoing research efforts in this direction in CMFRI is focussed on fish feeds based on the larvae of black soldier fly (*Hermetia illucens*) which has the potential

for large scale culture. Further, efforts may be directed to develop species specific feeds including microfeeds meant for larval nutrition, which are customized to suit the growth patterns of individual prioritized mariculture species for efficient and economically viable production. As mariculture picks-up momentum, commensurate efforts are needed to establish decentralized fish feed production centres both under private management as well as in PPP mode to meet the demand for feed.

Aquatic Animal Health Management

In Indian mariculture scenario, most of the bacterial diseases are caused by opportunistic pathogens such as *Vibrio* sp., *Photobacterium* sp. and *Streptococcus* sp. resulting in significant economic losses. Similarly, the disease caused by *Perkinsus olseni* and *P. beihaiensis* is a major threat to bivalve farming. Diagnostic tools to detect these pathogens have been developed by ICAR-CMFRI. Fish farming favours infectious diseases and therefore requires investment in disease management. A National Surveillance Programme Aquatic Animal Diseases (NSPAAD) was launched in India under the leadership of the National Bureau of Fish Genetic Resources (NBFGR) during 2013 for five years funded by the NFDB to monitor aquatic animal diseases including those which infect marine finfish and shellfish species covering fourteen Indian states with passive and active surveillance in more than 100 districts covers. In this project, CMFRI has been actively involved in screening of bivalve pathogens such as *P. olseni* and *P. beihaiensis* along the west coast of India.

Green Certification of Marine Ornamental Species

CMFRI has successfully standardized the breeding and seed production of 27 high value marine ornamental fish species which are highly demanded in the international market. Four crossbreeds/designer varieties of *Percula* clown fishes have also been developed. The potential for the development of ornamental fish trade in India is immense, though it is still in a nascent stage. The Government of India has identified this sector as one of the thrust areas for development to augment exports. The turn of the century has seen a spurt in the collection, culture and trade in marine ornamental fishes. For the trade to prosper, the three pre-requisites are quality, quantity and sustainability. A large part of ornamentals is still collected from the wild thereby damaging the fragile coral ecosystem and hence awareness generation among local communities and stakeholders to desist from

unlawful and illegal practices of catching ornamental fishes from the wild are important. The trade should also encourage protection of the habitat for an eco-friendly approach. Green certification is the certification given to a product to ensure its environmental and socioeconomic sustainability. It ensures product quality, safety and traceability. Keeping in line with these concepts, CMFRI is currently preparing guidelines on Green certification on Indian marine ornamental fishes.

Marketing and Value Chain Development

Due to the seasonality involved in Mariculture production systems, chances are high for market glut during harvest seasons. The highly perishable nature of the harvested produce makes it necessary to develop efficient value chains that ensure timely clearance of harvested produce at minimum cost. Value chain of Mariculture produce starts from the farm and extends through an elaborate set of intermediaries such as auctioneers, wholesalers, commission agents, retail outlets and processors till it reaches the consumer. Efficient market logistics (insulated trucks/refrigerated containers, cold/chilled storage facilities, packaging, etc.) play a major role in ensuring the quality of the final product. Post-harvest processing and value addition also constitute an important link of the extended value chains of Mariculture produce. In this backdrop, the government need to take special initiatives to ensure the comprehensive development of Mariculture value chains. Infrastructure requirements for Mariculture produce marketing should be addressed on a priority basis. Necessary financial assistance, capacity building, technical guidance and institutional support are to be extended to the various stakeholders involved in the value chain.

Way Forward

There is growing recognition that mariculture is an emerging sector that will evolve with innovative technologies and inputs as it grows. It is envisioned that by 2050, India can produce about 4.1 million tonnes per annum of marine fish through cage culture alone if one per cent of the total coast line (8118 kms) can be utilized for mariculture. For this, the country has to develop dedicated mariculture parks as envisaged in the draft NMP, which would be managed by local fishermen groups/co-operatives/entrepreneurs. It is to be ensured that, the requirements of seed (about 2460 million) and feed (about 6.15 million tonnes) are to be commensurately ensured to meet the future demand from the sector. Sustainable development of mariculture would go a long way in making India a leading seafood producer in the world.

References

- Gopalakrishnan, A, R Kirubakaran, John George, AG Ponniah, G Gopakumar, Sunilkumar K Mohamed, Krishnan, Imelda Joseph, Boby Ignatius, AK Abdul Nazar, R Jayakumar, MS Raju, RA Sreepada, P Shinoj and N Rajesh (2019) Draft National Mariculture Policy 2019 (NMP2019); Report of the Committee constituted by the National Fisheries Development Board (NFDB), Ministry of Fisheries, Animal Husbandry & Dairying, Govt. of India. CMFRI Marine Fisheries Policy Series No.17/2020, 22p.
- NPMF, National Policy on Marine Fisheries (2017) Ministry of Agriculture and Farmers Welfare (Department of Animal Husbandry, Dairying and Fisheries), Government of India, The Gazette of India Notification, Extraordinary, Part 1, Section 1, No. 121, New Delhi, the 1st May, 2017, F. No. 21001/05/2014-FY (Ind) Vol. V.

Fishing Technologies for Conservation of Marine Fish Genetic Resources

CN Ravishankar^{1*} and VR Madhu²

¹ICAR-Central Institute of Fisheries Education, Mumbai-400061, Maharashtra, India

²ICAR-Central Institute of Fisheries Technology, Willingdon Island, Cochin-682029, Kerala, India

The advancements in fishing technologies over the years, had helped to significantly increase the fish production from the capture fisheries and has helped to improve the per capita consumption of fish. Increase in the fishing effort, use of unsustainable fishing gears and lack of proper monitoring and surveillance, combined with the climate related changes in the oceans, have caused serious declines of fisheries worldwide. The bycatch, discards and pollution are also becoming concern in several fisheries world over including India. Despite the fact that numerous measures, including gear-based technical measures, have been evaluated and mandated by law, the use and implementation of these measures in the field, remained largely negligible.

Ecosystem-based approaches to fishing, reducing plastic pollution, spatial planning and creating biodiversity hotspots with no-take zones, strict regulations and effective implementation strategies, as well as a concerted effort to reduce the capacity of fisheries, are required for conservation of resources.

Introduction

The global production of fish has quadrupled over the past fifty years as a result of advancements in fishing industry relating to navigation, fish finding, and the introduction of synthetic materials. This has contributed increased availability for consumption.

Although total fish output has increased, it is observed that fish harvests from the wild have stayed relatively constant at roughly 90-95 million tonnes per year since the early 1990s. Globally, the fraction of fish stocks that are overexploited has more than doubled since the 1980s; as a result, the current levels of wild fish capture are unsustainable. The fall in catches has been caused by a decline in fish availability, rather than changes in fishing effort (Thurstan *et al.*, 2010).

A large range of gears are employed to harvest fish resources in the wild, in both coastal and inland waters. Among these, bottom trawling accounts for approximately 25 percent of all the fish harvested, while other types of trawling account for approximately 10 percent. Just over twenty percent of the catch is contributed by pelagic fishing techniques such as purse seine being the most prevalent (Pauly *et al.*, 2020). The majority of gear used in inland waters consists of gillnets, small encircling nets, hooks, and lines that vary greatly in design and technique of operation. In addition to illicit fishing techniques, climatic and land use changes made

a substantial impact on the fish diversity over the years (Miranda *et al.*, 2022).

The Indian Scenario

During 2019-20, India produced a total of 14.16 million metric tonnes (MT) of fish, which accounts for around 7.6 percent of the total global fish production. Fish and fish products are also a significant source of revenue from exports, generating around \$6.73 billion.

Marine Capture

More than 1500 species of finfishes and shellfish inhabit the coastal and offshore waters of India and approximately 300 are commercially significant. Numerous gears are used to harvest marine resources, including trawl nets, gillnets, seines, hooks and lines, and bag nets. The principal resources contributing to marine capture fisheries include the Indian oil sardine, lesser sardines, Indian mackerel, croakers, Bombay duck, threadfin breams, carangids and ribbonfishes among finfishes and penaeid and non-penaeid shrimps and cephalopods among shellfishes. The current marine production of 3.8 million tonnes represents approximately 71.6 percent of the revalidated fishing potential of 5.31 million tonnes. During the recent stock evaluations, it was found that the 86 percent, out of the evaluated 67 marine finfish and shellfish were being fished within biologically sustainable levels, i.e., yields from these stocks have not yet reached Maximum Sustainable Yield (MSY).

*Author for Correspondence: *Email-ravishankar@cife.edu.in

Major Issues that Affect the Sustainability of Fishing Operations

Overcapacity

The global fishing capacity and effort grew fast from the late 1970s through 2010 before levelling off. In terms of both capability and effort, the Asian fleet is more than an order of magnitude greater than that of any other region, and it continues to grow. In recent years, developed nations as a whole have declined in both metrics and cause for stabilising the worldwide trend. Developing and underdeveloped nations continue to expand, with the former possessing the biggest fleet and exhibiting the highest relative growth. In terms of watt days of fishing effort per tonne of wild marine catch, the efficiency of the global fleet is presently lower than it was in 1950, despite significant technological developments and expansion throughout the world's oceans (Bell *et al.*, 2017; FAO, 2020).

Bycatch

Bycatch refers to non-targeted species that are retained, sold, or disposed for whatever reason. Incidental catches are the retained non-targeted species, whereas discarded catch refers to the fraction of the catch that is returned to the ocean for economic, legal, or personal reasons (Alverson *et al.*, 1994). The non-targeted component consists primarily of juveniles of economically valuable species and other charismatic species, such as turtles, sharks, etc., that have diverse life strategies and are frequently negatively impacted by capture and subsequent release from fishing gear or vessels. Recent global estimates of bycatch are 9.1 million tonnes, with the biggest contribution coming from shrimp trawl fisheries, contributing approximately 4.2 million tonnes (Pérez Roda *et al.*, 2019).

There are only a few studies that report the bycatch from fishing vessels along the Indian coast. Pramod (2010) estimated the bycatch discards from mechanised trawlers operating in Indian EEZ at 1.2 million tonnes. The same study estimated 56.3% of the total catch of shrimp trawlers as bycatch. Estimates by Dinesh Babu *et al.* (2013), show that landing of low value bycatch (LVB) in trawl fisheries has increased from 14% in 2008 to 25% in 2011. There are reports on the significant interactions of cetaceans in high sea gillnets (Anderson *et al.*, 2020) and in surrounding gears like ring seines.

Though there are many technical measures that are used worldwide (Kennelly and Broadhurst, 2021),

the adoption of these measures is found to be limited including the tropical countries, where shrimp trawls are widely used. The low adoption of technical devices is attributed to the resistance from the fishers towards the modification and inherent fear of commercial catch losses (Suuronen, 2022). Abandoned, lost, or otherwise discarded fishing gear (ALDFG) is another issue that is gaining importance due to the negative impacts of plastics in the ocean and by affecting the food chains and ultimately the biodiversity of the oceans (Gilman *et al.*, 2021).

Lack of Implementation of Management Measures

Despite the fact that many nations have implemented localised management measures for the conservation of resources, the implementation in the fishery is frequently hampered by numerous obstacles: effective monitoring, control, and surveillance is frequently a problem, especially in the south Asian nations where the number and scale of fishing operations are quite high. Recent studies however show that the implementation of the CCRF has increased, as also the utilisation of bycatch as secondary raw material or for other traditional purposes (Roda *et al.*, 2019).

Technologies for Responsible Fishing

Use of Bycatch reduction devices (BRD) can address the problem of capture of non-targeted catches, provided the devices have been tested in the fishery for optimizing the size, shape and operational parameters. Square mesh cod-ends, Fisheye BRD, Sieve Net BRD, Juvenile Fish Excluder cum Shrimp Sorting Device (JFE-SSD), etc. are some of the BRDs that were field-tested for their effectiveness in the Indian scenario (Kennelly and Broadhurst, 2021). Species that are at risk due to climate-induced changes such as sea turtles and other Endangered Threatened and Protected (ETP) species often form a bycatch in different gear systems like trawls and long lines. Field studies have shown the utility of Turtle Excluder Devices (TEDs) to release turtles getting in the way of trawl nets and, use of circle hooks, to reduce mortality associated with hooking.

Modified designs of gears to reduce negative impacts, are also widely used. Trawls that reduce impacts to the bottom, include semi-pelagic trawls, short belly trawls, cut-away trawl designs. Modifications to the anterior region of the trawl net and other significant changes to the designs are reported (Kennelly and Broadhurst, 2021). The outcome of these designs has been the reduction in

the non-targeted catches, either by preventing entry to the gear (which is more beneficial in reducing post-release mortality) or the easy release from the anterior parts of the trawls that again benefits in terms of reducing post-release mortality.

Use of bird scare lines and chutes, to set the baited hooks deeper are often employed in longline fisheries to reduce the incidental capture of seabirds. While regulation in mesh size, is the common intervention to reduce capture of juveniles in surrounding gears, use of medina panels and backdown techniques etc. are the interventions to reduce cetaceans being accidentally caught during seining operations. Deterrent devices such as pingers, though effective in reducing the incidence of cetaceans in fishing gears, subsequent reduction in effectiveness of the device is reported due to habituation to the sound by the animals.

Future Prospective

The following action points are recommended

1. Ecosystem-based approaches to fisheries management
2. Spatial planning and effective implementation of temporal and/or spatial regulations
3. Effective quotas for regulating the catch and bycatch in different fisheries
4. Creation of marine protected areas and regulation of all activities that directly or indirectly affect the biodiversity
5. Measures to decrease plastic pollution from fishing operations, including rigorous rules, marking of fishing gear, and measures to reduce runoff contamination into the oceans.
6. Understanding the intricacies of marine fisheries through strong collaboration with fishermen is crucial for the development and implementation of diverse approaches to prevent the loss of fish diversity.
7. Restrictions on the use of illegal fishing gears and fishing methods that cause severe damage to flora and fauna

References

- Bell, JD, AR Watson and Ye Yimin (2017) Global fishing capacity and fishing effort from 1950 to 2012, *Fish and Fisheries* **18**(4): 792-793.
- Charles Anderson, R, Herrera Miguel, D Anoukchika, Ilangakoon, KM Koya, M Moazzam, L Mustika Putu and N Dipani Sutaria (2020) Cetacean bycatch in Indian Ocean tuna gillnet fisheries, *Endanger. Species Res.* **14**: 39-53.
- Dinesh Babu AP, EV Radhakrishnan, S Thomas, G Maheswarudu, PP Manojkumar, SJ Kizhakudan and PB Sawant (2013) Appraisal of trawl fisheries of India with special reference on the changing trends in bycatch utilization. *J. Mar. Biol. Assoc. India* **55**(2): 69-78.
- Eric Gilman, Michael Musyl, Suuronen Petri, Chaloupka Milani, Gorgin Saeid, Wilson Jono, Brandon Kuczenski, (2021) *Nature* **11**: 7195, <https://doi.org/10.1038/s41598-021-86123-3>
- FAO (2020) The State of World Fisheries and Aquaculture. Sustainability in action. Rome, 2020. <https://doi.org/10.4060/ca9229en>
- Food and Agriculture Organization (2020) FishStatJ – Software for fishery and aquaculture statistical time series. FAO.
- Kennelly SJ and MK Broadhurst (2021) A review of bycatch reduction in demersal fish trawls. *Reviews in Fish Biology and Fisheries* **31**: 289-318. <https://doi.org/10.1007/s11160-021-09644-0>
- Pauly D, D Zeller and MLD Palomares (Eds), 2020. Sea Around Us Concepts, Design and Data (seararoundus.org).
- Pérez Roda MA, E Gilman, T Huntington, SJ Kennelly, P Suuronen and MP Medley Chaloupka (2019) A third assessment of global marine fisheries discards. FAO Fisheries and Aquaculture Technical Paper No. 633. Rome, FAO. 78 pp. Licence: CC BY-NC-SA 3.0 IGO.
- Pramod G (2010) Illegal, Unreported and Unregulated Marine Fish Catches in the Indian Exclusive Economic Zone, Field Report (TJ Pitcher, Ed), Policy and Ecosystem Restoration in Fisheries, Fisheries Centre, University of British Columbia, BC, Vancouver, Canada: 29 p.
- Rafael Miranda, Imanol Miqueleiz, William Darwall, Catherine Sayer, Nicholas K Dulvy, Kent E Carpenter, Beth Polidoro, Nadia Dewhurst-Richman, Caroline Pollock, Craig Hilton-Taylor, Ruth Robin, H Thurstan, Simon Brockington and Callum M Roberts (2010) The effects of 118 years of industrial fishing on UK bottom trawl fisheries. *Nat. Commun.* **15**: 1-15 doi: 10.1038/ncomms1013.
- Suuronen P (2022) Understanding perspectives and barriers that affect fishers' responses to bycatch reduction technologies, *ICAR J. Mar. Sci.* **79**(4): 1015-1023.

Indian J. Plant Genet. Resour. 35(3): 325–329 (2022)
DOI 10.5958/0976-1926.2022.00093.6

Boosting Widespread Adoption of Sustainable Agriculture – New Metrics and the Role of Science

Natalia Estrada Carmona^{1*}, Sunayana Sharma², Jai Rana² and Kuldeep K Lal²

¹*Bioversity International, Parc Scientifique d'Agropolis II, 34397 Montpellier, France*

²*Bioversity International, G-1, B-Block, NASC Complex, DPS Marg, Pusa Campus, New Delhi-110012, India*

³*ICAR-National Bureau of Fish Genetic Resources, Canal Ring Road, PO Dilkhusha, Lucknow-226002, Uttar Pradesh, India*

Adapting to new climatic, social, and environmental realities demands deep and massive transformative changes in how humans manage, perceive, and relate with terrestrial and aquatic productive systems. In India, agriculture production is challenged by degraded soils, scarce and contested water, fragmented and degraded seminatural habitats, social conflict, and more frequent extreme events. Existing political will is enabling the adoption of sustainable agriculture. However, the pace and the extent of the adoption of promising strategies, practices, and approaches for achieving sustainable and resilient agriculture remains sparse. Accelerating a socially just transformation to sustainability in India requires a new systems-oriented, multidisciplinary, human well-being-centered research agenda. Specifically, the new agenda can expand, contest and reevaluate agriculture performance' in terms of how is evaluated and measured at the farm and landscape level. The new evidence will be critical for learning, innovating and re-designing sustainable, multifunctional and resilient agricultural landscapes.

Introduction

All base resources that maintain human populations and the economy (e.g., soil, water, terrestrial and aquatic biodiversity) are under threat, and have degraded at an alarming rate and level, cascading into widespread environmental crises and social inequalities (Martin *et al.*, 2020). Therefore, radical societal and economic transformations are essential to achieve sustainability as committed to and envisioned (Pascual *et al.*, 2022). This transformation includes recasting how humans relate, manage, and envision the role of agriculture (production of food and non-food products, livestock, fisheries, and forestry) in achieving multiple Sustainable Development Goals and global commitments

Accelerating the transformation and widespread adoption of sustainable agricultural systems country-wide is, therefore, a tremendous task that some countries started to embrace. For example, the Indian government is committed to transforming its agricultural land, and better managing its terrestrial and aquatic genetic resources through multiple mechanisms; including multilateral environmental agreements, enacted legislation, national biodiversity targets, and concrete action plans (NBA, ICAR-Bureau and ABC, 2020). Yet, translating existing

political will into accelerated crosssectoral and multilevel action for redesigning sustainable agricultural farms and landscapes remains challenging.

Multiple alternative approaches and practices to conventional agriculture can play (and are playing) a critical role in achieving healthy terrestrial and aquatic systems (Gupta *et al.*, 2021, Jones *et al.*, 2021, Leal *et al.*, 2020). These alternative and sustainable approaches share some commonalities, such as a) integrating traditional and scientific knowledge, b) putting farmers' wellbeing at the center to redesign aquatic and terrestrial agricultural systems that mobilize less or null external input use (including water), c) embedding more agricultural biodiversity in the farm, and landscape level, and d) retaining and restoring remnant patches of seminatural habitat (Gupta *et al.*, 2021). Despite multiple lines of evidence, adopting these actionable alternative approaches and practices remains rare and conventional agriculture still dominates (Gupta *et al.*, 2021).

Conventional agriculture played a crucial role in overcoming the challenges during the 60s by converting India's food availability from scarce to abundant (Kumar foreword in Gupta *et al.*, 2021). Conventional agriculture

*Author for Correspondence: Email-n.e.carmona@cgiar.org

research, planning, and performance assessment remains heavily centered around yields thus neglecting impacts beyond the field (e.g. landscape or watershed level) and beyond the agriculture sector (e.g. human health, environment, economy). This myopic vision to assess the performance of agricultural systems brought about disastrous social and environmental consequences, which are poorly accounted for in current economic and political models. In India, the severity and extent of degraded soils, contested water use across sectors, depleted groundwater reservoirs, fragmented and degraded natural habitats, and environmental conflict raise the urgency to rapidly move from unsustainable to sustainable agriculture production (Fig. 1).

As such, the transition towards sustainable agriculture production demands an environment of political will at national and global level and a new research agenda that better captures the comparative advantage of multifunctional, sustainable, and resilient agricultural systems. Ideally, this new research agenda will facilitate identifying local and context-relevant solutions that particularly supports small, subsistence, vulnerable, and marginal farmers to thrive and flourish in healthy and resilient ecosystems (EcoNetwork, 2022). This will require systems-level, multidisciplinary, and human wellbeing -centered research (EcoNetwork, 2022). Likewise, the new research agenda should question and challenge how agriculture performance is measured including what gets measured or considered in performance assessments. Here we elaborate on two specific questions that can push the role of science in guiding decision-making and actions towards a socially just transformation to sustainability in India.

Future Prospective and Concrete Action Points

What gets Measured – Making the Invisible, Visible

Wild and domesticated biodiversity are the invisible engineers of terrestrial and aquatic ecosystems. However, wild biodiversity is rarely monitored in agricultural landscapes, and domesticated biodiversity is rapidly disappearing from our plates and fields, leaving humans with limited genetic resources for future use (Jones et al., 2021, Dulloo et al., 2021, Leal et al., 2020). Maintaining and fostering diversity is a critical principle in attaining resilience and adaptability (Biggs et al., 2015). Hence, efforts to better capture the spatial and temporal dynamics of terrestrial and aquatic genetic resources (e.g., multi-taxa assessments) in agriculture-

dominated landscapes are crucial for establishing the connection between sustainable agriculture / biodiversity/ ecosystem functioning/ resilience.

Agriculture is also understood to be much more, such as “good food,” “identity,” “happiness,” “tradition,” “autonomy”, “resilience”, etc. Hence, developing, testing, and validating new metrics that capture sustainable agriculture’s ignored but highly relevant contributions to human wellbeing is key. For example, a study in Brazil shows that integrated interventions contribute more, and positively to multiple locally relevant aspects of human wellbeing than siloed interventions (e.g. land sparing strategy) (Carmenta et al., 2022).

Similarly, agriculture’s spillover effect on other sectors are often ignored. For example, un-sustainable crop production is compromising the 9.71 million tonnes per year of inland fish production due to pollution, sedimentation, water abstraction, obstruction of river flows by dams, and impaired environmental flows, among other pressures (ICAR-NBFG and ABC, 2020). Hence, understanding how terrestrial and aquatic managed systems interact is also critical for promoting synergistic multisectoral (e.g. agriculture, natural resources, human health) efforts, and collectively contribute towards achieving multiple objectives; such as, biodiversity conservation, nutritious and sustainable crop, livestock, forestry, and fisheries production.

Overall, re-designing agricultural land for resilience, multifunctionality and sustainability will require long-term and country-wide monitoring efforts a) tracking species, varietal and landrace diversity in productive systems (terrestrial and aquatic), b) capturing agriculture impacts beyond material aspects, and c) accounting for spillover effects across sectors. A network of landscapes with contrasting adoption levels of sustainable agriculture practices will enable innovation, cross-learning and advancing quantifying the comparative advantage of multifunctional and resilient agriculture against business as usual.

How Aquatic and Terrestrial Productive Systems Performance is Measured

Agriculture performance is often assessed through simplistic or static metrics such as yields, which keenly fall short of capturing the wide range and long-term contributions to people and nature from multifunctional and sustainable agriculture (Tittonell, 2014). A wide

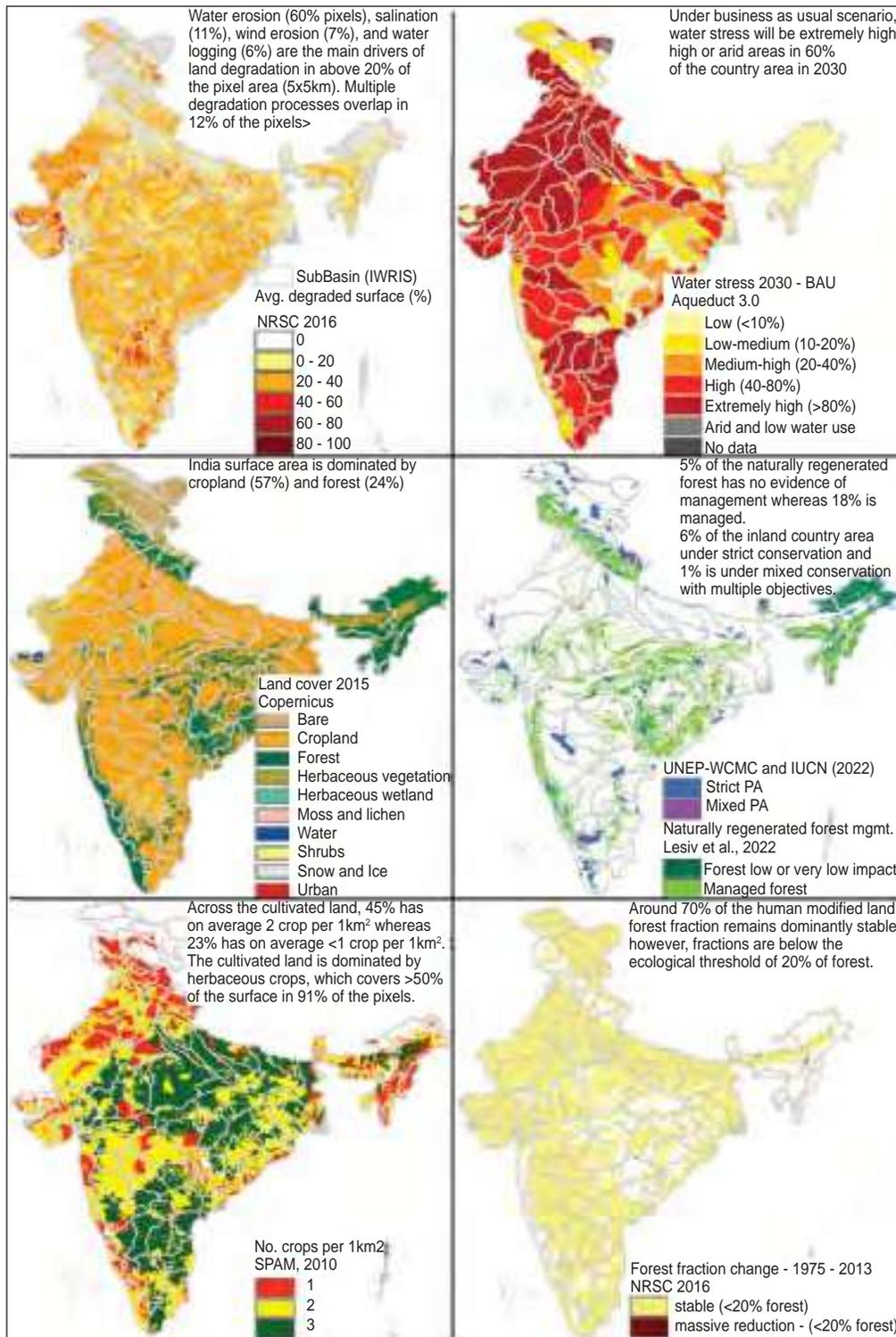


Fig. 1. Indian maps showing the severity and the urgency to move towards sustainable agriculture. In 2019, Indian agricultural land shared 4% of the global agricultural land area but contributed 11% of the global agriculture gross production and 16% of captured freshwater fisheries worldwide (India, 2nd largest global aquaculture producer) (FAOSTAT, *FishStatJ*, 2022). Social aspects are also critical but less commonly mapped, however, see in <https://ejatlas.org/> well-documented social-environmental conflicts often related to unsustainable agriculture. Hence, maintaining a viable economy and nourishing 17% of the global population without water, soil, biodiversity and in conflict with people and nature is a dead end.

range of promising holistic metrics exist but more innovation is also needed. Testing and uptaking the most adapted metrics will depend on an iterative, participatory, and truly collaborative process across sectors and stakeholders to test, validate and adjust holistic metrics to the different socio and agroecological contexts as well as national goals and global commitments.

For example, land equivalent ratios better capture combined production in diversified fields (Letourneau *et al.*, 2011), nutritional yield can capture nutritional contribution to humans (DeFries *et al.*, 2015), crop diversity can capture stability of national food production (Renard and Tilman, 2019) or contributions to wild-diversity (Sanchez *et al.*, 2022) to name a few. Other system-level metrics, although less developed, are also critical to measure the effectiveness of certain practices at the farm and landscape level in mitigating extreme events (e.g., hurricanes, Holt-Giménez *et al.*, 2002), or maintaining healthy terrestrial and aquatic wild and domesticated populations; along with the ecosystem services and resilience these communities provide (Gamez-Virues *et al.*, 2015, Feit *et al.*, 2021, Bailey and Buck, 2016).

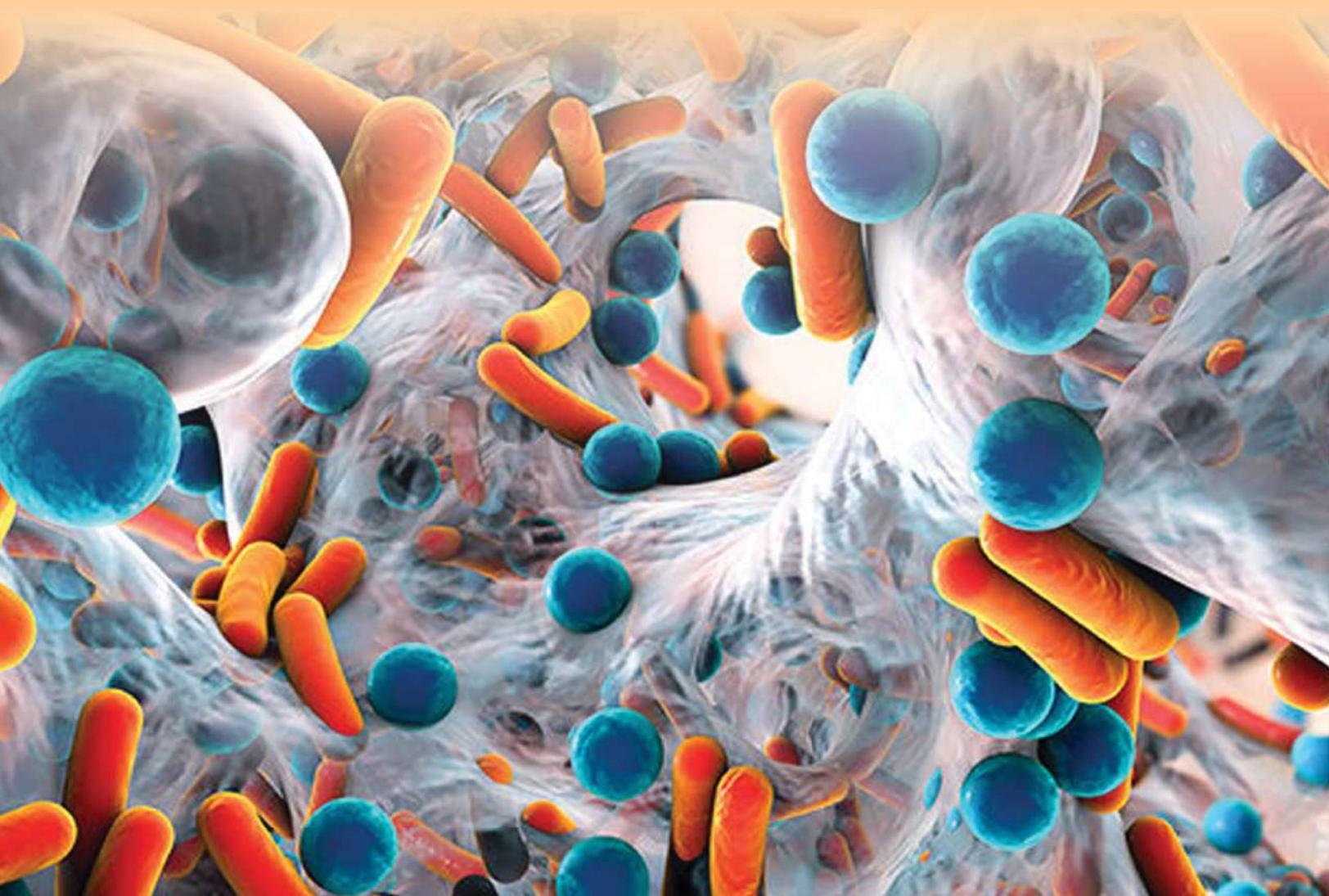
Overall, heading and maneuvering towards the de-sired socially just transformation to sustainability requires new holistic metrics that enable decision-makers to take timely, well-informed, and locally relevant decisions. How those, or new holistic metrics score India's efforts to transition remains an open question that a new research agenda could soon answer.

References

- Bailey Buck (2016) Managing for resilience: a landscape framework for food and livelihood security and ecosystem services. *Food Secur.* **8**: 477–490.
- Biggs, Reinette & Schler, Maja & Schoon, Michael (2015) Principles for building resilience: Sustaining ecosystem services in socio-ecological systems. 10.1017/CBO9781316014240.
- CCI Land Cover (2017b) Release of a 1992-2015 time series of annual global land cover maps at 300 m.^https://www.esa-landcover-647.cci.org/index.php?q=webfm_send/88
- Dulloo ME, NE Carmona, JC Rana, R Yadav and F Grazioli, (2021) Varietal threat index for monitoring crop diversity on farms in five agroecological regions in India. *Diversity*, **13**(11): https://doi.org/10.3390/d13110514
- EcoNetwork (2022) Sustainable agriculture and food systems in the Global South: An Indian Perspective.
- FAOSTAT statistical database (2022) Food and Agriculture Organization of the United Nations – FAO. Rome https://www.fao.org/faostat/en/
- Feit, *et al.* (2021) Landscape complexity promotes resilience of biological pest control to climate change. *Proc. R. Soc. B Biol. Sci.* 288.
- FishStatJ statistical database (2022) Fisheries and Aquaculture Division (NFI) of the Food and Agriculture Organization of the United Nations – FAO. Rome.https://www.fao.org/fishery/en/statistics/software/fishstatj/en
- Garibaldi LA, FJ Oddi, FE Miguez, I Bartomeus, MC Orr, EG Jobbágy, C Kremen, LA Schulte, AC Hughes, C Bagnato, G Abramson, P Bridgewater, DG Carella, S Díaz, LV Dicks, EC Ellis, M Goldenberg, CA Huaylla, M Kuperman, CD Zhu (2021) Working landscapes need at least 20% native habitat. *Conservation Letters* **14**(2): 1–10. https://doi.org/10.1111/conl.12773
- Gómez-Virúes, *et al.* (2015) Landscape simplification filters species traits and drives biotic homogenization. *Nat. Commun.* **6**.
- Gupta, Niti, Shanal Pradhan, Abhishek Jain, and Nayha Patel. (2021) Sustainable Agriculture in India 2021: What We Know and How to Scale Up. New Delhi: Council on Energy, Environment and Water.
- Holt-Giménez E (2002) Measuring farmers' agroecological resistance after Hurricane Mitch in Nicaragua: A case study in participatory, sustainable land management impact monitoring. *Agriculture, Ecosystems and Environment* **93**(1–3): 87–105. https://doi.org/10.1016/S0167-8809(02)00006-3
- Hofste R, S Kuzma, S Walker, EH Sutanudjaja, *et al.* (2019) “Aqueduct 3.0: Updated Decision-Relevant Global Water Risk Indicators.” Technical Note. Washington, DC: World Resources Institute. Available online at: https://www.wri.org/publication/aqueduct-30.
- International Food Policy Research Institute (2019) “Global Spatially-Disaggregated Crop Production Statistics Data for 2010 Version 2.0”, https://doi.org/10.7910/DVN/PRFF8V, Harvard Dataverse, V4
- ICAR-NBFGR & The Alliance of Bioversity and CIAT (2019-2020) Inland Aquatic resources of India and the ecosystem services management: status, issues and some policy suggestions.
- Jones SK, N Estrada-Carmona, SD Juventia, ME Dulloo, MA Laporte, C Villani and R Remans (2021) Agrobiodiversity Index scores show agrobiodiversity is underutilized in national food systems. *Nature Food*, **2**(September). https://doi.org/10.1038/s43016-021-00344-3
- Leal CC, GD Lennox, SFB Ferraz, J Ferreira, TA Gardner, JR Thomson, E Berenguer, AC Lees, A.C and RM Hughes (2020) Conservation of Tropical Aquatic Species. *Science*, **121**(October), 117–121.
- Lesiv M *et al.* (2022) Global forest management data for 2015 at a 100 m resolution. *Scientific Data* **9**(199): 1–14. https://doi.org/10.1038/s41597-022-01332-3
- Letourneau DK, I Armbrrecht, BS Rivera, JM Lerma, EJ Carmona, MC Daza, S Escobar, V Galindo, C Gutiérrez, SD López, JL Mejía, AMA Rangel, JH Rangel, L Rivera, CA Saavedra, AM Torres and AR Trujillo (2011) Does plant diversity benefit agroecosystems? A synthetic review. *Ecological Applications : A Publication of the Ecological*

- Society of America*, **21**(1): 9–21. <http://www.ncbi.nlm.nih.gov/pubmed/21516884>
- Martin A, MT Armijos, B Coolsaet, N AS Dawson, G Edwards, R Few, N Gross-Camp, I Rodriguez, H Schroeder, GL Tebboth M and CS White (2020) Environmental Justice and Transformations to Sustainability. *Environment* **62**(6): 19–30. <https://doi.org/10.1080/00139157.2020.1820294>
- NRSC (2016) Annual Cropland Data set 2013-14. NICES/DS(L)/LULC/1314/Jan2016.
- NRSC (2016) Indian Land Degradation Data set 2005-6. NICES/DS(L)/LD/2006/Jan2016.
- Pascual U, PD McElwee, SE Diamond, HT Ngo, X Bai, WWL Cheung, M Lim, N Steiner, J Agard, CI Donatti, CM Duarte, R Leemans, S Managi, APF Pires, V Reyes-García, C Trisos, RJ Scholes, and HO Pörtner (2022) Governing for Transformative Change across the Biodiversity–Climate–Society Nexus. *BioScience*, **XX**(Xx), 1–21. <https://doi.org/10.1093/biosci/biac031>
- Renard D and D Tilman (2019) National food production stabilized by crop diversity. *Nature*. <https://doi.org/10.1038/s41586-019-1316-y>
- Sanchez Bogado AC, SK Jones, A Purvis, N Estrada Carmona, and A De Palma (2022) Landscape and functional groups moderate the effect of diversified farming on biodiversity: A global meta-analysis. *Agriculture, Ecosystems and Environment*, 332.
- Tittonell P (2014) Ecological intensification of agriculture-sustainable by nature. *Current Opinion in Environmental Sustainability*, **8**: 53–61. <https://doi.org/10.1016/j.cosust.2014.08.006>
- UNEP-WCMC and IUCN (2022) Protected Planet: The world database on other effective area-based conservation measures, [March / 2022], Cambridge, UK: UNEP-WCMC and IUCN. Available at: www.protectedplanet.net.

ARTICLES ON
MICROBIAL GENETIC RESOURCES



ARTICLES ON MICROBIAL GENETIC RESOURCES

Title	Page No.
Microbial Genetic Resources: Some Aspects and Prospects	333
Conserving Microbial Diversity: Practices, Trends and Beyond	338
Diversity of Root Nodule Bacteria: Strengthening R&D and Inoculant Supply	343
Cyanobacterial Diversity Assessment Under Diverse Environments: A Molecular Approach	346
Synthetic Microbial Community (SynCom) for Sustainable Agriculture	351
Microbiome and Plant Health	355
Interlinking Soil Microbial Diversity and Rhizodeposition for Enhancing Nutrient Uptake and Productivity	360
Microbial Nitrification Paradox: A Paradigm Shift on Nitrogen Uptake by Rice	365
Current Status and Recent Developments in Microbial Pesticide Use in India	369
Blue Green Algae for Secondary Agriculture	375

Microbial Genetic Resources: Some Aspects and Prospects

Manoharachary Chakravarthula

Department of Botany, Osmania University Hyderabad-500007, Telangana, India

Nature is bountiful of microbes and fungi which form backbone for mankind. Only 5 to 70% of microbes and fungi are available in the form of cultures/germplasm. There is a need to unearth the hidden microbial and fungal wealth using innovative methods. Aspects and prospects of microbial genetic resources are discussed in this manuscript.

