RESEARCH ARTICLE

Diversity, genetic structure and core collection of mangaba (*Hancornia speciosa*) genebank

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Abstract This work was developed with the objective of characterizing the variability and genetic structure of matrices and progenies of the Mangaba genebank of Embrapa Tabuleiros Costeiros, to define a core collection. From the matrix of seven accessions, 289 individuals were generated, evaluated by nine SSR markers. The characterization of genetic variability was performed from the estimates: average number of alleles observed (Na = 5.63) and effective (Ne = 2.71); Shannon Information Index (I = 1.04); Heterozygosity observed (Ho = 0.45) and expected (He = 0.51); Fixation index (f = 0.13); Percent polymorphism (%P = 90.47); and, Number of private alleles (Nap = 20). The analysis of molecular variance (AMOVA) identified that the largest proportion of genetic variation is present within accessions (92%). The estimates of genetic differentiation (G_{ST} and R_{ST}) were considered low (< 0.05) to moderate (0.05–0.15) magnitude. Multivariate analyses (PCoA, Rogers and Bayesian genetic diversity) did not discriminate the accessions according to origin. The formation of the core collection allowed retaining 94.9% of the identified alleles. The accessions present genetic variability to be explored in mangaba conservation and genetic improvement programs. In the future data related to agronomic and morphological characterization should be used to support the formation of this core collection, since the combination of this information contributes to the design of more efficient strategies for the use of this genetic resource.

Introduction

The genus *Hancornia* is monospecific and consists of the species *Hancornia speciosa* Gomes (Jimenez et al. 2015). Popularly known as mangaba, the species belongs to the Apocynaceae family with a wide distribution in the Neotropics, being reported in Brazil, Paraguay, Bolivia and Peru (Collevatti et al. 2018; Fajardo et al. 2018). In Brazil, it can be seen from Amapá, in the North, to Paraná, in the South, in different phytophysiognomies associated with the Cerrado and the Atlantic Forest (Silva et al. 2021).

The mangaba is hermaphrodite and presents a self-incompatibility system, a mechanism that favors allogamy, being pollinated mainly by moths (Darrault and Schlindwein 2005). Its fruits have small oval seeds, which are mainly dispersed by medium and large mammals (Costa et al. 2017).

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The species is used in folk medicine and different parts of the plant have been used to treat various diseases, such as gastric ulcers and hypertension (Dórea et al. 2021). Additionally, it presents edible pulp with relevant nutritional properties (Jimenez et al. 2015), factors that have contributed to its use by the local population. The fruits are consumed *in natura* and used in the preparation of jellies, juices, cookies, ice cream, playing an important role for the local economy (Collevatti et al. 2016; Chaves et al. 2020). It is one of the most important raw materials for the juice and ice cream agroindustry among the native fruits in the Northeast and Midwest of Brazil, but the supply is unsatisfactory. According to Soares et al. (2019), despite its importance for local populations and its agro-industrial potential, the exploitation of the species has been carried out to a greater extent in natural populations, that is, in an extractivist manner (Soares et al. 2019).

The form of exploitation associated with the process of fragmentation of the areas of natural occurrence makes mangaba one of the genetically endangered fruit species in Brazil. Considering its ecological and economic importance, the collection, conservation, and characterization of available mangaba genetic resources are fundamental (Jimenez et al. 2015).

The adoption of in situ and ex situ conservation strategies is necessary for the maintenance of the remaining genetic variability of the species. The seeds are classified as recalcitrant, thus ex situ conservation should be accomplished through in vitro and/or in vivo field collections (Almeida et al. 2019).

Due to the reduction of natural areas and the social, economic and cultural importance of mangaba, Embrapa Tabuleiros Costeiros, a research center of Brazilian Agricultural Research Coporation (Embrapa) has been developing conservation strategies for the species for more than 15 years. The Mangaba genebank was established in 2006 and is accredited by the Ministry of the Environment (MMA)/Genetic Heritage Management Council (CGEN) as the depositary of the species in Brazil. It currently consists of 21 accessions represented by 299 plants, collected over a wide geographic range (Silva et al. 2021).

