



Assessment of genetic diversity of sesame accessions using sequence-related amplified polymorphism (SRAP) markers

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Abstract

Sequence-related amplified polymorphism (SRAP) markers were used to assess the genetic diversity among a collection of 30 sesame accessions representing different geographical environments. A combination of 109 primers generated a high number of alleles (1031) with 100% polymorphism. The polymorphic information content (PIC) and SRAP primer index (SPI) recorded overall means of 0.83 and 8.09, respectively. Genetic similarity values based on Jaccard coefficients ranged from 0.009 to 0.781, with an average similarity value of 0.40, indicating both high genetic distance and a wide genetic basis of the investigated accessions. The unweighted pair group method with arithmetic mean (UPGMA) dendrogram grouped 30 accessions into two main clusters, and six sub clusters and four accessions failed to form clusters and were separated individually. However, subclusters separated the accessions and, considering the relatedness of accessions and their geographical origin, formed distinct diversity among groups. JTS-8 from Vriddhachalam showed the widest genetic basis compared with other introduced accessions that were distributed throughout the dendrogram, indistinguishable by cluster analysis. SRAP analysis revealed a high degree of genetic polymorphism in sesame accessions investigated and showed weak association between geographical origin and SRAP patterns. This wide genetic variability should be considered for sesame breeding programs.

Keywords: Sesame, SRAP, genetic variability

Introduction

(*Sesamum indicum* L.) belongs to the *Pedaliaceae* family (Ashri, 1998) [6] and to the Tubiflorae order (Nayar, 1976) [22], which is considered one of the ancient oilseeds, it is a diploid ($2n = 26$) Sesame dicotyledonous species. Apart from the only cultivated species, there are about 20 species of the genus *Sesamum*; *Sesamum indicum* (Elleuch *et al.*, 2011) [12]. Sesame is very drought tolerant, grows well in most soil types and regions, and has been well studied in different crop rotations (Cagirgan, 2006) [9]. It is cultivated in tropical and subtropical regions of the world (Ashri, 1998) [6], with 96% of the world's sesame seeds produced in Africa and Asia, with Sudan, India and China being the main producing countries (Dossa *et al.*, 2016) [11]. With an oil content of 44–58%, sesame seeds are an important source of oil, protein (18–25%) and carbohydrates (13.5%) (Bedigian *et al.*, 1985) [7] and are traditionally consumed directly.

In order to conserve germplasm and use it in breeding operations, it seems to be crucial to characterize and assess the genetic diversity of sesame. Molecular markers or physical features can be used to characterize genetic variety. Due to their great efficiency, abundance of information, and lack of sensitivity to environmental influences compared to morphological data, molecular marker technologies have emerged as the primary approach for evaluating plant genetic diversity and the genetic foundation of specific features. Microsatellite markers, or short sequence repeats, are a specific type of marker system that has been utilized to explore the genetic diversity and evolutionary relationships among sesame accessions (Cho *et al.*, 2011 [10]; Li-Bin *et al.*,

2008 [21]; Zhang *et al.*, 2007 [25], Zhang *et al.*, 2010 [28]); inter-simple sequence repeats (ISSR) (Kumar and Sharma, 2011) [17]; amplified fragment length polymorphism (AFLP) (Laurentin and Karlovsky, 2007) [18]; and SRAP (Zhang *et al.*, 2012 [26], Zhang *et al.*, 2011 [27], Zhang *et al.*, 2010 [28], Zhang *et al.*, 2007 [25]). SRAP is an efficient marker system technique for the characterization of genetic diversity because it possesses a high degree of reproducibility and discriminatory power, simplicity, reasonable throughput rate, discloses numerous codominant markers, allows easy isolation of bands for sequencing more reproducible than RAPDs and is easier to assay than AFLPs and, most importantly, it targets open reading frames (Li and Quiros, 2001) [20]. The present study aimed to assess genetic variability among sesame accessions collected from different ecological regions using sequence-related amplified polymorphism markers.

Material and methods

Plant materials

Thirty sesame accessions and their origins are presented in Table 1. Out of them, thirty sesame accessions were kindly obtained from Regional research station, Vriddhachalam and Agricultural research station, JAU, Amreli were used in this study. These accessions were collected and conserved in seed gene banks representing sesame genetic resources for each region. The list of sesame accessions and their origin is given in Table 1 SRAP based PCR primers were applied on following accessions with different combinations.

