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Genetic diversity of avocado (*Persea americana* Mill.) germplasm in Vietnam using RAPD and ISSR molecular markers

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Abstract

Avocado is a significant cash crop in Vietnam, while little is known about its genetic diversification. Random amplified polymorphic DNA (RAPD) and inter-simple sequence repeat (ISSR) markers were performed to investigate the genetic diversity of twenty-eight imported and domestic avocado cultivars being maintained at Western Highlands Agriculture and Forestry Science Institute, Dak Lak, Vietnam. 18 RAPD and 15 ISSR primers produced 3183 and 2807 scorable bands, of which 83.92% and 71.72% were polymorphic, respectively. The RAPD markers exhibited an average PIC of 0.27 and Rp index of 12.63 whereas the mean PIC and Rp values of ISSR primers were 0.21 and 13.37, respectively. The correlation between RAPD and ISSR markers was low (r = 0.338), suggesting that the ability to resolve genetic variation among varieties may relate to the number of detected polymorphisms rather than the marker employed. Nevertheless, the correlation between ISSR and combined data was high (r = 0.791) and the correlation between RAPD and combined matrices was even higher (r = 0.897). This indicates that RAPD markers have slightly higher efficiency over ISSR to resolve genetic variation among 28 avocado cultivars. High genetic diversity among 28 avocado cultivars was revealed with the Jaccard's similarity coefficient ranging from 0.623 to 0.913 based on combined data analysis. The UPGMA dendrogram generated from combined RAPD and ISSR data grouped Vietnamese domestic and imported avocado cultivars into three different groups at 72% similarity. These results could be applied to the avocado conservation and breeding programs.

Keywords: Avocado; ISSR; genetic diversity; *Persea americana* Mill.; RAPD.
Abbreviations: ISSR_Inter-Simple Sequence Repeats; PCA_Principal Component Analysis; PIC_Polymorphism Information Content; RAPD_Random Amplified Polymorphic DNA; Rp_Resolving Power; UPGMA_Unweighted Pair-Group Method with Arithmetic Average; WASI_Western Highlands Agriculture and Forestry Science Institute.

Introduction

Avocado (*Persea americana* Mill.) is a nutritious fruit plant grown both in tropical and subtropical regions of many countries. Avocado is a polymorphic species with three botanic varieties including *P. americana* var. *Americana*, *P. americana* var. *guatemalensis* and *P. americana* var. *drymifolia* known as the West Indian, Guatemalan and Mexican, respectively. These avocado races are distinguishable ecologically. However, they are cross-compatible and hybridization could take place between two races (Bergh and Ellstrand, 1986).

Genetic diversity plays a paramount role in plant conservation and breeding programs (Govindaraj et al., 2015). Several techniques have been used to analyze the genetic diversity in plant germplasm including morphological, biochemical and DNA makers (Agarwal et al., 2008; Nguyen et al., 2009). Morphological and biochemical markers are simple and cheap. However, these markers are limited in number, and they are affected by the plant growth stages and environmental factors. The molecular markers offer many advantages over other markers since they are stable, detectable in all plant tissues and they are not subjected to environmental influence (Agarwal et al., 2008). In avocado, different molecular markers have been used to assess genetic diversity such as random amplified polymorphic DNA (RAPD) (Fiedler et al., 1998), restriction fragment length polymorphism (RFLP) (Davis et al., 1998; Furnier et al., 1990), simple sequence repeat (SSR) (Alcaraz and Hormaza, 2007; Guzmán et al., 2017; Juma et al., 2021; Schnell et al., 2003), inter-simple sequence repeat (ISSR) (Cuiris-Pérez et al., 2009), single nucleotide polymorphism (SNP) (Ge et al., 2019; Rubinstein et al., 2019; Talavera et al., 2019), minisatellites (Chen et al., 2009; Juma et al., 2021), and variable number tandem repeat (VNTR) (Mhameed et al., 1996).

