

# Development of Pearl Millet Minicore Collection for Enhanced Utilization of Germplasm

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## ABSTRACT

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] exhibits enormous genetic diversity in the global germplasm collection. The pearl millet core collection (~10% of the entire collection), with 2094 accessions, is still large for precise evaluation to identify parents for a genetic improvement program. A minicore collection of pearl millet comprising 238 accessions was constituted by using data on 10 qualitative and 8 quantitative traits of 2094 core collection accessions. The hierarchical cluster analysis of data using a phenotypic distance matrix resulted in 136 clusters. A proportional sampling strategy with 10% or a minimum of one accession from each cluster was used to form the minicore collection. The comparison of data in the minicore and core collections using various statistical parameters, such as homogeneity of distribution for geographical origin and frequency classes of qualitative traits, means, median, within and between-accession variances, diversity index, and phenotypic correlations, indicated that almost the entire genetic variation present in the core collection was captured in the minicore collection. The possible impact of high between- and within-accession variability in pearl millet on maintenance of genebank accessions is discussed. With its greatly reduced size, the pearl millet minicore collection can be used for precise evaluation of traits of agronomic importance and biotic and abiotic stresses as well as mapping with molecular markers for identification of trait-specific germplasm and discovery of new genes.

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**Abbreviations:** BLUPs, best linear unbiased predictors; CR%, coincidence rate; H', Shannon-Weaver diversity index; MD%, mean difference percentage; REML, residual (or restricted) maximum likelihood; VD%, variance difference percentage; VR%, variable rate.

PEARL MILLET [*Pennisetum glaucum* (L.) R. Br.] is a C<sub>4</sub> cereal crop grown mostly in the arid and semiarid regions of Africa and Asia on an area of 26 million ha (Rai et al., 2009) and is the sixth-ranked cereal after wheat (*Triticum aestivum* L.), maize (*Zea mays* L.), rice (*Oryza sativa* L.), barley (*Hordeum vulgare* L.), and sorghum [*Sorghum bicolor* (L.) Moench]. It is primarily cultivated for grain, but is also a valuable source of fodder (both stover and green forage). In nontraditional areas, such as the southern United States, Brazil, Australia, and Korea, it is grown for forage and silage production for dairy. Pearl millet is highly cross-pollinated and exhibits large genetic variability. The genebank at the International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Patancheru, conserves and maintains the largest collection of pearl millet germplasm, comprising 21,594 accessions from 50 countries, including 750 accessions of 24 wild species of genus *Pennisetum* (Upadhyaya et al., 2007). The large collection of germplasm in genebanks, coupled with limited information on traits of economic importance, has resulted in very limited use of germplasm by crop breeders (Bhattacharjee et al., 2007), thus contributing to a narrow genetic base in many crops (Dalrymple, 1986; Vellve, 1992; Cantrell et al., 1996; Upadhyaya et al., 2003). The concept of core collection (about 10% of the entire

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collection) was visualized to enhance the use of germplasm in breeding programs (Frankel and Brown, 1984a). Core collections have been reported in cereal crops; pearl millet (Bhattacharjee et al., 2007; Upadhyaya et al., 2009a), sorghum (Rao and Rao, 1995; Grenier et al., 2001), rice (Yan et al., 2007), maize (Li et al., 2004), finger millet [*Eleusine coracana* (L.) Gaertn.] (Upadhyaya et al., 2006a), and foxtail millet [*Setaria italica* (L.) Beauv.] (Upadhyaya et al., 2008). In pearl millet, Bhattacharjee et al. (2007) developed a core collection using data on 11 quantitative traits. This core collection was augmented with 501 accessions representing 4717 new accessions and exclusion of 7 (5 duplicates and 2 male sterile lines) accessions, resulting in a revised core collection of 2094 accessions (Upadhyaya et al., 2009a). Precise evaluation using multiple locations and replications of this core for economic traits that show high genotype  $\times$  environment interaction would be resource intensive. To overcome this, Upadhyaya and Ortiz (2001) suggested the minicore collection approach in crops having a large number of accessions. A minicore collection is a subset of the core, consisting of about 10% of the core collection, or 1% of the entire collection, and representing its diversity. Minicore collections have been developed in a number of crops such as chickpea (*Cicer arietinum* L.) (Upadhyaya and Ortiz, 2001), groundnut (*Arachis hypogaea* L.) (Upadhyaya et al., 2002), pigeonpea [*Cajanus cajan* (L.) Millsp.] (Upadhyaya et al., 2006b), rice (Ebana et al., 2008), sorghum (Upadhyaya et al., 2009b), and finger millet (Upadhyaya et al., 2010). The minicore collections in different crops have been evaluated for identifying useful traits (Upadhyaya, 2008; Upadhyaya et al., 2009c). The objective of this study was to establish a pearl millet minicore collection that is representative of the diversity of the pearl millet core and entire collection for effective use in breeding programs, molecular studies, and discovery of new genes.

