Indian Journal of Agricultural Sciences **93** (10): 1080–1085, October 2023/Article
https://doi.org/10.56093/ijas.v93i10.132658

SSR marker based profiling and population structure analysis in peach (*Prunus persica*) germplasm

RAJENDER KUMAR[1]*, D C DIMRI[1], KANCHAN KARKI[2], K M RAI[3], N K SINGH[1], JITENDRA SINGH SHIVRAN[1] and SWAPNIL BHARTI[1]

G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand 26 3145, India

Received: 26 January 2023; Accepted: 21 August 2023

ABSTRACT

For breeding programmes to be successful and for germplasm conservation, it is essential to characterize and analyze the genetic diversity of available germplasm. The present experiment was conducted at Molecular Biology and Genetic Engineering Laboratory of Uttarakhand Council for Biotechnology, Haldi, Uttarakhand during 2022 to study the molecular profile of 41 peach [*Prunus persica* (L.) Stokes] accessions using 23 polymorphic SSR markers. The number of alleles detected ranged from 3 to 8 with an average of 4.65 alleles per locus (Na) and a total of 107 alleles were amplified. The average effective number of alleles (Ne) were 2.89 per marker. The SSR marker MA015a produced maximum number of 8 alleles followed by BPPCT 015 and CPPCT14 which produced 7 alleles each. The polymorphic information content (PIC) varied between 0.317–0.836 with a mean value of 0.563. The observed heterozygosity examined was lower (Ho = 0.02) and the expected heterozygosity (He = 0.61) ranged between 0.34 to 0.85. The presence of a higher Shannon's information index (I) of 1.17 indicates higher diversity in the given set of peach genotypes. Jaccard's similarity coefficient ranging from 0.533 to 1, indicated a pair-wise relationship among the peach accessions. The cluster dendrogram partitioned the accessions into two main clusters. However, the total accessions were stratified into 3 groups (K=3) based on population structure analysis which was further confirmed by Principal Coordinate Analysis (PCoA). The information generated in the study may have great implications in molecular characterization, fingerprinting and documentation of accessions in the peach improvement programme.

Keywords: Cluster, Genetic diversity, Population structure, SSR marker

Peach [*Prunus persica* (L.) Stokes] is a species of genus *Prunus* belonging to the family Rosaceae and is diploid having chromosome number 2n = 2x = 16. It is considered to have originated in China (Faust and Timon 1995), however, few researchers suggest its origin in Persia. The primary center of peach diversity is in Tibet and southwest China, while Iran is a secondary center of diversity (Kumar *et al*. 2013). Genetic diversity plays an important role in ensuring diverse and novel alleles for yield, quality, biotic and abiotic stresses. Therefore, the evaluation of germplasm accessions is essential for determining genetic worth, reducing redundancies, building a core collection, effective comprehensive characterization and their utilization in genetic improvement programme (Dagnon 2022). A traditional approach of characterization and assessment of genetic diversity in perennial fruit crops is time-consuming and is highly influenced by environmental factors. The use of molecular markers has many advantages over phenotype based characterization with the potential to unravel even minor variations at the molecular level based on the differences in DNA and also determine the relationships among varieties of crops as well as crop populations. Molecular markers such as microsatellites or simple sequence repeats (SSRs) are tandemly repeated units of DNA that are abundant and widespread in a plant's genome and due to their co-dominant inheritance, high polymorphism, high discriminating power, relatively low cost and high reliability, SSR markers have proved invaluable in peach breeding programmes (Kumar *et al*. 2023).

In India, limited investigations have been conducted to characterize the available peach germplasm on the basis of molecular data and; to determine the molecular relationship among the germplasm for accelerating future breeding programmes. To develop new peach cultivars having desirable fruiting traits such as non-melting, non-browning and blood-red flesh with slow-ripening and peento types fruit (Kumar *et al*. 2015), it is important to understand the inheritance pattern of these traits as well as to identify

[1]G. B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand; [2]Uttarakhand Council for Biotechnology, Haldi, Pantnagar, Uttarakhand; [3]ICAR-National Bureau of Plant Genetic Resources, Regional Station, Bhowali, Nainital, Uttarakhand. *Corresponding author email: rajender.horti@gmail.com

possible donors. The aim of the present study was develop a molecular profile of 41 peach accessions using SSR markers and further determine the molecular relationship among accessions using marker data.

