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Genetic Diversity of Finger Millet [*Eleusine coracana* (L.) Gaertn] Landraces Characterized by Random Amplified Polymorphic DNA Analysis

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Abstract

Sixty-six finger millet accessions composed of 64 landraces and two improved varieties were evaluated using RAPD markers to study the genetic diversity in finger millet landraces. The study was conducted at the Laboratory of Department of Genetics, Faculty of Science, Kasetsart University, Bangkok, Thailand. The RAPD analysis was executed using 15 primers, which were screened based on their effectiveness to discriminate among the accessions. Among the 123 RAPD fragments amplified, 89 (72.35%) were polymorphic. The polymorphic information content (PIC) ranged from 0 to 0.50 with heterogeneous distribution and about 23% of the markers with a high discrimination power of ≥ 0.30. Genetic similarity between accessions estimated with simple matching coefficients ranged from 0.585 to 0.984. The RAPD cluster analysis successfully separated all accessions with the highest similarity value of approximately 98%. The 66 accessions were grouped into nine clusters at similarity index of approximately 0.83; however, there was no clear-cut separation among finger millet accessions in relation to the origin of their respective region. The result of the present study revealed the existence of ample variability in finger millet landraces that could be employed in the genetic improvement.

Key words: Finger millet, *Eleusine coracana*, Random amplified polymorphic DNA, Genetic diversity

1. Introduction

Ethiopia represents one of the major countries of genetic diversity. Eleven crops including finger millet (*Eleusine coracana*) are indicated as having their center of diversity in Ethiopia (Zonary 1970). The long history of cultivation and the large agro-ecological and cultural diversity in the country have resulted in large number of landraces of finger millet. Despite its importance as food and feed, the cultivation of finger millet is being pushed to the more marginal areas; therefore, genetic erosion is believed to occur due to several factors. Nevertheless, germplasm collection has been undertaken from different areas of the country and some reports indicated that about 2,051 finger millet germplasm accessions have been collected and preserved.

A fundamental goal of germplasm collection and conservation is the understanding of genetic relationships within and between the species of concern. A good understanding is critical for the effective organization and management of such germplasm collection. In plant breeding programs, estimates of genetic relationships can be useful for identification of parents for hybridization, and for reducing the number of accessions needed to maintain a broad range of genetic variability. Evaluation of numerous, highly similar accessions not only waste the plan of breeding resources but likely reduces the chance of identifying the truly unique and valuable accessions. To identify genetic materials that may contain useful traits for germplasm enhancement, and to understand the relationships among accessions and their corresponding collecting-site environments a systemic evaluation of genetic diversity is required (Steiner & Greene 1996). Comparison of parents using differences in DNA markers such as random amplified polymorphic DNA (RAPD) may be one of the method by which breeders can increase the probability of selecting those

parents with different gene sets.

Williams *et al*. (1990) proposed the use of random amplified polymorphic DNAs (RAPDs) as an additional form of molecular marker. This methodology offers the promise of virtually unlimited markers in plant species (Welsh & McClelland 1990; Williams *et al*. 1990). The power of RAPD technology lies in the following attributes; (1) no previous knowledge of the genome is required; (2) rapid results are obtained, especially when compared with the procedure involved in restricted fragment length polymorphism (RFLP) analysis; and (3) a universal set of random (arbitrary) primers can be used for genomic analysis of any organism (Welsh & McClelland 1990; Williams *et al*. 1990). This technology already applied to a wide range of plant species including maize (Welsh *et al*. 1991), tomato (Klein-Lankhorse *et al*. 1991), papaya (Stiles *et al*. 1993) *Panicum* millet (M'Ribu & Hilu 1994), sweat potato (Connolly *et al*. 1994), genus *Elusine* (Salimath *et al*. 1995), *Triticum* accessions (Cao *et al*. 1999) and finger millet (Fakrudin *et al*. 2004) .

A considerable number of diversity studies have been also done in Ethiopia using different markers on different crops; however, a study in finger millet was limited. Hence, this study was conducted to determine the level of DNA diversity, as measured with RAPD markers, among 66 finger millet accessions collected from Ethiopia and Eritrea. By understanding the genetic makeup, breeders will be better able to identify and exploit diversity in their programs.

