Genetic Resources and Crop Evolution (2006) 53: 679–685
DOI 10.1007/s10722-004-3228-3

Development of core subset of finger millet germplasm using geographical origin and data on 14 quantitative traits

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Received 10 May 2004; accepted in revised form 13 September 2004

Key words: Core subset, Correlation coefficients, Diversity index, *Eleusine coracana*, Finger millet

Abstract

Finger millet [*Eleusine coracana* (L.) Gaertn.] is an important cereal food crop in Africa and South Asia. It is a hardy crop that can be grown in very diverse environments from almost at sea level to about 2400 m.a.s.l. Finger millet has an excellent food value as its seeds contain protein ranging from 7 to 14% and are particularly rich in methionine amino acid, iron, and calcium. Despite all these merits, this crop has been neglected from the main stream of crop improvement research. One of the means to boost its production and productivity is to enhance utilization of finger millet germplasm to breed superior varieties. Keeping this objective in view, a core subset of finger millet germplasm (622 accessions) based on origin and data on 14 quantitative traits was developed from the entire global collection of 5940 accessions held in the genebank at ICRISAT, Patancheru, India. The comparison of means, variances, frequency distribution, Shannon-Weaver diversity index (H′) and phenotypic correlations indicated that the core subset represents the entire collection. These tests indicated that sampling was optimal and the diversity has been captured very well in the core subset. The correlation analysis indicated that panicle exsertion and longest finger length could be given lower priority in the future germplasm evaluation work of finger millet.

Introduction

Finger millet or ragi, *Eleusine coracana* (L.) Gaertn. is cultivated for human food in Africa and South Asia. Precise global area under finger millet is not known because this crop had often been clubbed with other millets. Global area under millets is 36.29 m ha (FAO 2003). The Consultative Group on International Agricultural Research (CGIAR) has estimated that 10% of the area under millets is with finger millet (www.cgiar.org/research/res_millet.html). Information is little better on some countries. For example, the crop was grown on 194,000 ha in Nepal in 1990, 11,000 ha in Sri Lanka in 1988, 227,000 ha in Ethiopia in 1986 (Riley et al. 1993), and 1.68 million ha in India in 2001–2002 (CMIE Feb. 2004). There are some more countries, namely, Uganda, Rwanda, Zaire, Kenya, Eritrea, Somalia, China, and Myanmar where finger millet is a common crop. Finger millet was domesticated about 5000 BC in Eastern Africa (possibly Ethiopia) and introduced into India as a crop 3000 years ago (Hilu et al. 1979). The closest wild relative of finger millet is *Eleusine coracana* subsp. *africana* (Kennedy-O'Byrne) Hilu and de Wet. *Eleusine coracana* subsp. *africana* is a native to Africa. These two taxa (finger millet and subsp.

africana) hybridize where they are sympatric in Africa and derivatives of such crosses often occur as weeds in cultivated fields.

Finger millet can perform better under adverse soil and weather conditions compared to other crops. Its climatic requirements match with upland paddy crop. The crop has a wide range of seasonal adaptation, and is grown in lands almost at sea level (in parts of Andhra Pradesh and Tamil Nadu, India) to about 2400 m.a.s.l. in hills of Uttranchal (India), and similarly at high altitudes in Uganda, Kenya and Ethiopia. In a trial of nine summer cereal species (rice, job's tears, pearl millet, sorghum, maize, common millet, barnyard millet, foxtail millet, and finger millet) finger millet was most resistant to water logging, except rice (Kono et al. 1988). Its reported yield potential is 4265 kg ha^{-1} in Uganda (Odelle 1993), 6060 kg ha^{-1} in Zimbabwe (Mushonga et al. 1993), 3700 kg ha^{-1} in Ethiopia (Mulatu and Kebebe 1993), and 4789 kg ha^{-1} in India (Bondale 1993).

Finger millet seeds are consumed in variety of forms, such as unleavened bread (roti), thin- or thick porridge, fermented porridge or extensively used in brewing. Finger millet food has high biological value. Seed protein content is about 7.4%, which is comparable to that of rice. However, some lines have been noted having as much as 14.2% protein (Iyengar et al. 1945–1946). Finger millet seeds are particularly rich for tryptophan, cystine, methionine, and total aromatic amino acids compared to other cereals. The seeds are exceptionally rich in calcium containing about 0.34% in whole seed compared with 0.01–0.06% calcium in most cereals (Kurien et al. 1959). The seeds are also rich in iron containing 46 mg kg^{-1} (Serna-Saldivar and Rooney 1995), which is much higher compared to wheat and rice.