## MATERIALS AND METHODS

Data of 18 phenotypic traits (10 qualitative and 8 quantitative) generated from evaluation of 2094 accessions from 46 countries in the pearl millet core collection developed at ICRISAT (Upadhyaya et al., 2009a) were used to develop the minicore subset. The accessions were grouped into early ( $\leq 50$  d), medium (51–80 d), and late ( $\geq 81$  d) maturity on the basis of days to flowering. They were then evaluated in an augmented design with one of the three control cultivars of different maturities, namely, ICTP 8203 (early), Raj 171 (medium), and IP 3616 (late), after every nine test accessions of specific maturity group, in the alfisol-Patancheru Soil Series (Udic Rhodustalfs) field in the rainy season (June to October), 2007, at Patancheru (18°N, 78°E, 545 m above sea level, and 600 km from the sea), Andhra Pradesh, India. ICTP 8203 (PI 537113) is an early-maturing, dark gray and large-seeded, open-pollinated, high-yielding variety developed by random mating of five  $S_2$  progenies selected from an *Iniadi* landrace, originating from northern Togo, at ICRISAT, Patancheru, India (Rai et al., 1990). Raj 171 is a medium-maturing, open-pollinated cultivar developed by random mating of

eight  $S_1$  progenies from an intervarietal composite selected at ICRISAT, Patancheru, India (Christinck et al., 1990). IP 3616 is a dual-purpose, late-maturing landrace from Tamil Nadu, India. Each plot consisted of a single row of 4-m length, with row spacing of 75 cm and plant-to-plant spacing of 10 cm. Care was taken to ensure uniform planting depth of 3 cm. The experimental field received 150 kg ha<sup>-1</sup> di-ammonium phosphate as basal fertilizer at the time of field preparation and 100 kg ha<sup>-1</sup> urea as topdressing 3 wk after sowing. The experiment received five life-saving irrigations each with 5 cm water. A recommended package of agronomic practices for raising a good crop was followed.

Data on 10 qualitative characters (synchrony of panicle maturity, panicle shape, panicle density, bristle length, seed shape, seed color, endosperm texture, green fodder yield potential, seed yield potential, and overall plant aspect) and 8 quantitative characters (days to flowering, plant height [centimeters], number of total and productive tillers, panicle length [centimeters], panicle thickness [millimeters], panicle exertion [centimeters] and 1000-seed weight [grams]) were recorded following pearl millet descriptors (IBPGR and ICRISAT, 1993). Observations were recorded on 30 representative plants in each accession for all the quantitative traits except 1000-seed weight, which was recorded on plot basis as weight of randomly taken 1000 seeds obtained from pooled seed from 30 representative plants of the plot. Days to flowering were recorded as days from sowing to the stage when main tiller panicle of a plant exhibited stigma emergence. Synchrony of panicle maturity, green fodder yield potential, seed yield potential, and overall plant aspect are visual observations and were recorded on a plot basis on a 1 to 9 scale, where 1 = poorest or lowest and 9 = excellent or highest; panicle shape on a 1 to 9 scale (1 = cylindrical and 9 = globose); panicle density on a 1 to 9 scale (1 = most loose and 9 = most compact); seed shape on a 1 to 5 scale (1 = obovate and 5 = globular); seed color on a 1 to 10 scale (1 = ivory, 5 = deep gray, and 10 = mixture of white and gray grains in a panicle); bristle length on a 1 to 9 scale (bristles below the seed apex level to  $>4$  cm above the seed apex level); and endosperm texture on a 1 to 9 scale (nine different combinations of percentage of corneous and starchy endosperm texture) following IBPGR and ICRISAT (1993).

