

## STATUS OF PEARL MILLET RESEARCH IN INDIA

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**Abstract:** Pearl millet [*Pennisetum glaucum* (L.)R.Br.] is grown mainly as a rainfed crop in north and northwestern parts of country. These regions are highly prone to scanty and unevenly distributed rainfall and hence are always drought-prone. High temperature and low nutrient status of soils further exaggerate the deleterious effect of drought. The pearl millet growing regions are also most vulnerable to climate change as the frequency of drought is likely to increase. The purpose of this review is to explore and document pearl millet research efforts and important milestones in its improvement and to discuss their implications for future pearl millet improvement as a grain and forage crop for the drought- and heat-stressed areas, especially in India.

**Keywords:** AICPMIP, All India Coordinated Pearl Millet Improvement Program; CMS, Cytoplasmic male sterility; DAiT, Diversity array technology; OPV, Open-pollinated variety

## INTRODUCTION

Millets are indigenous to many parts of the world. The most widely grown among major millet is pearl millet. Africa is considered to be centre of origin of pearl millet. The genetic name *Pennisetum* is derived from two Latin words *Penna* meaning feather and *sera* meaning bristles. The *Pennisetum* consist of six sub species as *P.ramosum*, *P.glaucum*, *P.purpureum*, *P.massaicum*, *P.orientale*, *P.squamulatum*.

In India, Millets are cultivated in an area of approximately 7.8 million ha with an average of about 9.25 million tons of grain production. The crop is favoured due to its productivity and short growing season under dry, high temperature conditions. Pearl millet grains are high in nutrient composition and are considered an inexpensive source of energy as compared to staple cereals such as wheat, rice and maize in terms of micronutrients (Zinc and Iron), protein content and amino acid composition for the resource-poor farmers. Pearl millet is a C4 plant having a very high photosynthetic efficiency and dry matter production capacity. It is usually grown under the most adverse agro-climatic conditions where other crops like sorghum and maize fail to produce economic yields. Besides, pearl millet has a remarkable ability to respond to favourable environments because of its short developmental stages and capacity for high growth rate, thus making it an excellent crop for short growing seasons under improved crop management.

In some of the hottest and driest regions where agriculture is possible in India and Africa, pearl millet is the only cereal that can be grown under dryland conditions and so plays a critical role in food security. However, people living in these regions have not yet benefited much from the current 'biotechnology revolution', or even the 'green

revolution' that dramatically increased food grain production on irrigated lands over a generation ago. Exotic germplasm in the genus *Pennisetum* comprises of primary, secondary and tertiary gene pools with many species possessing desirable genes for pearl millet improvement such as resistance to diseases, male sterility, improved fodder characteristics and apomixis.

**Plant breeding in pearl millet**

Genetic variability for agronomic traits is the key component of breeding programmes for broadening the gene pool of crops. However, the genetic variability for many traits is limited in cultivated germplasm. Genetic variation among landraces is of vital importance to breeding programmes that aim to produce improved landrace-based cultivars for marginal growing environments.

Breeding efforts in pearl millet are aimed at maximum exploitation of hybrid vigor for both grain and forage yields. Hybrid parents are bred mostly by recurrent selection which is based on the natural out-breeding behaviour of the crop; or by pedigree breeding from populations that have mostly been derived from single crosses. This approach continues to be widely used and quite successful, although the obvious effect is narrowing of the genetic base of hybrid cultivars. Cultivars developed from OPVs (Open pollinated varieties) and hybrids are highly heterogeneous and heterozygous, open to significant genetic changes. Though NILs and RILs hold great importance in breeding and mapping for traits, their limitations are the long time and/or high cost required for development, and these populations only detect the additive component but do not provide information on dominance relationships for any QTL.

Pearl millet is being exploited in breeding programs to develop DH but merely as the pollinator which

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will be further eliminated, resulting in haploids of the recipient species, eg. wheat, oat etc.

The genetic improvement of pearl millet in India started in 1930s and largely concentrated on improving the yield by mass selection and progeny testing, which led to development of some open-pollinated varieties (OPVs). For instance, mass selection in locally adapted material led to the development of C1, C2, Co.2 and Co.3 in Tamil Nadu; T5, T55, A1/3, S350 and S530 in Punjab; AKP1, AKP2 and AKP3 in Andhra Pradesh; Baroda 5, B117, B119 and 14D in Madhya Pradesh; RSJ, RSK, Bari N207 and S14 in Rajasthan; LM 38-39, Bajra 1, Bajra 2, S-14, S 28-25-2, Puri and Sadas 11 in Maharashtra; and Bijapur 11-11-7-14-6, Golagiri 1-8-5-5 and Sindagi 3-16-13-9-4 in Karnataka. Similarly, improvement for morphological uniformity in a landrace from Ghana at the Indian Agricultural Research Institute, New Delhi led to the development of 'Improved Ghana' and further mass selection in 'Improved Ghana' led to the development of Pusa Moti.

