Pearl Millet Breeding Lines Developed at ICRISAT: A Reservoir of Variability and Useful Source of Non-Target Traits

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Introduction

The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) conducts inter-disciplinary and partnership-based research for the genetic improvement of its mandate crops. During the 1970s and 1980s, the pearl millet improvement at ICRISAT, Patancheru, emphasized on: (i) developing a diverse range of trait-specific composites, based on the germplasm largely from the Western and Central Africa; (ii) improving them by the process of recurrent selection, principally for grain yield and downy mildew (Sclerospora graminicola (Sacc.) Schroet) resistance; and (iii) developing open-pollinated varieties (OPVs) (Rai and Anand Kumar 1994). The improved composites, OPVs, and breeding lines derived from them, were disseminated for worldwide utilization. These germplasm, improved populations, and breeding lines were also utilized at ICRISAT, Patancheru, mostly for pedigree breeding which was aimed at developing a wide and diverse range of improved trait-specific breeding lines and hybrid parents employing higher grain yield potential and downy mildew (DM) resistance, as the main selection criteria. Limited efforts were also made to develop composites, OPVs, and breeding lines with additional resistance to smut (Tolyposporium penicillariae Bref.) and ergot (Claviceps fusiformis Loveless).

In 1990s, pearl millet improvement program at ICRISAT adopted a regional strategy, by aligning its research and breeding product development priorities with the priorities of the regional players. In case of India, public sector breeding programs in the National Agricultural Research System (NARS) were largely directed towards hybrid breeding as well as private seed companies, which were rapidly emerging as dominant players and were engaged only in the hybrid development. Thus, pearl millet breeding at ICRISAT, Patancheru, made a strategic shift towards developing and disseminating a diverse range of high-yielding and downy mildew (DM) resistant, trait-based breeding lines and hybrid parents (seed parents and restorer parents) while their utilization in hybrid development and commercialization was taken up by the NARS and private seed companies. This strategy proved very successful, as reflected from the increase in the grain yield productivity from 6 kg ha^{-1} year^{-1} during the year 1966-1980 to 24 kg ha^{-1} year^{-1} during the year 1996-2012 (Yadav and Rai 2013). Much of this yield improvement occurred due to the development and adoption of a large number and diverse range of high-yielding and DM resistant hybrids, of which more than 85% were based on the ICRISAT-bred hybrid parents, or on the proprietary hybrid parents developed by the seed companies that included the improved breeding lines developed at ICRISAT in their parentage (Mula et al. 2007). The large on-farm hybrid cultivar diversity not only led to increases in grain yield, but also stemmed the large-scale DM epidemics, which were frequent events prior to 1990 (Rai et al. 2006). In this paper, we present, in an illustrative context, the variability for key agronomic traits and DM resistance, for which the targeted breeding was done. Additionally, we also examine the usefulness of the diversity in the breeding lines as a source for the non-target traits, if any, for which no deliberate selections were made during the breeding process.

Agronomic traits

Since 1981, ICRISAT has developed, designated, and disseminated more than 180 seed parents, i.e., male-sterile lines (A-lines) and their counterpart maintainers (B-lines).

These were characterized for their agronomic traits depending on the specific years they were developed. Ninety nine B-lines developed during the years1981-2004 were jointly evaluated in replicated trials during the summer and rainy seasons of 2005 at Patancheru with the aim of assessing the variability among these lines for key agronomic traits. While breeding for yield potential with specific traits, the principal strategy adopted at ICRISAT, Patancheru, was to breed d2 dwarf (a recessively inherited trait) seed parents on which hybrids of various heights could be developed, depending on the height of the restorer lines (R-lines). Besides, d2 dwarf seed parents have additional advantages from the view point of seed production (ease of *roguing* and lodging avoidance), which is done under high crop management conditions. Thus, 63 of the 99 B-lines were d2 dwarf with <110 cm height, and a few lines exceeded 120 cm height (Table1). Most of these lines took 45-54 days to 50% flowering and very few lines were very early (<42 days) or late (> 57 days). Twenty seven lines produced > 3 effective tillers plant-1 and 74 lines had 15-25 cm of panicle length. The panicle diameter in most of these lines was 2.0-3.0 cm and had 1000-grain weight mostly between 8-12 g. However, there were few lines that had >30 cm height or >3.5 cm thick panicles, and >12g of 1000-grain weight.

A similar evaluation of 116 restorer lines (R-lines) during the 2007 rainy and 2008 summer season, showed large variability for these agronomic traits. In contrast to B-lines, most of the R-lines had >120 cm height (some of them exceeding even 160 cm), but a few lines had no more than 100 cm height (Table 2). Majority of these lines took 42-54 days to 50 % flowering, thus providing an opportunity to breed hybrid parents and hybrids with a wide maturity range. Most of the lines had 1-3 effective tiller plant-1 but one line had >4 tillers plant-1. Panicle length in most of the lines ranged between 15-30 cm, but one line had >35 cm long panicles. Panicle diameter in most of the lines was ≤3.0 cm but one line had >3.5 cm thick panicles. During the period these lines were developed, there was no emphasis laid on breeding large-seeded R-lines. Thus, 41 R-lines had ≤ 8 g of 1000-grain weight. However, 22 lines had >11g of 1000-grain weight, and 3 lines had even >13 g of 1000-grain weight.

