

Comaparison of Morphological and Molecular Diversity of 34 Moringa (*Moringa Oleifera*. Lam) Genotypes

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ABSTRACT: The present investigation was conducted at the field of the Horticulture college and Research Institute, Tamil Nadu Agricultural University, Periyakulam during 2011-12 to study the nature of morphological and molecular diversity of 34 moringa ecotypes collected from Southern districts of Tamil Nadu. From the D² analysis under Tocher's method, seven clusters were formed representing the genetic diversity among the genotypes studied. Cluster I topped in having twelve numbers of genotypes among clusters formed, whereas cluster VI having ten genotypes and VI cluster having one genotype. Average intra and inter cluster distances in studied genotypes showed range from 0.00 to 19.97 it means the cluster posses highest intra cluster distance D= 19.97 which includes ten genotypes and maximum average inter cluster distance was observed between II and VII (31.38) and VI and VII (30.54). Study on genetic diversity of 34 moringa ecotypes of on the basis of molecular parameters. In the present study, 20 SSR markers produced 33 alleles and the number of alleles ranged from 2 to 3 with an average of 2.2 alleles per primer. The maximum number of amplified products was generated by primer MO 41 followed by MO 64. Allele sizes varied from 110-400 bp. PIC content of the SSR primers ranged from 0.34 to 0.76 with an average of 0.57. Hence, the primer pairs MO 6, MO 8 and MO 64 are considered to be worth in future studies in the field of taxonomical and genetic resource management.

Keywords: *Moringa oleifera*, morphological diversity, D² value, molecular diversity, SSR Markers, PIC value.

INTRODUCTION

Popularly known as "Drumstick" tree, horseradish tree, or Ben tree, *M. oleifera* is a deciduous to evergreen shrub or small tree with a height of 5 to 10 m (Morton, 1991). Its seedlings are fast growing with early sexual maturity and a height up to 4.5 m in 9 months and flowering in half a year (Von Maydell, 1986). *M. oleifera* used to distribute wildly in the forests of Western Himalaya, and then throughout India by cultivation (Selvam, 2005). Featured by richness in proteins, minerals, and vitamins, the leaves of *M. oleifera* are used as a highly nutrient vegetable and as cattle fodder (Mughal *et al.*, 1999). In addition, the seed powder is used in water purification (Ndabigengesere and Narasiah, 1998), and the seed oil is acquired for edibles, lubrication, and cosmetics (Anwar and Bhangar, 2003).

Since variability is a prime requisite for any selection programme, it is necessary to detect the

amount of variation in the population, which is subjected to selection. For morphological characterization, leaf characters like petiole colour, leaf colour and leaflet size, and flower characters like flower colour and flower size and fruit characters like fruit colour, fruit length, fruit girth and fruit weight are considered. When the variation existing between and within the population is not detectable through visual or morphological means, there is a need for a highly reliable and precise method to detect the variation without any environmental effects. The advent of molecular techniques, an analysis based on the polymorphisms found in protein or DNA has greatly facilitated research activities in the fields of phylogeny, taxonomy, genetics and plant breeding. At present, there are number of molecular markers available for such detection of variability, and they are not influenced by environmental effects. Simple Sequence Repeats (SSR), is a polymerase chain

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reaction (PCR) based DNA marker system, which is simple, cost effective and capable of revealing variation even at single nucleotide level. Based on co-dominant features and high allelic polymorphism, microsatellites [simple sequence repeats (SSRs)] have become a useful marker system in genetic diversity studies (Walter and Epperson, 2001); (Chaix *et al.*, 2003).

MATERIALS AND METHODS

The genetic diversity among thirty four accessions of moringa was assessed by using D^2 Mahalanobis statistics (Mahalanobis, 1936). The grouping of accessions was done using Tocher's method, as described by Rao (1952). Morphological data recorded were converted into similarity matrix, using the similarity coefficient of Rogers and Tanimoto (Rogers and Tanimoto, 1960) with the SIMQUAL programme of NTSYS-pc version 2.02i (Rohlf, 1998). A PCA was computed based on similarity coefficient for comparison among the 25 genotypes, based on morphological data.