The greatest push for the development of OPVs as a part of population improvement programme started in 1970s with the establishment of the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT). Exploiting a diverse range of germplasm, mostly from the African sources, several composites were developed and improved by recurrent selection, resulting in the development and dissemination of a large number and diverse range of breeding materials.

Pearl millet hybrids are largely generated from relatively narrow gene pools and current breeding programs do not make use of wild pearl millets except as donors of specific traits such as apomixis or resistance to pests and diseases and make only limited use of landrace germplasm.

Pearl millet being a highly cross-pollinated crop with outcrossing rate being more than 85% and displaying a high degree of heterosis for grain and stover yields, attempts were made to exploit heterosis in the 1950s utilizing the protogynous nature of flowering of this crop. The usual method at that time for production of hybrid seeds was growing the parental lines in mixture and allowing them to cross-pollinate. The resultant seed contained approximately 40% hybrid seed when the two parental lines in mixture and allowing them to cross-pollinate. The resultant seed contained approximately 40% hybrid seed when the two parental lines had synchronous flowering at about same time. These chance hybrids this produced outyielded local varieties by 10-15%. However, they could not become popular due to their limited yield advantage over OPVs, narrow range of adaptation and lack of seed production programmes. The above mentioned limitation in the exploitation of heterosis were circumvented with the discovery of cytoplasmic-nuclear male sterility and release of male-sterile lines Tift 23A and Tift 18A in early

1960s at Tifton, Georgia, USA. These lines were made available to Indian breeding programmes. The male-sterile line Tift 23A was extensively utilized, both at the Punjab Agricultural University and the Indian Agricultural Research Institute, because of its short stature, profuse tillering, uniform flowering and good combining ability. This laid the foundation of pearl millet hybrid breeding in India.

In the past few years most of the allele mining studies in this crop for improvement of various agronomic traits such as yield and stover quality as well as for abiotic and biotic stress resistance such as drought tolerance (DT) and downy mildew resistance have been achieved by generating and using bi-parental mapping populations.

Genetic maps based on molecular markers and identified quantitative trait loci (QTLs) have been used for developmental and breeding programs in pearl millet to enhance its productivity under the present scenario of global climatic change

Molecular markers like restriction fragment length polymorphism (RFLP), amplified fragment length polymorphism (AFLP), expressed sequence- simple sequence repeats (EST-SSRs), conserved intron spanning primer (CISP), single strand conformational polymorphism- single nucleotide polymorphism (SSCP-SNP) and diversity array technology (DArT) are dominant and proven to be useful in the evaluation of QTLs associated with various traits, terminal drought stress tolerance and adaptation and disease resistance. Along with genetic markers, core or mini core and reference collections are also helpful to identify new sources of abiotic and biotic stress resistance in pearl millet.

Association mapping, also known as linkage disequilibrium (LD) mapping opens a new platform for allele mining with the help of ancestral recombination events in natural populations or germplasm collections to make marker-phenotype associations. It has the advantage over QTLs associated linkage mapping in terms of less laborious, time efficient and generation of 1000s of recombinants with large and diverse gene pool. Recently a pearl millet inbred germplasm association panel (PMiGAP) of about 346 lines was generated that represented a collection of approximately 1000 diverse cultivars, landraces and parents of mapping population from various regions of Africa and Asia, out of which 250 lines were used for association mapping of DT traits. This PMiGAP is thought to provide new insights for fine mapping of QTLs and allele mining of favourable genes for important agronomic traits.

In order to understand whether control of water loss under non-limiting water conditions is implicated in terminal DT of pearl millet, a study was carried out involving two contrasting pair of parents of mapping population (PRLT2/88-33 X H77/833-2 and 863B-P2 X ICMB 841-P3) and near-isogenic lines (NILs) generated from introgression of a major DT-QTL

from PRLT2/88-33 (donor) into H77/833-2 (NILS-QTL) under water deficit conditions. Tolerant varieties and NILS-QTL showed low transpiration rate as compared to sensitive genotypes in well-watered conditions. Kholova (2010b) established that pearl millet genotypes carrying a DT-QTL for terminal DT, including tolerant and NIL-QTLs, have higher ABA levels and lower transpiration rate ( $Tr$ ) under well-watered conditions as compared to sensitive genotypes at all vapour pressure deficit (VPD) levels, ultimately contributing to absolute water saving during grain-filling. Furthermore, F7 recombinant inbred lines identify four QTLs that contributed to increase  $Tr$  under high VPD conditions, out of which the major QTL was mapped on LG6.