Molecular diversity

The expression of the quantitatively inherited morphological traits is highly influenced by the environments. For instance, two lines having similar flowering time may not be genetically similar with respect to the morphological trait but they may be similar with respect to the other traits. Such hidden differences are clearly unravelled by molecular analysis. From amongst the lines characterized for agronomic traits as described before, 98 B-lines and 115 R-lines were analysed for molecular diversity using 38 simple sequence repeats (SSR) markers. Most of the B-lines were grouped together in one cluster, and most of the R-lines were grouped together to form a second cluster, with four sub-clusters in B-lines and five sub-clusters in R-lines, respectively (Figure. 1). The 38 markers detected a total of 308 alleles with an average of 8.1 alleles per locus, and as many as 4-5 alleles each at 17 marker loci (Nepolean et al. 2012). Molecular variance analysis showed that differences between the B-lines and R-lines clusters accounted for 18% of the total variation, while 82% of the variation existed within the B-lines cluster and R-lines cluster. The polymorphic information content (PIC) was 0.56 in R-lines cluster, slightly higher as compared to the B-lines cluster (0.46). Similar was the case with respect to the gene diversity (0.52 in R-lines cluster and 0.50 in B-lines cluster). The results of the analysis of additional B-lines and R-lines with the same 38 markers, when combined with the results of this study, have shown even greater allelic richness, higher PIC, and gene diversity.

The parental lines used in the above studies represent a small sample of a large number of breeding lines and hybrids parents that have been developed and disseminated over the years. For instance, during the 2010 Scientists Field Days, 1792 breeding lines and hybrid parents were selected by breeders from NARS and seed companies, and during the 2012 Scientists Field Days, 2782 breeding lines and hybrid parents were selected. Apart from the supplies of the seed materials selected during these Scientists Field Days held on alternate years, a large number of breeding lines were evaluated in the trait-based nurseries run by NARS every year, under a collaborative research program with the All India Coordinated Pearl Millet Improvement Project. Dissemination of these lines has substantially diversified the genetic base of the hybrid programs conducted by both NARS and seed companies. A replicated evaluation of such large number of lines was economically prohibitive. In a recent effort to identify lines for constituting the heterotic gene pools, 606 breeding lines (400 from ICRISAT and 206 from public and private sector partners' organizations) are being evaluated at multiple locations for key agronomic traits. These are also being used for genome sequencing studies. The data generated from these studies will be an indicator of the magnitude of diversity that could be expected if almost all of the breeding lines produced and disseminated so far were to be evaluated.

Downy mildew resistance

Resistance to downy mildew (DM), the most dreaded disease of pearl millet in India, has been an integral part of the genetic improvement research program at ICRISAT. Breeding lines and germplasm sources with resistance to various pathotypes have been identified, but DM resistance breeding was mostly based on the utilization of resistance sources identified in the breeding materials. This, coupled with effective large-scale screening techniques (Singh et al. 1997) has significantly accelerated the breeding process. One hundred and forty-one B-lines were screened in the greenhouse under high disease pressure using seedling inoculation for checking their resistance against the following five diverse pathotypes: (1) Sg 200 from Jamnagar (Gujarat), (2) Sg 212 from Durgapura (Rajasthan), (3) Sg 139 from Jodhpur (Rajasthan), (4) Sg 298 from New Delhi, and (5) Sg 150 from Jalna (Maharashtra). It was found that 11 lines were resistant (<10% DM incidence) to all the five pathotypes while 12 lines were resistant to any four of the pathotypes. Based on agronomic traits and DM resistance, 9 of these lines were designated as ICMB 11111 through ICMB 11999 for the purpose of dissemination (Table 3). Following the same procedure and using the same five pathotypes, 131 B-lines were screened in the year 2012, out of which 33 lines were found resistant to all five pathotypes while 21 lines were resistant to any four of the pathotypes. Also, based on agronomic traits and DM resistance, five of these lines were designated as ICMB 12111 through ICMB 12555 for the purpose of dissemination. These results illustrated the variability for DM resistance to multiple pathotypes in the advanced breeding lines that have been derived from the crosses among the elite breeding lines available in the large pool of diverse breeding materials developed at ICRISAT.

Cytoplasmic diversity of A-lines

The A1 system of cytoplasmic-nuclear male sterility (CMS), discovered more than 50 years ago (Burton 1958), still continues to be the only CMS source being used for breeding A-lines at almost all of the research centres worldwide. During 1990s, ICRISAT initiated a research to identify, introduce, and characterize alternative CMS sources, and showed that the two distinct A4 and A5 CMS systems were more promising than the A1 system (Rai et al. 2006). While de-emphasizing the use of A1 CMS system, ICRISAT developed 68 A-lines using this CMS system during the years 1995-2013 (Table 4). Utilization of the A4 CMS system at ICRISAT was initiated in the year 1998 and further expanded in the years that followed, producing a total of 71 A-lines during the years 1998-2013. Utilization of the A5 CMS system was initiated in the year 2001, producing 18 A-lines during the years 2001-2013. Although ICRISAT documented the various advantages of the A4 and A5 CMS systems, and developed as well as disseminated large number of A-lines, their utilization by breeders for hybrid development in both the public and private sectors did not start, due to lack of suitable restorers in their breeding programs. ICRISAT then initiated limited breeding efforts for developing and disseminating restorers, especially for the A5 CMS system, for which the restorers in the breeding materials at ICRISAT and elsewhere were almost non-existent. Just recently, the advantages of these two CMS systems for genetic diversification of A-lines have been widely realized in India as reflected in a research prioritization consultation meeting of Hybrid Parents Research Consortia (Rai et al 2012).Consequently, several research programs in India, both in the public and private sectors, have started their own A-line and R-line breeding for these two CMS systems.