Molecular diversity was done by DNA extraction, DNA quality and quantity check, DNA amplification, PCR Programme, Gel electrophoresis and dendrogram analysis. Scoring of SSR marker was carried out by considering only the clear and unambiguous bands. Markers were scored for the presence and absence of the corresponding band among the different accessions. The scores '1' and '0' were given for the presence and absence of bands, respectively. Polymorphism information content (PIC) for each SSR marker was calculated based on the formula $PIC_i = 2f_i(1-f_i)$, where PIC_i is the polymorphic information content of the marker i , f_i is the frequency of marker bands present and $(1-f_i)$ is the frequency of absent marker bands (Roldan-Ruiz *et al.*, 2000).

The data obtained by scoring the SSR profiles of different primers were subject to cluster analysis. Similarity matrix was constructed using Jaccard's coefficient (Jaccard, 1908) and the similarity values were used for cluster analysis and dendrogram was constructed by Unweighted Pair-Group Method using Arithmetic averages (UPGMA) with the Sequential Agglomerative Hierarchical and Nested (SHAN) function (Sneath and Sokal, 1973). Data analysis was done using NTSYS-pc version 2.02i (Rohlf, 1998).

RESULTS AND DISCUSSION

Mahalanobis D^2 Analysis

Thirty four accessions of moringa were subjected to analysis based on 25 characters following

Mahalanobis D^2 statistics. The per plant mean of thirty four accessions was transformed into standardized uncorrelated mean values.

The D^2 values corresponding to all possible combinations among thirty four accessions were computed. By application of clustering technique, thirty four genotypes were grouped into eight clusters.

The pattern of distribution of genotypes into various clusters with their source is indicated in **Table 1**. Among the eight clusters, cluster I was the largest with 17 accessions, followed by cluster VI and cluster VII with 5 accessions each; cluster III with 3 accessions, clusters II, IV, V and VIII with one accession each.

Table 1
Distribution of genotypes into different clusters

Cluster No.	No. of types	Accession No.	Source/ Tamil Nadu
I	17	MO 2	Salaipudhur
		MO 3	Palakanoothu
		MO 4	Veriyappur
		MO 6	Kaveiyampatty
		MO 7	Kaveiyampatty
		MO 8	Idayarkottai
		MO 13	Murugamalai hills
		MO 14	Murugamalai hills
		MO 17	Seelayampatty
		MO 18	Kamatchipuram
		MO 20	Kamatchipuram
		MO 23	Sadayandipatty
		MO 27	Periyakulam
		MO 28	Endapuli
		MO 29	Endapuli
		MO 30	Endapuli
		MO 31	M. Vaipatty
II	1	MO 26	Periyakulam
III	3	MO 5	Pachaloor
		MO 24	Kandamanoor
		MO 25	Kadamalaigundu
IV	1	MO 32	Ganesapuram
V	1	MO 11	Kamatchipuram
VI	5	MO 9	Idayarkottai
		MO 19	Kamatchipuram
		MO 21	Kurangani
		MO 22	Kamatchipuram
VII	5	MO 10	Periyakottai
		MO 15	Murugamalai hills
		MO 16	Rajathanikottai
		MO 33	Puur
		MO 34	Meenakshi oothu
VIII	1	MO 1	Salaipudhur

Intra and inter cluster distance

The inter and intra cluster D^2 and D values among the eight clusters are presented in **Table 2**. Cluster VII recorded the maximum intra cluster distance of 26.27 followed by cluster VI with an intra cluster distance of 22.58.

The highest inter cluster distance was found between cluster VI and VII (38.24) which was followed by cluster VII and cluster VIII (36.42) and cluster III and VII (35.03). The least inter cluster distance was found between cluster II and V (18.46) followed by cluster V and VI (19.63).

Table 2
Cluster distance values for 25 characters in moringa

	I	II	III	IV	V	VI	VII	VIII
I	18.98	22.23	24.40	23.72	23.11	25.90	27.39	26.81
II		0.00	20.46	24.82	18.46	20.34	34.36	27.04
III			20.54	26.64	24.90	28.45	35.03	29.46
IV				0.00	27.99	33.22	24.69	27.87
V					0.00	19.63	35.22	24.22
VI						22.58	38.24	31.25
VII							26.27	36.42
VIII								0.00

Relative contribution of each character to genetic divergence

The rating technique was adopted to rank the characters in the order of their contribution to total genetic divergence. The data on contribution of each character towards the distance are furnished in **Table 3**. The highest contribution to genetic divergence was by Fruit weight (24.78 per cent), followed by trunk girth (23.53 per cent), number of seeds/ fruit (16.93 per cent), Leaf Area Index (LAI) (6.77 per cent). The contribution of fruit girth was 6.24 per cent, magnesium (4.28 per cent) and β carotene (3.92 per cent). The contribution by other characters ranged from 0.00 to 3.57 per cent.