Despite so many merits, finger millet has remained neglected from the main stream of crop improvement research compared to the cereals like maize, rice, and wheat. One of the means to make the crop competitive is to breed superior and high yielding cultivars utilizing diverse germplasm resources. The genebank at ICRISAT, Patancheru, India is holding 5940 accessions of finger millet from 23 countries. These accessions should be evaluated for various agronomic traits including yield at number of locations in replicated trials. However, this is a costly exercise due to large size of collection and financial constraints. To overcome this problem, Frankel (1984) proposed sampling of the collection to a manageable sample or 'core collection'. A core collection contains a subset of accessions from entire collection that captures most of available diversity of species (Brown 1989a). The core subset thus formed can be evaluated extensively and the information derived could be used to guide more efficient utilization of the entire collection (Brown 1989b). Core subsets of entire germplasm collections have been established in number of cereal crops, namely, barley (Knüpffer and van Hintum 1995), sorghum (Prasada Rao and Ramanatha Rao 1995; Grenier et al. 2001), maize (Taba et al. 1994, 1998), wheat (Zeuli and Qualset 1993), quinoa (Ortiz et al. 1998), and pearl millet (Bhattacharjee 2000), to name only some. The objective of this study was to develop a core subset of finger millet germplasm accessions using data on geographical origin and quantitative traits.

Materials and methods

The finger millet germplasm collection in the ICRISAT, Patancheru genebank (5940 accessions) is the largest collection compared to any other finger millet collection in the world. These collections were characterized at ICRISAT, Patancheru research farm over the years from 1974 to 2003. The characterization site is located at 18°N and 78°E, at an altitude of 545 m, and about 600 km from the sea. Annual rainfall is about 750 mm, most of which occurs during June–September. The germplasm accessions were sown on red soils (alfisols), on 60 cm apart ridges, each accessions occupying single row of 4 m length, spacing being 60×10 cm. A doze of 20 kg nitrogen and 50 kg phosphorus ha^{-1} was applied as basal fertilizers and 45 kg nitrogen as top dressing. In all the years, sowings were done towards end of July. Irrigation and hand weeding were done when necessary. In all the germplasm sets grown over the years, crop was reasonably free from any disease or insect damage and no chemical sprays were applied.

Data was recorded on eight qualitative (description in discrete classes) and 14 quantitative (continuous variation) traits following the Descriptors of Finger Millet (IBPGR 1985). The data on all the eight qualitative traits (plant pigmenta-

tion, growth habit, inflorescence compactness, glume prominence, seed color, lodging, senescence, and overall disease free 'score' and one quantitative trait (days to 50% flowering) were recorded on plot basis. Data on basal tiller number was taken on five representative plants of the plot. Data for the remaining 12 quantitative traits: plant height (mm), number of culm branches, flag leaf blade length (mm), flag leaf blade width (mm), flag leaf sheath length (mm), peduncle length (mm), panicle exsertion (mm), inflorescence length (mm), inflorescence width (mm), longest finger length (mm), longest finger width (mm) and number of panicle branches was recorded on main culms of the five representative plants of the plots. During field evaluation, accessions were also classified into botanical races.

The entire germplasm collection of finger millet was stratified into four regions: Africa, Asia, America, and Europe. The information on country of origin was not available on 181 accessions. They were grouped into 'unknown' region. The data recorded on the 14 quantitative traits in each group was standardized using range of each variable to eliminate scale differences. The principal component analysis (PCA) was performed on the accessions from each region. This analysis provides a reduced dimension model that would indicate measured differences among the groups. The variation captured by first five scores in the five regions ranged from 61.24 to 99.28%. A hierarchical cluster analysis (Ward 1963) was conducted on first five scores in each region separately. From each cluster, about 10% accessions were randomly picked to form the core subset. If number of accessions in a cluster was <10, one accession was picked to be included in the core subset.

The frequency distribution of accessions according to regions, country of origin, botanical races, and biological status in the entire collection and the core subset was analyzed by the χ^2-test. The means of the entire collection and core subset were compared using Newman–Keuls procedure (Newman 1939; Keuls 1952) for all the 14 quantitative traits. The homogeneity of variances of the entire collection and core subset was tested with Levene's test (Levene 1960). The percentage of significant differences between the entire collection and the core subset were calculated for the coefficient of variation (%) and the range % (Hu et al. 2000). The diversity index (H′) of Shannon and Weaver (1949) was estimated and used as a measure of phenotypic diversity in the entire collection and core subset for each trait. The phenotypic correlations between the traits in entire collection and core subset were estimated separately. This was done to know to what extent the trait associations (presumed to be under genetic control) were captured in the core subset.