Non-target traits

Deliberate selection for the various agronomic traits and DM resistance has generated large variability for these traits in the breeding lines as described above. Utilization of the existing breeding lines in crosses and selection continues to further generate more variability for these traits. During the past 10 years or so, new issues have emerged, triggering the search for new traits and their utilization in cultivar development. Since there had been no prior selection for these so-called' non-target traits' during the course of breeding, search for these can be made in the germplasm. However, the transfer of genes for these traits, from germplasm into adapted elite agronomic backgrounds devoid of any detrimental effects on agronomic traits is a lengthy process. If the sources of these non-target traits are found in the elite breeding lines, it can significantly accelerate the breeding process. Therefore, a wide range of breeding lines have been screened for several non-target traits and sources of these have been identified.

Resistance to leaf diseases

Leaf blast on pearl millet, caused by *Pyricularia grisea* (Cooke) Sacc. [teleomorph:Magnaporthe grisea (Herbert) Barr], has emerged as a serious disease during the past 5-6 years in several parts of India. This disease not only affects grain and fodder yield but also the fodder quality. This disease becomes more severe during humid weather conditions, especially in the dense plant stands. Rust, caused by Puccinia substriata var. indica, is another foliar disease, which also affects the grain and fodder yield as well as the fodder quality. Rust infection and disease development is favored by lower temperatures. In northern India, the disease does not frequently occur until the flowering time in September, when temperatures are somewhat moderate. In other regions of the country, rust may attack even at the seedling stage. Rust is of major concern in the peninsular India where pearl millet is planted during the post-rainy (rabi) season. However, pearl millet rust has also been reported in the central and peninsular India during the cultivation of the summer season (March–May) crop where seed production is undertaken. Effective large-scale field and greenhouse screening techniques have been developed; both for leaf blast (Sharma et al. 2013) and rust (Singh et al. 1997). Blast reaction is recorded on the scale of 1 to 9, where 1 = no lesions to small brown specks of pinhead size, and 9 = >75% leaf area covered with lesions or all the dead leaves. Lines showing ≤3.0 score are categorized as resistant, and those with score between 3.1 to 5.0 are categorized as moderately resistant. In case of rust, lines with <10% rust severity are categorized as resistant and those with 10-20% rust severity are categorized as moderately resistant.

One hundred sixty-two hybrid parents (B-lines and R-lines) were screened during the year 2012, in the greenhouse under high disease pressure for blast resistance against the following five pathotypes of M. grisea: (1) Pg 45 from Patancheru (Andhra Pradesh), (2) Pg 53 from Kherpa (Rajasthan),(3) Pg 56 from Gotan (Rajasthan), (4) Pg 118 from Rewari (Haryana), and (5) Pg 119 from Bhojawas (Haryana). While several lines were found resistant to one or the other pathotypes, seven B-lines and two R-lines were found resistant to all the five pathotypes (Table 5). During the year 2008-09, post rainy (rabi) season, 214 advanced breeding lines, including 126 B-lines, 23 R-lines and 65 potential R-lines, were evaluated for checking the rust resistance in the disease nursery at Patancheru, under natural epiphytotic conditions. Eight lines (1 B-line, 7 R-lines) that showed resistance (≤10% rust severity) in the field screen were further evaluated (or screened) in the greenhouse under high pressure using artificial inoculation. One B-line and three R-lines were found resistant to rust, with all three R-lines (including the two sister lines) showing hypersensitive reaction (Table 6). In another experiment conducted during the year 2012, 69 hybrid parents and populations with high biomass yield potential were screened for rust resistance under greenhouse conditions. Of these, 8 hybrid parents were categorized as resistant (<10% rust severity) (Table 6) and 27 were categorized as moderately resistant (10-20% rust severity). However, some of the rust resistant lines such as ICMB 03222, ICMR 06999, and ICMB 96222, were found having 35-40% rust severity in a later evaluation done under field conditions during Feb-March, 2014. This indicated existence of pathogenic variability and a change in the virulence of the pathogen population at Patancheru. Even though, these three lines are now susceptible to the pathogen population at Patancheru, they may be tested for resistance against pathotypes/races prevalent in other pearl millet growing areas of India.