Table 3
Relative contribution of characters to genetic divergence

S.no	Source	Times ranked 1 st	Contribution %
1.	Fruit weight (g)	139	24.78
2.	Trunk girth (cm)	132	23.53
3.	Number of seeds/fruit	95	16.93
4.	Leaf Area Index (LAI)	38	6.77
5.	Fruit girth (cm)	35	6.24
6.	Magnesium (mg/100g)	24	4.28
7.	β carotene (mg/100g)	22	3.92
8.	IAA oxidase activity	20	3.57
9.	Leaf area (cm ²)	13	2.32
10.	Pod numbers	9	1.60
11.	Number of flowers/ Inflorescence	9	1.60
12.	100 seed weight	8	1.43
13.	Seed number/kg	5	0.89
14.	Length of fruit (cm)	5	0.89
15.	Calcium (mg/100g)	3	0.53
16.	Plant height (m)	1	0.18
17.	Chlorophyll (SPAD value)	1	0.18
18.	Chlorophyll Stability Index (CSI) (%)	1	0.18
19.	Nitrate reductase activity	1	0.18
20.	OTHERS	0	0.00
21.	TOTAL	561	100

Cluster mean values for characters

The mean values of 25 characters for eight clusters are presented in **Table 4**. Cluster VI recorded the highest mean value for days to flower appearance (45.33) followed by cluster III (42.48) and cluster II (41.85). Cluster IV recorded the lowest mean value for days to flower appearance (31.88). Cluster VII recorded the highest mean value for plant height (50.27) followed by clusters IV (47.07) and II (46.07). Cluster III recorded the lowest mean value for plant height (37.05) and cluster VI (37.51).

Cluster VIII recorded the maximum leaf length of 71.83 followed by cluster V (52.00) Cluster I recorded the minimum petiole length of 44.16. The highest mean values for Leaf area (cm²) was recorded by cluster VIII (581.33), followed by cluster V (523.00) and the lowest was recorded by cluster II and VII (359.33). The highest mean for Leaf Area Index was recorded by cluster VIII (3.30) and the lowest by cluster III (0.98). The highest mean for trunk girth was recorded by cluster VII (189.32) while, the lowest mean for trunk girth was recorded by cluster II (77.23). The highest mean value for Number of flowers/ Inflorescence was recorded by cluster V (27.45) and cluster VIII recorded the lowest mean (16.76).

Cluster VI recorded the maximum mean value for length of fruit (71.66) and the lowest length of fruit was recorded by cluster IV (28.30). Cluster VI recorded the maximum value for fruit girth (11.72) followed by cluster I (9.11) while, cluster V recorded the lowest mean value for fruit girth (6.73). Cluster VI recorded the highest mean value for fruit weight (183.38) and cluster III recorded lowest mean value for oxalate content (84.39). Cluster II recorded highest mean value for number of seeds/fruit (23.64) and lowest in cluster VII (16.21).

Cluster VIII recorded the highest mean value for pod numbers (133.58) followed by cluster II (115.83) while cluster V recorded the lowest mean value for pod numbers (73.91). The highest mean value for seed number/kg was recorded in cluster V (4179.00) followed by cluster VII (4015.40). Cluster VIII recorded the lowest mean value for seed number/kg (3144.00).

Cluster V recorded the maximum mean value for oil content (30.40) and the lowest oil content was recorded by cluster IV (19.43). Cluster VIII recorded the maximum value for chlorophyll content (52.80) followed by cluster III (49.11) while, cluster IV recorded the lowest mean value for chlorophyll content (42.30). Cluster IV recorded the highest mean value for Chlorophyll Stability Index (85.67) and

cluster V recorded lowest mean value for oxalate content (77.47). Cluster II recorded highest mean value for IAA oxidase activity (3.74) and lowest in cluster VIII (1.40).

Cluster VII recorded the lowest mean value for Nitrate reductase activity (29.08) followed by cluster VIII (28.67) while cluster V recorded the lowest mean value for Nitrate reductase activity (22.93). The highest mean value for relative water content was recorded in cluster II (94.63) followed by cluster III (94.40). Cluster V recorded the lowest mean value for relative water content (81.03).