Key Words: Biodiversity, Cultures, Fungi, Microbes, WFCC

Introduction

Microbiome is a unique term applied to denote the prokaryotes and some fungi like yeasts which colonize and survive in different ecological niches under varied ecological conditions including extremophilic habitats. Microbes have a greater role in waste recycling, recycling of elements, plant productivity, management of biotic and abiotic stress, in industry, agriculture, pharmaceuticals and others. It is known that several agricultural practices including non-judicious use of pesticides, fungicides, weedicides, biocides, and over-fertilization besides human interference have caused damage to the ecological functioning of an ecosystem which in turn resulted in the loss of helpful microbes and fungi that are essential for plant and soil health. Thus, the complications that arose have impacted the influence of microbes and their role in agro-ecosystems.

The microbial world and microbial germplasms are the unseen national and international resources that deserve greater attention. Microbes include viruses, bacteria, mycoplasma, actinomycetes, fungi, algae and others (Wald, 2001).

Few microbial species were described till 1995, but later more than 2000 microbial species have been added. Maybe as of today, 400 bacterial species must have been encountered from India. However, the actinomycetes in particular with reference to India need to be attempted with utmost care and attention due to the fact that only around 30 to 40 actinomycetes are reported from India. The stipulated rules of CBD and National biodiversity Authority do not permit the exchange of microbial or

fungal cultures, though they permit the scientists to discuss with their respective partners in person. As per the international nomenclatural rules, one has to deposit the new taxon of microbial and fungal germplasm, in two culture collections in two countries.

Microbial and Fungal Diversity

The diversity of microbes existing on earth is still not known fully to mankind and it is mainly because out of millions of organisms existing and colonising diversified ecological niches have not been discovered fully. Only 5 to 10% of microbes and fungi have been discovered and 95% remain unexplored in nature. The exploration of such wealth may take thousands of years. At present what we know about microbial diversity is only a drop of water present in Atlantic Sea. It is known that microbes have been playing a very important role with reference to mankind besides being the foundation of the coming bioeconomy. The Convention of Biological Diversity (CBD) makes *ex situ* of conservation of microbes and fungi as mandatory. These organisms have become increasingly important and coordinated with shared best practices including data systems and engaged with many genome programs envisaged by the World Federation for Culture Collections (WFCC). Article 9 of CBD mandates that every country needs to have *ex situ* microbial germplasm repository to manage the microbial and fungal diversity unique to the country. Microbial and fungal genetic resources represent a complete genome, genes and some parts of gene that are relevant to human welfare. Genetic resource can be referred as the diversity of microbial and fungal and other

*Author for Correspondence: Email-cmchary@rediffmail.com

living entities or species that make up an ecosystem. Mutation, genetic drift, gene flow and selection act as evolutionary factors due to the fact that every population may contain a distinctive gene pool different from other population. Thus, genetic resources are complementary to type culture collections and biodiversity collections. Microbial resource centres may maintain diversified groups of microbes and fungi whereas genetic resources may maintain thousands of isolates including hundreds of mutants at the same genetic locus. Genetic resources are not configured as intellectual property due to the fact that they are the creations of nature but not that of the human mind. It is important to control biodiversity loss, as humans are dependent on plants, animals, microbes, fungi and others for their survival. In this context, it is imperative to mention that 1/3rd of global microbial and fungal diversity exists in India and there is a need to explore diversified habitats for the microbes and fungi for human sustenance.

Fungi are heterotrophs, flourish in various environments as saprophytes and also as pathogens due to the absence of chlorophyll and are able to proliferate on materials originating from animals, plants, and others. Their diversified existence in a complex ecosystem enriches the life of man and other living beings. Fungal diversity is immeasurable and its loss will near irretrievable damage to the intricate network of life. Fungi form prospective candidates of the biological diversity omnipresent in the environment and attracted greater attention from many due to their vital role in human welfare. There is a great threat to the fungal world due to the deterioration of the environmental conditions. Many fungi have become already endangered and it's high time that everyone should work for the protection of nature in terms of biodiversity. Recently it has been estimated that 13 million fungi may exist on earth of which only 150000 species are so far described and India is bestowed with 29000 fungal species (Karon *et al.*, 2004, Manoharachary *et al.*, 2005, Smith, 2012).

Enumeration, Isolation, Growth and Identification of Microbes and Fungi

Various habitats form microcosms and are inhabited by a wide range of microbes and fungi whose predominance is influenced by various ecological factors and biological factors. The population and functional dynamics of microbes and fungi differ from habitat to habitat. The complex activities happening in each ecological niche due to microbes and fungi put tremendous influence in

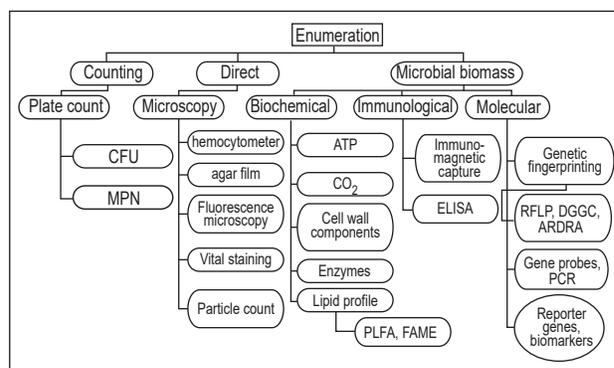


Fig. 1. Enumeration, Isolation, Growth and Identification of Microbes

the form of beneficial activities for human sustenance. Therefore, methods of isolation, enumeration, culturing of microbes and other activities form important aspects of understanding microbial and fungal diversity. The study of a microbial and fungal population in a given habitat is a challenging task due to the fact that some organisms may be represented by large numbers or some by small numbers. Therefore, various approaches have been evolved to enumerate microbes. After enumeration and isolation of microbes and fungi, different culture media have to be selected for their growth and multiplication.

Synthetic, semi-synthetic and non-synthetic or natural media are used for culturing the microorganisms. Enriched media contains substances that encourage the growth and multiplication of required organisms. This is followed by isolation, growth and characterization of microorganisms. Identification of microorganisms and fungi is based on morphological characters, molecular characters and serological techniques including DNA

Conservation of Microbes and Fungi

Biopreservation is nothing but the preservation of the integrity and functionality of the organism. The main aim is to maintain the organism alive, and uncontaminated without affecting the original status and also without causing any variation and mutation. Unlike for plants and animals, less attention is paid for the conservation of microbes and fungi. Microbial germplasm deposition form backbone and is the gold mine for all the ecosystems services including production of novel therapeutics. Community and ecosystem preservation is very difficult to understand as per current technology. Hence, gene pool in the form of extracted DNA has to be preserved. Researchers have to focus on preservation of intact

communities including co cultures, enrichment cultures and natural communities.

It is of immense importance to culture non-culturable microbes and fungi. Preservations include agar slant culture, agar slant cultured with paraffin oil, saline suspension, cryopreservation, preservation in dry vacuum, freeze drying (lyophilization) and refrigeration. Fungi form an important component for human welfare and therefore deserve to be conserved *in situ* and *ex situ* methods. Some of the fungi like rusts, smuts, powdery mildews and others cannot be cultured artificially. Therefore, they have to be maintained on living hosts which is considered in-situ conservation. It is also possible to preserve them in the form of herbarium specimens wherein the fungal spores can survive for years together. In some fungal species, *ex situ* conservation include preservations in agar slants, glycerol oil, soil culture, water culture, cryopreservation, preservation on dry vacuum, lyophilization and refrigeration (Om Prakash *et al.*, 2013). Further, the identification of microbes and fungi is an important aspect due to the fact that many microbiologists and fungal taxonomists have expired, some of them are not in a position to render help in identification due to bad health and there are only few endangered taxonomists who can be helpful in the identification of bacteria and fungi. Unfortunately, scientific organisations, governmental organisations, research institutions, policymakers, and universities have not emphasized the importance of taxonomy and taxonomists though taxonomy is the mother of all sciences. Taxonomy is important as it is useful to identify the species that are useful to mankind.

Microbial Genetic Resources

Mother earth possesses greater biomass and rich diversity of microbes and fungi. Microbial and fungal culture collections have a long history of making significant contributions to various branches of science. The stakeholders of microbes, fungi and others have found it difficult to convince the funding agencies and others with a common and unified voice. The world's largest microbial and fungal collection centre, American Type Culture Collection (ATCC), was established in 1925. ATCC distributes microbial cultures which are certified and recognized under Budapest treaty. Further, the whole genome sequence that is generated for microbes

is not publicly available. Regulatory issues like Nagoya protocol on access and benefit-sharing as well as biosecurity issues impose a new compliance burden on collection staff. Thus there are diversified issues and problems that need perfect answers. Diverse living collections are guardians of characterized germplasm for agriculture and health which form the foundation for biotechnology (Kovin, 2017; WFCC 2010). Some collection centres are more than a hundred years old. A total of 476 culture depositories are registered with World Federation of Culture Collection, but only a few are funded. However, the nonfunded organisations are getting closed, thus paving the way for losing millions of pounds. All the national and international societies and also organisations have to be united to stress up on funding microbial and fungal resource centres around the world. Table 1 shows the microbial germplasm centres located around the world and India.

Table 1. Microbial spectrum barcoding

Microbes	India	World
Viruses/bacteria etc.	1200	15000
Algae	8000	40000
Fungi	29000	1,50,000

Asia

Countries and Regions	Culture Collections	Cultures
Bangladesh	1	25
China	48	343,831
Chinese Taipei	2	75,746
Hong Kong	2	3,201
India	33	221,241
Indonesia	22	15,774
Iran	23	112,486
Israel	4	776
Japan	26	265,863
Korea (Rep. of)	26	175,183
Malaysia	13	16,828
Mongolia	3	7,131
Nepal	1	14
Pakistan	9	4,168
Philippines	6	4,197
Qatar	1	2,300
Saudi Arabia	1	9
Singapore	3	6,289
Sri Lanka	9	997
Thailand	66	124,597
Viet Nam	4	10,306
Total	303	1,390,962

Africa

Countries and Regions	Culture Collections	Cultures
Egypt	6	2,973
Morocco	2	1,742
Nigeria	3	385
Senegal	1	210
South Africa	4	13,660
Uganda	1	550
Zimbabwe	2	702
Total	19	20,222

America

Countries and Regions	Culture Collections	Cultures
Argentina	15	10,280
Brazil	89	137,292
Canada	20	88,741
Chile	5	2,491
Colombia	4	8,035
Cuba	13	6,440
Ecuador	3	4,481
Mexico	18	9,757
USA	37	343,835
Uruguay	1	20
Venezuela	3	3,476
Total	208	614,848

Europe

Countries and Regions	Culture Collections	Cultures
Armenia	1	11,520
Austria	2	6,070
Belarus	1	1,645
Belgium	7	295,229
Bulgaria	4	12,807
Czech	14	14,995
Denmark	3	112,066
Estonia	5	30,412
Finland	2	10,857

Conclusion

There is a need to strengthen biodiversity, taxonomy, conservation, and utilization of microbes and fungi.

Table. Holding in important culture collection centres of India

S. No	Name	Bacteria	Fungi	Yeasts	BGA
1	NCIM, Pune	1400	950	600	20
2	NFMC, Bharatidasan University, Tiruchirappalli	---	----	----	290
3	ITCC, IARI, New Delhi	20	3800	--	--
4	MCC, Pune	149314	15338	--	--
5	MTCC, Chandigarh	1124	1245	575	---
6	NAIMCC, NBAIM, MAU, UP	3049	4179	63	356
7	NCCPF, Chandigarh	4341	---	---	----
8	NCDC, Karnal	400	15	20	---
9	NFCCI, Pune	3050	-	-	-

Countries and Regions	Culture Collections	Cultures
France	38	98,685
Germany	15	107,024
Greece(Hellenic Rep.)	8	7,962
Hungary	8	15,451
Ireland	1	380
Italy	20	49,972
Kazakhstan	2	398
Latvia	1	1,452
Macedonia	1	12
Netherlands	6	106,275
Norway	2	3,028
Poland	10	9,206
Portugal	9	14,940
Romania	2	760
Russian Federation	29	68,427
Slovak	4	4,825
Slovenia	3	17,142
Spain	5	12,821
Sweden	4	52,940
Switzerland	4	3,965
Turkey	12	7,769
U.K.	19	87,415
Ukraine	11	18,695
Uzbekistan	4	3,436
Yugoslavia	2	897
Total	259	1,189,478

Oceania

Countries and Regions	Culture Collections	Cultures
Australia	34	97,120
Fiji	1	49
New Zealand	6	28,148
Papua New Guinea	1	270
Total	42	125,587

Source: World Data Centre for Microorganisms and World Federation for Culture Collection.

Human resource development is essential. Hence, skilled scientists/technicians human resources need to be developed in the whole world. Microbial genetic resources contain several metabolites that are useful

to mankind. Therefore, germplasm centres have to be funded adequately to encourage diversity, taxonomy and conservation studies and research.

Acknowledgements

Thanks to NASI, Prayagraj for encouragement and to Dr Anil Kumar Saxena, Ex-Director NBAIM for encouragement and support.

References

- Nakasone KK, SW Peterson and S Jong (2004) Preservation and distribution of fungal cultures. In: Biodiversity of fungi (GG Mueller, GF Bills and MS Foster Eds) Elsevier Academic Press, USA.
- Kevin M (2017) A review of living collections with special emphasis on sustainability and its impact on research across multiple disciplines. *Biopreserv. Biobank* **115**: 20-30.
- Manoharachary C, K Sridhar, RA Singh, A Adholeya, S Rawat, and BN Johri (2006) Fungal diversity, distribution, conservation, and prospects of fungi from India. *Curr. Sci.* **89**: 59-70.
- Om Prakash, Y Nimonkar and YS Shouche (2013) Practice and prospects of microbial preservation. *FEMS Microbiol. Lett.* **339**: 1-9.
- Smith D (2012) Culture Collections. *Adv. Appl. Microbiol.* **79**: 73-118.
- Wald S (2001) Biological resource centres underpinning future of life sciences and biotechnology. OECD Publishing, 2001. <https://www.oecd.org/sti/biotech/2487.422>
- WFCC (2010) World federation from culture collections- guidelines for the establishment and generation of collections of cultures of microorganisms. www.wfcc.inte/guidelines

Conserving Microbial Diversity: Practices, Trends and Beyond

Alok Kumar Srivastava, Hillol Chakdar*, Pramod Kumar Sahu and Murugan Kumar

ICAR-National Bureau of Agriculturally Important Microorganisms, Kushmaur-275103, Uttar Pradesh, India

Interests in preservation of microbes for *ex situ* conservation have grown hand in hand with the microbial diversity studies flourishing worldwide. For a successful exploration study, preservation of isolated strains without any change in original traits is as important as collection of pure culture of microbes from different habitats, their characterization and exploration for economical use. Preservation of microbes is also important to realize the potential of characterized strains on a large scale. This review focusses on the different methods available for preservation of microbial strains for short to long term. In addition, specific preservation techniques for certain microbes and problems and concerns in the routinely used preservation techniques and revival of preserved microbes have been dealt in detail. The review highlights the importance of research in the area of preservation of microbes.

Key Words: Conservation, Cryopreservation, Lyophilization, Microbial Diversity, Mineral Oil Storage

Introduction

Microorganisms are vitally important life forms on the earth for humankind not only because of their ubiquitous nature and cosmopolitan presence but due to their obvious role and direct involvement in the natural biogeochemical cycles, cycling of biologically important elements, minerals and nutrients, food web, environmental remediation, crop production or protection. Therefore, acquisition, characterization, cataloguing and conservation of microbes from different habitats are most important task for any culture collection. The World Data Centre for Microorganisms (WDCM) currently lists 820 culture collections, in 78 countries, holding more than 3,348,121 microbial strains, registered with the World Data Centre for Microbes. At present, WDCM database of culture collections encompass 1,521,992 bacteria, 952,933 fungi, 32,839 viruses and some of them are cell line (42,223). The status of Collections of Asian countries and its holdings depicts that India ranks third after Japan and China with the total holdings of 221,241 accessions and is 5th in the top 20 ranking countries maintaining the microbial culture collections. In India, Microbial Type Culture Collection (MTCC), Chandigarh, National Centre for Microbial Resource (NCMR), Pune, National Agriculturally Important Microbial Culture Collection (NAIMCC), Mau, National Fungal Culture Collection of India (NFCCI), Pune, National

Collection of Industrial Microorganisms (NCIM), Pune and Indian Type Culture Collection (ITCC), New Delhi are the important microbial resource centres catering the needs of the research and industrial applications. Recognizing the importance of industrial applications of microorganisms, three culture collections in India viz. MTCC, NCMR and NAIMCC have been recognized as International Depository Authority (IDA) for patent purposes under the Budapest Treaty by World Intellectual Property Organization (WIPO). The conservation of microorganisms is key to their application in agriculture and industry. Microbial culture collections in India and abroad generally follow both short- and long-term preservation methods which are subsequently supplied to a wide range of clientele including academia and industry for diverse applications and basic research. In this paper, we aim to briefly describe various methods of *ex situ* microbial preservation and future directions to develop more efficient techniques to conserve more of the microbial diversity for future applications.

Ex-situ Conservation of Microorganisms

The importance of microbes in modern civilization is incomparable due to their multifaceted application in food, pharmaceuticals, and biotechnology industry. Working with microbes requires isolating, characterizing, and preserving the potential ones for a longer period of time. Microbes have a brilliant capability of surviving

*Author for Correspondence: Email- hillol.chakdar@gmail.com

long-term preservation with the least effect on their physiology than any other organisms (Hawksworth and Kirsop, 1988). This gives the opportunity to maintain microbial cultures for an almost indefinite time. The diversity of microorganisms is enormous and so are the methods of their conservation. Some of the microbes can survive in sterile water itself (e.g. *Ralstonia*) whereas some of the classes of microbes are very difficult to preserve for a long time (e.g. non-sporulating fungi and cyanobacteria). Different preservation methods for microorganisms are used, such as slant, stab, mineral oil stock, cryopreservation, lyophilization, drying in an inert material, etc. The following sections briefly describe various preservation methods for microbial cultures-

Short-term Preservation

Researchers follow varied techniques for preserving microbial strains for a short period of time. It is the simplest method for preserving microbes, however, has more chances of developing mutants, loss of culture, and compromising culture purity. Different methods used for short-term preservation are as follows-

Preservation in Slants

Both bacterial and fungal cultures growing on agar slants can be stored at 4-8°C for a period of 6-8 months. Some additives such as acacia gum could further enhance survival. Oil overlay is also practiced to improve the survival of slants (Nakasone *et al.*, 2004).

Storage in Sterile Water

Many bacteria, as well as fungi, show stability in sterile water. It is inexpensive and successfully reported for preserving members of fungal groups like oomycetes, basidiomycetes, ectomycorrhiza, ascomycetes, yeast, etc. for varying periods. Though the survival varies from class to class, a maximum survival of upto 10 years is reported in sterile water for wood-inhabiting ascomycetes when stored at 20°C (Johnson and Martin, 1992). Bacteria such as *Ralstonia solanacearum*, *Pseudomonas fluorescens*, *Xanthomonas campestris*, and *Staphylococcus aureus* are also reported to have different degrees of survival in sterile water.

Mineral Oil Storage

Fungal cultures are preserved submerged in high-quality mineral oil. The storage can be done at ambient temperatures. Cultures can be preserved for several years immersed in mineral oil. The high-density mineral

oils restrict the oxygen diffusion and thereby slow the metabolic activities leading to survival for a longer period. This technique is suitable for nonsporulating fungi which can't be preserved through cryopreservation and lyophilization.

Long-term Preservation

Continuous sub-culturing affects the cultural purity as well as compromise the important traits of the microbes, thus long-term preservation methods are followed to keep the microbial cultures for a period of 5-10 years. Preservation in -80°C, -196°C, and lyophilized storage are done for long-term storage. The long-term storage in form of cryopreservation and lyophilization is suitable for many microorganisms, the research has to be poured into developing protocols for preserving recalcitrant microbes.

Lyophilized Storage

Lyophilization/freeze-drying is one of the most reliable methods for the long-term preservation of microbes. This is practiced regularly in most of the culture collections due to low maintenance and ease of transport. In the process of lyophilization, the cell suspension is frozen, dried under vacuum, and sealed. This dry powder under vacuum can be kept at ambient temperature for a period of 5-10 years, or even more, depending on the type of culture. Stability in lyophilized culture is achieved by the addition of cryoprotective agents. Skim milk (10%) is commonly used as a cryoprotective agent. Other cryoprotectants like inositol, sucrose, etc. are also used. Vacuum is maintained in the lyophilized culture vial throughout the preservation time. Though the lyophilized ampoules can be stored at ambient temperatures, the viability can be further improved by storing the ampoules at 4°C in dark.

Cryopreservation

Cryopreservation of microbes refers to conserving microbial cells at cryogenic temperature. Preservation at ultra-low temperature (-80°C or below) halts cellular activities and prevents damage to DNA and proteins. Halting the physiological activities of the cell helps in maintaining life for a longer period of time. Cryoprotectants such as glycerol (10-20%) and DMSO (5%) can prevent cellular damage due to the formation of ice crystals inside cells (Wowk, 2007). Polymer-based (polyethylene glycol) cryopreservation has also been reported for bacteriophages (Marton *et al.*, 2021).

Different temperature storages are in practice viz. -80 and -196°C. The storage at -196°C is done through liquid-N₂ and requires constant refilling. As the rate of survival of the cryopreserved microorganisms decreases with the increase in temperature, preservation below -80°C is not recommended. The microorganisms can be preserved in 10-20% glycerol stocks for 8-10 months period (some microbes can survive even longer) at -20°C which, however, is not suitable for long-term preservation. Besides storage temperature, the success of cryopreservation depends largely on the rate of cooling, cell size, cell wall physiology, the permeability of water, and nature of the cryoprotectants used. To cover the preservation of diverse groups of microbes, specific attention is required to optimize suitable cryoprotectants and other conditions. The length of survival also depends on the rate of freezing and thawing.

Specific Preservation Techniques and Modern Development in Preservation

Apart from the above-mentioned routine approaches, specific preservation techniques have also been developed for recalcitrant microbes which have low viability under common long-term storage protocols. To cover the preservation of diverse groups of microbes, specific attention is required to optimize suitable cryoprotectants and other conditions after considering the physiology of such microbes. The vegetative hyphae of saprophytic fungi *Lentinus crinitus* successfully cryopreserved using wheat grain along with other cryoprotectants (glycerol, DMSO, sucrose, glucose) (Bertéli et al., 2022). A collective effort has been taken by the European culture collections under the EMbaRC project for improving the survival and storage of lyo-recalcitrant bacteria (Peiren et al., 2015). This study concluded that horse serum supplemented with trehalose and skim milk supplemented with trehalose were better-suited lyoprotectant. Cryopreservation for microalgae is still a challenge. Recent reports suggest that cold acclimation at 4°C for 24 h before lyophilization along with controlled freezing, plunging, and thawing using an accurate programmable freezer could enhance the survival and storage of microalgae *Ettlia* sp. (Ha et al., 2019). In the case of anaerobic microorganisms, handling and storage is difficult. A novel kit-based strategy called BIOME-Preserve is used for transporting and preserving anaerobic microorganisms such as *Bifidobacterium*, *Bacteroides*, *Blautia*, and *Anaerobutyricum* (Hyde et al., 2022) at cryogenic temperatures. Resting structures

of some fungi such as sclerotia or microsclerotia can be preserved for 2-3 years at 4°C. Soil fungi such as *Magnaporthe*, *Physarum*, *Cylindrocladium*, etc can be preserved by this method (Singleton et al., 1992). Preserving fungi on inorganic substrates such as grains, agar strips, wood chips, filter paper, straw, etc. is also a useful practice (Nakasone et al., 2004; Bertéli et al., 2022). Preservation in the sterile soil-sand mixture is also practiced for fungi like *Rhizoctonia solani* (Sneh et al., 1991). Silica gel can be effectively used for sporulating fungi.

Problems and Prospects in Microbial Conservation

The methods used for both short- or long-term preservation of microorganisms suffer from certain disadvantages. Short-term preservation like subculturing on slants or maintenance of live cultures in liquid medium are prone to contamination and affect the stability of important traits due to mutation. The loss of traits after subculturing or maintenance as live cultures are more frequent in case of plasmid borne characters. However, these methods are the most popular ones as they do not require sophisticated instruments like ultra-low freezers or freeze dryers. The key to the preservation of fungi in mineral oil is the quality of the oil used. Ideally the oils should have a density 0.83-0.89 g/cm³ which otherwise may not be effective in preservation. Many of the fungi preserved in mineral oil show slow revival and may also require a number of subcultures to remove the oils. However, there are also chances of contamination of the overlaying oil with airborne fungal spores if the tubes containing the fungal cultures are not kept tightly capped and not placed in a clean place. Mineral oil preservation is more effective when kept at low temperature.

Cryopreservation of microbes in glycerol or DMSO stock is yet another popular approach. Although many other cryoprotectants have been used but glycerol or DMSO have been mostly used. It has been reported that a number of aerobic bacteria like *Staphylococcus*, *Micrococcus*, *Pseudomonas*, *Streptococcus*, *Lactococcus*, *Corynebacterium* and *E. coli* show toxic effects with 10% DMSO (Fomin et al., 1973). Even some microalgae like *Chlorella* have shown toxic effect at very low concentration (<2.5%) of DMSO (Morris, 1976). Some microorganisms like *Methylomonas*, *Methylocystis* have been reported to show very minimal cryoprotective effect while a few others like *Rhodospirillum*, *Micrococcus*,

Pseudomonas have even shown toxicity to glycerol (Fomin *et al.*, 1973; Green and Woodford, 1992). A large number of other cryoprotectants like polyalcohols, saccharides, heterocyclic compounds have also been used as cryoprotectants in microbial preservation. Moreover, a combination of cryoprotectants may be more useful for longer preservation as compared to using a single or routine one. Cryopreservation under temperature at or below -80°C requires ultralow temperature freezers or liquid nitrogen which may not be available at every microbiology laboratory. The rate of cooling and thawing is another important factor affecting the cell viability. Cell type and cell size is also crucial for cryopreservation as the response to low temperature varies with such factors. Initial cell density should be high enough as the microbial cells will die throughout the storage period.

Though lyophilization is very successful in preserving many bacteria, actinomycetes, archaea, and fungi, some of the microbial classes such as non-sporulating fungi, yeast (e.g. *Lipomyces*, *Leucosporidium*, *Brettanomyces*), and bacteria (e.g. *Clostridium botulinum*, *Helicobacter pylori*) can't survive lyophilization with routine protocols (Smith *et al.*, 2008). In addition to cell density and type of cryoprotectants used during freeze drying, the growth phase of the cells is also a critical factor for survival of bacteria upon lyophilization. Desiccation tolerance of microorganisms is also important factor for freeze drying as desiccation sensitive cells may lose the viability in a short period. Rehydration of the freeze-dried microbial cultures is also crucial. Use of inappropriate rehydration media may also result in poor or no growth during revival. Usually, 10% non-fat skimmed milk can be a good rehydration medium.

In most of the laboratories and culture collections, a generic preservation methodology is followed which may,

however, not be very useful always. The physiological responses of the microbial cell to low temperature, desiccation need to be considered along with the type and organization of the cell. As the known microbial diversity is only minor fraction of the actual diversity, we need to focus more on developing novel methodologies to preserve more diverse microflora. In nature, even the recalcitrant microbial cells can survive for a considerable period which otherwise either cannot be preserved in laboratories at all (e.g. obligate pathogens) or can be preserved for a very short period. Therefore, mimicking the natural conditions like preserving in freeze dried host or soil or sand or natural polysaccharides may help to preserve for longer period. Certainly, other approaches like optimizing conditions of cryopreservation or lyophilization and using appropriate cryoprotectants should also be carried out. Besides freeze drying, other drying technologies like fluid bed drying or spray drying should also be largely tested for wide range of microorganisms.

Conclusion

The research on preservation of microbes is not given its due credit, even after understanding the importance of microbes in various fields. In most culture collections worldwide, a generic method is followed for medium term and long term preservation. This is not advisable especially in case of storage of cells in deep freezers (-20°C and -80°C) in the presence of cryoprotectants like glycerol and lyophilization. With regards to medium term storage in deep freezers methodologies are to be standardized for efficient preservation by optimizing the different cryoprotectants and their concentration for different groups of microorganisms. Similarly, for lyophilization, methodologies are to be optimized with different lyoprotectants for different group of

FUTURE DIRECTIONS

- Instead of hit and trial with methods or cryoprotectants, a biology/physiology driven approach should be taken up. Based on the biology of the microbes and environmental tolerance, the method of preservation should be chosen
- The natural survival of the microbes in diverse environments should be mimicked for longer preservation.
- Efforts should also be devoted towards optimizing conditions and media to induce resting structures which can be easily preserved for longer durations
- Developing long term preservation methods for obligate pathogens retaining their pathogenicity. Host factors should also be considered while developing such methods

microorganisms. Culture collections worldwide should in future focus on optimizing the routine preservation techniques of microorganisms of paramount importance. They should also workout on the methodologies for long term preservation of recalcitrant microbes like archaea and others.

References

- Bertéli MBD, CR Pinheiro, BO Philadelpho, DM Otero, CDF Ribeiro, CO de Souza, E de Souza Ferreira, SP Ruiz, JS do Valle, GA Linde and NB Colauto (2022) Long-term cryopreservation of *Lentinus crinitus* strains by wheat grain technique. *J. Microbiol. Methods* **198**: 106491.
- Fomin DC, IS Alycheva, LI Veselovskaya and SM Tatchin (1973) Antibacterial properties of dimethyl sulphoxide (in Russian). In: *Khimioterapia infektsiy I lekarstvennoy ustoychivosti patogennykh mikroorganizmov*, Moskva, 63–64.
- Green PN and SK Woodford (1992) Preservation studies on some obligately methanotrophic bacteria. *Lett. Appl. Microbiol.* **14**: 158–162.
- Ha JS, JW Lee, SH Seo, CY Ahn, GJ Rho, HG Lee and HM Oh (2019) Optimized cryopreservation of *Ettlia* sp. using short cold acclimation and controlled freezing procedure. *J. Appl. Phycol.* **3**: 2277–2287.
- Hawksworth DL and BE Kirsop (1988). (Eds) 'Living resources for biotechnology. Filamentous fungi.' (Cambridge University Press: Cambridge).
- Hyde ER, H Lozano and S Cox (2022) BIOME-Preserve: A novel storage and transport medium for preserving anaerobic microbiota samples for culture recovery. *Plos one* **17**(1): e0261820.
- Johnson GC and AK Martin (1992) Survival of wood-inhabiting fungi stored for 10 years in water and under oil. *Can. J. Microbiol.* **38**: 861–864.
- Marton HL, KM Styles, P Kilbride, AP Sagona and MI Gibson (2021) Polymer-Mediated Cryopreservation of Bacteriophages. *Biomacromolecules* **22**: 5281–5289.
- Morris GJ (1976) The cryopreservation of *Chlorella* 1. Interactions of rate of cooling, protective additive and warming rate. *Arch. Microbiol.* **107**: 57–62.
- Peiren J, J Buyse, P De E Vos, Lang, D Clermont, S Hamon, E Bégau, C Bizet, J Pascual, MA Ruvira and MC Macián (2015) Improving survival and storage stability of bacteria recalcitrant to freeze-drying: a coordinated study by European culture collections. *Appl. Microbiol. Biotechnol.* **99**: 3559–3571.
- Singleton LL, JD Mihail and CM Rush 1992 (Eds) 'Methods for research on soil-borne phytopathogenic fungi'. (American Phytopathological Society Press: St. Paul, Minnesota).
- Sneh B, L Burpee and A Ogoshi (1991) Identification of *Rhizoctonia* species. APS press.
- Wolk B (2007) How cryoprotectants work. *Cryonics* **28**: 3–7.

Diversity of Root Nodule Bacteria: Strengthening R&D and Inoculant Supply

DLN Rao

Ex-Project Coordinator (AICRP on BNF), ICAR-Indian Institute of Soil Science, Bhopal-462038, Madhya Pradesh., India

There is a renewed global interest in biological nitrogen fixation (BNF) for the last several decades, particularly in assessing the diversity of root nodule bacterial populations in diverse geographies. This article focuses on the current status and argues for the establishment of centres of excellence on Nitrogen Fixation, Microbial Inoculants and Soil Ecology to provide sharper focus and impetus for the research and development effort needed for rhizobial strain improvement and for streamlining inoculants supply to help achieve improved soil health and N sustainability.

Introduction

The world nitrogen fertilizer demand continues to grow but the scenario has undergone a significant change with China and India accounting for nearly half the consumption. Overuse and imbalanced application, discontinuance of traditional practices like crop residue recycling, composts and green manuring, legume rotations etc., have all led to adverse effects on soil physical properties, reduced soil organic matter content and diminished soil biodiversity. Climate change will further exacerbate these problems. Rightly, therefore, the ecofriendly processes like biological nitrogen fixation (BNF) by legume-rhizobia symbiosis are regaining importance because BNF can supply a major portion of N in low external input agriculture.

Legume BNF

Global agricultural BNF is estimated at 50-70 Tg yr⁻¹; cultivated legumes supply 24 Tg of N annually with soybean alone accounting for over three-fourths of it. The BNF input added by various crops of India was estimated at 5.20-5.76 Tg yr⁻¹ (Rao and Balachandar, 2017) representing 9.5 to 10.6 % of the global agricultural BNF, with legumes averaging about 2.24Tg yr⁻¹. About 50 million hectares of crop legumes, equivalent to about 25% of the area sown globally are inoculated with rhizobia each year with average yield benefits ranging from 15-20%. The primary goal of any legume rhizobial inoculant programme is to select those strains that compete well with native rhizobia for nodule occupancy, are stress-tolerant and most effective in improving BNF and yields. An overview of the status of BNF in Indian Agriculture is given in Rao (2014).

The bacteria which form nitrogen-fixing symbiosis with legumes are collectively called rhizobia. Until 1992, there were only four genera of root nodulating bacteria: *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium* and *Azorhizobium*. Later, four more were added, *Mesorhizobium*, *Allorhizobium*, *Methylobacterium* and *Burkholderia*. The classification of rhizobia is in great flux for the last 3 decades. The nomenclature of some of the old species is under revision and many new genera and species are being continuously discovered. The majority of legume nodulating bacteria (LNB) described so far belong to α -*Proteobacteria* class. The isolation and characterization of new and often unusual LNB including *Agrobacterium* on diversified plant hosts have resulted in the naming of many new rhizobial species. On the basis of the 16S rRNA gene sequence, the currently described legume symbionts of α , β and γ -*Proteobacteria* belong to 238 species in 18 genera and two clades. The number of rhizobial species that can nodulate cultivated legumes has expanded, e.g., for common bean (14 species) soybean (11), cowpea (6), chickpea (5), peanut (5), lentils (4), faba bean (3) and pea (2 species) (Shamseldin *et al.*, 2017). The currently recognized important genera and species of rhizobia are shown in the box.

Diversity of Rhizobia

The outcome of the huge amount of work done on the diversity of *Rhizobium* in the last four decades is apparent in the constant flux in the taxonomy of root nodulating rhizobia owing to the rapid progress in molecular biology-based omics technologies. The recent use of whole-genome sequencing-based taxonomy will

*Author for Correspondence: Email- dlnrao.icar@gmail.com

obviously again change the current taxonomy. The increase in legume explorations in different geographical regions is leading to isolation and characterization of more rhizobial species and the discovery of newer ones. The major focus in the last three decades has been on soybean rhizobia in China, Brazil and India.

Rhizobial Collections

Extensive, organised collections of root nodule bacteria were made all over India beginning from the late 1960's at IARI, New Delhi under a PL-480 project and under the ICAR's AICRP on Pulses Improvement Project from the 1970's. ICRISAT, Hyderabad from the mid-1970's created an extensive collection of rhizobia not only from India but of many elite global strains, these were later transferred to IARI under the DBT-*Rhizobium* germ plasm collections in early 1990's. Extensive surveys were made on nodulation of tree legumes at CSSRI, Karnal during mid-1990's in a DBT project and rhizobia deposited in the above collection. However, intensive efforts on biogeography of rhizobia were made only later, under the AINP on Soil Biodiversity-Biofertilizers, IISS, Bhopal first for soybean in Madhya Pradesh from 2002 onwards and later for all the major legumes grown in MP, Haryana, Rajasthan and Gujarat from 2009 onwards. The improved strains of soybean *Bradyrhizobium japonicum* are in supply for inoculant manufacture in central India. In the DBT-funded India-UK Nitrogen Fixation Centre (IUNFC) from 2016, effective strains of *Bradyrhizobium yuan mingense* and *Sinorhizobium* sp. of pigeonpea were isolated and field tested in central India at IISS, Bhopal and in North Indian Plains at NBAIM, Mau. The improved strains of rhizobia from the studies in the

Rhizobial Taxonomy

Rhizobium strains
Class α-Proteobacteria
I Order Rhizobiales
I Family Rhizobiaceae
Genus <i>Rhizobium</i> (98); Genus <i>Ensifer</i> (formerly <i>Sinorhizobium</i>) (21); Genus <i>Allorhizobium</i> (1); Genus <i>Shinella</i> (1); Genus <i>Pararhizobium</i> (5).
II Family Phyllobacteriaceae
Genus <i>Mesorhizobium</i> (40); Genus <i>Phyllobacterium</i> (8); Genus <i>Aminobacter</i> (1)
III Family Bradyrhizobiaceae
Genus <i>Bradyrhizobium</i> (37); Genus <i>Blastobacter</i> (2); Genus <i>Photrhizobium</i> (1)
IV Family Hyphomicrobiaceae
Genus <i>Devosia</i> (1); Genus <i>Azorhizobium</i> (3)
V Family Methylobacteriaceae

last 15 years in India are available in the depository of microbial germplasm at ICAR-NBAIM, Mau.

Notwithstanding the above, it can be asserted that rhizobial research in India has been comparatively neglected by microbiologists, attributable mainly to excessive attention towards plant growth-promoting rhizobacteria (PGPR). The rigorous efforts required in extraction of rhizobia from root nodules, differentiation from non-rhizobia, batteries of nodulation tests, MPN assays for soil rhizobial populations, ARA assays of nitrogenase activity in root nodules, continuous greenhouse testing of the strains every year, measurements of competition with native rhizobial strains for nodule occupancy by inoculant strains in fields using marked strains or their intrinsic antibiotic resistance, field measurements of N fixation either by N difference with non-legumes, isotope dilution methods or long-term N balance over several years summarize some of the reasons for the diversion towards more convenient research avenues.

Perspective

The neglect of rhizobial research is dangerous and needs urgent redressal. The only solution is to establish a centre on the pattern of countries like Australia devoted exclusively to research on rhizobia, maintain the culture collections, undertake continuous testing and centralise mother culture supply each crop season to inoculant manufacturers as per the need of various geographies.

Such a centre also needs to conserve rhizobia from wild legume relatives of cultivated legumes, tree legumes etc. It needs to be adequately staffed with scientists and technicians and equipped with advanced instruments for BNF measurements by isotope abundance.

There is an urgent need to improve rhizobial inoculant quality standards in India, both in terms of rhizobial counts and fewer contaminants. Also, develop technologies for manufacturing high-count inoculants and novel delivery systems.

During the PM's Vaibhav Summit in the vertical on "Microbial Resources for Sustainable Agriculture" held on Oct 10, 2020, I concluded my presentation (*Soil Ecological Stewardship-BNF, Microbial Inoculants, Soil Biological Indicators*) with a need for creating a long-term institutional framework, firstly, strengthen the existing networks of AINP Soil Biodiversity-Biofertilizers; AMAAS project and Crop and Commodity AICRPs; and secondly establish 3 advanced centres of excellence

at NBAIM, Mau, IARI, New Delhi and IISS, Bhopal with collaborations with ICAR institutes and SAU's.