Pearl millet is cultivated as a rotational mulch crop on about 2 million ha in no-till soybean production system in central Brazil. Adriana Seed Company, a member of the Pearl Millet Hybrid Parents Research Consortium, is evaluating pearl millet for grain and fodder production. ICRISAT had supplied 446 hybrid parents and breeding lines to Adriana Seed Company during the year 2009-12. Several of these were found useful for various yield components and are currently being used in the breeding program of the Adriana Seed Company. In the lowland regions of Central Brazil at altitudes below 600 m, leaf spot caused by Bipolaris setariae and leaf blast, are the two major leaf diseases prevalent in February-early April planting caused due to the gradual decrease of precipitation and day time temperatures, and high relative humidity. In later plantings, at altitudes above 600 m, rust is a major disease caused due to lower night time temperatures and higher dew levels. As per Saari and Prescott (1975), during the course of evaluation, for agronomic traits breeding lines introduced from ICRISAT were scored for both rust and blast respectively. Lines with the score (a) 1-2 were categorized as highly resistant (HR), (b) scores 3-4 were categorized as resistant (R), and (c) score 5-6 were categorized as moderately resistant (MR). Most of the lines were susceptible to both the diseases, while, based on a 3-year evaluation; it was found that 36 lines had moderate resistance to both diseases. Five lines were resistant to Bipolaris setariae and were moderately resistant to rust (Table 7). Based on a 1-year evaluation, it was found that one line was resistant to both diseases but only further evaluation will be able to confirm whether this line holds resistance, especially against rust, as learned from the observations in ICRISAT studies.

Flowering-period heat tolerance

Pearl millet has recently emerged as a highly productive and remunerative summer season crop in parts of Gujarat, Rajasthan, Uttar Pradesh, and Maharashtra states of India. During the flowering period in this season, the air temperature often exceeds 42°C and may reach as high as 45°C, leading to poor seed set and low grain yield. However, a few hybrids developed by the seed companies, based on the improved breeding lines and hybrid parents bred at ICRISAT, have shown excellent seed set and give very high grain yield. One such hybrid that has been under cultivation for a long time and has shown the highest level of flowering-period heat tolerance is Proagro 9444, which is based on a reselection within ICRISAT-bred A- line ICMA 92777 and its maintainer, ICMB 92777. Based on the previous field screening of 173 B-lines grouped in three sets and evaluated at 4-6 locations during 2009-2011 (different set for each year), 14 B-lines were found to be tolerant. These were re-evaluated together in trials conducted at 3 locations in 2012 and 4 locations in 2013. Sanchor in Rajasthan was found to be the most stressful location in 2012 with the air temperatures exceeding 450C during the flowering-period, while Aligarh was the most stressful location in 2013 with the air temperatures x exceeding 430C during flowering-period. Six B-lines (including ICMB 92777) were identified as the most heat tolerant (Table 8). Five of these lines had 68-74% mean seed set, which were comparable to that of Proagro 9444. Our preliminary observations indicate heterosis for seed set under high air temperatures, implying that hybrids developed from the use of parental lines identified above would have higher seed set than Proagro 9444. These lines along with others identified as heat tolerant based on the 2-year screening, were used to develop a heat- tolerant composite. These lines were used by some of the seed companies and heat-tolerant breeding lines were derived. The seed of the heat-tolerant composite was disseminated to NARS and private seed companies to provide a broad-based source of heat tolerance.

C. Grain iron and zinc density

Iron (Fe) and Zinc (Zn) deficiencies have been recognised worldwide as serious public health problems, especially in the developing countries where rural population and the urban poor are heavily dependent on the staple cereals, as a major source of their dietary energy and nutritional requirements. For instance, about 80% of the pregnant women, 52 % of the non-pregnant women, and 74% of the children in 6-35 months age group in India suffer from iron deficiency (Chakravarty and Ghose, 2004). On the other hand, about 52% of the children below 5 years are zinc deficient. The HarvestPlus Challenge Program of the CGIAR, initiated the resource mobilization and coordination efforts in 2004 with the aim to support the biofortified cultivar development of several crops, including pearl millet. Also, this initiative was considered as a cost-effective and sustainable approach to address this problem. While germplasm evaluation was undertaken to identify the sources of high Fe and Zn densities, hybrid parents and advanced breeding lines from mainstream breeding, developed from the use of a large number and diverse range of germplasm, were screened to assess the variability and identify possible sources of high levels of these micronutrients in elite agronomic backgrounds.

More than 480 advanced seed parent progenies and more than 560 restorer parent progenies developed in the mainstream breeding were evaluated in the rainy season of the year 2010 at Patancheru, and the open-pollinated grain samples were analyzed using X-ray Fluorescence Spectroscopy (XRF) technique. Large variability was observed for both the micronutrients (Table 9). While 12% of the seed parents progenies and 31% of the restorer parent progenies had <40 mg kg^{-1} of Fe density, about 21% of the seed parent progenies and 15% of the restorer parent progenies had >70 mg kg^{-1} Fe density, with few progenies in both the sets exceeding even 100 mg kg^{-1} Fe density. Similarly, while 26% of the seed parent progenies and 42% of the restorer parent progenies had <40 mg kg^{-1} Zn density, about 8-10% of both the progenies had >60 mg kg^{-1} Zn density. Thus, identification of these progenies with such high levels of Fe and Zn density from amongst the breeding lines otherwise not bred for these micronutrients as target traits was quite significant, considering the fact that the highest-Fe commercial cultivar ICTP 8203, identified to date, had 77 mg kg^{-1} Fe density and 65 mg kg^{-1} Zn density.