The data on quantitative traits were analyzed following Residual Maximum Likelihood (REML; Patterson and Thompson, 1971) using GENSTAT 12th edition (<http://www.vsnl.co.uk>, verified 27 Oct. 2010). For all the quantitative traits, except 1000-seed weight, genotypic variance was partitioned into between and within-accession variances and tested for significance against respective standard errors. Best linear unbiased predictors (BLUPs) (Schönfeld and Werner, 1986) were calculated for all the eight quantitative characters for each accession. On the basis of BLUPs, mean, range, and variances for all quantitative traits were calculated for the core and minicore collection separately. A distance matrix using data on 18 traits was created following Gower (1985) and subjected to hierarchical cluster algorithm of Ward (1963) at an  $R^2$  (squared multiple correlation value) of 0.75 for clustering of accessions. This method optimizes an objective function because it minimizes the sum of squares between groups. A proportional sampling strategy with 10% or a minimum of one accession from each cluster was used to form a minicore collection. The 46 countries of origin were grouped into 11 regions (Table 1). Frequencies for geographic regions, countries within regions, and all the

qualitative traits in the core and minicore collections were tested by  $\chi^2$ . Heterogeneity of  $\chi^2$  was calculated as the difference between the sum of  $\chi^2$  values of countries within a region and the pooled  $\chi^2$  value of the region. The Yates (1934) correction was applied if the number of accessions in a particular region or country was less than five in the core collection. Means for the core and minicore collections were compared by the Newman-Keuls procedure (Newman, 1939; Keuls, 1952). Homogeneity of variances between accessions was tested by Levene's test (Levene, 1960). Based on predicted means of each plant, within-accession variance was calculated for each accession. The percentage of significant differences between the core and minicore collections was calculated for the mean difference (MD%) and variance difference (VD%) (Hu et al., 2000). The coincidence rate (CR%) and variable rate (VR%) were calculated to compare the core and minicore collections (Hu et al., 2000). The Wilcoxon (1945) rank-sum nonparametric test was performed using the SAS NPAR1WAY procedure to compare the medians of quantitative traits between the core and minicore collections (SAS Institute Inc., 2009). The Shannon-Weaver diversity index ( $H'$ ) (Shannon and Weaver, 1949) was used to measure and compare the phenotypic diversity for qualitative and quantitative traits in the core and minicore collections. The phenotypic correlations among eight quantitative traits using accession means in the core and minicore collections were estimated independently to determine whether the associations, which may be under the same genetic control, were conserved in the minicore collection.

## RESULTS AND DISCUSSION

### Constitution of the Minicore

The hierarchical cluster analysis on phenotypic distance matrix using Ward's (1963) method resulted in 136 clusters. Number of accessions in individual clusters varied from 2 to 50. A minicore collection of 238 accessions (11.4% of the core collection and 1.1% of the entire collection) was formed using the proportional sampling strategy (10% or minimum of one accession).