Table 1: List and source of Sesame accessions used for the current study

Sr. No.	Accessions	Types	Source
1	TMV-4	Brown	Regional research station, Vriddhachalam
2	TMV-7	Brown	Regional research station, Vriddhachalam
3	VRI-1	Brown	Regional research station, Vriddhachalam
4	VRI-2	Brown	Regional research station, Vriddhachalam
5	VRI-3	White	Regional research station, Vriddhachalam
6	VRI-4	Brown	Regional research station, Vriddhachalam
7	VRI-5	White	Regional research station, Vriddhachalam
8	Paiyur-1	Black	Regional research station, Vriddhachalam
9	CO-1	Black	Regional research station, Vriddhachalam
10	Smak	White	Regional research station, Vriddhachalam
11	Amrit	Brown	Regional research station, Vriddhachalam
12	Nirmala	White	Regional research station, Vriddhachalam
13	Kalinga Sesame 3-1	White	Regional research station, Vriddhachalam
14	Prachi	Brown	Regional research station, Vriddhachalam
15	Subhra	White	Regional research station, Vriddhachalam
16	TKG-21	White	Regional research station, Vriddhachalam
17	TKG-22	White	Regional research station, Vriddhachalam
18	TKG-55	White	Regional research station, Vriddhachalam
19	TKG-306	White	Regional research station, Vriddhachalam
20	TKG-308	White	Regional research station, Vriddhachalam
21	JTS-8	White	Regional research station, Vriddhachalam
22	G-Til-1	White	Agricultural research station, JAU, Amreli
23	G-Til-2	White	Agricultural research station, JAU, Amreli
24	G-Til-3	White	Agricultural research station, JAU, Amreli
25	G-Til-4	White	Agricultural research station, JAU, Amreli
26	G-Til-5	White	Agricultural research station, JAU, Amreli
27	G-Til-6	White	Agricultural research station, JAU, Amreli
28	G-Til-10	Black	Agricultural research station, JAU, Amreli
29	G-Til-11	Black	Agricultural research station, JAU, Amreli
30	Purva	Brown	Agricultural research station, JAU, Amreli

DNA Extraction

DNA extraction was carried out using CTAB method. The leaf sample was grinded to a fine powder of extraction buffer was added and mixed and then incubated at 65°C for 30 min. An equal volume of chloroform: isoamyl alcohol (24:1 v/v) was added for protein precipitation. The samples were centrifuged at 12000 rpm for 15 min at 4°C. DNA was precipitated using ice-cold isopropanol, and the pellet was washed using 80% ethanol. The quality and concentration of extracted DNA were detected using 0.8% agarose gel electrophoresis. Dilution with TE was carried out.

Sequence-related amplified polymorphism (SRAP) markers

Sequence-related amplified polymorphism (SRAP) markers SRAP-PCR amplification was performed following the methods of Ammar *et al.*, (2015)^[4]. The PCR reaction was

performed with different combinations of the forward and reverse primers, 50 ng template DNA, and nuclease-free water up to 20 ul. Amplification was carried out on a Variti thermal cyclers (Applied Biosystems). The PCR was set with initial denaturation at 94°C for 5 min followed by five cycles of denaturation at 94°C for 1 min, annealing at 35°C for 1 min and elongation at 72°C for 1 min. In the remaining 30 cycles, the annealing temperature was increased to 50°C for 1 min with a final extension step at 72°C for 15 min.

Separation and photographing of PCR products

PCR products were separated on 1.8% agarose gel in 1X TBE buffer (89 mM Tris, 89 mM Boric acid, 2 mM EDTA) at 115 volt for 1.5-2 h. A 100 bp standard DNA ladder as the molecular standard marker was used to confirm the appropriate (SRAP bands). The banding patterns were photographed under UV light for further analysis.

Table 2: SRAP Primer sequence utilized for polymorphism

Primer code	Primer sequence (5'—3') Forward primer	Primer code	Primer sequence (5'—3') Reverse primer
Me01	TGAGTCCAAACCGGATA	Em01	GACTGCGTACGAATTAAT
Me02	TGAGTCCAAACCGGAGC	Em02	GACTGCGTACGAATTTGC
Me03	TGAGTCCAAACCGGAAT	Em03	GACTGCGTACGAATTGAC
Me04	TGAGTCCAAACCGGACC	Em04	GACTGCGTACGAATTTGA
Me05	TGAGTCCAAACCGGAAG	Em05	GACTGCGTACGAATTAAC
Me06	TGAGTCCAAACCGGTAA	Em06	GACTGCGTACGAATTGCA
Me07	TGAGTCCAAACCGGTCC	Em07	GACTGCGTACGAATTCOA
Me08	TGAGTCCAAACCGGTGC	Em08	GACTGCGTACGAATTCTG
Me09	TGAGTCCAAACCGGACG	Em09	GACTGCGTACGAATTCCGA
Me10	TGAGTCCAAACCGGACT	Em10	GACTGCGTACGAATTCAG
Me11	TGAGTCCAAACCGGAGG	Em11	GACTGCGTACGAATTCCA

Molecular Data analysis

DNA-bands were scored as present (1) or absent (0) for each primer pair. The genetic similarities were calculated using the data matrix was read by NTSYS-pc version 2.2 (numerical taxonomy and multivariate analysis system for personal computers, exeter software) (Rohlf, 1998) [24]. Data generated from SRAP analysis were analyzed using the Jaccard similarity coefficient (Jaccard, 1908) [14]. These similarity coefficients were used to construct a dendrogram using the unweighted pair group method with arithmetic average (UPGMA) program. The polymorphism information $j=1$ content (PIC) for each primer was calculated to estimate its allelic variation as follows: $PIC_i=1-\sum P_{ij}^2$, where P_{ij} is the frequency of the i th allele for marker j and the summation extends over n alleles, calculated for each SRAP marker (Anderson *et al.*, 1993) [5]. Discrimination power was calculated by dividing the number of polymorphic alleles amplified for each primer by the total number of polymorphic alleles obtained (Khierallah *et al.*, 2011) [15].