Terminal drought stress (flowering through grain filling) is more damaging to pearl millet's asynchronous tillering behaviour and rapid growth rate allow it to recover rapidly from intermittent drought stress during these earlier stages of plant development, but provide no advantages under unrelieved terminal drought stress. Improving the adaptation of pearl millet to terminal drought stress environments is, therefore, a major objective for breeding programmes aimed at improving both the crop's productivity and its yield stability.

A number of landraces have been identified which could serve as a rich source of diversity for abiotic stress tolerance in pearl millet. CZP9802, the first open pollen pearl millet variety derived from the landraces of Rajasthan is not only highly adapted to drought but is also high yielding (14-33% higher grain yield and 18-36% higher stover yield) as compared to the controls Pusa 266 and ICTP 8203. CZP9802 is endowed with the property to escape terminal drought stress owing to its exceptional attributes of flowering within 48 days and maturity in 75 days making it a suitable cultivar for arid zones of India. ICMV 221 is being cultivated in India, is terminal DT, downy mildew resistant and has improved grain yield potential.

As compared to other cereal crops only limited information has been available on response to soil salinity in pearl millet. Reduced shoot N content and increased  $K^+$  and  $Na^+$  content is usually associated with salinity tolerance in pearl millet. According to Krishnamurthy (2007), shoot biomass ratio associated with salt tolerance and shoot  $Na^+$  concentration could be used as potential selection criteria for screening of pearl millet germplasm at vegetative stage. ICRISAT has carried out some basic research on salinity tolerance of pearl millet in collaboration with the International Centre for Biosaline Agriculture (ICAB) along with NARS partners in both India and WANA region. For the screening of salinity tolerant breeding materials of pearl millet, pot culture method has been followed in the salinity affected fields. Screening of pearl millet germplasm has resulted in the development of

advanced breeding materials, improved population including OPVs, gene pools and composites, parental lines of potential hybrids, and germplasm accessions with high grain and forage yield presumably with a high degree of salinity tolerance. In the short-to-medium terms, some of these materials can be released for cultivation after extensive validation of their yield performances at on-farm trials.

H77/833-2, an elite pearl millet inbred line, hybrids of which are widely used by farmers in north-western India, is sensitive to terminal drought stress but tolerant to high-temperature stress. This cultivar has been used as one of the parents (recurrent) to generate a set of mapping population (NILs) to map QTLs for terminal DT of grain and stover yield as well as their component traits in pearl millet.

Like other cereal crops, pearl millet is also prone to various biotic stresses including fungal, viral and bacterial infections. A combination of various disease management practices can be helpful in combating fungal disease including cultural methods, chemical methods, and host plant resistance. Further identification and utilization of new genes for host plant resistance for developing varieties resistant to biotic stresses are very important. It can be achieved by developing precise phenotyping methods, exploiting natural genetic mechanisms of resistance.

Hybrids of pearl millet have been mass-produced using cytoplasmic-nuclear male-sterile (CMS) female parents. Apomixis has the potential to become as important in millet hybrid production as is CMS, allowing breeders to more rapidly and efficiently use available germplasm to produce hybrids. CMS millet plants are male-sterile because their pollen is not viable. Female fertility, however, is usually normal. CMS results from specific interaction of the cytoplasm and the nucleus. Cytoplasmic-nuclear male sterility was first documented in pearl millet by Burton (1958). A1, released by Burton (1965), has been the most widely used cytoplasm for producing commercial hybrids. Tift 23A was developed as a female parent with A1 cytoplasm and became the seed parent in India for the first two millet hybrids released. The five distinct cytoplasm (A1 to A5) reported in pearl millet have been distinguished mainly by restoration and maintained relationships when crossed by known sterility maintainer and fertility restorer-lines.

Mitochondrial DNA restriction endonuclease fragment and maize mitochondrial gene probe hybridization patterns have been used to distinguish the cytoplasm in pearl millet. Rajeshwari *et al.* (1994) characterized diverse cytoplasm of pearl millet by Southern blot hybridization and maize mitochondrial DNA probes.