There is no improvement program, nor mangaba cultivars. In the Mangaba genebank some accessions were preliminarily identified and selected as early (Silva et al. 2019a, b, c), with low seasonality (Machado et al. 2020), and as excellent sources of vitamin C and antioxidants (Silva et al. 2012, 2021; Silva et al. 2017a, b; Santos et al. 2020; Santana et al. 2021).

Knowledge of the genetic variability present in a BAG is essential for the efficient management and use of this resource (Bernard et al. 2018). This characterization can be accomplished through morphological, agronomic, chemical, and molecular variables. DNA markers have been used as key tools in the molecular characterization of different accessions, as they are not influenced by environment and plant development stage, and are more accurate, efficient and reliable for the discrimination of related genotypes. The genetic diversity of the accessions of the Mangaba genabank was carried out following its expansion (Costa et al. 2011; Silva et al. 2011; Silva et al. 2019a, b, c). With the need to form an area of progenies from this genebank, aiming at a breeding program, the proposal of this work was built.

Microsatellites (SSR) are important tools for characterizing genetic diversity and structure due to technical simplicity, speed, high resolving power, reproducibility, high level of polymorphism, and codominant inheritance (Kölliker et al. 2010; Silva et al. 2019a, b, c). These markers have been successfully used for different purposes in mangaba, such as in research to estimate gene flow among cultivars (Collevatti et al. 2016), characterize spatial genetic structure (Costa et al. 2017), and quantify the genetic variability present in natural populations (Amorim et al. 2015; Rodrigues et al. 2015; Chaves et al. 2020), in germplasm (Silva et al. 2019a, b, c) and progenies (Soares et al. 2018).

Genebanks are considered valuable reservoirs of allelic variability for different traits of economic and ecological importance that can be exploited in conservation and genetic improvement programs (Campoy et al. 2016). However, the preservation and maintenance of a large number of accesses is labor intensive and expensive. The establishment of a core collection is a strategy that allows maximizing genetic diversity in a smaller number of accessions, reducing the number of redundant genotypes, which is an alternative for reducing maintenance costs (Pereira et al. 2020).

In view of the above, this work was carried out with the objective of characterizing the variability and genetic structure of mangaba progenies from matrices of the genebank, using SSR markers, and to define a core collection for the germplasm, with the purpose of generating information for the conservation, management and efficient use of this genetic resource.

Material and methods

The Mangaba genebank (Fig. 1) is located in Itaporanga d'Ajuda, Sergipe state, Brazil (11° 06′ 40″ S and 37° 11′ 15″ W).

Seven accessions with matrices in fruiting were selected for the production of the progenies, which were evaluated together with the 289 progenies generated, totaling 296 accessions (seven matrices and 289 progenies) (Table 1).

To obtain the progenies, fruits were collected from each access used as matrix. The fruits were pulped, and the seeds treated with 2.5% sodium hypochlorite solution for 3 min. After, they were washed and dried in the shade. For seedling production, the seeds were sown in polyethylene bags containing washed

Table 1 Accessions, procedence and number of progenies present in the Mangaba genebank of Embrapa Tabuleiros Costeiros. Itaporanga d'Ajuda, Sergipe, Brazil

Acession/Code	Procedence	N° of progeny
Preguiça—PR	Indiaroba, SE	17
Lagoa Grande—LG	Mata de São João, Bahia	59
Pontal—PT	Indiaroba, Sergipe	56
Barra de Itariri—BI	Conde, Bahia	32
Terra Caída—TC	Indiaroba, Sergipe	52
Água Boa—AB	Salvaterra, Pará	55
Costa Azul—CA	Jandaíra, Bahia	18
		289
Total	7 matrices + 289 progenies	296

sand and identified according to the matrix accession (Fig. 2). At 6 months after sowing, leaves were taken from each progeny for genomic DNA extraction. The collected leaves were stored at − 80 °C until DNA extraction.