2. Materials and methods

2.1 Plant materials

Sixty-six finger millet accessions consisting of 64 landraces and two improved varieties (Padet and Tadesse) were used in this study. They landraces were collected from five former regions of Ethiopia (Tigray, Gonder, Gojam, Welega, and Gamo Gofa) and Eritrea covering different agro-ecologies (Table 1).

2.2 DNA extraction

The experiment was conducted at the Laboratory of Department of Genetics, Faculty of Science, Kasetsart University, Bangkok, Thailand. The seedlings of 66 accessions were grown in pot for three weeks and the leaves of each accession were harvested separately, and then washed and ground into a fine powder in liquid nitrogen and used directly for DNA extraction following the protocol of Dellaporta *et al.* (1983) as modified by Rueda *et al.* (1998).

2.3 Polymerase chain reaction (PCR) amplification and electrophoresis

RAPD primers were screened based on their ability to detect distinct and clearly resolved amplified products. PCR amplification and electrophoresis were undertaken to identify the polymorphism among the accessions. The PCR was performed in a volume of 25 µl containing 2.5 µl of 10x PCR buffer, 1.0 µl of 50 mM magnesium chloride, 0.5 µl of each of the 2.5 mM deoxyribonucleotide triphosphates (dATP, dCTP, dGTP, dTTP), 1.5 µl of a single PCR primer, 1.25 U of *Taq* DNA polymerase, approximately 15 ng of genomic DNA template and 16.75 µl ddH_2O. Amplification was carried out in programmable thermal cycler for one initial denaturation cycle at 94^0C for 2 minutes followed by 40 cycles at 94^0C for 30 seconds for denatuation, at 36^0C for 30 seconds for annealing, at 72^0c for 1 minute for extension and at 72^0C for 3 minutes for final extension, then held at 4^0C for 5 minutes. On completion of the cycles the samples were stored at 4^0c before electrophoresis. The PCR amplified products were separated out through 1% agarose and visualized through staining with ethidium bromide and photographed under UV light.

2.4 Data analysis

Data were recorded as presence (1) or absence (0) of DNA band from the examination of photographic negatives. Each amplified fragment was named by the source of the primer, the kit number and its approximate size in base pairs. The percent of polymorphism was calculated as P%= (P/n) x 100, where: P is the number of polymorphic bands and n is the total number of bands produced from the reaction. The polymorphism information content (PIC) of each fragment was determined as described by Weir (1996) as $PIC=1-\sum P^2i$. Where: Pi is the frequency of the i^{th} allele in the observed accessions. Genetic similarity between pairs of accessions was estimated using simple matching coefficient (Sneath & Sokal 1973), SM= (a+d)/(a+b+c+d). Where: a= number of 1, 1 matches, b= number of 1, 0 matches, c= number of 0, 1 matches and d= number of 0, 0 matches. Cluster analysis was carried out on similarity estimates using

unweighted pair-group methods, arithmetic average (UPGMA) and the resulting cluster was represented as dendrogram. Data were analyzed using NTSYS-pc version 2.01 computer program (Roholf 1997).

3. Results and discussion

3.1 Primer screening

A total of 164 RAPD primers were screened for amplification and 111 primers generated amplified bands, while 53 primers did not generate amplified bands. Based on consistent production of strong amplification and uniform reproducible products, 23 primers were chosen and used for further RAPD analysis in all the 66 accessions. Among the 23 primers employed for RAPD analysis, five of them showed monomorphic bands across all accessions and another three were not consistent. As a result, the remaining 15 primers (Table 2) were used for the genetic analysis.

3.2 RAPD polymorphism and power of discrimination

Using the 15 RAPD primers, the RAPD analysis generated a total of 123 RAPD fragments (Table 2 & 3). The band size, detected by comparing the amplified fragments with Gene Ruler^{TM} DNA Ladder mix ranged from about 200 bp (OPD-12) to 2400 bp (OPA-18) in length. The RAPD markers were found to be informative markers in assessing the genetic diversity of 66 finger millet accessions.

Each primer produced a different set of polymorphism with average number of 8.2±2.833 bands per primer (Table 2 & 3). Primer OPD-08 generated the maximum (16) number of scorable bands all of them being polymorphic (100%). The minimum number (5) of bands was generated by primer OPC-02 with only one monomorphic band presented across all accessions and the other four were polymorphic bands with 80% polymorphism. Though primer OPE-1 produced 10 scorable bands, the majority of bands (60%) were monomorphic across all accessions.