Results and discussion

121 primer pair combinations were screened on 30 accessions selected from different resources to study the ability of primers to amplify DNA fragments. The characteristics of the 109 primers combination are listed in Table 2. These primers showed good amplification. All primers showed 100% polymorphism percentage. Primers produced high numbers of bands across accessions; in total, they generated 1031 bands ranging from 4 bands for primer pair combinations (SRAP-31) to 18 for (SRAP-15) with an average of 9.45 bands per primer pair combination across accessions. The polymorphic information content (PIC) recorded for each primer combination ranged from 0.390 for primer combination (SRAP-31) to 0.920 for primer combination (SRAP-15). The overall mean for PIC values was 0.835. The SRAP primer index (SPI) recorded for each primer combination ranged from 1.550 for primer combination (SRAP-31) to 16.510 for primer combination (SRAP-15). The overall mean for PIC values was 8.089 (Table 3). Similarity matrix using Dice coefficient varied from 0.011 between V25 to 1 between V15 and V13, as shown in Table 4.

The dendrogram generated is presented in Figure 1. The molecular analysis showed that variation among the accessions was low 30-40%. The dendrogram revealed two main clusters, A and B, at similarity index of 16%. Cluster A was subdivided into three subclusters, I, II and III. Cluster B was also subclustered into two, IV and V. Subcluster I consisted of 18 accessions from Vriddhachalam 1, 4, 5, 6, 8, 9, 3, 15, 2, 7, 16, 10, 11, 12, 14, 17, 18 and 19. Another Vriddhachalam accession, 13, lonely in cluster II and accession 20 was also lonely in cluster III. They both related to Subcluster I accessions at similarity index of 36%. Subcluster IV consists of all accessions from Amreli 22, 23, 24, 26, 27, 25, 28 and 29. Another Amreli accession, 30, lonely in cluster V. Accession 21 from Vriddhachalam stood alone in subcluster VI. Accessions from a subcluster I, II and III that are found in one cluster (A) may be more closely related to that particular cluster mates than the members outside that cluster.

The UPGMA cluster analysis of the accessions based on the SRAP data was cut at a similarity of 0.16 (which represented the distance from the maximum similarity of

0.78 to the minimum of 0.01). Cutting the dendrogram at this similarity value resulted in two main clusters comprising 97% of the accessions, two subcluster II and IV comprising 59.4% and 26.4% respectively, and four single clusters comprising 14.13% of the accessions.

A data matrix plot estimating genetic differentiation among 30 accessions of sesame. The scattered plot disclosed a pattern of five groups. The plot showed that the accessions, TMV-4 and JTS-8 were distinctly different from each other. Most of the accessions were scattered between these two (TMV-4 and JTS-8) varieties with extremely low factor scores along the dimensions Figure 2. The cophenetic correlation coefficient (0.97) indicated little distortion between the original similarity values from the similarity matrix and the values used to construct the dendrogram. The 30 sesame accessions could be classified into five clusters. Result was consistent with bipolt of PCoA analysis shown in Figure 2.

In order to quantify the level of polymorphism detected through Jaccard's estimate of similarity coefficients based on the probability that an amplified fragment from one plant will also be found in another was used to generate a similarity matrix. All accessions fall in the range of 0.009 to 0.781 (shown by bold figure in Table 4) with the average being 0.40. The similarity coefficients values were highest between the accessions G-Til-1 and G-Til-2 (0.781), followed by G-Til-5 and G-Til-6 (0.761), TKG-55 and TKG-306 (0.752) and by G-Til-2 and G-Til-3 (0.742). These accessions showed maximum degree of similarity (> 78%) in their genetic makeup. However, the minimum values of similarity coefficients were observed between VRI-5 and JTS-8 (0.009) followed by Smark and JTS-8, Amrit and JTS-8 (0.010) and VRI-2 and JTS-8, TMV-7 and JTS-8, Subhra and JTS-8, G-Til-1 and JTS-8, G-Til-2 and JTS-8, G-Til-3 and JTS-8, G-Til-4 and JTS-8, G-Til-5 and JTS-8, G-Til-6 and JTS-8, G-Til-10 and JTS-8, G-Til-11 and JTS-8, and Purva and JTS-8, (0.011). Thus these accessions were highly diverse. The crossing among these accessions would likely to produce highly heterotic individuals and could be used in heterosis breeding (Table 4). These findings were in accordance with the previous studies of Aboelnaga *et al.*, (2020) [1] showed that similarity matrix using Dice coefficient varied from 0.78 between V32 and V25 to 1 between V15 and V13.