- India Nitrogen Fixation Research Centre (INFRC)
- Indian Microbial Inoculants Technology Centre (IMTC)
- India Soil Ecology Research Centre (ISERC)

As a follow-up action of the summit, the establishment of the above centres along with assured core manpower and budget and linking up with entrepreneurial start-ups for inoculant manufacture would help achieve the goal of timely supply of quality inoculants to farmers through assured supply chains and translate into improved BNF, soil biodiversity and soil health, and ultimately to lesser

use of externally applied fertilizers, greater input use efficiency and improved factor productivity.

References

- Rao DLN (2014) Recent advances in biological nitrogen fixation in agricultural systems. *Proc. Ind. Nat. Sci. Acad.* **80**: 359-378.
- Rao DLN and D Balachandar (2017) Nitrogen inputs from Biological Nitrogen Fixation in Indian Agriculture. In: YP Abrol *et al.* (eds) The Indian Nitrogen Assessment. Sources of reactive nitrogen, environmental and climate effects, management options and policies. Elsevier, pp. 117-132.
- Shamseldin A, A Abdelkhalek, MJ Sadowsky (2017) Recent changes to the classification of symbiotic, nitrogen-fixing, legume-associating bacteria: a review. *Symbiosis* **71**:91-109.

Cyanobacterial Diversity Assessment Under Diverse Environments: A Molecular Approach

Krishna Kumar Rai, Ruchi Rai, Shilpi Singh and LC Rai*

Molecular Biology Section, Centre of Advanced Study in Botany, Institute of Science, Banaras Hindu University, Varanasi-221005, Uttar Pradesh, India

Cyanobacteria are photosynthetic microorganisms that inhabit diverse environmental conditions worldwide, ranging from freshwater and soil to extreme conditions. Apart from their photosynthesis and nitrogen-fixing abilities, they also produce a large variety of molecules with high potential in pharmaceutical and industrial applications. Recent investigations have elucidated that most of the cyanobacteria that belong to orders Oscillatoriales, Nostocales, Chroococcales, and Synechococcales have been extensively characterized. In contrast, those that belong to Pleurocapsales, Chroococciopsales, and Gloeobacterales remain unexplored in terms of their molecular diversity and relative bioactivity. In recent decades, molecular biology techniques have revealed valuable insights into the role and functions of various prokaryotes, including cyanobacteria, by precisely mapping their diversity in their natural environment. This review provides an overview of molecular techniques and their advantages and limitations for studying cyanobacterial diversity.

Key Words: Diversity, Molecular tools, Microarray, Sequencing

Introduction

Cyanobacteria are a distinctive phylogenetic group of gram-negative prokaryotes capable of performing oxygenic photosynthesis (Kumari *et al.*, 2012; Soo *et al.*, 2017). Approximately 3.6 billion years ago, they played a remarkable role in the oxygenation of the atmosphere by generating molecular O₂, thereby accentuating life on earth (Soo *et al.*, 2017). Cyanobacteria are also considered the progenitor of chloroplasts and are involved in the fixation of atmospheric N₂, thus playing a substantial role in maintaining the nitrogen economy (Kumari and Rai, 2019). Cyanobacteria are ubiquitous in diverse ecological niches and conceal enormous diversity in their habits, morphology, physiology, and metabolism. They are present in freshwater, marine, hot, and cold springs and symbiotic associations (Gaysina *et al.*, 2019). Furthermore, cyanobacteria produce a wide range of toxins and secondary metabolites; therefore, they have attracted broad interest in biofuel and biotechnological/pharmaceutical applications (Demay *et al.*, 2019).

The quantification of numbers and types of cyanobacteria within a community is imperative to get functional insight into their role in regulating the fundamental structure of the ecosystem (Gaysina *et al.*, 2019). Since cyanobacteria exhibit a high degree of

morphological diversity compared to other prokaryotes, therefore, until recently, they were identified and categorized based on their morphological traits per-se., cell size, shape, colour, branching pattern, and cell contents (Demay *et al.*, 2019). With the technological advancements and their unicellular, colonial and filamentous nature, researchers have managed to assess their diversity at morphological, physiological, biochemical, and molecular levels to reveal the hidden potential of cyanobacteria for industrial/agricultural applications (Kumari and Rai, 2019). The morphological and physiological diversity assessment methods have several limitations, like the low resolution of the light microscope failing to provide a clear view of spores, akinetes, and other taxonomic traits (Anand *et al.*, 2019).

Biochemical diversity analysis involves assessing fatty acid composition, polar/nonpolar glycolipids, and interestingly lipid profiling of various cyanobacteria such as *Synechocystis* sp. strain PCC 6308, *Prochloron* sp. and *Synechococcus* strain revealed valuable insight into their phylogenetic distributions (Sallal *et al.*, 1990). However, the biochemical approach also has its limitations in assessing cyanobacterial diversity due to their growth limitations and nutrient composition variations (Sallal *et al.*, 1990). Therefore, to overcome

*Author for Correspondence: Email-lcrbhu15@gmail.com; lcr Rai@bhu.ac.in

these constraints, present-day cyanobacterial diversity analysis is being made at the molecular level by using various molecular biology tools based on DNA and RNA, such as RT-PCR, microarray, and fluorescence in-situ hybridization, and metagenomics (Anand *et al.*, 2019). Several researchers have exploited the above-mentioned molecular techniques to get novel insights into the structure and functions of microbial communities at genomic, proteomic, and metabolomic levels (Rott *et al.*, 2018). This article will summarize the state-of-the-art molecular techniques currently used for cyanobacterial diversity assessment. In addition, a comprehensive understanding of their mechanisms, functions, and limitations will also be discussed.

Molecular tools for assessing cyanobacterial diversity

Several molecular biology tools have been employed in recent decades to study the diversity and function of cyanobacteria and other prokaryotes (Anand *et al.*, 2019). DNA or RNA profiling/fingerprinting is one of the robust molecular tools readily exploited to unveil microbial diversity in a specific environment by ascertaining variation at the gene level (Madigan *et al.*, 2009). A wide variety of microbial communities has been categorized phylogenetically and functionally using this approach (Fig. 1). However, the application of this technique becomes limiting wherever it was difficult to establish microbial cultures in the laboratory (Madigan

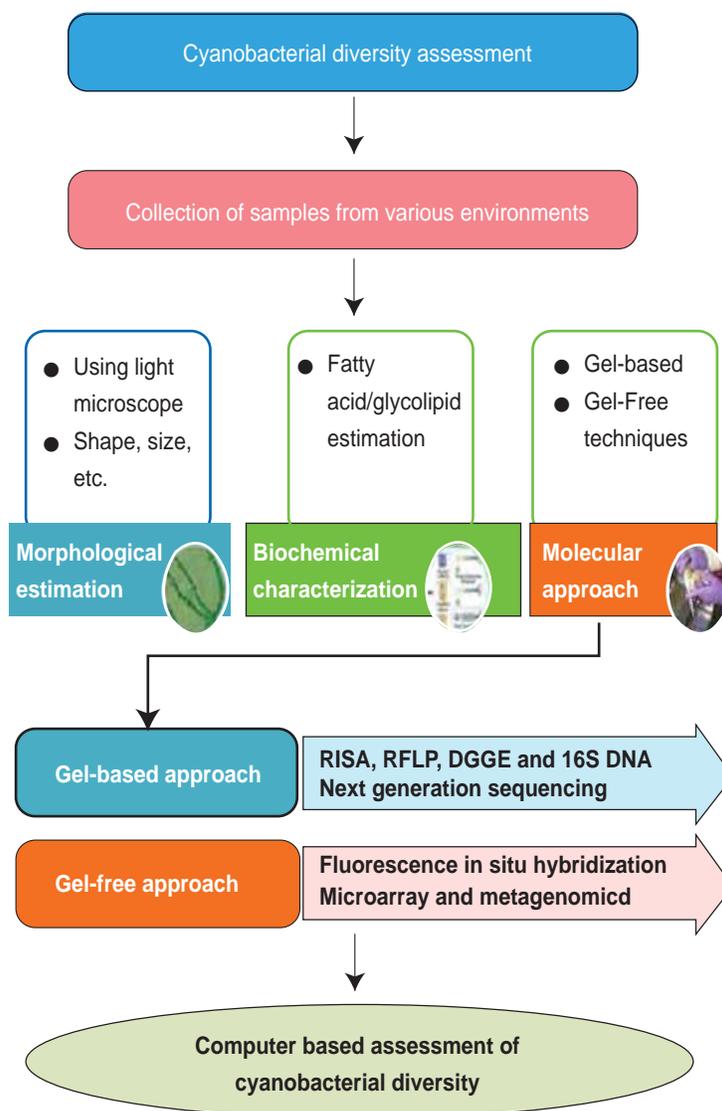


Fig. 1. A schematic overview of molecular techniques used for cyanobacterial diversity assessment

et al., 2009). Therefore, the above method has been revamped by integrating ultra-throughput sequencing technologies (NGS), metagenomics, DNA microarray for analysing gene expression patterns, and 16S rRNA gene library construction. Overall, the molecular techniques employed for cyanobacterial diversity and functional assessment have been grouped into two broad categories (i) gel-based and (ii) gel-free techniques.

Gel-based Techniques

Gel-based techniques mainly involving ribosomal intergenic spacer analysis (RISA), restriction fragment length polymorphism-PCR (RFLP-PCR), and denaturing gradient gel electrophoresis (DGGE) have been widely used to study cyanobacterial community (Kumari and Rai, 2019). RISA assesses microbial diversity using oligonucleotide designed using an intergenic spacer region between the 16S and 23S rRNA gene (Rottet et al., 2018). The functional mechanisms of RISA involve PCR amplification of DNA from different microbial samples with the specifically designed primer combinations, and the difference between banding patterns is visualized. Similarly, RFLP has also been extensively used to reveal the phylogenetic profile of several dominant microbial genera, including cyanobacteria, namely *Scytonema* and *Leptolyngbya* (Srivastava et al., 2016). RFLP involves restriction digestion of microbial DNA samples using specific restriction enzymes and amplifying the digested DNA fragments using particular primers (Lukow et al., 2000). The difference between amplicon size is evaluated, which is the basis of diversity assessment. Several studies have confirmed the applicability of both RISA and RFLP for ascertaining microbial diversity in *Anabaena* and other cyanobacteria in a precise, efficient, timely, and cost-effective way (Neilan et al., 1995).

DGGE is also a molecular tool that offers an extra edge for analysing microbial diversity, which involves the separation of different DNA samples based on increasing denaturing conditions (Kumari and Rai, 2019). DGGE technique offers high-resolution microbial diversity assessment by analysing the DNA molecules that differ by a single nucleotide (Song et al., 2005). Further, its robustness relies on the fact that it could work with lengthy DNA sequence analysis without compromising its specificity and efficiency. The DGGE technique work on a sequence-dependent separation mechanism where PCR products are visualized on polyacrylamide gel electrophoresis after denaturing the DNA samples either with formamide or urea (Jungblut

et al., 2005). As the denatured DNA fragment migrates on the gel, the shape of the DNA fragments changes dramatically, resulting in the slow migration of DNA along the established denaturing gradient, thus facilitating the visualization of the slightest change in the base pairs (Anand et al., 2019). A large body of literature has confirmed the efficiency and robustness of this technique in evaluating diversity analysis of toxin-producing cyanobacteria e.g. *Microcystis* and other prokaryotes (Kardinaal et al., 2007).

The 16S rRNA gene technique of library construction and diversity analysis involves the construction of primer sets from the variable region of the 16S rRNA gene, followed by the sequencing of the amplified products (Semenova et al., 2001). The 16S rRNA gene is ubiquitous to all microbes except viruses therefore used as the signature sequence for taxonomic characterization of microorganisms (Svenning et al., 2005). Various online tools and software packages such as *SILVIA*, *GreenGenes*, and *EzBioCloud* serve as the repository of 16S rRNA gene sequences of most of the microbes, which can be exploited for designing primers for diversity analysis (Yoon et al., 2017). In general, the fingerprinting of the microbial community relies on the PCR amplification of 16S rRNA gene fragments and evaluating the banding patterns on gel electrophoresis (Kumari and Rai, 2019). Further, researchers have employed quantitative RealTime PCR (qPCR) using a specific set of 16S rDNA-designed primers (Anand et al., 2019). The qPCR is a sensitive technique that evaluates diversity based on fold change in the expression of the concerned in the heterogeneous collection of microbial RNA samples (Palinska et al., 2018). Both the techniques are now being extensively used to analyse diversity in various prokaryotes and cyanobacteria such as *Nostoc* and *Synechococcus* (Palinska et al., 2018). However, amplification efficiency of both the techniques depends upon the quality, quantity, and purity of DNA/RNA samples and specific primers.

Gel-free techniques

Fluorescence *in situ* hybridization (FISH) is a molecular cytogenetic tool that detects specific DNA positions on the chromosome using sequence-specific fluorescent probes (Kumari and Rai, 2019). Several researchers have exploited FISH as a tool for species identification using a specifically designed oligonucleotide or polynucleotide probes, which are fluorescently labelled at 5' end using fluorescein and rhodamine (Threon and Cloete, 2000).

The FISH technique has been successfully used for the taxonomic classification of *Nodularia* strains from brackish water and several other cyanobacterial species using DNA-DNA and DNA-RNA hybridization (Suda *et al.*, 2002). Microarray is a high-throughput technique that has been extensively employed for microbial diversity assessment using a specific antibody against the gene of interest (Anand *et al.*, 2019). Microarray is gene-chip technology that can test many biological samples by performing DNA-DNA, DNA-RNA, and DNA-protein hybridization (Baudart *et al.*, 2017). This technique has contributed significantly to revealing the diversity of freshwater pathogens, including cyanobacteria, based on the hybridization of toxin encoding genes (Medlin, 2018). However, the efficiency of microarray techniques' depends on the specificity of the probe designed for the molecular assessment of the diversity.

Metagenomics is DNA sequencing-based technology and a culture-independent tool for analysing DNA samples isolated directly from the environmental samples (Lu *et al.*, 2016). This technique performs taxonomic and diversity analysis in detail and provides valuable insight into their physiology by studying the expression pattern of genes under distinct environments (Lu *et al.*, 2016). Compared to other molecular diversity assessment tools such as RISA, RFLP, DGGE, and 16S rRNA gene, metagenomics provides comprehensive information about the biodiversity and physiological behaviour of the concerned microbes (Laver *et al.*, 2015). Metagenomics techniques involve DNA/RNA library preparations followed by their assembly, sequencing, and data interpretations. The metagenomics technique is a complex molecular diversity analysis tool that is less biased than PCR and offers in-depth information related to the physiology and diversity of microbial communities.

Researchers have also identified and exploited various PCR-based molecular markers for cyanobacterial diversity analysis in conjunction with the earlier techniques. For example, 16SrRNA-based molecular markers are extensively used to decipher phylogenetic relationships (Komarek, 2016). Likewise, molecular identification based on the RNA polymerase β subunit (*rpoB*) gene has also been used to study cyanobacterial diversity under different environmental conditions (Lyra *et al.*, 2005). The *rpoB* gene is highly conserved and ubiquitously distributed, offering a more significant advantage over the 16S rRNA gene for diversity analysis

(Kumari and Rai, 2019). Microbiologists have also designed molecular markers based on nitrogen-fixing (*nif*) genes and have used them to analyse diversity among different microbial communities (Anand *et al.*, 2019). A marker based on the *nifH* gene has also been identified in cyanobacteria such as *Anabaena variabilis*, and other photosynthetic cyanobacteria to gain valuable insight into their phylogenetic relationship by correlating the data with morphological and biochemical characters (Thiel *et al.*, 1995).

Conclusion

Cyanobacteria represent immense diversity, as exhibited by the high degree of morphological variations. Quantifying their number and types within a community is central to understanding an ecosystem's structure and function. Recent advancements in molecular biology tools, especially in genomics, metagenomics, and proteomics, are imperative for the taxonomic assessment of cyanobacteria by outlining more significant differences among the lineages. These molecular techniques can precisely resolve species relationships by decoding their genome and can provide an in-depth understanding of cyanobacterial diversity under extreme environments.

Acknowledgments

LC Rai thanks the National Academy of Science, India for the NASI Senior Scientist Platinum Jubilee Fellowship, the Indian Council of Agricultural Research-National Bureau of Agriculturally Important Microorganisms (ICAR-NBAIM) for financial support; Dr Krishna Kumar Rai is thankful to the National Academy of Science, India for RA ship. Dr Ruchi Rai is thankful to DST, New Delhi, for Women Scientist Scheme A (WOSA) award. Dr Shilpi Singh thanks CSIR for the Senior Research Associate award.

References

- Anand N, N Thajuddin, PK Dadheech (2019) Cyanobacterial taxonomy: morphometry to molecular studies. In: Mishra AK, DN Tiwari, AN Rai, (eds.) *Cyanobacteria—From Basic Science to Applications*. Academic Press, Elsevier, pp. 43–64.
- Baudart J, D Guillebault, E Mielke, T Meyer, N Tandon, S Fischer *et al* (2017) Microarray (phylochip) analysis of freshwater pathogens at several sites along the Northern German coast transecting both estuarine and freshwaters. *Appl. Microbiol. Biotechnol.* **101**: 871–886.
- Demay J, C Bernard, A Reinhardt, B Marie (2019) Natural products from cyanobacteria: Focus on beneficial activities. *Mar. Drugs* **17**: 320.

- Gaysina LA, A Saraf, P Singh (2019) Cyanobacteria in diverse habitats. In: Mishra AK, DN Tiwari, AN Rai (Eds.) *Cyanobacteria—From Basic Science to Applications*. Academic Press, Elsevier, pp. 1–28.
- Jungblut AD, I Hawes, D Mountfort, B Hitzfeld, DR Dietrich, BP Burns, BA Neilan (2005) Diversity within cyanobacterial mat communities in variable salinity meltwater ponds of McMurdo Ice Shelf, Antarctica. *Environ. Microbiol.* **7**: 519–529.
- Kardinaal WEA, I Janse, M Kamst-van Agterveld, M Meima, J Snoek, LR Mur, J Huisman, G Zwart, PM Visser (2007) Microcystis genotype succession in relation to microcystin concentrations in freshwater lakes. *Aquat. Microb. Ecol.* **48**: 1–12.
- Komárek J (2016) A polyphasic approach for the taxonomy of cyanobacteria: principles and applications. *Eur. J. Phycol.* **51**: 346–353.
- Kumari N, LC Rai (2020) Cyanobacterial diversity: molecular insights under multifarious environmental conditions. In PK Singh (ed.) *Advances in cyanobacterial biology*. Academic Press, pp. 17–33.
- Kumari N, OP Narayan, LC Rai (2012) Cyanobacterial diversity-shifts induced by butachlor in selected Indian rice fields in eastern Uttar Pradesh and western Bihar analyzed with PCR and DGGE. *J. Microbiol. Biotechnol.* **22**: 1–12.
- Laver T, J Harrison, PA O'Neill, K Moore, A Farbos, K Paszkiewicz (2015) Assessing the performance of the Oxford Nanopore Technologies MinION. *Biomol. Detec. Quantif.* **3**: 1–8
- Lu H, F Giordano, Z Ning (2016) Oxford Nanopore MinION sequencing and genome assembly. *Genom. Proteom. Bioinform.* **14**: 265–279.
- Lukow T, PF Dunfield, W Liesack (2000) Use of the T-RFLP technique to assess spatial and temporal changes in the bacterial community structure within an agricultural soil planted with transgenic and non-transgenic potato plants. *FEMS Microbiol. Ecol.* **32**: 241–247.
- Lyra C, M Laamanen, JM Lehtimäki, A Surakka, K Sivonen (2005) Benthic cyanobacteria of the genus *Nodularia* are non-toxic, without gas vacuoles, able to glide and genetically more diverse than planktonic *Nodularia*. *Int. J. Syst. Evol. Microbiol.* **55**: 555–568.
- Madigan MT, DP Clark, D Stahl, JM Martinko (2010) Brock biology of microorganisms. 13th edition. Benjamin Cummings.
- Medlin LK (2018) CYANO RT-Microarray: A Novel Tool to Detect Gene Expression in Cyanobacteria. *Environ. Microbiol.* **1**: 17–21.
- Neilan BA, D Jacobs, AE Goodman (1995) Genetic diversity and phylogeny of toxic cyanobacteria determined by DNA polymorphisms within the phycocyanin locus. *Appl. Environ. Microbiol.* **61**: 3875–3883.
- Palinska KA, JC Vogt, W Surosz (2018) Biodiversity analysis of the unique geothermal microbial ecosystem of the Blue Lagoon (Iceland) using next-generation sequencing (NGS). *Hydrobiologia* **811**: 93–102.
- Rott E, A Pentecost, J Mareš (2018) Introduction: Recent developments in cyanobacterial research with special reference to aquatic habitats, molecular ecology and phylogenetic taxonomy. *Hydrobiologia* **811**: 1–6.
- Sallal AK, NA Nimer, SS Radwan (1990) Lipid and fatty acid composition of freshwater cyanobacteria. *Microbiology* **136**: 2043–2048.
- Semenova EA, KD Kuznedelov, MA Grachev (2001) Nucleotide sequences of fragments of 16S rRNA of the Baikal natural populations and laboratory cultures of cyanobacteria. *Mol. Biol.* **35**: 405–410.
- Song T, L Mårtensson, T Eriksson, W Zheng, U Rasmussen (2005) Biodiversity and seasonal variation of the cyanobacterial assemblage in a rice paddy field in Fujian, China. *FEMS Microbiol. Ecol.* **54**: 131–140
- Soo RM, J Hemp, DH Parks, WW Fischer, P Hugenholtz (2017) On the origins of oxygenic photosynthesis and aerobic respiration in cyanobacteria. *Science* **355**: 1436–1440.
- Srivastava M, MS Kaushik, AK Mishra (2016) Linking the physicochemical properties with the abundance and diversity of rhizospheric bacterial population inhabiting paddy soil based on a concerted multivariate analysis of PCR-DGGE and RISA. *Geomicrobiol. J.* **33**: 894–905.
- Suda S, MM Watanabe, S Otsuka, A Mahakahant, W Yongmanitchai, N Nopartnaraporn, Y Liu, JG Day (2002) Taxonomic revision of water-bloom-forming species of oscillatorioid cyanobacteria. *Int. J. Syst. Evol. Microbiol.* **52**: 1577–1595.
- Svenning MM, T Eriksson, U Rasmussen (2005) Phylogeny of symbiotic cyanobacteria within the genus *Nostoc* based on 16S rDNA sequence analyses. *Arch. Microbiol.* **183**: 19–26.
- Theron J, TE Cloete (2000) Molecular techniques for determining microbial diversity and community structure in natural environments. *Crit. Rev. Microbiol.* **26**: 37–57.
- Thiel T, EM Lyons, JC Erker, A Ernst (1995) A second nitrogenase in vegetative cells of a heterocyst-forming cyanobacterium. *Proc. Natl. Acad. Sci.* **92**: 9358–9362.
- Yoon SH, SM Ha, S Kwon, J Lim, Y Kim, H Seo et al (2017) Introducing EzBioCloud: A taxonomically united database of 16S rRNA and whole genome assemblies. *Int. J. Syst. Evol. Microbiol.* **67**: 1613–1617.

Synthetic Microbial Community (SynCom) for Sustainable Agriculture

Nunna Sai Aparna Devi and Dananjeyan Balachandar*

Department of Agricultural Microbiology, Tamil Nadu Agricultural University, Coimbatore-641003, Tamil Nadu, India

Agricultural sustainability is possible only when reducing crops' dependency on chemical fertilizers, improving crops' ability to grow on marginal soil types, and improving their resilience against biotic and abiotic stresses. Though bio inoculants play a crucial role in agricultural sustainability, they are still limited due to low reproducibility in fields due to different crops and cultivars, soil types, and agro-ecological conditions. Traditionally, inoculants are developed through a single isolate study model, i.e., *in vitro* screening of strains for plant-growth promotion, omitting their interaction with the host plant and soil ecosystem. Hence, a paradigm shift is needed in developing the inoculant to improve the microbe-mediated crop fitness and productivity. In this paper, we framework the synthetic microbial community (SynCom) as a potential resource for developing ecology-based inoculants to augment nutrient acquisition, drought mitigation, and pathogen resistance of crops to ensure sustainability.

Key Words: Biocontrol agent, Biofertilizer, Core microbiome, Plant holobiont, Plant microbiome, Synthetic microbial community (SynCom)

Introduction

Exploring the soil and plant-associated microbes to improve agriculture productivity is an extremely attractive approach as it ensures sustainability. Till now, the plant-growth promoting bacteria residing in the crop's sphere and soil were isolated, screened, and commercially utilized as 'biofertilizers', or 'biocontrol agents' (collectively referred to as microbial inoculants). This conventional approach-based commercial formulation could ensure a 10-15% yield increase with a 15-20% reduction in chemical fertilizer usage. The inconsistent performance of these inoculants within and between crops and different soil types leads to low farmers' preferences. Though the microbial inoculants have the potential, it is believed that we have not yet explored them to replace the chemical fertilizers completely. Hence, a paradigm shift in developing microbial inoculants is needed to extend its benefits beyond the present scope. Synthetic microbial community (SynCom) is a recently introduced approach that facilitates to design the microbial inoculant using microbial ecology and genetics.

What is SynCom?

The plant recruits its microbial partners through the release of nutrient-rich rhizo-deposits. Upon colonization, a range of microbial functions modulates the phenome

of the plant positively to enhance the nutrient and water uptake, growth, and health. These microbial assemblages also mitigate environmental stresses. The plant has a strategy to recruit its core microbiome assemblage. The genetic and physiological traits of the plant are crucial for the recruitment and microbial reservoir of the soil. So, the holobiont (plant with its core microbiome) functioning of crop plants has significance in agricultural sustainability. However, the complexity of plant microbiome in terms of community and functioning often does not allow to proceed the resources for application in agriculture. The synthetic microbial community represents the less-complex consortium selected from the core microbiome of a plant with uncompromised functioning as that of the core microbiome. SynCom is an ecology and genetic-based approach to developing the inoculant.

SynCom Inoculants

Based on the number of strains being used for SynCom development, the SynComs were grouped as small (less than 10), medium (10-100), and large (more than 100) SynComs (Table 1). The large SynComs were tested in Arabidopsis to understand the basic features of SynCom. At the same time, the small and medium SynComs were potentially explored in several crops with a specific beneficial target.

*Author for Correspondence: Email-dbalu@tau.ac.in

SynCom Development Strategies

The roadmap for successful SynCom-based inoculant development is (1) understanding the microbiome of a crop through culturable and metagenomic approaches; (2) Identifying and tailoring the microbiome of a crop; (3) screening the potential candidates for persistence and specific benefits; (4) designing the SynCom inoculant, (5) understanding SynCom-crop interaction (6) Assessing the influence of SynCom inoculant on crop's physiology and productivity. This bottom-up approach will develop an efficient SynCom-based inoculant for low-input sustainable rice production (Fig. 1).

Future Prospects

Though SynCom is accepted as a potential alternate for present inoculants to improve sustainability in agriculture, several researchable issues and technological gaps are still to be addressed to make SynCom viable and sustainable technology for agricultural application.

1. **Candidate selection for SynCom:** The plant microbiome comprises rhizospheric, epiphytic, endophytic and phyllospheric microbes, and the abundance and diversity are dynamic. Under this condition, selecting candidates from a plant's core microbiome is challenging. The modern tools, including omic approaches, bioinformatics, and modelling, will pave the way to identifying the possible potential candidates for SynCom.
2. **Number of species in SynCom:** As SynCom is a reductionist approach, using large SynCom (>100) would always ensure better results across the crops, soils, and environmental conditions. However, managing a large number of strains and their equimolar ratio would be a challenging issue.
3. **Microbe-microbe interactions:** The interaction between two strains and among more than two strains would be complex in nature. Understanding the overall interactions of SynCom candidates across the cell density would be a challenging and decisive factor for SynCom's performance.
4. **SynCom as functional representative of the core microbiome:** SynCom should have a limited number of core microbiome candidates. But, it should function in the plant ecosystem like that of the original microbiome without compromising the overall functionality.
5. **Role of each taxon in SynCom:** The contribution of each strain (or taxon) present in the SynCom should be decoded. For this, the drop-out experiment (SynCom without one strain) could be more helpful.
6. **SynCom stability:** SynCom should be robust and prevalent throughout the crop growth stages. The inoculated community should not

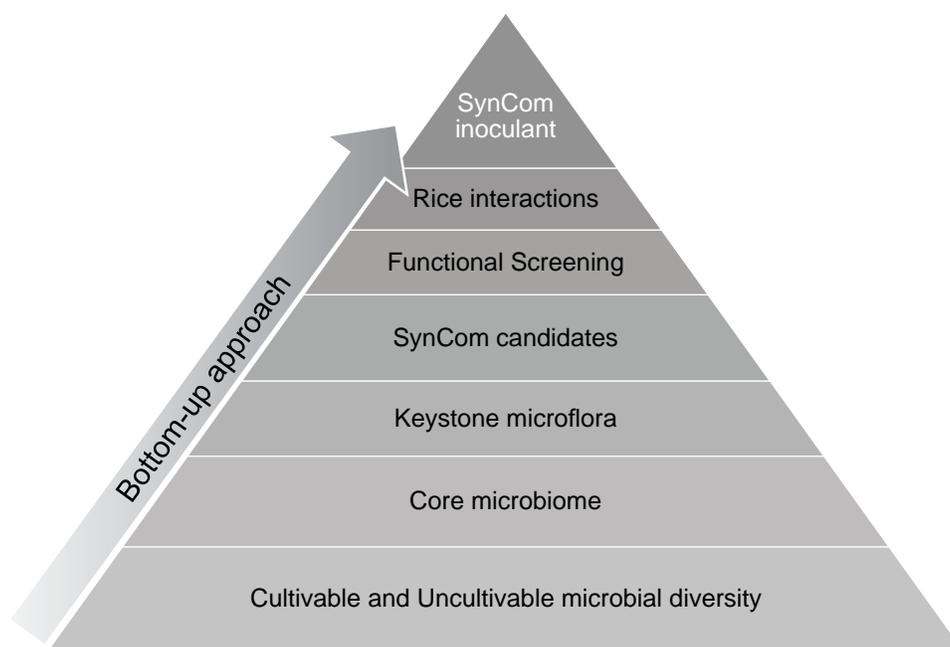


Fig. 1. The proposed model describing the bottom-up approach for SynCom inoculant development

Table 1. Overview of some SynComs and their functioning in plants

Type of SynCom	SynCom candidates		Model plant	Significance	Reference
	No. of species	Candidates			
Small	6	Pseudomonads	Garlic	Plant growth	Zhuang <i>et al.</i> (2020)
Small	8	<i>Williamsia, Pantoea</i>	Alfalfa	Stability of Syncom; Species domination	Moccia <i>et al.</i> (2020)
Medium	12	<i>Bacillus, Paenibacillus, Pantoea, Bradyrhizobium, Kluyvera, Actinobacter, Streptococcus, Chryso bacterium, Spingobium.</i>	Soybean	Stability of SynCom, N and P acquisition	Wang <i>et al.</i> (2021)
Medium	15	Members of Actinobacteria, Alpha, beta and gamma proteobacteria, Firmicutes	Tomato	Salt stress resilience	Schmitz <i>et al.</i> (2022)
Large	183	Members of rhizosphere microflora	Arabidopsis	Tryptophan metabolism	Wolinska <i>et al.</i> (2021)
Large	188	Leaf and root derived microflora	Arabidopsis	SynCom colonization pattern	Bai <i>et al.</i> (2015)

be overcome by the native soil microbiome. Hence, efforts should be taken to identify such candidates during SynCom development.

- Validation of SynCom:** To validate the performance of SynCom in terms of stability, robustness in colonization and effectiveness, sequencing techniques and quantitative PCR-based assays can be performed.
- Resources for SynCom:** The microbiome of wild species and landraces of a crop have co-evolved with their host plants. These microbes are well-adapted to extreme environments and excel multiple benefits to the plants. Hence, exploring these microbiomes for SynCom development could be a practical approach for agricultural purposes.
- SynCom–Crop interactions:** Understanding the molecular talk between the SynCom community and the host plant is essential to assess SynCom’s effectiveness. The transcriptomic, proteomic,

and metabolomic tools can be explored for this purpose.

- Scale up of SynCom:** Industrial scale-up of all the strains of SynCom formulated as a single product is a significant constraint. For this, the use of a minimal number of candidates would be recommended.

Microbial Inoculants versus SynCom Inoculants

Microbial inoculants are inevitable bio-inputs for sustainable agriculture. They are economically viable technology, successfully demonstrated for their nutrient supplementation and stress management in several crops. However, their potential is less explored due to several environmental, scientific, and social issues. SynCom approach will resolve the deficits of current inoculant technology (Table 2).

References

de Souza RSC, JSL Armanhi and P Arruda (2020) From microbiome to traits: designing synthetic microbial

Table 2. Comparison between the present microbial inoculants and Syncom inoculants

Microbial inoculant	SynCom inoculant
One or two potential strains	Group of strains as a consortium
Develop through single isolate study model	Develop through ecology-based study
Potential strains from culturable plant microbiome	Involves both culturable and culture-independent tools to identify the strains
Each strain with specific beneficial trait	A Group of strains provides multiple benefits to crop on a labor-sharing basis
No need to be the representative of the core microbiome	Functional representatives of the core microbiome.
Top-down approach and inoculant development and evaluation are easy and quick.	The bottom-up approach needs a robust and methodological approach for inoculant development and evaluation.
Performance varies depending on crops, cultivars, management, soils, and agro-ecological conditions	Consistent performance is possible across varied crops and soils.
Less interaction with soil ecosystem and hence less persistence	High persistence due to high interaction with soil ecosystems
Uncertainty in robust colonization and prevalence throughout the crop’s growth stages.	Ensures robust colonization and prevalence.
It can be supplementary for chemical fertilizers and synthetic chemicals.	Potential substitute the chemical fertilizers and synthetic chemicals

- communities for improved crop resiliency. *Front. Plant Sci.* **11**: 1179.
- Pozo MJ, I Zabalgogazcoa, BR Vazquez de Aldana and Martinez-Medina A (2021) Untapping the potential of plant mycobiomes for applications in agriculture. *Curr. Opin. Plant Biol.* **60**: 102034.
- Ray P, V Lakshmanan, JL Labbé and KD Craven (2020) Microbe to microbiome: A Paradigm Shift in the application of microorganisms for sustainable agriculture. *Front. Microbiol.* **11**: 622926.
- Saad MM, AA Eida and H Hirt (2020) Tailoring plant-associated microbial inoculants in agriculture: a roadmap for successful application. *J. Exp. Bot.* **71**: 3878-3901.
- Schmitz L, Z Yan, M Schneijderberg, *et al.* (2022) Synthetic bacterial community derived from a desert rhizosphere confers salt stress resilience to tomato in the presence of a soil microbiome. *ISME J.* doi: 10.1038/s41396-022-01238-3.
- Suman A, V Govindasamy, B Ramakrishnan, K Aswini, J Sai Prasad, P Sharma, D Pathak and K Annapurna (2022) Microbial community and function-based synthetic bioinoculants: a perspective for sustainable agriculture. *Front. Microbiol.* **12**: 805498.

Key Words

Biofertilizer: Commercial preparation containing microorganisms used to provide macro- and micronutrients to the crops.

Biocontrol agent: Commercial microbial preparation applied to crops for pest and disease management.

Core microbiome: Represents the keystone microbes established through evolutionary mechanisms of selection that are important for plant fitness. These species harbor important genes for the fitness of the plant.

Microbial inoculant: The commercial preparation containing microorganisms applied to improve the crop's nutrient uptake, biotic and abiotic stress amelioration and to improve soil health.

Plant holobiont: plant and its associated microbiomes as a single ecological and evolutionary unit.

Plant microbiome: Refers the characteristic microbial community of specific ecological niches of plant. It also includes its functionality. The plant microbiome can be from rhizosphere, phyllosphere, epiphytic and endosphere.

Rhizosphere: The thin layer of soil in close proximity of plant root which is directly influenced by the root.

Synthetic microbial community (SynCom): Represents the consortia of microorganisms designed to mimic, at some scale, the observed function and structure of the microbiome in natural conditions.

Microbiome and Plant Health

Pushp Lata¹, Kiran Bala², Piyush Goel³, Utkarsh Sood^{4,5}, Princy Hira⁵, Vatsala Dwivedi⁶, Abhilash Kumar⁷, Charu Dogra Rawat⁷ and Rup Lal^{*1,4}

¹Department of Zoology, University of Delhi, New Delhi-110007, India

²Department of Zoology, Deshbandhu College, University of Delhi, New Delhi-110059, India

³Department of Biosciences and Biomedical Engineering, Indian Institute of Technology Indore, Simrol, Indore-453552, India

⁴Phixgen Pvt. Ltd., Gurugram-122001, Haryana, India

⁵Department of Zoology, Maitreyi College, University of New Delhi-110021, India

⁶Department of Zoology, Dayal Singh College, University of New Delhi-110003, India

⁷Department of Zoology, Ramjas College, University of New Delhi-110007, India

Recent researches have shown that plants are associated with plethora of microorganisms also called plant microbiome. Plant microbiome interactions primarily affect plant growth, development, plant -fitness and productivity. Depending upon the specific habitat of plant, microbial communities are mainly predominant in three different regions namely rhizosphere, phyllosphere and endosphere. The full range of microbes and their association with plants and hence maintenance of crop productivity has recently begun to be explored. In this article, we primarily focus on the interaction of plants and root associated bacteria enhancing plant mineral nutrition, summarizing the current knowledge in several research fields that can converge to improve our understanding of the molecular mechanisms underpinning this phenomenon.

Keywords: Inoculum, Microbiome, Plant health, Rhizosphere, Seed, Phyllosphere

Introduction

The plant microbiome also known as phytomicrobiome has recently become a very important aspect of research with plant scientist. This is mainly due to the strong evidences that have emerged that plant microbiome plays a very important role in maintaining plant health and crop production (Witton *et al.*, 2018). A better understanding of the plant microbe interactions and plant microbiome is expected to contribute significantly to the crop productivity in future. This article gives a glimpse of the recent developments in this area to indicate that this area is extremely important to explore in order to increase crop productivity in future.

The Plant Microbiome

The plant microbiome comprises of huge amounts of beneficial, commensal and pathogenic microorganisms that play critical essential roles in plant growth and maintaining plant health. (Naylor *et al.*, 2022). Recent studies have revealed that soil microorganism in association with plant microbiome especially in the rhizosphere are the facilitators of several plant processes. This they do by secreting hormones like auxins that help

in root growth changes. Some microbes take up iron from the soil and they solubilise phosphorus and make this nutrient available to the plants (Nelson, 2018). Plant microbiome provides protection against pathogens to the plants by indirectly activating the immune responses. The conversion of atmospheric nitrogen to ammonia is also taken by nitrogen fixers.

The plant microbiomes are very specific and their role to complex interconnected microbial networks that are important in plant health and ecosystem functioning has recently been explored (Babalola *et al.*, 2020). It will not be out of place to mention here that the plant microbiome is also specific to the genotype of plant, plant species, edaphic and other environmental factors, but the roles of this makeup are difficult to separate from each other in natural environments (Compant *et al.*, 2019). These microbial inhabitants in soil and around the plants are also responsible for production of antibiotics and fungal cell wall degrading enzymes. It has also been reported that by changing the genetic makeup of keystone microbial community, the overall performance of the plant can be enhanced. There are

*Author for Correspondence: Email-ruplal@gmail.com

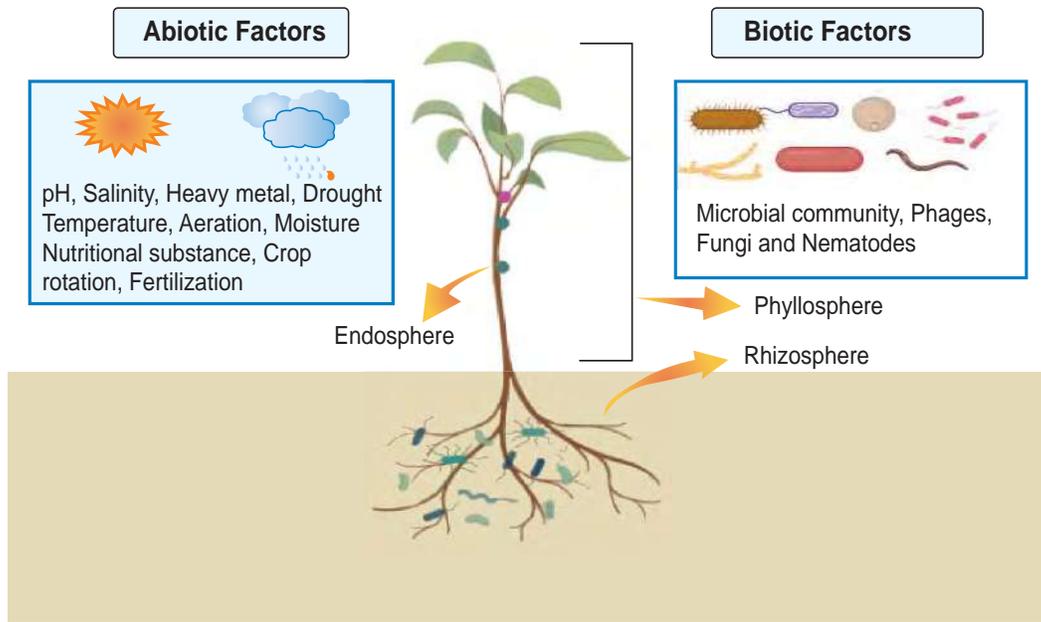


Fig. 1. Biotic and abiotic factors affecting the plant-microbe interaction

various biotic and abiotic factors that affect the soil microbiome interaction (Fig. 1).