The average number of bands per primer of 8.2±2.833 observed in this study was relatively high compared with 6.86 reported by Fakrudin *et al.* (2004) in finger millet from Africa and India but almost similar with the report of Salimath *et al.* (1995) who found 8 bands per primers in finger millet from Africa, Asia and Brazil. This difference could be attributed to the genotypes evaluated, the primers used and the selection of scorable bands in each study (Cansian & Echeverrigarya 2000).

As shown in Table 2 & 3, among the 123 RAPD fragments, 89 (72.35%) were polymorphic and 34 (27.64%) were monomorphic. This is an evidence for high degree of variability among all accessions. The high polymorphic rate (72.35%) was in agreement with the findings of Fakrudin *et al.* (2004) who reported polymorphic rate of 85.82%. However, Salimath *et al.* (1995) reported only 10% polymorphism in 17 accessions of finger millet from Africa, Asia and Brazil. The polymorphism observed in RAPD markers among finger millet accession in this study demonstrated the effectiveness of RAPD technique in determining intraspecific variation.

The polymorphic rate for each primer is shown in Table 2. It ranged from 33.3 to 100%. Primer OPC-20 exhibited the lowest (33.3%) polymorphism whereas three primers, namely OPA-09, OPD-08 and OPD-12 exhibited 100% polymorphism despite the fact that each of them generated variable number of RAPD fragments. The overall average percentage of polymorphism (72.35) was high, indicating the existence of a very high level of DNA sequence variability in finger millet landraces from Ethiopia and Eritrea. This in turn revealed the existence of high variability.

Comparison of more primers generally provides additional confirmatory evidence for genetic variation; however, due to the existence of wide gene pool of Ethiopian and Eritrean finger millet, 89 polymorphic bands generated by 15 primers could distinguish the accession evaluated in this study. Hence, this revealed the ability of RAPD to discriminate among the accessions and suggested its application for diversity study.

The discrimination power of each marker was estimated by polymorphic information content. The PIC scores ranged from 0.0 to 0.500 (OPD-08-1800) with a mean value ±SD of 0.0.162±0.175. The RAPD markers showed heterogeneous distribution of PIC scores and about 23% of markers had a high discrimination power of ≥ 0.30 (Fig. 1).

3.3 Estimates of genetic similarity

Similarity matrix based on the simple matching coefficient (Sneath & Sokal 1973) using the RAPD data

were used to assess the extent of genetic relatedness among the 66 finger millet accessions (results not shown here). The similarity coefficient value ranged from 0.585 to 0.984 among the finger millet accessions assessed in this study. The most similar genotypes appeared to be accessions 234205 (43) from Tigray, 230722 (40) and 230714 (39) both from Eritrea. Of the 123, amplified products, 121 products (98.4%) showed the same presence or absence behavior whereas the remaining two products (1.6%) exhibited different presence or absence behavior in these accessions.

The minimum genetic similarity 0.585 was between accession 215874 (14) from Gojam with accessions 211474(9) from Gamo Gofa, 236447 (50) from Welega and 238319 (59) from Tigray. Accession 215874 (14) showed 41.5 % difference of absence or presence behavior for the amplified products with these three accessions. Relatively the small number of pair wise differences in traits/markers (high genetic similarity values) among some accessions is likely their genetic relatedness. On the other hand, large number of pair wise differences (low genetic similarity values) revealed that accessions are genetically far distant.

3.4 RAPD clustering

The dendrogram constructed from cluster analysis and the number of accessions in each cluster and their origin of regions are shown in Fig. 2 and Table 4, respectively. The cluster analysis separated the accessions into two major groups at similarity scale of 0.79. In the first group only one accession, 215874 (14) from Gojam was clearly distinguished from all other accessions, implying that this accession was genetically more distinct from other accessions. However, its exclusion from others in the RAPD dendrogram was rather difficult to conclude whether this was due to morphological difference, as it was closely clustered with other 14 accessions in the morphological dendrogram reported by Kebere *et al.* (2007). The second group contained all the remaining 65 accessions, which could be further grouped into several sub-clusters depending on the similarity index employed.