DNA extraction was performed using the method described by Doyle and Doyle (1990) modified by Alzate-Marin et al. (2009). The extracted DNA was diluted in 50 µL of TE and its quantification was

Fig. 1 Mangaba Genebank of Embrapa Tabuleiros Costeiros. Itaporanga d'Ajuda, Sergipe, Brazil

Fig. 2 Fruits, processing and mangaba progeny formation

performed by spectrophotometry using Nanodrop 2000c (Thermo Scientific, USA). Nine pairs of microsatellite primers (Table 2) developed by Rodrigues et al. (2015) were used to characterize genetic diversity and structure.

In each amplification reaction a total of 13 µL of solution containing: 9 ng DNA, 0.25 mg mL^{-1} BSA (bovine serum albumin), 0.2 µM of each primer (25 pmol), $1 \times$ PCR buffer (10X), 0.25 mM dNTPs mix (2.5 mM), 1 U of Taq DNA polymerase and sterile ultrapure water.

The amplifications were performed in a Veriti 96 Fast thermal cycler (Applied Biosystems/USA). The PCR reaction consisted of an initial step at 94 °C for 1 min, followed by 35 cycles (95 °C for 1 min for denaturation of the DNA strands, primer annealing temperature (Table 2) for 1 min, and 72 °C for DNA strand extension for 1 min), and a final extension step performed at 72 °C for 20 min. To test for contamination by exogenous DNA, a negative control containing all PCR components except DNA (replaced by water) was included in each experiment.

The amplification results were visualized by electrophoresis on a 2% (w/v) agarose gel stained with ethidium bromide (0.5 µg mL^{-1}) (Sambrook et al. 1989) and photodocumented under ultraviolet light (Kodac Gel Logic 200 Imaging System). For size comparison of the amplified fragments the DNA marker 1 Kb Ladder (Invitrogen) was used.

For the analyses of the fragments obtained by each SSR primer pair used, 1 µL of each reaction was mixed with 10 µL of HiDi formamide (Applied Biosystems, Foster City, CA) and 1 µL of the internal marker carboxy-X-rhodamine (ROX), developed by Brondani and Grattapaglia (2001). The resulting solution was denatured for 5 min at 95 °C. Then, fragment separation was performed in an ABI 3730 automated DNA analyzer (Applied Biosystems, Foster City, CA). Fluorescence peak detection and genotyping were performed with the program Genemapper version 4.1 (Applied Biosystems). Allele size was

Table 2 Annealing temperature (AT), repeat motifs, allele amplitude (pb) and fluorescence of microsatellites used to characterize the diversity and genetic structure of mangaba genebank of Embrapa Tabuleiros Costeiros

Primer	AT (°C)	Repeat motifs	Allele amplitude (bp)	Fluorescence
HS01	56	(GCA)6(TC)20(GCA)8	250 a 310	HEX
HS03	56	(CT)5(CT)5	120 a 180	6-FAM
HS05	56	(GA)15(TGC)6	200 a 300	HEX
HS06	54	(GA)14	100 a 150	HEX
HS08	52	(CA)6(CT)17	200 a 250	6-FAM
HS10	56	(CT)14(CT)8	100 a 200	HEX
HS16	54	(GA)12	100 a 150	6-FAM
HS27	54	(GA)14	100 a 150	6-FAM
HS33	56	(AG)24	80 a 120	6-FAM

bp base pairs

adjusted for allele classes defined by the AlleloBin program (Prasanth and Chandra 2006).

The statistics allele frequency, number of alleles per *locus*, expected and observed heterozygosity, and PIC (Polymorphic Information Content) were estimated for each SSR primer pair using Power Marker 3.25 software (Liu and Muse 2005).