The plant microbiome is primarily divided into: Rhizosphere, phyllosphere, endosphere and seed microbiome.

Rhizosphere Microbiome

Rhizosphere microbiome is very important for maintaining plant health. For instance, rhizosphere microbiome interactions have been proved to be beneficial in increasing stress tolerance and providing pathogenic resistance to plants. (Qu *et al.*, 2020) It was observed that in soyabean rhizosphere, the presence of limonene, naphthalene and pinene resulted in increased detoxification, reduced carbon metabolism pathways such as starch, mannose pathways and the available compounds were used as a carbon source. Rhizosphere bacteria like *Pseudomonas fluorescens* have utilised pinene as the source of carbon while the naphthalene was metabolized by several other bacterial genera including *Ralstonia*, *Pseudomonas*, *Streptomyces*, *Mycobacterium* etc (Lui *et al.* 2019). In other study, it was observed that the disease resistance in Cassava against *Xanthomonas axonopodis* pv. *manihotis* (Xam) was conferred by the *Lactococcus* sp., which produces nisin, a lantibiotic. It was also observed that the amylose content of cassava can be attributed to *Enterobacter cloacae* while the starch content and viscosity was provided by *Pseudomonas* and *Stenotrophomonas* sp (Zhang *et al.*,

2021). Genera like *Lysobacter* and *Aeromicrobium* which are prevalent in high pH agricultural soils produces some secondary metabolites. These metabolites are responsible for antimicrobial activities against many soil-borne pathogens in common bean (*Phaseolus vulgaris*) plant (Pérez-Jaramillo *et al.*, 2019). Recent-rhizospheric microbiome analysis of maize plant revealed that the most recent germplasm has evolved mechanisms for reduced nitrogen conversion and enhanced amino-acid synthesis when compared to the decade old microbiome (Van *et al.*, 2018). It suggested that the maize plant has a microbiome full of copiotrophs which resulted in decreased yields in modern agro-systems. Such results demand for inter-breeding processes to enhance the microbiome and can lead to differential soil-management practices. Rhizospheric microbiome can also act as a good indicator of soil-pollution and hence plant health (Wang *et al.*, 2019). Bamboo soil contaminated with heavy-metals like chromium has shown lesser concentrations of phylum Proteobacteria. Some species of phylum Acidobacteria (*Bryobacter* and *Granulicella*) and Actinobacteria (*Lamia* and *Arthrobacter*) have the ability to reduce the heavy metals like chromium in soil (Zhang *et al.*, 2020). Phylum Chloroflexi is known to perform anoxygenic photosynthesis using several inorganic elements as the electron donors. Drought-stress tolerance can also be brought about by engineering the rhizospheric microbiome of plants. Microbiota of phylum Betaproteobacteria and Actinobacteria

produces extracellular polysaccharides which resulted in better plant growth, enhanced water retention and high respiration rate in wheat seedlings (Jochum *et al.*, 2019).

Phyllosphere Microbiome

The plant leaf surface or phyllosphere represents a distinctive and diverse microbiome even though it is exposed to various abiotic stresses such as nutrient stress, ultraviolet radiations and desiccation. Bacteria, fungi, viruses, and cyanobacteria are the predominant colonizers of phyllosphere. Studies reveal that amongst bacteria, Proteobacteria (alpha-, beta-, and gammaproteobacteria), Bacteroidetes, Actinobacteria, and Firmicutes are the dominant phyla with *Bacillus* and *Pseudomonas* representing the most abundant genera. The other common species found are *Pantoea*, *Erwinia*, *Sphingomonas*, *Acinetobacter*, *Xanthomonas*, and *Gluconobacter*. (Thapa *et al.*, 2018). Yeast like fungus *Aureobasidium pullulans* is dominant in the phyllosphere as well on the surface of fruits. Common fungal genera occurring in the leaves are *Cladosporium*, *Alternaria*, *Penicillium*, *Acremonium*, *Mucor*, and *Aspergillus*, whereas commonly occurring yeasts are *Cryptococcus*, *Sporobolomyces*, and *Rhodotorula* (Thapa *et al.*, 2018). The diversity of Cyanobacteria is determined by the species of plant it inhabits and also on ecosystem. The dominant orders are Nostocales and Oscillatoriales (Stone *et al.*, 2018). These microbes interact with the host plant in several ways and promote plant growth directly by modulating phytohormones, improved nutrient acquisition by increasing their bioavailability and indirectly by pathogen suppression (Bashir *et al.*, 2022). The analysis of phyllosphere microbiome and community plays a vital role in framing policy related to sustainable agriculture and design of consortia for their use as inoculants for rhizoremediation, phytostimulation, and biocontrol and overall plant growth enhancement. Recent evidence suggests that neighboring plants also modulate the host phyllosphere microbiota via aerial dispersal and genetic factors (Shakir *et al.*, 2021; Meyer *et al.*, 2022).

Endosphere Microbiome

Complex microbial communities and microorganisms colonize internal parts of the plants (endosphere) and play significant role in plant's growth and health by, for example, making the plants resistant to many diseases or by other mutualistic interactions (Hassani *et al.*, 2018).

Studying endosphere microbiome, thus, constitutes an important aspect to understand and modulate plant traits, physiology and ecosystem functioning.

The structure of endosphere microbiome was found to be continuously varying during growth and wilting processes in plants (Liu *et al.*, 2019). For instance bacterial and fungal diversity increased with the development of banana plantlets with bacterial groups belonging to Enterobacteriaceae family found to be present with different relative abundance in all the samples (Dong *et al.*, 2019). The endosphere microbiome was engineered to improve *Fusarium* wilt resistance in banana plant (Liu *et al.*, 2019). Inoculation with engineered *Enterobacter sp.* and *Kosakonia sp.* for expression 1-Amino Cyclopropane-1-Carboxylate (ACC) deaminase on their cell walls increased resistance to *Fusarium* wilt disease as compared to controls without inoculations (Liu *et al.*, 2019). Similarly, *Pseudomonas* strain (LTGT-11-2Z) was found to be widespread in root endosphere microbiome of a desert plant *Alhagis parsifolia*. LTGT-11-2Z was found to promote drought resistance in wheat plant which pointed towards its potential to be a biotechnological agent that can make crop plants more resistant to drought conditions which would be very useful for agriculture (Zhang *et al.*, 2020).

To ascertain the influence of soil on the diversity of endosphere microbiome of roots and leaves (Pangesti *et al.*, 2020) mixed sterilize soil with 10% grass-grown soil and planted *Chrysanthemum* plants in the mixed soils. Control plants were raised in sterilized soils only. All the plants were exposed to thrips and then roots and leaves were collected from both the *Chrysanthemum* and control plants. Analysis of endosphere microbiome revealed increase in bacterial diversity post-inoculation only in the roots and not in the leaf tissues. Endosphere of both roots and leaves showed dominance of the Pseudomonadaceae family. In case of leaves Pseudomonadaceae was higher in abundance in inoculated plants than in control ones. Whereas in case of roots, abundance is higher in control plants. Rare bacterial families were mostly found in leaves, showing that bacterial communities in roots and leaves differed substantially and thus had differential impacts on their growth (Pangesti *et al.*, 2020).

Recently (Goodwin, 2022), endosphere microbiome study of Ginseng (*Panax sp.*) has shown that large number and types of fungal, bacterial, archeal and viral endophytes are present in its endosphere. Bacterial and

fungal endophytes are abundantly present in its roots and some of these endophytes are specific to tissues present above and below the ground. Many endophytes are found to metabolize ginsenosides which are major active pharmacological component of Ginseng. The ability of endosphere microbes to produce/metabolize ginsenosides shows that they acquire certain specific or altered genes which help them to grow in Ginseng apoplast.

It is thus very clear that endosphere microbiome studies, provide information for better understanding of the endophytes and plants interactions, adaptations and evolution. This can contribute towards designing and developing strategies for more sustainable microbe-based agriculture.

Seed Microbiome

Microbiome in plants has been extensively studied as microbes associated with both root (rhizosphere) and shoot (phyllosphere). To date, very less emphasis is given to the role of the microbiota that exists as part of the seed. Recent studies have shown that microbiomes affect the germination, health, and productivity of the plants in both agricultural and natural ecosystems (Nelson, 2018)

As all plant tissues have been reported to have endophytic and epiphytic microbial associations, seeds have also been reported to have both types of microbial associations reported as Endospermosphere and Spermosphere (Nelson, 2018). Seeds of different plants are associated with diverse microbial communities that depend on the genotype of the plant, its environment, and to some extent anthropogenic interventions. Bacterial and fungal populations are reported to be the abundant inhabitants of the seed microbiome, however, in the seeds of native alpine plants, Archaea has also been reported as newly discovered seed endophytes (Wassermann et al., 2019).

Deciphering the healthy microbiome can be essential for improving crop yield, and improved plant tolerance to biotic and abiotic stress. It can be targeted by harnessing seed microbiomes of wild crop cultivars and re-introduce missing beneficial seed microbes (Berg and Raaijmakers, 2018). The role of soil has also been shown to be pertinent as the transmission of microbiota to seed is majorly from the soil (Rocheffort et al. 2021). Recently, it has been shown that the endophytic seed microbiome can be altered by abiotic treatment and also by climatic stress (Bintarti et al., 2022)

Seeds being the carrier of the next generation of the plants and act as a primary inoculant for microbiota, future 'omic' studies targeted on seeds and will surely play a significant role in microbiome and plant.

References

- Bashir I, AF War, I Rafiq, ZA Reshi, I Rashid, YS Shouche (2022) Phyllosphere microbiome: Diversity and functions. *Microbiol. Res.* **254**: 126888.
- Berg, G and JM Raaijmakers (2018) Saving seed microbiomes. *ISME J* **12**: 1167–1170.
- Bintarti AF et al. (2022) Abiotic Treatment to Common Bean Plants Results in an Altered Endophytic Seed Microbiome. *Microbiol. Spectr.* **10**: e0021021.
- Compant S, MC Cambon, C Vacher, B Mitter, A Samad, and A Sessitsch (2021) The plant endosphere world–bacterial life within plants. *Environ. Microbiol.* **23**: 1812–1829.
- Dong CJ, LL Wang, Q Li and QM Shang (2019) Bacterial communities in the rhizosphere, phyllosphere and endosphere of tomato plants. *PLoS One* **14**: e0223847.
- Liu F, T Hewezi, SL Lebeis, V Pantalone, PS Grewal, and ME Staton (2019) Soil indigenous microbiome and plant genotypes cooperatively modify soybean rhizosphere microbiome assembly. *BMC Microbiol.* **19**: 201.
- Goodwin PH (2022) The Endosphere Microbiome of Ginseng. *Plants* **11**: 415.
- Hassani, M, PDurán and SHacquard (2018) Microbial interactions within the plant holobiont. *Microbiome* **6**: 1-17.
- Pérez-Jaramillo JE, M de Hollander, CA Ramírez, R Mendes, JM Raaijmakers and VJ Carrión (2019) Deciphering rhizosphere microbiome assembly of wild and modern common bean (*Phaseolus vulgaris*) in native and agricultural soils from Colombia. *Microbiome* **7**: 114.
- Zhang L, J Zhang, Y Wei, W Hu, G Liu, H Zeng and H Shi (2021) Microbiome-wide association studies reveal correlations between the structure and metabolism of the rhizosphere microbiome and disease resistance in cassava. *Plant Biotechnol J*, **19**: 689–701.
- Liu Y, A Zhu, H Tan, L Cao and R Zhang (2019) Engineering banana endosphere microbiome to improve Fusarium wilt resistance in banana. *Microbiome* **7**: 1-5.
- Jochum MD, KL McWilliams, EA Pierson, YK Jo (2019) Host-mediated microbiome engineering (HMME) of drought tolerance in the wheat rhizosphere. *PLoS One* **14**: e0225933.
- Meyer KM, R Porch, IE Muscettola, AL Vasconcelos, JK Sherman, CJ Metcalf, SE Lindow and B Koskella (2022) Plant neighborhood shapes diversity and reduces interspecific variation of the phyllosphere microbiome. *ISME J.* **16**: 1376-1387.
- Nelson EB (2018) The seed microbiome: Origins, interactions, and impacts. *Plant and Soil*, **422**: 7–34.
- Pangesti N, A Pineda, SE Hannula and TM Bezemer (2020) Soil inoculation alters the endosphere microbiome of chrysanthemum roots and leaves. *Plant and Soil* **455**: 107-119.

- Pascale A, S Proietti, IS Pantelides and IA Stringlis (2020) Modulation of the root microbiome by plant molecules: the basis for targeted disease suppression and plant growth promotion. *Front Plant Sci.* **10**: 1741.
- Qu Q, Z Zhang, WJGM Peijnenburg, W Liu, T Lu, B Hu, J Chen, J Chen, Z Lin and H Qian (2020) Rhizosphere microbiome assembly and its impact on plant growth. *J. Agric. Food Chem.* **68**: 5024-5038.
- Rocheftort, A. *et al.* (2021) Transmission of Seed and Soil Microbiota to Seedling', *mSystems.* **6**: e0044621
- Shakir S, SS Zaidi, FT de Vries and S Mansoor (2021) Plant genetic networks shaping phyllosphere microbial community. *Trends Genet.* **37**: 306-316.
- Stone BW, EA Weingarten and CR Jackson (2018) The role of the phyllosphere microbiome in plant health and function. *Annual plant reviews online* 533-556.
- Thapa S and R Prasanna (2018) Prospecting the characteristics and significance of the phyllosphere microbiome. *Ann. Microbiol.* **68**: 229-245.
- Van Deynze A, P Zamora, PM Delaux *et al.* (2018) Nitrogen fixation in a landrace of maize is supported by a mucilage-associated diazotrophic microbiota. *PLoS biology* **16**: e2006352.
- Wang L and X Li (2019) Steering soil microbiome to enhance soil system resilience. *Crit. Rev. Microbiol.* **45**: 743-753.
- Wassermann B, T Cernava, H Müller, C Berg and G Berg (2019) Seeds of native alpine plants host unique microbial communities embedded in cross-kingdom networks. *Microbiome* **7**: 108.
- Wei F, L Zhao, X Xu, H Feng, Y Shi, G Deakin, Z Feng and H Zhu (2019) Cultivar-dependent variation of the cotton rhizosphere and endosphere microbiome under field conditions. *Front. Plant Sci.* **10**: 1659.
- Zhang X, F Bian, Z Zhong, X Gai and C Yang (2020) Deciphering the rhizosphere microbiome of a bamboo plant in response to different chromium contamination levels. *J Hazard Mater.* **399**: 123107.
- Zhang L, W Zhang, Q Li, R Cui, Z Wang, Y Wang, YZ Zhang, W Ding and X Shen (2020) Deciphering the root endosphere microbiome of the desert plant *Alhagisparisifolia* for drought resistance-promoting bacteria. *Appl Environ. Microbiol.* **86**: e02863-19

Interlinking Soil Microbial Diversity and Rhizodeposition for Enhancing Nutrient Uptake and Productivity

KK Pal* and Rinku Dey

ICAR-Directorate of Groundnut Research, Junagadh-362001, Gujarat, India

To provide sufficient food and nutrition to an estimated world population of 9.6 billion by 2050, the present level of global food production has to be increased by 50-70%. This monumental task will be abysmally difficult to achieve, owing to the reduction in arable land, increase in frequency and intensity of climate extremes, shrinkage in available agricultural inputs, reduction in soil fertility and organic carbon, predicted inundation of coastal ecosystems due to ingress of seawater, reduction in the diversity of plants due to rapid afforestation, the incidence of newer pests and diseases, etc. Plant species allow specific groups of microorganisms to proliferate in the rhizosphere, depending upon the qualitative and quantitative nature of photo-assimilated carbon flow in the subterranean system in the form of rhizo deposition, the primary source of food and nutrition to the growing organisms. Thus, the quality and quantity of rhizodeposition in a crop ecosystem will determine the diversity of crop-associated rhizosphere microbes vis-à-vis the fate of biogeochemical cycles operating in the rhizosphere, and the availability of essential nutrients for the growing crops to realize genetic yield potential. Therefore, enhancing and or altering the structural and functional diversity of beneficial microbes in the rhizosphere by changing the nature of photo-assimilated carbon in rhizodeposition, quantitatively and qualitatively, through genetic augmentation of crop plants, would facilitate the identification of superior plant varieties for a given ecosystem for ensuring improved plant growth and yield by enhanced nutrient availability.

Introduction

One of the influencing factors that determine the composition of rhizomicrobiome is the composition of rhizodeposition, the primary source of food and nutrition for organisms to proliferate (Tian *et al.*, 2019) in the rhizosphere. The translocation and release of photo-assimilated carbon, entrapping solar energy, water and carbon dioxide, from plant roots into its surroundings is called rhizodeposition, which is composed of root-released tissue cells, sloughed-off cells, mucilages, exudates, lysates, volatile compounds, etc., individually called rhizodeposits. The composition of rhizodeposits varies from plant to plant and within the plant varieties grown in different climatic conditions, quantitatively and qualitatively. Among the rhizodeposits, root exudates are the most dynamic and versatile and the quality and quantity of which vary with plant types and soil, environment, and age of the plants (Crowley and Rengel, 1999). That is why the rhizomicrobiome of rice is different from that of maize, groundnut, soybean, pigeon pea, etc., and even within rice varieties grown in India and Japan. The root exudates include organic compounds (carboxylate anions, phenolics, carbohydrates, amino acids, enzymes, proteins, organic

acids, etc.) and inorganic ions (protons, phosphate, and other nutrients, etc.) (Dakora and Philips, 2002; Bias *et al.*, 2004), the composition of which will vary from plant to plant (Table 1). Though the quantitative and qualitative nature of root exudates significantly influence the functional and structural diversity of microbes in the rhizosphere, other rhizodeposits also play significant roles in disease suppression, production of antimicrobial agents, and messenger molecules for communication, etc. The final product of the rhizodeposition, after microbial utilization, is the soil organic carbon, the composition of which will be determined by the composition of rhizodeposition (Villarino *et al.*, 2021). As soil organic matter content is the indicator of soil health, the composition of rhizodeposition vis-à-vis the composition of soil organic matter will influence the status of soil health. Depending upon the plant species and age of the plant, 20-50% of photo-assimilated carbon is distributed to the roots (Johnson *et al.* 2006)) to sustain the subterranean system. For example, in cereal crops, 50% of the 50% photo-assimilated carbon is required for root respiration and biomass and the rest 50% meant for root rhizodeposition (Fig. 1). However, in legumes, the situation is little different considering the fact that

*Author for Correspondence: Email-kkpal9426476749@gmail.com

Table 1. Composition of root exudates of different plant species (peanut, rape, tomato, flax, etc.) that shape the beneficial microbial dynamics in the rhizosphere

Exudate component	Rhizosphere function	Compounds identified in root exudates	Reference
Organic Acids	Nutrient source, chemo-attractants, chelators, etc.	Citric, oxalic, malic, fumaric, succinic, acetic, butyric, formic, lactic, malonic, glutaric, rosmarinic, vanillic, etc.	Dakora and Philips (2002); Nguyen (2003), Bias <i>et al.</i> (2004)
Amino acids	Nutrient source, chemo-attractants, chelators, etc.	Alanine, asparagine, aspartate, cystein, cystine, glutamate, glycine, lysine, methionine, serine, threonine, proline, tryptophan, arginine, etc.	
Sugars & Vitamins	Promoters of plant and microbial growth, nutrient source, etc.	Glucose, fructose, galactose, maltose, ribose, xylose, rhamnase, arabinose, raffinose, deoxyribose, oligosaccharides, biotin, thiamin, niacin, pantothenate, riboflavin, etc.	
Enzymes	Catalyst for P release from organic molecules, biocatalyst for organic matter transformation, etc.	Acid/alkaline phosphatase, invertase, amylase, protease, etc.	
Purines	Nutrient source	Adenine, guanine, cytidine, uridine	
Inorganic ions and gases	Chemoattractant signals to microbes	HCO ₃ ⁻ , OH ⁻ , H ⁺ , CO ₂ , H ₂ , etc.	
Phenolics	Nutrient source, chemo attractant signals to microbes, microbial growth promoters, nod gene inducer/ inhibitors, chetalors, phytoalexins, etc.	Liquiritigenin, diadzein, genistein, coumetrol, eriodictyol, naringenin, isoliquiritigenin, umbelliferone, formonoetin, quercetin, leteolin, 4',7-dihydroxyflavone and derivatives, chalcone, etc.	
Root border cells	Produce signals that control mitosis, produces signals, stimulate microbial growth, release chemo-attractants, synthesize defense molecule, etc.		

30% of the 50% photo-assimilated carbon is required for sustaining root nodules and nitrogen fixation (Fig 1) and the rest 40% and 30% will be recruited for sustaining root respiration and biomass and rhizodeposition, respectively. Given that nearly 50% of photo-assimilated carbon is required to sustain a subterranean system, tailoring the rhizodeposition, qualitatively and quantitatively, will be pivotal in changing the structural and functional dynamics of microbial profile in soil for enhancing the nutrient availability and uptake for sustainable food production in future.

How Rhizodeposition Influences Microbial Diversity

Plants translocate an array of photo-assimilated carbon to sustain the subterranean system including root respiration and biomass gain and rhizodeposition. The dynamics of structural and functional diversity of microbial community play the most important role in nutrient mobilization, availability and subsequent uptake by the plants from the limited available pool of nutrients in the soil and thus influencing the plant growth and yield. As rhizodeposits, particularly root exudates, are primary substrates for the proliferation of different microorganisms in the rhizosphere, enhanced flow of photo-assimilated carbon (Fig. 1) in the form

of root exudates and its differential composition, qualitatively and quantitatively, will play a pivotal role in influencing the structural and functional beneficial microbial community in the rhizosphere vis-à-vis nutrient availability. Suppose enhanced exudation of organic acids is targeted by genetic augmentation in the varietal development programme, there will be likely an enhancement in the solubilisation and availability of non-available forms of nutrients directly or indirectly. Directly by mineralization and indirectly by enhancing the population of microbes which are capable of utilizing organic acids as primary sources of substrates for growth and multiplication, thereby enhancing the solubilisation of essential elements from unavailable to available forms. Similarly, enhanced exudation of monosaccharides will encourage rapid multiplication of microbes than exudation of oligo- and poly-saccharides. Enhanced exudation of acid/alkaline phosphatase by a particular crop variety will enhance the availability of inorganic phosphate than the variety which exudates out fewer phosphatases (Fig. 1). The same is true for all other rhizodeposits in different manners. Therefore, enhancing the quantity of preferred substrates in the root exudates, by genetic augmentation, for a desired beneficial group of microorganisms involved in different biogeochemical cycles would have the potential to

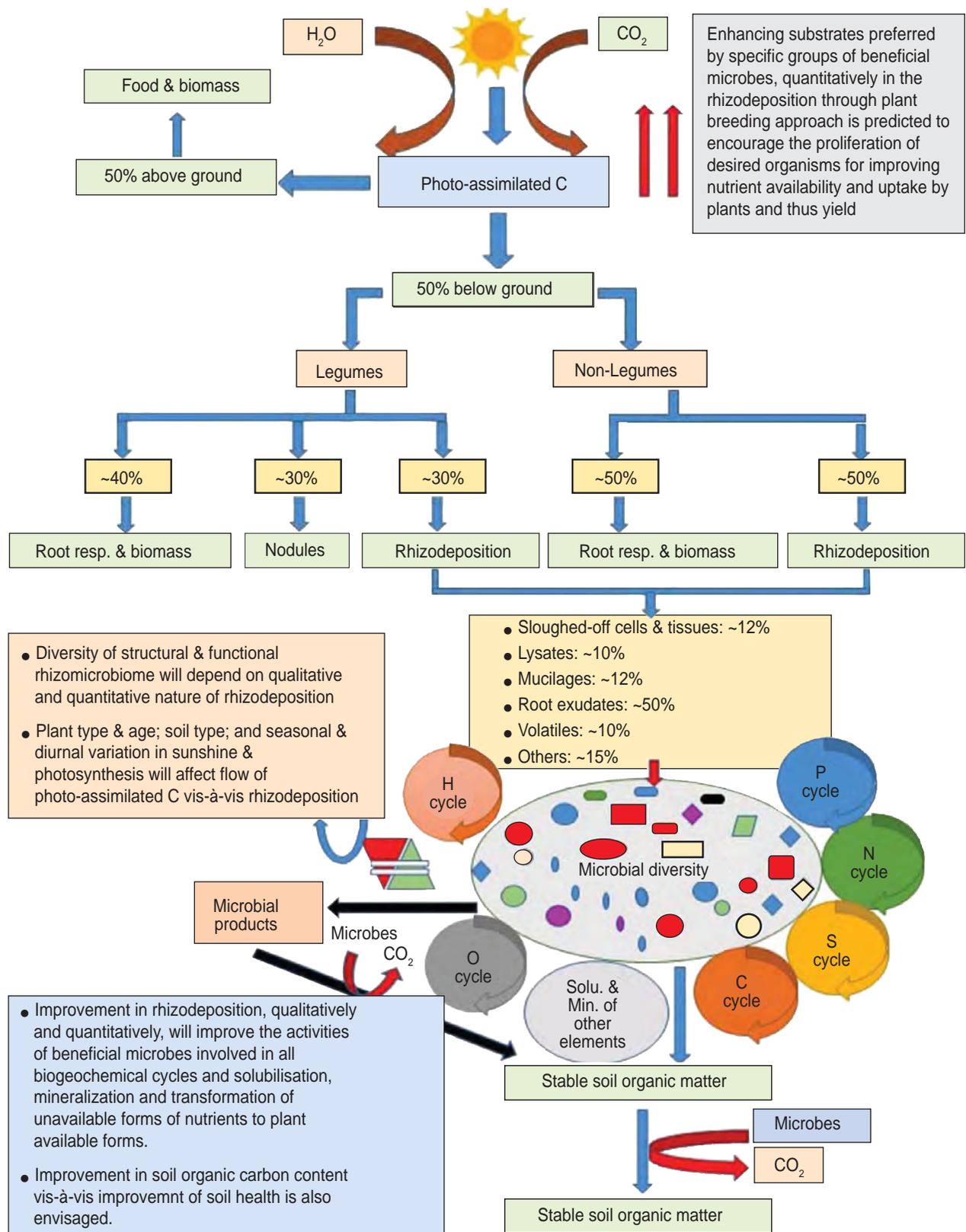


Fig. 1. Predicted fate of photo-assimilated carbon and possible role of rhizodeposition in nutrient dynamics and soil organic matter content

alter the patterns of nutrient mobilization, and disease suppression, and other vital soil processes defining outcome of plant-microbe interaction. Because of the flow of photo-assimilated carbon, the rhizosphere is abundant in various nutrients due to an aggregation of root exudates (Dakora and Philips, 2002) including amino acids, sugars, organic acids, vitamins, and enzymes (Bias *et al.* 2004, Gray and Smith, 2005) (Table 1). Root exudates not only function as a source of food but also function as repellents against pathogens and as attractants that aggregate beneficial microbes (Ahemad and Kibert, 2014, Hassan *et al.*, 2019). However, the flow of carbon in the rhizosphere varies with the physiological status, species of plants, and microorganisms that can be present in a given environment (Kang *et al.*, 2010). It has also been reported that soybean root exudates trigger an early symbiotic association between *Bacillus diazoefficiens* and soybean and also act as a messenger (Liu *et al.*, 2015, Walker *et al.*, 2004). Therefore, for shaping the rhizosphere interaction and its beneficial outcome, the root exudates play a crucial role. The classification and functions of root exudates are shown in Table 1.

Therefore, the population dynamics of predominant genera (*Agrobacterium*, *Arthrobacter*, *Azotobacter*, *Azospirillum*, *Bacillus*, *Burkholderia*, *Caulobacter*, *Chromobacterium*, *Erwinia*, *Flavobacterium*, *Micrococcus*, *Pseudomonas*, *Serratia*, *Cellulomonas*, *Allorhizobium*, *Azorhizobium*, *Bradyrhizobium*, *Sinorhizobium*, *Mesorhizobium*, *Rhizobium*, *Pantoea*, *Methylobacterium*, *Exiguobacterium*, *Paenibacillus*, *Azoarcus*, *Azospirillum*, *Alcaligenes*, *Streptomyces*, *Streptosporangium*, *Thermobifida*, *Micromonospora*, etc.) of bacteria and actinomycetes, representing plant growth-promoting group, will be determined by the quality and quantity of rhizodeposition, preferably root exudates as described above, from the flow of photo-assimilated carbon into the subterranean system. In turn, the dynamics of the above microflora will influence the availability of nutrients for the plant to take up for influencing the yield.

Future Perspective and Action Points

The genetic yield potential of any crop variety can only be realized with the favourable environmental condition along with the availability of essential nutrients. As such the availability of nutrients will be the outcome of the interaction of functional diversity of the microbes and bio-geochemical cycles and transformation and mineralization processes in the soil. Keeping other

factors constant, the qualitative and quantitative nature of rhizodeposition will be the determinant of the structural and functional diversity of the rhizomicrobiome. However, to understand the roles of rhizodeposition in influencing the rhizomicrobiome vis-à-vis availability of essential nutrients for realizing the genetic yield potential of crops, a culture-independent approach would be required along with complete profiling of rhizodeposition using omics approaches. However, all these approaches are in their infancy. As there is much variability among the wheat genotypes in root growth rates, exudates, and responses to the structural and functional microflora of un-ploughed soil, there is probably a lot of variation in rhizodeposition patterns among different plant species and that is the reason why there is wide variation in microbial profiles across different crop species. Therefore, it is possible to alter the dynamics of beneficial microbes in the rhizosphere by altering the photo-assimilated carbon flow through breeding processes. However, technological innovations are required to quantitatively and qualitatively assess both the flow of carbon as well as a shift in community structure during the augmentation of the genetic makeup of the plant. Therefore, for future agriculture to flourish in the increasingly parched planet to produce sufficient food for all in the backdrop of dwindling land, inputs and adverse climatic factors, the focus should be diverted to harness most from the interaction of photo-assimilated carbon flow-rhizodeposition-rhizomicrobiome in a given production system.

References

- Ahemad M and M Kibret (2014) Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. *J. King Saud Univ. Sci.* **26**: 1-20.
- Bias HP, SW Park, TL Weir, RM Callaway and JM Vivanco (2004) How plants communicate using the underground information superhighway? *Trends Plant Sci.* **9**: 26–32.
- Crowley DE and Z Rengel (1999) Biology and chemistry of nutrient availability in the rhizosphere. In: Mineral Nutrition of Crops: Fundamental Mechanisms and Implications, Z Rengel (Ed) Food Products Press, New York. pp. 1-40.
- Dakora FD and DA Phillips (2002) Root exudates as mediators of mineral acquisition in low-nutrient environments. In Food Security in Nutrient-Stressed Environments: Exploiting Plants' Genetic Capabilities, Springer: Dordrecht, The Netherlands, pp. 201–213.
- Gray E and D Smith (2005) Intracellular and extracellular PGPR: Commonalities and distinctions in the plant-bacterium signaling processes. *Soil Biol. Biochem.* **37**: 395–412.
- Hassan MK, JA McInroy and JW Klopffer (2019) The Interactions of Rhizodeposits with Plant Growth-Promoting Rhizobacteria in

- the Rhizosphere: A Review. *Agriculture*, **9(142)**: doi:10.3390/agriculture9070142.
- Johnson JMF, RR Allmaras and DC Reicosky (2006) Estimating source carbon from crop residues, roots and rhizodeposits using the national grain-yield database. *Agron. J.* **98**: 622–636.
- Kang BG, WT Kim, HS Yun and SC Chang (2010) Use of plant growth-promoting rhizobacteria to control stress responses of plant roots. *Plant Biotechnol. Rep.* **4**: 179–183.
- Liu Y, D Guan, X Jiang, M Ma, L Li, F Cao, H Chen, D Shen and JLi (2015) Proteins involved in nodulation competitiveness of two *Bradyrhizobium diazoefficiens* strains induced by soybean root exudates. *Biol. Fertil. Soils* **51**: 251–260.
- Nguyen C (2003) Rhizodeposition of organic C by plants: Mechanisms and controls. *Agronomie* **23**: 375–396.
- Tian T, A Reverdy, Q She, B Sun and Y Chai (2019) The role of rhizodeposits in shaping rhizomicrobiome. *Environ. Microbiol. Rep.* doi:10.1111/1758-2229.12816 published: 19 December 2019 **12(2)**: 160-172 pp
- Villarino SH, P Pinto, RB Jackson and G Piñeiro (2021) Plant rhizodeposition: A key factor for soil organic matter formation in stable fractions. *Sci. Adv.* **7**: eabd3176.
- Walker TS, HP Bais, E Grotewold and JM Vivanco (2003) Root exudation and rhizosphere biology. *Plant Physiol.* **132**: 44–51.

Microbial Nitrification Paradox: A Paradigm Shift on Nitrogen Uptake by Rice

Babanpreet Kour and Balasubramanian Ramakrishnan*

Division of Microbiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012, India

Nitrogen fertilization is an integral agronomic practice to increasing productivity and profitability in agriculture. But the poor nitrogen use efficiency (NUE, about < 40%) causes many economic and environmental challenges. The microbial oxidative process of converting ammonia to nitrite to nitrate (nitrification) is the rate-limiting step in the N loss. The century-old, conventional theory of nitrification with the involvement of two functionally different bacterial groups as ammonia-oxidizing bacteria and nitrite-oxidizing bacteria has been upturned by the recent discoveries of archaeal members involved in ammonia oxidation (Ammonia-oxidizing archaea, AOA), anaerobic ammonia-oxidizing bacteria (Anammox bacteria) and complete ammonia oxidizing bacteria (Comammox bacteria) in the last two decades, largely due to the advances in molecular and metagenomic methods introduced to study the microbial ecology. What is interesting to know is that nitrate-transporters of host plants, as in rice, are involved in the assembly of the microbiome associated with roots. Besides, rice plants produce the biological nitrification inhibiting (BNI) compounds released through root exudates. Involvement of diverse microbiome members and the plant genome through nitrate transporters on the rice rhizosphere microbiome assembly necessitates the reappraisal of the nitrogen fertilization management options. This paper also highlights the need for gathering new knowledge on the plant-microbe interactions, from the genome to metabolite levels, and conserving these resources for sustainable rice cultivation.

Introduction

Prokaryotic microorganisms can react with molecular dinitrogen (N_2) to convert it to ammonium, the first step in the biological nitrogen cycle. An alternative to this step is the application of chemical nitrogen fertilizers, which is practiced mainly in intensive agriculture. The nitrogen input in the form of ammonium (NH_4^+), nitrate (NO_3^-), or urea provides the substrates for microbial processes, such as nitrification and denitrification. The discovery of nitrifying microorganisms by Winogradsky (1891) led to an understanding that the oxidation of ammonia to nitrite to nitrate involves two bacterial functional groups: (i) autotrophic ammonia-oxidizing bacteria (AOB) and (ii) autotrophic nitrite-oxidizing bacteria (NOB). These autotrophic bacteria, along with heterotrophic (bacterial- as well as fungal) nitrifiers, have adverse consequences, depending on the type and amount of N fertilizers applied, such as (i) poor N-fertilizer use efficiency (NUE), (ii) losses of different N forms, (iii) nitrate-contamination of freshwater resources, and (iv) increased nitrous oxide emissions. The century-old, conventional theory about nitrification has been challenged in the last two decades, by the reports on the presence of anaerobic ammonia-oxidizing bacteria (Anammox bacteria, discovered in 1999), ammonia-

oxidizing archaea (AOA in 2005), the coupled ferric-iron reducing- and ammonia-oxidizing bacteria (Feammox in 2006), and complete ammonia-oxidizing bacteria (Comammox in 2015) in soils (Hayatsu *et al.*, 2021). These discoveries are primarily due to the advances in molecular detection and metagenomic analyses of nitrifying microorganisms. A better understanding of nitrification processes is necessary to create opportunities for improving the NUE and minimizing the adverse environmental impact of intensive cultivation.

Traditionally, rice is cultivated under lowland (waterlogged) conditions where ammonium is considered the major form of available N to plants. The predominantly anaerobic soil conditions in the rice-paddy ecosystem impede nitrate accumulation or retention. However, some ammonium gets oxidized to nitrate in the rhizosphere by the molecular oxygen, which is transported through specialized aerenchyma; nitrate subsequently gets converted to N_2 or other gaseous forms. In modern, intensive rice cultivation, urea or ammoniacal fertilizers are applied to increase productivity. About 50-70% of applied N is lost either via volatilization or nitrate leaching, contributing to the poor nitrogen use efficiency (NUE) in rice. The oxidative process of ammonium to nitrate via nitrite can occur in

*Author for Correspondence: Email-b.ramakrishnan@icar.gov.in ; ramakrishnanbala@yahoo.com

the presence of AOB and AOA, and nitrite to nitrate by the NOB, or ammonium to N_2 by the anammox bacteria (i.e., *Candidatus Brocadia*, *Candidatus Kuenenia*, *Candidatus Anammoxglobus*, and *Candidatus Jettenia*) and comammox bacteria (i.e., *Nitrospira* sublineage II). Until the discoveries of these novel microbial entities in the last two decades, nitrification has been considered the foremost and rate-limiting step in the N-loss. The nitrification process has been the key target for mitigating nitrogen loss from agricultural fields. Therefore, synthetic nitrification inhibitors (NIs such as 2-amino-4-chloro-6-methyl pyrimidine, dicyandiamide (DCD), 3, 4-Dimethylpyrazole phosphate (DMPP) and nitrapyrin) are employed to inhibit the activities of nitrifiers from decreasing the accumulation of nitrate in soils and nitrous oxide emissions from soils, including from the rice-paddy fields. Nevertheless, the application of these synthetic NIs has several drawbacks, such as high cost, poor efficiency, increased ammonia volatilization, non-target effects, and the risk potential related to trophic transfer, food safety, and the environment (Subbarao *et al.*, 2006).

Nitrification: A Conundrum in the Rice-Paddy Fields

While nitrification inhibition appears promising to prevent oxidative N losses via nitrate, the paradox is that nitrate is another available form of N for plant uptake. Interestingly, nitrate has recently been recognized as an equally important N source as ammonium for rice cultivation (Fig. 1). Even the lowland rice plants can take up about 15–40% of total N in the form of nitrate.

Additionally, the plant-microbe interactions related to nitrification have never received enough attention. The rice plant genome has several nitrate transporters, such as nitrate and peptide transporter family (NPF) members (about 93), nitrate transporters (NRT2, 4), and high-affinity nitrate transport-activating protein members (NAR2, 2), suggesting their significant involvement in the N uptake and distribution of N within the plant (Cai *et al.*, 2009; Li *et al.*, 2017). These nitrate transporters are critical for improving nitrogen use efficiency (NUE) in rice. Many *Indica* rice cultivars exhibit superior growth on nitrate than ammonium, and their combined presence [i.e., $NH_4^+ : NO_3^-$ at 75:25] acts synergistically. Thus, nitrate availability is equally essential as ammonium for plant uptake. The predominantly anaerobic flooded rice fields are neither conducive to nitrate formation nor nitrate retention. Even if a small amount of nitrate is produced, subsequent denitrification processes prevent nitrate accumulation. Likewise, only a small amount of N is lost via nitrate leaching.