All accessions were successfully separated from one another with the highest similarity value of approximately 0.98. Hence, this again confirmed RAPD could be used routinely by plant breeders to identify genetic variation (Keil & Griffin 1994), locate region of genome linked to agronomically important genes (Pillary & Kenny 1996) and facilitate introgression of desirable genes to commercial accessions (Stuber 1992).

At similarity index corresponding approximately to 0.83, the 66 accessions were clustered into nine groups. Clusters I, II, V and VI each constituted only one accession. Accession 215824 (14) in cluster I and accession 215877 (15) in cluster V originated from Gojam whereas accession 242131 (65) in cluster II was from Gonder. The improved variety, Tadesse, originally from Kenya was grouped in cluster VI.

Four accessions, 242123 (63) from Gonder, 241769 (62) from Gamo Gofa and both 237459 (56) and 237449 (53) from Tigray made cluster III. Cluster IV and VII each constituted only two accessions. Accessions 237452(55) and 235830(47), which were sampled from Tigray and Gonder, respectively made cluster IV whereas accessions, 237451 (54) and 237447 (52) both from Tigray were member of cluster VII. The majority of the accessions including one improved variety (Padet) were grouped in cluster VIII. Forty-nine accessions that was about 74.24% fall in this group. However, none of them were found to be absolutely similar. Eight accessions from Eritrea, seven from Tigray, seven from Gonder, 13 from Gojam, 10 from Welega, three from Gamo Gofa and one improved variety, Padet, were included in cluster VIII. One accession, 235138 (44) from Gonder, two accessions from Welega, 216024 (22) and 216025 (23), 2 accessions from Gamo Gofa, 100084 (2) and 100055 (1) made-up cluster IX.

Unlike the findings of Fakrudin *et al*. (2004) who found that a clear apportionment of finger millet accessions in concordance with geographical origin and pedigree history, the RAPD data result in the present study did not provide a clear-cut separation among finger millet accessions in relation to the origin of their respective geographical region although there was irregular trend that accessions from the same region were clustered together. Lack of a clear pattern of variation in relation to geographic origin was in agreement with the result of Hilu (1995) where RAPD data did not discriminate between African and Indian finger millet cultivars. Though the accessions assessed in this study mainly represented landraces from different geographical regions of Ethiopia and Eritrea, the analysis of RAPD data did not show a clear-cut pattern of variation in relation to geographical region could be due to the long history of domestication and cultivation of finger millet that might be contributed to the dispersion of alleles through

out the country, lessening the influence of geography on pattern of variation among them.

4. Conclusion

The RAPD analysis was effective in assessing the genetic diversity and the result of the present study revealed the existence of ample variability in Ethiopian finger millet landraces that could be employed in the genetic improvement. The most divergent materials and those having complementary characters could be employed in the improvement program.

5. References

Cansian, R.L. & Echeverrigaray, S. (2000), Discrimination among cultivars of cabbage using randomly amplified polymorphic DNA markers, *Horticultural Science* **35**: 1155-1158.

Cao, W., Scoles, G., Hucl, P. & Chibbar, R.N. (1999), The use of RAPD analysis to classify *Triticum* accessions, *Theoretical and Applied Genetics* **98**: 602 – 607.

Connolly, A.G., Godwin, I.D. & Cooper, M. (1994), Interpretation of randomly amplified polymorphic DNA marker for fingerprinting sweet potato (*Ipomoea batatas* L.) genotypes, *Theoretical and Applied Genetics* **88**: 332 – 336.

Dellaporta, S.L., Wood, J. & Hicks, J.B. (1983), A plant DNA mini preparation, Version II, *Plant Molecular Biology Reporter* **1**: 19-21.

Fakrudin B., Kulkarni, R.S., Shashidhar, H.E. & Hittalmani, S. (2004), Genetic diversity assessment of finger millet, *Eleusine coracana* (Gaertn), germplasm through RAPD analysis. *Plant Genetics Resources Newsletter* **138**: 50-54.

Hilu, K.W. (1995), Evolution of finger millet: Evidence from random amplified polymorphic DNA, *Genome* **38**: 232 –238.