### Frequency Distribution

$\chi^2$  probabilities for frequency distribution of accessions in geographic regions and countries within regions in the core and minicore collections were nonsignificant for most of the 46 countries except Gambia ( $P = 0.006$ ) in West Africa (Table 1), indicating representative similarity.  $\chi^2$  probabilities for all the 11 geographic regions were also nonsignificant except for North Africa ( $P = 0.0002$ ) and Europe ( $P = 0.009$ ). Heterogeneity values were nonsignificant for all the regions except North Africa ( $P = 0.007$ ), indicating that the countries within each region were represented adequately. The overall  $\chi^2$  (38.65 at 45 df) was nonsignificant ( $P = 0.736$ ).

$\chi^2$  probabilities for frequency distribution of classes in all 10 qualitative traits were nonsignificant ( $P = 0.0859$  to 0.9495) (Table 2). Uniform distribution of classes in the core and minicore collections indicated that the sampling technique to constitute the minicore was appropriate and that the minicore represented the core collection for qualitative traits.

**Table 1.  $\chi^2$  test for the frequency distribution of pearl millet [*Pennisetum glaucum* (L.) R. Br.] core and minicore collection accessions in different regions and countries within region.**

Region/Country	Core	Minicore	df	$\chi^2$	P
<b>Americas</b>	24	5	1	2.0035	0.1569
Brazil	1	1	1	1.3489	0.2455
Mexico	1	1	1	1.3489	0.2455
United States of America	22	3	1	0.1193	0.7298
Heterogeneity			2	0.8136	0.6658
<b>North Africa</b>	3	3	1	13.9784	0.0002
Algeria	1	1	1	1.3489	0.2455
Morocco	1	1	1	1.3489	0.2455
Tunisia	1	1	1	1.3489	0.2455
Heterogeneity			2	9.9316	0.0070
<b>Southern Africa</b>	255	26	1	0.2186	0.6401
Botswana	7	1	1	0.0606	0.8055
Mozambique	3	1	1	0.0810	0.7760
Namibia	86	11	1	0.2010	0.6540
South Africa	15	1	1	0.2728	0.6015
Zambia	20	2	1	0.0247	0.8751
Zimbabwe	124	10	1	1.0734	0.3002
Heterogeneity			5	1.4949	0.9137
<b>Central Africa</b>	115	16	1	0.7716	0.3797
Cameroon	91	10	1	0.0028	0.9577
Central African Republic	11	2	1	0.4833	0.4869
Chad	11	2	1	0.4833	0.4869
Congo	1	1	1	1.3489	0.2455
Zaire	1	1	1	1.3489	0.2455
Heterogeneity			4	2.8956	0.5754
<b>East Africa</b>	187	19	1	0.1722	0.6782
Ethiopia	1	1	1	1.3489	0.2455
Kenya	11	2	1	0.4833	0.4869
Malawi	35	3	1	0.2123	0.6450
Somalia	1	1	1	1.3489	0.2455
Sudan	70	8	1	0.0040	0.9494
Tanzania	56	3	1	1.6961	0.1928
Uganda	13	1	1	0.1411	0.7072
Heterogeneity			6	5.0624	0.5358
<b>West Africa</b>	672	72	1	0.2510	0.6164
Benin	6	1	1	0.1619	0.6874
Burkina Faso	80	6	1	0.9668	0.3255
Cape Verde	1	1	1	1.3489	0.2455
Gambia	2	2	1	7.7143	0.0055
Ghana	37	4	1	0.0044	0.9472
Mali	112	12	1	0.0213	0.8841
Mauritania	1	1	1	1.3489	0.2455
Niger	110	15	1	0.5965	0.4399
Nigeria	236	22	1	0.4270	0.5135
Senegal	40	4	1	0.0494	0.8241
Sierra Leone	9	2	1	0.9829	0.3215
Togo	38	2	1	1.1884	0.2757
Heterogeneity			11	14.5596	0.2036
<b>Europe</b>	5	3	1	6.7446	0.0094
France	2	1	1	0.3421	0.5586
Germany	1	1	1	1.3489	0.2455
United Kingdom	2	1	1	0.3421	0.5586
Heterogeneity			2	4.7115	0.0948
<b>Oceania</b>	1	1	1	1.3489	0.2455
Australia	1	1	1	1.3489	0.2455
<b>South Asia</b>	788	88	1	0.0000	0.9951
India	774	86	1	0.0022	0.9625
Myanmar	1	1	1	1.3489	0.2455
Pakistan	13	1	1	0.1411	0.7072
Heterogeneity			2	1.4920	0.4743
<b>East Asia</b>	1	1	1	1.3489	0.2455
Union of Soviet Socialist Republics	1	1	1	1.3489	0.2455
<b>West Asia</b>	43	4	1	0.1349	0.7134
Lebanon	10	1	1	0.0124	0.9115
Yemen, Republic of	33	3	1	0.1282	0.7203
Heterogeneity			1	0.0057	0.9399
<b>Overall</b>	2094	238	45	38.6540	0.7363