Apomixis is a reproductive mechanism by which seed is produced from somatic cells that develop into embryos without fertilization. These cells and the resulting embryos have the same chromosomal and genetic constitution as the plant on which the seed is

produced. Of the three basic types of apomixis, apospory is the only type confirmed in millet. As a result of these types of apomixis, all progeny of a plant are derived from somatic cells and are therefore identical to the plant on which the seed is formed. Interest in apomixis of millet stems come from the need for efficiency of breeding and seed production. With apomixis, one could explore all the available germplasm to make hybrids. Apomixis is not known to occur naturally in pearl millet but it has been induced in mutation studies.

Apomixis does occur in other species of *Pennisetum*, and a project was initiated in the late 1970s to transfer apomixis from a wild apomictic *Pennisetum* species into pearl millet. A number of wild apomictic species were investigated, but the most success was obtained with hexaploid ( $2n=6x=54$ ) *P. squamulatum* crossed with tetraploid ( $2n=4x=28$ ) pearl millet (Hanna *et al.* 1992). Apomixis would have the greatest impact for producing pearl millet hybrids in countries where hybrids are not used. It would rapidly make available the increased yield and vigor of hybrids, regardless of the heterozygosity of the parental lines.

This is an obligate aposporous species and has widely been used to study inheritance and transfer of genes controlling apomixis in pearl millet background, as well to produce high quality perennial fodder types in combination with pearl millet and napier grass (*P. purpureum*), in the form of tri-specific or double cross hybrids. Recently, a  $2n=56$  cytotype of *P. squamulatum* was identified, characterized by obligate apomictic mode of reproduction, perenniality and tolerance to ergot, smut, frost and drought under field conditions.

The cross-pollinated breeding system, availability of commercially usable cytoplasmic-nuclear male-sterility systems, and heterosis for grain and fodder yield and several other traits of agronomic and economic importance, provide for open-pollinated varieties (OPVs) and hybrids as the two broad cultivar options. Pearl millet hybrids have 25-30% grain yield advantage over OPVs. Therefore, the major emphasis in India has been on hybrid development, initially targeted to relatively better endowed agro-ecoregions, with grain yield as the highest priority trait.

#### Utilization of Molecular markers

Pearl millet is one of the most important small-grained  $C_4$  Panicoid crops with a large genome size (~2352Mb), short life cycle and outbreeding nature. Pearl millet has excellent nutrient composition and exceptional buffering capacity against variable climatic conditions and pathogen attack makes pearl millet a wonderful model crop for stress tolerance studies.

Genetic diversity studies in *Pennisetum* germplasm offer possibilities of identifying diverse germplasm for utilization in improving pearl millet open-pollinated varieties and hybrids. These efforts require

effective DNA marker-based fingerprinting strategies for rapid assessment of genetic relationships.

As molecular markers play an important role in improving our understanding the genetic basis of economically important traits and are efficient tools to speed up crop improvement. During the past decades, various molecular markers such as RFLPs, STSs, AFLPs, single-strand conformational polymorphisms, genomic SSRs and genomic SSRs have been developed for pearl millet. It has been found that DArT performs well in polyploidy species, can be rapidly developed for practically any genome in contrast to SNPs, and offers a practical solution to the problems associated with development and application of markers for orphan crops. It is a cost effective, solid state platform, hybridization-based marker technology that offers a high multiplexing level (being able to simultaneously genotype several thousand loci per assay), while being independent of sequence information. Genetic differences in pearl millet have been studied previously by morphological and isozyme analysis. Subsequently, RAPDs, RFLPs, and SSRs with other PCR-compatible markers have been used to estimate pearl millet genetic diversity. The power of DArT fingerprinting method lies in its ability to compare different genomes at a large number of loci in a single assay, at an average cost per marker locus that is very low compared with PCR-compatible markers. The large numbers of markers that are simultaneously assayed by DArT provide a high level of resolution in genetic diversity studies. Genetic distance estimated derived using DArT provides a high level of resolution in genetic diversity studies. Genetic distance estimates derived using DArT are more likely to be accurate because the 'random' nature of DArT markers should reduce the ascertainment bias when compared with technologies relying on targeted marker development.