The high-Fe progenies identified in 2010 have been evaluated for Fe and Zn over the years and seasons in different trials, using ICTP 8203 as a standard high-Fe control. Similarly, high-Fe progenies identified from among the additional mainstream breeding lines in the year 2011 had also been evaluated over the years and seasons in various trials, using ICTP 8203 as a control. Since the data on Fe and Zn densities of these lines were from different trials and different years/seasons, and the Fe and Zn densities of ICTP 8203 itself varied across the trials, Fe and Zn densities of each line was also presented as a percent of ICTP 8203, for a valid assessment of the relative superiority of these lines over the control. Based on six year x season environments, several high-Fe progenies were identified, out of which nine exceeded the Fe density of ICTP 8203 by an average of 15 to 48% (Table 10). All these progenies had >80 mg kg^{-1} Fe density with one having exceptionally high-Fe density of 104 mg kg^{-1}. The mean Zn density in these progenies varied from 46 to 57 mg kg^{-1}, being 3-4% less than ICTP 8203 in two lines and exceeding by an average of 4 to 25% in seven lines. Except for the two progenies that involved a NCD2 (Nigerian Composite Dwarf) progeny as one of the parents in the cross, all the other progenies were derived from crosses that had both parents developed from iniadi germplasm, along with a progeny from Extra-early B-composite (EEBC) which was involved in most of the crosses.

A similar procedure of successive evaluation of the restorer parent progenies in 4-8 repeat trials over the years and seasons led to the identification of several high-Fe progenies, which exceeded the Fe density of ICTP 8203. Fifteen of these derived from diverse parentage are given in Table 11. The mean Fe density in these progenies varied from 76 to 115 mg kg^{-1}, exceeding the Fe density of ICTP 8203 by an average of 5 to 66%. Thirteen of these progenies had mean Zn density of 53-64 mg kg^{-1}, thereby exceeding the Zn density of ICTP 8203 by an average of 3 to 23%. All these progenies were derived from populations (composites and OPVs) or from crosses that are entirely or largely based on iniadi germplasm with the exception of MRC-1-15-130-2-2-1-B-B-3-B-B-B-1-3-1, which was derived from Mandor Restorer Composite (MRC). It should be noted that MRC was derived by inter-mating high-tillering, early-maturity, and small-seeded inbred lines from ICRISAT and five State Agricultural Universities in Northern India (Yadav et al. 2012). This high-Fe progeny, however, has generally 1-2 effective tillers plant-1 and has large seed size, which are typical characteristics of iniadi germplasm. This may imply that MRC or its progeny during the course of breeding perhaps got out-crossed with some iniadi line, producing large-seeded and low-tillering segregants, out of which one produced this high-Fe breeding line. While the true parentage of this high-Fe line remains as a matter of speculation, it is one of the few restorer parent progenies' which was found to be stable for very high level of Fe density.

Conclusions

Pearl millet improvement research at ICRISAT, Patancheru, has made extensive use of germplasm from Western Africa, the primary center of diversity, for the genetic enhancement of this crop. Composites and OPVs, largely based on this germplasm, and the breeding lines derived from them, were further used over the years, for breeding a diverse range and large number of trait-specific breeding lines and hybrid parents with improved yield potential and DM resistance as the principal selection criteria. These breeding lines were disseminated and used worldwide, predominantly in India. Evaluation of a rather small sample of hybrid parents (seed parents and restorer parents) and breeding lines showed large variability for the key agronomic traits and DM resistance, for which targeted selections were made. Molecular diversity analysis grouped seed parents in one cluster and restorer parents in another separate cluster, with large within-cluster diversity, indicating good prospects of further genetic diversification of seed parents and restorer parents using these lines in the breeding programs. During the breeding process, no selection was made for resistance to leaf blast and rust, flowering-period heat tolerance, and high Fe and Zn densities. Yet, several lines were found promising for these non-target traits. This indicated that besides the target traits for which ICRISAT breeding lines were selected, the large pool of breeding lines could be an useful source of new traits for which intended selections wasn't done (i.e. non-target traits), but which might be necessary for breeding pearl millet cultivars which are adaptable to the changing environmental conditions and also in addressing changing farmers and consumer requirements.

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Table 1. Frequency distribution of designated pearl millet B-lines for agronomic traits. Mean of 2005 summer and rainy season, Patancheru.

Trait		Number of B-lines in trait class							
Plant height (cm)	Trait class	60–70	71–80	81–90	91–100	101–110	111–120	121–130	131–140
	No. of lines	6	17	18	16	19	13	8	2
Time to 50% flower (days)	Trait class	<42	42.1–45	45.1–48	48.1–51	51.1–54	54.1–57	57.1–60	
	No. of lines	3	8	21	44	12	9	2	
Number of productive tillers plant^{-1}	Trait class	1	1.1–2	2.1–3	3.1–4	4.1–5	5.1–6		
	No. of lines	0	31	41	22	4	1		
Panicle length (cm)	Trait class	<15	15.1–20	20.1–25	25.1–30	30.1–35			
	No. of lines	18	64	10	5	2			
Panicle diameter (cm)	Trait class	2.0–2.5	2.6–3.0	3.1–3.5	3.6–4.0				
	No. of lines	39	43	15	2				
1000-grain weight (g)	Trait class	6–7	7.1–8	8.1–9	9.1–10	10.1–11	11.1–12	12.1–13	
	No. of lines	1	8	15	25	22	21	7	

Source: Rai et al. (2009)

Table 2. Frequency distribution of designated pearl millet R-lines for agronomic traits. Mean of 2007 rainy season and 2008 summer season, Patancheru.