On the contrary, ammonium toxicity is of utmost concern in rice-paddy fields. Increased ammonium levels can suppress the uptake of other cations (e.g. K^+ , Ca^{2+} , and Mg^{2+}) and even inhibit plant growth. What is more interesting is that nitrate transporters of rice are a significant factor in the rhizosphere microbiome assembly (Zhang *et al.*, 2019). The rhizosphere microbiome, especially the diversity and composition and functions such as niche creation, resource competition, antagonism, and cooperation, influences the plant growth and development, resistance to pathogens, and stress

Non-target effects of synthetic nitrification inhibitors (SNIs)

Synthetic nitrification inhibitors (SNIs) have numerous modulating effects on the non-target organisms, including the loss of diversity and functions of different microbial guilds. For example, higher concentrations of Chloro-methyl-pyrazole phosphate (CIMPP), Dicyandiamide (DCD), and 4-dimethylpyrazolephosphate (DMPP) inhibited the growth of non-target microorganisms. SNIs suppress not only the abundance of ammonia-oxidizing archaea, but also the total population of archaea. Nitrapyrin has substantial effects on the relative abundance of non-target archaea (*Thaumarchaeota*), bacteria (*Proteobacteria* and *Nitrospirae*) and fungi (*Basidiomycota*) in the wheat rhizosphere. Specifically, the abundances of bacteria (*Gaiella*, *Nitrososphaera* and *Rhodoplanes*) and fungi (*Gliomastix*, and *Ganoderma*) are more susceptible to the nitrapyrin addition (Schmidt *et al.*, 2022). The soil treatment with nitrapyrin leads to the selective enrichment of copiotrophs (bacteria- Alphaproteo bacteria and Betaproteo bacteria; *Ascomycota* fungi) with the suppression of oligotrophs (bacteria – *Acidobacteria* and *Planctomycetes* and *Basidiomycota* fungi). These findings suggest that SNIs can disturb the plant-microbe interactions and natural soil functioning processes that contravene the desired effect of nitrification inhibition.

or harmful, the host plant determines the community structure by attracting the desired microorganisms using specific chemical compounds through root exudation. The rhizosphere-associated microorganisms provide additional genetic and functional diversity to the host plants; the rhizosphere microbiome is considered the secondary genome of plants. Interestingly, the root exudates of rice contain a nitrification inhibitor (i.e., 1,9-decanediol) which can inhibit the ammonia monooxygenase (AMO) and there is a strong correlation between the amounts of 1,9-decanediol in the root exudates and the biological nitrification inhibition (BNI) abilities (Sun *et al.*, 2016). The regulation of the N cycle in the rhizosphere depends primarily on the plant-microbe interactions. The mechanistic understanding of the microbiome assembly processes, factors that drive the assembly, and niches the specific microbial members can occupy are essential to benefit from these plant-microbe interactions, improve the NUE, and protect the environment sustainably.

Future Perspectives

1. The chemical dialogues between the rhizosphere microbiome and the host plants require intensive studies. The biological nitrification inhibitors can be superior to the synthetic nitrification inhibitors. It will be better to breed, design, or engineer rice cultivars with higher capacities to modulate nitrification processes for better plant N uptake.
2. The chemical dialogues in the rhizosphere or phyllosphere as the driver of the microbiome assembly also need scrutiny in all the crops to identify the crop-specific stimulators or inhibitors.
3. The beneficial characteristics of BNIs necessitate breeding rice cultivars with desired traits associated with the plant-microbe interactions. The relevant fundamental researches on the identification of genetic loci, specificity, compatibility with other useful traits, and biosynthetic pathways associated with the plant-microbe interactions are critical for making a positive impact in agriculture.
4. Due to their critical roles in the plant-microbe interactions, the microbial genomic resources need conservation and utilization. Future strategies need to include the creation of the genomic database of beneficial microorganisms, besides the culture collections for utilizing them as bio-stimulants or as the sources of genes for use in agriculture.

References

- Cai H, Y Zhou, J Xiao, X Li, Q Zhang and X Lian (2009) Overexpressed glutamine synthetase gene modifies nitrogen metabolism and abiotic stress responses in rice. *Plant Cell Rep.* **28**: 527-537.
- Dong D, W Yang, H Sun, S Kong and H Xu (2021) Nitrous oxide emissions in response to long-term application of the nitrification inhibitor DMPP in an acidic luvisol. *Appl. Soil Ecol.* **159**: 103861.
- Hayatsu M, C Katsuyama and K Tago (2021) Overview of recent researches on nitrifying microorganisms in soil *Soil Sci. Plant Nutr.* **67**: 619-632.
- Li H, B Hu and C Chu (2017) Nitrogen use efficiency in crops: lessons from *Arabidopsis* and rice. *J. Exp. Bot.* **68**: 2477-2488.
- Schmidt R, XB Wang, P Garbeva and É Yergeau (2022) The nitrification inhibitor nitrapyrin has non-target effects on the soil microbial community structure, composition, and functions. *Appl. Soil Ecol.* **171**: 104350.
- Subbarao GV, O Ito, KL Sahrawat, WL Berry, K Nakahara, T Ishikawa, T Watanabe, K Suenaga, M Rondon and IM Rao (2006) Scope and strategies for regulation of nitrification in agricultural systems—challenges and opportunities. *Crit. Rev. Plant Sci.* **25**: 303-335.
- Sun L, Y Lu, F Yu, HJ Kronzucker and W Shi (2016) Biological nitrification inhibition by rice root exudates and its relationship with nitrogen-use efficiency. *New Phytol.* **212**: 646-656.
- Winogradsky S (1891) The formation of nitrates in the process of nitrification. In: *Annales de l'Institut Pasteur* (Vol. 5, pp. 577-616).
- Zhang J, YX Liu, N Zhang, B Hu, T Jin, H Xu, Y Qin, P Yan, X Zhang, X Guo and J Hui (2019) NRT1.1B is associated with root microbiota composition and nitrogen use in field-grown rice. *Nature Biotech.* **37**: 676-684.

Current Status and Recent Developments in Microbial Pesticide Use in India

HB Singh

Department of Biotechnology, GLA University, Mathura-281406, Uttar Pradesh, India

For ensuring sustainable food security scientists are working hard to develop alternative eco-friendly technologies for crop protection to combat the ill effect of chemical pesticides. A large number of agriculturally important microorganisms (AIMs) are being used as biopesticides or biostimulants for the management of plant diseases. Despite extensive research on biopesticides globally their reach to end-users i.e. farmer is very low. The main barrier to not accepting biopesticides is poor quality products available in the Indian market. Issues related to commercialization, regulatory requirements in India, slow market growth and less acceptance of biopesticides by the farmers in India are discussed.

Introduction

In order to effectively manage plant diseases and stimulate plant growth, microbial pesticides and bio pesticides (PGPR, bio stimulants) based on living microorganisms and their bioactive components have been extensively studied, published, and promoted. They have, however, typically been consigned to niche items due to their lack of efficacy, uneven field performance, short shelf lives, and stringent regulation requirements by Central Insecticide Board and Registration Committee (CIBRC). Despite significant market penetration gains, bio-pesticides still only account for a small portion of Agri-bio input goods. Numerous microorganisms associated with plants are known to suppress pathogenic organisms, produce hormones that stimulate plant development, and increase plant health and resilience to disease. To safeguard crop production and boost output, managing pests and illnesses in agriculture is crucial. The awareness and demand for AIMs used as biopesticides, biofertilizers, and bio stimulants is rising slowly all over the world. Microbial pesticides are based on bacteria, fungus, viruses, nematodes, protozoa, and other microorganisms. *Bacillus subtilis*, *Gliocladium* spp., *Trichoderma*, *Pseudomonas fluorescens*, *Beauveria bassiana*, and *Metarrhizium anisopliae* are among the nine microbes included in a schedule that was published in the Gazette of India on March 26, 1999, as an amendment to the Insecticides Act, 1968 for the commercial production of biopesticides. The Insecticide Act of 1968 added 26 new microorganisms to its agenda for the manufacture of microbial biopesticides.

In India, the biocontrol agents *Trichoderma viride*, *T. harzianum*, *P. fluorescens*, *B. bassiana*, *M. anisopliae*, and *B. subtilis* have carved out a place for themselves as crucial components in the management of numerous pests and illnesses (Singh *et al.* 2016). Its reach is still confined to a few specific states in our nation, though. The proliferation of low-quality producers of biopesticides is the main cause of this situation. In our opinion, biopesticides have a possible role to play in the creation of a future integrated pest management program. Hopefully, the government will soon adopt a more sensible stance regarding microbial biopesticides.

The research on microbial pesticides would be beneficial only if the product based on novel strains is commercialized and registered. To accomplish this, it is necessary to adhere to the Central Insecticide Board's standards. Technology advancement also heavily depends on the mass manufacture of formulations based on microorganisms. These CIB-set parameters will be discussed in this manuscript, along with other aspects of the marketing and registration of biopesticides (Singh *et al.*, 2016).

Need for Commercialization of Microbial Pesticides in India?

Prior to independence, our research in plant disease management was primarily theoretical and exploratory in nature. Biological control agents seem to be the same. To combat disease in micro-plots, scientists have tested a vast number of antagonists, narrowed them down to a few that were effective, and then put them to the test in

*Author for Correspondence: Email-hbs1@rediffmail.com

the field. The most effective antagonists were produced in this way. Commercialization of biocontrol products would have been the next obvious step in this sequence. Biocontrol products make up less than 1-2 % of the crop protection market, according to estimates from a firm. About 80 biocontrol products have crossed the finish line to become commercial merchandise worldwide. The products have a confined range of applications because they have only been assessed on a small range of crops and can only control one or two infections. Additionally, there hasn't been a lot of investment in the development of commercial formulations of biocontrol-active microbes, perhaps as a result of the high costs associated with creating, testing, registering, and selling these products. Based on both the growing degree of complexity and the declining potential for repeat sales, the efforts made in the direction of the commercialization of biocontrol agents may be divided into three groups. As with the application of soothing balm in medicine, the first group suggested applying the antagonist precisely and directly to the infection site as needed. This is done to prevent or overwhelm the pathogen by applying a large population of antagonist organisms, such as antagonists applied to seeds to protect germination against damping-off, antagonists applied to fruits to prevent fruit decay in storage, and bacteria growing at temperatures below 0°C to protect plants from frost damage. With the exception of *Gliocladium virens* GL-21, which is placed in the soil near seeds or plants with a sufficiently high inoculum density to ensure an initially high population in the infection court, nearly all microorganisms that have been commercially available to date fall into this category. This collection of bioagents is crucial as it represents the first logical step in their commercialization for use against plant diseases and because it makes a substantial contribution to raising awareness of microbes among business people, farmers, and society at large. The second group, which contains antagonists administered to seeds or other single points with the intention of spreading with plant growth and defending roots and shoots, is supported by Plant Growth Promoting Rhizobacteria (PGPR). This group of antagonists may include examples of the inundative and augmentative application strategy, where the antagonist is expected to multiply, persist for some or all of the plant's life, and boost the population of an insufficient supply of a linked or pretty close antagonist already present in the rhizosphere. For instance, colonized wheat grains strewn on the soil surface were employed to

spread an oxygenic *Aspergillus flavus*, which prevents the growth of toxic strains of *A. flavus* on the floral portions of cotton plants. Due to the antagonists' increased susceptibility to the impacts of competition and environmental factors, this category of examples presents a more difficult biocontrol approach. At this time, there are no commercially available alternatives in this group.

The following are examples of biological control failure or variable performance in the rhizosphere with introduced microbes:

- a. Low disease pressure for an effective test
- b. Very poor carbon content in the soil
- c. Heavy dosage of chemicals used in the field prior to use of biologicals
- d. The use of biopesticides encourages the expansion of non-target disease harm
- e. Variable colonization of the transplanted strain's impacted roots or loss of the strain's ecological competency
- f. Wherever it is required for the efficient action of an antagonist, the expression of antibiotic synthesis occurs perhaps too late or insufficiently to actively control the disease.

However, these restrictions can be overcome by choosing efficient antagonist strains and enhancing their activity using cutting-edge biotechnological techniques. A combination of institutional issues, technical constraints, and unreasonably high expectations for these goods are some of the additional difficulties associated with the commercialization of biocontrol products. But if we're serious about creating marketable biological control products, we'll have to get over these challenges. As a result of years of research, the formulation's rhizosphere competency varies with shifting soil conditions. All research must move in the direction of the creation and commercial manufacturing of the unique superior strain after isolating a rhizosphere competent strain of an efficient biocontrol agent or antagonist. In order to develop a broad-spectrum product that can effectively control a large number of pathogenic fungi and bacteria under different conditions, field trials are essential for the development of a product. These trials must be executed under varied soil conditions in various fields, on crop varieties, and also with multiple pathogenic fungi or bacteria. In order to help prevent illnesses, biocontrol fungi offer

other advantages, such as reducing the physiological stresses that seedlings experience naturally and boosting resilience to abiotic pressures (Singh *et al.*, 2017). Another restriction is selecting an appropriate substrate for the creation of an efficient formulation. The development of biocontrol formulations has utilized a variety of substrates. Utilizing agricultural wastes, such as wheat bran, coffee and tea grounds, rice hay, distilled waste from oil-producing plants, and rice waste would be financially viable. For use in the field, we have created a cheap technology for mass-producing biocontrol fungi. Different substrates, such as powdered rye grass seed, Diatomaceous earth granules and molasses, wheat bran formulations, wheat bran sawdust formulations, molasses-yeast medium, and others, are employed in laboratories to mass-produce biocontrol fungi. Many commercially viable products for disease management are either region- or disease- or both-specific. Therefore, it is imperative to go forward with the selection of the most efficient strains of biocontrol fungus and their production as broad-spectrum formulations for use against a variety of soil-borne illnesses under various soil conditions. The essential factor in the commercialization of microbial agents for use in agriculture, as well as a significant barrier, is cost. Another significant barrier is that most nations lack the infrastructure needed to scale up and commercialize biocontrol products. It is necessary to create a framework for the commercialization and distribution of biocontrol agents to farmers that is akin to the one used by breeders to release breakthrough varieties.

The optimum parameters for the commercialization of a biocontrol agent are as follows:

- Appropriate and suitable strain selection.
- Prolong shelf life and storage of bioformulations at room temperature.
- Novel application technology viz., seed biopriming.
- Scale up and quality control at production and distribution sites.
- Registration of bioformulation with regulatory body.

Registration of the organisms used to produce biocontrol products is one of the biggest obstacles that must be overcome during the commercialization process. Through Directive 91/414/EEC, which is “Concerning the

Placement of Plant Protection Products on the Market,” an effort has been made in the European countries to offer a uniform legislative framework throughout the European Union. However, there has been much discussion on this matter and it is still not completely settled, so there remains uncertainty in the domain. The EPA maintains four levels of testing for possible adverse impacts of microbial pesticides in the USA. These include adverse impacts on plants and animals that are pathogenic or poisonous but not targeted. The Insecticide Act of 1968 now requires that antagonistic organisms, including *Trichoderma* species, *Gliocladium* species, *Bacillus* species, *Pseudomonas* species, etc., be listed in the schedule. This regulation was put into effect by the Indian government in March 2009. India has a completely different situation as the market for crop protection has been monopolized by the chemical pesticide business. It is also discouraging to see how the administration is acting. A paradigm shift in business practices is required to establish a market for biocontrol products in India. Any microbiological product must be registered in accordance with specific legal requirements before it can be sold. The Insecticides Act of 1968 and its implementing regulations govern the import, manufacturing, sale, transportation, and distribution of biopesticides in India (Singh *et al.*, 2016). The following are the key components of this requirement:

The Central Insecticides Board approved adding more microorganisms to the schedule of the Insecticides Act to control production and use in India (Table 1).

After inclusion of an organism in the schedule, an applicant can submit Form I along with prescribed fees to the Registration Committee (RC) under Section 9(3B) as per the guidelines of data generation. Permanent certification is issued under section 9(3) as per guidelines.

Data Requirements

Usually, the CIB-RC allows commercialization in terms of import, manufacture, sale, transport, and distribution of biopesticides only after the grant of regular registration. The data requirements for all microbes to be registered are provided at <http://www.cibrc.nic.in>. Currently, 970 microbial pesticides are registered with CIBRC for commercial production and use by farmers (Fig. 1). There are 355 products based on *Trichoderma* spp. manufactured in India by several companies (Fig. 2).

Table 1. Microbes listed in Gazette of India in the 1968 Insecticide Act's for production of biopesticides

<i>Agrobacterium radiobacter</i> strain 84	<i>Fusarium oxysporum</i> (non pathogenic)	<i>Paecilomyces lilanicus</i>
<i>Agrobacterium tumefaciens</i>	<i>Gliocladium</i> spp.	<i>Photobacterium luminescens</i> akhurstii strain K-1
<i>Alcaligenes</i> spp.	<i>Grannulosis</i> viruses	<i>Piriformaspora indica</i>
<i>Ampelomyces quisqualis</i>	<i>Hirsutella</i> spp.	<i>Serratia marcescens</i> GPS5
<i>Aspergillus niger</i> – strain 27	<i>Metarrhizium anisopliae</i>	<i>Streptomyces griseoviridis</i>
<i>Bacillus subtilis</i>	<i>Myrotheium verrucaria</i>	<i>Streptomyces lydicus</i>
<i>Beauveria bassiana</i>	Nuclear Polyhedrosis Viruses (NPV)	<i>Trichoderma</i> spp.
<i>Burkholderia cepacia</i>	<i>Nomurea rileyi</i>	<i>Verticillium chlamydosporium</i>
<i>Candida oleophila</i>	<i>Pseudomonas fluorescens</i>	<i>Verticillium lecanii</i>
<i>Chaetomium globosum</i>	<i>Penicillium islanidicum</i> (for groundnut)	VAM (fungus)
<i>Coniocytrium minitans</i>	<i>Pythium oligandrum</i>	
<i>Erwinia amylovora</i> (hairpin protein)	<i>Phlebia gigantean</i>	

In India, we have 361 biocontrol laboratories/units for the production of biocontrol agents (Fig. 3) with annual production and consumption of 7203 Metric Tons during 2018-19 (Fig. 4).

Why AIMs Are Not So Popular in India?

In India, a large number of academic institutions, universities, ICAR Institutes, CSIR Institutes are working on agriculturally important microorganisms (AIMs) for management of pests and diseases, plant growth promotion, bioremediation etc., but there is no connectivity among academia, industry, and regulatory bodies (Fig. 5). Therefore, a very strong bond is to be developed among these three partners so that the microbes get a substantial place in agriculture and the environment.

Future Prospects

What is the outlook for biological management of plant pathogens that are responsible for various diseases around

the globe? The main issue is how to get the technology from the lab to the commercial growers in order to change the outlook for biological control. More scientific efficacy trials in commercial or almost commercial contexts are required, along with thorough replication and statistical analysis. Biocontrol products are either sold separately or blended with other microbial metabolites when they are formulated. However, extensive research needs to be done in order to create mixes that might lead to greater success. The biopesticide Industry Alliance is building a certification method to guarantee industry standards for efficacy, quality, and consistency in order to aid in improving the global market perception of biopesticides as efficient products. The availability of all these facts to growers and extension agents will make a stronger impression than relying just on company advertisements. Discovering the causes of the dearth of biological control data is not that tricky anymore.

A few are mentioned below:

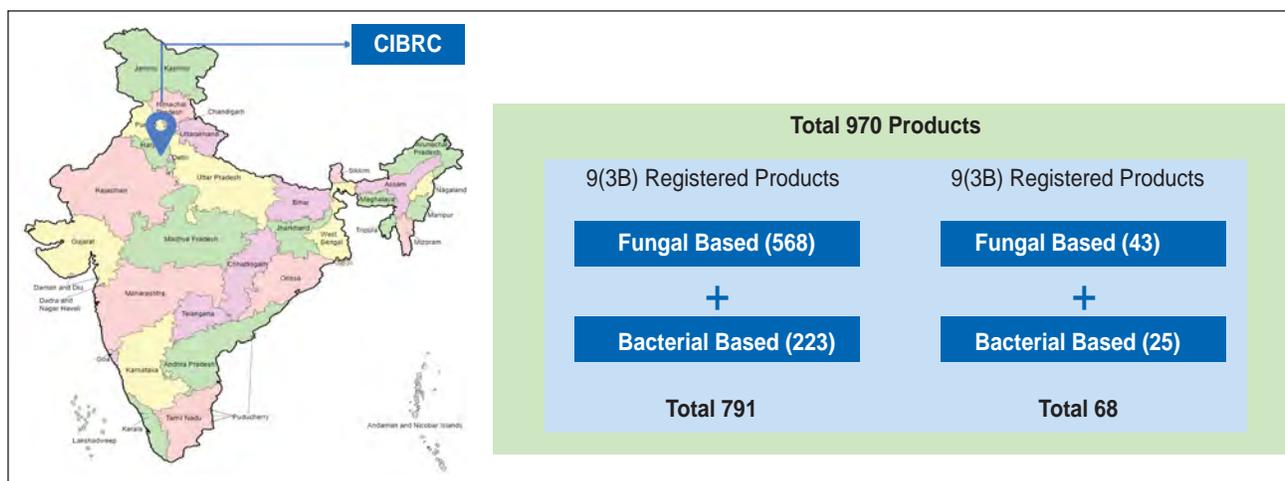


Fig. 1. Commercially Registered Microbial pesticides from CIBRC

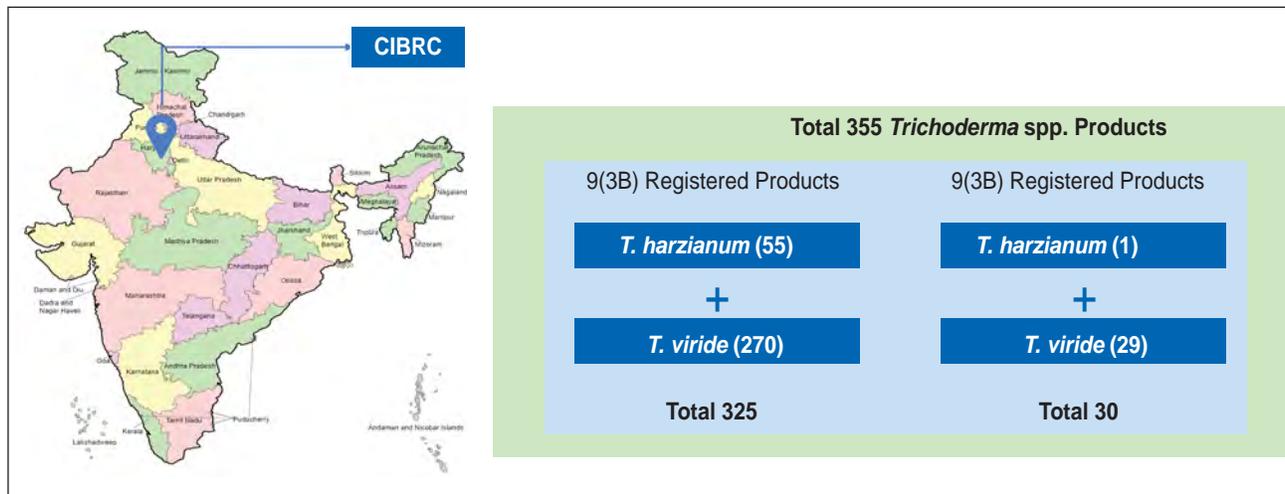


Fig. 2. Commercially Registered Microbial pesticides based on *Trichoderma* spp. from CIBRC

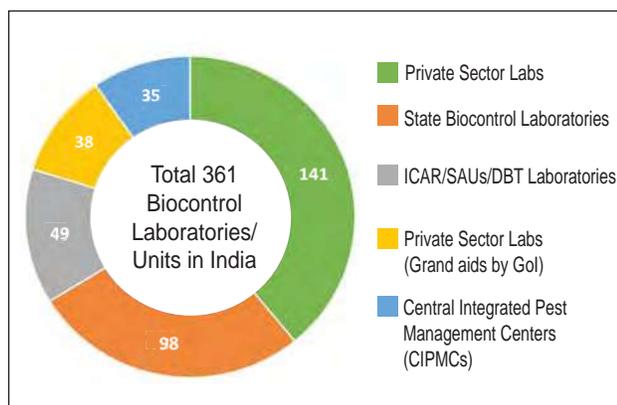


Fig. 3. Infrastructure for production on Microbial pesticides in India

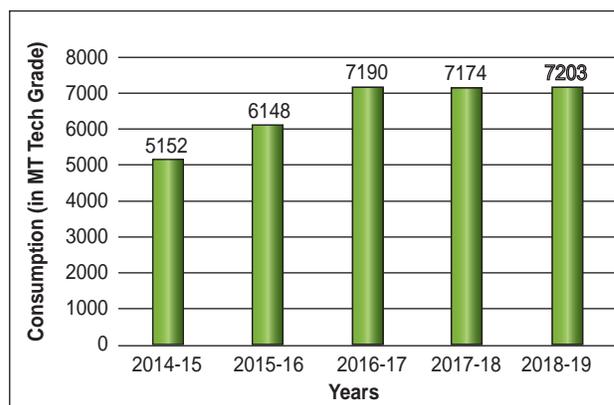


Fig. 4. Consumption of Biopesticides in India

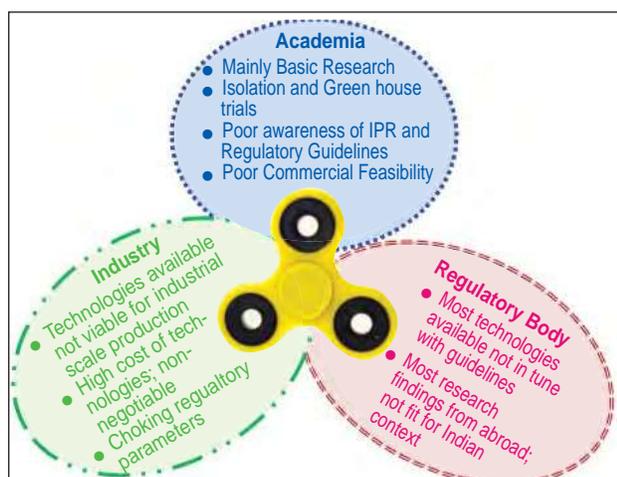


Fig. 5. Gap areas between academia, industry and regulatory body

1. A lot of the information is confidential and is only available in corporate reports or in registration dossiers submitted to regulatory bodies.
2. Universities and research institutes no longer test products on a regular basis; instead, findings are reported in research station bulletins, which are not indexed by citation databases.
3. Many trials may demonstrate a lack of efficacy and are not published.
4. Scientific publications do not publish biocontrol product effectiveness studies due to deficit innovative ideas.

More research is needed on the pathogen’s ecology and epidemiology, which are particularly understudied for diseases that are soil-borne. Before extolling the virtues of biological control over chemical control

techniques, a number of additional issues also need to be addressed.

For example:

- a. How is the pathogen introduced and how does it spread?
- b. How does the connection exist between damage and population density?
- c. How do different environmental factors affect the effectiveness of newly introduced microorganisms?

Every change we make has an impact on the environment, including the one of doing nothing. But we need to find a way out of this mess so that we may presume that the main qualities of our commercial products for the biocontrol of plant diseases are safety and effectiveness. Only then can it be anticipated that the marketing of biocontrol agents will be effective. The idea that the disease should be managed rather than

entirely controlled must be ingrained in end users' minds for biocontrol to be more widely accepted. Additionally, commercial companies are hesitant to finance ongoing efforts in biocontrol research because of the market size, variability, and methods of production, formulation, and distribution. For the development of next-generation goods with longer shelf lives, recent research methods on *Trichoderma*, in particular genomics, transcriptomics, proteomics, and metabolomics, can be applied. To get over these challenges, more research must be done across all aspects of biocontrol.

References

- Singh HB, BK Sarma and C Keswani (Eds.) 2017. Advances in PGPR Research. CABI-UK. 408 pages, ISBN-9781786390325.
- Singh HB, BK Sarma and C Keswani (Eds.) (2016) Agriculturally Important Microorganisms: Commercialization and Regulatory Requirements in Asia. Springer, Singapore. 336 pages, ISBN-13: 978-9811025754.

Blue Green Algae for Secondary Agriculture

Hillo Chakdar¹, Shaloo Verma¹ and Sunil Pabbi^{2*}

¹ICAR-National Bureau of Agriculturally Important Microorganisms, Mau-275103, Uttar Pradesh, India

²Division of Microbiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012, India

Blue Green Algae (BGA) or cyanobacteria are photoautotrophic microorganisms widely distributed in terrestrial and aquatic habitats. These have tremendous metabolic diversity with the ability to produce a range of metabolites like pigments, polysaccharides, amino acids, lipids, vitamins, enzymes, antimicrobial compounds and many more. Till now the major application of cyanobacteria is in the form of biofertilizers, especially in India and a few South Asian nations, nonetheless due to the production of a number of industrially important molecules like phycobiliproteins, amino acids, bioactive lipids, biofuels, their mass cultivation has tremendous scope in secondary agriculture. Many of these compounds are of high value and have applications in the food, feed and pharmaceutical industries. Nevertheless, commercial exploitation of cyanobacteria in secondary agriculture is still in its infancy. Appropriate research efforts and suitable policy decisions are required to establish these versatile organisms as a secondary agriculture option and a profitable commercial venture.

Key Words: Bioactive metabolites, Biorefinery approach, Cyanobacteria, Secondary agriculture, *Spirulina*

Introduction

Blue Green Algae are the most primitive prokaryotic photoautotrophs responsible for the oxygenation of the earth's atmosphere. These are considered as the most successful and sustained prokaryotic organisms during the course of evolution. BGA can produce an array of metabolites, such as phenols, flavonoids, carotenoids, phycobiliproteins, steroids, vitamins, amino acids, enzymes, polysaccharides etc. which can be potentially used for agricultural and industrial applications. BGA are used as food or health supplements, superfood bar, or fine chemicals in nutraceutical industries, their biomass are used as a source of biofertilizers, natural soil conditioner, and their pigments such as phycobiliproteins (phycocyanin and phycoerythrin) are used in different cosmetic products or natural food colorants. However, the major application of cyanobacteria in most countries is in the form of biofertilizers for rice.

Primary agriculture mainly aims to produce or cultivate natural resources for human consumption. However, many a time value addition or the processing is generally avoided which can otherwise render more return to the producers. On the other hand, secondary agriculture is generally defined as processing or adding value to the primary agricultural commodities and can make a remarkable positive contribution towards the national

economy (Yadav *et al.*, 2020). Secondary agriculture contributes more than 25% of India's GDP (Yadav *et al.*, 2020). It utilizes renewable agro-bioresources or agri-waste by value addition. Nurseries, biofertilizers, biopesticides, compost, flavours, dyes, essential oils, and biofuels are some of the avenues of secondary agriculture (Yadav *et al.*, 2020). The demand for bioactive foods, and green health products is increasing with increased health awareness. Such products can be one of the important constituents of secondary agriculture. Cyanobacteria can be a useful resource for secondary agriculture due to the production of the wide range of metabolites with commercial application, photoautotrophic growth and ease of mass multiplication. Further to this, the biorefinery approach toward commercial exploitation of cyanobacteria can multiply the return by many folds.

In this present review, we aim to briefly present the applications and prospects of cyanobacteria pertinent to secondary agriculture and challenges for commercial exploitation.

BGA as a Source of Natural Pigments

Cyanobacterial pigments such as phycobiliproteins, chlorophyll, and carotenoids possess commercial significance and have sparked research interest. Phycobiliproteins (PBP) can be a natural alternative to harmful synthetic dyes and colorants. Due to its

*Author for Correspondence: Email-sunil.pabbi@gmail.com

carcinogenic nature and potential allergic reactions, the use of synthetic food colors has been restricted by European Food Safety Authority (AESAs) and the American Food and Drug Administration (FDA) (Vigani *et al.*, 2015). Cyanobacterial phycocyanin (C-PC) has been used as a natural colorant in food industries such as in chewing gum, ice cream, candies, dairy products, beverages such as soft drinks; drug and cosmetics; and have economic potential due to its non-toxic properties and health benefits (Chakdar *et al.*, 2012). Phycocyanin has been approved as a permitted food colorant by FDAs of USA and Japan which has expanded its application. In the aquaculture industry, C-PCs are used as feed supplements due to their high nutritional value (Yusoff *et al.*, 2020). Species of genera such as *Spirulina*, *Synechococcus*, and *Anabaena* contain abundant natural blue pigment. Phycocyanin with a purity >0.70 is considered a food grade while a purity above 4.0 is considered a pharmaceutical grade. The cost of pharmaceutical-grade phycocyanin can go up to thousands of USD per milligram. According to Future Market Insights, the overall market of phycobiliproteins was USD 112.3 million in 2018 which is predicted to double by 2028 (Pagels *et al.*, 2019). Phycocyanin pigment alone has a market of about \$100 million USD.

BGA as a Source of Cosmeceuticals

Globally, India ranks fourth for generating revenue from beauty and health care products and the Indian cosmetic industry is expected to reach 20 billion USD by 2025. With the rising awareness among consumers regarding the side effects of chemical cosmetic formulations, the demand for green cosmetics is increasing. Being phototrophic microorganisms with ubiquitous distribution, cyanobacteria possess excellent photoprotective and antioxidative mechanisms. The metabolites like pigments, exopolysaccharides, and amino acids have tremendous potential to be used in green cosmeceuticals. Mycosporin-like amino acids (MAA) and Scytonemin (SCY), extracted from different cyanobacteria are potential UV protectants and provide protection against harmful solar radiations (Kageyama *et al.*, 2018). Tetrahydropyridines, an MAA derivative, used as sunscreen pigments not only prevent UV damage but also suppress inflammation and have antioxidant activity. SCY extracted from *Nostoc commune* and *Rivularia* sp., can have significant antioxidant and photoprotective activities.

Cyanobacterial exopolysaccharides (EPS) can be a potential source of natural moisturizer as they protect cells from dehydration. Li *et al.* (2010) reported that EPS obtained from *N. commune* could be exploited as a natural humectant in cosmetic industries demonstrating 10.1% water absorption and 28% water retention capacity as compared to urea with 5.8% water absorption and 15.9% water retention capacity. Sacran, an exopolysaccharide obtained from *Aphanothece sacrum* possess higher water absorption efficiency than hyaluronic acid, the most widely used ingredient in moisturizing products.

Due to their beneficial effect on human skin, EU has included Methylsilanol spirulinate (CAS 188012–54-6) *Arthrospira platensis* (CAS 223751–80-2), *Limnospira maxima* in the Cosmetic Ingredient Database (Verma *et al.*, 2022). *Spirulina* extracts are used as moisturizers and skin softeners in beauty products manufactured by Nykaa E-Retail Pvt. Ltd. (India). Blue Green Algae Hair Rescue Conditioning Mask marketed by Aubrey Organics, Inc. (USA) has been reported to help in hair strengthening. Phormiskin Bioprotech G from Codif Recherche and Nature (Paris, France) is derived from *Phormidium persicinum* which has unique photoprotective properties (Chakdar and Pabbi, 2017). Blue Therapy (Lift and Blur) manufactured and marketed by Biotherm (France) uses extracts of *Aphanizomenon flos-aquae* and *Laminaria ochroleuca* which confer anti-ageing effects. Global leaders in the cosmetics industry like Louis Vuitton (France) and Danial Jouvance (France) have developed their own microalgal production systems for manufacturing microalgae-based cosmeceuticals (Verma *et al.*, 2022).

BGA as a Source of Nutraceuticals

Cyanobacteria are a rich source of protein, minerals, vitamins, healthy lipids and antioxidants (Pulz and Gross, 2004; Rosenberg *et al.*, 2008). Cyanobacterial biomass has been utilized as a source of traditional food in many Asian and African countries for centuries. *Spirulina* was consumed as nutritious food in central Africa by Kanembu people, the indigenous population of Asia and North America for a long time. “Dihé” a sun-dried hardened mat of *Spirulina* (*Arthrospira*) collected from Lake Chad is used by the locals as nutrient-rich food (Carcea *et al.* 2015). *Nostoc*, *Anabaena* and *Spirulina* are used as popular food supplements in many South American countries. *Spirulina* is regarded as a superfood

with $\approx 70\%$ protein content as compared to 15-25% from animals or fish. Free fatty acids may account for up to 70% to 80% of the total lipids in *Spirulina* which is also a rich source of gamma-linolenic acid (GLA). Besides, *Spirulina* is also an excellent source of Vitamin B-complex. According to NASA, the nutritional value of 1 kg of *Spirulina* equals 1000 kg of assorted vegetables and fruits. Hence, NASA (CELSS) and the European space agency (MELISSA) proposed *Spirulina* as a major source of nutrition in long-term space missions (Sies, 1996).

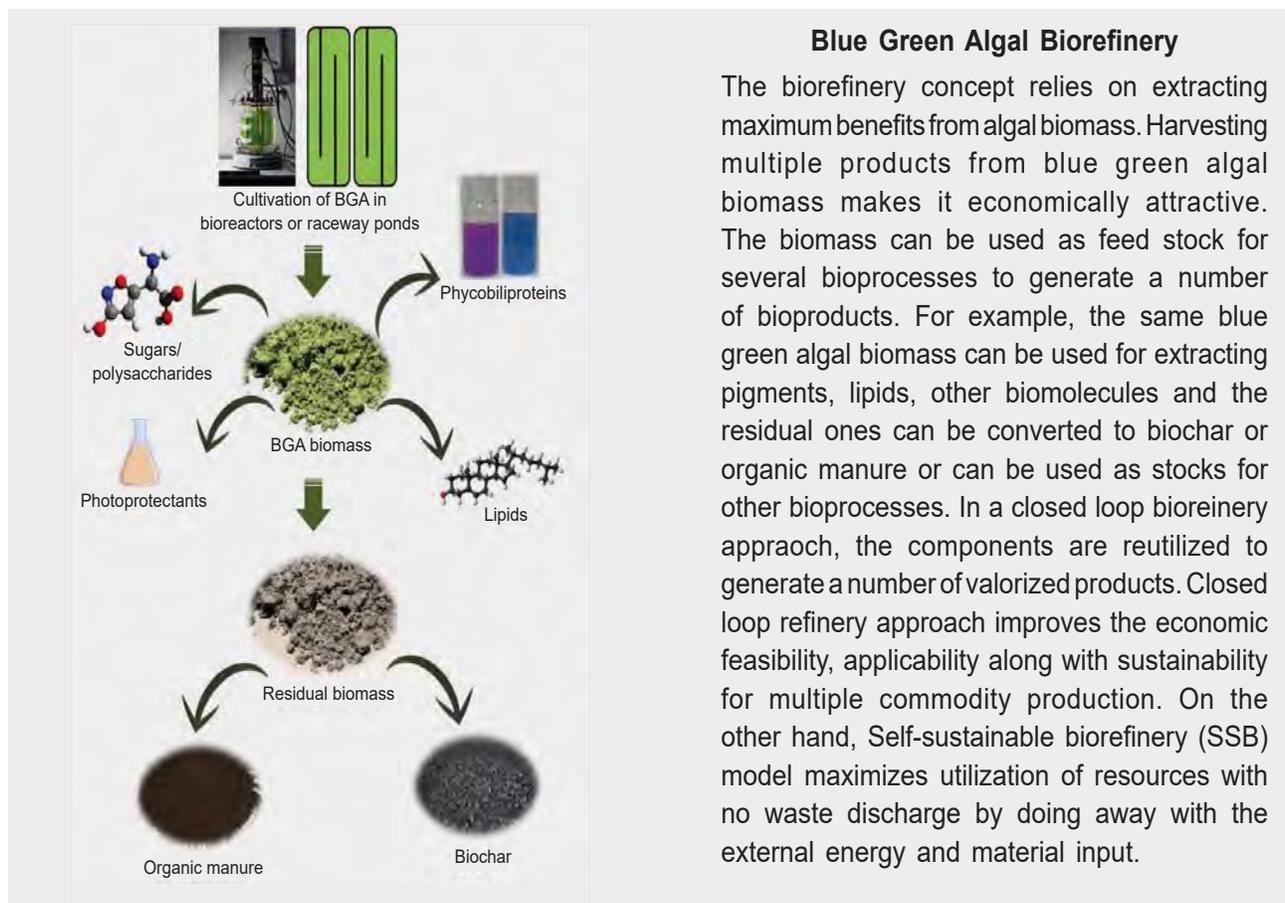
Spirulina is reported as the most dominant commercial cyanobacteria in biomass production with >10000 tons annual production globally (Guidi *et al.*, 2021). A number of commercial preparations are available in many countries including India. In India, EID Parry, Tamil Nadu, Sannat Group, Tamil Nadu, NB Laboratories, Maharashtra, Shibin Exports, Tamil Nadu are some of the major *Spirulina* growers. Annual production of *Spirulina* in India is more than 600 tons per annum. *Spirulina* production can be easily taken up by farmers and entrepreneurs as *Spirulina* powder itself can be sold at a price of Rs. 600-1200.00 per kg. Further, when sold in the form of capsules, each capsule (500 mg) can cost up to Rs. 3-5.0. Conservatively, a *Spirulina* production unit with a capacity of 1000 kg (dried) biomass per year will cost around Rs. 6.0-7.0 lakhs. Therefore, *Spirulina* farming can be an excellent option for secondary agriculture. Apart from *Spirulina*, other cyanobacteria like *Aphanizomenon flos-aquae* (Kalmath's Best ® Blue Green Algae) have also been marketed by Kalmath Valley Botanicals LLC, USA. This product has a considerable market in the USA, Germany, Canada, Korea, Japan and Austria.