Kebere, B., Scripichitt, P., Wongyai, W. & Hongtrakul, V. (2007), Cluster and principal component analysis of finger millet [*Eleusine coracana* (L.) Gaertn] landraces from Ethiopia and Eritrea. *Kamphaengsean Academic. Journal* **5**:40-52.

Keil, M. & Griffin A.R. (1994), Use of random amplified polymorphic DNA (RAPD) markers in the discrimination and verification of genotypes in *Eucalyptus*, *Theoretical and Applied Genetics* **89**: 442-450.

Klein-Lankhorst, R.M., Vermunt, A., Wiide, R., Liharska, T. & Zabel, P. (1991), Isolation of molecular markers for tomato (*L.* esculentium) using random amplified polymorphic DNA (RAPD), *Theoretical and Applied Genetics* **83**: 108-114.

M'Ribu, H.K. & Hilu, K.W. (1994), Detection of interspecific and intraspecific variation in *Panicum* millets through random amplified polymorphic DNA, *Theoretical and Applied Genetics* **88**: 412 – 416.

Pillary, M. and Kenny, S.T. (1996), Random amplified polymorphic DNA (RAPD) markers in hop *Humulus lupulus*: level of genetic variability and segregation in F1 progeny, *Theoretical and Applied Genetics* **92**: 334-339.

Roholf, F.J. (1997), *NTSYS-PC Numerical Taxonomy and Multivariate Analysis System*, Version 2.01. Exseter Software, New York: Setauket.

Rueda, J., Linacero, R. & Vazquez, A.M. (1998), Plant total DNA extraction, in Karp, A., Isaac, P.G. & Ingram, D.S. (eds), *Molecular Tools for Screening Biodiversity*, London: Chapman, pp. 10-14.

Salimath, S.S., de Olivera, A.C., Godwin, I.D. & Bennetzen, J.L. (1995), Assessment of genome origins and genetic diversity in the genus *Eleusine* with DNA markers, *Genome* **38**: 757 –763.

Sneath, P.H.A. & Sokal, R.R. (1973), *Numerical Taxonomy*, San Francisco: Freeman.

Steiner, J.J. & Greene, S.L. (1996), Proposed ecological descriptors and their utility for plant germplasm collection, *Crop Science* **36**: 439-451.

Stiles, J.L., Lemme, C., Sondur, S., Morshidi, M.B. & Manshardt, R. (1993), Using randomly amplified polymorphic DNA for evaluating genetic relationships among papaya cultivars, *Theoretical and Applied Genetics* **85**: 697-701

Stuber, C.W. (1992), Biochemical and molecular marker in plant breeding, *Plant Breeding Reviews* **9**: 37-61.

Weir, B.S. (1996), *Genetic Data Analysis II* (2nd edition). Sunderland: Sinauer.

Welsh, J. & McClelland, M. (1990), Fingerprinting genomes using PCR with arbitrary primers, *Nucleic Acids Research* **18**: 6531-6535.

Welsh, J., Honeyyecutt, R.J., McClelland, M. & Sobral B.W.S. (1991), Parentage determination in maize hybrids using the arbitrarily primed polymerase chain reaction (AP-PCR), *Theoretical and Applied Genetics* **91**: 1078-1085.

Williams, J.G.K., Kubelik, A.R., Livak, K.J., Rafalski, J.A. & Tingey, S.V. (1990), DNA polymorphisms amplified by arbitrary primers are useful as genetic markers, *Nucleic Acids Research* **18**: 6531-6535.

Zonary, D. (1970). Centers of diversity and centers of origin, in Frankel, O.M. & E. Bennett, E. (eds), *Genetic Resources in Plants, Their Exploration and Conservation*, Oxford: Blackwell, pp. 33-42.

