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## Research Article

### SSR marker assisted assessment of genetic diversity among mango (*Mangifera indica* L.) genotypes

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#### Abstract

Mango (*Mangifera indica* L.) is one of the most economically important tropical fruit crops, valued for its nutritional quality, sensory attributes, and wide genetic variability. Assessment of genetic diversity among cultivated genotypes is essential for effective germplasm conservation and crop improvement. The present study was conducted to evaluate genetic diversity and relationships among 50 popular mango genotypes cultivated in Telangana, India, using simple sequence repeat (SSR) markers. A total of 60 SSR markers were screened, of which 42 produced clear and reproducible polymorphic amplification. These markers generated a total of 109 alleles. Allele size ranged from 100 bp (MillHR 06b) to 330 bp (MillHR 14b), with an average of 2.21 alleles per locus. Polymorphic information content (PIC) values ranged from 0.24 to 0.80, with a mean of 0.53, indicating moderate to high informativeness of the markers. Major allele frequency varied from 0.36 to 0.94, observed heterozygosity from 0.00 to 0.98, and gene diversity from 0.11 to 0.73, reflecting substantial genetic variation among the genotypes. Cluster analysis using Jaccard's similarity coefficient and UPGMA grouped the genotypes into three major clusters at a similarity coefficient of 0.62. The results demonstrate the effectiveness of SSR markers in discriminating mango genotypes and provide a valuable basis for selecting diverse parental material for mango breeding and improvement programmes.

**Keywords:** Mango, *Mangifera indica*, genetic diversity, SSR markers, molecular characterization, cluster analysis

#### INTRODUCTION

Mango (*Mangifera indica* L.,  $2n = 40$ ) belongs to the family Anacardiaceae and is one of the most important tropical fruit crops cultivated in India, which harbours nearly 1000 cultivars and represents the world's largest mango germplasm resource (Mukherjee, 1950; Karihaloo *et al.*, 2003). Cultivated since prehistoric times, mango occupies approximately 2.29 million ha in India with a production of 20.44 million tonnes, and is widely grown across tropical and subtropical regions (Mitra, 2016; Yamanaka *et al.*, 2019). Major mango-producing countries include India, China, Thailand, Mexico and Pakistan, while within India, Uttar Pradesh, Andhra Pradesh, Odisha, Karnataka and Telangana are the principal mango-growing states (National Horticulture Database, 2023-24). India is recognized as the primary centre of origin and diversity, with domestication dating back nearly 4000 years and extending to the Malay Peninsula in Southeast Asia.

Mango fruits are rich in dietary fibre, antioxidant vitamins A and C, and vitamin B6, and contain bioactive compounds such as triterpenes and lupeol with anticancer properties. In addition, mango leaves are an important source of essential minerals and vitamins and are widely used in traditional medicine for the management of diabetes, asthma and renal disorders (Kumar *et al.*, 2021).

The mango genome size is estimated at  $4.39 \times 10^8$  bp (Arumuganathan and Earle, 1991). Commercial mango cultivars exhibit high heterozygosity and region-specific adaptation, resulting in substantial genetic diversity. Although morphological descriptors have traditionally been used for diversity assessment (IPGRI, 2006), such traits are often environmentally influenced and may yield misleading inferences (Sankar *et al.*, 2011). DNA-based molecular markers provide a more precise

and reliable approach for genetic characterization (Varshney *et al.*, 2004). Among these, simple sequence repeat (SSR) markers are widely preferred due to their co-dominant inheritance, abundance and high reproducibility (Gupta and Varshney, 2000). Despite this, limited information is available on mango-specific SSR-based diversity studies. Telangana possesses rich mango germplasm conserved at the Fruit Research Station, Sangareddy, and the present study was undertaken to assess genetic diversity and relationships among popular mango cultivars of the region using SSR- markers.

## MATERIALS AND METHODS

The experimental material used in the present study comprised of 50 (Table: 1-36 and 37-50 table and juicy cultivars, respectively) cultivars of mango were selected and taken from Fruit Research Station, Sangareddy, Telangana, India (Table 1). These cultivars represent widely grown and commercially important mango genotypes of the region.

**DNA isolation and SSR analysis:** Molecular characterization was carried out at the College of Horticulture, Sri Konda Laxman Telangana State Horticultural University, Rajendranagar; the Institute of Biotechnology, Professor Jayashankar Telangana State Agricultural University, Rajendranagar; and PRR Biotech Private Limited, Mehdiapatnam, Hyderabad. Total genomic DNA was extracted from approximately 100 mg of fresh young leaf tissue using the cetyltrimethyl ammonium bromide (CTAB) method following Doyle and Doyle (1990). DNA quantity was measured using a biophotometer (Eppendorf, India), and quality was assessed by 1% agarose gel electrophoresis. DNA samples were diluted with TE buffer to a final concentration of 50 ng  $\mu\text{l}^{-1}$  and stored at  $-20^{\circ}\text{C}$  until use.

PCR amplification was performed using 60 SSR markers (Table 2) following the protocol described by Sambrook and Russel (2012). The SSR primers were selected based on previously published reports (Ravishankar *et al.*, 2011; Begum *et al.*, 2012; Schnell *et al.*, 2005; Viruel *et al.*, 2005). Amplified products were resolved on 3% agarose gels prepared in 1× TBE buffer at 80 V using a horizontal gel electrophoresis unit, stained with ethidium bromide, and documented using a gel Bio-Rad gel documentation system.

**Data analysis:** Clear and reproducible SSR bands were scored as presence (1) or absence (0) to generate binary data. Genetic parameters, including polymorphic information content (PIC), number of alleles, major allele frequency and gene diversity, were calculated using PowerMarker v3.25. Cluster analysis and similarity matrix construction were performed using NTSYS-pc software version 2.02e (Rohlf, 1998) based on Jaccard's similarity coefficient, and a dendrogram was generated using the Unweighted Pair Group Method with Arithmetic Mean (UPGMA) to visualize genetic relationships among the genotypes.