In Vietnam, avocado was first introduced to Lam Dong province by the French in the 1940s (Nguyen and Vo, 1999). The plant has been propagated mainly through seed propagation for more than 80 years and has adapted to a broad range of climates, topography, and vegetation. Consequently, high genetic diversity has been accumulated in this germplasm. To date, analyses of genetic diversity based on DNA polymorphisms in avocados grown in Vietnam

have been described. Le et al. (2016) used 10 ISSR primers to evaluate the genetic variation of 11 domestic avocado accessions while Pham et al. (2019) evaluated the genetic diversity of 24 avocado accessions by using 18 SSR markers. In this study, we analyzed 28 domestic and imported avocado cultivars collected worldwide and being maintained at Western Highlands Agriculture and Forestry Science Institute (WASI), Dak Lak, Vietnam by using RAPD and ISSR markers. The detailed genetic diversity analysis among 28 avocado cultivars provides important information for breeding programs and preservation of avocado germplasm.

Results

Molecular analysis using RAPD markers

The 18 RAPD markers generated a total of 176 loci ranging in size from 0.2 to 3.0 kb with an average of 9.78 loci per primer (Table 1; Figure 1A). The proportion of polymorphic loci varied from 50.0% for primer OPQ-18 to 100% for primers OPA-01, OPB-03, OPC-01, OPC-08 and OPD-01 with a mean of 83.92% polymorphism per primer. Polymorphism Information Content (PIC) value of 18 RAPD markers ranged from a highest of 0.42 (OPD-01) to a lowest of 0.14 (OPA-02) with an average of 0.27. The resolving power (Rp) index was found to be highest for primer OPC-13 (19.50) and the lowest value was found in the primer OPB-03 (5.93) with an average of 12.63 per primer (Table 1).

The Jaccard's similarity coefficients based on RAPD data ranged from 0.534 between cultivar TA-Hb6 and Pinkerton to 0.903 between cultivar Hass and Lamb Hass, with an overall mean of 0.718 (Supplemental Table 1). The dendrogram obtained from Unweighted Pair-Group Method with Arithmetic Average (UPGMA) analysis of genetic similarity based on the RAPD markers is shown in Figure 2. With 68% similarity, the 28 avocado cultivars were grouped into three main clusters. Cluster I included 17 cultivars (034, Blackman, TA1, TA-Hb2, TA-Hb5, TA02-20, TA-Hb1, TA-Hb6, Booth 7, Reed, ThanhBich, Ruotdo, TA40, GEM, Sharwil, Fuerte and Duke 7). Cluster II was composed of ten cultivars including Lamb Hass, Hass, GA, Pinkerton, Edranol, TA17, TA21, TA-Hb3, TA-Hb4 and CuBa. Cluster III had only one cultivar TA-Hb7 (Figure 2).

Molecular analysis using ISSR markers

A total of 145 loci were produced by 15 ISSR primers with an average of 9.67 loci per primer (Table 2, Figure 1B). The primer UBC-857 exhibited the largest loci (15) whereas primer UBC-889 generated the least number of loci (6). The polymorphism ranged from 37.50% for primer UBC-840 to 100% for primer UBC-809 with an average of 71.72% polymorphic bands per primer. The highest PIC value was found for primer ISSR-T1 (0.31), whereas the lowest PIC value was for primer UBC-856 (0.04) with an average PIC index of 0.21 per primer. The average Rp value for the 15 ISSR markers was 13.37 with the highest value of 20.29 for primer UBC-857 and the lowest value of 8.86 for primer UBC-889 (Table 2).

The genetic similarity values using Jaccard's coefficient based on ISSR data varied from 0.624 to 0.934 with an average of 0.787 (Supplemental Table 2). Lamb Hass and Hass cultivars showed the highest similarity (0.934) while TA21 and GEM cultivars exhibited the lowest similarity index (0.624). The dendrogram shows the formation of three main groups of 28 avocado cultivars at a coefficient value of 75%. Cluster I was the largest cluster having 18 cultivars: 034, CuBa, ThanhBich, Ruotdo, TA02-20, TA1, TA-Hb5, TA-Hb1, TA-Hb6, TA-Hb2, TA-Hb4, Booth 7, TA-Hb7, Blackman, TA-Hb3, TA-40, TA17, and TA21. Cluster II consisted of 9 cultivars counting Lamb Hass, Hass, GEM, Pinkerton, Reed, GA, Sharwil, Fuerte and Edranol. Cluster III included only one cultivar Duke 7 (Figure 3).