Trait	No. of R-lines in trait classes									
Plant height (cm)	Trait class	<90	90.1-100	100.1-110	111.1-120	120.1-130	130.1-140	140.1-150	150.1-160	>160
	No. of lines	1	3	4	6	19	25	19	19	20
Time to 50% flowering (d)	Trait class	<42	42.1-45	45.1-48	48.1-51	51.1-54	54.1-57	57.1-60	>60	-
	No. of lines	-	21	31	35	20	8	1	-	-
Number of productive tillers plant^{-1}	Trait class	1	1.1-2	2.1-3	3.1-4	4.1-5	5.1-6	>6	-	-
	No. of lines	-	47	63	5	1	-	-	-	-
Panicle length (cm)	Trait class	<15	15.1-20	20.1-25	25.1-30	30.1-35	>35	-	-	-
	No. of lines	9	68	28	10	-	1	-	-	-
Panicle diameter (cm)	Trait class	< 2.0	2.0-2.5	2.6-3.0	3.1-3.5	3.6-4.0	-	-	-	-
	No. of lines	21	63	27	4	1	-	-	-	-
1000-grain weight (g)	Trait class	<6	6-7	7.1-8	8.1-9	9.1-10	10.1-11	11.1-12	12.1-13	>13
	No. of lines	6	12	23	24	18	11	11	8	3

Source: Gupta et al. (2011)

Table 3. Downy mildew incidence in B-lines designated in the years 2011 and 2012 to five diverse pathotypes of Sclerospora graminicola

	DM incidence (%) against pathotype				
B-line	Sg200	Sg212	Sg139	Sg298	Sg150
ICMB 11111	17	18	22	0	0
ICMB 11222	2	0	4	52	51
ICMB 11333	7	0	13	0	0
ICMB 11444	7	2	4	0	0
ICMB 11555	4	5	0	0	18
ICMB 11666	22	6	8	0	0
ICMB 11777	0	0	0	4	0
ICMB 11888	0	19	23	61	0
ICMB 11999	51	50	8	30	9
ICMB 12111	0	0	0	8	0
ICMB 12222	2	3	0	13	0
ICMB 12333	100	18	0	4	2
ICMB 12444	23	7	41	17	9
ICMB 12555	0	6	2	14	4
7042 (Susc. control)	97	99	99	99	91

Note: DM= Downy mildew.

Table 4. Pearl millet A-lines developed and designated at ICRISAT, Patancheru during 1981-2013

A-line designation period	No. of A lines with cytoplasm		
	A1	A4	A5
1981 - 1994	26	-	-
1995 - 2000	28	9	-
2001 - 2006	14	35	4
2007 - 2013	26	27	14
Total	94	71	18

Table 5. Leaf blast resistant B-lines and R-lines of pearl millet

B/R Line	Blast severity score (1-9 scale) against pathotype				
	Pg 45	Pg 53	Pg 56	Pg 118	Pg 119
ICMB 88004	3	2	3	2	2
ICMB 92444	3	2	2	1	2
ICMB 02111	3	3	3	3	2
ICMR 06444	2	3	3	1	1
ICMB 07111	2	1	1	1	2
ICMB 09333	2	1	1	1	1
ICMB 09999	2	1	1	1	2
ICMB 93333	3	1	1	2	3
ICMR 11003	2	2	1	2	2
ICMB 95444 (Susceptible control)	7	8	8	9	8

Table 6. Rust resistant designated hybrid parents and restorer-parent progenies of pearl millet

	Rust severity (%)		
	Field screen, Patancheru		Greenhouse screen
Line	Lower leaves	Top four leaves	
ICMB 96222	10	3	2
ICMR 06999	3	0	Hypersensitive response
ICMP 451-P8	10	0	Hypersensitive response
ICMP 451-P6	10	0	Hypersensitive response
ICMB 03222	*	*	5
ICMB 91444	*	*	5
ICMB 02666	*	*	5
ICMB 09888	*	*	5
HHVBC tall (C1) S1-33-3-1-1-1-2-B-B-3-2-3	*	*	5
MRC HS-86-1-1-5-B-B-B-B-B-B-2	*	*	5
ICMS 7704-S1-52-3-1-1-3-2-3-3-B-B-11	*	*	7
ICMS 7704-S1-126-5-2-2-5-1-3-B-B-1-5	*	*	8
ICMB 89111 (Susceptible control)	58	21	51

** Not screened under field condition*

Table 7. Pearl millet breeding lines identified as resistant to leaf diseases at Campo Grande, Brazil.