## RESULTS AND DISCUSSION

Genetic diversity among 50 mango genotypes was assessed using 60 SSR markers, of which 42 primers produced clear and reproducible polymorphic amplification (Table 3). These markers generated a total of 109 alleles, indicating a high level of allelic variation. Similar level of polymorphism was reported earlier by Wahdan *et al.* (2011), who observed polymorphic amplification in 36 of 42 SSR primers among Egyptian mango strains. Allele sizes ranged from 100 bp (MillHR 06b) to 330 bp (MillHR 14b), which is comparable with previously reported ranges of 90–370 bp (Begum *et al.*, 2012) and 100–400 bp (Anshuman Singh *et al.*, 2012) in mango.

The number of alleles per locus varied from two to four, with an average of 2.21 alleles per SSR marker. These results are consistent with earlier reports in mango by Schnell *et al.* (2006), Singh and Bhat (2009), Anshuman *et al.* (2012), Kumar *et al.* (2013) and Malathi *et al.* (2013), who reported allele numbers ranging from moderate to high across different germplasm sets. The polymorphic information content (PIC) values ranged from 0.24 to 0.80, with a mean of 0.53, indicating moderate to high discriminatory power of the SSR markers employed. Markers such as MillHR 19a, MillHR 02c, SSR-39, MiSHRS-32, MillHR 26a and MillHR 32a exhibited higher PIC values, demonstrating their effectiveness in distinguishing mango genotypes. Comparable PIC ranges have been reported by Begum *et al.* (2016) and Ravishankar *et al.* (2011) in mango, further validating the robustness of the marker system used in this study.

Major allele frequency ranged from 0.36 to 0.94, with a mean value of 0.65, corroborating earlier findings by Malathi *et al.* (2013). Observed heterozygosity varied widely from 0.00 to 0.98, with an average of 0.41, reflecting the highly heterozygous nature of mango. Similar heterozygosity trends have been reported in mango by Anju Bajpai *et al.* (2008), Malathi *et al.* (2013) and May Sandar Kyaing *et al.* (2019). Gene diversity values ranged from 0.11 to 0.73, with a mean of 0.45, consistent with previous studies in mango by Anju *et al.* (2008) and Lokesh *et al.* (2018), indicating substantial genetic variation among the evaluated genotypes. The gel images of SSR markers and banding pattern generated in all mango cultivars are given in Fig 1-4.

Cluster analysis based on Jaccard's similarity coefficient and UPGMA grouped the 50 mango genotypes into three major clusters at a similarity coefficient of 0.62 (Table 4; Fig. 5), revealing considerable genetic divergence could be attributed to the cross-pollinated nature of mango crop. Similarity coefficients ranged from 0.61 to 0.88, with the highest similarity observed between Mahamooda Vikarabad and Manjeera, followed by Ranitellakaya and Shajahan, and Baneshan and Vaddepalli Selection. Cluster I was the largest, comprising 43 genotypes further subdivided into three sub-clusters. Sub-cluster IA with 0.66 similarity comprised 21 cultivars, primarily table

Table 1. List of mango cultivars and collection site

S.No.	Genotypes	Sampling Unit	Collection Site
1	Dashehari 35	Mother Block FRS	Sangareddy, Telangana
2	Allampur Baneshan	Mother Block FRS	Sangareddy, Telangana
3	Asif Us Samar	Mother Block FRS	Sangareddy, Telangana
4	Azam Us samar	Mother Block FRS	Sangareddy, Telangana
5	Baneshan	Mother Block FRS	Sangareddy, Telangana
6	Chinna Suvarnarekha	Mother Block FRS	Sangareddy, Telangana
7	Dashehari	Mother Block FRS	Sangareddy, Telangana
8	Dilpasand	Mother Block FRS	Sangareddy, Telangana
9	Goa Bandar	Mother Block FRS	Sangareddy, Telangana
10	Himayath	Mother Block FRS	Sangareddy, Telangana
11	Jehangir	Mother Block FRS	Sangareddy, Telangana
12	Kaju	Mother Block FRS	Sangareddy, Telangana
13	Kalepahad	Mother Block FRS	Sangareddy, Telangana
14	Kesar	Mother Block FRS	Sangareddy, Telangana
15	Lalmuni	Mother Block FRS	Sangareddy, Telangana
16	Latif Us Samar	Mother Block FRS	Sangareddy, Telangana
17	Mahamooda Uppal	Mother Block FRS	Sangareddy, Telangana
18	Mahamooda Vikarabad	Mother Block FRS	Sangareddy, Telangana
19	Manjeera	Mother Block FRS	Sangareddy, Telangana
20	Mulgoa	Mother Block FRS	Sangareddy, Telangana
21	Nazeem Pasand	Mother Block FRS	Sangareddy, Telangana
22	Neeleshan	Mother Block FRS	Sangareddy, Telangana
23	Neelum	Mother Block FRS	Sangareddy, Telangana
24	Parasapalli Doodiya	Mother Block FRS	Sangareddy, Telangana
25	Pulihora	Mother Block FRS	Sangareddy, Telangana
26	Ranitellakaya	Mother Block FRS	Sangareddy, Telangana
27	Rumani	Mother Block FRS	Sangareddy, Telangana
28	Sannakulu	Mother Block FRS	Sangareddy, Telangana
29	Shajahan	Mother Block FRS	Sangareddy, Telangana
30	Shendriya	Mother Block FRS	Sangareddy, Telangana
31	Sora	Mother Block FRS	Sangareddy, Telangana
32	Suvarnarekha	Mother Block FRS	Sangareddy, Telangana
33	Totapari	Mother Block FRS	Sangareddy, Telangana
34	Vaddepalli Selection	Mother Block FRS	Sangareddy, Telangana
35	Vanraj	Mother Block FRS	Sangareddy, Telangana
36	Yerra Mulgoa	Mother Block FRS	Sangareddy, Telangana
37	Aryavrtham Irsalu	Mother Block FRS	Sangareddy, Telangana
38	Cherukuramam	Mother Block FRS	Sangareddy, Telangana
39	Chinnarasam	Mother Block FRS	Sangareddy, Telangana
40	Kothapalli Kobbari	Mother Block FRS	Sangareddy, Telangana
41	Meetavari Peechumanu	Mother Block FRS	Sangareddy, Telangana
42	Nagulapalli Irsalu	Mother Block FRS	Sangareddy, Telangana
43	Navaneetham	Mother Block FRS	Sangareddy, Telangana
44	Panakalu	Mother Block FRS	Sangareddy, Telangana
45	Panchavarnam	Mother Block FRS	Sangareddy, Telangana
46	Pandurivari Mamidi	Mother Block FRS	Sangareddy, Telangana
47	Peddarasam	Mother Block FRS	Sangareddy, Telangana
48	Yellow Arati	Mother Block FRS	Sangareddy, Telangana
49	Yerra Arati	Mother Block FRS	Sangareddy, Telangana
50	Zardalu	Mother Block FRS	Sangareddy, Telangana