Challenges and Prospects

No doubt, the application of cyanobacteria can be a potential avenue in secondary agriculture. The beneficial effects of cyanobacterial biomass, pigments and extracts in nutraceuticals, pharmaceutical, and cosmeceutical industries cannot be overlooked and these have gained much attention in recent years. Increased demand for safer and more efficient natural raw ingredients has further proved the potential of cyanobacteria in the industrial sector. The major advantage of using cyanobacteria for industrial application is their photoautotrophic and diazotrophic growth which can significantly cut down

the cost of commercial cultivation considering a very low or no requirement of C and N sources. Furthermore, this can also reduce the chances of bacterial or fungal contamination. Despite such advantages, the commercial exploitation of cyanobacteria is comparatively low as compared to bacteria or fungi and interestingly, most of the products commercialized are based on *Spirulina*. The majority of the cyanobacterial products in the market are available either as food supplements or food products containing cyanobacterial pigments. It is true that *Spirulina* is a rich source of various nutritional factors and pigments but there are a number of other cyanobacteria which have tremendous commercial potential but have not been exploited. For example, *Nostoc* spp. is rich in nutritional components and *Lyngbya* spp. are a rich source of antimicrobial compounds. Though the research efforts towards understanding the diversity and metabolic potential of cyanobacteria are meagre as compared to bacteria or fungi, and to exploit cyanobacteria for fine chemicals and pharmaceuticals at a commercial scale for use in secondary agriculture, microbiologists and algologists need to reorient their research focus. Cyanobacterial biomass production under outdoor open cultivation is highly influenced by changing environmental conditions which ultimately affect the economic viability. Especially, light quality, quantity and temperature are very critical to cyanobacterial biomass production. Therefore, it is required to identify cyanobacterial species which can sustain a wide range of environmental conditions and result in economically viable biomass production. Lack of high-yielding strains and limitations in improving the inherent yields are the major factors behind relatively lower commercial exploitation. Furthermore, isolation of specific compounds requires downstream processing including extraction and purification.

Large-scale cultivation of cyanobacteria is also critical for commercial exploitation. Raceway ponds and photobioreactors are generally used worldwide for the mass cultivation of cyanobacteria. However, such facilities require huge investments in terms of infrastructure and operational costs. In many instances, a combination of both raceway and photobioreactor systems (Hybrid) has been recommended to cut down the cost of mass cultivation (Bravo-Fritz *et al.*, 2016). The profitability of commercial cultivation of cyanobacteria can be further increased by a biorefinery approach



Blue Green Algal Biorefinery

The biorefinery concept relies on extracting maximum benefits from algal biomass. Harvesting multiple products from blue green algal biomass makes it economically attractive. The biomass can be used as feed stock for several bioprocesses to generate a number of bioproducts. For example, the same blue green algal biomass can be used for extracting pigments, lipids, other biomolecules and the residual ones can be converted to biochar or organic manure or can be used as stocks for other bioprocesses. In a closed loop biorefinery approach, the components are reutilized to generate a number of valorized products. Closed loop refinery approach improves the economic feasibility, applicability along with sustainability for multiple commodity production. On the other hand, Self-sustainable biorefinery (SSB) model maximizes utilization of resources with no waste discharge by doing away with the external energy and material input.

which enhances the economic feasibility by allowing the exploitation of different co-products that can be individually utilized (Bastiaens *et al.*, 2017).

Conclusion

Blue-green algae have tremendous commercial potential and can significantly contribute to the Indian economy. However, lack of awareness, poor availability of commercial strains and process optimisation for cost minimization with output maximization are some of the bottlenecks for their limited commercial exploitation. Nonetheless, industrial exploitation in India has increased significantly in the last two decades with a considerable increase in the number of commercial algae growers. Despite many advantages as a secondary agriculture resource, it is not yet very popular as compared to crop-based or medicinal plant-based products. A concerted effort to explore more avenues in the biorefinery approach and policy decisions to provide capital support from the government (as provided in the case of biofertilizers or biopesticide production units) is urgently required to harness the potential of BGA.

References

- Bastiaens L, S Van Roy, G Thomassen and K Elst (2017) Biorefinery of algae: Technical and economic considerations. In: Gonzalez-Fernandez C and R Muñoz (Eds) Microalgae based biofuels and bioproducts. Woodhead Publishing, Cambridge, UK, pp 327–345.
- Bravo-Fritz CP, CA Sáez-Navarrete, LA Herrera-Zepelin and F Varas-Concha (2016) Multi-scenario energy-economic evaluation for a biorefinery based on microalgae biomass with application of anaerobic digestion. *Algal Res.* **16**: 292–307.
- Carcea M, M Sorto, C Batello, V Narducci, A Aguzzi, E Azzini *et al.* (2015) Nutritional characterization of traditional and improved Dihé, alimentary blue-green algae from the lake Chad region in Africa. *LWT Food Sci. Technol.* **62**: 753–763.
- Chakdar H, S Jadhav, D Dhar, S Pabbi (2012) Potential applications of blue green algae. *J. Sci. Ind. Res.* **71**:13–20.
- Chakdar H and S Pabbi. 2017. Algal pigments for human health and cosmeceuticals. *Algal Green Chemistry*: Elsevier 171-188.
- Guidi F, Z Gojkovic, M Venuleo, PACJ Assunção and E Portillo (2021) Long-term cultivation of a native *Arthrospira platensis* (*Spirulina*) strain in Pozo Izquierdo (Gran Canaria, Spain): Technical evidence for a viable production of food-grade biomass. *Processes* **9**: 1333.

- Kageyama H and R Waditee-Sirisattha (2018) Chapter 5 – Mycosporine-like amino acids as multifunctional secondary metabolites in cyanobacteria: from biochemical to application aspects. In: Attaur R, (ed) *Studies in Natural Products Chemistry*: Elsevier **59**:153–194.
- Li H, J Xu, Y Liu, S Ai, F Qin and Z Li *et al.* (2010). Antioxidant and moisture-retention activities of the polysaccharide from *Nostoc commune*. *Carbohydr. Polym.* **83**:1821-1827.
- Pagels F, AC Guedes, HM Amaro, A Kijjoa, V Vasconcelos (2019) Phycobiliproteins from Cyanobacteria: Chemistry and biotechnological applications. *Biotechnol. Adv.* **37**: 422–443.
- Pulz O and W Gross (2004) Valuable products from biotechnology of microalgae. *Appl. Microbiol. Biotechnol.* **65**: 635–648.
- Rosenberg JN, GA Oyler, L Wikinson, MJ Betenbaugh (2008) A green light for engineered algae: redirecting metabolism to fuel a biotechnology revolution. *Curr. Opin. Biotechnol.* **19**: 430–436.
- Sies H (1996) Antioxidants in Disease, Mechanisms and Therapy, *Academic Press*, New York.
- Verma S, S Thapa, N Siddiqui and H Chakdar (2022) Cyanobacterial secondary metabolites towards improved commercial significance through multiomics approaches. *World J. Microbiol. Biotechnol.* **38** :1-22.
- Vigani M, C Parisi, E Rodriguez-Cerezo, MJ Barbosa, L Sijtsma, M Ploeg and C Enzing (2015) Food and feed products from microalgae: Market opportunities and challenges for the EU. *Trends Food Sci. Technol.* **42**: 81–92.
- Yadav SK, BS Kauldhar, PP Sandhu, K Thakur and TR Sharma (2020) Retrospect and prospects of secondary agriculture and bioprocessing. *J. Plant Biochem. Biotechnol.* **29**: 1-4. <https://doi.org/10.1007/s13562-020-00550-3>
- Yusoff FM, S Banerjee, N Nagao, Y Imaizumi, M Shariff and T Toda (2020) Use of microalgae pigments in aquaculture. In: Jacob-Lopes E, MI Queiroz, LQ Zepka (Eds.) *Pigments from Microalgae Handbook*, Springer International Publishing: Cham, Switzerland pp 471–513.

ARTICLES ON
INSECT GENETIC RESOURCES



ARTICLES ON INSECT GENETIC RESOURCES

Title	Page No.
Role of Invertebrate Diversity with Special Reference to Insects in Agro-biodiversity and Ecological Services –Indian Efforts	383
Insect Diversity and International Cooperation in Invasive Species Research	386
Diversity of Scarab Fauna (Coleoptera: Scarabaeidae) – Importance and Implications	389
Conserving Floral and Faunal Diversity of Rice Paddies	393
Diversity of Bee Pollinators – Global and Indian Perspective	397
Ecological Implications on Insect Biodiversity	401
Insect Collections as a Reservoir of Insect Genetic Resources	406
Arthropod Germplasm Information System (AGIS) in India	408

Role of Invertebrate Diversity with Special Reference to Insects in Agro-biodiversity and Ecological Services –Indian Efforts

M Nagesh* and K Sreedevi

ICAR-National Bureau of Agricultural Insect Resources, Bellary Road, Bengaluru-560024, Karnataka, India

Invertebrates occupy larger proportion in the animal species diversity in almost all habitats and are important components of all ecosystems. The importance of the ecological role played by these invertebrates are not given due consideration and the diversity is under-sampled and understudied. Insects belonging to phylum Arthropoda form a major component that renders several ecological functions and ecosystem services. In this chapter, the importance of the insect services are highlighted and the role of ICAR-National Bureau of Agricultural Insect Resources, Bengaluru in documentation of the insect diversity, its utilization and conservation is emphasized, which are in line with the mandate of the Bureau and Delhi declaration on Agrobiodiversity management.

Introduction

Invertebrates are the most prolific animals in terms of numbers of species and numbers of individuals; abundant in every ecological niche and successful animals on the earth for over 400 million years. Invertebrates belong to the Phylum Arthropoda with segmented bodies, jointed appendages and chitinous exoskeletons. In addition to insects, arthropods include spiders, centipedes, millipedes, scorpions, mites, lobsters, and crabs. Insects are the largest group of arthropods. To date, 1.7 million invertebrate species have been documented out of estimated numbers ranging from 5-30 million. Unfortunately, species numbers are declining faster than we could record their existence. Invertebrates occur at all levels of highly complex food webs, and comprise an enormous biomass that is an important food source for birds and mammals. The diversity of invertebrate faunae in agroecosystems and their relationships to soil processes suggests that they are potential bioindicators.

Insects are the largest and most diverse group of organisms on Earth, comprising of 30 orders with nearly 1 million described species and estimated number exceeding 20 million. Ecological roles of insects are wide and far, contributing to every ecological niche and function. Many are herbivores while others function as predators and parasitoids feeding on other insects and, other arthropods and vertebrates; others as decomposers, feeding on dead wood and leaf litter, carcasses of larger animals, or as soil builders. Importantly insects/

invertebrates sustain pollination and recycle soil nutrients to plants. This illustrates the contribution that insects and other invertebrates make to the composition and functioning of all terrestrial ecosystems, including those in urban greenspaces. The breadth of ecosystem services provided by insects corresponds to an estimated annual economic value of US\$57 billion. Insect pollinators (e.g. bees, flower-flies, and butterflies) pollinate over 85% of wild flowering plants and over 75% of agricultural crop species.

The evidence for decline of global insect diversity is irrefutable. A long-term study found declines of more than 75% of insects in protected areas in Germany. Multiple factors are driving insect declines, including loss and fragmentation of habitat; pollution from light, microplastics, and use of synthetic pesticide and climate change. A world without insects, the loss of partial or whole insect communities can have disastrous effects for food web and reduce an area's ability to recover and be productive.

ICAR-NBAIR and Delhi Declaration on Agrobiodiversity Management

ICAR-NBAIR is the youngest Bureau of ICAR which came in to existence in 2014. ICAR-NBAIR has three Divisions with specific mandate drawn in line with the Agrobiodiversity (ABD) conservation and sustainable use towards achieving the SDGs and the Aichi Targets of CBD, as mentioned below.

*Author for Correspondence: Email-director.nbair@icar.gov.in, Nagesh.m@icar.gov.in

- Biodiversity inventory: biosystematics, molecular diagnostics, genomics
- Biosecurity, ecological services, benefit sharing:
- Invasives, biological control, pollinators, soil builders, organic waste, as feed and food
- Capacity building, social awareness, community participation
- *In situ* biodiversity conservation technologies in cropping systems

Other Activities of ICAR-NBAIR include:

- Identification of traditional agrobiodiversity knowledge: ITKs related to entomophagy (insects as food & supplement) and biodiversity – Erisilk worm, white grubs; Surveys conducted in Pasighat, Arunachal Pradesh and Jorhat, Meghalaya to document insects used as food.
- Waste-to-Wealth (Ecological service): Utilizing Black soldier fly for conversion of organic waste from farm and urban sources to environmentally-safe compost and its by-products.
- Agrobiodiversity conservation: Insect repository and identification services where in cumulative preserved insect specimens in specimens in National Insect Museum at ICAR-NBAIR are 1,92,883 with an average annual additions at 15,000-18,000 specimens and addition of new species 300-310 annually; while *Ex situ* conservation of 136 live-insect cultures maintained in ICAR-NBAIR Insectary and Mass Production Unit; Documentation of potential *Apis* and non-*Apis* pollinator species in different crop ecosystems.
- Coordinated & harmonized management of INVASIVES with SMDs, NARES, SAUs, State line Depts including FAW in maize; Rugose white fly in coconut and palms in southern states, east, west coast etc., movement of planting material; broad mite in mulberry; black thrips in chilli in south and west India, and cassava mealybug in cassava in TN and Kerala; Continuous monitoring and surveillance through AICRP BC centres, SAUs and CIBXRC etc.
- Exchange of natural enemies (both multi- and bi-lateral) for global biological control : Importation of natural enemies on case-to-case basis including *Smicronyx lutulentus* – seed predator on parthenium weed from Queensland, Australia and *Anagyrus*

lopezi – parasitoid on cassava mealybug from IITA, Benin, West Africa.

- Strategies adopted for mitigation of alien invasive species and biocontrol systems at ICAR-NBAIR include identification of indigenous natural enemies against introduced insect pests and suppression of invasives through conservation of indigenous natural enemies.

Priorities in Insect Biodiversity in Agro-ecological Systems for Ecological Services

With the intense anthropogenic activities and rapid climate change patterns, global agriculture in particular and eroding biodiversity there is an urgent need to meet the challenges of global food and nutritional security some of the priorities are

- Distributional information, delimitation of biogeographical areas and their biological importance: Focus on (1) inventory and estimation of faunal richness; (2) monitoring for conservation management and the selection and use of bioindicators; and (3) assessment of conservation status and recovery of threatened species. We then explore the capacity of biosystematics to complement and enhance these programmes.
- INVENTORIZATION and database of agro-ecological zones; MONITORING temporal changes in species abundances, species richness or species assemblages; Bioindication in terms of three different categories of biological indicators: environmental, ecological and biodiversity.
- ENLIST THREATENED SPECIES and ecological communities at international, national and regional levels.
- Evolve strategies for *in situ* conservation of beneficials in soil and crop ecosystems.

References

- Patch EM (1938) Without Benefit of Insects, *Bulletin of the Brooklyn Entomological Society* **33**(1): 1-9.
- Carson R (1962) *Silent Spring*. Boston: Houghton Mifflin Company. Cambridge, MA.
- Montgomery GA, RR Dunn, R Fox, E Jongejans, SR Leather, *et al.* (2020) Is the insect apocalypse upon us? How to find out. *Biological Conservation* **241**: e108327.
- Thomas CD, TH Jones and SE Hartley (2019) “Insectageddon”: A call for more robust data and rigorous analyses. *Global Change Biology* **25**: 1891-1892.

Forister ML, EM Pelton and SH Black (2019) Declines in insect abundance and diversity: We know enough to act now. *Conservation Science and Practice* **1**: e80.

Sanchez-Bayo F and KAG Wyckhuys (2019) Worldwide decline of the entomofauna: A review of its drivers. *Biological Conservation* **232**: 8-27.

Losey JE and M Vaughan (2006) The economic value of ecological services provided by insects. *Bioscience* **56**(4): 311-323.

Ollerton J, Winfree R and S Tarrant (2011) How many flowering plants are pollinated by animals? *Oikos* **120**(3): 321-326.

Isbell F, D Tilman, PB Reich and AT Clark (2019) Deficits of biodiversity and productivity linger a century after agricultural abandonment. *Nat. Ecol. Evol.* **3**(11): 1533-1538.

Insect Invasive Species: Threat Posed and Collaborative Efforts for Management

Richa Varshney*, RS Ramya and Omprakash Navik

ICAR-National Bureau of Agricultural Insect Resources, Bellary Road, Bengaluru-560024, Karnataka, India

Insects are a highly diverse group of organisms and their rich diversity is essential for ecosystem functioning. Insects have more influence on agriculture and most agricultural pests are invasive species that have been introduced into a new ecosystem. These invasive species have the capacity to compete, establish and displace the native species and therefore can be a threat to biodiversity. Introduction of such invasive species has increased with the increase in trade across the globe. Therefore, it is pertinent to strengthen the International cooperation and coordination between the affected countries and the country of native origin to identify the threat for the execution of different mitigation strategies. International Organization for Biological Control (IOBC), Centre for Agriculture and Bioscience International (CABI) and the Biotechnology and Biological Control Agency facilitate international collaboration and play a vital role in managing invasions by alien pests. In India, Directorate of Plant Protection, Quarantine and Storage (DPPQ&S), ICAR-NBPGR, ICAR-NBAIR and other crop-specific institutes of ICAR, Ministry of Agriculture and Farmers' Welfare, Government of India are the bodies engaged in monitoring of invasive insects/invasive alien species. Thus, international collaboration plays a key role in research on the prevention of future invasions, the development of more effective surveillance and eradication methods and the implementation of classical biological control.

Introduction

India is one of the mega biodiversity centres of the world constituting nearly 7% of the world's insect species. In India, the diversity of insects is known for 27 orders, and Coleoptera remains the most speciose insect order. Of the 27 orders, Coleoptera, Lepidoptera, Orthoptera, Diptera, Hemiptera, Hymenoptera, Odonata and Thysanoptera constitute about 94% of insect fauna and the remaining 21 orders represents 6% of insect species (ZSI, 2012). Worldwide, Coleoptera (3,87,000), Lepidoptera (1,57,000), Diptera (1,55,000) and Hymenoptera (1,17,000) are the most speciose insect orders (Stork, 2018).

Insects due to their rich diversity, ecological role, and influence on agricultural, human health and natural resources contribute more for ecosystem functioning (Scudder, 2017). Most major agricultural pests are non-native species that have been introduced into a new ecosystem and cause potential damage to crops. Agricultural intensification and climate change are rapidly decreasing the diversity of insects worldwide.

Additionally, the rise of invasive species also competes, displaces and changes the community structure of several native insect species in various habitats (Fortuna *et al.* 2022).

Threat to Biodiversity and Invasive Alien Insect Species

Invasive alien species are plants, animals, or any other organisms that are introduced unintentionally or deliberately into a new area that is outside their natural habitat (IUCN). These species have the capacity to establish and can become a threat to biodiversity, disrupt ecosystem services, human health, etc. With globalisation, came the dissolution of the natural barriers, and the tremendous boost to trade, tourism and transport have made it easy for many alien pests to enter and invade new habitats.

Once introduced, they engaged in competition for food and space and eliminate native species. Invasive alien species caused 40% of animal extinction since 17th century (CBD 2006). Invasive alien insect species

*Author for Correspondence: Email-Richa.Varshney@icar.gov.in

cause direct damage to crops by feeding or indirectly by transmitting diseases and viruses to many crops and thus poses a serious threat to food security. In India, twenty-three species of insects have been reported until 2019 as invasive species (Naveena *et al.*, 2020). These species caused a huge loss in India. For ex., Papaya mealybug in India caused immense loss. Later importation of parasitoid, *Acerophagus papayae* Noyes and Schauff saved a crop loss of Rs. 435 crores (papaya, tapioca and mulberry) and input cost on pesticides to the tune of Rs. 244.5 crores annually in Tamil Nadu (AICRP-BC Annual Report).

There are many examples of invasive alien insects causing harm to native plants and associated fauna. In the Galapagos Islands, the invasion of *Icerya purchasi* Maskell adversely affected endangered plant *Darwiniothamnus tenuifolius* and significantly reduced the abundance of associated three species of Lepidoptera (Roque-Albelo, 2003). The harlequin ladybird, *Harmonia axyridis* (Pallas), a predator which was introduced to North America and Europe to protect crops is now a globally expanding invasive alien species and has spread to almost every continent (de Groot and Haelewaters, 2022). Arrival of this predator resulted in decline in population of two-spot ladybird, *Adalia bipunctata* (Linnaeus) by 30-40 % in Europe over 5 years (Roy *et al.*, 2012).

International Cooperation to Mitigate Invasive Species

Introduction of species has increased with the increase in trade across the globe. Therefore, it is pertinent to foster international cooperation and coordination between the affected countries and the country of native origin to identify the threat and execution of different management strategies. Management strategies for biological invasion can be accomplished by three strategies, by prevention of introduction, by eradication after establishment and by containment to prevent further spread. Information available on invasive species is very much required for prevention. The information on the potentially damaging species along with their management practices can be accessed through various websites like Global Plant Protection News (<https://iapps2010.me>), Centre for Agriculture and Bioscience International's (CABI) Invasive Species Compendium (<https://www.cabi.org/isc>) etc. Official pest reports are also released by the corresponding National Plant Protection Organizations

of countries of origin to conform to International Plant Protection Convention's (IPPC) Standard on Pest Reporting (ISPM-17) (Liebhold *et al.*, 2021).

International Plant Protection Convention (IPPC) has been identified by the World Trade Organization's Agreement on the Application of Sanitary and Phytosanitary Standards (SPS) as the international body responsible for putting in place standards for phytosanitary treatments and other plant quarantine activities (MacLeod *et al.*, 2010). Critical information necessary for implementing phytosanitary measures is generated by elucidating invasion pathways and helps in decision making to apply different prophylactic phytosanitary treatments (Hennessey *et al.*, 2014). International Organization for Biological Control (IOBC), CABI and the Biotechnology and Biological Control Agency facilitate international collaboration and play a vital role in managing invasions by alien pests. However, collaborative research on biological control must adhere to international treaties, one of which is the Convention on Biological Diversity (CBD) with broad objectives on biodiversity conservation; sustainable use of biodiversity; and on fair and equitable benefit sharing (Liebhold *et al.*, 2021).

In India, Directorate of Plant Protection, Quarantine and Storage (DPPQ&S), ICAR-NBPGR, ICAR-NBAIR and other crop-specific institutes of ICAR, Ministry of Agriculture and Farmers' Welfare, Government of India are the bodies engaged in monitoring Invasive insects/Invasive Alien species (Naveena *et al.*, 2020). DPPQ&S is the responsible body for intercepting invasive insects/weeds/pathogens to the country *via* international trade. Thus, international collaboration plays a key role in research on the prevention of future invasions, the development of more effective surveillance and eradication methods, the implementation of classical biological control, and effective coordination with foreign scientists so as to facilitate collaborative research and exchange of data.

References

- De Groot MD and D Haelewaters (2022) Double Infections of the Invasive Ladybird *Harmonia axyridis*. *Frontiers in Ecology and Evolution* **10**: 756972. <https://doi.org/10.3389/fevo.2022.756972>
- Fortuna TM, P Le Gall, S Mezdour and PA Calatayud (2022) Impact of invasive insects on native insect communities. *Current Opinion in Insect Science* 100904. <https://doi.org/10.1016/j.cois.2022.100904>

- Liebhold AM, T Faith, FT Campbell, DR Gordon, Q Guo, N Havill, B Kinder, R MacKenzie, DR Lance, DE Pearson, SE Sing, T Warziniack, RC Venette and D Yemshanov (2021) The Role of International Cooperation in Invasive Species Research. In Poland TM, Patel-Weynand *et al.* (Eds.). *Invasive species in forests and rangelands of the United States—A comprehensive science synthesis for the United States forest sector*, Springer. pp. 293–304. <https://doi.org/10.1007/978-3-030-45367-1>
- MacLeod A, M Pautasso, MJ Jeger and R Haines-Young (2010) Evolution of the international regulation of plant pests and challenges for future plant health. *Food Secur*, **2**: 49–70.
- Naveena NL, PR Shashank, D Raghavendra and J Mallikarjuna (2020) Invasive insect pests in India: Current scenario and future perspective. *Indian Entomologist* **1**(1): 23-28.
- Roque-Albelo L (2003) Population decline of Galapagos endemic Lepidoptera on Volcano Alcedo (Isabela Island, Galapagos Islands, Ecuador): an effect of the introduction of the cottony cushion scale? *Bull Inst R Sci Nat Belg Entomologie* **73**: 1–4.
- Roy HE, T Adriaens, NJ Isaac, M Kenis, T Onkelinx, GS Martin, *et al.* (2012) Invasive alien predator causes rapid declines of native European ladybirds. *Diversity and Distribution* **18**: 717–725. doi: 10.1111/j.1472-4642.2012.00883.x
- Stork NE (2018) How many species of insects and other terrestrial arthropods are there on earth? *Annual Review of Entomology* **63**: 31-45. <https://doi.org/10.1146/annurev-ento-020117-043348>
- ZSI (2012) COP XI publications. www.zsi.gov.in/Cop-11/cop-11.html/

Diversity of Scarab Fauna (Coleoptera: Scarabaeidae) – Importance and Implications

Kolla Sreedevi*, Judith Corolyn and M Nagesh

ICAR-National Bureau of Agricultural Insect Resources, Bengaluru-560 024, Karnataka, India

Scarab fauna include most economically important dung rollers and white grubs, which are diverse and speciose. These belong to the family Scarabaeidae of Coleoptera. The dung beetles or dung rollers being beneficial play an important role in cleaning the debris/faeces and thus improve the soil structure and fertility through nutrient recycling. On the other hand white grubs are destructive and herbivores causing damage to the plants of several commercial agricultural and horticultural crops affecting the yields. Understanding the species diversity of the dung rollers and white grubs aids in conservation of the former group and management of the latter for the benefit of all lives on this earth. The chapter focuses on the classification, diversity and its importance of both the groups.

Introduction

Scarabaeids belonging to the family Scarabaeidae of Coleoptera comprises of both beneficial coprophagous and harmful phytophagous groups. The former is known as Laprosticti and the latter as Pleurosticti. The family belongs to the superfamily Scarabaeoidea, which is one of the largest superfamilies of the order Coleoptera. It constitutes a very diverse and cosmopolitan group comprising of nearly 31,000 species world wide of which Scarabaeidae constitutes about 91% representing 27,800 species worldwide (Ratcliffe and Jameson, 2004). The superfamily Scarabaeoidea is divided into 12 families, 43 subfamilies, 118 tribes and 94 subtribes (Smith *et al.*, 2006) of which Scarabaeidae is the largest family. The scarab species are widely adapted to most terrestrial habitats comprising of phytophages, fungivores, herbivores, necrophages, coprophages and saprophages and are ubiquitous. The degradation of natural habitats and changes in the land use patterns are posing threat to the biodiversity worldwide (Newbold *et al.*, 2015). The conservation of dung beetles and management of white grubs is utmost crucial and understanding the diversity (both alpha and beta) will help in working towards the targeted goals. The decline in species richness of the dung beetles will threaten their functionality and the ecosystem services rendered (Verdu *et al.*, 2020). So, the present chapter presents a brief overview with respect to the classification and diversity of coprophagous and phytophagous groups of Scarabaeidae.

Classification

Scarabaeidae is the largest family of Scarabaeoidea and according to Lawrence and Newton (1995), it comprises of 11 subfamilies *viz.*, Melolonthinae, Rutelinae, Dynastinae, Cetoniinae, Scarabaeinae, Aphodiinae, Trichiinae, Valginae, Aclopiniae, Orphininae and Allidiostomatinae while Grebennikov and Scholtz (2004) recognized 13 subfamilies, Aphodiinae, Scarabaeinae, Pachypodinae, Orphininae, Allidiostomatinae, Dynamopodinae, Aclopiniae, Euchiriinae, Phaenomeridinae, Melolonthinae, Rutelinae, Dynastinae and Cetoninae and Smith (2006) documented 16 subfamilies, 82 tribes and 94 subtribes under Scarabaeidae. In earlier classification of Ritcher (1966) and Booth *et al.* (1990), Scarabaeidae comprised of six major subfamilies *viz.*, Scarabaeinae, Aphodiinae, Cetoniinae, Rutelinae, Melolonthinae and Dynastinae, where the former two are grouped as Laprosticti that are coprophagous and latter four are grouped as Pleurosticti that are phytophagous. The four groups *viz.*, Scarabaeinae, Rutelinae, Cetoniinae, Dynastinae along with the small subfamily Euchiriinae of the Indian subcontinent were comprehensively explored taxonomically by Arrow (1910, 1917 and 1931) in the three volumes of his 'Fauna of British India' series. Other major groups of taxa that require comprehensive taxonomic treatises are Melolonthinae and Aphodiinae.

The beetles belonging to Laprosticti are primarily dung feeders, while that of Pleurosticti are phytophagous. Among the Pleurosticti, Melolonthinae, Rutelinae and

*Author for Correspondence: Email-kolla.sreedevi@icar.gov.in

few species of Dynastinae comprises of white grubs where either or both larval and adult stages are phytophagous in nature. In certain species of Dynastinae and Cetoniinae, the adults form the damaging stages and their larvae feed on soil organic matter or humus (detritivorous). Of all, Melolonthinae is the largest subfamily comprising of 10 tribes viz., Oncerini, Podolasiini, Sericini, Melolonthini, Diplotaxini, Pachydemini, Dichelonychini, Macroductylini, Lichniini and Hopliini (Evans and Smith, 2009) followed by Rutelinae. According to several workers, Melolonthinae comprises of 10 to 13 tribes, of which Melolonthini, Sericini, Diplotaxini, Macroductylini, Hopliini (Sabatinelli, 1993) are known to occur in India. A comprehensive work on Indian Sericini was carried out by Ahrens and Fabrizi (2016).

Species Diversity in India

The fauna of the Indian subcontinent is very prosperous and diverse, yet to be fully explored. The estimated species of Laprosticti Scarabaeidae will be around 6500+ worldwide, precisely 6,200 species covering 267 genera (Tarasov and Genier, 2015). Pleurosticti Scarabaeidae constitutes nearly 25,000 described species worldwide (Scholtz and Grebennikov, 2005) and occupies more than two thirds of all species of Scarabaeoidea (Ahrens and Vogler, 2008). The maximum number occurs in the tropical areas of the world, particularly in the African and Oriental regions. Ali (2001) reported that the family Scarabaeidae represents about 2500 species from the Indian subcontinent, of which two-thirds are white grubs. Pleurosticti Scarabaeidae include four subfamilies viz., Melolonthinae, Rutelinae, Dynastinae, and Cetoniinae, of which Melolonthinae is the largest subfamily with 750 genera and 11,000 species known to occur in the world and 75 genera with 932 species in India (Houston and Weir, 1992).

An account on diversity of Coprinae belonging to Laprosticti scarab fauna (Arrow, 1931) and the most exhaustive account on Pleurosticti scarab fauna of India and adjacent countries was published by Arrow (1910, 1917) in 'Fauna volumes of British India' wherein 690 species under five subfamilies namely Cetoniinae (241 species), Dynastinae (46 species), Rutelinae (398 species), Desmonychinae (1 species) and Euchirinae (4 species) have been included. Desmonychinae erected by Arrow (1917) as a monotypic taxon with a single genus *Desmonyx* Arrow is unique to Indian subcontinent, which is now merged with Rutelinae by certain workers ([\[sv.wikipedia.org/wiki/Rutelidae\]\(https://sv.wikipedia.org/wiki/Rutelidae\)\) and the Desmonychini Arrow is treated as a tribe of Rutelinae.](https://</p>
</div>
<div data-bbox=)

The second largest subfamily is Rutelinae, and it comprises approximately 200 genera and 4,100 species worldwide (Machatschke, 1972). The subfamily is divided into six tribes, five of which occur in the New World. The tribe Spodochlamyini is found only in Central and South America; the tribe Anoplognathini occurs in Australia and Western Central and South America; the tribe Geniatini is distributed in Central and South America; the tribe Rutelini is widely distributed, but is most speciose in the Neotropics; the tribe Anomalini is widely distributed and is most speciose in the Old World. The tribe Adoretini is exclusively distributed in the Old World. Five tribes Peltonotini, Paratasiini, Anomalini, Adorrhinyptiini and Adoretini are known to occur in India (Arrow, 1917). The economically important genus *Anomala* comprises of 800 species in the Old World (Jameson et al., 2003). Majority of the Ruteline species in India belong to tribes Anomalini and Adoretini. Arrow (1917) reported 398 species to occur in British India, of which 261 species belong to genera *Anomala* and *Adoretus*.

Importance and Implications

Coprophagous: Dung beetles are economically important species that uses the dung of mammals for feeding and nesting. Thus primarily they aid in cleaning the debris by breaking, disintegrating and recycling into the soil. This is an important ecosystem service rendered by these dung beetles. While carrying and decomposing the dung balls, inadvertently the dung beetles disperse the seeds thus indirectly contributing to the multiplication of the plant species diversity. The soil structure is improved by the organic carbon incorporated into the soil through decomposed dung balls. Another added advantage of timely decomposition of dung and faeces is reduction of parasitic flies that are harmful to the livestock and human beings. Certain dung beetles also serve as good bioindicators and thus assessing the coprophagous scarab diversity aid in conserving these dung beetles, which has its multitude benefits.

Phytophagous: White grubs that cause damage to the commercial crops like sugarcane, groundnut, potato, maize, arecanut, soybean, turmeric, ginger, etc. by feeding on the underground portion of the plant at larval stages and aerial parts by the adult beetles are of serious concern. The yield losses by these white grubs

are estimated to range from 40-70% in different crops at different locations in India. The management of the white grub population below the economic threshold level is need of the hour to save the yield losses. In this direction, documentation and knowledge on the diversity and predominance of the white grub species helps in strategizing and timing the management practices in the specified crop and locations.

Conclusion

The diversity in any insect group is yet to be fully explored but specifically in scarab diversity, more explorations and at faster pace needs to be taken up to address the challenges associated with its functional importance. Documentation of the species diversity is the baseline for any further scientific studies that supports database generation. Ahrens and Fabrizi (2016) monograph on Indian Sericini has given a comprehensive account, after which the explorations and taxonomic studies revealed 23 new species of white grubs belonging to tribe Sericini (Sreedevi *et al.*, 2018; Sreedevi *et al.*, 2019; Chandra *et al.*, 2021). This gives us a strong indication of much more taxa to be discovered to explore the diversity. Concerted efforts are to be made towards documenting the dung beetles and white grub diversity of Indian fauna.

References

- Ahrens D and AP Vogler (2008) Towards the phylogeny of chafers (Sericini): Analysis of alignment-variable sequences and the evolution of segment numbers in the antennal club. *Mol. Phylogenet. Evol.* **47**: 783–798.
- Ahrens D and S Fabrizi (2016) A monograph of Sericini of India (Coleoptera: Scarabaeidae). *Bonn Zoological Bulletin* **65**(1): 1-355.
- Ali ATM (2001) Biosystematics of phytophagous scarabaeidae - An Indian Overview. In: Indian Phytophagous Scarabs and their Management: Present Status and Future Strategies (eds. Sharma G., Mathur YS. and Gupta RBL), *Agrobios (India)*, Jodhpur, pp 5-37.
- Arrow GJ (1910) Lamellicornia I: Cetoniinae and Dynastinae. In: *The Fauna of British India, Including Ceylon and Burma*, Thacker Spink and Co., Calcutta, pp. 322.
- Arrow GJ (1917) Lamellicornia II: Rutelinae, Desmoryicinae, Euchirinae. In: *The Fauna of British India, Including Ceylon and Burma*, Thacker Spink and Co., Calcutta, pp. 387.
- Arrow GJ (1931) The Fauna of British India III, Coprinae, Taylor & Francis. London, 1-428
- Booth RG, ML Cox and RB Madge (1990) Guide to insects of importance to man (Coleoptera). International Institute of Entomology, the Natural History Museum United Kingdom.
- Chandra K, D Ahrens, D Bhunia, K Sreedevi and D Gupta (2021) New species and New records of Tribe Sericini (Coleoptera: Scarabaeidae: Melolonthinae) of India. *Zootaxa*, **4951**(3), 492–510. <https://doi.org/10.11646/zootaxa.4951.3.4>
- Evans AV and Smith ABT (2009) *An Electronic Checklist of the New World Chafers (Coleoptera: Scarabaeidae: Melolonthinae). Version 3*. Electronically published, Ottawa, Ontario, Canada, 353 pp. <http://www.museum.unl.edu/research/entomology/SSSA/NW-Melo-v3.pdf>
- Houston WWK and TA Weir (1992) Melolonthinae. In: Houston WWK (Ed), *Zoological catalogue of Australia. Coleoptera: Scarabaeoidea*. Australian Government Printing Service, Canberra, pp.174–358.
- Jameson ML, Paucar-Cabrera, Aura, Solis and Angel (2003) “Synopsis of the New World Genera of Anomalini (Coleoptera: Scarabaeidae: Rutelinae) and Description of a New Genus from Costa Rica and Nicaragua”. *Papers in Entomology*, **96**(4): 415- 432.
- Lawrence JF and AF Newton (1995) Families and subfamilies of Coleoptera (with selected genera, notes, references and data on family-group names). In: *Biology, Phylogeny and classification of Coleoptera* (Pakaluk J and Slipinski, SA eds.) Muzeum Instytut Zoologii PAN, Warszawa. pp.779-1006.
- Machatschke JW (1972) Scarabaeoidea: Melolonthidae, Rutelinae. *Coleopterorum Catalogus Supplementa* **66**: 1-361.
- Newbold T., LN Hudson, SLL Hill, S Contu, I Lysenko, RA Senior, L Borger, DJ Bennett, A Choimes, B Collen, J Day, A De Palma, S Diaz, Londoño S Echeverria, MJ Edgar, A Feldman, M Garon, MLK Harrison, T Alhuseini DJ Ingram, Y Itescu, J Kattge, V Kemp, L Kirkpatrick, M Kleyer, DLP Correia, CD Martin, S Meiri, M Novosolov, Y Pan, HRP Phillips, DW Purves, A Robinson, J Simpson, SL Tuck, E Weiher HJ White RM Ewers, GM Mace, JPW Scharlemann, A Purvis (2015) Global effects of land use on local terrestrial biodiversity. *Nature* **520**: 45–50. <https://doi.org/10.1038/nature14324>
- Ratcliffe BC and ML Jameson (2004) The revised classification for Scarabaeoidea: what the hell is going on. *Scarabs* **15**: 3-10.
- Ritcher PO (1966) White grubs and their allies: A study of North American Scarabaeoid larvae. In: *Oregon State Monographs: Studies in Entomology* (Latyin, JD (ed), Oregon State University Press, pp 6-10.
- Sabatini G (1993) Catalogue of the Scarabaeoidea Melolonthidae from Himalaya and North India: Macroductylini, Hopliini, Diplotaxini, Melolonthini and Sericini. *Memorie della Societa Entomologica Italiana, Genova* **71**(2): 593-636.
- Scholtz CH and Grebennikov VV (2005) 13. Scarabaeoidea Latreille, 1802. In: *Coleoptera, Beetles. vol. 1: Morphology and Systematics (Archostemata, Adepaga, Myxophaga, Polyphaga partim)* (Beutel RG, Leschen, RAB (eds.)). Handbook of Zoology. vol. IV Arthropoda: Insecta part 38. W. de Gruyter-Berlin, New York, pp. 367–426
- Smith ABT (2006) A review of the family-group names for the superfamily Scarabaeoidea (Coleoptera) with corrections to nomenclature and a current classification. *Coleopterists Society Monograph* **5**: 144-204.
- Sreedevi K, J Speer, S Fabrizi and D Ahrens (2018) New species and records of Sericini scarab beetles from the

- Indian subcontinent (Coleoptera, Scarabaeidae). *ZooKeys* **772**: 97–128. <https://doi.org/10.3897/zookeys.772.25320>
- Sreedevi, K., Ranasinghe, S., Fabrizi, S. & Ahrens, D. 2019. New species and records of Sericini from the Indian subcontinent (Coleoptera, Scarabaeidae) II. *Eur. J. Taxon.*, **567**, 1–26. <https://doi.org/10.5852/ejt.2019.567>
- Tarasov S and F Genier (2015) Innovative bayesian and parsimony phylogeny of dung beetles (Coleoptera: Scarabaeidae: Scarabaeinae) enhanced by ontology-based partitioning of morphological characters. *Plos one*, **10**(3)
- Verdú JR, F Sanchez-Pinero, JM Lobo and V Cortez (2020) Evaluating long-term ivermectin use and the role of dung beetles in reducing short-term CH₄ and CO₂ emissions from livestock faeces: a mesocosm design under Mediterranean conditions. *Ecological Entomology* **45**: 109–120. <https://doi.org/10.1111/een.12777>

Conserving Floral and Faunal Diversity of Rice Paddies

Chitra Shanker* and Jhansi Rani Billa

ICAR-Indian Institute of Rice Research, Rajendranagar, Hyderabad, Telangana-500030, India

Rice paddies have complex trophic food webs of flora and fauna. Integrated biodiversity management of flowering forbs can help conserve the beneficial insects of rice ecosystem, in turn enhancing natural biological control of pests. A concept of habitat management through ecological engineering has been developed at the ICAR-Indian Institute of Rice Research, tested, and validated in farmers' fields over several seasons of crop. The strategies are low cost, easy to adopt, ecological sound and environmentally friendly, while providing farmers with monetary returns and nutritional benefits. One of the techniques advocated, is increasing floral diversity. Floral diversity can be maintained by conserving commonly occurring flowering forbs in rice fields or by growing a crop plant that provides monetary and nutritional benefits. Flowering plants on rice bunds had significant impact on biodiversity of parasitoids such as *Anagrus*, *Oligosita* and others. Field studies through baiting and laboratory studies for longevity indicated enhanced parasitization rates and six to ten times increase in longevity of parasitoids. Significant differences were observed in parasitism of hopper eggs in plots with different crop borders in comparison to plots without flower borders ($F = 4.91$; $df 7$; $p < 0.01$).