Results and discussions

The 5940 accessions in the entire collection were grouped into 104 clusters. Region wise the number of clusters was 42 in Africa, 32 in Asia, 5 in America, 7 in Europe, and 18 in the unknown group. The number of accessions in individual clusters ranged from 1 (0.017%) to 213 (3.58%). The number of clusters with 1–30 accessions was 39, and for each of the three subsequent classes i.e., 31–60, 61–90, and 91–120 accessions, number of clusters was 19. For the remaining 8 clusters, the number of accessions ranged from 123 to 213. Following the procedure mentioned above, a core subset of finger millet consisting of 622 accessions was formed from the entire collection of 5940 accessions held in the genebank at ICRISAT, Patancheru. The core subset constituted 10.47% of the entire collection. In the core subset, Africa region was represented by 365 accessions (58.68% of core subset), Asia by 223 accessions (35.85%), America by 5 accessions (0.80%), and Europe by 7 accessions (1.13%). The 'unknown' region was represented by 22 accessions (3.54% of the core subset). The accessions from Africa, Asia, and those from unknown origin were represented adequately in the core subset and corresponded well with their number in entire collection. The accessions from America and Europe were over represented in the core subset (Table 1). America region had 7 accessions and Europe had 22 accessions. However, clustering based on data on 14 quantitative traits indicated 5 clusters for American and 7 clusters for European accessions. Including at least one accession from each of the clusters resulted in higher (significant) representation in core subset from these two regions. Individual countries within each region were represented adequately and corresponded very well with the number in the entire collection

Table 1. Number and percentage of accessions in different regions, and countries within region, of entire collection and core subset of finger millet germplasm.

Class	Entire collection		Core subset		χ^2	p
	Number	Percentage	Number	Percentage		
Region/Country						
Africa	3567	60.0[a]	365	58.68	0.194	0.660
Burundi	15	0.42	3	0.82	0.607	0.436
Cameroon	7	0.20	1	0.27	0.065	0.798
Ethiopia	31	0.87	3	0.82	0.142	0.706
Kenya	946	26.52	107	29.32	1.075	0.300
Malawi	252	7.06	25	6.85	0.024	0.877
Mozambique	1	0.03	1	0.27	1.545	0.214
Nigeria	19	0.53	5	1.37	3.360	0.067
Senegal	5	0.14	1	0.27	0.000	0.987
South Africa	1	0.03	1	0.27	1.545	0.214
Tanzania	41	1.15	3	0.82	0.685	0.408
Uganda	958	26.86	81	22.19	2.958	0.085
Zaire	2	0.06	1	0.27	0.426	0.514
Zambia	136	3.81	21	5.75	3.606	0.058
Zimbabwe	1153	32.32	112	30.68	0.303	0.582
Asia	2163	36.41	223	35.85	0.054	0.816
India	1361	62.92	149	66.82	0.537	0.464
Maldives	4	0.18	1	0.45	0.019	0.892
Nepal	779	36.01	70	31.39	1.324	0.250
Pakistan	1	0.05	1	0.45	1.528	0.216
Sri Lanka	18	0.83	2	0.90	0.068	0.794
Americas (USA)	7	0.12	5	0.80	19.359	0.000
Europe	22	0.37	7	1.13	7.644	0.006
Germany	1	4.55	1	14.29	0.104	0.747
Italy	7	31.82	3	42.86	0.033	0.855
United Kingdom	14	63.64	3	42.86	0.858	0.354
Unknown	181	3.05	22	3.54	0.490	0.484

[a]Figures given in this column are percentage of accessions in a region over total accessions, and in a country within region.

(Table 1) indicating that the sampling technique to constitute the core subset was optimal.

Of the five races of finger millet genetic resources (de Wet et al. 1984), the germplasm is most represented by race *vulgaris* (61.16%), followed by *plana* (16.99%), *compacta* (11.40%), *elongata* (8.55%), and race *africana* (1.90%). The first four races are cultivated whereas the fifth race, *africana* is a wild form. The representation of races in the core subset was similar to that in the entire collection (Table 2). The frequency distribution of accessions in the entire collection and core subset was also compared for their agricultural status. All the four groups, namely, improved cultivars, breeding materials, landraces, and the wild types were represented similarly in both the entire collection and core subset (Table 2) indicating optimal composition of the core subset.

