

Advanced Breeding Approaches for Small Millet Crops to Accelerate Improvements for a New Green Revolution

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ARTICLE ID: 04

Introduction

Small millets include a group of small-seeded cereal crops of the grass family Poaceae. This includes finger millet, foxtail millet, proso millet, barnyard millet, kodo millet, and little millet (see details of millet table-1). These crops once ruled as food and fodder but the area under millets cultivation has been drastically decreasing over the last six decades in India, especially since the green revolution. The biggest small millets-producing state is Karnataka (56% of total production) followed distantly by Tamil Nadu (14.0%) and Uttarakhand (9.3%). In terms of cultivated area, Karnataka has the highest proportion (48%) followed by Maharashtra (10.5%) (Hariprasanna, 2023).

Table 1:Small millet genomic resources and features

Details	Fingermillet	Littlemillet	Kodomillet	Barnyard millet	Foxtailmillet	Prosomillet
Common Name	Ragi,Mundua	Kutki,Savan	Kodon	Sanwa	Kangni,Kaku n	Cheena,Bari
Scientific Name	Elusinecoracana	Panicumsumat rense	Paspalumscro biculatum	Echinochloafru mentacea	Setariaitalica	Panicummiliace um
Chromoso me number	2n=4x=36, AABB	2n=4x=36, AABB	2n=4x=40	2n=6x=54	2n=2x=18, AA	2n=4x=36
Ploidy Level	Tetraploid	Tetraploid	Tetraploid	Hexaploid	Diploid	Tetraploid
Genome size estimate (pg)	3.34–3.87	N/A	1.91–1.98		1.02–1.04	2.08
Domesticati on	Ethiopia and Uganda	India	India	India	China, Europe and Afghanistan- Lebanon	China and Europe

Nevertheless, small millets are rich in micronutrients and essential amino acids for regulatory activities. Small millets are being considered for their paramount importance in



nutritional aspects not provided by other staple crops. These species have profound nutritional benefits, especially their micronutrient and protein profiles (see Table2) (Lydia et al., 2023).

Hence, international and national organizations have recently aimed to restore these lost crops for their desirable traits, and for that, they set new goals and breeding road maps for accelerated improvements of small millets crops (see Figure 1). small millets have received minimal research attention in terms of development of genetic and genomic resources and breeding for yield enhancement. Crop breeding is a rapidly advancing science. It is able to make use of genetic and biotechnological innovations to efficiently develop better crop varieties.

Table 2 Nutritional composition of small millets comparison with major cereals(Source: Lydiaet al.2023 and Hariprasanna, 2023)

Сгор	CHO(g)	Protein(g)	Fat(g)	Crude fiber (g)	Ash(g)	Ca(mg)	P(mg)	Fe(mg)	Zn(mg)	Mg (mg)
Finger millet	72.60	7.70	1.50	3.60	2.70	344.00	250.00	6.30	2.30	130.00
Foxtail millet	60.90	12.30	4.30	8.00	3.30	31.00	290.00	2.80	2.40	81.00
Pros millet	60.90	12.50	1.10	5.20	1.90	14.00	206.00	0.80	1.40	81.00
Barnyard millet	65.50	6.20	4. 40	13.60	2.20	20.00	280.00	8.00	3.00	137.00
Little millet	65.60	10.40	1.30	7.60	1.30	16.10	220.00	1.30	3.70	133.00
Kodo millet	66.20	8.90	2.60	5.20	1.70	15.30	188.00	2.30	0.70	147.00
Rice	78.20	7.90	0.50	1.00	11.19	7.50	160.00	0.70	1.30	64.00
Wheat	64.00	10.60	1.50	2.00	0.94	41.00	306.00	5.30	2.70	138.00
Maize	18.70	3.27	1.35	2.00	4.83	10.00	89.00	0.52	0.46	37.00



Figure 1 Breeding objectives and Road map for revitalizing small millets

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Research and development efforts

Compared to other crops, attention to small millets was limited till the establishment of separate AICRP on small millets in 1986.