Cluster IV recorded the maximum mean value for leaf soluble protein (28.00) and the lowest length of fruit was recorded by cluster II (23.00). Cluster III and

V recorded the maximum value for β carotene (12.90) followed by cluster I (11.55) while, cluster IV recorded the lowest mean value for β carotene (9.80). Cluster IV recorded the highest mean value for crude fibre (12.68) and cluster II recorded lowest mean value for crude fibre (10.79). Cluster V recorded highest mean value for iron content (131.97) and lowest in cluster VIII (109.80).

Cluster II recorded the maximum calcium content of 3.87 followed by cluster VIII (3.07) Cluster III recorded the minimum petiole length of 1.82. The highest mean values for magnesium was recorded by cluster IV (0.72), followed by cluster II (0.68) and the lowest was recorded by cluster VIII (0.32).

Table 4
Cluster mean value for 25 characters in moringa

Clusters	Plant height (m)	Leaf length (cm)	Leaf area (cm ²)	Leaf Area Index (LAI)	Trunk girth (cm)	Number of flowers/ Inflorescence	Length of fruit (cm)	Fruit girth (cm)	Fruit weight (g)	Number of seeds/ fruit	Pod numbers	Seed number/ kg	100 seed weight
I	5.28	44.16	436.31	1.76	149.13	24.72	61.77	9.11	111.68	21.80	105.49	3202.26	33.27
II	4.73	49.00	359.33	1.25	100.65	18.47	58.70	7.63	153.87	23.64	115.83	3822.00	30.13
III	4.84	47.11	491.00	0.98	77.23	19.71	58.78	8.23	84.39	23.06	100.93	3640.67	30.09
IV	5.57	49.33	436.33	1.74	109.78	25.42	28.30	8.30	106.68	12.89	109.41	3240.67	29.70
V	5.29	52.00	523.00	2.15	123.63	27.45	55.00	6.73	177.62	21.39	73.91	4179.00	33.60
VI	5.38	48.13	468.27	1.22	136.73	25.96	71.66	8.02	183.38	23.47	98.48	3423.60	35.01
VII	5.55	51.27	359.33	1.78	189.32	26.41	49.52	7.53	95.60	16.21	86.41	4015.40	32.78
VIII	4.32	71.83	581.33	3.30	80.13	16.76	51.84	11.72	120.48	19.40	133.58	3144.00	38.27

Clusters	Oil content (%)	Chlorophyll (SPAD value)	Chlorophyll Stability Index (CSI) (%)	IAA oxidase activity	Nitrate reductase activity	Relative water content (RWC)	Leaf soluble protein (mg/100g)	β carotene (mg/100g)	Crude fibre (%)	Iron (mg/100g)	Calcium (mg/100g)	Magnesium (mg/100g)
I	25.91	45.17	84.83	3.44	28.01	89.78	27.61	11.55	11.99	118.92	2.15	0.48
II	26.40	48.70	84.80	3.74	25.20	94.63	23.00	11.37	10.79	119.87	3.87	0.68
III	29.71	49.11	84.59	3.13	24.73	94.40	26.40	12.90	11.18	123.79	1.82	0.53
IV	19.43	42.30	85.67	3.63	24.83	83.40	28.00	9.80	12.68	128.70	2.95	0.72
V	30.40	43.60	77.47	3.53	22.93	81.03	22.17	12.90	12.47	131.97	2.00	0.48
VI	28.43	44.41	85.05	2.92	27.73	88.46	26.97	11.33	12.09	125.21	2.29	0.53
VII	26.69	47.57	85.64	2.94	29.08	86.11	26.50	10.30	11.79	123.12	2.30	0.38
VIII	24.53	52.80	82.70	1.40	28.67	94.30	24.80	10.40	11.43	109.80	3.07	0.32

D² analysis was carried out using the data gathered from twenty five characters. The presence of high variability among the accessions studied for different characters were further confirmed through the pattern of distribution of 34 moringa accessions into eight clusters based on genetic divergence D² statistics in which cluster I contained maximum

number of accessions (17), Cluster VI (5), Cluster VII (5), cluster III (3), clusters II, IV, V and VIII have one accession in each. Clusters I and VI revealed a number of accessions from different geographical sources which indicated that genetic divergence has no relationship with geographical divergence. Such failure of relationship between genetic diversity and

geographical diversity suggested that forces such as exchange of genetic stocks, genetic drift, spontaneous variation, natural and artificial selection are perhaps responsible for genetic diversity which is in line with the findings of Nagarajan and Prasad (1980); Pawar (1995) and Kamble (2000). Such clustering pattern also implied that there is distinct divergence between different genotypes of moringa.