Introduction

Rice is the staple food crop of 3.5 billion people around the world, mostly from Asia. Rice paddies are unique eco-systems having aquatic, benthic and terrestrial niches that harbour a complexity of flora and fauna which interact with each other. An approach for "Integrated biodiversity management" was proposed by Kiritani (2000). Habitat management through ecological engineering is an ecofriendly technique that is gaining popularity among biocontrol advocates. Though the term was coined by Odum (1971) in the broader sense, it was adapted to pest management in crops (Gurr *et al.*, 2004). It encompasses habitat manipulation through cultural methods to enhance biological control (Gurr *et al.*, 2004) and help harness the existing biodiversity in agro-ecosystems for sustainable pest management. Despite the potential in enhancing the effectiveness of natural enemies of rice pests by manipulating nearby habitat (Gurr *et al.*, 2011), efforts to do so have been limited. One way to conserve natural enemies is through increasing the diversity and density of nectar bearing flowering plants in the rice fields.

Such plants may provide nectar and pollen, important alternate foods for many parasitoids and predators by increasing their longevity and fecundity (Landis *et al.*, 2000). Specific natural enemies whose activities and impact are increased from the presence of specific flowering plants can be identified through research on nectar and pollen qualities. Flower species that meet the needs of natural enemies, while minimising support to pests should be selected (Gurr *et al.*, 2011).

We studied the biodiversity of flowering forbs surrounding rice fields and their potential for utilization in habitat management. A total of eighty plant species were observed belonging to 29 families viz., Acanthaceae, Aizoaceae, Amaranthaceae, Apiaceae, Apocynaceae, Asteraceae, Brassicaceae, Capparaceae, Commelinaceae, Convolvulaceae, Euphorbiaceae, Fabaceae, Juncaceae, Lamiaceae, Lythraceae, Malvaceae, Menispermaceae, Nyctaginaceae, Oxalidaceae, Plantaginaceae, Poaceae, Portulacaceae, Primulaceae, Solanaceae, Tiliaceae, Verbenaceae, Verbenaceae, Verbenaceae and Zygo-phyllaceae. Of these the most amenable for habitat management belonged to the two families Fabaceae and Asteraceae (Table 1).

*Author for Correspondence: Email-chitrashanker@gmail.com; Chitra.Shanker@icar.gov.in

Table 1. Candidate plant species for *in situ* conservation in rice paddies

S.No.	Plant species	Family	Suitable for habitat management	Benefits	Natural enemies
1	<i>Ageratum conyzoides</i> L.	Asteraceae	Y	Pollen, nectar	coccinellids, parasitoid wasps, spiders
2	<i>Parthenium hysterophorus</i> L.	Asteraceae	N	Pollen, nectar	coccinellids
3	<i>Spheranthus indicus</i> Linn.	Asteraceae	N	Pollen, nectar	coccinellids, mirids
4	<i>Acmella uliginosa</i> (Sw.) Cass.	Asteraceae	Y	Pollen, nectar	coccinellids, mirids, spiders
5	<i>Eclipta prostrata</i> L.	Asteraceae	Y	Pollen, nectar	coccinellids, parasitoid wasps, spiders
6	<i>Tridax procumbens</i> L.	Asteraceae	Y	Pollen, nectar, alternate prey	coccinellids, parasitoid wasps
7	<i>Caesulia axillarisi</i> Roxb.	Asteraceae	N	Pollen, nectar	spiders
8	<i>Vigna trilobata</i> (L.) Verdc.	Fabaceae	Y	Nectar, alternate prey, honey dew	coccinellids, parasitoid wasps, spiders
9	<i>Macropitium atropurpureum</i> (DC.) Urb.	Fabaceae	N	Nectar, honey dew, alternate prey	coccinellids, parasitoid wasps, spiders
10	<i>Melilotus alba</i> Medik.	Fabaceae	Y	Nectar, alternate prey, honey dew	coccinellids, parasitoid wasps, spiders

Functional Significance of Floral Biodiversity

The seasonal abundance of coccinellids and their prey was recorded on the rice crop and the surrounding flora (Shanker *et al.*, 2018). Coccinellids abundant in rice ecosystem was found to establish on weed flora surrounding rice fields before moving into the rice crop. Similarly, an outward movement from rice crop to weeds was observed after harvest of the rice crop. Coccinellid prey range assessed using PAGE electrophoresis indicated that the leafhoppers and aphids on the weeds were the prey of the coccinellids before they colonized the rice fields. The alternate prey such as *Aphis gossypii* Glover, *Aphis craccivora* (Koch), *Cicadulina bipunctata* (Melichar), *Schizaphis graminum* (Rondani), *Sitobion* sp., *Thaia oryzivora* Ghauri and *Zygina maculifrons*

Matsumura infesting weed flora surrounding rice fields were utilized by coccinellid predators - *Harmonia octomaculata* (Fabricius), *Micraspis discolor* (F.), *Propylea dissecta* (Mulsant), *Coccinella transversalis* Fabricius, *Cheilomenes sexmaculata* (Fabricius), *Scymnus nubilus* Mulsant and *Brumoides suturalis* (Fabricius).

Potted rice plants exposed to leafhopper/ planthopper adults for oviposition were used as bait for assessing parasitisation in the field with and without a flower border. The freshly emerged parasitoids were exposed to newly opened flowers of the border plants in the laboratory. The survival of adults was observed in comparison to the parasitoids directly offered honey and those without any nectar source. The mean percent parasitisation of

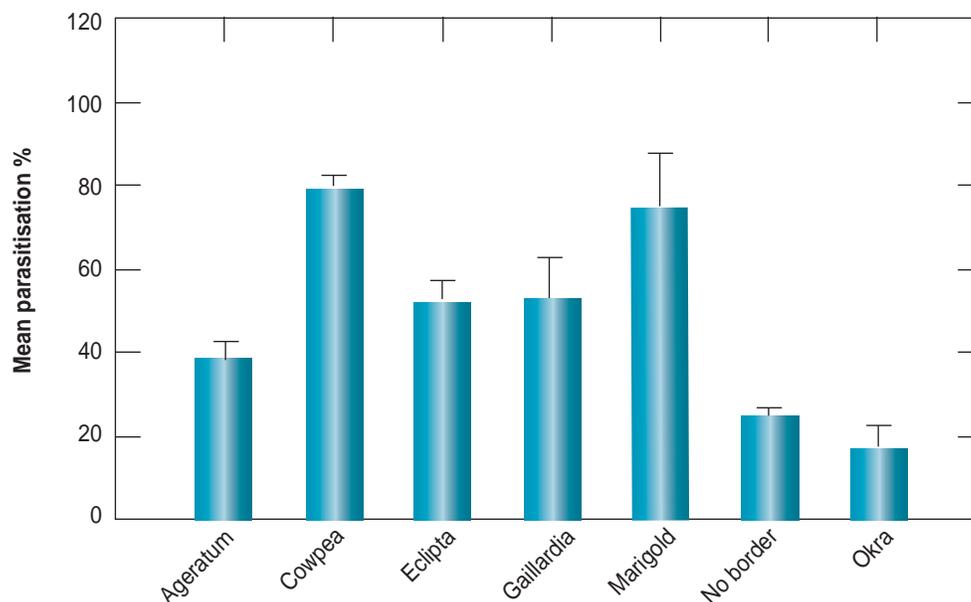


Fig. 1. Impact of floral diversity on rice bunds on egg parasitisation of plant hoppers

hopper eggs ranged from 22.53–77.91 per cent and was significantly higher in plots with flowering borders of *Vigna*, *Ageratum* and *Eclipta* (LSD = 24.15, P=0.01). The longevity of the parasitoid, *Gonatocerus* sp., (in days) was highest on *Eclipta* (7.11±1.45) followed by *Tagetus* (6.83±1.40) and *Tridax* (6.35±1.37). The longevity of *Oligosita* sp. increased 10-fold when offered food/ nectar source. Flowering plants both enhanced longevity and parasitisation.

In addition to *in situ* conservation of flowering weeds, studies on various bund crops such as *Tagetus erecta* L. (Asteraceae), *Vigna unguiculata* L., *V. mungo* (L.) Hepper, *V. radiata* (L.) R. Wilczek, *Crotalaria juncea* L. (Fabaceae), *Coriandrum sativum* L. (Apiaceae), and *Abelmoschus esculentus* (L.) Moench (Malvaceae) were tested and validated in multi-locations. Commercial flowers, pulses, vegetables green manure crops can be grown on bunds. Hopper populations were significantly lower in fields with a bund crop and limited use of insecticide (0.11 to 7.11/ hill) compared to farmers’ practice fields with average of five sprays in a season and with no bund crop (9.22/hill) and untreated control (433/ hill). Bund crops had significant impact on parasitism of hopper eggs compared to plots without flower borders (F= 4.91; df 7; p <0.01). Highest parasitism of hopper eggs was observed in plots with black gram border (45.6 %) while the lowest was observed in farmer’s practice plots

without flower border (11.4%). However, highest benefit: cost ratio of 5.25 was recorded in rice fields with marigold as bund crop due to additional economic gain while the lowest ratio of 2.64 was observed in farmers’ fields without bund crop. Integrated biodiversity conservation of flora and fauna can reap rich ecological benefits (Fig 2).

Future Prospective and Action Points

1. The impetus on organic farming and the use of alternative methods of pest management point towards the use of habitat management strategies for in situ conservation of floral and faunal diversity of agro ecosystems and harnessing their ecosystem services.
2. Government support for organic methods and the need to double the income of farmers is a two-edged sword. The use of low-cost technologies that rely on conservation tactics can help farmers to reduce pest management costs while reaping some extra monetary returns.
3. Rice paddies can also be considered as conservation habitat for avian biodiversity and host numerous winter migrants. Our observations indicate rice fields and surrounds to harbour 170 species of birds over the year.
4. Odonate diversity is also the highest in rice paddies. This group has many endangered species and

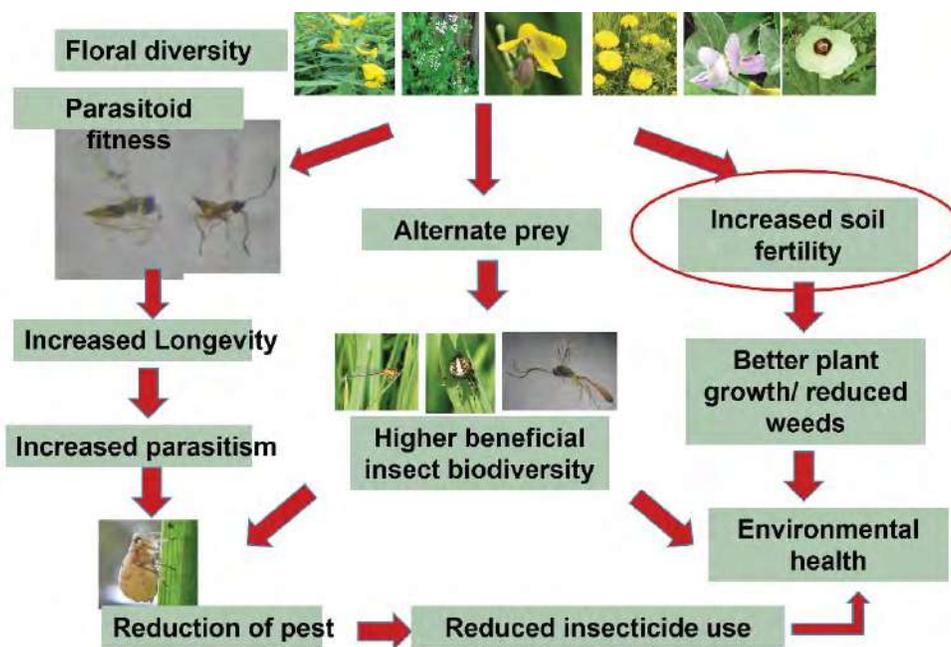


Fig. 2. Ecological benefits of Biodiversity management in rice paddies

biodiversity management in rice paddies can be the way forward for conservation of such threatened taxa.

References

- Gurr GM, SL Scarratt, SD Wratten, L Berndt and N Irvin (2004) Ecological engineering, habitat manipulation and pest management. In: GM Gurr, SD Wratten and MA Altieri (Eds.), *Ecological engineering for pest management: Advances in habitat manipulation for arthropods* (pp. 1-12). Wallingford, UK: CABI Publishing.
- Gurr GM, J Liu, DMY Read, JLA Catindig, JA Cheng, LPLan and KL Heong (2011) Parasitoids of Asian rice planthopper (Hemiptera: Delphacidae) pests and prospects for enhancing biological control. *Ann. Appl. Biol.* 158: 149-176.
- Kiritani K (2000) Integrated Biodiversity Management in Paddy Fields: Shift of Paradigm from IPM toward IBM. *Integr. pest manag. rev.* 5: 175–183.
- Landis D., SD Wratten and G Gurr (2000) Habitat manipulation to conserve natural enemies of arthropod pests in agriculture. *Annu. Rev. Entomol.* 45: 175-201.
- Odum HT (1971) *Environment, power and society*. New York: John Wiley & Sons
- Shanker C, L Chintagunta, S Muthusamy, S Vailla, A Srinivasan and G Katti (2018) Flora surrounding rice fields as a source of alternative prey for coccinellids feeding on the pests of rice. *Eur. J. Entomol.* 115: 364–371.

Diversity of Bee Pollinators – Global and Indian Perspective

Amala Udayakumar, Kesavan Subaharan* and TM Shivalingaswamy

ICAR-National Bureau of Agricultural Insect Resources, Bellary Road, Bengaluru-560024, Karnataka, India

Bees are important diverse group of pollinators that contribute to pollination and reproductive success of many angiosperms. Of the hundred principal crops that accounts for world's food supply, only 15 % was pollinated by domestic bees (honeybees and bumble bees) and 80% are pollinated by wild non-apis bees (leaf cutter bees, sweat bees, digger bees). Honeybees, bumble bees and stingless bees were social in habit with typical caste system (queen, drones and workers) and division of labour. Seven different species of honeybees were reported from India viz., *Apis mellifera*, *A. cerana*, *A. florea*, *A. dorsata* and *A. labriosa*. Among the different species of honeybees, *A. mellifera* and *A. cerana* were domesticated in India for the honey production. Native bees like leaf cutter bees, sweat bees, digger bees were solitary in habit and constructs their habit in hollow cavities, pithy stems and underground. The native bees buzz pollinates plants with typical floral morphology results in increased fruit/pod set and yield in different crops.

Pollination

Pollination is an ecosystem process that has evolved over millions of years to benefit both flowering plants and pollinators. Pollinators visit flowers for many reasons, including feeding, pollen collection and for gaining warmth. When pollinators visit flowers, pollen rubs or drops onto their bodies. The pollen is then transferred to another flower or a different part of the same flower as the pollinator moves from one location to the next. This process is a vital stage in the life cycle of all flowering plants and is necessary to start seed and fruit production in flowers. Not only do pollinators provide essential services in nature, they are also necessary for healthy, productive agricultural ecosystems as they ensure the production of full-bodied fruit and fertile seed sets in many crops.

Contribution of Bee pollinators to agriculture

Pollination services in economic terms was valued to the tune of about \$219 billion per year contributing to 9.5% of global crop value (Calderone, 2011) both the services provided by managed honeybees and wild non-apis bees. Native bees complement the role of honeybees in many crops contributing substantially to the crop pollination.

Melittophily-Bees as Pollinators

Diversity of Bee Pollinators

Bees are important group of pollinators with around 20,000 described species worldwide (Michener, 2007).

Apidae is the well distributed first largest family that includes honeybees, bumble bees and carpenter bees with around 5700 species reported worldwide. The family Apidae comprises of three major subfamilies viz., Apinae, Xylocopinae and Bombinae with around 25 genera of bees (Pannure, 2016). The native solitary bees belong to the families Megachilidae (leaf cutter bees), Halictidae (sweat bees), Colletidae (minner bees) and Melittidae (digger bees).

Honeybees

Honeybees are major group of insect pollinators that belongs to the genus *Apis*, family Apidae and order Hymenoptera. Seven different species of honeybees were reported from Asia viz., European honeybee *Apis mellifera*, Indian honeybee *A. cerana*, dwarf or little honeybee *A. florea*, rock bee or giant bee *A. dorsata* and giant mountain honeybee *A. labriosa* (Yadav *et al.*, 2017).

Apis mellifera

This is a domesticated bee species with habits similar to Indian honeybee *A. cerana indica* except for the behaviour of construction of parallelly arranged multiple combs with a uniform bee space. This species of honeybee is widely domesticated in America, Australia, Asia, Europe and Africa. Across the continents of distribution of this species of bee, *A. mellifera* around 20 subspecies or races were designated globally due to the tremendous variations in the species. Based on the morphometric and

*Author for Correspondence: Email-Kesavan.Subaharan@icar.gov.in

genetic analysis supported with ecological, physiological and behavioural traits, four major groups of *A. mellifera* was reported (Franck *et al.*, 2001; Miguel *et al.*, 2011). The four reported races of *A. mellifera* includes group A that comprised of all subspecies throughout Africa; group M that includes subspecies from Western and Northern Europe; group C includes subspecies from Eastern Europe and group O includes species from Turkey and Middle East.

The Africanized bees were reported to be hybrid species of *A. mellifera* often referred as ‘killer bees’ have a habit of aggressive stinging with a habit of chasing the intruders over greater distances during the instances of miss handling and excitation (Nogueira-Neto, 1964; Rinderer & Hellmich, 1991). This aggressive race of *A. mellifera* were reported to establish when *A. mellifera* from South Africa mated with Brazilian population and this strain was first reported in Brazil in the 1950s later deduced through America.

Apis cerana

This species commonly referred as Indian honeybee of Asian honeybee is native to Asia. This is widely domesticated species in India with gentle temperament and easy to handle. *A. cerana* has a strong tendency of swarming and absconding. The honey yield is less compared to the Western honeybee, *A. mellifera* but with wider adaptability for domestication. This species is relatively resistant to all pest and diseases infesting the colonies. In India, two races of *A. cerana* was reported (Kapil 1956; Narayanan et al. 1960a&b) viz., black ‘hill’ morph adapted to thrive in higher elevations and yellow ‘plain’ morph in lower elevations. Three subspecies of honeybees were also reported by International Centre for Integrated Mountain Development (ICIMOD) from India *A. cerana indica* found in plains, *A. cerana cerana* in the north-west Himalayas, parts of Pakistan and Nepal and *A. cerana himalaya* in hills of Nepal, Uttar Pradesh, the north-east Himalayas and Bhutan.

Apis florea

This species of bee commonly referred as ‘little bee’ or ‘dwarf bee’ construct nests with single small sized comb (Vaudo *et al.*, 2012). Similar to *A. dorsata* they have an open nesting habit usually nesting in trees or shrubs or branches. They are highly prone to swarm with gentle in temperament. The honey yield of this bees is very low and due to its higher swarming behaviour, this species of bee is not domesticated in India.

Apis dorsata

This species commonly referred as ‘rock bee’ or ‘giant honeybee’ distributed across India occurring commonly in foothills of mountains, forest areas and swarms of this bees often can be seen in urban dwellings. They construct open exposed nest with a single comb with a strong defensive behaviour (Woyke *et al.*, 2001; Liu *et al.*, 2007). Because of the higher migratory behaviour with aggressiveness, comb structure with a strong temperament during handling, the domestication of this species is meagrely attempted in India.

Apis labriosa

This species of bees was reported from Nepal, Bhutan, Sikkim and Yunnan between altitudes of 1200 and 1400 m. They are relatively larger in size compared to *A. dorsata* and were reported to be active in lower temperatures and greater heights (Gupta, 2014). They construct exposed single combs in mountainous areas.

Dammer Bees or Stingless Bees

Stingless bees belonging to the genus *Tetragonula* and *Melipona* are smaller in size, easily amenable for domestication; construct their nests of irregular combs of colonies using wax and resin in cracks, walls and other places (Leonhardt *et al.*, 2007; Kumar *et al.*, 2012). In Indian subcontinent, eight species of stingless bees were reported viz., *Lepidotrigona arcifera* (Cockerell), *Lisotrigona cacciae* (Nurse), *Lisotrigona mohandasi* Jobiraj & Narendran, *Tetragonula aff. laeviceps* (Smith), *Tetragonula bengalensis* (Cameron), *Tetragonula gressitti* (Sakagami), *Tetragonula iridipennis* (Smith), *Tetragonula praeterita* (Walker), and *Tetragonula ruficornis* (Smith) (Rasmussen 2013). Stingless bees have lesser absconding and swarming behaviour compared to honeybees and they are relatively more tolerant to diseases as their nests are made of resin/propolis. Stingless bees play a crucial role in the pollination of mango, strawberry, coconut, lablab, gooseberry and cucurbits.

Bumble Bees

Bumble bees belongs to the subfamily Bombinae are social in habit with typical caste system ie., queen, worker and drone like honeybees thrive in the higher elevations and temperate climatic conditions. In India, around 62 species of bumble bees were reported from the Himalayan regions (Williams, 2022). Bumble bees are commercially utilised buzz pollinators for enhancing

fruit/seed set in crops grown under protected cultivations like strawberry, tomato, brinjal and cucurbits.

Native Solitary Bees

Leaf Cutter Bees

Leafcutter bees are solitary in habit belonging to the family Megachilidae. Adult females of leaf cutting bees belonging to the genus *Megachile* sp uses leaf pieces/resin/soil particles/minute pebbles to construct nests in the pre-existing cavities, hollow stems, dead woods and manmade holes (Cane *et al.*, 2007). The genus *Megachile* with 1489 species is the largest in the family Megachilidae (Kunjwal *et al.*, 2020). In India, 237 megachilid bee species including 96 species of the genus *Megachile* are reported (Ascher & Pickering, 2018). These bees are specialised pollinators of plants with ‘papilionaceous flowers’ and they trip the flower to get the pollen reward (Abrol *et al.*, 1990). They are important commercially utilised pollinators of alfalfa (Pitts Singer & Cane, 2011), legumes (Amala & Shivalingaswamy, 2021) *etc.*

Sweat Bees

They are solitary in habit belongs to the family Halictidae with subterranean nesting habit. The world fauna counts over 250 species of halictid bees (Astafurova, 2013). These bees were attracted to human perspiration. Few species of Halictids, *viz.*, *Hoplonomia westwoodi*, *Nomia* sp, *Lasioglossum* sp were reported to be the buzz pollinators of solanaceous crops like tomato and brinjal (Amala & Shivalingaswamy, 2018).

Carpenter Bees

Carpenter bees belongs to the subfamily Xylocopinae are a widespread bee fauna actively foraging on wide range of crops plants (Michener, 2007), polylectic and were reported to be an efficient pollinator of wide range of crops temperate fruits and oilseed crops such as niger, safflower, mustard, linseed (Navatha & Sreedevi, 2015), apple, raspberry, cranberry, cosmos, sunflower, red gram, tomato, winged bean, mustard, alfalfa (Mattu & Kumar, 2016). Two groups of carpenter bees *viz.*, large carpenter bees (*Xylocopa* sp) construct their nests in hard woods like bamboos and other trees and small carpenter bees (*Ceratina* sp) construct their nests in plants with softwood pithy stems (Amala & Shivalingaswamy, 2019). Large carpenter bees are one of the world’s most diverse groups of solitary bees, with 373 described

species in the world of which 29 species are from India (Prasantha and Belavadi, 2016).

Digger Bees

Digger bees belongs to the subfamily Anthophorinae, families Colletidae, Melittidae and Andrenidae. Around 50 species of Melittid bees are reported in India (Saini *et al.*, 2019). Bees belonging to the genus, *Amegilla* (Anthophorinae) builds a solitary nest, in burrows in dried-up river banks, old clay homes, and mortar between bricks, but may also burrow in soft sandstone. Cells, at the end of tunnels, contain an egg with a pollen/nectar mixture for the larval food. *Amegilla* sp is reported to be an efficient buzz pollinator of Solanaceous vegetables across the globe (Hoogendorn *et al.*, 2007).

References

- Amala U and TM Shivalingaswamy (2018) Nesting biology, seasonality and host range of sweat bee, *Hoplonomia westwoodi* (Gribodo) (Hymenoptera: Halictidae: Nomiinae). *Sociobiology* **65**(3): 491-496.
- Amala U and TM Shivalingaswamy (2019) Nest architecture and life cycle of Small Carpenter bee, *Ceratina binghami* Cockerell (Xylocopinae: Apidae: Hymenoptera). *Sociobiology* **66**(1): 29-33.
- Amala U and TM Shivalingaswamy (2022) Leafcutter Bees (Hymenoptera: Megachilidae) as Pollinators of Pigeon Pea (*Cajanus cajan* (L.) Millsp., Fabaceae): Artificial Trap Nests as a strategy for their conservation. *Sociobiology* **69**(1): DOI: 10.13102/sociobiology.v69i1.7202
- Ascher J and J Pickering (2018) Discover life bee species guide and world checklist (Hymenoptera:Apoidea:Anthophila). https://www.discoverlife.org/mp/20q?guide=Apoidea_species
- Ascher JS and J Pickering (2010) Apoidea Species Guide. Available from URL: http://www.discoverlife.org/mp/20q?guide=Apoidea_species.
- Astafurova YV (2013) Geographic Distribution of Halictid Bees of the Subfamilies Rophitinae and Nomiinae (Hymenoptera, Halictidae) in the Palaearctic. *Entomol. Rev.* **93**(4): 437–451.
- Claderone MW (2013) Insect Pollinated crops, Insect Pollinators and US Agriculture, Trend analysis of Aggregate data for the period 1999-2009, *Plos One*, 372-375.
- Franck P, L Garnery, A Loiseau, BP Oldroyd, HR Hepburn, M Salignac et al (2001) Genetic diversity of the honeybee in Africa: microsatellite and mitochondrial data. *Heredity* **86**: 420–430
- Gupta RK (2010) An annotated catalogue of the bees of the Indian region. URL: beesind.com/beesind2.
- Gupta RK (2014) Taxonomy and distribution of different honeybee species (Eds) Beekeeping for Poverty Alleviation and Livelihood Security, pp 63–101.
- Hoogendorn K, N Coventry and MA Keller (2007) Foraging behaviour of a blue banded bee, *Amegilla chlorocyanea*

- in greenhouses: implications for use as tomato pollinators. *Apidologie* **38**: 86-92.
- Kapil RP (1956) Variation in the biometric characters of the Indian honeybee *Apis cerana indica*. *Indian J. Ent.* **28**: 440-457
- Kumar MS, AJAR Singh and G Alagumuthu (2012) Traditional beekeeping of stingless bees (sp.) by Kani tribes of Western Ghats, Tamil Nadu, India, **11**: 342-345.
- Kunjwal N, MS Khan, G Kumar and P Srivastava (2020) Notes on the nesting ecology of the Megachile bees from North India. *J. Apic. Res.* doi.org/10.1080/00218839.2020.1774151.
- Leonhardt SD, K Kworschak, T Eltz and N Bluthgen (2007) Foraging loads of stingless bees and utilization of stored nectar for pollen harvesting. *Apidologie* **38**: 125-137. doi: 10.1051/apido: 2006059.
- Liu F, DW Roubik, D He and J Li (2007) Old comb for nesting site recognition by *Apis dorsata* Field experiments in China. *J. Insect. Sci.* **54**: 424-426.
- Mattu VK and A Kumar (2016) Diversity and relative abundance of solitary bees on *Jatropha curcas* crop in Sirmour and Solan Hills of Himachal Pradesh, India. *Int. J. Sci. Res.* **5**(5): 1815-1818.
- Michener CD (2007) *The Bees of the World* (Second Edition), The Johns Hopkins University Press.
- Miguel I, M Baylac, M Iriondo, C Manzano, L Garnery and A Estonba (2011) Both geometric morphometric and microsatellite data consistently support the differentiation of the *Apis mellifera* evolutionary branch. *Apidologie* **42**: 150-161
- Narayanan ES, PL Sharma and KG Phadke (1960a) Studies on biometry of the Indian bees III. Tongue length and number of hooks on the hind wings of *Apis cerana indica* F. collected from Madras State. *Indian Bee J.* **23**: 3-9
- Narayanan ES, PL Sharma and KG Phadke (1960b) Studies on biometry of the Indian bees. 1. Tongue length and number of hooks on the hind wings of *Apis cerana indica* F. *Indian Bee J.* **22**: 81-88
- Navatha L and K Sreedevi (2015) Pollinator diversity of solitary bees in oilseed crops. *Curr. Biot.* **8**(4): 375-381.
- Nogueira-Neto P (1964) The spread of a fierce African bee in Brazil. *Bee World* **45**: 119-121. doi: 10.1080/0005772x.1964.11097060.
- Pannure A (2016) Bee Pollinators decline: Perspectives from India. *Int. Res. J. Nat. Sci.* **3**(5): 1-10.
- Pitts Singer TL and JH Cane (2011) The Alfalfa Leafcutting Bee, *Megachile rotundata*: The World's Most Intensively Managed Solitary Bee. *Annu. Rev. Entomol.* **56**: 221-237.
- Prashantha C and VV Belavadi (2016) Large carpenter bees, *Xylocopa* from central Western Ghats of India: taxonomic notes on subgenus *Koptortosoma* (Hymenoptera: Apidae: Xylocopinae). *Oriental Insects* doi.org/10.1080/00305316.2016.1270233.
- Rasmussen C (2013) Stingless bees (Hymenoptera: Apidae: Meliponini) of the Indian subcontinent: Diversity, taxonomy and current status of knowledge. *Zootaxa* **3647**: 401-28. doi: 10.11646/zootaxa.3647.3.1.
- Rinderer TE, and RL Hellmich (1991) "The Process of Africanization," in *The "African" Honey Bee*, M Spivak, DJC Fletcher and MD Breed (Eds) (Boulder CO: Westview Press), 95-117. doi: 10.1525/ahu.2004.29.1.95
- Saini J, K Chandra and H Kumar (2016) Description of a new species of genus *Melitta* (Hymenoptera: Melittidae) from India. *Oriental Insects* **54**(3): DOI: 10.1080/00305316.2019.1624221
- Vaudo AD, JD Ellis, GA Cambray and M Hills (2012) Honey bee (*Apis mellifera capensis*/A. m. *scutellata* hybrid) nesting behavior in the Eastern Cape, *South Afr. J. Sci.* **59**(3): 323-331.
- Williams PH (2022) *The Bumblebees of the Himalaya – an Identification Guide*. Abc Taxa ISBN: 978-9-0732-4256-2.
- Woyke J, J Wilde and M Wilde (2001). Swarming, migration and absconding of *Apis dorsata* colonies. In: *Proceedings of the 7th international conference on apiculture in tropical climates*, Chiang Mai, pp 183-188
- Yadav S, Y Kumar and BL Jat (2017) Honeybee: Diversity, Castes and Life Cycle. In: Omkar (eds) *Industrial Entomology*. Springer, Singapore. https://doi.org/10.1007/978-981-10-3304-9_2

Ecological Implications on Insect Biodiversity

Amala Udayakumar and Kesavan Subaharan

ICAR-National Bureau of Agricultural Insect Resources, Bellary Road, Bengaluru-560024, Karnataka, India

Insects are important group of organisms that are diverse forming an important component of food web. They play vital ecological functions like nutrient recyclers, soil decomposers, pollinators, biocontrol agents, soil builders and scavengers. Anthropogenic factors like habitat destruction, land fragmentation, deforestation, climate change, monocropping and urbanization threaten the native diversity of the insects with possible loss in their abundance, richness and extinction. There is a growing need to conserve the diversity of insects using tactics like maintenance of crop diversity amidst the crop areas to serve as refugia and breeding grounds for the beneficial insects, creating of breeding habitats for natural multiplication of beneficials. Adoption of minor agronomic practices leading to crop and habitat diversification benefits native insect population for their natural proliferation in agro-ecosystems.

Insect Biodiversity—A Crucial Factor for Ecological Balance

Insect are largest and diverse group of organisms in Earth that constitutes to around 75% of all described animal species. The abundance of the insects is contributed by factors like flight ability, faster rates of multiplication, plasticity in their behavior and increased fecundity. Insects like butterflies, bees, dragonflies are referred as ‘bio-indicator’ species as they their occurrence and abundance rapidly respond to anthropogenic disturbances in ecosystem (Pollard, 1991). Many groups of insects are vital components of food web by serving as pollinators, nutrient recyclers, decomposers and biocontrol agents that regulate insect pest populations. The role of insects as scavengers is of paramount importance as they are first to visit organisms on carcass that aids in its faster degradation. Anthropogenic interventions directly threaten the diversity of insects resulting in species extinction. According to the Zoological Survey of India, 1,318 species of butterflies in the country, of which, according to the International Union for Conservation of Nature (IUCN), 35 species are critically endangered. There is a need to conserve the insect diversity with an aim to keep the food web intact in any ecosystem.

Vital Ecological Role Played by Insects

Insects in Nutrient Recycling

Insects plays a major role in nutrient recycling through degradation of leaf litters and organic matter. Dung beetles and few species of flies are referred as

‘coprophages (faeces eaters) as they oviposit in the fecal matter of other animals. There are three different functional types of dung beetles (Doubé, 1990),

- Tunnelers (paracoprids) – they build tunnels under the pile of dung and move the dung into the tunnels, oviposits on it and develops inside the tunnel.
- Dwellers (endocoprids) – they construct burrows or develops just underneath the fresh dung piles.
- Rollers (telecoprids) – they remove a ball of dung roll in to tunnels far away from the dung pile.

The tunnelers group of dung beetles were reported to improve aeration to the roots of pasture plants and aiding in soaking of water at the root zones improves aeration to the crops. During the process of decomposition of the dung, the beetles feed upon the dung converting it into nitrogen that gets assimilated by the plants for their growth and development. The physical and chemical properties of soil was influenced by the dung relocation and burrowing activities of the dung beetles. The decomposition process of the dung aids in the homogenization of microbial communities during the course of action (Slade *et al.*, 2016).

Insects as Pollinators

Pollination is an ecosystem process that has evolved over millions of years to benefit both flowering plants and pollinators. Pollinators visit flowers for many reasons, including feeding, pollen collection, and warmth. When pollinators visit flowers, pollen rubs or drops onto their bodies. The pollen is then transferred to another flower

*Author for Correspondence: Email-Kesavan.Subaharan@icar.gov.in

or a different part of the same flower as the pollinator moves from one location to the next. This process is a vital stage in the life cycle of all flowering plants and is necessary to start seed and fruit production in flowers. Animals pollinate approximately 75 percent of the crop plants grown worldwide for food, fiber, beverages, condiments, spices, and medicines. The principal pollinators are bees. Approximately 73 % of the world's cultivated crop such as cashew, squash, mango, cocoa, cranberries and blueberries were pollinated by a variety of bees, 19% by flies, 6.5% by bats, 5% by wasps and beetles, 4% by birds, butterflies and moths. Of the hundred principal crops that accounts for world's food supply, only 15 % was pollinated by domestic bees (mostly honeybees and bumble bees) and at least 80% are pollinated by wild non-apis pollinators (mostly leaf cutter bees, sweat bees, mason bees, flies, butterflies).

Insects as Soil Builders

Collembolans are the major group of microarthropods referred as 'key indicator species' of richness of soil biota as they feed on decaying organic substances, plant litters and that feed upon the soil microbiota viz., algae, bacteria and actinomycetes (Chen 1995). Deposition of frass during feeding on plants were reported to increase soil nitrogen and carbon pools with increasing the rate of soil respiration and leaching of nitric oxide that has direct and indirect effect on nutrient dynamics of the edaphic ecosystem (Frost and Hunter, 2004). Ground beetles belonging to the family Carabidae are important group of insects which are reported as predators as well as weed killers. The larvae of the ground beetles are voracious feeders of soil dwelling insects like wireworms, grubs, maggots resulting in curbing of the population of soil insects. Adult beetles were also reported to feed on the seeds of notorious weeds helping in regulation of weed population in crop fields (Liebman & Gallandt, 1997).

Insects as Scavengers

Insects play a vital role as scavengers by feeding in the dead matter helping in the breakdown of the decaying organic material and recycling them into the soil. The carrion feeding insects provide a natural sanitation to the environment through decomposition of dead organism's time to time. Blow flies (Calliphoridae) and bottle flies are the first to visit insects on any dead organisms. They oviposit in the decaying flesh and the hatching larvae feed upon the decaying matter. The occurrence of flies in the dead matter is of immense importance in

forensic studies that helps in investigations to figure out the time of death of the organism. In addition to flies, social insects like termites decomposes decaying plant material in soil during their nest construction process. The decomposed plant material is used as food material inside their nests. Hydrophilids commonly referred as 'water scavenger beetles' play a vital role in feeding and recycling of larvae /maggots of insects and snails in water bodies (Inoda *et al.*, 2014). They are active feeders of mosquito larvae that directly beneficial to contain the immature stages of the vector. This important ecosystem service rendered by different insects like flies, beetles in the form of scavengers form a component of food web.

Biocontrol – Predators and Parasitoids

Natural enemies viz., predators and parasitoids provide a very vital pest regulatory service effecting natural reduction of harmful insects through natural predation and parasitization. The natural enemies keep the insect pest population under the economical threshold levels and prevent them from flaring up causing the total crop destruction. The concept of natural biological control holds good in case of invasives where a natural enemy (parasitoid/predator) specific to the invasive pest would occur to contain it.

Factors Threatening the Insect Biodiversity

Habitat destruction

Destruction of natural breeding grounds or dwelling habitats of insects is a major cause for loss of biodiversity of insects as it negatively impacts their abundance and distribution in the natural ecosystem (Krueß & Tschardt, 2000). Among the different group of insects, butterflies belonging to the order Lepidoptera were reported to be worst affected through habitat destruction by anthropogenic factors (Warren & Dennis, 2004). The butterflies need floral rich habitats to meet the nectar requirement of the adults with sufficient host plants to complete their larval stage in their natural breeding places (Thomas *et al.*, 2001).

Deforestation

Forest ecosystem serves as undisturbed buffer zone for insects where anthropogenic interventions to threat insect diversity is at minimal levels. Deforestation results in loss of habitats for a wide group insect negatively impacting their diversity and abundance in the natural ecosystems (Chilima *et al.*, 2016). The extinction of forest dwelling dung beetles was correlated with the

deforestation in Madagascar (Hanski *et al.*, 2007). The plant-pollinator community interactions were reported to be affected through deforestation that challenges the reproductive success of many plants in the forest ecosystem (McCain, 2005).

Monocropping

Monocropping is a practice of cultivating single crop over a larger area. The practice of monocropping narrows down the food choice for the insects which has an innate preference to select and forage upon the plants/flowers. The survival and multiplication of bee pollinators largely on polyfloral diet rather than monofloral diet as the collected pollen influence the brood development in the colonies. Monocrop also provides nutrient inferior pollen that negatively impacts the fitness of the pollinators.