Note: 1-36 and 37-50 table and juicy cultivars, respectively

Table 2. SSR markers used for DNA amplification in 50 mango genotypes

S. No.	Marker	Forward	Reverse
1	MiIHR 01a	GGATGCACAACAACAAGCAC	TCAGCAAGCAATCCCTTCTT
2	MiIHR 02c	CCCCAACATTCATAAACACA	CCTCCTTACATGCCTCCTTG
3	MiIHR 03a	GTCGATGCCTGGAATGAAGT	AAGCATCGAACAGCTCCAAT
4	MiIHR 04c	CGTTTTTGACCCTCTTGAGC	CCGCATACTTCCCTTCACAT
5	MiIHR 05c	CTCTCCCTCACTTGCTCCAC	AGACCACCGACAACGAAAAC
6	MiIHR 06 b	CGCCGAGCCTATAACCTCTA	ATCATGCCCTAAACGACGAC
7	MiIHR 08 b	TGCTCTCTACTGCCCGGTAT	GTCACACCAATCGGGAATCT
8	MiIHR 11a	CAGTGAAACCACCGGTCAA	TGGCCAGCTGATACCTTCTT
9	MiIHR 12a	GCCCCATCAATACGATTGTC	ATTTCCCACCATTGTGCTTG
10	MiIHR 13	CCCAGTTCCAACATCATCAG	TTCTCTGGAAGAGGGAAGA
11	MiIHR 14 b	CCGAAACAACCTTCTCTCCA	TGCTCTCTGGCCTCTTCTTC
12	MiIHR 15b	CTAACCATTTCGGCATCCTCT	TCTGTGATAGAATGGCAAAGAA
13	MiIHR 16a	TTTCACTTGGTTCTGGATTGC	ATTTCCCACCATTGTGCTTG
14	MiIHR 17 b	GCTTGCTTCCAACCTGAGAC	GCAAAATGCTCGGAGAAGAC
15	MiIHR 18 b	TCTGAGTCACTCCTTTTCA	ATACTCGTGCCTCGTCTGT
16	MiIHR 19a	TGATATTTTCAGGGCCCAAG	AAATGGCACAAGTGGGAAAG
17	MiIHR 22a	TGGCCGAAGTAGCAAACCTCT	CCCCATTTGAGAAAATTCC
18	MiIHR 23a	TCTGACCCAACAAGAACCA	TCCTCCTCGTCTCATCATC
19	MiIHR 24 b	GCTCAACGAACCCAACCTGAT	TCCAGCATTCAATGAAGAAGTT
20	MiIHR 25a	TGTGAGTCTCCGTTTGTGCT	CCCTCTCATTTTCCCAGTCA
21	MiIHR 26a	GCGAAAGAGGAGAGTGCAAG	TCTATAAGTGCCCCCTCACG
22	MiIHR 28c	GCGGTCGCAGACAAATCTATA	ACAAGTCGAGATTGTCACATCTTT
23	MiIHR 29a	CGATGAGGATGGTTGGTTTT	CATCAACAGTCGCCATCAAT
24	MiIHR 30a	AGCTATCGCCACAGCAAATC	GTCTTCTTCTGGCTGCCAAC
25	MiIHR 31 b	TTCTGTTAGTGGCGGTGTTG	CACCTCCTCCTCCTCCTCTT
26	MiIHR 32a	TGGTGGTGTGTTGTTGCACT	ACCACCCGCAGTATTGAAAG
27	MiIHR 33a	GAAGCACTTGTCTCCCTTGC	CCTCACACTCCTCCACCTGT
28	MiIHR 34 b	CTGAGTTTGGCAAGGGAGAG	TTGATCCTTACCACCATCA
29	MiIHR 35a	TGGTGAAGCTTGTGTCTGC	TGGCTTGACTGTTTTTCAGC
30	MiIHR 36 b	TCTATAAGTGCCCCCTCACG	ACTGCCACCGTGAAAGTAG
31	SSR-15	TTTACCAAGCTAGGGTCA	CACTCTTAAACTATTCAACCA
32	SSR-18	CGTCATCCTTACAGCGAACT	CATCTTTGATCATCCGAAAC
33	SSR-20	CGCTCTGTGAGAATCAAATGGT	GGACTCTTATTAGCCAATGGGATG
34	SSR-36	CCTCAATCTCACTCAACA	ACCCCAACATAAATCTAC
35	SSR-39	TGTCTACCATCAAGTTCCG	GCTGTTGTTGCTTTACTG
36	SSR-46	TCATTGCTGTCCCTTTTC	ATCGCTCAACAATCC
37	SSR-52	AAAAACCTTACATAAGTGAATC	CAGTTAACCTGTTACTTTTTT
38	SSR-55	ATATCTCACGGCTTCAATGA	TATTAATTTTACAGACTATGTTCA
39	SSR-57	CATGGAGTTGTGATACCTAC	CAGAGTTAGCCATATAGAGTG
40	SSR-60	ATTATTTACCCTACAGAGTGC	GTATTATCGGTAATGTCTTCAT
41	SSR-61	AAAGATAGCATTAAATTAAGGA	GTAAGTATCGCTGCTGTTTGTATT
42	SSR-65	ATAGATTCATATCTTCTTGCAT	TATAAATTATCATCTTCACTGC
43	SSR-82	TCTGACCCAACAAGAACCA	TCCTCCTCGTCTCATCATC
44	SSR-83	AGCTATCGCCACAGCAAATC	GTCTTCTTCTGGCTGCCAAC
45	SSR-84	TCTATAAGTGCCCCCTCACG	ACTGCCACCGTGAAAGTAG
46	SSR-88	CTGAGTTTGGCAAGGGAGAG	TTGATCCTTACCACCATCA
47	MiSHRS-1	TAACAGCTTTGCTTGCCTCC	TCCGCCGATAAACATCAGAC
48	MiSHRS-4	CCACGAATATCAACTGCTGCC	TCTGACACTGCTTCCACC
49	MiSHRS-32	TTGATGCAACTTTCTGCC	ATGTGATTGTTAGAATGAACCT
50	MiSHRS-36	GTTTTTATTCTCAAAATGTGTG	CTTTCATGTTTATAGATGCAA
51	MiSHRS-44	AACCCATCTAGCCAACCC	TTGACAGTTACCAACAGCAC
52	MiSHRS-48	TTTACCAAGCTAGGGTCA	CACTCTTAAACTATTCAACCA
53	LMMA-1	ATGGAGACTAGAATGTACAGAG	ATTAAATCTCGTCCACAAGT
54	LMMA-2	AAATAAGATGAAGCAACTAAAG	TTAGTGATTTTGTATGTTCTTG
55	LMMA-4	AAAAACCTTACATAAGTGAATC	CAGTTAACCTGTTACCTTTTT
56	LMMA-6	ATATCTCAGGCTTCAATGA	TATTAATTTTACAGACTATGTTCA
57	LMMA-7	ATTTAACTCTTCAACTTTCAAC	AGATTTAGTTTTGATTATGGAG
58	LMMA-8	CATGGAGTTGTGATACCTAC	CAGAGTTAGCCATATAGAGTG
59	LMMA-9	TTGCAACTGATAACAAATATAG	TTACATGACAGATATACACTT
60	MngSSR-14	TCATTAAGCTGTGGCAACCA	CATTGCATAGATGTGGTCATT