The following goal should be set for the breeding and improvement of small millets crops

- Improvement of grain yield including maximization of biomass and the harvest index.
- Developed location-specific cultivar as per requirements of soil, rainfall, temperature, humidity, day length, and cropping patterns.
- Small millets are generally sown in poor soils so, develop cultivar which has a high Nutrient-use efficiency, particularly NPK
- Breeding of dwarf varieties to overcome lodging.
- Devolve drought-tolerant cultivars, which have a high water use efficiency
- Develop non-shattering cultivars to prevent yield loss in the field,
- Developing post-harvest technology demands based varieties which has easy threshing of grains and easy dehulling

Conventional breeding approaches

Various traditional breeding methods such as pure line selection, pedigree selection, mass selection, and mutation breeding, are applicable to self-pollinating crops in small millets as well. Reports on small millets cultivars released over a period of time showed that a majority of them were released following selection from local landraces/cultivars, followed by pedigree selection (hybridization and selection). For example, in India, of the 248 varieties of six small millets (finger millet-121, foxtail millet-32, proso millet-24, Kodo millet-33, barnyard millet-18, and little millet—20), about 65% were released following selection from landraces, about 30% through pedigree selection, and 5% through mutation breeding.

Molecular markers and Genomics assisted breeding for small millets improvement

The discovery and application of molecular markers for valuable traits reduce the requirement for complex phenotypic analysis and the time required for the development of new commercial varieties. DNA markers that are tightly linked to agronomically important genes (called gene tagging) may be used as molecular tools for MAS in plant breeding (Collard and Mackill, 2008). Germplasm collections including tolerant crops, landraces and wild relatives of crops can be used to identify or isolate QTL(s), gene(s) or allele(s) that



confer tolerance to abiotic stresses such as drought and high temperature by using modern genomics approaches. Although candidate QTL(s) can be deployed through MB approaches such as MABC, MARS and GS, the most promising candidate genes along with appropriate promoters can be used by using a GE approach in conventional breeding programs. Recent biotechnological developments are helping breeders make desired genetic changes with much greater precision. Breeders can cut out, add in, or otherwise "edit" genes so a plant can be more productive.

Molecular Breeding (MB) approach						
 Marker genotyping platforms 	Using diverse	Validation of QTLs/ markers in diverse lines	Genome wide selection (GWS)			
→ Single Nucleotide Polymorphism (SNPs)	populations having viral disease	↑ Identification of	↑ Marker assisted recurrent selection			
 Simple Sequence Repeats (SSRs) 	resistance	Identification of markers	(MARS) ↑ Marker Assisted			
Molecular markers development	→ parental lines by crossing	linked with viral disease resistance ▲	Backcrossing (MABC)			
Genomic resources M	apping population	Linkage mapping/	Use of pertinent			
development ★	development	association mapping	MB approaches ▲			

(Source:Mishraet al. 2017)

Multi-omics approaches for gene discovery and crop improvement

At present, it is a need to identify and characterize novel traits in small millets to generate stress-resistant and high-yielding cultivars, which is not possible by conventional breeding approaches. With the advancement of long sequencing tools and multi-omics techniques, it is now easier to generate larger germplasm and genome database.

The genome-editing techniquesare now getting better with time, the novel techniques like homologous recombination, RNAi and CRISPR/Cas based genome editing have revolutionized the process. As the approaches are cost-effective, easy to use, and prone to negligible error, will help to counter the climate change challenges and to eradicate world food security along with may attract, public acceptance to genome-engineered plants.



Genomic selection and speed breeding

Integrating genomic selection and speed breeding to fast-forward genetic gain in crops. The breeding cycle length decreased by genomic selection could be further reduced by performing population development under speed breeding conditions. Selection candidates could be phenotyped for grain yield secondary traits (*eg*, root architecture) in the glasshouse, and plants that carry the desired trait could be selected.

Context-dependent selection of the trait will enable plants to be better adapted to the target environment prior to selecting more complex traits, such as yield.

Phenotyping and selecting plants under speed breeding in the glasshouse could further improve selection intensity and the rate of genetic gain (see Figure2)



Figure 2: Integration of different techniques with speed breeding platform to enhance the result. GS, genomic selection; MABC, marker assisted backcrossing; MAS, markerassisted selection; NGS, next-generation sequencing (Source: Krishnappa*et al.* 2021; Pandey et al. 2022)



Conclusion:

Conventional breeding approaches have been successful in developing several cultivars. However, genomics assisted breeding will facilitate the identification of novel alleles and genes with superior agronomic performance and resistance to biotic and abiotic stresses to accelerate small millets improvement. Integration of different techniques with speed breeding platform will fast-forward the genetic gains in small millets.

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