The first major milestone was achieved in 1993 with the creation of a molecular marker-based genetic linkage map of the pearl millet genome with 181 restriction fragment length polymorphism (RFLP) markers-the marker system of choice in the early 1990s. Now more than 699 molecular markers have been created and mapped for pearl millet, a consensus linkage map has been produced, and quantitative trait loci (QTL) for disease resistance, drought tolerance, flowering time and grain and stover yield, and ruminant nutritional quality of straw have been mapped. ICRISAT and its partners have successfully demonstrated the use of RFLP markers in the marker-assisted backcross transfer of additional downy mildew resistance into a parental line of popular pearl millet hybrid "HHB 67" and a new version of this hybrid, "HHB 67 Improved" based on the improved parental line, was released for commercial cultivation in 2005 as the first public-bred product of marker-assisted breeding in India.

However, RFLP markers are too labor-intensive and high cost for applied use, as well as having potential health and environmental hazards. Thus these markers are not considered suitable for large-scale genotyping applications in an applied plant breeding program.

The identification of a major QTL for drought tolerance on linkage group (LG 2) in pearl millet and its validation in two independent MAS programmes provides a strong impetus to pursue this key genomic region further to answer the remaining challenging questions.

Pearl millet germplasm show a large range of genotypic and phenotypic variations including tolerance to abiotic and biotic stresses. Conventional breeding for enhancing abiotic and biotic stress resistance in pearl millet have met with considerable success, however, in last few years various novel approaches including functional genomics and molecular breeding have been attempted in this crop for augmenting yield under adverse environmental conditions, and there is still a lot of scope for further improvement using genomic tools. Discovery and use of various DNA-based markers such as EST-SSRs, DArT, CISP and SSCP-SNP in pearl millet not only help in determining population structure and genetic diversity but also prove to be important for developing strategies for crop improvement at a faster rate and greater precision. Molecular marker-based genetic linkage maps and identification of genomic regions determining yield under abiotic stresses particularly terminal drought have paved way for marker-assisted selection and breeding of pearl millet cultivars. Reference collections and marker-assisted backcrossing have also been used to improve biotic stress resistance in pearl millet specifically to downy mildew. Whole genome sequencing of pearl millet genome will give new insights for processing of functional genes and assist in crop improvement programs through molecular breeding approaches.

#### **Conservation efforts**

*Ex situ* seed storage is one of the most extensively used methods for conserving pearl millet genetic resources. Pearl millet germplasm has been preserved in various national and international gene banks. India with 11243 accessions has the largest collection of pearl millet germplasm followed by Brazil (7225) and Canada (3764). The initiative for the conservation of pearl millet genetic resources in India was taken up by International Crop Research Institute for Semi-Arid Tropics (ICRISAT), Patancheru in collaboration with different national and international organizations including several national organizations such as National Bureau of Plant Genetic Resources (NBPGR), All India Coordinated Pearl Millet Improvement Project (AICPMIP), National Agricultural Research Systems (NARS), Agricultural Universities, and other ICAR Institutes etc. About 65 organizations had contributed

approximately 10,764 accessions of pearl millet in different disciplines of ICRISAT, in past few years.

Pearl millet exhibits a vast range of genetic diversity in its global germplasm collection. Core and mini-core collections of pearl millet germplasm could serve as wonderful cost effective resources for allele mining and identification of genotyping variants for resistance to biotic and abiotic stresses as well as other important agronomic traits. A core collection comprises of about 10% of the entire germplasm collection while a mini core is comprised of ~1% of the total collection. Upadhyaya (2009) reported a core collection of 2094 pearl millet accessions which was huge in itself to be evaluated by breeders for crop improvement. Hence, a mini –core collection of 238 pearl millet accessions representing all accessions of the core collection was constituted using data on 10 qualitative and eight quantitative traits. This mini-core collection with its greatly reduced size could be effectively utilized for accurate evaluation of desirable traits including tolerance to abiotic and biotic stresses as well as for mapping with molecular markers and identification of trait-specific germplasm and discovery of candidate genes. Other than the core and mini-core collections, a genotype based reference set comprised of 300 accessions is also available in pearl millet. A global composite collection of accessions in pearl millet based on SSRs and high-throughput assays has also been developed at ICRISAT prior to the formation of reference sets for assessing population structure and diversity. This reference set reportedly capture 87-95% of the allelic diversity of the composite collections (ICRISAT, 2008/2009).