Table 3. Polymorphic SSR markers used for characterization of 50 mango genotypes

S. No.	Marker	Product size in bp	Number of alleles	PIC values	Major allele frequency	Heterozygosity	Gene diversity
1	MilIHR 01a	240-250	2	0.45	0.84	0.32	0.27
2	MilIHR 02c	190-220	3	0.76	0.38	0.40	0.66
3	MilIHR 04c	160-180	2	0.38	0.76	0.48	0.36
4	MilIHR 05c	190-210	2	0.36	0.74	0.52	0.38
5	MilIHR 06 b	100-120	2	0.46	0.86	0.28	0.24
6	MilIHR 12a	170-180	2	0.38	0.76	0.48	0.36
7	MilIHR 13	170-190	2	0.53	0.71	0.30	0.41
8	MilIHR 14 b	320-330	2	0.24	0.64	0.72	0.46
9	MilIHR 15b	130-160	3	0.69	0.71	0.38	0.45
10	MilIHR 16a	210-220	2	0.47	0.65	0.54	0.46
11	MilIHR 17 b	220-240	2	0.44	0.67	0.46	0.44
12	MilIHR 18 b	170-185	2	0.56	0.94	0.00	0.11
13	MilIHR 19a	170-210	4	0.80	0.36	0.69	0.73
14	MilIHR 23a	130-155	3	0.50	0.48	0.98	0.62
15	MilIHR 24 b	240-250	2	0.31	0.64	0.64	0.46
16	MilIHR 25a	145-150	2	0.54	0.65	0.34	0.46
17	MilIHR 26a	140-160	3	0.71	0.49	0.56	0.63
18	MilIHR 28c	120-130	2	0.57	0.59	0.30	0.48
19	MilIHR 30a	190-200	2	0.61	0.59	0.26	0.48
20	MilIHR 31 b	250-265	2	0.52	0.73	0.30	0.39
21	MilIHR 32a	200-210	2	0.71	0.60	0.08	0.48
22	MilIHR 33a	160-170	2	0.52	0.53	0.38	0.50
23	MilIHR 36 b	220-230	2	0.52	0.70	0.32	0.42
24	SSR-20	300-310	2	0.39	0.74	0.48	0.38
25	SSR-36	230-240	2	0.62	0.55	0.30	0.50
26	SSR-39	170-190	3	0.75	0.52	0.40	0.57
27	SSR-46	190-200	2	0.56	0.68	0.28	0.44
28	SSR-52	120-210	2	0.48	0.58	0.44	0.49
29	SSR-55	120-130	2	0.42	0.77	0.42	0.35
30	SSR-57	260-270	2	0.38	0.65	0.62	0.46
31	SSR-65	240-260	3	0.56	0.54	0.86	0.60
32	SSR-82	120-140	3	0.56	0.56	0.82	0.58
33	SSR-83	190-200	2	0.50	0.60	0.40	0.48
34	SSR-88	180-200	2	0.47	0.83	0.30	0.28
35	MiSHRS-1	190-200	2	0.67	0.75	0.06	0.38
36	MiSHRS-4	150-160	2	0.60	0.70	0.20	0.42
37	MiSHRS-32	190-200	2	0.74	0.52	0.04	0.51
38	MiSHRS-36	180-190	2	0.59	0.69	0.24	0.43
39	MiSHRS-48	210-220	2	0.65	0.57	0.18	0.49
40	LMMA-6	110-120	2	0.51	0.78	0.28	0.34
41	LMMA-8	250-260	2	0.25	0.58	0.72	0.49
42	MngSSR-14	170-180	2	0.63	0.51	0.22	0.50
<b>Total/Mean</b>		-	<b>109</b>	<b>0.53</b>	<b>0.65</b>	<b>0.41</b>	<b>0.45</b>