Climate Change

Predictions reveal an increase in precipitation across the globe owing to climate change that might cause change in cropping pattern which will affect the species composition of insect herbivores in different crops. Change in day length might alter the overwintering phase of insects (Dhillon *et al.*, 2017). The more predicted impacts of climate change on insect herbivores were i) faster rates of development ii) increased risk of invasive insects iii) increased number of generations of insect iv) extended geographical range of insect pests v) increase in overwintering stage of insects (Porter *et al.*, 1991). Studies conducted across the globe indicated that there occurs a temporal mis-synchrony between the flowering of plants and pollinators that negatively impacts their abundance and diversity across the ecosystem.

Urbanization

Urbanization is a phenomenon that affects the diversity and richness of insect population in long term. The ecological traits of insects and plants are directly impacted by the process in terms of shift in niches (Kamdem *et al.*, 2012), phenology changes in crop that affects the visitation of insects (Diamond *et al.*, 2014). With urbanization and rise in environmental pollution, drastic changes in insect behavior were predicted over a period of time that might impact the plant-insect interactions at the trophic levels. The insects were also reported to extend their geographical range of distribution and possibly invade newer areas.

Land Fragmentation

Land fragmentation is a rising concern in the current era owing to the urbanization factors. Land fragmentation is a factor that may threaten the insect diversity through reduction in the area that cause a habitat loss to the insects, edge effect that impact the insect species distribution and richness in the ecosystem. The increased distance between the fragmented habitats demand an additional energy toll for the bees to access the floral resources that might hinder the movement of bees to the detached habitats in a long run.

Measures to Boost up Insect Diversity

Creation of Crop Diversity and Semi-natural Habitats

The role of creation of semi-natural habitats to support the diverse arthropods like parasitoids, predators (Thomas *et al.*, 1997) and pollinators (Lagerlof *et al.*, 1992; Meek *et al.*, 2002) was reported as a vital component of agricultural landscaping in the recent times. The easiest way to attract native pollinators is to plant gardens or meadows that contain a diversity of native wildflowers, trees, and shrubs. A variety of wildflowers and native grasses will provide native pollinators with food (nectar, pollen, and/or larval host plants). Trees and dense shrubbery provide important shelter, nesting, and overwintering areas for beneficial insects. To maximize food and shelter, landowners can include gardens, fruit-bearing trees and shrubs, thickets and hedgerows of flowering shrubs, and set-asides (areas that are not mowed) in their plans. Due to differing preferences among pollinator species, planted areas should contain varying levels of vegetation and areas of sun, partial shade, and full shade.

Plants native to the region should be selected. Native plants are adapted to the local climate, soils, and the native pollinators with which they co-evolved. Native plants should comprise at least 75 percent of a habitat area. Invasive species should not be planted because they will degrade pollinator and other wildlife habitat by interfering with the natural structure and composition of the ecosystem.

Mowed lawn area should be minimized in favor of patches of native wildflowers, shrubs, and grasses. Lawn areas that do exist should be mowed less frequently to allow the vegetation to provide habitat for pollinators.

Perennials can be chosen over annuals. Perennials are generally richer in nectar and, because they bloom year after year, provide a more dependable food source than annuals. Both nectar and pollen flowers should be available throughout the growing season.

In high value perennial crops, narrow bands of intercropped resource-rich plants can be planted within fields to create permanent patches for beneficial insects. This will not only increase the stability of insect populations in the field, it will provide resources for a large number of other beneficial insects. These crops can be chosen with other farm goals in mind also, for example, to improve soil quality.

Creation of Habitats for Breeding of Beneficials

Beetle Banks

Ground beetles are soil inhabiting generalist predators that plays a major role in reduction of harmful insect pests. Beetle banks includes the concept of raising field strips with typically with grasses running through the middle of the field that serves as refugia for the overwintering stages of predatory beetles like carabids, coccinellids that can shift to the main crop during appropriate stages of insect pest infestation (Woodcock *et al.*, 2005). Such habitats also serve as hiding spots for the beneficials protecting them from pesticidal sprays in the main crop. The increased abundance and activity of ground beetles and rove beetles in the beetle banks during the winter period which later shifts to the cultivated crop causing natural suppression in farmlands (Varchola & Dunn, 1999). Carabids get an additional advantage of protection in the field strips during the agricultural field preparations like tillage, weeding etc.

Butterfly Gardens

Butterflies are important group of model insects that indicates healthiness of an environment. 'Butter fly garden' concept is a classical approach to conserve butterflies and maintain their diversity. The butter fly garden should contain nectar rich flowering plants for their survival. The larvae of butterflies require suitable host plants for feeding and multiplication. Identification of host plants preferred by butterflies for oviposition is crucial to integrate such host plants in the gardens for their proliferation.

Conclusion

Growers can adopt some relatively simple practices in and

around their fields to enhance farm suitability for these important beneficial insects. Enhancing the suitability of farm landscapes for native pollinators will also provide a diversified strategy for achieving good crop yields in pollination-dependent crops year after year.

References

- Kamdem C, B Tene Fossog, F Simard, J Etouana, C Ndo, P Kengne, P Boussès, FX Etoa, PAwono-Ambene, D Fontenille, *et al.* (2012) Anthropogenic habitat disturbance and ecological divergence between incipient species of the malaria mosquito *Anopheles gambiae*. *PLoS One*, 7: Article e39453.
- Chen B (1995) Food preference and effects of food type on the life history of some soil Collembola. *Pedobiologia* 39: 496–505.
- Chilima C, VM Bulambo and S Chiotha (2016) The impact of deforestation and forest conversion on abundance and diversity of insects and plants in a miombo forest *Afr. J. Ecol.* <https://doi.org/10.1111/aje.12291>
- Dhillon MK and F Hasan (2017) Temperature-dependent development of diapausing larvae of *Chilo partellus* (Swinhoe) (Lepidoptera: Crambidae). *J. Therm. Biol.* 69: 213–220. doi:10.1016/j.jtherbio.2017.07.016
- Diamond, SE, H Cayton, T Wepprich, CN Jenkins, RR Dunn, NM Haddad and L Ries (2014) Unexpected phenological responses of butterflies to the interaction of urbanization and geographic temperature. *Ecology* <https://doi.org/10.1890/13-1848.1>
- Frost CJ and MD Hunter (2004) Insect canopy herbivory and frass deposition affect soil nutrient dynamics and export in oak mesocosms. *Ecology* 85(12): 3335–3347. DOI:10.1890/04-0003
- Hanski Koivulehto H, A Cameron, P Pierre Rahagalala (2007) Deforestation and apparent extinctions of endemic forest beetles in Madagascar. *Pop. Ecol.* <https://doi.org/10.1098/rsbl.2007.0043>.
- Heath ACG (2012) Beneficial aspects of blowflies (Diptera: Calliphoridae). *New Zealand Entomol.* 7(3): doi.org/10.1080/0/00779962.1982.9722422
- Inoda T, Y Indoa and JKH Rulan (2014) Larvae of the water scavenger beetle, *Hydrophilus acuminatus* (Coleoptera: Hydrophilidae) are specialist predators of snails. *European J. Entomol.* 112(1): DOI: 10.14411/eje.2015.016.
- Kruess A. and T Scharntke (2000) Effects of Habitat Fragmentation on Plant-Insect Communities. In: Ekbom B, Irwin ME, Robert Y (eds) Interchanges of Insects between Agricultural and Surrounding Landscapes. Springer, Dordrecht. https://doi.org/10.1007/978-94-017-1913-1_4
- Lagerlöf, J., JD Stark, and B Svensson (1992) Margins of agricultural fields as habitats for pollinating insects. *Agric. Ecosyst. Environ.* 40: 117–224
- Liebman M and ER Gallandt (1997) Many little hammers: Ecological approaches for management of crop-weed interactions. In *Ecology in Agriculture*, LE Jackson (ed) San Diego: Academic Press.

- Mccain CD (2005) Deforestation affects the web of plant-insect interactions affecting reproductive success in the Amazonian herb, *Rapatea ulei*. Dissertation pp 231.
- Meek B., D Loxton, T Sparks, R Pywell, H Pickett and M Nowakowski (2002) The effect of arable field margin composition on invertebrate biodiversity. *Biol. Conserv.* **106**: 259–271.
- Pollard E (1991) Monitoring butterfly numbers, pp. 87–111. In: Goldsmith, FB (ed.). *Monitoring for Conservation and Ecology*. Chapman and Hall, London, 275pp.
- Porter JH, ML Parry and TR Carter (1991) The potential effects of climatic change on agricultural insect pests. *Agric. For. Meteorol.* **57**: 221–240. doi:10.1016/0168-1923(91)90088-8
- Slade EM, T Roslin, M Santalahti and T Bell (2016) Disentangling the ‘brown world’ faecal-detritus interaction web: dung beetle effects on soil microbial properties *Oikos* **125**: 629–635.
- Thomas JA, DJ Simcox, JC Wardlaw, WG Elmes, ME Hochberg and RT Clark (1998) Effects of latitude, altitude and climate on the habitat and conservation of the endangered butterfly *Maculinea arion* and its *Myrmica* ant host. *J. Insect Conserv.* **2**: 39–46.
- Thomas CFG, F Green and EJP Marshall (1997) Distribution, dispersal and population size of the ground beetles, *Pterostichus melanarius* (Illiger) and *Harpalus rufipes* (Degeer) (Coleoptera, Carabidae), in field margin habitats. *Biol. Agric. Hortic.* **15**: 337–352.
- Varchola JM and JP Dunn (1999) Changes in ground beetle (Coleoptera: Carabidae) assemblages in farming systems bordered by complex or simple roadside vegetation. *Agric. Ecosyst. Environ.* **73**(1): 41–49.
- Woodcock BA, DB Westbury, SG Potts, SJ Harris and VK Brown (2005) Establishing field margins to promote beetle conservation in arable farms. *Agric. Ecosyst. Environ.* **107**: 255–266.

Insect Collections as a Reservoir of Insect Genetic Resources

Ankita Gupta

ICAR-National Bureau of Agricultural Insect Resources, Bellary Road, Bengaluru-560024, Karnataka, India

Insect museums across the globe are gaining immense attention post issues of biodiversity loss estimation. The biggest question during the present times is the accurate assessment of insect species diversity and biodiversity accountability in terms of what amount of insect biodiversity is lost due to anthropogenic factors. However, many of the countries still do not possess a precise and standard dataset of their insect biodiversity and thus it is not possible to assess the loss holistically. Here comes the inevitable role of insect museums in documentation of the species present in their collections and furthermore translation of their dataset into extant and extinct species. This paper presents the role of insect collections and their databases in Insect Genetic Resources.

Introduction

Often the concept of Insect Genetic Resources is considered to be confined to the live forms present in the fields or the ones amenable to mass culturing in the laboratories. Since decades, the insect museums across the globe have contributed immensely in unravelling and documenting the mystery of insect species diversity. These collections lay a strong foundation for research scientists, quarantine personnel, taxonomists, farmers, and students as based on the dataset of these, precise and timely identification of pests and their natural enemies can be undertaken. They are also authoritative resources for systematics, evolutionary biology, ecology, natural resource management, biosecurity and biogeography. Mostly all renowned museums offer identification services as it serves as the first line of defence in protecting their respective biological resources and environment. The species identification is key to understanding the native biodiversity and threats that may destabilize it. In this article, a brief overview of various online insect sources is mentioned which serve as a reservoir of knowledge for insect genetic resources in multiple forms.

Insect Databases as Identification and Information Tools

A Glimpse of a Few Global Insect Collections and their Databases

Presently the United States Department of Agriculture-APHIS-Identification Technology Program-ITP is one of the most successful and reliable source for insect identification in the United States. Under ITP, three

means are in use with the aim of sustainable agriculture: ID tool websites, mobile apps, and CAPS screening aids (USDA, 2022) (<https://idtools.org/identify.php>). Likewise, the renowned Natural History Museum, London has some of the most unique insect specimen databases besides having the information rich database like the Universal Chalcidoidea Database (Noyes, 2019). The digitized insect records of Australian National Insect Collection (ANIC), Australia are served through Online Zoological Collections of Australian Museums, the Atlas of Living Australia, and the Global Biodiversity Information Facility. The U.S. National Entomological Collection (USNM), one of the world's largest and most important accessible entomological collections with over 33 million specimens, which is collectively maintained by: Smithsonian Institution; Systematic Entomology Laboratory (Agricultural Research Service, United States Department of Agriculture); and Walter Reed Biosystematics Unit (Walter Reed Army Institute of Research), has online portal for its primary types, specimens and species.

Indian Insect Museums and their Databases

Even though India comes under 17 mega-diverse countries of the world, our insect collections and museum digitization are still not at par with the insect museums of developed countries and needs lots of attention on priority basis. In India, the premier and historic insect collections are housed at the Zoological Survey of India- Kolkata, the National Pusa Collection, ICAR-IARI-New Delhi, the National Insect Museum of ICAR- NBAIR-Bengaluru, Forest Research Institute, Dehradun, etc. The species

*Author for Correspondence: Email-Ankita.Gupta@icar.gov.in

rich and neatly illustrated exhaustive insect databases of ICAR-NBAIR serve as major identification aid and taxonomic source, which is globally available, for many agriculturally important and diversity rich speciose taxa <https://www.nbair.res.in/databases> (NBAIR, 2022). The National PUSA Collection, IARI-New Delhi has also initiated its online insect database for selected taxa. Few of the insect repositories in India have been nominated as designated repositories by the Ministry of Environment, Forest and Climate Change which amass vouchers of National Biodiversity Authority of India.

Citizen Science Concept in Insect Diversity Documentation

Some of the most voluminous Indian insect databases are with the involvement of citizen science thus making the conservation of insect biodiversity more appealing and approachable. 'Butterflies of India' by Kunte *et al.*, 2022- <https://www.ifoundbutterflies.org> and 'Moths of India' <https://www.mothsofindia.org> by Sondhi *et al.*, 2022 are some of the successful examples of involvement of 'citizen science concept' in documentation and conservation of insect diversity.

Insect DNA Database

Cockburn detailed the need and referenced majority of the insect related databases way back in 1998. With the present era of DNA barcoding, all the insect museums are presently aiming for establishing their DNA library for the species housed in their collections. Many insect museums are already sharing their data with some of the upcoming reliable DNA database like Consortium

for the Barcode of Life (CBOL). However, the NCBI Taxonomy Browser, still remains as one of the most exhaustive, dependable and expanding resources for insect germplasm.

Conclusion

The need for authentically identified and scientifically well preserved and catalogued insect collections is gaining momentum as the world is realising the need for insect conservation and biodiversity assessment. These collections should be looked upon as one of the national assets rather than considering them as a liability and a resource sink.

Acknowledgement

The author is grateful to the Director NBAIR Dr M Nagesh for research encouragement and support.

References

- Cockburn AF (1998) Insect Germplasm and Genome Databases. *American Entomologist* 16-19.
- Kunte K, S Sondhi, and P Roy (Chief Editors) *Butterflies of India*, v. 3.06. Indian Foundation for Butterflies. <https://www.ifoundbutterflies.org>
- NBAIR (2022) Databases. <https://www.nbair.res.in/databases>
- Noyes JS (2019) Universal Chalcidoidea Database. World Wide Web electronic publication. <http://www.nhm.ac.uk/chalcidoids>
- Sondhi S, Y Sondhi, P Roy and K Kunte (Chief Editors) (2022) *Moths of India*, v. 3.30. Indian Foundation for Butterflies. <https://www.mothsofindia.org>
- USDA (2022) Identification Technology Program. <https://idtools.org/identify.php>

Arthropod Germplasm Information System (AGIS) in India

M Pratheepa*, Richa Varshney and R Gandhi Gracy

ICAR-National Bureau of Agricultural Insect Resources, Bellary Road, Hebbal, Bengaluru-560024, Karnataka, India

Arthropods are significant components of diverse ecosystems and are the major players in functioning of ecosystem processes. They must be preserved because of their natural values but also because we require them for human survival. Thus, arthropods must become a main and essential part of the conservation strategy. Insect conservation aims at for saving both endangered species and ecosystem processes with a multitude of approaches. Germplasm is a term used to describe a collection of genetic resources for an organism. Arthropod germplasm is essential in agriculture. Accessibility to the germplasm information like location of availability and characteristics of the arthropod organisms is necessary for the researchers in the form of web based database. Hence, Arthropod Germplasm Information System (AGIS) – an online database has been developed in the country during 2014, for the storage of information about the live arthropod genetic resources maintained at different institutions in India. The exact aim of this AGIS database is to develop, maintain and exchange arthropod germplasm information. AGIS would furnish to the needs of the entomologists especially who working on agriculturally important insects. The website address for this resource is <https://databases.nbair.res.in/germplasm/index.php> and institutions can register their live arthropod germplasm accessions by accessing this link.

Key Words: Agriculture, Arthropod germplasm, Germplasm database, Insect genetic resources, Passport details

Introduction

Arthropods have to be preserved because we require them for human survival. Therefore, arthropods are essential part of the conservation strategy. Insect conservation aims at for saving both endangered species and ecosystem processes with a multitude of approaches (Kim, 1993). Germplasm is a term used to describe a collection of genetic resources for an organism. Arthropod germplasm is essential in agriculture. In India, ICAR-National Bureau of Agricultural Insect Resources (NBAIR), Bengaluru acts as a nodal organization for collection, characterization, documentation and conservation of insects and related arthropods for commercial and research purposes. NBAIR being the national repository for live arthropod germplasm collection and maintenance, the present database aims to promote the arthropod genetic resources of various categories like host insects, parasitoids, predators, detritivores insects, silkworm resources and veterinary pests, mites and spiders. Accessibility to the germplasm information like location of availability and characteristics of these arthropod organisms is necessary for the researchers in the form of web based database. Keeping these points in view, Arthropod Germplasm Information System

(AGIS)-an online software tool has been developed in the country during 2014, for the storage of information about the live arthropod genetic resources maintained at different institutions in India. The arthropod germplasm collections are unique because scientists maintain them by continuously rearing live specimens and they serve as genetic resources. The website address for this resource is <https://databases.nbair.res.in/germplasm/index.php>. The home page of the AGIS is depicted in Fig 1.

AGIS Key Elements

The AGIS contains the key elements such as Home, Germplasm Information, Germplasm Registration, Downloads, Contact Us, Admin Login and User Login. Germplasm Information option provide the passport information of the arthropod genetic resources submitted by different institutions in India. Presently, passport information is available in AGIS for silkworm genetic resources maintained at Central Sericultural Germplasm Resources Centre (CSGRC), Hosur, Tamil Nadu, Central Silk Technological Research Institute (CSTRI), Berhampore, West Bengal and Mysore, Karnataka; Central Tasar Research and Training Institute, Ranchi, Jharkhand. Apart from silkworm, AGIS contains

*Author for Correspondence: Email-Pratheepa.M@icar.gov.in



Fig. 1. Home page of Arthropod Germplasm Information System

information on veterinary pests maintained in the entomology laboratory of ICAR-Indian Veterinary Research Institute (IVRI), Bareilly, UP and host insects, parasitoids, predators and detritivores insects, mites and spiders maintained at ICAR-National Bureau of Agricultural Insect Resources (NBAIR), Bengaluru through this online database tool. An Institute Accession Number and a National Accession Number are the unique identification numbers for each germplasm registered. The passport information for the silkworm germplasm includes voltinism, race name, donor, origin, class, parentage, egg color, yolk color, larval pattern, eye spot, crescent, star (Faint/Present/Absent), cocoon color and cocoon shape. Passport information for veterinary pests includes scientific name of the veterinary pest, systematic position, origin, common name of the veterinary pest, common name of the host animals and locality details. The passport information for the predatory insects maintained at ICAR-NBAIR includes systematic position, target pest details, host plant, locality details and their's utility for biocontrol. The passport information for parasitoids includes systematic position, target pest details, host plant, locality details and information on how they could be utilised for biological control of pests. The passport information of host insects includes systematic position, common name of the host insect, the stage of the insect that could be supplied, locality details, etc. The passport information of detritivores insect includes systematic position, etc. One can register their live

arthropod germplasm accessions by accessing the link <https://databases.nbair.res.in/germplasm/index.php>.

Conclusion

The exact aim of this AGIS database is to develop, maintain and exchange arthropod germplasm information. The AGIS gives provision for multi-user accessibility based on Client-Server technology with Add/update options and the server is located at ICAR-NBAIR, Bengaluru, India. The information on different groups of arthropod germplasm like Silkworm, Veterinary Pests, Host Insects, Predators, Parasitoids, Detritivores insects are being regularly updated into the online database AGIS by the entomologists from various institutions in India. Researchers from all over the world can browse into the website <https://databases.nbair.res.in/germplasm/index.php> and obtain information on the live arthropod germplasm accessions available in India. The AGIS – online tool is a ready reference for researchers and can also obtain the passport information on the arthropod germplasm culture maintained at different organisations.

Acknowledgement

Authors sincerely thank the Director, ICAR-NBAIR, Bengaluru India for providing the infrastructure facilities and constant encouragement.

Reference

Kim KC (1993) Biodiversity, conservation and inventory: why insects matter. *Biodiversity and Conservation* 2(3): pp. 191-214.

GUIDELINES TO AUTHORS

GENERAL

Indian Journal of Plant Genetic Resources (IJPGR) is the official publication of the Indian Society of Plant Genetic Resources. Aim of the Journal is to disseminate knowledge on plant genetic resources (PGR) research and application. Being the only journal in the area of PGR, the journal aims to provide a forum for discussion and debate on current issues of PGR. For publication in the journal, the authors must be a member of the Society. IJPGR publishes full-length papers or short communications of original scientific research in the field of plant genetic resources. Review articles (with prior consent or invitation only) summarizing the existing state of knowledge in topics related to plant genetic resources will also be published.

Contributions should be as concise as possible. The maximum length of the review article, full-length papers and short communications is usually restricted to 12, 6, 3 printed pages including illustrations and tables, respectively.

SCOPE

- Basic research on biosystematics, genetics and genomics related to PGR
- Applied research on field and laboratory evaluation of PGR
- Supportive research in conservation and quarantine of PGR
- Policy research on access and benefit sharing; IPRs
- Status reports on ecology, conservation, traditional knowledge and use of PGR

EDITORIAL BOARD

Editor-in-Chief:

Sunil ARCHAK

Indian Journal of Plant Genetic Resources
H-204, ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110012, India
editor.pgr@gmail.com

Foreign Editors:

Michael HALEWOOD

Head of Policy Unit
Bioversity International
Rome, Italy
m.halewood@cgiar.org

Adriana ALERCIA

Germplasm Documentation Specialist
ITPGRFA, FAO
Rome, Italy
adriana.alercia@fao.org

Ronnie VERNOOY

Genetic Resources Policy Specialist
Bioversity International
Rome, Italy
r.vernooy@cgiar.org

Ehsan DULLOO

Team Leader, Integrated Conservation Strategies
Bioversity International
Mauritius
e.dulloo.cgiar.org

Indian Editors:

Anjula PANDEY

Principal Scientist
Division of Plant Exploration and Germplasm Collection
ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
anjula.Pandey@icar.gov.in

Ruchira PANDEY

Principal Scientist (Retired)
Tissue Culture and Cryopreservation
Unit National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
ruchira.Pandey@icar.gov.in

K. Joseph JOHN

Principal Scientist
ICAR-NBPGR Regional Station – Thrissur
Thrissur-680 656, Kerala
joseph.k@icar.gov.in; nbpgrtsr@gmail.com

V. Celia CHALAM

Principal Scientist
Division of Plant Quarantine
National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
celia.chalam@icar.gov.in

N. SIVARAJ

Principal Scientist
ICAR-NBPGR Regional Station – Hyderabad
ARI Campus, Rajendra Nagar
Hyderabad-500 030, Andhra Pradesh
n.sivaraj@icar.gov.in

R.K. SALGOTRA

Professor & Coordinator
School of Biotechnology
Sher-e-Kashmir University of Agricultural Sciences &
Technology of Jammu-180 009
rks_2959@rediffmail.com

Rakesh SINGH

Principal Scientist
Division of Genomic Resources
National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
rakesh.singh2@icar.gov.in

Lalit ARYA

Principal Scientist
Division of Genomic Resources
ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
lalit.arya@icar.gov.in

K. PRADHEEP

Principal Scientist
ICAR-NBPGR Regional Station - Thrissur, Vellanikkara,
KAU P.O. Thrissur-680656, Kerala
k.pradheep@icar.gov.in

Pitchaimuthu M.

Principal Scientist
Division of Vegetable Crops
ICAR-Indian Institute of Horticultural Research
Hessaraghatta Lake Post, Bengaluru-560 089
m.pitchaimuthu@icar.gov.in

Aditya PRATAP

Principal Scientist
ICAR-Indian Institute of Pulses Research
Kanpur-208 024, Uttar Pradesh
Aditya.Pratap@icar.gov.in

Vandana TYAGI

Principal Scientist
Germplasm Exchange Unit
ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
vandana.tyagi@icar.gov.in

Kavita GUPTA

Principal Scientist
Division of Plant Quarantine
National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
kavita.Gupta@icar.gov.in

Sheikh M. SULTAN

Principal Scientist & Officer-In-Charge
ICAR-NBPGR, Regional Station
Srinagar-190 005
Jammu & Kashmir
mohmmad.sheikh@icar.gov.in

Manjusha VERMA

Principal Scientist
Division of Genomic Resources
ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
manjusha.verma@icar.gov.in

Amit SINGH

Senior Scientist
Division of Genomic Resources
ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110 012
Amit.singh5@icar.gov.in

Sherry Rachel JACOB

Senior Scientist
 Division of Germplasm Conservation
 ICAR-National Bureau of Plant Genetic Resources
 Pusa Campus, New Delhi-110 012
sherry.jacob@icar.gov.in

S. RAJKUMAR

Senior Scientist
 Division of Genomic Resources
 ICAR-National Bureau of Plant Genetic Resources
 Pusa Campus, New Delhi-110 012
S.Rajkumar@icar.gov.in

C.S. MOHANTY

Principal Scientist
 CSIR-National Botanical Research Institute
 436, Rana Pratap Marg, Prem Nagar, Hazratganj
 Lucknow-226 001, Uttar Pradesh
cs.mohanti@nbri.res.in

Kamala VENKATESWARAN

Principal Scientist
 ICAR-NBPGR Regional Station – Hyderabad
 ARI Campus, Rajendra Nagar
 Hyderabad-500030, Andhra Pradesh
kamala.Venkateswaran@icar.gov.in

ORGANIZATION OF THE MANUSCRIPT**Full-length papers**

Title Page: The title page of the manuscript should be the first page and should include the title, names and addresses of the authors, abstract and keywords.

Title: Keep the title brief, specific and informative and amendable to indexing. It should be typed in running text with first letter of word as capital and latin names in italics.

Name and Address: The name of authors and the address of the institution where the work was carried out should be mentioned below the title. Present address of correspondence, if different, should be given as footnote indicating by asterisk (*) the author to whom the correspondence and reprint requests are to be made. E-mail addresses should also be indicated, if any.

Abstract: The abstract should clearly state the rationale, objectives, methods, and important conclusions of the study. It should not exceed 150 words.

Key Words: The abstract should be followed by not more than five key words indicating the contents of the paper and useful for abstracting purposes.

Main Text: The main text of the paper should start from the second page which should contain the title of the paper followed by the text divided into following main headings which are to be typed in running text and flushed with the margin: Introduction, Materials and Methods, Results, Discussion, Acknowledgements, References. Wherever appropriate, results and discussion can be combined and acknowledgements be omitted.

Introduction should be brief and limited to the statement of the problem and aim of the experiment.

Materials and Methods should include relevant details on the nature of material, experimental design, the techniques employed and the statistical method used. For well-known methods, citation of reference will suffice.

Results and Discussion should be clear to readers in different disciplines. Units of measurement should be SI.

Tables should be typed on separate sheets, each with a heading stating its contents clearly and concisely. Numerical data and calculations should be thoroughly checked.

Figures of only good quality that are essential to a clear understanding of the paper shall be accepted. Legends to the illustrations should be typed on separate paper. Information in the legend should not be repeated in the text and similarly, the same data should not be represented in both graph and table form. All figures, whether photographs, maps, graphs or line drawings should be numbered consecutively. Illustration number and title of the article with authors' name should be given at the back of the plates in soft pencil.

Line drawings of high quality, preferable in the desired final size would be accepted. The inscriptions should be clearly legible.

Photographs for publication should be of high contrast, black and white, glossy print, trimmed at right angles. Magnification should be indicated with a bar scale on the photo. Authors need to indicate colour reproduction of photographs (cost of colour printing will be borne by the authors).

Acknowledgements should mention only guidance or assistance received in real terms, and financial grant provided by an agency. Acknowledgements for inspiration, typing etc., need not be mentioned.

References in text should be cited by author, year of publication (e.g. Joshi, 1995) and multiple citations should be in chronological order (Withers and Englemann, 1998; Rao et al., 2001). References should be listed in alphabetical order under the first authors' name. The names of journals should be abbreviated according to the latest edition of the World List of Scientific Periodicals (eds P Brown and GB Stratton), Butterworths, London.

The following examples may be used for citations:

Bisht IS, RK Mahajan, TR Loknathan and RC Agarwal (1998) Diversity in Indian sesame collection and stratification of germplasm accessions in different diversity groups. *Genet. Resour. Crop Evol.* **45**: 325-335.

Withers LA and F Englemann (1998) In vitro conservation of plant genetic resources. In: A Altman (ed.) *Agricultural Biotechnology*. Marcell Dekker Inc., New York, pp 57-58.

WOI (1985) *The Wealth of India - Raw Materials. A Dictionary of Indian Raw Materials and Industrial Products – Raw Material Vol 1: A* (Revised). Publications and Information Directorate, Council of Scientific and Industrial Research, New Delhi, 513 p.

Engels, JMM and V Ramanatha Rao (eds) (1988) *Regeneration of seed crop and their wild relatives*. Proceedings of a Consultation Meeting, 4-7 December 1995. ICRISAT, Hyderabad, India and IPGRI, Rome, Italy, 167 p.

Short Communications

The style and format as mentioned for full-length papers should be followed for Short Communications. However, the abstract should be restricted to not more than 50 words and the remainder text should be continuous (without headings). Illustrative material should be kept at minimum, usually not more than one table or figure and only few references should be included (not more than 10). Authors can also submit meeting reports after consulting EIC.

Submission of the Manuscript Submission of an article will be held to imply that it has not been previously published or submitted for publication elsewhere. A cover letter including a statement to this effect should be submitted with the manuscript. Article are submitted through online submission system (http://www.indianjournals.com/ijor.aspx?target=manuscript_submission) or by email to:

Editor-in-Chief, Indian Journal of Plant Genetic Resources

E-mail: ispgr2015@gmail.com

- Experts in the subject will review all the submitted manuscripts and the final decision about the acceptance of the manuscript rests with the Editorial Board. If manuscript is accepted for publication, the revised manuscript should be accompanied by electronic copy on CD or through electronic mail.
- Publication of a paper in the Journal does not imply the responsibility for an agreement with the statements or view written therein, and rests entirely on the authors thereof.
- The authors will receive page proofs, which should be corrected and returned without delay. Corrections must be kept to the minimum, and the proof stage should not be regarded as an opportunity for further editing and additions. Although, every effort is made by the editors to correct proofs of all the papers they assume no responsibility for errors that may remain in the final print.

ETHICS STATEMENT

IJPGR approves the spirit of the guidelines for journal editors developed by the Committee on Publication Ethics (COPE).

1. Manuscripts submitted to IJPGR are evaluated entirely on the basis of their scientific content and relevance to PGR research community.
2. IJPGR does not levy any publication charges to members of ISPGR.

3. ISPGR, the publishers of the Journal, take all possible measures to uphold the highest standards of publication ethics and to prevent any malpractices.
4. Authors who submit manuscripts to IJPGR declare that the submissions are original and unpublished and are not under consideration for publication elsewhere.
5. Authors also declare that the manuscript is based on their own original work devoid of any nature and amount of plagiarism.
6. IJPGR makes full efforts to eliminate any kind of conflict of interest of authors, editors or reviewers, resulting from competitive, collaborative or other relationship with any person, institution or agency connected to the submissions.

Ethics expected to be followed by IJPGR Editors

1. IJPGR Editorial Team is fully responsible for the decision on acceptance or rejection of a manuscript submitted to the Journal. An established procedure is followed to ensure unbiased decision making. Submissions are handled by different editors based on their specialization. Editors obtain the reports of three referees with expertise in the topic dealt in the submission. Each editor recommends rejection or acceptance (direct or after revisions) to the Editor-in-Chief who takes the final decision.
2. The evaluation of manuscripts is made on the basis of their scholarly content in terms of relevance, novelty and scope as well as their presentation in terms of language and syntax. IJPGR does not consider any other factors like gender, race, religious belief, ethnic origin, citizenship, or political philosophy of the authors.
3. IJPGR strictly follows transparency in the evaluation process and confidentiality of the submission. Reviewers, as on today, remain anonymous to authors.

Ethics expected to be followed by IJPGR Reviewers

1. Peer review is the backbone of IJPGR manuscript flow. IJPGR depends upon the services of reviewers for maintaining ethical integrity and scholarly quality.
2. IJPGR does not follow double blind review and therefore the reviewers are expected to maintain absolute confidentiality with regard to the contents of manuscripts.
3. The reviews are conducted objectively with every comment/decision supported by clear reasons. Reviewers follow a non-ranking format provided by the IJPGR to facilitate decision making by Editor.

Ethics expected to be followed by Authors

1. Authorship should be restricted to only those persons who have made a significant contribution to the conception, design, execution, or interpretation of the reported study. In recent times, symbiotic authorship has become a serious ethical issue.
2. The authors must ensure that the experiment and manuscript are both entirely original works. Every instance of use of work and/or words of other authors must be appropriately cited or quoted. Any kind of plagiarism detected at review stage results in rejection of the manuscript. If the plagiarism is detected post-publication, the article will be summarily retracted. If any fundamental errors are found in published papers, authors will inform IJPGR which will take corrective measures including retraction.
3. IJPGR persuades authors to provide the basic data related to manuscripts such as description of germplasm and its availability to ensure greater utilization of diverse germplasm. Authors are also encouraged to provide relevant raw data for editorial review. Authors must prepare such data in a format to provide public access. Specific requests of data confidentiality shall be entertained by IJPGR on case to case basis.
4. Authors must avoid any kind of duplication in their submissions i.e. attempting to publish same/significantly similar manuscripts in more than one journal. IJPGR does not consider the following as a prior publication: (i) Abstract in a conference/symposium; (ii) Academic thesis/dissertation; (iii) Invited talks and interviews on specific topics.

5. Submission to IJPGR implies that authors have complied with ethical and technical standards of use of hazardous material, use of animal or human subjects, access and use of genetic resources, access and use of IP protected data. It is authors' duty to ensure no national or international legal requirement is compromised.
6. Submission of a manuscript to IJPGR implies that the corresponding author has ensured that all co-authors have seen and approved the final version of the paper and have agreed to its submission for publication to IJPGR. All sources of financial support should also be disclosed.

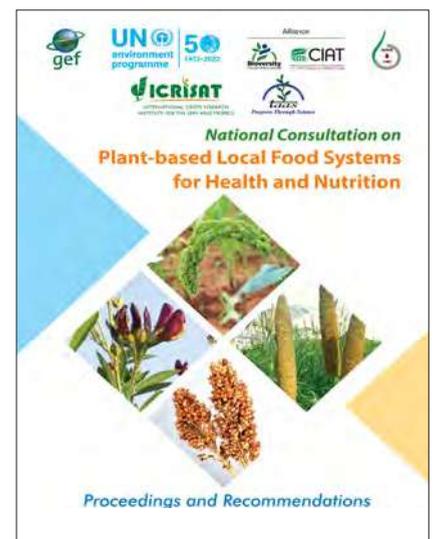
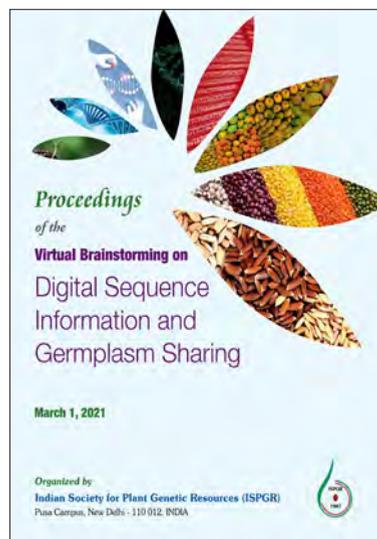
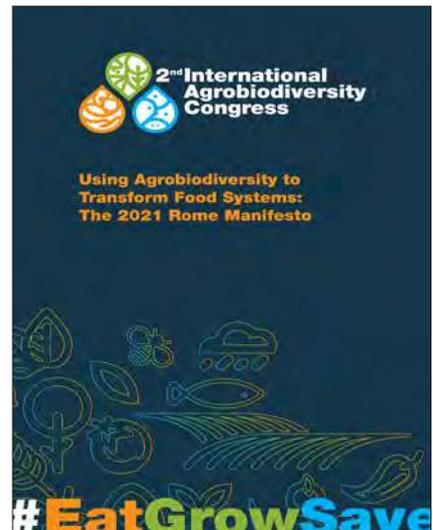
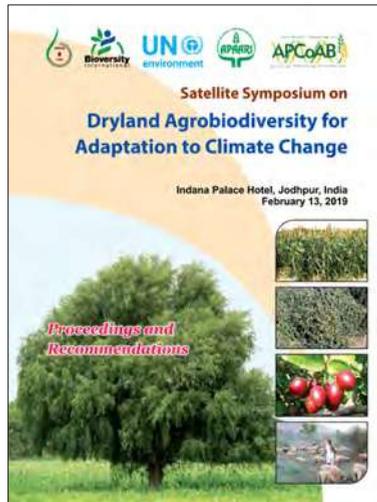
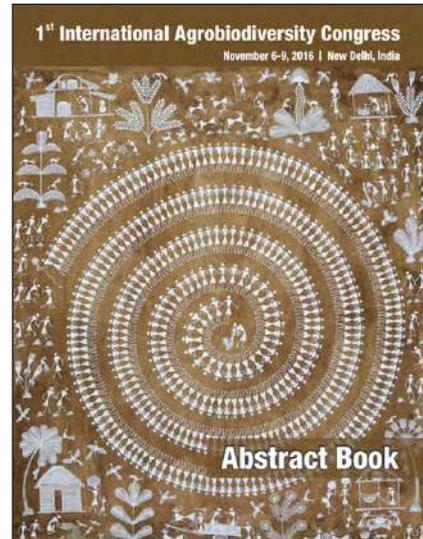
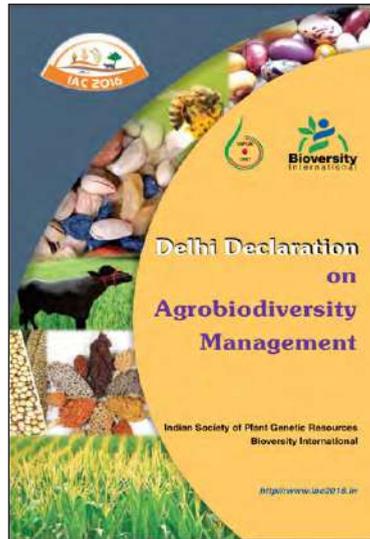
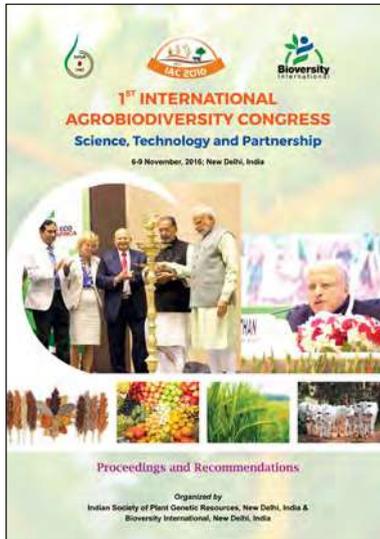
PUBLISHER/EDITORIAL OFFICE ADDRESS

H-204, ICAR-National Bureau of Plant Genetic Resources
Pusa Campus, New Delhi-110012, India

E-mail: *ispgr2015@gmail.com*

Publisher website: *www.nbpr.ernet.in/ispgr*

Journal hosting and online submission of manuscript: *www.indianjournals.com*





“Genetic resources need to be conserved through use and efficient scientific management for present requirements and posterity. Genetic resources must be accessible for research without hurdles to mitigate the impacts of climate change and to achieve food and environmental security”

– Dr RS Paroda