MATERIALS AND METHODS

A study was carried out at Molecular Biology and Genetic Engineering Laboratory of Uttarakhand Council for Biotechnology, Haldi, Pantnagar, Uttarakhand, during 2022 for molecular characterization of 41 peach genotypes. The experimental materials consisted of low and high chill peach accessions (Table 1). The low chill peaches consisting of 15 accessions are maintained at Horticulture Research Center, Patharchatta of G. B. Pant University of Agriculture and Technology, Pantnagar, US Nagar, Uttarakhand, whereas 26 high chill peach accessions are maintained at ICAR-National Bureau of Plant Genetic Resources, Regional Station, Bhowali, Nainital, Uttarakhand.

DNA extraction: The genomic DNA was extracted from fresh young leaves following the cetyltrimethyl ammonium bromide (CTAB) protocol (Doyle and Doyle 1987) with some minor modifications. Quality and quantity of genomic DNA was estimated through a NanoDrop spectrophotometer (Thermo Scientific, USA) and was diluted to 50 ng/µl to carry out the PCR amplification reactions.

Primers used and PCR amplification: A set of 25 SSR primers selected from earlier studies (Cipriani *et al*. 1999, Sosinski *et al*. 2000, Aranzana *et al*. 2002, Dirlewanger *et al*. 2002, Yamamoto *et al*. 2002) based on their reproducibility and high discrimination power was used in the molecular profiling of peach accessions using genomic DNA. PCR reaction mixture consisting of 50 ng of template DNA, 0.2 mM dNTPs, 0.5 µM primer, 1.0 µl of 10X PCR buffer, 1.5 mM magnesium chloride and 1 unit of Taq polymerase to amplify DNA in 10 µl reactions. The PCR cycle was programmed at initial denaturation for 4 min at 94°C (1 cycle), followed by 35 cycles each of denaturation (94°C for 1 min), annealing (52–60°C for 1 min) and elongation (72°C for 1 min). The final extension cycle was performed at 72°C for 7 min (1 cycle).

Table 1 List of peach germplasm used in molecular characterization study

Low chill peach germplasm maintained at Pantnagar, Uttarakhand	*IC/EC No.	High chill peach germplasm maintained at Bhowali, Nainital, Uttarakhand	*IC/EC No.
Saharanpur Prabhat	-	Ramgarh Selection	IC-360682
Pratap	-	Red June	IC-360683
Early Grand	-	Red June	IC-360691
Florda Prince	-	Red June	IC-360680
Florda Gold	-	Early June	IC-360685
Florda Red	-	Bhatauida	IC-360698
Pant Peach-1	-	Ashadhiya	IC-360689
Red June	-	Totapari	IC-360690
Sharbati Late	-	Totapari	IC-247431
Sharbati Surkha	-	Alexander	IC-320193
Shan-i-Punjab	-	Paradelux Chapta	IC-247432
Selection-12	-	Paradelux Gola	IC-320194
Fla-16	-	Jethiya Chonch	IC-247430
Selection-1	-	Alton	EC-19377
Selection-2	-	Co-smith	EC-552641
		-	IC-209014
		Summer Glo	EC-198817
		Sone Peach	IC-413953
		Okubo	EC-280765
		July Elberta	EC-27793
		Nishika	EC-38736
		Nunowaisi	EC-280767
		Early Redhaven	EC-313954
		Kanto-5	EC-38737
		Crest Haven	-
		Red Globe	-

*IC, Indigenous collection; EC, Exotic collection

Gel electrophoresis: The PCR reaction mixture after the completion of a cycle was taken from the PCR machine and mixed with 6X loading dye and loaded on gel consisting of 2.5% agarose in 0.5X TBE buffer fused with ethidium bromide (1.0 µg/ml) and finally separated using horizontal gel electrophoresis at 90 V current for 1.5 hrs. The gel matrices were then visualized under a UV transilluminator and photographs were taken on the UV light gel documentation system.