Combined RAPD and ISSR analysis

A combination of 33 RAPD and ISSR primers produced 253 polymorphic loci out of 321 total loci indicating 78.36% polymorphism. The average PIC and Rp values of combined RAPD and ISSR data analysis were 0.24 and 12.96, respectively (Table 3).

Combined RAPD and ISSR data analysis revealed the similarity coefficients ranged from 0.632 to 0.913 with a mean of 0.745. The greatest coefficient was between Lamb Hass and Hass cultivars, whereas CuBa and Duke 7 showed the least similarity value of 0.632 (Supplemental Table 3). The UPGMA dendrogram obtained from the cluster analysis of combining data of both markers separated 28 avocado cultivars into three distinct clusters at 72% similarity. Interestingly, the dendrogram generated by combined data was similar to the one constructed from ISSR data analysis (Figure 4).

The Mantel test and Pearson's correlation test (r) were used to measure the correlation between similarity matrices obtained from RAPD, ISSR data (separately and in combination). A low correlation between ISSR and RAPD matrices was obtained with a correlation coefficient of 0.338 ($p<0.0001$) while matrices of RAPD or ISSR and combined data exhibited a good correlation with $r = 0.897$ and 0.719 ($p<0.0001$), respectively (Table 4).

In addition to UPGMA clustering analysis, principal component analysis (PCA) based on the pooled RAPD and ISSR data showed genetic relationships among avocado cultivars in two-dimensional space accounted for 36.06% and 18.46% of the total genetic variation. The PCA data also grouped 28 avocado cultivars into major clusters which are similar to the dendrogram produced by the cluster analysis (Figure 5). Cultivars Lamb Hass, Hass, Pinkerton, Edranol, GEM, Sharwil, Fuerte, Reed and GA were grouped into one cluster whereas 034, ThanhBich, RuotDo, TA40, Blackman, TA1, TA-Hb5, TA-Hb2, TA02-20, TA-Hb1, TA-Hb6 and Booth 7 were in the same cluster. TA-Hb3, TA-Hb4, TA-Hb7, TA17, TA21 and Cuba were gathered in one cluster. As expected, Duke 7 was out-grouped from the remaining cultivars (Figure 5).

Discussion

Molecular markers have proven to be useful in clarifying genetic relationships among individuals in avocado germplasm (Cuiris-Pérez et al., 2009; Rubinstein et al., 2019; Schnell et al., 2003; Talavera et al., 2019). Here, we analyzed the genetic diversity of 28 avocado cultivars collected from different geographical zones of the world by using RAPD and ISSR markers. Previous reports have indicated that the ISSR marker has a higher capacity to detect polymorphism and genetic diversity than RAPD markers (Pham et al., 2021; Ziekiewicz et al., 1994). However, other studies have mentioned that RAPD markers identified more polymorphism than ISSR markers in many species such as *Vigna umbellata* (Muthusamy et al., 2008); *Citrullus colocynthis* (Verma et al., 2017); *Jatropha curcas* (Gupta et al., 2008). In avocado, Fiedler et al. (1998) have shown that

Table 1. Amplification of 18 RAPD markers in 28 avocado cultivars.

Primer	Sequence	Ta (°C)	Total No. of loci	Polymorphic loci	Polymorphism (%)	Total No. of bands	PIC	Rp
OPA-01	GAGGCCCTTC	34	10	10	100.00	188	0.23	13.43
OPA-02	TGCCGAGCTG	34	6	4	66.67	134	0.14	9.57
OPA-03	AGTCAGCCAC	32	9	6	66.67	175	0.23	12.50
OPA-04	AATCGGGCTG	32	13	9	69.23	270	0.24	19.29
OPA-05	AGGGGTCTTG	32	10	8	80.00	166	0.22	11.86
OPB-01	GTTTCGCTC C	32	11	9	81.82	226	0.28	16.14
OPB-02	TGATCCCTGG	32	11	9	81.82	170	0.29	12.14
OPB-03	CATCCCCCTG	34	6	6	100.00	83	0.25	5.93
OPB-04	GGACTGGAGT	32	11	9	81.82	197	0.32	14.07
OPB-05	TGCGCCCTTC	34	10	10	100.00	173	0.34	12.36
OPB-17	AGGGAACGAG	32	10	8	80,00	193	0.27	13.79
OPC-01	TTCGAGCCAG	32	8	8	100,00	146	0.29	10.43
OPC-03	GGGGGTCTTT	32	8	7	87.50	168	0.33	12.00
OPC-04	CCGCATCTAC	32	11	8	72.73	188	0.29	13.43
OPC-08	TGGACCGGTG	34	11	11	100.00	159	0.27	11.36
OPC-13	AAGCCTCGTC	32	13	12	92.31	273	0.19	19.50
OPD-01	ACCGCGAAGG	34	12	12	100.00	143	0.42	10.21
OPQ-18	AGGCTGGGTG	34	6	3	50.00	131	0.18	9.36
Total			176	149		3183		
Average/primer			9.78	8.28	83.92	176.83	0.27	12.63
Average/cultivar						113.68		

