

Performance and Quality Management Experiences in the Indian National Genebank



S K. Sharma

Director

National Bureau of Plant Genetic Resources

New Delhi – 110 012, INDIA



Need for Conservation : Rapid Extinction of PGR

- **International**

- 50,000 to 2,50,000 of plants and 800 vertebrates will be lost by 2020 AD (Raven 1987)
- IUCN estimates loss of 25% of world species by 2050 AD

- **National**

- IUCN list 17 Indian species in “Extinct Plant Species of the World”
- Red Data Book of Botanical Survey of India list around 1500 plant species as endangered
- About 60 of the 250 wild crop relatives are either rare or under threat
- Until recently 30,000 rice varieties were grown in India, which are feared to reduce to 50 by 2000 (envfor.nic.in)



Role of National Genebank

- **Long-term conservation:** of germplasm for posterity following international standards
- **Information documentation:** Passport and management database provide full exposure about the variability available to user
- **Restoration of active collections:** Conservation of germplasm to support present use with restoration of lost germplasm and after recommended regeneration cycle to NAGS
- **Technical support to National Active Germplasm Sites** in the maintenance of facilities and national network
- **Germplasm distribution:** Supply germplasm with minimum or desired information in emergency to facilitate sustainable use in crop improvement
- **Registration of plant Germplasm:** Registration of plant germplasm with traits of potential value
- **Protecting farmers' and breeders' privileges:** with appropriate documentation of information



Conservation of PGR at the National Genebank

- ***Ex situ* plant conservation in Field Genebank**
- ***Ex situ* seed conservation in Seed Genebank**
- ***Ex situ-in vitro* conservation of explants or organs under aseptic conditions**
 - * ***In vitro* slow growth conservation of morphogenic cultures in *in vitro* Bank**
 - * ***In vitro* cryopreservation in Cyrobank, and**
 - * **DNA library conservation in DNA bank**



Seedbank Standards

- **Seed-moisture level : 3-7% for base collections (Long Term Storage)**
- **Seed-moisture level : 8-10% for active collections (Medium Term Storage)**
- **Accession size:**
 - 4000 seed in out-crossing species
 - 2000 in self-pollinated species
 - 500-1000 in difficult crops and wild species
 - quantity as much as possible to reduce regeneration load
- **Viability monitoring:**
 - Initial viability upon storage
 - 5 year period in active collections
 - 10 year interval in base collections
- **Storage containers: Temperature and moisture proof and sealable**
 - Tri-layered aluminium foil pouches
- **Regeneration: 100 or more plants for regeneration to avoid the loss of alleles.**



Seed Storage Conditions

Short-term storage:

- For seed processing
- Temperature 15 - 20°C and relative humidity 30 - 40%.

Medium-term storage:

- Temperature 0 - 10°C, and relative humidity 25- 30%.
- Adequate to maintain seed viability above 65% for 25-35 years
- suitable for active collections

Long-term storage:

- Acceptable: Sub-zero with seed moisture 3 -7%
- Preferred: -18°C or cooler with seed moisture 3 -7%
- Adequate to maintain seed viability for 50-100 years
- Suitable for base collections.
- Seeds kept in hermetically sealed containers with 4-5% seed moisture



Regeneration Standards – for Base collection

- Seeds for base collection are of the highest quality and pathogen-free.
- Regeneration should be undertaken when viability falls to 85% of initial value
- Seeds from original sample taken after 3 regenerations
- Proper regeneration protocols are followed for individual crops and species
 - Reproductive rates of individual genotypes
 - The breeding system
 - Seed dormancy
 - Variability in flowering time
 - Isolation of accessions from one another to protect against pollen migration and controlled hand-pollination
 - caging or larger separation distances
- 100 or more plants for regeneration to avoid large losses of alleles.
- Seeds used for regeneration should be as close as possible genetically to original sample.