Data analysis: Based on the allelic amplification, a binary matrix was generated. The polymorphic information content was computed using the Power Marker 3.5 software (Liu and Muse 2005). The GenAlex 6.5 (Peakall *et al*. 2012) software was used to assess the total number of alleles per locus (Na), number of effective alleles per locus (Ne), Shannon's information index (I), observed heterozygosity (Ho), expected heterozygosity (He) and fixation index (F). NTSYS-pc 2.11 (Rohlf 2000) software was used to compute the Jaccard's similarity coefficient and subsequently for dendrogram construction. Based on the Bayesian clustering, the population structure of the 41 peach accessions was determined using STRUCTURE version 2.3.2 (Pritchard *et al*. 2000). The length of the burn-in period and Markov Chain Monte Carlo (MCMC) were set at 10,000 iterations. For each K-value ranging from 1 to 10, ten runs were performed to accurately estimate the number of populations. The methodology of Evanno *et al*. (2005) was employed to determine the Delta K values and further, the best K value was estimated using the STRUCTURE Harvester program (Earl *et al*. 2012). In order to represent the spatial distribution of individuals, the Principal Coordinates Analysis was performed using GenAlEx 6.5 (Peakall *et al*. 2012).

RESULTS AND DISCUSSION

A total of 25 SSR markers were screened initially and 23 of these showed polymorphisms (Table 2). The amplified alleles across the 41 peach accessions varied between 100–320 bp. The major allele frequencies varied from 0.232 (MA015a) to 0.805 (MA020a). The polymorphism information content (PIC), a measure of allelic diversity, ranged from 0.317 to 0.836 for specific locus with a mean value of 0.563. The highest PIC value was estimated for the primer MA015a followed by BPPCT033 (0.732), CPPCT22 (0.694), MA023a (0.663), UDP98-407 (0.661) and BPPCT025 (0.654). Therefore, these primers were demonstrated to be highly effective in differentiating the peach accessions.

Across the 41 accessions, 23 markers amplified a total of 107 alleles (Table 2). The number of alleles amplified per SSR marker ranged between 3 to 8 with an average of 4.65 alleles per locus. The SSR marker MA015a produced a maximum number of 8 alleles followed by 7 alleles each by the markers BPPCT 015 and CPPCT14. The number of effective alleles per marker (Ne) ranged between 1.51 (MA020a) to 6.79 (MA015a) with an average value of 2.89 alleles per marker. Similarly, Shannon's Information Index (I) varied from a minimum value of 0.68 (MA020a) to a maximum value of 1.99 (MA015a) with an average value of 1.17. Unique allelic amplification was observed in accession Early June and Alton with markers BPPCT 015 and CPPCT 30, respectively. The observed heterozygosity (Ho) was very low which ranged between 0 to 0.27 with an average value of 0.02. However, the expected heterozygosity (He) was moderately high and varied from 0.34 to 0.85 with an average value of 0.61. The values for the inbreeding coefficient expressed by the fixation index (F) were lower for the marker CPPCT22 (0.67), MA015a (0.69), MA009b (0.96) and UDP98-412 (0.96) while all other markers showed a value of 1.

Based on the high percentage of polymorphic loci, the average number of alleles per locus (4.65), and the average expected heterozygosity (0.61) observed, the 23 SSR markers are assumed to be of great potential for genetic diversity studies in peaches. The number of alleles per locus observed in the investigation varied from 3 to 8 which seems to be quite similar to the results of Bouhadida *et al*. (2011) who noted 2 to 11 alleles per locus but higher than the number of alleles per locus noted by Bedo *et al*. (2018) while analyzing peach accessions. Differences in the number of alleles per locus and the total number of alleles detected may be due to the nature of accessions included in the investigation and selection of markers. Accessions with less diversity may result in a low number of alleles amplification.