DIVERSITY ANALYSIS USING SSR MARKERS

SSR Analysis

Thirty four accessions of moringa were also analyzed using twenty SSR markers (Table 5). Twenty primer

Table 5
List of primers used for SSR analysis

S. No. (Max. 15 Characters)	Primer Name	Primer Sequence 5' to 3'	Annealing temperature
1	MO1 F	TTGCTGCCTCCTTTTGCA	58
	MO1 R	AACTGTCACCTCCTATCCA	
2	MO6 F	GCATAGCCACCTTTACTCCT	61
	MO6 R	GACTTTTGAACTCCACCACC	
3	MO8 F	GTAGATGGTGCAGTACTCA	58
	MO8 R	TGGGGTTCTTGTTCCTTAT	
4	MO10 F	CTTTACACCTCAGTATCCCT	58
	MO10 R	GTTCCGGCTTATGTTCTCGTT	
5	MO12 F	ACCGAAGATGATAAGGTGGG	59
	MO12 R	CAAAAAGGAAGAACGCAAGAG	
6	MO13 F	TTTCGGGTTTTCTTCACGC	58
	MO13 R	AGCTCACTTCCATCTCCAT	
7	MO15 F	CCCCTCTATTTCCATTTCC	59
	MO15 R	GCTCCATAAAACCTCTTGCT	
8	MO18 F	TTTTCTCCCTTATTGTGCC	58
	MO18 R	CCGTTGCCCTTTGTGGTTCA	
9	MO41 F	TGGGATTAGGGCATTAGAAA	55
	MO41 R	TAGTGGGTCCAAGACAAAGC	
10	MO44 F	GGCACATAGGCACGCAATAC	59
	MO44 R	CAACAAAACCCATCCAGAAA	
11	MO45 F	CCTTTGAAGTTGAAAATCTC	58
	MO45 R	TTCTAGGGTAGTTGAATCCA	
12	MO46 F	ACCAAGGGTTTCAACTGCTG	61
	MO46 R	CATTTTGGCAGCGGTCTCACG	
13	MO48 F	AGAAGAACCAACAGAGGAT	58
	MO48 R	CTTTTCACTAACCACCACC	
14	MO55 F	ATTACAGAACGATGAAACCA	56
	MO55 R	CTCTTTCCCTCCATTCAACC	
15	MO56 F	TCAATACGCCAAGTAAGCAA	60
	MO56 R	AAGCACTTCACGCATAAAAC	
16	MO58 F	TGGATTTCTTCTCTGCTAT	58
	MO58 R	CACAGTTCTTATTGTATTGG	
17	MO61 F	TGTGGGTCTTCTCTCTCTC	60
	MO61 R	CTTCTGTCTTCTCTCTCTGCT	
18	MO62 F	AAACATAGCAACTGTGAGAT	55
	MO62 R	CTCCAACAACATACAAAATC	
19	MO64 F	TCCGCACCTTCTCTCTCTT	58
	MO64 R	AACTCCCTTGACGGACACCAG	
20	MO68 F	TGCTTCGCTTCTCTATTCT	56
	MO68 R	ACCACAGGCTTGCTTCAGTA	

pairs were screened out of which fifteen pairs showed amplification. Out of fifteen primer pairs eleven were found to be polymorphic and four of them were monomorphic (Table 6).

The fifteen primers generated a total of 28 amplicons and 21 of them show polymorphism (61.54). The number of amplicons per primer ranged from one to three and polymorphic amplicons ranged from one to two. The highest number of polymorphic amplicons (2) was generated by primer MO 1, MO 6, MO 8, MO 41, MO 46, MO 48, MO 61, MO 62, MO 64 and MO 68. Primers MO 1, MO 6, MO 8, MO 41, MO 46, MO 48, MO 62, MO 64 showed the highest per cent of polymorphism (100%) while the least per cent of polymorphism was recorded in primer MO 12 with 50 %. In the present study, 15 SSR markers produced 33 alleles and the number of alleles ranged from 2 to 3 with an average of 2.2 alleles per primer. The number of alleles amplified ranged from 2 to 4 in cassava (Moyib *et al.*, 2007). The maximum number of amplified products was generated by primer MO 41 followed by MO 64. Allele sizes varied from 110-400 bp which were in close agreement with allelic size reported by Asare *et al.* (2011) in cassava.