Sesamum indicum L. has a large genetic variability, which should be taken into account when planning conservation strategies or when sesame variability is used in breeding programs. This high level of polymorphism in sesame has been reported before for its morphology. Early molecular studies have not confirmed this. A DNA-based marker SRAP has been used in studying genetic variability in sesame.

In the present study, SRAP was successfully applied on sesame genetic diversity in a representative sample of the collection of 30 sesame accessions representing different geographical environments. The combination of 109 primers generated a higher number of alleles (1031) with 100% polymorphism, indicating the suitability of selected primers for detecting genetic variability among the accessions tested. Although differences in both the number of primers and accessions used in this study compared with other studies, the polymorphisms were higher than those reported by Zhang *et al.*, (2010 [28], 2011 [27], 2012 [26]), who also

reported an equivalent amount of polymorphic alleles per locus in 67 sesame cultivars. In contrast to these findings, Zhang *et al.*, (2011) [27] recorded an average of 14.5 and 12.5 polymorphic alleles per primer combination, compared with 21.6 polymorphic alleles in this study. The higher number of amplified fragments detected in this study associated with higher polymorphism percentages compared with other studies is mainly due to the accessions, primer combination, used as well as the laser-based detection system in capillary electrophoresis with more extraordinary resolution power than ordinary gel electrophoresis (Ammar *et al.*, 2015) [4].

The higher values of Polymorphic Information Content (PIC) (range = 0.39–0.92, average = 0.83) and SRAP Primer Index (SPI) (range = 1.55–16.50, average = 8.09) recorded in this study indicated the genetic diversity of the accessions. The results of PIC values in this study exceeded those of Zhang *et al.*, (2012) [26], who recorded an average of 0.20 in minicore and 0.18 in Chinese sesame core collection. SRAP primers yielded more than twice the number of polymorphic bands per primer with the highest average polymorphism information content (PIC) in comparative studies including different molecular markers (Robarts and Wolfe, 2014) [23], where markers with higher PIC values possessed greater potential to reveal allelic variation. High PIC values were used as selection criteria to develop a mini-core collection of Chinese sesame core collection (Zhang *et al.*, 2012) [26]. PIC was also used to measure informativeness of primer combinations in many crops including soybean, wheat, maize, and different *Medicago* species. In this study, pair wise genetic similarity indices between all 30 accessions (range = 0.01–0.78, average = 0.40) divided the accessions to six clusters. The Jaccard genetic similarity coefficient indicated that genetic relationships were the closest between accessions in group three (0.36) and the most distant in the second group (0.54). Accessions in group three were introduced from Vriddhachalam, while those in the second group were introduced from Amreli. This result compare with other

study, pair wise genetic similarity indices between all 52 accessions (range = 0.12–0.49, average = 0.30) divided the accessions to seven main clusters. The Jaccard genetic similarity coefficient indicated that genetic relationships were the closest between accessions in group seven (0.34–0.40) and the most distant in the second group (0.23–0.42). Accessions in group seven were introduced from Pakistan, while those in the second group were introduced from different geographical regions (Ali *et al.*, 2017) [2].

For sesame, the conventional wisdom that choosing accessions from various geographic origins will optimize the diversity accessible to a breeding operation is not true (Laurentin and Karlovsky, 2006) [19]. The extent of gene flow in a species depends on the distribution of the habitats it occupies, the size and degree of isolation of its populations, and the movement of pollen and seeds between populations, as Hartings *et al.*, (2008) [13] demonstrated. Genetic diversity is distributed both within and among populations and is a function of the rate of gene flow between populations. The reduction in genetic diversity and the fuzziness of the borders between genetic and geographic groupings may also be caused by natural out-crossing (Kumar *et al.*, 2012 [16] and Ali *et al.*, 2007 [3]). however, suggested that

AFLP markers and geographic origin dictated the close genetic relationships amongst the accessions. The results were corroborated by Bedigian (2010) [8], who noted patterns of genotypic diversity in sesame that followed regional lines. The accessions were primarily clustered according to their geographical origin as well as morphological traits. Zhang *et al.*, (2010) [28] have found that the higher genetic diversity may be partially explained by the complexity of the climate and geographical elements, planting practices, and the relative importance of the crop in the region. Because of this, cultivar improvement was not given much thought, and many ancient landraces were maintained while maintaining a higher level of genetic diversity.