Material	Identity/pedigree	No. of years evaluated	Rust rating	Bipolaris rating
Designated B-line	ICMB 07999	3	MR	R
Seed parent progeny	(ICMB 95444 x ICMB 94555)-12-1-5-3-3	3	MR	R
	(ICMB 99555 x ICMB 00555)-5-4-3-B-B-2-1	3	MR	R
	[[{ICMV 88908-11-12-3-2-B x B-bulk)-8-B-3 x {(843B x ICMPS 900-9-3-2-2)-41-2-5-5 S2-34-1-2-1-1 x B-bulk}-5-B-B]-11-1-1-B-B x ICMB 04111]-69-2-3-1	1	R	R
Restorer progeny	JBV 3 S1-171-2-1-1-1-B	3	MR	R
	{[((MC 94 S1-34-1-B x HHVBC)-16-2-1) × (IP 19626-4-2-3)]-B-28-3-2-2-2}×{ICMS 8511-S1-62-2-1-2-2-2-4-1-3-B}-B-13-1-1	3	MR	R

Table 8. Seed set in designated B-lines of pearl millet identified for flowering-period heat tolerance

B-line	Seed set (%) 2012 (Sanchor)	2013 (Aligarh)	Mean
ICMB 92777	73	65	69
ICMB 00333	73	75	74
ICMB 03555	72	72	72
ICMB 05666	71	74	73
ICMB 07222	74	62	68
ICMB 09111	66	60	63
Proagro 9444 (Heat- tolerant control)	73	70	72

Table 9. Frequency distribution of mainstream breeding lines for grain iron (Fe) and zinc (Zn) densities in pearl millet, 2010 rainy season, Patancheru.

			Percent lines in micronutrient class (mgkg-1)							
Breeding line	No. of lines	Micronutrient	<40	41-50	51-60	61-70	71-80	81-90	91-100	>100
Seed parent progenies	487	Fe	12	20	29	17	11	6	2	2
		Zn	26	38	25	9	<1	<1	0	0
Restorer parent progenies	565	Fe	31	25	16	12	7	5	1	2
		Zn	42	33	17	5	2	<1	0	0

Note. High-Fe control ICTP 8203 had 77 mg kg-1 Fe and 65 mg kg-1 Zn density.

Table 10. Iron (Fe) and Zinc (Zn) density in high-Fe seed parents progenies, Mean of 6 (year x season) environments (2010-2013), Patancheru

	Fe density (mg kg-1)		Fe density as % of ICTP 8203		Zn density (mg kg-1)		Zn density as % of ICTP 8203	
Parentage of breeding line	Mean	Range	Mean	Range	Mean	Range	Mean	Range
(EEBC S1-407-1-B-B-B-B-B-1-B-1-B-10-1 x B-bulk (3981-3989 G))-2-4-1	82	57-101	117	93-131	51	35-67	106	82-133
(EEBC S1-407-1-B-B-B-B-B-1-B-1-B-13-1 x B-bulk (3981-3989/S06 G1))-1-2-3	81	68-104	117	89-161	50	29-65	104	85-133
(EEBC S1-407-1-B-B-B-B-B-1-B-1-B-5-1x B-bulk (3981-3989/S06 G1))-2-1-3	88	61-113	125	100-146	55	31-81	114	80-130
(ICMB 04888 x ICMB 02333)-1-1-3-2	77	68-88	110	106-115	49	33-64	101	87-114
(ICMB 95111 x EEBC S1-407-1-B-B)-17-3-1-B-B-B-B-4-Bx 3981-4011 G2}-1-4-2	90	76-104	129	119-150	57	40-72	119	91-140
(ICMB 99555 x ICMB 99111)-2-1-1-B-B-B-1	81	55-101	115	90-132	46	33-57	97	75-116
(NC D2 BC7F4-34-3-1-2-B-2-B x EEBC 407)-12-1-2	86	54-97	121	88-149	56	36-75	117	82-151
(NC D2 BC7F4-34-3-1-2-B-2-B x EEBC 407)-4-2-2-2	86	62-102	123	101-153	52	35-66	111	78-147
{[(843B x ICTP 8202-161-5)-20-3-B-B-3 x B-bulk]-2-B-9 x [(ICMB 96555 x LaGrap C2 S1-32-1)-10 x IP 14758-2-1]-8-2}-1-1-1-2	86	80-92	123	110-142	46	34-53	96	78-117
{[(BESCBPT/91-40 x SPF3/S91-3)-1-2-2-3 x B-bulk]-8-1-1-3-B-B-B-B-3-1 x B-bulk (3981-4011/S06 G1)}-1-3-2	104	79-123	148	129-168	57	38-82	125	86-161

Table 11. Iron (Fe) and Zinc (Zn) density in high-Fe restorer parents progenies, Mean of 4-8 (year x season) environments (2010-2013), Patancheru.