Table 6
List of SSR primers showing total and polymorphic amplicons generated pattern for 34 genotypes of moringa accessions

S.No	Primer Name	Total no. of bands (a)	Total no. of polymorphic bands (b)	% Polymorphism (b/a x 100)
1.	MO 1	2	2	100
2.	MO 6	2	2	100
3.	MO 8	2	2	100
4.	MO 10	1	0	0
5.	MO 12	2	1	50
6.	MO 41	2	2	100
7.	MO 46	2	2	100
8.	MO 48	2	2	100
9.	MO 55	1	0	0
10.	MO 56	1	0	0
11.	MO 58	1	0	0
12.	MO 61	3	2	66.66
13.	MO 62	2	2	100
14.	MO 64	2	2	100
15.	MO 68	3	2	66.66
	Total	28	21	75.00

Polymorphism Information Content

Polymorphism Information Content (PIC) value was calculated for 15 polymorphic primers and given in the Table 7. PIC value was highest for the primer MO 6 (0.76) followed by primer MO 8 (0.70) while, the lowest PIC value recorded by the primer MO 61 (0.34). The mean PIC value for 15 polymorphic primers was

0.575. Polymorphic Information Content (PIC) reveals the quantity of information that can be obtained from a particular primer. In the present study, PIC content of the SSR primers ranged from 0.34 to 0.76 with an average of 0.57. Similar PIC values were obtained in earlier studies using SSR markers. PIC values ranged from 0.07 to 0.77 with an average of 0.52 (Asare *et al.*, 2011) in cassava. The higher PIC value indicated the informativeness of the primer pairs. Hence, the primer pairs MO 6, MO 8 and MO 64 are considered to be worth in future studies in the field of taxonomical and genetic resource management particularly in moringa.

Cluster Analysis

The cluster analysis of 34 moringa accessions based on UPGMA suggested the formation of six clusters (Fig.1). Among the six clusters, cluster I was the largest with 9 accessions followed by cluster II (7 accessions). Cluster III comprised of 6 accessions, cluster IV comprised of 5 accessions, cluster V consisted of 5 accessions and Cluster VI comprises of 2 accessions. Cluster I and II has many sub clusters. It is inferred from the cluster that Cluster 1 and VI have more differences in molecular level and it may be used in molecular breeding.

Table 7
List of SSR primers used and the level of polymorphism detected

S.No	Primer	PIC Value	Allele size range (bp)	Number of alleles
1.	MO 1	0.64	150-160	2
2.	MO 6	0.76	350-400	2
3.	MO 8	0.70	150-200	2
4.	MO 10	0.66	200-250	2
5.	MO 12	0.56	150-250	2
6.	MO 41	0.39	150-170	3
7.	MO 46	0.58	150-250	2
8.	MO 48	0.60	120-130	2
9.	MO 55	0.60	150-250	2
10.	MO 56	0.58	250-300	2
11.	MO 58	0.57	250-280	2
12.	MO 61	0.34	280-310	3
13.	MO 62	0.65	110-120	2
14.	MO 64	0.62	200-300	2
15.	MO 68	0.37	200-300	3
	Total	8.62		33
	Mean	0.575		2.2

Comparison between morphological and molecular markers

The grouping of the genotypes as observed in cluster analysis based on SSR data was also compared with the available morphological data. The grouping of genotypes based on cluster analysis was not similar between morphological data and SSR analysis. Both morphological descriptors and SSR markers were able to group the accessions into distinct clusters independent of locality of collection. However, where the morphological descriptors indicated some accessions were the same, SSR markers were able to distinguish them into distinct genotypes with some located in different clusters. The wide genetic variability observed using SSR markers would be valuable for efficient management of germplasm and for effective utilization of materials in breeding programmes to produce hybrids of desirable