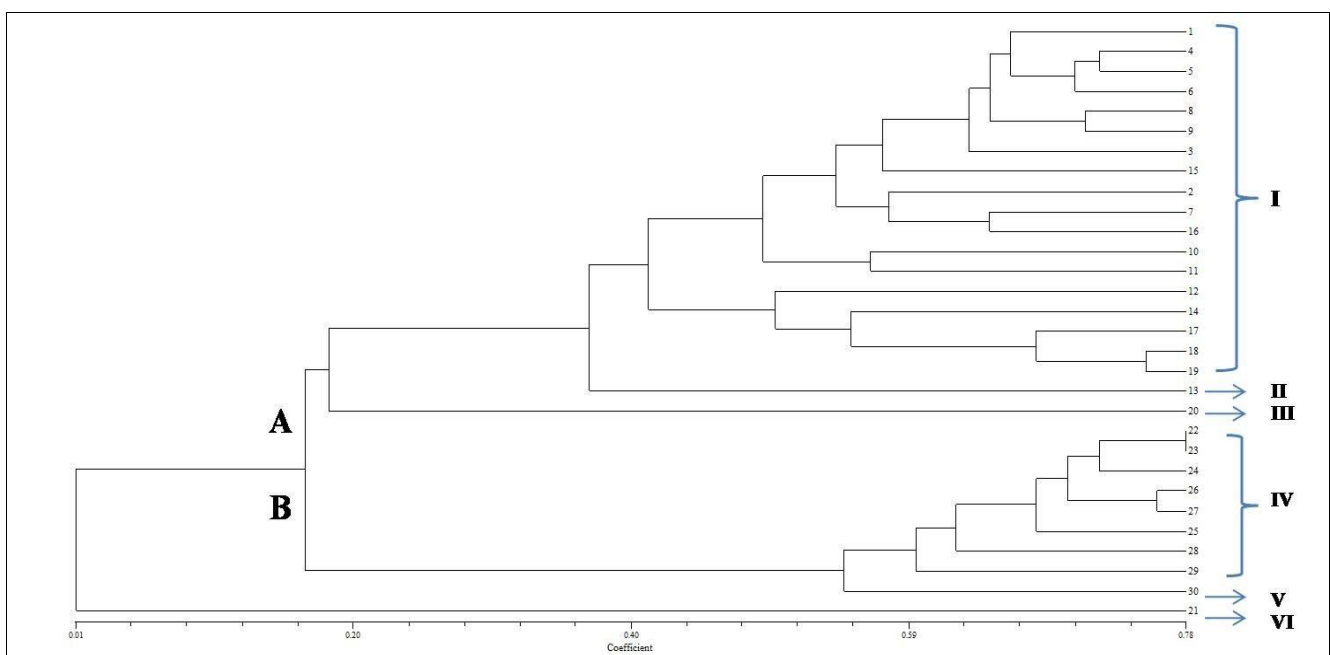


Fig 1: Dendrogram produced by Jaccard's coefficient and the unweighted pair group method with arithmetic average (UPGMA) clustering method based on SRAP data in 30 sesame accessions.

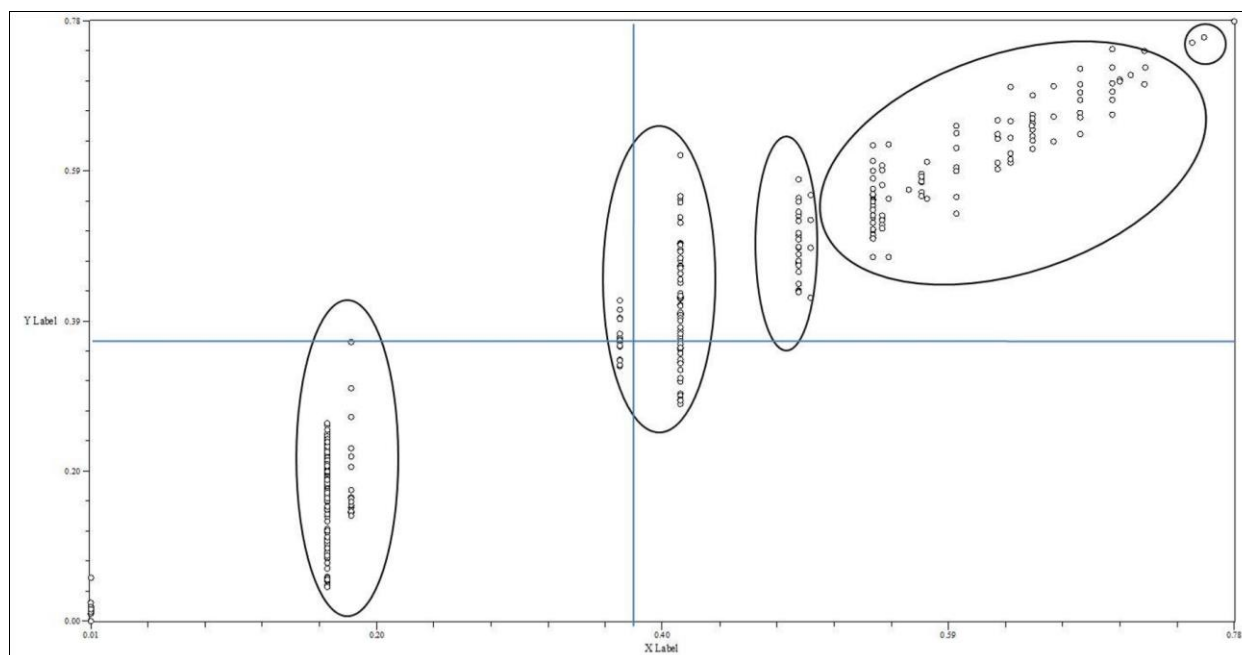


Fig 2: Two-dimensional matrix plot of 30 accessions was computed through of principal component analysis.