The average PIC value in the investigation was 0.56 which is similar to the PIC value (0.55) reported by Bouhadida *et al*. (2011) and higher (0.49) than Trifonova *et al*. (2021). The higher PIC indicates the high efficiency power of SSR markers in discriminating different accessions and also in quantifying genetic diversity among peach accessions. The 10 SSR loci (MA015a, BPPCT033, CPPCT22, MA023a, UDP98-407, BPPCT025, pchcms5, UDP96-003, CPPCT14 and BPPCT015) have PIC values greater than 0.60 which indicate more significance of these markers in the peach research programme.

Compared to expected heterozygosity (He), we observed lower heterozygosity (0.024), suggesting a lesser per cent of heterozygotes in the evaluated accessions. This observation is further supported by the high inbreeding coefficient (F= 0.97) value. The He value in the investigation was 0.61 which is higher than the He value of 0.41 reported by Perez *et al*. (2020) while investigating 142 local peach accessions from Canary Islands, Spain. These observations may be explained by the degree of relatedness in the ancestry among the accessions used in the investigation. High F values indicate an excess of homozygotes at most of the loci. Limited pollen dispersal and gene flow due to the self-pollinated nature of peaches may be the reason for homozygosity at most of the loci (Rajesh *et al*. 2008).

Jaccard's similarity coefficient among the 41 peach accessions based on SSR profiles of 23 markers varied from 0.533 to 1 with an average value of 0.724 which indicates the high degree of genetic diversity in the peach accessions (Supplemenatry Table 1). Accessions namely,

Table 2 Molecular diversity analysis of peach germplasm

Marker	Annealing temperature (˚C)	Amplified product (bp)	Major allele frequency	PIC	Na	Ne	I	Ho	He	F
BPPCT001	57	160–220	0.634	0.482	4	2.15	0.97	0.00	0.53	1.00
BPPCT015	57	120–240	0.512	0.625	7	2.99	1.41	0.00	0.67	1.00
BPPCT017	57	160–200	0.585	0.470	3	2.21	0.90	0.00	0.55	1.00
BPPCT025	57	160–240	0.390	0.654	5	3.38	1.37	0.00	0.70	1.00
BPPCT033	59	180–260	0.293	0.732	5	4.34	1.53	0.00	0.77	1.00
CPPCT14	50	120–240	0.512	0.625	7	2.99	1.41	0.00	0.67	1.00
CPPCT22	55	240–320	0.329	0.694	5	3.84	1.42	0.24	0.74	0.67
CPPCT28	55	100–160	0.488	0.511	3	2.47	0.97	0.00	0.59	1.00
CPPCT30	55	160–260	0.756	0.375	5	1.68	0.82	0.00	0.40	1.00
CPPCT31	55	160–220	0.756	0.380	4	1.68	0.81	0.00	0.41	1.00
MA009b	53	120–180	0.500	0.550	3	2.64	1.03	0.02	0.62	0.96
MA015a	55	180–300	0.232	0.836	8	6.79	1.99	0.27	0.85	0.69
MA017a	55	160–240	0.585	0.516	5	2.35	1.07	0.00	0.57	1.00
MA020a	53	180–240	0.805	0.317	4	1.51	0.68	0.00	0.34	1.00
MA023a	53	180–280	0.415	0.663	5	3.45	1.38	0.00	0.71	1.00
pchcms1	57	180–260	0.488	0.526	5	2.52	1.09	0.00	0.60	1.00
pchcms5	57	240–320	0.439	0.643	5	3.25	1.34	0.00	0.69	1.00
pchgms2	55	160–220	0.732	0.411	4	1.78	0.87	0.00	0.44	1.00
UDP96-003	57	120–220	0.390	0.637	5	3.24	1.33	0.00	0.69	1.00
UDP96-005	57	140–190	0.561	0.522	3	2.43	0.99	0.00	0.59	1.00
UDP98-021	57	140–200	0.585	0.540	4	2.43	1.09	0.00	0.59	1.00
UDP98-407	57	190–260	0.415	0.661	4	3.45	1.31	0.00	0.71	1.00
UDP98-412	57	100–180	0.427	0.587	4	2.92	1.13	0.02	0.66	0.96
Mean			0.514	0.563	4.65	2.891	1.170	0.024	0.613	0.969

PIC, Polymorphism information content; Na, No. of different alleles; Ne, No. of effective alleles; I, Shannon's Information index; Ho, Observed heterozygosity; He, Expected heterozygosity; F, Fixation index.