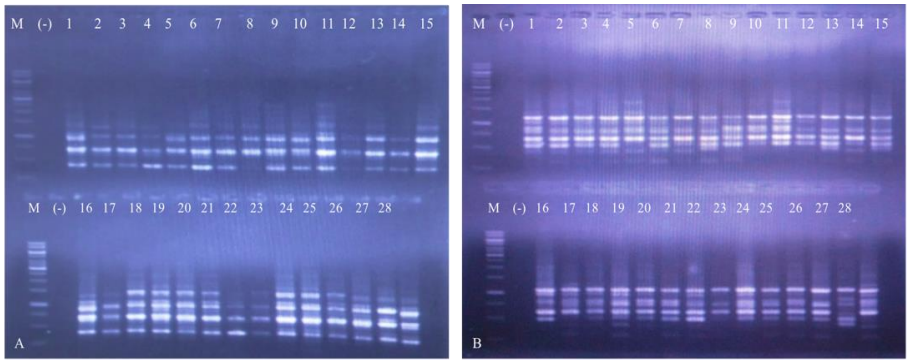

Fig 1. RAPD banding profile obtained with primer OPC-04 (A) and ISSR banding profile obtained with primer UBC-824 (B). Lane M: 1kb ladder, lane (-): blank control; lane 1 – 28: PCR products from cultivars in terms of No. given in Table 5.

Table 2. Amplification of 15 ISSR markers in 28 avocado cultivars.

Primer	Sequence	Ta (°C)	Total No. of loci	Polymorphic loci	Polymorphism (%)	Total No. of bands	PIC	Rp
UBC-809	$(AG)_8G$	52.4	7	7	100.00	206	0.25	9.64
UBC-810	$(GA)_8T$	50	7	4	57.14	284	0.09	13.29
UBC-813	$(CT)_8T$	50	10	9	90.00	135	0.22	14.14
UBC-815	$(CT)_8G$	52.4	12	8	66.67	224	0.28	16.00
UBC-824	$(TC)_8G$	52.4	10	8	80.00	178	0.27	12.71
UBC-840	$(GA)_8YT$	51.6	8	3	37.50	183	0.10	13.07
UBC-848	$(CA)_8RG$	53.9	10	7	70.00	145	0.24	14.71
UBC-856	$(AC)_8YA$	51.6	9	4	44.44	184	0.04	10.36
UBC-857	$(AC)_8YG$	53.9	15	11	73.33	142	0.23	20.29
UBC-858	$(TG)_8RT$	51.6	11	8	72.73	186	0.21	13.14
UBC-860	$(TG)_8RA$	51.6	8	6	75.00	198	0.20	10.14
UBC-889	$ACTCGTAGT(CA)_6$	47.6	6	5	83.33	124	0.20	8.86
ISSR-3	$(GACA)_4$	53.9	11	8	72.73	211	0.24	15.07
ISSR-T1	$(GT)_6CC$	44	10	8	80.00	189	0.31	13.50
ISSR-T3	$(AC)_6CG$	44	11	8	72.73	218	0.23	15.57
Total			145	104		2807		
Average/primer			9.67	6.93	71.72	187.13	0.21	13.37
Average/cultivar						100.25		

In ISSR primer sequences: R = A/G; Y = C/T.