Table 3: Size, number of amplified bands, per cent polymorphism and PIC obtained by SRAP primers in 30 sesame varieties.

Primer	Combinations	SRAP primer	Polymorphic bands	PIC	SPI
1	ME-01 EM-01	SRAP-01	6	0.82	4.92
2	ME-01 EM-02	SRAP-02	9	0.83	7.51
3	ME-01 EM-03	SRAP-03	13	0.89	11.6
4	ME-01 EM-04	SRAP-04	14	0.9	12.7
5	ME-01 EM-05	SRAP-05	5	0.75	3.74
6	ME-01 EM-06	SRAP-06	14	0.92	12.9
7	ME-01 EM-07	SRAP-07	12	0.87	10.38
8	ME-01 EM-08	SRAP-08	8	0.82	6.55
9	ME-01 EM-09	SRAP-09	13	0.91	11.84
10	ME-01 EM-10	SRAP-10	10	0.87	8.74
11	ME-01 EM-11	SRAP-11	6	0.72	4.35
12	ME-02 EM-01	SRAP-12	11	0.89	9.78
13	ME-02 EM-02	SRAP-13	10	0.81	8.16
14	ME-02 EM-03	SRAP-14	7	0.77	5.41
15	ME-02 EM-04	SRAP-15	18	0.92	16.51
16	ME-02 EM-05	SRAP-16	15	0.91	13.64
17	ME-02 EM-06	SRAP-17	9	0.85	7.74
18	ME-02 EM-07	SRAP-18	11	0.89	9.79
19	ME-02 EM-08	SRAP-19	9	0.85	7.73
20	ME-02 EM-09	SRAP-20	8	0.83	6.67
21	ME-02 EM-10	SRAP-21	8	0.84	6.73
22	ME-02 EM-11	SRAP-22	7	0.82	5.72
23	ME-03 EM-01	SRAP-23	10	0.82	8.25
24	ME-03 EM-02	SRAP-24	9	0.84	7.59
25	ME-03 EM-03	SRAP-25	14	0.9	12.64
26	ME-03 EM-04	SRAP-26	10	0.86	8.64
27	ME-03 EM-05	SRAP-27	14	0.89	12.41
28	ME-03 EM-06	SRAP-28	12	0.89	10.65
29	ME-03 EM-07	SRAP-29	9	0.8	7.19
30	ME-03 EM-08	SRAP-30	11	0.88	9.72
31	ME-03 EM-09	SRAP-31	4	0.39	1.55
32	ME-03 EM-10	SRAP-32	5	0.71	3.57
33	ME-03 EM-11	SRAP-33	14	0.9	12.61
34	ME-04 EM-01	SRAP-34	9	0.87	7.83
35	ME-04 EM-02	SRAP-35	9	0.86	7.74
36	ME-04 EM-03	SRAP-36	6	0.76	4.54
37	ME-04 EM-04	SRAP-37	7	0.79	5.52
38	ME-04 EM-06	SRAP-38	9	0.85	7.66
39	ME-04 EM-07	SRAP-39	7	0.83	5.88
40	ME-04 EM-08	SRAP-40	4	0.68	2.71