**Table 2.**  $\chi^2$  value and probability for frequency distribution of classes in 10 qualitative traits in pearl millet [*Pennisetum glaucum* (L.) R. Br.] core and minicore collection accessions.

Character	df	$\chi^2$	P
Panicle shape	8	4.3660	0.8226
Panicle density	8	10.7313	0.2174
Bristle length	8	3.7539	0.8786
Synchrony of panicle maturity	6	2.2527	0.8951
Green fodder yield potential	7	7.1424	0.4142
Seed yield potential	6	11.0800	0.0859
Overall plant aspect	6	9.9851	0.1253
Seed shape	4	0.7147	0.9495
Seed color	9	13.0721	0.1594
Endosperm texture	7	8.1773	0.3172

## Mean, Median, and Variances

Differences between the means of the core and minicore collections were nonsignificant for all eight quantitative traits, resulting in 0 MD% (Table 3). There were no significant differences between the medians of the core and minicore for any of the quantitative traits as tested by the Wilcoxon (1945) rank-sum non-parametric test NPAR-1WAY procedure, indicating that the minicore represents the core collection for each of the eight quantitative traits.

The variances in the core and minicore collections were homogeneous ( $P = 0.060$  to  $0.737$ ) for all the quantitative traits, resulting in 0 VD% (Table 3). The zero value of MD% and VD% (<20%) indicated that the minicore adequately represented the core collection (Hu et al., 2000).

The data recorded on 30 representative plants in each accession were used to partition total variance into between- and within-accession variance. In the core, between- and within-accession variance component for all quantitative characters was highly significant, indicating sufficient variability (data not shown). However, in the case of number of total and productive tillers, panicle exertion, and thickness, the within-accession variance component was higher compared to between-accession variance, indicating presence of greater intra-accession variability for these traits. To compare the core and minicore, simple variances were obtained for each accession by using predicted means of each plant. Similar to the core collection, the minicore also showed greater within-accession variances for number of total and productive tillers, panicle exertion, and panicle thickness.

Between-accession variances were greater for days to flowering, plant height, and panicle length (data not shown). The within-accession variance range for each of the quantitative traits in the core and minicore was comparable. The mean values of within-accession variances were slightly higher in the minicore than the core for all the traits except panicle thickness, indicating that the minicore captured sufficient variability at both the levels (Table 4). The highly cross-pollinated nature of the crop coupled with varying degrees of self-pollination, uncertain environmental conditions, and farmers' preferences have played a major role during domestication of pearl millet, thus ensuring greater variability between and within accessions. Within-accession variation for most of the traits may also be the consequence of some sort of buffering mechanism adapted by different genotypes to different environmental conditions (Bradshaw, 1965). For example, intravarietal variation for flowering time would ensure that in case of a dry spell, all plants in a field will not be affected by drought in their most sensitive flowering stage (Hausmann et al., 2007). The pearl millet allogamous mating system in conjunction with farmers' seed management and regional exchange activities has also led to regionally similar but highly heterogeneous populations adapted to withstand the vagaries of extreme climatic conditions (vom Brocke et al., 2003). The enormous diversity creates challenges for genebank managers in terms of redundancy, regeneration, and conservation of a large number of accessions of this crop. The traits that show more within-accession variation will need careful planning for maintenance of the variability. The minicore, as representative of the core and entire collection, may help overcome some of these difficulties. To avoid inbreeding and loss of diversity during regeneration, a greater number of plants per accession need to be used for seed multiplication and subsequent conservation. Thus, a greater quantity of seed can be supplied to a researcher, ensuring delivery of the full spectrum of variability present within and between accessions.