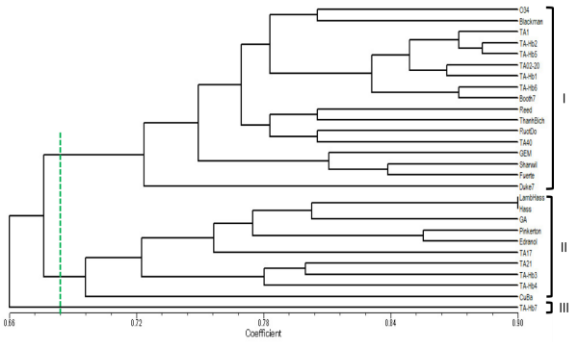

Fig 2. Dendrogram obtained with UPGMA from the Jaccard's similarity coefficients of 28 avocado cultivars based on RAPD data analysis.

Table 3. Efficiency of molecular markers for determining polymorphism in 28 avocado cultivars.

Parameters for marker efficiency	Molecular marker system		
	RAPD	ISSR	Combined RAPD and ISSR
Number of cultivars	28	28	28
Total number of primers	18	15	33
Total number of loci	176	145	321
Total number of polymorphic loci	149	104	253
Polymorphism (%)	83.92	71.72	78.36
Total number of scorable bands	3183	2807	5990
Polymorphism information content (PIC)	0.27	0.21	0.24
Resolving power (Rp)	12.63	13.37	12.96

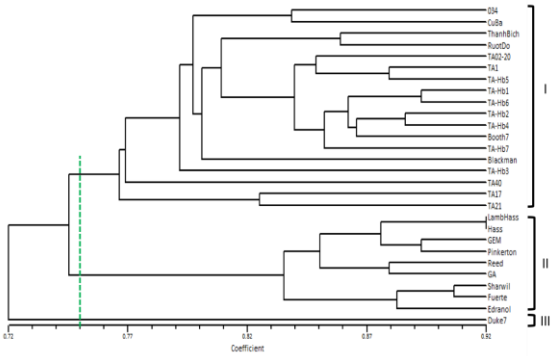

Fig 3. Dendrogram obtained with UPGMA from the Jaccard's similarity coefficients of 28 avocado cultivars based on ISSR data analysis.

Table 4. Matrix comparisons of Mantel test between markers.

Comparison	Matrix correlation (*r*)	p-value (two-tailed)	Alpha
RAPD *vs*. ISSR	0.338	< 0.0001	0.05
RAPD *vs*. combined RAPD and ISSR	0.897	< 0.0001	0.05
ISSR *vs*. combined RAPD and ISSR	0.719	< 0.0001	0.05

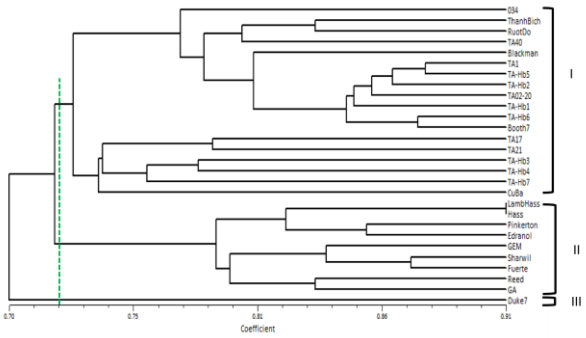

Fig 4. Dendrogram obtained with UPGMA from the Jaccard's similarity coefficients of 28 avocado cultivars based on combined RAPD and ISSR data analysis.

Table 5. List of the avocado cultivars used in this study.

No.	Cultivar	Origin	No.	Cultivar	Origin
1	034	Lam Dong (Vietnam)	15	Fuerte	USA
2	Lamb Hass	USA	16	Blackman	Mexico
3	Hass	USA	17	TA21	Dak Lak (Vietnam)
4	GEM	USA	18	TA1	Dak Lak (Vietnam)
5	TA17	Dak Lak (Vietnam)	19	TA02-20	Dak Lak (Vietnam)
6	Reed	USA	20	TA – Hb1	Dak Lak (Vietnam)
7	Pinkerton	Australia	21	TA – Hb2	Dak Lak (Vietnam)
8	GA	USA	22	TA – Hb3	Dak Lak (Vietnam)
9	ThanhBich	Dak Lak (Vietnam)	23	TA – Hb4	Dak Lak (Vietnam)
10	Ruotdo	Dak Lak (Vietnam)	24	TA – Hb5	Dak Lak (Vietnam)
11	TA40	Dak Lak (Vietnam)	25	TA – Hb6	Dak Lak (Vietnam)
12	CuBa	Dak Lak (Vietnam)	26	TA – Hb7	Dak Lak (Vietnam)
13	Sharwil	USA	27	Booth 7	USA
14	Edranol	Cuba	28	Duke 7	Australia