41	ME-04 EM-09	SRAP-41	8	0.85	6.76
42	ME-04 EM-10	SRAP-42	9	0.84	7.6
43	ME-04 EM-11	SRAP-43	6	0.82	4.93
44	ME-05 EM-01	SRAP-44	13	0.9	11.73
45	ME-05 EM-02	SRAP-45	12	0.89	10.73
46	ME-05 EM-03	SRAP-46	11	0.88	9.73
47	ME-05 EM-04	SRAP-47	5	0.66	3.32
48	ME-05 EM-05	SRAP-48	5	0.78	3.9
49	ME-05 EM-06	SRAP-49	7	0.81	5.65
50	ME-05 EM-07	SRAP-50	4	0.69	2.77
51	ME-05 EM-08	SRAP-51	9	0.85	7.64
52	ME-05 EM-09	SRAP-52	8	0.83	6.67
53	ME-05 EM-10	SRAP-53	7	0.79	5.52
54	ME-05 EM-11	SRAP-54	9	0.79	7.1
55	ME-06 EM-01	SRAP-55	10	0.86	8.85
56	ME-06 EM-02	SRAP-56	11	0.89	9.75
57	ME-06 EM-03	SRAP-57	14	0.91	12.8
58	ME-06 EM-04	SRAP-58	15	0.9	13.56
59	ME-06 EM-05	SRAP-59	14	0.9	12.67
60	ME-06 EM-06	SRAP-60	8	0.84	6.72
61	ME-06 EM-07	SRAP-61	8	0.81	6.49
62	ME-06 EM-08	SRAP-62	13	0.9	11.71
63	ME-06 EM-09	SRAP-63	5	0.76	3.78
64	ME-06 EM-10	SRAP-64	11	0.86	9.49
65	ME-06 EM-11	SRAP-65	9	0.88	7.88
66	ME-07 EM-01	SRAP-66	11	0.88	9.65
67	ME-07 EM-02	SRAP-67	13	0.86	11.22
68	ME-07 EM-03	SRAP-68	14	0.9	12.67
69	ME-07 EM-04	SRAP-69	9	0.83	7.46
70	ME-07 EM-05	SRAP-70	13	0.89	11.63
71	ME-07 EM-06	SRAP-71	8	0.85	6.81
72	ME-07 EM-07	SRAP-72	7	0.83	5.83
73	ME-07 EM-08	SRAP-73	9	0.87	7.82
74	ME-07 EM-09	SRAP-74	15	0.9	13.44
75	ME-07 EM-10	SRAP-75	6	0.81	4.84
76	ME-07 EM-11	SRAP-76	5	0.7	3.5
77	ME-08 EM-01	SRAP-77	12	0.89	10.72
78	ME-08 EM-02	SRAP-78	10	0.88	8.81
79	ME-08 EM-03	SRAP-79	15	0.9	13.42
80	ME-08 EM-04	SRAP-80	10	0.85	8.51
81	ME-08 EM-05	SRAP-81	14	0.91	12.68
82	ME-08 EM-06	SRAP-82	6	0.79	4.71
83	ME-08 EM-07	SRAP-83	10	0.86	8.61
84	ME-08 EM-08	SRAP-84	10	0.87	8.74
85	ME-08 EM-09	SRAP-85	12	0.9	10.76
86	ME-08 EM-10	SRAP-86	17	0.92	15.65
87	ME-08 EM-11	SRAP-87	7	0.77	5.41
88	ME-09 EM-01	SRAP-88	8	0.85	6.77
89	ME-09 EM-02	SRAP-89	6	0.67	4
90	ME-09 EM-03	SRAP-90	6	0.77	4.59
91	ME-09 EM-04	SRAP-91	7	0.77	5.38
92	ME-09 EM-05	SRAP-92	10	0.89	8.94
93	ME-09 EM-06	SRAP-93	11	0.86	9.43
94	ME-09 EM-07	SRAP-94	7	0.8	5.59
95	ME-09 EM-08	SRAP-95	8	0.81	6.52
96	ME-09 EM-09	SRAP-96	6	0.76	4.59
97	ME-09 EM-11	SRAP-97	7	0.79	5.55
98	ME-10 EM-08	SRAP-98	7	0.82	5.71
99	ME-10 EM-10	SRAP-99	9	0.84	7.6
100	ME-10 EM-11	SRAP-100	9	0.88	7.89
101	ME-11 EM-01	SRAP-101	4	0.65	2.59
102	ME-11 EM-02	SRAP-102	9	0.82	7.41
103	ME-11 EM-03	SRAP-103	9	0.84	7.56
104	ME-11 EM-04	SRAP-104	5	0.79	3.96
105	ME-11 EM-05	SRAP-105	7	0.84	5.91
106	ME-11 EM-07	SRAP-106	13	0.91	11.88
107	ME-11 EM-08	SRAP-107	13	0.91	11.78
108	ME-11 EM-09	SRAP-108	8	0.84	6.71

109	ME-11 EM-11	SRAP-109	12	0.91	10.86
	Total no of bands		1031		
	Average		9.46	0.84	8.09

Table 4: Dice similarity coefficient of 30 sesame accessions based on SRAP data analysis.