The genetic variability of the accessions was characterized by estimates of the number of alleles observed (Na) and effective (Ne); Shannon Information Index (I); Heterozygosity observed (Ho) and expected (He); calculated according to the proportions expected by Hardy–Weinberg Equilibrium (Nei 1978); Fixation index (f); Percentage polymorphism (%P); and; Number of private alleles (Nap).

Analysis of Molecular Variance (AMOVA) was performed to estimate the genetic variance existing between and within accessions, and the level of significance was determined at 9999 permutations. Genetic differentiation among accessions (G_{ST}) (Nei 1973), which corresponds to the proportion of genetic variation within accessions compared to the total genetic variation, was estimated and its significance tested using 10,000 bootstraps. Additionally, Nei genetic distance was estimated among the accessions. Analyses were performed using Genalex 6.5 software (Peakall and Smouse 2012).

Genetic distance among the accessions was assessed using Rogers (1972) genetic distance and visualized by constructing a dendrogram using the UPGMA (Unweighted Pair Group Method with Arithmetic Means) algorithm. The analysis was performed with the help of the poppr package (Kamvar et al. 2014) for R (R Core Team 2016). Ten thousand bootstraps were performed to infer on the reliability of the clusters. The software FigTree 1.4.1 was used to format the obtained dendrogram. Principal coordinate analysis (PCoA), at the individual level, was performed using Genalex 6.5 software (Peakall and Smouse 2012).

Bayesian analysis was performed to estimate the genetic structure of the accessions using Structure v.2.3.4 software (Pritchard et al. 2000). Genetic clustering values (k) ranging from 1 to 7 (number of accessions) were tested, and for each k, 10 independent repetitions were performed. Each repetition consisted of a burn in period of 50,000 iterations, followed by 100,000 MCMC (Markov Chain Monte Carlo) iterations, assuming the admixture ancestry model and uncorrelated allele frequency. The number of gene groups (k) was identified by the ΔK method described by Evanno et al. (2005), implemented in Structure Harvester software (Earl and Vonholdt 2012). Accessions with membership values lower than 0.8 were considered to be of mixed ancestry.

The core collection is a subsample of the germplasm and is established to represent the genetic diversity present in the collection in a smaller number of accessions. The maximum length sub tree function implemented in DARwin 6.0.14 (Perrier and Jacquemoud-Collet 2006) was used to identify the accessions to compose the mangaba core collection. This function eliminates accessions that are considered redundant and allows the selection of accessions that retain the most genetic variation. Ten thousand bootstraps were performed to infer on the reliability of the result.

Results and discussion

A total of 98 alleles were detected using nine SSR markers (Table 3). The number of alleles per locus ranged from six (HS03, HS10 and HS27) to 20 (HS01), with an average of 10.89, and fragment size ranged from 103 to 343 bp. The expected heterozygosity (He) ranged from 0.04 (HS03) to 0.87 (HS01), and the observed heterozygosity (Ho) ranged from 0.01 (HS03) to 0.72 (HS06). The HS03 marker

Table 3 Maximum frequence (*f*max), number of alleles observed (Na), expected heterozygosity (He), observed heterozygosity (Ho) and polymorphic information content (PIC) for nine SSR markers

SSR markers	*f*max	Na	He	Ho	PIC
HS01	0,23	20	0.87	0.67	0.86
HS03	0,98	6	0.04	0.01	0.04
HS05	0,34	14	0.83	0.40	0.82
HS06	0,30	13	0.78	0.72	0.75
HS08	0,40	7	0.68	0.68	0.62
HS10	0,68	6	0.49	0.31	0.44
HS16	0,48	17	0.66	0.69	0.61
HS27	0,58	6	0.56	0.15	0.48
HS33	0,40	9	0.76	0.43	0.72
Average	0.49	10.89	0.63	0.45	0.59

showed the lowest heterozygosity value (0.04), indicating that the analyzed accessions did not show diversity for this locus. The PIC values ranged from 0.04 (HS03) to 0.86 (HS01).