IC-360682 and IC-360683; IC-360691 and IC-360680, and Totapari and IC-247431 were found synonymous with a similarity-coefficients of 1.0. However, the minimum similarity coefficient was noted between IC-209014 and Red June (0.533) followed by IC-209014 and Shan-i-Punjab (0.542), Sone Peach and Red June (0.542) and Alton and Red June (542). The existence of similarity coefficients of 1 among some accessions indicates no dis-similarity between those accessions for all the loci investigated using the SSR markers, which prove their similar genetic constitution. Sharma and Sharma (2018) also assessed the genetic diversity and genetic relationships among 45 peach cultivars in Himachal Pradesh, India using 48 RAPD and 46 ISSR markers and observed Jaccard's similarity coefficients ranging between 0.37 to 0.95 and 0.43 to 0.95, respectively.

The phylogenetic tree constructed using UPGMA (Un-weighted pair group method with arithmetic mean) grouped 41 peach genotypes into two major clusters (cluster I and II) at 64% similarity. Cluster I contained 33 accessions, of these 26 were high chill and 7 were low chill peach accessions. Dendrograms provide an effective way to summarize microsatellite data, allowing the identification of relationships among germplasm, as well as documentation of identical genotypes. The present study revealed that clustering of peach accessions is influenced by their geographical adaptation. Observations on geographic affinity in the grouping of peach accessions were also noted earlier by Bouhadida *et al*. (2011).

For the purpose of determining the genetic structure of the peach accessions, the Bayesian clustering approach was used. A population structure analysis based on the Delta K value stratified the 41 accessions into three clusters with few admixtures (Fig 1). Cluster 1 included 7 genotypes (in red bars) while Cluster 2 included 15 genotypes (in blue bars). Cluster 3 had 8 genotypes (in green bars). The analysis revealed admixture in genotypes Saharanpur Prabhat, Pratap, Early Grand, Selection-12, Selection-1, Selection-2, Bhatauida, Jethiya Chonch, Nunowaisi and Early Redhaven. Furthermore, the principal coordinate analysis (PCoA) was also performed to reveal the genetic