Row /Cols	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30
1	1.000																													
2	0.556	1.000																												
3	0.650	0.508	1.000																											
4	0.696	0.541	0.670	1.000																										
5	0.625	0.549	0.629	0.721	1.000																									
6	0.652	0.554	0.596	0.705	0.702	1.000																								
7	0.563	0.698	0.526	0.555	0.600	0.619	1.000																							
8	0.632	0.499	0.602	0.684	0.615	0.625	0.507	1.000																						
9	0.640	0.528	0.609	0.659	0.648	0.655	0.576	0.711	1.000																					
10	0.502	0.478	0.486	0.505	0.528	0.497	0.551	0.533	0.575	1.000																				
11	0.465	0.469	0.430	0.455	0.430	0.429	0.470	0.439	0.463	0.562	1.000																			
12	0.489	0.549	0.452	0.464	0.469	0.462	0.526	0.459	0.491	0.484	0.492	1.000																		
13	0.340	0.365	0.358	0.367	0.332	0.339	0.333	0.367	0.369	0.365	0.363	0.394	1.000																	
14	0.296	0.378	0.313	0.311	0.293	0.283	0.312	0.288	0.289	0.288	0.316	0.421	0.405	1.000																
15	0.572	0.502	0.573	0.583	0.558	0.573	0.548	0.554	0.578	0.546	0.463	0.491	0.374	0.328	1.000															
16	0.509	0.551	0.503	0.589	0.546	0.498	0.645	0.475	0.536	0.521	0.488	0.591	0.358	0.337	0.587	1.000														
17	0.427	0.606	0.398	0.422	0.440	0.423	0.490	0.383	0.420	0.398	0.400	0.544	0.363	0.474	0.460	0.545	1.000													
18	0.361	0.481	0.361	0.368	0.353	0.340	0.371	0.349	0.355	0.336	0.356	0.486	0.417	0.621	0.374	0.410	0.634	1.000												
19	0.400	0.553	0.402	0.421	0.412	0.399	0.423	0.394	0.410	0.382	0.391	0.522	0.393	0.550	0.446	0.473	0.720	0.752	1.000											
20	0.154	0.171	0.162	0.160	0.148	0.142	0.140	0.143	0.155	0.137	0.150	0.201	0.225	0.364	0.151	0.156	0.214	0.303	0.266	1.000										
21	0.013	0.011	0.015	0.011	0.013	0.013	0.009	0.012	0.012	0.010	0.010	0.012	0.019	0.024	0.011	0.009	0.012	0.018	0.156	0.056	1.000									
22	0.191	0.193	0.178	0.183	0.214	0.199	0.210	0.164	0.178	0.229	0.212	0.186	0.100	0.101	0.196	0.204	0.190	0.117	0.147	0.053	0.011	1.000								
23	0.189	0.199	0.182	0.183	0.213	0.203	0.231	0.167	0.185	0.237	0.256	0.195	0.094	0.090	0.195	0.225	0.197	0.115	0.145	0.057	0.011	0.781	1.000							
24	0.211	0.235	0.205	0.202	0.242	0.222	0.255	0.188	0.209	0.257	0.238	0.213	0.095	0.088	0.211	0.240	0.204	0.119	0.149	0.047	0.011	0.699	0.742	1.000						
25	0.214	0.217	0.198	0.205	0.233	0.217	0.244	0.181	0.130	0.249	0.233	0.215	0.091	0.094	0.199	0.241	0.203	0.118	0.149	0.057	0.011	0.679	0.662	0.699	1.000					
26	0.186	0.201	0.165	0.179	0.213	0.195	0.217	0.155	0.180	0.236	0.213	0.194	0.087	0.082	0.189	0.213	0.181	0.100	0.130	0.044	0.011	0.721	0.745	0.700	0.688	1.000				
27	0.186	0.203	0.177	0.186	0.207	0.209	0.228	0.156	0.195	0.233	0.206	0.194	0.087	0.082	0.182	0.213	0.187	0.100	0.136	0.052	0.011	0.660	0.690	0.679	0.656	0.761	1.000			
28	0.178	0.188	0.160	0.169	0.204	0.189	0.209	0.145	0.173	0.220	0.200	0.177	0.085	0.076	0.176	0.199	0.179	0.107	0.130	0.053	0.011	0.597	0.630	0.629	0.588	0.652	0.634	1.000		
29	0.180	0.177	0.163	0.169	0.204	0.194	0.196	0.155	0.176	0.201	0.195	0.181	0.089	0.089	0.180	0.188	0.185	0.104	0.130	0.053	0.011	0.590	0.586	0.552	0.530	0.635	0.645	0.617	1.000	
30	0.157	0.166	0.148	0.156	0.172	0.169	0.163	0.140	0.160	0.184	0.167	0.157	0.860	0.068	0.146	0.160	0.149	0.095	0.109	0.054	0.011	0.524	0.528	0.516	0.523	0.594	0.568	0.511	0.587	1.000

Conclusion

In conclusion, the aim of this analysis was to analyze molecular characterization, marker polymorphisms, morphometric and yield-related traits and genetic variability analysis between 30 genotypes of *Sesamum indicum* L. from different geographical region. Our SRAP-based study on sesame accessions collected from different geographical regions found a high level of polymorphism in this DNA-based marker and did not show an association between geographical origin and SRAP patterns. 109 SRAP primers showed clear amplification among the 30 sesame genotypes with 100% polymorphism. The large number of polymorphic-amplified fragments produced in this study (1031), with an average of 9.45 fragments per primer pair indicates that this system is a reliable and powerful tool to evaluate genetic polymorphisms and relationships among sesame accessions. Such information will be useful to determine optimal breeding strategies, and to allow continued progress in sesame breeding. Diverse genetic backgrounds among parental lines provide a large supply for allelic variations that can be used to develop new favourable gene combinations in sesame breeding programs. It is necessary to reinforce the collection and protection of sesame germplasm resources from agro-ecological zones with higher diversity. Furthermore, genetic distance between parents should be considered for sesame breeding programs.

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