The PIC value represents the probability of detecting polymorphism between two random samples (Ismail et al. 2019). With the exception of primer HS03, the primers showed high discrimination power, with PIC values higher than 0.44 (HS10). Considering the PIC formula, the observed values are dependent on the number of detected alleles and their relative frequency (Guzmán et al. 2020). Therefore, the observation of one or two alleles with high frequency will contribute to low PIC values, as could be observed for primer HS03 that presented one allele with frequency equal to 0.98 (Table 3).

The use of nine SSR primers allowed the characterization of the variability and genetic structure among the progenies and seven matrix accessions of the Mangaba genebank. The number of alleles per locus detected (6–20) is indicative of the allelic richness of the population, and was considered sufficient to meet the objective of the present study since for SSR markers the detection of two to seven alleles per locus is considered satisfactory (Aljumaili et al. 2018). The high heterozygosity values detected for the SSR markers used may be related to the reproductive system (interbreeding) of the species (Table 3).

The average number of alleles per locus is an indication of genetic diversity, and ranged from 3.67 (CA) to 7.22 (PT) (Table 4). The average was lower than that observed by Collevatti el al. (2018) in a study with 28 natural mangaba subpopulations (9.6) using SSR markers. The effective number of alleles was lower than the observed number of alleles, suggesting that many alleles are rare ($p < 0.05$) or have low frequency ($0.05 > p < 0.25$) (Viegas et al. 2011). The accessions that presented the lowest number of alleles were PR and CA, which may be related to the smaller number of individuals analyzed (20). The effective number of alleles was lower than the observed number of alleles, ranging from 1.98 (CA) to 3.46 (BI). And, unique alleles were observed for five accessions (Table 4).

Considering the Shannon index, here was less genetic diversity in the CA accession (0.76), and greater in the BI accession (1.31) (Table 4). The high values observed indicate the existence of high genetic variability in materials. The estimated values were higher than those of their matrices for all accessions, when observed by Santos et al. (2017), using ISSR markers, varying from 0.28 (CA) to 0.42 (BI), proving the occurrence of random crossings between the accessions.

Ho ranged from 0.37 (CA) to 0.53 (PR), with a mean of 0.45, and He ranged from 0.39 (CA) to 0.62 (BI), with a average of 0.51 (Table 4). This index refers to the proportion of accessions that

Table 4 Estimates of genetic variability parameters for mangaba genebank accessions of Embrapa Tabuleiros Costeiros

Accession	Na	Ne	I	Ho	He	f	%P	Nap
PR	3.89	2.33	0.85	0.53	0.44	−0.19	77.78	0
LG	5.67	3.02	1.04	0.41	0.51	0.25	100	1
PT	7.22	3.20	1.22	0.45	0.59	0.28	100	6
BI	6.78	3.46	1.31	0.48	0.62	0.27	88.89	3
TC	5.78	2.22	0.91	0.43	0.45	0.02	88.89	4
AB	6.44	2.76	1.19	0.48	0.58	0.14	100	6
CA	3.67	1.98	0.76	0.37	0.39	0.04	77.78	0
Average	5.63	2.71	1.04	0.45	0.51	0.13	90.47	20

Na Average number of alleles per locus, *Ne* Effective number of alleles per locus, *I* Shannon index, *Ho* Observed heterozygosity, *He* Expected heterozygosity, *f* fixation index, *%P* Polymorphism percentage, *Nap* Number of private alleles

are heterozygous for a given locus. With the exception of the PR access, the others presented He > Ho, according to the Hardy–Weinberg Equilibrium. This result is an indication of heterozygote deficiency in the population, and suggests the occurrence of crossing between related individuals (Bernard et al. 2018), and can be proven by the estimated values of the Fixation Index (f) for these accessions (greater than zero). The estimates for Ho and He were lower than expected for allogamous species (0.63 and 0.65, respectively) and for long-lived species (0.63 and 0.68, respectively) (Nybom 2004). The estimates for Ho were lower than those reported by Costa et al. (2017), studying mangaba genotypes using SSR markers (0.679–0.714), and similar to those observed by Chaves et al. (2020), also for mangaba genotypes evaluated using SSR markers (0.428–0.581).