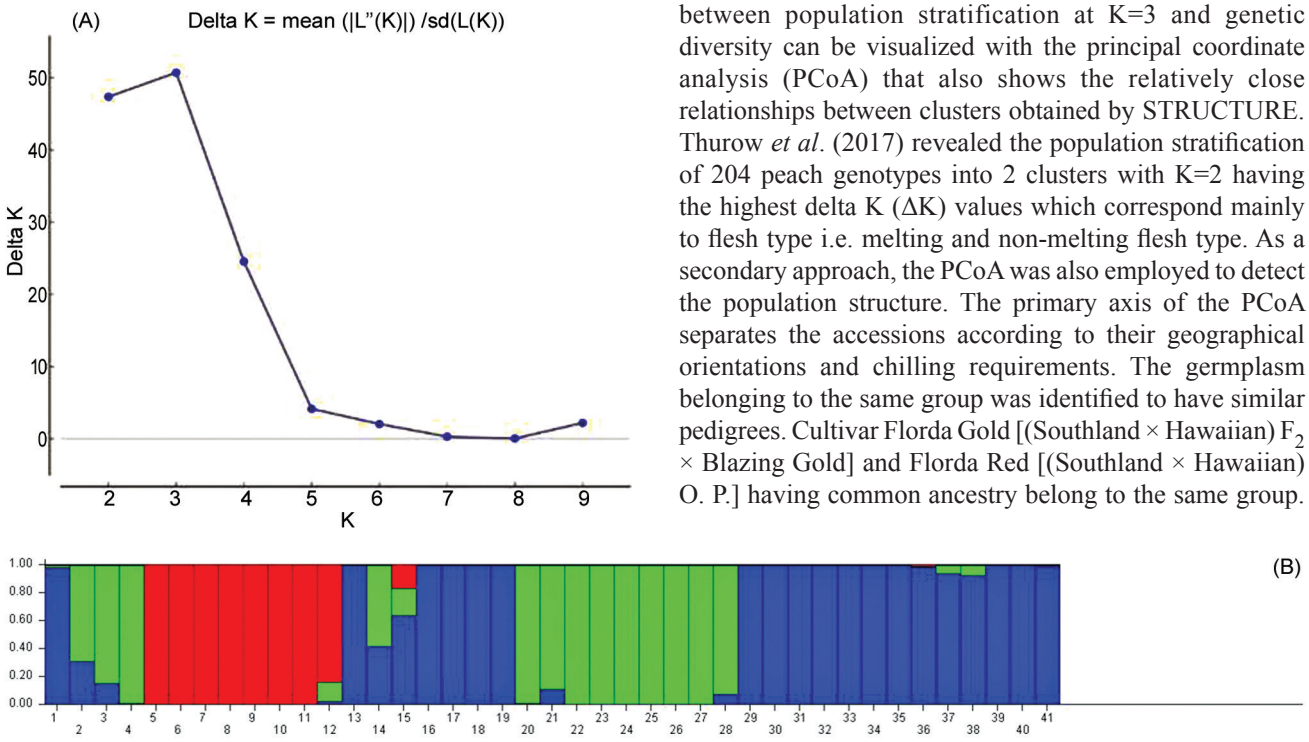

Fig 1 Population genetic structure based on 23 SSR markers in 41 peach accessions
(a) ΔK graph showing peak value at K = 3, (b) population structure at ΔK = 3.

between population stratification at K=3 and genetic diversity can be visualized with the principal coordinate analysis (PCoA) that also shows the relatively close relationships between clusters obtained by STRUCTURE. Thurow *et al*. (2017) revealed the population stratification of 204 peach genotypes into 2 clusters with K=2 having the highest delta K (ΔK) values which correspond mainly to flesh type i.e. melting and non-melting flesh type. As a secondary approach, the PCoA was also employed to detect the population structure. The primary axis of the PCoA separates the accessions according to their geographical orientations and chilling requirements. The germplasm belonging to the same group was identified to have similar pedigrees. Cultivar Florda Gold [(Southland × Hawaiian) F_2 × Blazing Gold] and Florda Red [(Southland × Hawaiian) O. P.] having common ancestry belong to the same group. Similarly, cultivars Early Grand [Selection from Fla 5-58 × Early Amber] and Florda Prince [Fla.2-7 × Fla. 13-72 (Maravilha)] shared similar ancestry and hence belonged to the same group.

relationships among the selected accessions, which classified the 41 peach accessions into three groups (Fig 2). The first three principal coordinates explained 20.12%, 10.77% and 10.08% of the variance.

With the help of the STRUCTURE software, we analyzed the population structure of 41 peach accessions and found three distinct subpopulations that correspond to the result of the phylogenetic analysis. A clear agreement

The investigation was successful in generating the molecular profile of 41 peach accessions using 23 SSR markers. Higher allelic forms locus and high PIC values indicate the effectiveness of SSR markers in characterizing