## Shannon-Weaver Diversity Index

Shannon-Weaver diversity index is used to measure the allelic richness and evenness in the population. A low  $H'$  indicates an extremely unbalanced frequency class

**Table 3.** Range, means, medians, and variances in core and minicore collections of pearl millet [*Pennisetum glaucum* (L.) R. Br.].

Character	Range		Mean <sup>†</sup>		Median <sup>‡</sup>			Variance <sup>¶</sup>			
	Core	Minicore	Core	Minicore	Core	Minicore	P	Core	Minicore	F	P
Days to flowering	30.55–129.82	30.55–123.38	64.6a	64.9a	63.8	63.2	0.408	117.68	137.54	0.94	0.333
Plant height (cm)	45.86–411.08	89.59–372.78	258.1a	253.7a	260	255.1	0.130	1723.80	1646.62	0.75	0.388
Total tillers per plant (no.)	0.00–12.93	0.00–9.18	1.9a	2.0a	1.8	1.8	0.472	0.89	1.15	0.47	0.492
Productive tillers per plant (no.)	0.00–6.47	0.00–5.80	1.5a	1.5a	1.4	1.4	0.287	0.20	0.32	0.55	0.458
Panicle exertion (cm)	–10.83–14.71	–4.75–14.14	4.8a	5.0a	5.1	5.2	0.554	7.80	6.76	0.97	0.325
Panicle length (cm)	7.08–64.99	10.93–58.98	24.5a	24.1a	23.4	23.3	0.783	39.21	45.21	0.56	0.458
Panicle thickness (mm)	11.00–35.41	12.16–31.50	21.1a	21.3a	20.8	21.1	0.215	8.20	10.14	3.53	0.060
1000-seed weight (g)	2.85–19.30	3.00–19.30	8.7a	8.8a	8.5	8.7	0.149	4.54	4.76	0.11	0.737

<sup>†</sup>Differences between means of core and minicore were tested by Newman-Keuls test, and means followed by different letters are significantly different at  $P = 0.05$ .

<sup>‡</sup>Medians were tested by NPAR1WAY procedure in SAS.

<sup>¶</sup>Variance homogeneity tested by Levene's test at  $P = 0.05$ .

**Table 4. Range and mean of within-accession variance in pearl millet [*Pennisetum glaucum* (L.) R. Br.] core and minicore collections.**

Character	Minimum		Maximum		Mean	
	Core	Minicore	Core	Minicore	Core	Minicore
Days to flowering	0.46	1.21	541.59	541.59	30.72	31.26
Plant height (cm)	29.00	32.00	13635.00	13635.00	1483.72	1483.98
Total tillers per plant (no.)	0.03	0.03	79.43	79.43	1.79	2.36
Productive tillers per plant (no.)	0.03	0.03	20.55	20.55	0.59	0.75
Panicle exertion (cm)	1.43	1.43	231.96	204.08	26.38	27.44
Panicle length (cm)	0.88	4.05	469.93	469.93	36.33	37.28
Panicle thickness (mm)	1.95	2.86	143.03	143.03	17.29	17.07

for an individual trait and a lack of genetic diversity. The average  $H'$  for all 18 traits in the minicore collection ( $0.602 \pm 0.0195$ ) was comparable to that in the core collection ( $0.597 \pm 0.0171$ ), suggesting that the minicore has captured adequate diversity from the core collection (Table 5). Endosperm texture had the highest  $H'$  values of 0.770 and 0.805 in the core and minicore collection, respectively. Bristle length had the lowest  $H'$  values of 0.447 and 0.444 in the core and minicore collection, respectively.