The access PR presented higher Ho than He. This result is an indication of excess heterozygosity in this progeny (Yun et al. 2020), confirming the result obtained for the Fixation Index (f = − 0.19) (Table 4). The Fixation Index is one of the most important parameters in population genetics, as it presents the balance between homozygotes and heterozygotes present in the population (Pereira et al. 2020). The average value for the Fixation Index was 0.13, indicating a low level of inbreeding for the accessions evaluated.

Mangaba has a self-incompatibility mechanism (Darrault and Schlindwein 2005), which favors cross-fertilization and reduces the occurrence of inbreeding. Thus, the excess of heterozygotes observed for the PR access can be explained by the reproductive system of the species and, probably, the deficit of heterozygotes observed for the other accesses occurred due to crossing between related individuals. Biparental inbreeding was reported to be the cause of the high values observed for the endogamy coefficient in natural populations of mangaba sampled in the Midwest region of Brazil (Costa et al. 2017).

The percentage of polymorphic loci was higher than 75% (Table 4), confirming the presence of genetic variability for the accessions evaluated. The results were higher than those observed in remaining mangaba populations (73.77%) (Silva et al. 2017a, b), which may be related to the marker (SSR), which is considered more informative than ISSR, and the number of individuals evaluated (296). The high genetic variability detected for the accessions evaluated is often related to species that have wide geographic distribution (Al Salameen et al. 2018), as is the case with mangaba.

The estimate for Nei's genetic distance (Table 5) among the accessions ranged from 0.098 to 0.607. The lowest genetic distance of Nei was observed between the PT and TC accessions (0.098), and the

Table 5 Nei's genetic distance (above the diagonal) and GST (below the diagonal) between mangaba accessions

	PR	LG	PT	BI	TC	AB	CA
PR	–	0.478	0.198	0.340	0.188	0.324	0.229
LG	0.162	–	0.261	0.239	0.574	0.607	0.589
PT	0.071	0.08	–	0.182	0.098	0.172	0.155
BI	0.105	0.069	0.043	–	0.250	0.309	0.312
TC	0.087	0.186	0.040	0.085	–	0.144	0.153
AB	0.110	0.154	0.047	0.074	0.058	–	0.341
CA	0.111	0.201	0.064	0.108	0.079	0.125	–

Table 6 Molecular analysis of variance (AMOVA) among the seven mangaba accessions

**Significant at 1% probability

Source of variation	GL	SQ	QM	Variância	%	p-value	R_{ST}
Between accessions	6	1,880,524.440	313,420.740	3230.208	8	0.001**	0.076
Within accessions	605	23,650,842.429	39,092.302	39,092.302	92		
Total	611	25,531,366.869		42,322.510	100		

highest, between the AB and LG accessions (0.607). The genetic differentiation among accessions (GST) ranged from 0.040 (PT and TC) to 0.201 (LG and CA) (Table 5).

The values observed for Nei and GST genetic distance corroborate with the estimates obtained for AMOVA (Table 6), in which the smallest proportion of genetic variation (8%) was detected among accessions, while the largest proportion (92%) was observed within accessions. In general, it is observed for species with cross-fertilization, such as mangaba, that 10–20% of the genetic variation is found between populations and that for autogamous species this value is higher than 50% (Al Salameen et al. 2018). This pattern was also observed in a study conducted on natural populations of mangaba using RAPD markers (Fajardo et al. 2018).