Differences between means of the core subset and entire collection were non-significant for all the quantitative traits used in developing core collection (Table 3). The homogeneity test of the variances between core subset and the entire collection revealed that variances for all the 14 quantitative traits were homogeneous. The coefficient of variation (CV%) and the range (%) of the 14 traits are indicative of how precisely the variability for these traits has been captured in the core subset from the entire collection. For the quantitative traits, CV (%) ranged from 100.10 to 130.97, indicating a good representation in the core subset. So was the sampling of range of characteristics for various traits. The values ranged from 70.59 to 100.0%. This indicated that for these two characteristics also the core subset was adequately representative of the entire collection (Table 3). The χ^2 values of the comparison of frequency distri-

Table 2. Number and percentage of accessions belonging to different races and biological status in entire collection and core subset of finger millet germplasm.

Class	Entire collection		Core subset		χ^2	p
	Number	Percentage	Number	Percentage		
Race						
africana	113	1.90	16	2.57	1.470	0.225
compacta	677	11.40	75	12.06	0.238	0.626
elongata	508	8.55	50	8.04	0.192	0.661
plana	1009	16.99	102	16.40	0.127	0.722
vulgaris	3633	61.16	379	60.93	0.005	0.942
Agricultural Status						
Improved cultivar	135	2.27	12	1.93	0.323	0.570
Breeding line	50	0.84	4	0.64	0.292	0.589
Landrace	5642	94.98	590	94.86	0.001	0.974
Wild	113	1.90	16	2.57	1.468	0.226

Table 3. Comparison of means, variances, coefficient of variation (CV %), and range (%) for the quantitative traits in the entire collection and core subset of finger millet germplasm.

Characters	Mean			Variances				CV(%)	Range (%)
	Entire collection	Core subset	Differences[a]	Entire collection	Core subset	F Value	p[b]		
Days to 50% flowering	80.4 ± 0.11	80.0 ± 0.35	NS	72.68	74.62	0.02	0.890	101.78	82.86
Plant height (mm)	100.7 ± 0.24	100.7 ± 0.77	NS	337.88	394.06	1.17	0.280	108.01	95.24
Basal tillers (no.)	5.2 ± 0.04	5.3 ± 0.16	NS	9.82	15.52	1.94	0.164	123.09	100
Culm branching (no.)	2.3 ± 0.02	2.2 ± 0.05	NS	1.76	1.84	0.14	0.706	105.67	71.43
Flag leaf blade length (mm)	358.1 ± 0.98	356.8 ± 3.05	NS	5741.09	5708.52	0.0001	0.995	100.10	92.31
Flag leaf blade width (mm)	12.6 ± 0.04	12.7 ± 0.12	NS	8.54	8.86	0.60	0.439	101.25	100
Flag leaf sheath length (mm)	102.5 ± 0.25	102.2 ± 0.78	NS	358.46	379.40	0.27	0.604	103.16	70.59
Peduncle length (mm)	215.4 ± 0.57	215.5 ± 1.87	NS	1947.27	2178.32	2.18	0.140	105.75	91.44
Panicle exsertion (mm)	113.5 ± 0.51	113.4 ± 1.62	NS	1551.06	1623.10	0.30	0.585	102.37	76.39
Inflorescence length (mm)	93.1 ± 0.36	94.0 ± 1.21	NS	772.51	913.46	2.31	0.128	107.68	90.32
Inflorescence width (mm)	78.4 ± 0.40	79.3 ± 1.31	NS	953.59	1672.42	0.13	0.720	130.97	95.24
Longest finger length (mm)	72.6 ± 0.28	73.3 ± 0.93	NS	473.86	529.90	1.02	0.312	104.82	95.83
Longest finger width (mm)	11.6 ± 0.03	11.5 ± 0.10	NS	6.73	8.86	0.07	0.797	115.22	100
Panicle branches (no.)	7.7 ± 0.02	7.7 ± 0.07	NS	3.07	3.72	0.04	0.851	111.25	100
Mean								108.65	90.12

[a] Differences between means of entire and core collection were tested by Newman–Keuls test; NS indicates significant at $p = 0.05$.
[b]Variances were tested by Lenene's test.

bution of the eight qualitative traits (data not given) were non-significant ($p = 0.308$–0.695), indicating high precision of the sampling technique followed.