The VR% compares the coefficient of variation values of the traits measured in the core collection with the minicore in general and determines how well the variance is being represented in the minicore. The CR% indicates whether the distribution ranges of each trait in the minicore are well represented when compared to the core collection. Core collection with low MD% and VD% (<20%) and large CR% (>80%) and VR% (>100%) are considered to provide a good representation of the genetic diversity of the initial collection (Hu et al., 2000). The coefficient of variation values were higher in the minicore than in the core collection, resulting in a high VR% of 109.3 for the quantitative traits (Table 5). The high CR% captured for quantitative traits (83.4%) in the minicore indicated homogeneous distribution ranges of the traits and that it was representative of the core collection.

### Trait Associations

Phenotypic correlations were calculated among eight quantitative traits in the core and minicore collection separately (Table 6). In the core collection, all the correlations were significant at  $P = 0.01$  except between days to flowering and 1000-seed weight and between panicle length and thickness. In the minicore, the pattern of association between most of the trait combinations was similar to that observed in the core. Further, the proportion of variance in one trait that can be attributed to its linear relationship with a second trait is indicated by the square of the correlation coefficient (Snedecor and Cochran, 1980). Estimates of correlations  $\geq 0.707$  or  $\leq -0.707$  have been suggested as meaningful correlations (Skinner et al., 1999). In the present study, we found such high correlation between number of total and productive tillers in the core ( $r = 0.890$ ) as well as minicore ( $r = 0.901$ ), respectively. A few other important correlations in the core as well as minicore were: negative correlation between panicle length and exertion, positive association between panicle length and plant height,

negative correlation between panicle thickness and number of total and productive tillers, and positive association between panicle thickness and 1000-seed weight.

The pearl millet minicore developed in this study captured almost the full diversity of the core collection for all the traits. The minicore provides an opportunity to identify phenotypic variability in preliminary analyses of a more modest-sized sample. For pearl millet, specifically, the minicore collection can be extensively screened for desirable alleles in multiple environments for traits such as pathotype-specific downy mildew resistance and heat and drought tolerance; major agronomic traits; for biochemical components of human nutrition (minerals such as iron and zinc and vitamins, etc.), industrial use (starch, sugar, etc.), and livestock feed (phenols, tannins, etc.); and for identification of maintainers and fertility restorers of male sterility of different cytoplasmic male sterility systems. For example, this minicore captured two downy mildew resistant accessions (IP 21187 and IP 21201), four

**Table 5. Shannon-Weaver diversity ( $H'$ ) index, coincidence rate (CR%), and variable rate (VR%) in pearl millet [*Pennisetum glaucum* (L.) R. Br.] core and minicore collections.**

Character	$H'$		CR%	VR%
	Core	Minicore		
<b>Quantitative</b>				
Days to flowering	0.609	0.591	93.5	107.4
Plant height (cm)	0.616	0.618	77.5	108.2
Total tillers per plant (no.)	0.518	0.499	71.0	110.7
Productive tillers per plant (no.)	0.540	0.488	89.6	136.7
Panicle exertion (cm)	0.620	0.621	74.0	90.4
Panicle length (cm)	0.558	0.546	83.0	109.0
Panicle thickness (mm)	0.619	0.619	79.2	110.2
1000-seed weight (g)	0.613	0.566	99.1	102.0
Mean—quantitative	0.586	0.569	83.4	109.3
SE	0.0145	0.0189	3.491	4.567
<b>Qualitative</b>				
Panicle shape	0.566	0.633	—†	—
Panicle density	0.640	0.651	—	—
Bristle length	0.447	0.444	—	—
Synchrony of panicle maturity	0.664	0.678	—	—
Green fodder yield potential	0.628	0.647	—	—
Seed yield potential	0.536	0.564	—	—
Overall plant aspect	0.537	0.576	—	—
Seed shape	0.692	0.690	—	—
Seed color	0.574	0.608	—	—
Endosperm texture	0.770	0.805	—	—
Mean—qualitative	0.605	0.630	—	—
SE	0.0292	0.0297	—	—
Mean—all	0.597	0.602	—	—
SE	0.0171	0.0195	—	—