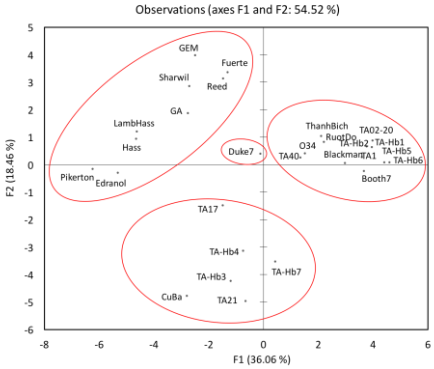

Fig 5. Principal component analysis of 28 avocado cultivars based on combined RAPD and ISSR markers. The first and second principal components account for 36.06% and 18.46% of the genetic variation, respectively.

the average percentage of polymorphism among 16 avocado accessions identified by RAPD markers was 83% whereas Cuiris-Pérez et al. (2009) indicated that the overall polymorphism of 77 avocado accessions investigated by ISSR markers ranged from 82.3% to 95.4%. Sánchez-González et al. (2020) found that the percentage of polymorphic bands produced by ISSR primers in the population of Mexico race avocado ranged from 93% up to 100%. In this study, the RAPD markers identified a higher level of polymorphism (83.92%) among 28 avocado cultivars than ISSR markers (71.72%). In addition, RAPD markers had higher PIC values than ISSR, suggesting that RAPD is more efficient than ISSR with regard to polymorphism detection. Similar results have been reported by Reyes-Alemán et al. (2018) who also used RAPD and ISSR to employ the genetic diversity analysis in avocados and indicated the average PIC value of RAPD (0.345) was greater than that of ISSR markers (0.222).

The resolving power (Rp) is used as a parameter to measure the ability of molecular markers to discriminate among accessions (Prevost and Wilkinson, 1999). In this study, the average Rp value of ISSR primers was 13.37 which is slightly higher than that of RAPD primers (12.63) (Table 3). These results suggested that ISSR would be a better marker for the determination of genetic relationships among avocado cultivars. UPGMA clustering analysis showed that both ISSR and combined RAPD+ISSR dendrograms grouped 28 avocado cultivars in the same clusters (Figure 3 and Figure 4), whereas the formation of clusters generated by RAPD profiling data was different (Figure 2, 3 and 4). In agreement with our findings, multiple studies have shown that ISSR markers provided several times more information than RAPD primers; and therefore, had high efficiency and reliability for evaluation of genetic diversity in many species (Elmeer et al., 2017; Galvan et al., 2003; Nagaoka and Ogihara, 1997).

Our findings showed that RAPD markers were more efficient for the detection of polymorphism, whereas ISSR primers were found to be more efficient for resolving genetic variation among avocado cultivars. Furthermore, the regression test showed low correlation between RAPD- and ISSR-based similarities with r = 0.338 but high regression for RAPD or ISRR and combined data (r = 0.897 and 0.719, respectively) was found (Table 4). Hence, even though both RAPD and ISSR marker systems individually can be used to study genetic diversity in avocados, we recommended the combination of two markers should be used for better discrimination and more reliable results. Combined different types of molecular markers have been usually used for assessing genetic diversity in many species (Elmeer et al., 2017; Noormohammadi et al., 2012; Pham et al., 2021, 2022).

The similarity coefficient ranged from 0.534 - 0.903 in RAPD, 0.624 – 0.934 in ISSR and 0.623 – 0.913 in combined data indicating high genetic variations among the 28 avocado cultivars evaluated. Our findings are similar to other studies that have reported high diversity in avocado germplasm (Reyes-Alemán et al., 2008; Juma et al., 2021; Pham et al., 2019; Rubinstein et al., 2019; Schnell et al., 2003). It was found that cultivar Lamb Hass was closely related to Hass on the basic similarity matrix obtained from all three analyses.