The Shannon–Weaver diversity index (H′) was calculated to compare the diversity for the phenotypic characters in the core subset and entire collection. The index is used in genetic studies as a convenient measure of both allelic richness and allelic evenness. A low H′ indicates an extremely unbalanced frequency of classes for an individual trait and a lack of genetic diversity. The average H′ for the eight qualitative traits and 14 quantitative traits in the core subset and the entire collection were similar (Table 4), indicating that diversity of entire collection was well represented in the core subset.

An adequate and proper sampling, essential in developing a representative core subset, should consider the sampling of phenotypic associations arising out of co-adapted gene complexes (Ortiz et al. 1998). Phenotypic correlations were calculated between the 14 quantitative traits in core subset as well as entire collection, separately (data not given). In the entire collection, 79 correlations were significant at $p = 0.05$ whereas in core subset,

Table 4. Shannon diversity index in the entire collection and core collection for various qualitative and quantitative characters in finger millet.

Characters	Entire collection	Core collection
Qualitative traits		
Plant pigmentation	0.270	0.271
Growth habit	0.279	0.286
Inflorescence compactness	0.635	0.642
Glume prominence	0.445	0.453
Fruit color	0.508	0.519
Lodging	0.650	0.649
Senescence	0.490	0.480
Disease resistance	0.468	0.459
Mean ± SE	0.468 ± 0.050	0.470 ± 0.050
Quantitative traits		
Days to 50% flowering	0.632	0.621
Plant height (mm)	0.635	0.619
Basal tillers (no.)	0.496	0.496
Culm branching (no.)	0.613	0.596
Flag leaf blade length (mm)	0.621	0.612
Flag leaf blade width (mm)	0.411	0.402
Flag leaf sheath length (mm)	0.579	0.595
Peduncle length (mm)	0.634	0.632
Panicle exsertion (mm)	0.623	0.624
Inflorescence length (mm)	0.575	0.571
Inflorescence width (mm)	0.525	0.549
Longest finger length (mm)	0.606	0.578
Longest finger width (mm)	0.403	0.420
Panicle branches (no.)	0.563	0.562
Mean ± SE	0.565 ± 0.021	0.563 ± 0.020
Over all Mean ± SE	0.530 ± 0.024	0.529 ± 0.023

only 45 were significant and remaining 34 non-significant. This indicated a fairly good representation of co-adapted gene complexes from entire collection to the core subset. However, the reverse was true in one combination (inflorescence length vs. culm branches number) in which correlation was significant in core subset (−0.110) but was non-significant in the entire collection (−0.005).

Data on character associations could be used to identify a few traits, which are less relevant and could be of low priority in the germplasm evaluation. This will simplify the work and save resources. In the present experiment, of the 91 character associations estimated, two associations: peduncle length vs. panicle exsertion (*r* value 0.818 for core subset and 0.839 for entire collection) and longest finger length vs. inflorescence length (*r* value 0.799 for core subset and 0.795 for entire collection) had reasonably high and similar estimates. This indicates that in the future characterization of finger millet germplasm, only peduncle length, and inflorescence length could be used. Both of these traits are easier to measure than the panicle exsertion and longest finger length.

Finger millet germplasm possess vast diversity for agronomic traits. For example when a germplasm collection of finger millet was evaluated at ICRISAT, Patancheru farm, the accessions revealed a range of 54–120 days for flowering time, 45–165 cm for plant height, 1–70 tillers plant^{-1} and inflorescence length from 1–32 cm (Prasada Rao and de Wet 1997). Presumably, much of this diversity has been captured in the core subset of 622 accessions that can be used effectively as a starting point for research projects involving screening of the germplasm collection to identify sources of desirable traits. The information on clusters to which particular accessions with traits of interest belong will assist in looking extensively for more accessions with similar traits. The core subset should be evaluated extensively under the potential environments and the sources for economic traits identified must find their place in the finger millet improvement programs. The composition of the core subset should not be considered as permanent entity, rather it should be dynamic as additional accessions and information become available.

The list of finger millet accessions included in the core subset and seeds can be obtained on request, free of charge from the genebank at ICRISAT, Patancheru, India.

The information can also be accessed from ICRISAT (www.icrisat.org) and singer (www.singer.cgiar.org) websites.

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