† = not estimated

**Table 6. Phenotypic correlation coefficients between eight quantitative traits in the pearl millet [*Pennisetum glaucum* (L.) R. Br.] core (below diagonal) and minicore (above diagonal) collections.†**

Character	Days to flowering	Plant height	Total tillers	Productive tillers	Panicle exertion	Panicle length	Panicle thickness	1000-seed weight
Days to flowering		0.227	-0.117	-0.077	-0.248	0.091	0.138	-0.001
Plant height (cm)	0.220		-0.102	-0.150	-0.051	0.314	0.134	0.183
Total tillers per plant (no.)	-0.171	-0.071		0.901	0.146	-0.220	-0.312	-0.202
Productive tillers per plant (no.)	-0.121	-0.076	0.890		0.070	-0.197	-0.254	-0.182
Panicle exertion (cm)	-0.242	-0.077	0.195	0.143		-0.353	-0.222	-0.061
Panicle length (cm)	0.119	0.309	-0.196	-0.177	-0.498		0.031	0.099
Panicle thickness (mm)	0.099	0.207	-0.268	-0.229	-0.138	0.029		0.287
1000-seed weight (g)	0.004	0.131	-0.159	-0.133	-0.104	0.099	0.264	

†Correlation coefficients >0.050 and 0.036 are significant at  $P = 0.01$  and  $0.05$ , respectively, in the core collection at df 2092, and correlation coefficients >0.150 and 0.107 are significant at  $P = 0.01$  and  $0.05$ , respectively, in the minicore at df 236.

ergot-resistant accessions (IPs 21244, 21283, 21312, and 21438), five smut-resistant accessions (IPs 19722, 19816, 19851, 19913, 19964), and an early-flowering (IP 7846) and a high-tillering accession (IP 22269). The minicore with a manageable number of accessions can be used for combining-ability studies of economic traits (nonobservable characters) (Frankel and Brown, 1984b), thus adding a new and important facet to the evaluation and subsequent use of germplasm collection. This will lead to greater use of diverse germplasm for discovery of new genes, especially in the context of climate change scenario. Furthermore, additional sources of new genes can be found in the larger core collection or the entire collection by selectively examining the clusters from which the minicore accessions have been selected for a particular trait. Minicore collections have been evaluated to identify new sources for important biotic stresses such as multiple disease resistance in chickpea (Pande et al., 2006), rust and late leaf spot resistance in groundnut (Kusuma et al., 2007), grain mold and downy mildew resistance in sorghum (Sharma et al., 2010), and abiotic stresses such as drought tolerance in chickpea (Kashiwagi et al., 2005) and groundnut (Upadhyaya, 2005); salinity tolerance in chickpea (Vadez et al., 2007), groundnut, and pigeonpea (Srivastava et al., 2006, 2007). The minicore collections have also been evaluated for agronomic traits of importance in various crops such as chickpea, groundnut, and pigeonpea and are being made more meaningful by addition of molecular marker data for different traits (summarized in Upadhyaya, 2008; Upadhyaya et al., 2009c). The minicore collections can also be used to study the population structure and linkage disequilibrium in crops, a prerequisite for carrying out association mapping studies (Wang et al., 2008). In conclusion, the minicore of 238 accessions constituted in this study is a good representative of the ICRISAT pearl millet core collection of 2094 accessions as well as the entire collection of 20,766 cultivated accessions. The research community can avail itself of limited seeds of the pearl millet minicore accessions free of charge from the genebank at ICRISAT, Patancheru, India, following the terms and conditions of the Standard Material Transfer Agreement.

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