purpose varieties. Notably, the highest genetic similarity within this group was observed between Ranitellakaya and Shajahan (86%), followed by Baneshan and Vaddepalli Selection (83%). The inclusion of the new clone Dashehari-35 alongside Dashehari (0.73 similarity) validates its genetic identity for cultivation in Telangana. Sub-cluster IB at 0.67 similarity a heterogeneous group of 20 table and juicy cultivars. Mahamooda Vikarabad and Manjeera exhibited the highest similarity (88%), indicating

identical allelic profiles. Sub-cluster IC with 0.70 similarity contained two prominent juicy cultivars, chinna rasam and Navaneetham. While Cluster II included four genotypes and Cluster III comprised three genotypes.

The selection and hybridization programmes in mango can be affected based on the clustering. The clustering pattern revealed that genotypes did not segregate strictly according to their utility (table or juicy types) at

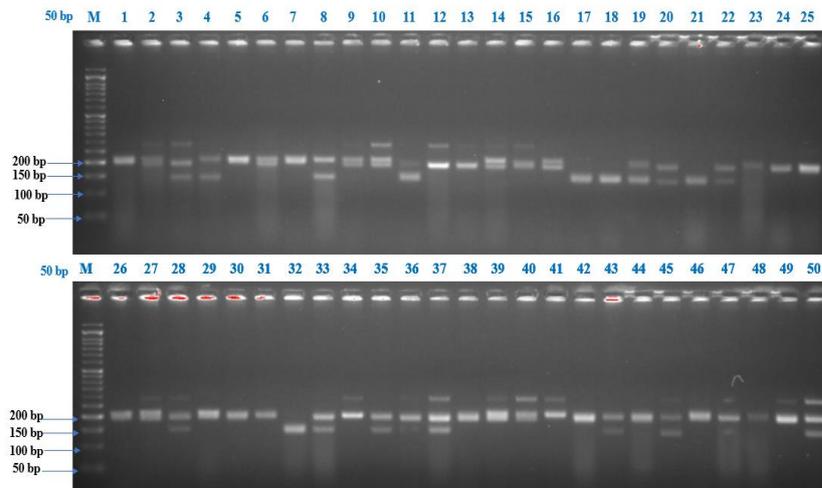


Fig. 1. SSR amplification profile of 50 mango genotypes generated using primer MillHR 19a

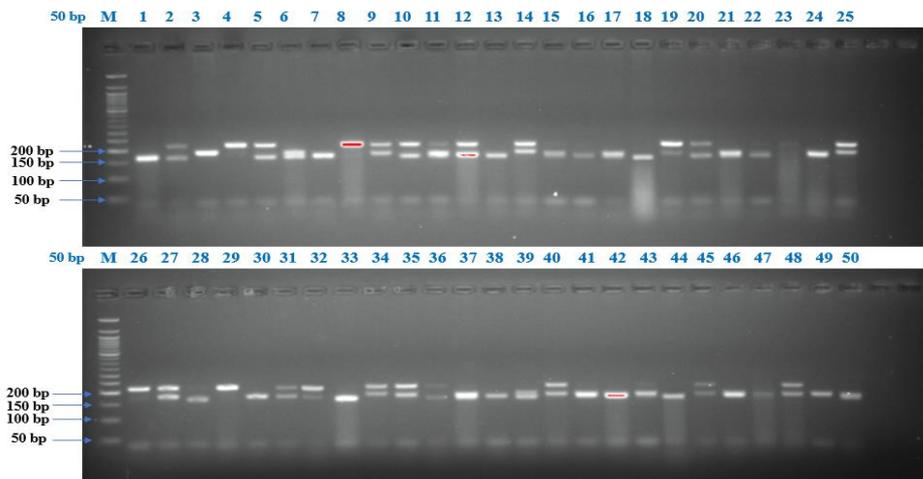


Fig. 2. SSR amplification profile of 50 mango genotypes generated using primer MillHR 02c

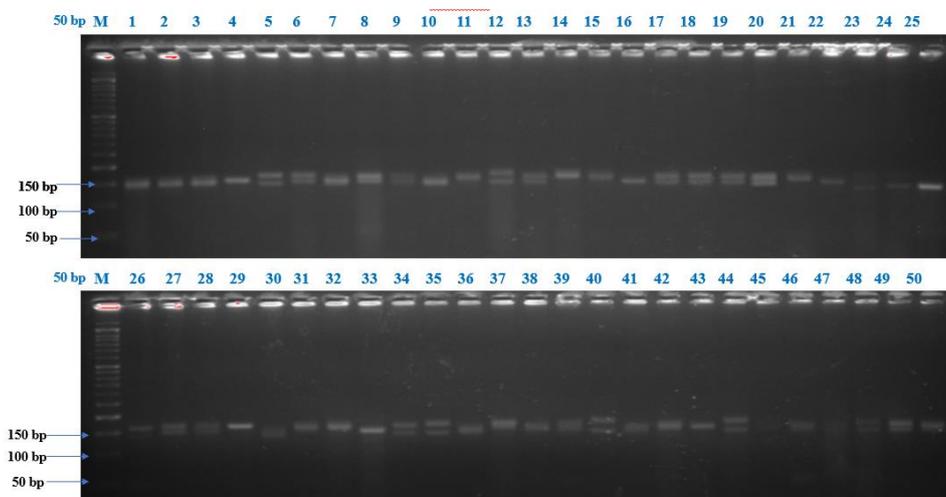


Fig. 3. SSR amplification profile of 50 mango genotypes generated using primer MillHR 26a