Table 1. Finger millet accessions with their collection administrative region and altitude used in the study

Code	Accession number	Local (vernacular) name	Collection adm. Region	Altitude	Code	Accession number	Local (vernacular) name	Collection adm. Region	Altitude
1	100055	Dagussa	Gamo Gofa	1450	34	229728	-	Gojam	1440
2	100084	Dagussa	Gamo Gofa	1820	35	230101	Dagusa	Eritrea	1740
3	100094	Key dagussa	Welega	1290	36	230117	Dagusa	Eritrea	1650
4	100095	Dagussa	Welega	1480	37	230130	-	Eritrea	1800
5	204747	Dagussa	Eritrea	2250	38	230136	-	Eritrea	1900
6	208444	Dagussa	Gonder	2500	39	230714	Dagusa	Eritrea	1950
7	208448	Dagussa	Gojam	1250	40	230722	Dagusa	Eritrea	2000
8	208730	Daguja	Welega	1900	41	230724	Dagusa	Eritrea	2040
9	211474	Baracha	Gamo Gofa	1560	42	234178	Dagusa	Tigray	1840
10	213035	Bercha	Gamo Gofa	1380	43	234205	Dagusa	Tigray	2100
11	215841	Dagussa	Gojam	2050	44	235138	Dagusa	Gonder	2200
12	215850	Dagussa	Gojam	1800	45	235141	Dagusa	Gonder	1870
13	215867	Dagussa (Tikur)	Gojam	1990	46	235700	Berecha	Gamo Gofa	1530
14	215874	Dagussa	Gojam	2350	47	235830	-	Gonder	1640
15	215877	Dagussa	Gojam	2230	48	235838	-	Gonder	1290
16	215879	Dagussa	Gojam	2400	49	235842	-	Gonder	940
17	215883	Dagussa	Gojam	2400	50	236447	Daguso	Welega	1630
18	215889	Dagussa (Nech)	Gojam	2100	51	236450	Daguso	Welega	2230
19	215896	Dagussa	Gojam	2160	52	237447	Dagusa	Tigray	1570
20	215973	Dagussa	Gonder	2090	53	237449	Dagusa	Tigray	1470
21	215977	Dagussa	Gonder	1940	54	237451	Dagusa	Tigray	1450
22	216024	Daguja	Welega	2330	55	237452	Dagusa	Tigray	1430
23	216025	Daguja	Welega	2330	56	237459	Dagusa	Tigray	1940
24	216028	Guracha Dagnja	Welega	2150	57	237462	Dagusa	Tigray	2100
25	216041	Daguja	Welega	1960	58	237477	Dagusa	Tigray	1710
26	216043	Daguja	Welega	1980	59	238319	Dagusa	Tigray	2130
27	216045	Daguja	Welega	1880	60	238331	Dagusa	Tigray	1300
28	216051	Daguja	Welega	1910	61	238336	Dagusa	Tigray	1020
29	216052	Daguja	Welega	1660	62	241769	Persheka	Gamo Gofa	1500
30	225895	Tikur Dagussa	Gojam	1845	63	242123	Tikur Dagusa	Gonder	1720
31	229723	Dagusa	Gojam	1300	64	242125	Tikur Dagusa	Gonder	2015
32	229726	-	Gojam	1600	65	242131	Tikur Dagusa	Gonder	2350
33	KNE#411	Padet*	Kenya	-	66	KNE#1098	Tadesse*	-	-

- Information not available, * Improved variety, developed from introduction.

Table 2. List of selected RAPD primers, their sequence, number of bands and polymorphic (%) of the RAPD analysis result in 66 finger millet accessions

Primer No.	Primer sequence	Amplified DNA bands			Polymorphic (%)
		Polymorphic	Monomorphic	Total	
OPA-09	5´GGGTAACGCC3´	6	0	6	100
OPA-10	5´CAATCGCCGT 3´	5	3	8	62.5
OPA-11	5´GTGATCGCAG 3´	7	3	10	70.0
OPA-18	5´AGGTGACCGT 3´	6	2	8	75.0
OPAA-03	5´TTAGCGCCCC 3´	4	2	6	66.8
OPC-02	5´GTGAGGCGTC3´	4	1	5	80.0
OPC-05	5´GATGACCGCC3´	5	2	7	71.4
OPC-20	5´ACTTCGCCAC 3´	3	6	9	33.3
OPD-08	5'GTGTGCCCCA 3'	16	0	16	100
OPD-12	5'CACCGTATCC 3'	11	0	11	100
OPD-18	5'GAGAGCCAAC 3'	5	1	6	83.3
OPE-01	5'CCCAAGGTCC 3'	4	6	10	40.0
OPX-05	5´ CCTTTCCCTC 3´	5	1	6	83.3
OPY-02	5´ CATCGCCGCA3´	3	3	6	50.0
UBC-06	5'GGGCCGTTTA 3'	5	4	9	55.6
Total		89	34	123	
Average		5.93	2.27	8.20	72.35
SD		3.390	1.944	2.833	