The estimated value for the R_{ST} statistic was 0.076 (Table 6), proving the existence of moderate genetic differentiation among the accessions evaluated. The lowest genetic distance of Nei was observed between the accessions PT and TC (0.098, Table 5), which may be associated with the origin of these accessions (Indiaroba, Sergipe, Brazil). The G_{ST} values between PT and BI (0.043); PT and TC (0.040); and PT and AB (0.047) are considered low according to the classification proposed by Wright (1978). The other values observed are considered moderate. The presence of private alleles is an indication of differentiation among accessions and demands strategies for the conservation of accessions possessing these alleles. The PR and CA accessions showed no private alleles (Table 2).
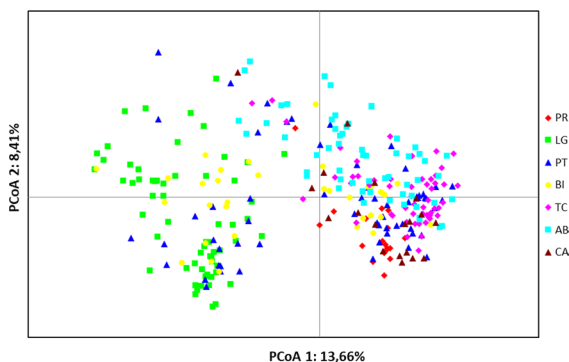

Fig. 3 Principal coordinate analysis among mangaba accessions

The principal coordinates analysis (Fig. 3), performed to evaluate the distribution of genetic variability among the accessions, did not allow us to distinguish them according to origin. The first two principal coordinates explained 22.07% of the total genetic variance of the 296 accessions, with 13.66% explained by coordinate 1 and 8.41% by coordinate 2.

The genetic distance between the accessions was estimated using Rogers' coefficient (1972) and ranged from 0.0 (between accessions BIP2.3 and BIP2.5) to 1.0 (between accessions PTP2.14 and LGP1.10; PTP2.14 and PTP2.11; PTP2.14 and PTP4; PTP2.14 and ABP2.1) (Fig. 4).

The population genetic structure analysis based on Bayesian statistics allowed the identification of two clusters (k = 2) (Fig. 5). The first cluster was composed of 197 progenies, generated from arrays of the accessions PR, PT, BI, TC, AB, and CA. The second cluster was composed of 90 accessions, generated from the LG, PT, and BI accessions. A total of nine accessions (membership values less than 80% for the two clusters detected) showed mixed ancestry.

Knowledge of the genetic structure of the germplasm is essential for the design of efficient strategies for the conservation and genetic improvement of the species. Cluster identification allows for the selection of genitors for breeding programs, which can contribute to increased genetic diversity and potential gain from selection (Campoy et al. 2016).

Population genetic structure analysis based on Bayesian statistics was used to infer on the ancestry of the accessions from the molecular information (Bernard et al. 2018). This analysis did not discriminate the accessions according to their origin, confirming the results obtained for the principal coordinates analysis (Fig. 2) and Rogers genetic distance analysis (Fig. 3). These results indicate that there is no correlation between the molecular data and the geographical origin of the analyzed accessions (Ismail et al. 2019). It was observed that 57 LG accessions, 17 PT accessions and 15 BI accessions were grouped in the same cluster with the analysis in Structure (green color, Fig. 4) and considering the first principal coordinate (Fig. 2), these accessions are grouped on the negative side. Analyzing the dendrogram (Fig. 3),

Fig. 4 Dendrogram obtained using the UPGMA clustering method based on the Rogers's genetic distance (Rogers 1972) between mangaba accessions

Fig. 5 Estimated population structure for 296 Mangaba accessions (k = 2)
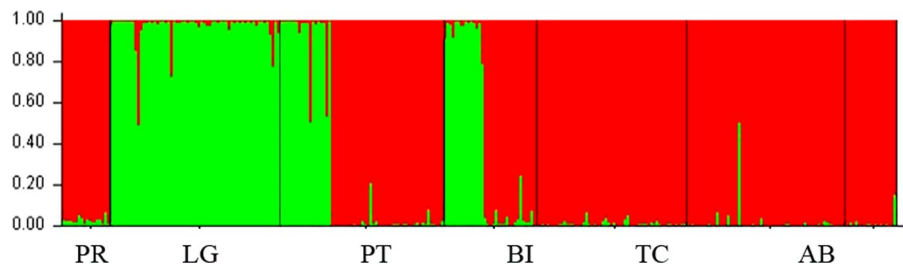
these accessions are also grouped together (blue, yellow, and green).