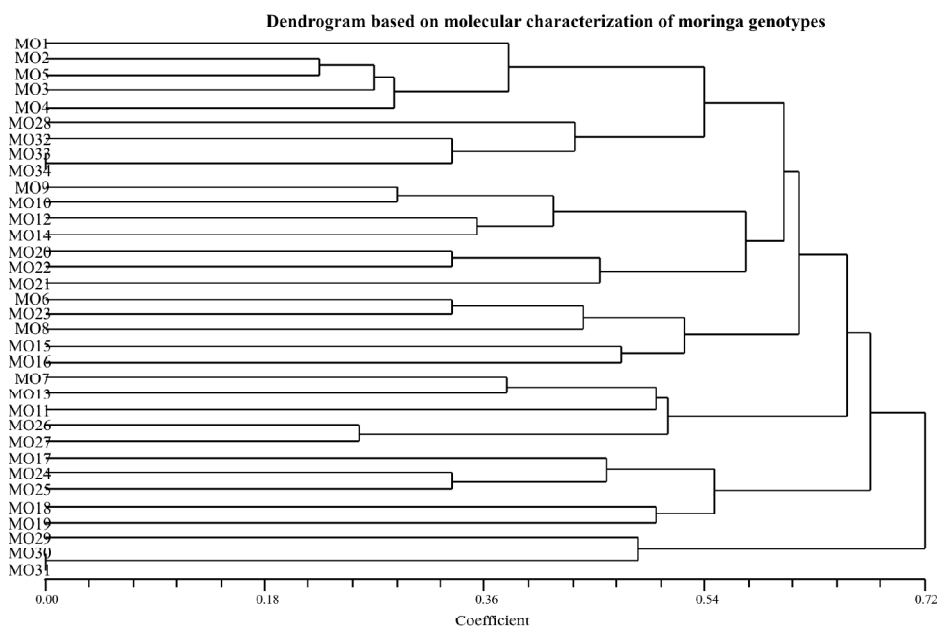


Figure 1: Dendrogram showing the distribution of the thirty four moringa accessions

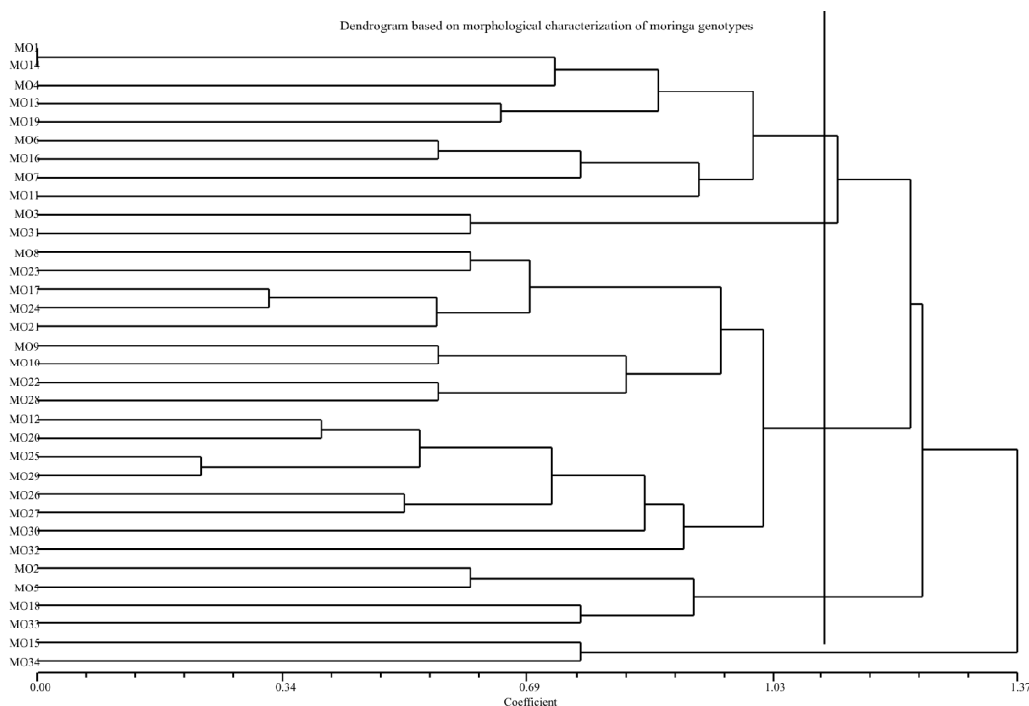


Figure 2: Dendrogram based on morphological characterization of moringa genotypes

characteristics. Therefore, the application of morphological descriptors in management of germplasm should be backed by the use of molecular markers. Despite this low correlation between morphological and SSR analysis, there were few similar groups formed in respective dendrograms. The genotypes *viz.*, MO 1, MO 3 and MO 4 are grouped under I cluster in both morphological and SSR analysis and the genotypes *viz.*, MO 10, MO 12, MO 20, MO 21 and MO 22 are grouped under II cluster in both morphological and SSR analysis. However, some discrepancies between the two dendrograms can be found. In morphological analysis, MO 15 and MO 34 formed a separate cluster were grouped in cluster IV, while in the SSR analysis, the genotypes MO 15 and MO 34 are grouped under different sub clusters. Similar results were also reported in mandarins by Koehler-Santos *et al.* (2003), who detected differences between dendrograms generated from morphological and SSR data and they suggested that morphological and molecular differences are apparently independent, due to different selection and evolutionary factors.