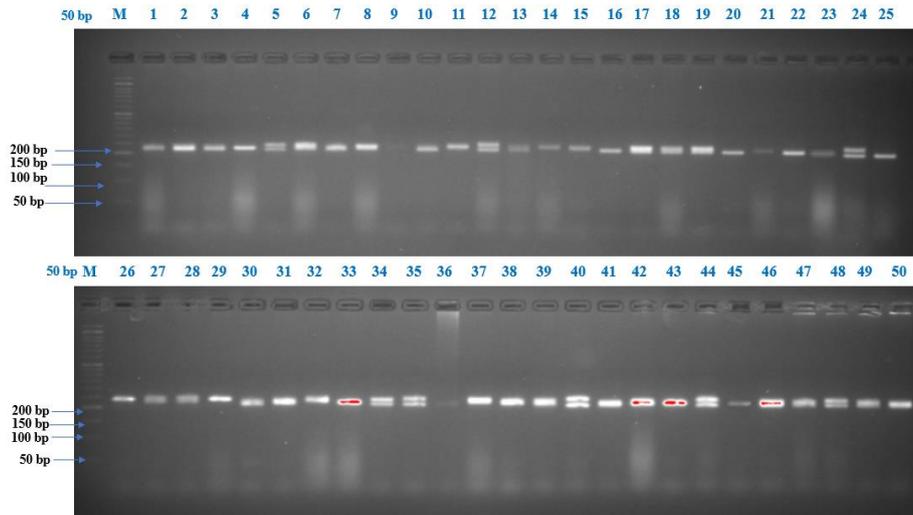


Fig. 4. SSR amplification profile of 50 mango genotypes generated using primer SSR-36. (Refer Table 1 for name of the genotypes)

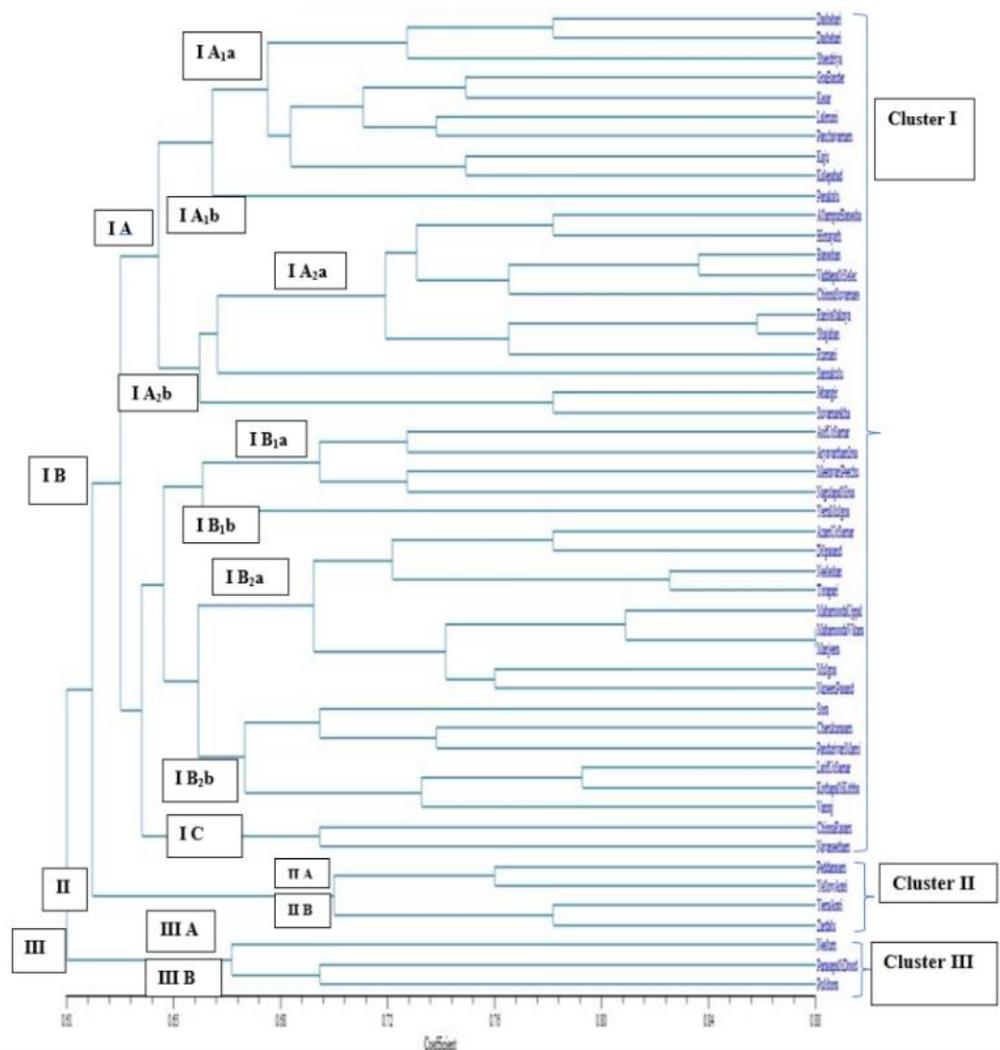


Fig. 5. Dendrogram constructed using Jaccard's similarity coefficient based on UPGMA

Table 4. Cluster-wise grouping of mango genotypes

Cluster	Genotypes	Number of genotypes	
Cluster IA (21 genotypes)	Cluster IA <sub>1</sub> a	Dashehari-35, Dashehari, Goa Bander, Kaju, Kalepahad, Kesar, Lalmuni, Panchavarnam, Shendriya	9
	Cluster IA <sub>1</sub> b	Panakalu	1
	Cluster IA <sub>2</sub> a	Allampur Baneshan, Baneshan, Chinna Suvarnarekha, Himayath, Ranitellakaya, Rumani, Vaddepalli Selection, Shajahan	8
	Cluster IA <sub>2</sub> b	Jehangir, Sannakulu, Suvarnarekha	3
Cluster IB (20 genotypes)	Cluster IB <sub>1</sub> a	Aryavartham Irsalu, Asif Us Samar, Meetavari Peechumanu, Nagulapalli Irsalu	4
	Cluster IB <sub>1</sub> b	Yerra Mulgoa	1
	Cluster IB <sub>2</sub> a	Azam Us Samar, Dilpasand, Mahamooda Uppal, Mahamooda Vikarabad, Manjeera, Mulgoa Nazeem Pasand, Neeleshan, Totapari,	9
	Cluster IB <sub>2</sub> b	Cherukurasam, Kothapalli Kobbari, Latif Us Samar, , Pandurivari Mamidi, Sora, Vanraj	6
Cluster IC (2 genotypes)	Chinna Rasam, Navaneetham	2	
Cluster II (4 genotypes)	Cluster II A <sub>1</sub>	Pedda Rasam, Yellow Arati	2
	Cluster II A <sub>2</sub>	Yerra Arati, Zardalu	2
Cluster III (3 genotypes)	Cluster IIIA <sub>1</sub>	Neelum	1
	Cluster IIIA <sub>2</sub>	Parasapalli Doodiya, Pulihora	2