Table 3. Summary of statistics of RAPD analysis of 66 finger millet accessions

Parameter	Estimates
Total marker	123
Total number of polymorphic bands	89
Maximum number of bands produced by a primer	16
Minimum number of bands produced by a primer	5
Average number of polymorphic bands per primer±SD	5.93±3.39
Average number of bands produced by primer±SD	8.2±2.833
Average polymorphic information content (PIC)±SD	0.162±0.175
Average polymorphism rate	72.35

Table 4. Number of finger millet accessions in each cluster and region of origin

Cluster	Total number of accessions/cluster	Region of Origin	Number of accessions/region
I	1	Gojam	1
II	1	Gonder	1
III	4	Tigray	2
		Gonder	1
		Gamo Gofa	1
IV	2	Tigray	1
		Gonder	1
	1	Gojam	1
VI	1	Improved Variety*	1
VII	2	Tigray	2
VIII	49	Eritrea	8
		Tigray	7
		Gonder	7
		Gojam	13
		Welega	10
		Gamo Gofa	3
		Improved variety*	1
IX	5	Gonder	1
		Welega	2
		Gamo Gofa	2

* Developed from introduction

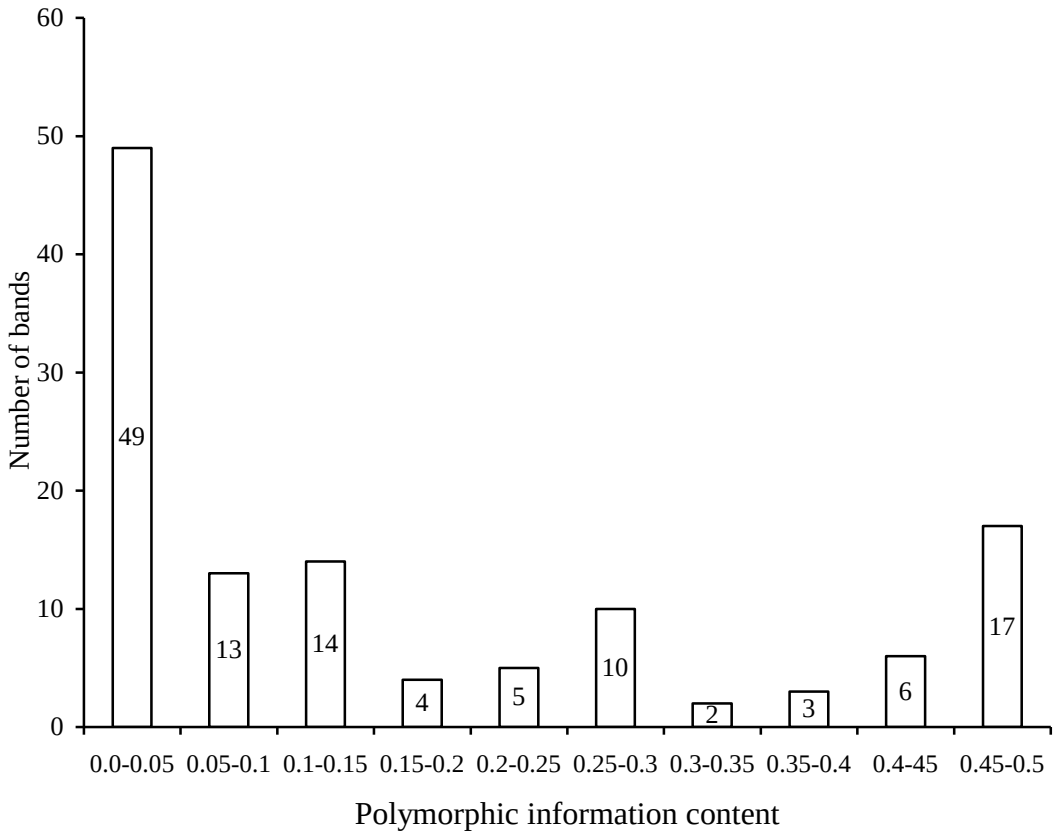

Figure 1. Distribution of polymorphic information content scores for 123 RAPD markers among 66 finger millet accessions