DNA-based molecular markers, genetic linkage maps, and sequence information are essential genomic resources to carry out genetic studies or marker-aided breeding in any crop. Though the discovery of molecular markers and construction of genetic maps in millets lag far behind those of staple cereals, pearl millet reportedly has comparatively substantial stock of genetic and genomic resources in the form of DNA-based markers, such as AFLP, RFLP, RAPD, expressed sequence tag-based (EST) markers, sequence-tagged sites (STSs), simple sequence repeat (SSRs/microsatellites), DArTs, CISP and SNP have been developed to distinguish genetic variability, linkage map analysis and marker assisted screening to expedite the breeding programs. Molecular markers help in evaluating genetic differences in germplasm collections for appropriate selection of mating parents for hybrid breeding, studying population structure, and analysis of QTLs for abiotic and biotic stress resistance. Pearl millet exhibits a very high level of DNA marker polymorphism even between elite inbred parental lines of popular hybrids. A set of 21 polymorphic EST-based SSRs markers and six genomic markers were developed in pearl millet and were tested to detect polymorphism across 11 pairs of pearl millet

mapping population parental lines. These EST-based markers are also used for marker assisted breeding and crop improvement programs in pearl millet at ICRISAT. Senthilvel (2010) also reported the development of an array of about 6900 DArT clones using PstI/BanII complexity reduction, and identification of a total of 256-277 polymorphic DArT markers in three RILs of pearl millet. A total of 574 polymorphic DArT markers were identified in a set of 24 genetically diverse pearl millet inbreds from 7000 DArT clones obtained from 95 diverse genotypes using PstI/BanII complexity reduction. With the help of DArT markers, comparative genomics and genome organization studies can be easily done and the cost of marker-assisted backcrossing (MABC) is also low as compared to others. Sehgal (2012) used available pearl millet ESTs and developed 75 SNP and CISP markers and demonstrated their use in identifying candidate genes underlying a major DT-QTL using four genotypes namely, H77/833-2, PRLT 2/89-33, ICMR 01029 and ICMR 01004, representing parents of two mapping populations. There is another recent research on identification of 83,875 SNP markers using genotyping by sequencing (GBS) of PstI-MspI reduced representation libraries in 500 genotypes of pearl millet, comprised of 252 global accessions and 248 Senegalese landraces, which exhibited high genetic diversity relative to other genotypes of Africa and Asia. Ambavat (2016) constructed a linkage map comprising 286 loci (229 DArT and 57 SSR markers) through the genotyping of 256 DArT 7 RILs derived from the cross between 81B-P6 and ICMP 451-P8. Using these markers, QTL for rust resistance have been mapped on LG1 with LOD score of 27. These molecular markers have been used to construct a pearl millet genetic linkage map and to identify and map QTLs for terminal drought, reduced salt uptake, grain and stover yield, and for downy mildew resistance, rust and blast resistance. These genetic tools have also been used for diversity assessment, studying recombination rates, analyzing domestication syndrome and comparative genetics. It has also been proposed that pearl millet genome has undergone several structural rearrangements that could be possibly associated with its evolution and adaptation in severe abiotic stresses and nutrient deficient soils although until now marker relationships are almost collinear across all the 10 mapping populations developed and mapped. There have been only a few reports on transcriptome analysis in pearl millet for delineating the mechanism of abiotic stress tolerance till date. Mishra (2007) reported the identification of about 2494 differentially regulated transcripts in response to drought, salinity and cold stress indicating the existence of a complex gene regulatory network in this stress tolerant crop. Transcriptome analysis using 454 Roche NGS tool was done to understand the mechanisms underlying resistance to downy mildew

in pearl millet. A comprehensive pearl millet transcriptome assembly (~43 Mbp) has also been developed by integrating data from three independent investigations carried out by Zeng (2011) and Rajaram (2013) and is comprised of about 69,398 tentative assembly contigs. This transcriptome assembly also includes pearl millet ovule transcriptome data. Of late development of cultivars using genetic engineering approaches is gaining significance in plant biology and stress physiology. The understanding of mechanisms that regulate gene expression and the capacity to transfer important genes from other organisms into plants will expand the ways in which plants can be utilized. In pearl millet, the biolistic method for gene delivery has been frequently used for transformation. Girgi *et al* (2006), introduced an *afp* gene from the mold *Aspergillus giganteus*, encoding an antifungal protein AFP into pearl millet through particle bombardment. A significant reduction in disease symptoms against rust and downy mildew infection was shown. Similarly, a *Brassica juncea* non-expressor of pathogenesis-related gene 1 (BjNPR1) had been stably integrated and expressed into a pearl millet male fertility restorer line ICMP451 through *Agrobacterium*-mediated transformation. Development of genetically engineered pearl millet varieties is still in nascent stage despite its economic importance.

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