the major cluster level, but rather at sub-cluster levels. Understanding the genetic diversity among the varieties is important in mango production, improvement and breeding, knowledge on this field can supply useful information for further scientific progress in developing new genotypes (Rajwana *et al.*, 2010). Hybrids such as Manjeera and Neeleshan grouped closely with one of their parental lines, while the other parent was positioned in a different cluster, indicating considerable parental divergence. Similar observations were reported by Shareefa (2008) and Malathi *et al.* (2018). Genotypes originating from the same geographical region were distributed into different sub-clusters. This pattern indicates that the clustering of cultivars was largely independent of geographical boundaries, demonstrating that geographic isolation alone is not the sole determinant of genetic diversity. These findings are in agreement with Eiadthong *et al.* (2000). The inability to distinctly separate table and juicy cultivars corroborates earlier reports by Rahman *et al.* (2007); Anju *et al.* (2008); Kumar *et al.* (2013); Ariffin *et al.* (2015); Begum *et al.* (2016) and Lokesh *et al.* (2018) in mango.

Overall, the SSR-based clustering and diversity parameters highlight substantial genetic variability among mango genotypes of Telangana, underscoring the effectiveness of SSR markers in resolving genetic relationships and identifying diverse parental material for mango improvement programmes.

The present study demonstrates that SSR markers are highly effective in resolving genetic diversity and

relationships among mango genotypes cultivated in Telangana. The moderate to high polymorphism detected, particularly with markers such as MillHR 19a, MillHR 02c, SSR-39, MiSHRS-32, MillHR 26a and MillHR 32a, highlights their utility for molecular characterization and germplasm management. UPGMA-based clustering revealed substantial genetic divergence among genotypes, with grouping patterns independent of fruit usage type and geographical origin. The use of a larger number of SSR markers with broader genome coverage could enable a more accurate assessment of genetic diversity. Overall, the findings confirm the robustness of SSR-based diversity analysis and provide a reliable foundation for the selection of genetically diverse parental material, conservation of mango germplasm, and future applications in mango breeding, genetic purity assessment and marker-assisted improvement programmes.

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#### REFERENCES

- Anju, B., Chandra, R., Rajan, S. and Navin, S. 2008. RAPD and minisatellite markers for genetic diversity and relationship in guava varieties. *Indian Journal of Genetics*, **68**: 441–445.