These findings were not surprising since Lamb Hass was a cross between a Hass and Gwen avocado (Rubinstein et al., 2019). The least similarity value (0.534) was obtained

between Pinkerton and TA-Hb6 cultivars based on RAPD analysis, whereas the least similarity (0.634) was found between GEM and TA21 cultivars according to ISSR analysis (Supplemental Table 1 and 2). In addition, combined RAPD and ISSR data analysis showed that the minimum similarity value (0.632) was between Duke 7 and CuBa cultivars (Supplemental Table 3). These results can be explained by the fact that TA-Hb6, TA21 and CuBa were all collected in Dak Lak (Vietnam) while Pinkerton and Duke 7 were collected from Australia, and GEM originated in America. The dendrogram of the combined data separated 28 avocado cultivars into three main clusters. Cluster I had 16 domestic and two imported avocado cultivars (Booth 7 and Blackman) while cluster II grouped 9 imported cultivars. Cultivar Duke 7 collected from Australia was singled out from the rest of the cultivars. The separation of Vietnamese domestic and imported avocado cultivars into distinct groups suggested that the genetic relationship among studied avocado cultivars could be related to the geographical environment. The mixing of two imported cultivars Booth 7 and Blackman in the group of domestic cultivars could be caused by the region-specific variations that arise when they were grown in Vietnam.

Materials and methods

Plant materials

Leaves of 28 domestic and imported avocado cultivars were sampled and stored at -20°C till further use. All the 28 cultivars are being maintained at Western Highlands Agriculture and Forestry Science Institute, Dak Lak, Vietnam. The list of avocado cultivars is presented in Table 5.

DNA extraction

Genomic DNA avocados were extracted from the leaf using TopPURE® Plant DNA Extraction Kit (ABT Biological Solutions Co., Ltd., Vietnam) following the manufacturer's instructions. DNA integrity was examined in 1.0% agarose gel electrophoresis and quantified by a spectrophotometer (BioRad, Germany). The DNA was diluted into the final concentration of 10 ng/µl and stored at -20°C for PCR amplification.

PCR amplification of RAPD and ISSR markers

A total of 18 RAPD and 15 ISSR primers were used for PCR amplification. Background information about RAPD and ISSR primers including names, sequences and annealing temperature (Ta) is presented in Table 1 and Table 2. PCR reactions were conducted with 2 µl of primer (5 µM), 0.5 µl DNA, 5 µl MyTaq Mix (Meridian Bioscience, Bioline) and 2.5 µl autoclaved distilled water. The PCR was run in ASTEC Thermal Cyclers (Gene Atlas, ASTEC, Japan) using a program of initial denaturation at 95^0C for 5 min, 35 cycles of 30 seconds denaturation at 95^0C, 30 seconds annealing at Ta that depends on specific primers and 90 seconds extension at 72^0C, followed by a 7 min final extension at 72^0C. PCR products were separated on 1% agarose gel in 1×TAE buffer by electrophoresis at 80 V for 45 min and photographed by Bio-image System (BioRad, Germany).

Data analysis

Bands amplified with ISSR, and RAPD primers were manually scored as presence (1) or absence (0). Only clear and reproducible bands were counted.

The polymorphism information content (PIC) value for each primer was calculated as proposed by Roldán-Ruiz et al. (2000): $PIC = 2f(1 - f)$. In which, f is the frequency of the present bands and ($1 - f$) is the frequency of the absent bands.

The resolving power (Rp) of each primer was calculated according to Prevost and Wilkinson (1999): $Rp = \Sigma BI$, where BI is band informativeness and takes the values of $1 - [2\times (0.5 - p)]$, in which p is the proportion of cultivars containing a particular band.

Dendrograms were generated using unweighted pair-group method with arithmetic average based on the Jaccard's similarity coefficient TREE program of NTSYS 2.1 software. The principal component analysis for three-dimensional distribution analysis and Mantel test was conducted by XLSTAT 2018 software.

Conclusion

High genetic variation among 28 analyzed avocado cultivars collected in Vietnam and from other countries was investigated by using combined RAPD and ISSR markers. The results on genetic diversity in avocado germplasm could be further used in avocado conservation, hybridization, and selection programs.

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