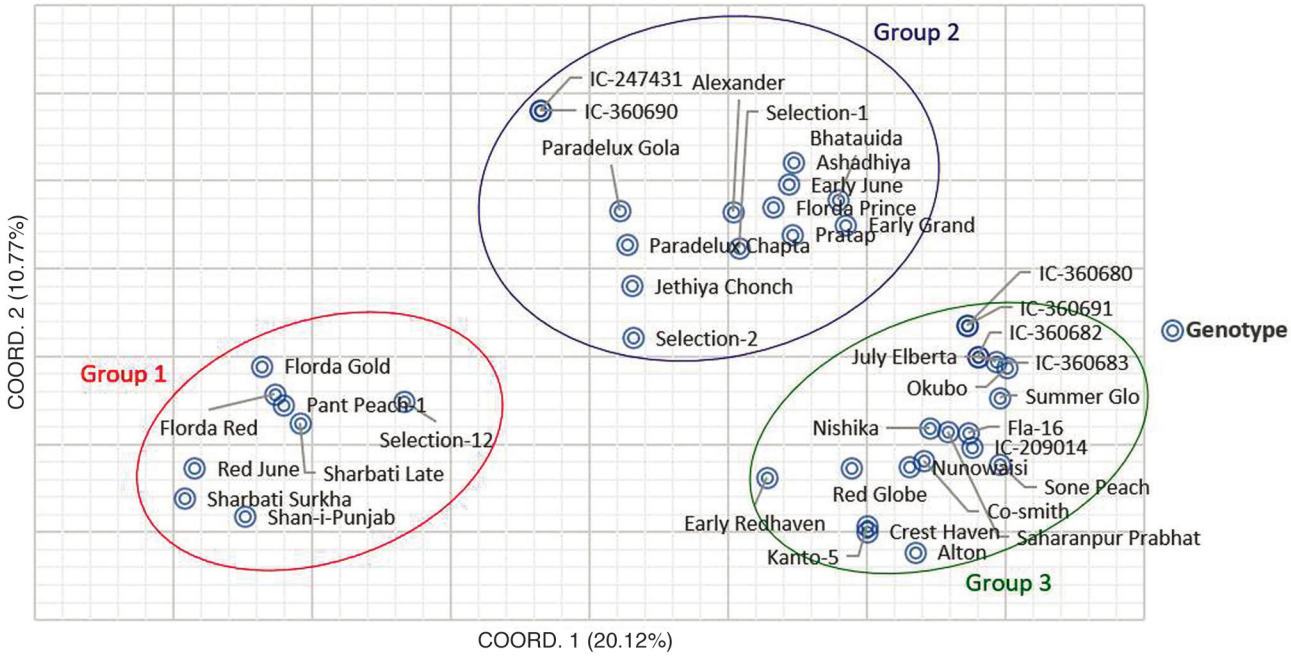

Fig 2 PCoA based on pairwise Nei's genetic distance estimates for the populations.

a large number of peach accessions. The presence of different clusters in both phylogenetic as well as population structure analysis indicates a high level of genetic variations in the investigated peach accessions which can be used as a criterion for the selection of diverse parents in the improvement programme to get better transgressive segregants with respect to different traits of interest. The molecular profile of 41 peach accessions may be used as a tag for documentation. Further, the identified primers to be highly polymorphic may constitute a set of markers for fingerprinting peach accessions.

ACKNOWLEDGMENT

Author is grateful to the G. B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand for financial support.

REFERENCES

Aranzana M J, Garcia-Mas J, Carbo J and Arus P. 2002. Development and variability analysis of microsatellite markers in peach. *Plant Breeding* **121**: 87–92.

Bedo J, Bariş E, Szani Z, Surányi D, Kiss E and Veres A. 2018. SSR based characterization of peach (*Prunus persica* L.) and apricot (*Prunus armeniaca* L.) varieties cultivated in Hungary. *Acta agraria Debreceniensis* **74**: 17–24.

Bouhadida M, Moreno M, Gonzalo M, Alonso S J and Gogorcena Y. 2011. Genetic variability of introduced and local Spanish peach cultivars determined by SSR markers. *Tree Genetics and Genomes* **7**: 257–70.

Cipriani G, Lot G, Huang W G, Marrazzo M T, Peterlunger E and Testolin R. 1999. AC/GT and AG/CT microsatellite repeats in peach [*Prunus persica* (L) Batsch]: Isolation, characterization and cross-species amplification in *Prunus*. *Theoretical and Applied Genetics* **99**: 65–72.

Dagnon Y D, Palanga K K, Bammite D, Bodian A, Akabassi G C, Fonceka D and Tozo K. 2022. Genetic diversity and population structure of cowpea [*Vigna unguiculata* (L.) Walp.] accessions from Togo using SSR markers. *PLoS ONE* **17**(10): e0252362.

Dirlewanger E, Crosson A, Tavaud P, Aranzana M J, Poizat C, Zanetto A, Arus P and Laigret L. 2002. Development of microsatellite markers in peach and their use in genetic diversity analysis in peach and sweet cherry. *Theoretical and Applied Genetics* **105**: 127–38.