Parentage of breeding line	No. of environments	Fe density (mg kg-1)		Fe density as % of ICTP 8203		Zn density (mg kg-1)		Zn density as % of ICTP 8203	
		Mean	Range	Mean	Range	Mean	Range	Mean	Range
AIMP 92901 S1-15-1-2-3-B-3-B-9-2-1	8	92	71-119	126	107-152	53	30-78	102	74-121
AIMP 92901 S1-296-2-1-1-4-2-B-7-3-1	6	102	90-118	142	129-162	57	42-72	114	101-129
HHVBC Tall S1-51-1-P1-3-B	7	100	77-129	138	119-155	53	38-62	103	88-116
ICMR 312 S1-59-1	5	97	83-118	135	118-159	60	46-77	114	105-127
ICMV 221 S1 - 366	5	86	80-92	123	116-132	58	42-73	112	104-116
ICMV 96490-S1-15-1-2-2-1-2	7	115	95-138	166	147-215	62	41-77	123	92-158
ICTP 8203 S1-386	5	98	84-124	145	132-168	61	41-83	122	99-147
LaGrap C2-S1-14-4-1-3-4-4	7	89	72-112	124	111-145	57	43-70	112	85-132
MRC HS-130-2-2-1-B-B-3-B-B-B-1-3-1	8	110	93-128	149	135-182	64	42-76	120	106-137
SDMV 90031-S1-11-1-1-3-3-B-4-B-2-1-B	6	87	64-107	119	99-138	57	38-79	109	91-123
(EERC-HS-8)-B-2-1-2-1	7	100	85-119	139	119-153	48	28-78	104	73-181
(MC 94 C2-S1-3-1-3-3-1-2-1 x ICMR 312 S1-3-2-3-2-1-1-B-B)-B-46-P1-1	4	92	87-96	121	113-124	51	36-69	96	69-108
(MC 94 C2-S1-3-1-3-3-1-2-1 x SDMV 90031 S1-3-3-2-2-2-2-2)-B-8-2-1	7	76	65-87	105	84-128	62	44-74	119	91-140
(MC 94 C2-S1-3-2-2-2-1-3-B-B x AIMP 92901 S1-488-2-1-1-4-B-B)-B-30-1-3	4	79	66-105	110	94-144	54	41-67	103	90-120
[(IPC 1617×SDMV 90031-S1-84-1-1-1-1)×AIMP 92901 S1-296-2-1-1-3-B-1]-4-4-2-1	7	90	69-121	123	105-157	59	36-79	112	104-122

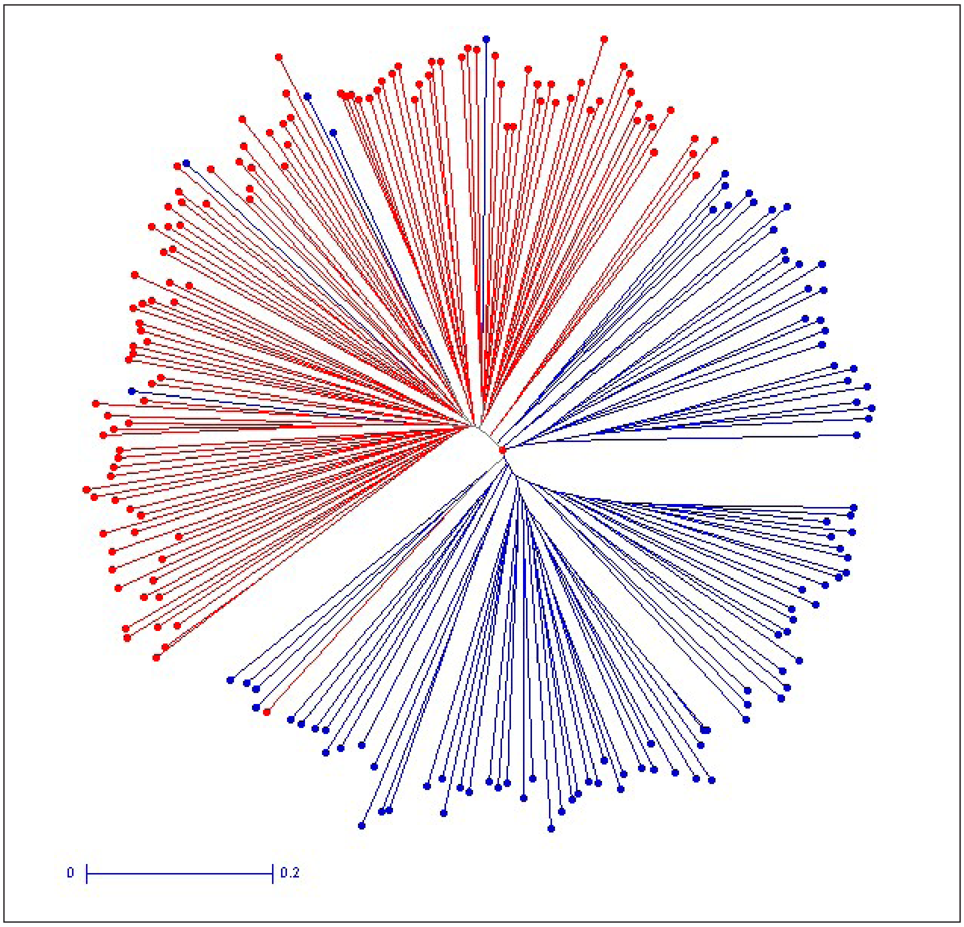

Figure 1: Clustering pattern of 98 B-lines and 115 R-lines based on genotypic data (B-lines are shown in blue color and R-lines in red color)

Source: Nepolean et al. (2012)