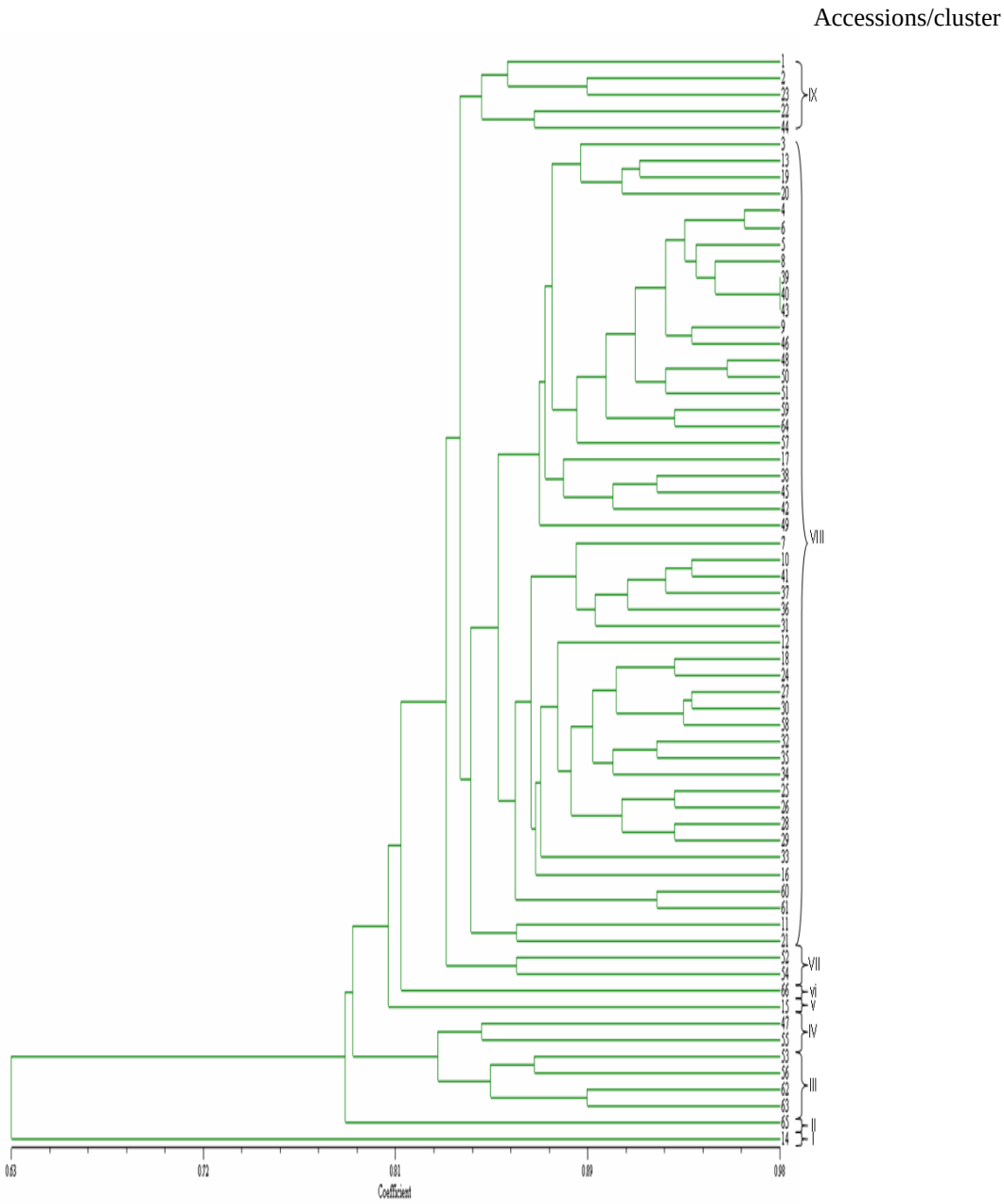

Figure 2. Dendrogram showing genetic relationship among 66 finger millet accessions generated by UPGMA cluster using 123 RAPD markers amplified by 15 RAPD primers