REFERENCES

- Anwar, F. and M.I. Bhangar. (2003), Analytical characterization of *Moringa oleifera* seed oil grown in temperate regions of Pakistan. *J. Agr. Food Chem.*, **51**: 6558-6563.
- Asare, P.A., I. K. A. Galyuon, J. K. Sarfo and J. P. Tetteh. (2011), Morphological and molecular based diversity studies of some cassava (*Manihot esculenta* crantz) germplasm in Ghana. *Afr. J. of Biotechnol.*, **10** (63): 13900-13908.
- Chaix, G., S. Gerber, V. Razafimaharo, P. Vigneron, D. Verhaegen, and S. Hamon. (2003), Gene flow estimation with microsatellites in a Malagasy seed orchard of *Eucalyptus grandis*. *Theor. Appl. Genet.*, **107**: 705-712.
- Jaccard, P. (1908), Nouvelles recherches sur la distribution florale. *Bull. Soc. Vaud. Sci. Nat.*, **44**: 223-270.
- Mahalanobis, P.C. (1936), On the generalized distance in statistic. *Proc. Natn. Inst. Sci., India*, **132**: 49-50.
- Morton, J. F. (1991), The horseradish tree, *Moringa pterigosperma*(Moringaceae): A boon to arid lands. *Econ. Bot.*, **45**: 318-333.
- Moyib, O. K, O. A. Odunloa and A. G. O. Dixon. (2007), SSR markers reveal genetic variation between improved cassava cultivars and landraces within a collection of Nigerian cassava germplasm. *Afr. J. Biotechnol.*, **6**(23): 2670-2674.
- Mughal, M. H., P.S. Saba-Srivastava, and M. Iqbal. (1999), Drumstick (*Moringa pterygisperma*): A unique source of food and medicine. *J. Econ. Taxon. Bot.*, **23**:47-61.
- Nagarajan, K. and M. N. Prasad (1980), Studies in genetic diversity in foxtail millet (*Setaria italica* B.). *Madras Agri. J.*, **67**: 28- 38.

- Ndabigengesere, A. and K.S. Narasiah. (1998), Quality of water treated by coagulation using *Moringa oleifera* seeds. *Water Res.*, **32**:781-791.
- Pawar, A.N. (1995), Genetic Diversity and path analysis in *Amaranthus*. M.Sc., (Ag.) Thesis, Mahatma Phule Krishi Vidyapeeth, Rahuri.
- Rao, C.R. (1952), *Advanced Statistical Methods in Biometrical Research*. John Wiley and Sons, Inc., New York, pp. 390.
- Rohlf, F.J. (1998), NTSYS-PC: Numerical taxonomy and multivariate analysis system. Version 2.02. Exeter Publications Setauket, New York.
- Roldan-Ruiz, I., J. Dendauw, E. VanBockstaele, A. Depicker and M. De Loose. (2000), AFLP markers reveal high polymorphic rates in ryegrasses. (*Lolium* spp.) *Mol. Breed.*, **6**: 125-134.
- Selvam, A.B.D. (2005), Distribution, phenology and utilization of *Moringa oleifera* Lamk.: An indigenous medicinal plant of India. *J. Econ. Taxon. Bot.*, **29**: 102-108.
- Sneath, P.H.A. and R.R. Sokal. (1973), *Numerical Taxonomy: the principles and practice of numerical classification*. Freeman, San Francisco. 573p.
- Von Maydell, H. J. (1986), *Trees and shrubs of sahel: Their characterization and uses*. Deutsche Gesellschaft für Technische Zusammenarbeit, Eschborn, Germany. p. 334-337.
- Walter, R. and B. K. Epperson. (2001), Geographic pattern of genetic variation in *Pinus resinosa*: Area of greatest diversity is not the origin of postglacial populations. *Mol. Ecol.*, **10**:103-111.