Registration of Sample

- **Verification of identity of the material and assigning an identification number**
- **Physical verification of the seed material and selection of physiologically mature seed**
- **Genetic verification of seed material with assessment of seed morphology for uniformity, particularly in relation to quantitative traits, such as seed size (with variation within expected limits) and qualitative traits like seed coat colour etc.**
- **Removal of undesirable adulterants such as weed seeds, other crop seeds, inert matter, immature/broken/shriveled seeds, seeds damaged by insects and diseases etc.**
- **Record the data in the data file or database**



Standards for Information Maintenance

- **Passport**
- **Management**
- **Characterisation**
- **Evaluation**
- **Mode of reproduction**



Standards for Germplasm Exchange

- Seed be supplied in suitable containers
- Adequate information should accompany for effective use
- Seed lot should have high viability level
- Quarantine regulations are strictly followed
- Should accompany Material Transfer Agreement (MTA) as per national requirements



Standards for Maintenance of Modules

- A voltage stabiliser to protect from fluctuation of the incoming electric supply.
- A built-in electrical control panel providing complete operation information.
- Indicator lights to display operating condition.
- **Spares:** compressor unit, thermostat, fan motor, expansion valves, compressor contractor, overload relays, and fuses etc. should always be kept for immediate replacement



Standards for Safety and Security of Modules

- Ensured uninterrupted power supply
- Fire precautions
- Security
- Refrigeration standards and equipment used as per Design of Seed Storage Facilities for Genetic Conservation (DSSF)
- Construction and installation- following guidance given by DSSF
- Safety of personnel: Protective clothing etc.



Methods used for monitoring seed viability

Fixed sample size Germination test : This test is done using hundred seeds, each in two replications, following a suitable method of germination test (top of paper or between a paper). The germination proceeding is calculated on the basis of seed germinated. If the present germination is above 90, the test is accepted as valid. If it is below 90%, the test is repeated using another 200 seeds. The mean germination percentage is calculated, and if the germination percentage is below 85, the accessions are labelled for regeneration.

Sequential Germination Test: This method is used with accessions, with less quantity of seeds. In this method 40 seeds are used for germination test. The number of seed germinated are counted and the germination percentage is compared, with the number of seeds germinated to identify the accessions requiring regeneration.



Base Collections in National Genebank -20°C

(As on 30th September, 2007)

S.No.	Crop group	Present status
1.	Cereals	138725
2.	Millets and forages	49617
3.	Pseudo cereals	5886
4.	Legumes	54629
5.	Oilseeds	49187
6.	Fibre	9714
7.	Vegetables	22248
8.	Fruits	268
8.	Medicinal & Aromatic Plants and Narcotics	5704
9.	Spices	1995
10.	Agro-forestry	2330
11.	Duplicate Safety Samples	10235
	Total	350666*

- The figure includes 2838 Released varieties and 1454 Genetic stocks
- No. of crop species conserved-1289



Monitoring of Germplasm Conserved over Ten Years



Crop	Acc	Range of viability		Acceptable (%)	% for Regen.
		Initial	Present		
Wheat	103	85-100	85-100	100	Nil
Maize	90	90-100	88-100	100	Nil
Barley	195	80-100	80-100	100	Nil
Amaranth	60	90-100	85-100	100	Nil
Kodo millet	8	86-98	86-98	100	Nil
Finger millet	10	86-98	84-96	100	Nil
Sorghum	10	80-100	80-96	100	Nil
Pearl millet	10	85-100	90-100	100	Nil
Barnyard millet	9	90-100	88-100	100	Nil
Chickpea	125	85-100	88-100	100	Nil
Pigeonpea	95	88-100	85-100	100	Nil
Urd bean	15	80-90	85-90	100	Nil



Monitoring of Germplasm Conserved over Ten Years

Crop	Acc.	Range of viability		Acceptable (%)	% for Regen.
		Initial	Present		
Groundnut	53	90-100	90-100	100	Nil
Brassica	49	95-100	94-100	100	Nil
Sunflower	40	90-100	90-100	100	Nil
Linseed	50	95-100	95-100	100	Nil
Soybean	44	80-100	48-68	66	44l
Brinjal	52	70-80	70-80	100	Nil
Cabbage	45	80-70	80-70	100	Nil
Carrot	38	80-90	80-90	100	Nil
Raddish	92	70-80	70-80	100	Nil
Sowa	53	85-100	90-100	100	Nil
Jute	334	85-100	85-100	100	Nil
Kenaf	194	85-100	85-100	100	Nil
Roselle	51	85-100	85-100	100	Nil



Germplasm Conservation: Supportive Research

- Investigating seed storage behaviour
- Identification of factors contributing to seed deterioration in storage
- Investigating cost effective conventional seed storage methods (Ultra-desiccation)
- Dormancy breaking protocol
- Priming
- Zero energy based seed conservation

Monitoring of seed viability of the accessions kept for two years at Leh and Khardungla yielded positive results

- Establishment of a National Repository for safety duplicate in collaboration with DRDO



Thanks!