The creation/maintenance of a germplasm bank presents logistical and economical limitations. Thus, the creation of a core collection, which represents most of the genetic diversity present in the BAG in a smaller number of accessions, is an efficient way to reduce costs (Campoy et al. 2016; Bernard et al. 2018) and increase the efficiency of the design of conservation and genetic improvement strategies for the species. The maximum length sub tree function of the DARwin 6.0.14 software was used iteratively to eliminate redundant accessions, based on the molecular data, and allowed the selection of 225 accessions to compose the core collection of the Mangaba BAG. The seven accessions had representatives in the
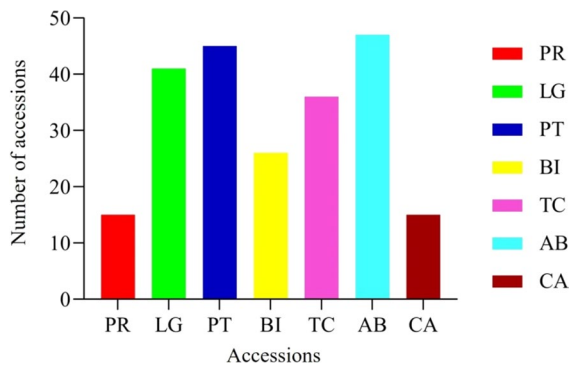
Fig. 6 Number of accessions selected to compose the Mangaba Core Collection

composition of the core collection, being 6.67% PR, 18.22% LG, 20.89% AB, 11.55% BI, 6.67% CA, 20% PT, and 16% TC (Fig. 6). The selected accessions retained 94.90% of the detected alleles.

The three approaches used to study the genetic structure of mangaba accessions (Structure, PCoA, and Dendrogram) indicated that the accessions used as a matrix have the same genetic background and share common alleles among them (Ahmed et al. 2021). Moreover, as the species mangaba presents a self-incompatibility mechanism (Darrault and Schlindwein 2005), which favors allogamy, the maintenance of the parent accessions in the same experimental field (BAG) contributed to the occurrence of gene flow and, consequently, to the genetic similarity observed among the progenies.

The use of SSR molecular markers allowed the identification of genetic variability within

and between progenies and matrices of the accessions of the Mangaba genebank, and contributed to the selection of materials to compose the core collection of this BAG, implemented in the field 15 months after sowing (Fig. 7).

Additionally, data related to agronomic and morphological characterization should be used to support the formation of this core collection, since the combination of this information contributes to the design of more efficient strategies for the use of this genetic resource.

Conclusion

Mangaba progenies from the matrices of seven accessions of the Mangaba genebank show genetic diversity within and among accessions. This diversity can be exploited for conservation and direct use of this genetic resource, and the formation of the core collection is considered essential for the rational and economic management of the collection. The identification of clusters by analyzing the genetic structure of germplasm allows the efficient design of crossings (selection of parents) for breeding programs, corroborating the increase in genetic diversity and gain with selection. Additionally, the data obtained can subsidize research related to the identification of genes associated with traits of economic interest, through the correlation between the molecular marker data with the agronomic/morphological.

Fig. 7 Production of mangaba progenies and implementation of the core collection area. Itaporanga d'Ajuda, Sergipe, Brazil

Declarations

Conflict of interest The authors have no relevant financial or non-financial interests to disclose.

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