Doyle J J and Doyle J L. 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochemical Bulletin* **19**: 11–15.

Earl D A and vonHoldt B M. 2012. STRUCTURE HARVESTER: A website and program for visualizing STRUCTURE output and implementing the Evanno method. *Conservation Genetics Resources* **4**(2): 359–61.

Evanno G, Regnaut S and Goudet J. 2005. Detecting the number of clusters of individuals using the software STRUCTURE: A simulation study. *Molecular ecology* **14**: 2611–20.

Faust M and Timon B. 1995. Origin and dissemination of peach. *Horticultural Reviews* **17**: 267–379.

Kumar K, Khokhar A and Singh D. 2013. Peach. *Fruit Production in India*, 1st edn, pp. 457–58. Dhillon W S (Ed). Narendra Publishing House, Delhi, India.

Kumar J, Thakur A, Verma M K, Kumar K, Singh H, Singh D, Thakur D, Jawandha S K and Banyal S K. 2015. Peach. *Temperate Fruit Breeding: Domestication to Cultivar Development*, 1st edn, pp. 438. Ghosh S N (Ed). Jaya Publishing House, Delhi, India.

Kumar R, Dimri D C, Singh N K, Singh A K, Karki K, Uniyal S, Kumar R, Shivran J S and Bajeli J. 2023. Stratification and SSR markers integration for promoting low chill peach (*Prunus persica*) hybridization in foot hills of Himalayas. *The Indian Journal of Agricultural Sciences* **93**(5): 484–88.

Liu K and Muse S V. 2005. Power marker: Integrated analysis environment for genetic marker data. *Bioinformatics* **21**: 2128–29.

Peakall R and Smouse P E. 2012. GenAlEx 6.5: Genetic analysis in Excel. Population genetic software for teaching and research-an update. *Bioinformatics* **28**: 2537–39.

Perez V, Larrañaga N, Abdallah D, Wünsch A and Hormaza J I. 2020. Genetic diversity of local peach (*Prunus persica*) accessions from La Palma island (Canary Islands, Spain). *Agronomy* **10**(4): 457.

Pritchard J K, Stephens M and Donnelly P. 2000. Inference of population structure using multilocus genotype data. *Genetics* **155**: 945–59.

Rajesh M K, Arunachalam V, Nagarajan P, Lebrun P, Samsudeen K and Thamban C. 2008. Genetic survey of 10 Indian coconut landraces by simple sequence repeats (SSRs). *Scientia Horticulturae* **118**: 282–87.

Rohlf FJ. 2000. NTSYS-pc: Numerical taxonomy and multivariate analysis system, version 2.11. Exeter Software: Setauket, NY.

Sharma P and Sharma R. 2018. DNA fingerprinting of peach (*Prunus persica*) germplasm in accessing genetic variation using arbitrary oligonucleotide markers system. *Indian Journal of Biotechnology* **17**(3): 484–91.

Sosinski B, Gannavarapu M, Hager L D, Beck L E, King G J, Ryder C D, Rajapakse S, Baird W V, Ballard R E and Abbott A G. 2000. Characterization of microsatellite markers in peach [*Prunus persica* (L.) Batsch]. *Theoretical and Applied Genetics* **101**: 421–28.

Thurow L, Raseira M, Bonow S, Arge L and Castro C. 2017. Population genetic analysis of Brazilian peach breeding germplasm. *Revista Brasileira de Fruticultura* **39**(5): (e-166).

Trifonova A A, Boris K V, Mesyats N V, Tsiupka V A, Smykov A V and Mitrofanova I V. 2021. Genetic diversity of peach cultivars from the collection of the Nikita Botanical Garden based on SSR markers. *Plants* **10**(12): 2609.

Yamamoto T, Mochida K, Imai T, Shi Y Z and Ogiwara I. 2002. Microsatellite markers in peach [*Prunus persica* (L.) Batsch] derived from an enriched genomic and cDNA libraries. *Molecular Ecology Notes* **23**: 298–301.