- Anshuman, S., Singh, A. and Singh, S.K. 2012. SSR markers reveal genetic diversity in closely related mango hybrids. *Indian Journal of Horticulture*, **69**(3): 299–305.
- Ariffin, Z., Sah Md., Idris, M.S. and Hashim, N. 2015. Genetic diversity of selected *Mangifera* species revealed by inter simple sequence repeat markers. *International Journal of Biodiversity*, **2015**: 1–8. [Cross Ref]
- Arumuganathan, K. and Earle, E.D. 1991. Nuclear DNA content of some important plant species. *Plant Molecular Biology Reports*, **9**: 208–218. [Cross Ref]
- Begum, H., Medagam, T.R., Surapaneni, M., Boreddy, P.R., Gonela, N., Javaregowda, N. and Ebrahimali, A.S. 2016. Morphological variability and microsatellite diversity of cultivated mango (*Mangifera indica* L.) from Andhra Pradesh, India. *Jordan Journal of Agricultural Sciences*, **12**(3): 815–843. [Cross Ref]
- Begum, H., Medagam, T.R., Surapaneni, M., Boreddy, P.R., Gonela, N., Javaregowda, N. and Ebrahimali, A.S. 2012. Molecular analysis of intracultivar polymorphism in 'Peddarasam' mango (*Mangifera indica* L.) using microsatellite markers. *Asia Pacific Journal of Molecular Biology and Biotechnology*, **21**: 97–113.
- Doyle, J.J. and Doyle, J.L. 1990. Isolation of plant DNA from fresh tissue. *Focus*, **12**: 13–15. [Cross Ref]
- Eiadthong, W., Yonemori, K., Sugiura, A., Utsunomiya, N. and Subhadrabandhu, S. 2000. Identification of mango cultivars of Thailand and evaluation of their genetic variation using SSR-anchored primers. *Scientia Horticulturae*, **82**: 57–66. [Cross Ref]
- Gupta, P.K. and Varshney, R.K. 2000. The development and use of microsatellite markers for genetic analysis and plant breeding with emphasis in bread wheat. *Euphytica*, **113**: 163–185. [Cross Ref]
- IPGRI. 2006. Descriptors for Mango (*Mangifera indica* L.). International Plant Genetic Resources Institute, Rome, Italy.
- Karihaloo, J.L., Dwivedi, Y.K., Sunil, A., Ambika, B. and Gaikwad. 2003. Analysis of genetic diversity of Indian mango cultivars using RAPD markers. *Journal of Horticultural Science and Biotechnology*, **78**(3): 285–289. [Cross Ref]
- Kumar, M., Ponnuswami, V., Nagarajan, P., Jeyakumar, P. and Senthil, N. 2013. Molecular characterization of ten mango cultivars using simple sequence repeat markers. *African Journal of Biotechnology*, **12**(47): 6568–6573. [Cross Ref]
- Kumar, M., Saurabh, V., Tomar, M., Hasan, M., Changan, S., Sasi, M., Maheshwari, C., Prajapati, U., Singh, S., Prajapat, R.K. and Dhumal, S. 2021. Mango (*Mangifera indica* L.) leaves: Nutritional composition, phytochemical profile and health-promoting bioactivities. *Antioxidants*, **10**(2): 299. [Cross Ref]
- Lokesh, B., Singh, A.K. and Singh, C.P. 2018. Characterization of mango (*Mangifera indica* L.) genotypes based on physico-chemical quality attributes. *Journal of Applied and Natural Science*, **9**(4): 2199–2204. [Cross Ref]
- Malathi, S., Lakshminarayan, R.V., Hameedunnisa, B., Purushotham, R., Neetasri, B., Nagaraju, C., Anwar, J. and Siddiq, E.A. 2013. Population structure and genetic analysis of different utility types of mango (*Mangifera indica* L.) germplasm of Andhra Pradesh using microsatellite markers. *Plant Systematics and Evolution*, **299**(7): 1215–1229. [Cross Ref]
- Malathi, S., Lakshminarayan, R.V., Hameedunnisa, B., Purushotham, R., Neetasri, B., Nagaraju, C., Anwar, J. and Siddiq, E.A. 2018. Genetic variability and DNA fingerprinting of elite mango genotypes of India using microsatellite markers. *Proceedings of the National Academy of Sciences, India, Section B: Biological Sciences*, **89**(2): 1–8. [Cross Ref]
- May, S.K., April, N.Y.S., Moe, M.M., Honey, T.P.H., Khin, P.Y., Seinn, S.M.P. and Nwe, S.H. 2019. Morphological and molecular analysis of indigenous Myanmar mango (*Mangifera indica* L.) landraces around Kyaukse district. *Journal of Plant Biotechnology*, **46**: 61–70. [Cross Ref]
- Mitra, S.K. 2016. Mango production in the world—present situation and future prospects. *Acta Horticulturae*, **1111**: 287–296. [Cross Ref]
- Mukherjee, S.K. 1950. Mango: Its allopolyploid nature. *Nature*, **166**(4213): 196–197. [Cross Ref]
- National Horticulture Board. 2023–2024. *National Horticulture Database*. Ministry of Agriculture, Government of India, Gurugram, India.
- Rajwana, I.A., Aman, U.M., Ahmad, S.K. and Ahmad, M. 2010. A new mango hybrid shows better shelf life and fruit quality. *Pakistan Journal of Botany*, **42**(4): 2503–2512.
- Rahman, M.L., Rabbani, M.G., Siddique, M.N.A., Rahman, M.A., Garvey, E.J. and Rahaman, E.H.M.S. 2007. Molecular characterization of 28 mango germplasm using RAPD markers. *Plant Tissue Culture and Biotechnology*, **17**(1): 71–77. [Cross Ref]
- Ravishankar, K.V., Mani, H.R., Lalitha, A. and Dinesh, M.R. 2011. Development of new microsatellite markers from mango (*Mangifera indica* L.) and cross-species amplification. *American Journal of Botany*, **98**: 96–98. [Cross Ref]

- Rohlf, F.J. 2000. *NTSYS-pc: Numerical taxonomy and multivariate analysis system*, Version 2.1. Exeter Software, Setauket, New York, USA.
- Sambrook, J. and Russell, D.W. 2012. *Molecular Cloning: A Laboratory Manual*. 4th Edition. Cold Spring Harbor Laboratory Press.
- Sankar, J., Rosaiah, G., Kurapati, R.K. and Pinnamaneni, R. 2011. Molecular identification of mango (*Mangifera indica* L.) var. Totapuri. *Bioinformation*, **5**: 405–409. [\[Cross Ref\]](#)
- Schnell, R.J., Brown, J.S., Olano, C.T., Meerow, A., Campbell, R.J. and Kuhn, D.N. 2006. Mango genetic diversity analysis and pedigree inferences for Florida cultivars using microsatellite markers. *Journal of the American Society for Horticultural Science*, **131(2)**: 214–224. [\[Cross Ref\]](#)
- Shareefa, M. 2008. DNA fingerprinting of mango (*Mangifera indica* L.) genotypes using molecular markers. Ph.D. Thesis, Indian Agricultural Research Institute, New Delhi.
- Singh, S. and Bhat, K.V. 2009. Molecular characterization and analysis of geographical differentiation of Indian mango (*Mangifera indica* L.) germplasm. *Acta Horticulturae*, **839**: 599–606. [\[Cross Ref\]](#)
- Varshney, A., Mohapatra, T. and Sharma, R.P. 2004. Molecular mapping and marker-assisted selection of traits for crop improvement. In: Srivastava, A. and Narula, S. (Eds.), *Plant Biotechnology and Molecular Markers*. Anamaya Publishers, New Delhi, pp. 289. [\[Cross Ref\]](#)
- Viruel, M.A., Escribano, P., Barbieri, M., Ferri, M. and Hormaza, J.I. 2005. Fingerprinting, embryo type and geographic differentiation in mango (*Mangifera indica* L., Anacardiaceae) with microsatellites. *Molecular Breeding*, **15**: 383–393. [\[Cross Ref\]](#)
- Wahdan, M.T., Abdelsalam, A.Z., El-Naggar, A.A. and Hussein, M.A. 2011. Preliminary horticultural studies to describe and identify two new Egyptian mango strains using DNA fingerprinting. *Journal of American Science*, **7(2)**: 641–650.
- Yamanaka, S., Hosaka, F., Matsumura, M., Onoue-Makishi, Y., Nashima, K., Urasaki, N., Ogata, T., Shoda, M. and Yamamoto, T. 2019. Genetic diversity and relatedness of mango cultivars assessed by SSR markers. *Breeding Science*, **69 (2)**: 332–344. [\[Cross Ref\]](#)