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USE OF TRAP MARKERS TO DETERMINE GENETIC RELATEDNESS IN NEW CASTOR BEAN (*RICINUS COMMUNIS* L.) LINES

Martin Vivodík [1], Emre Sevindik[2], Želmíra Balážová[1], Milan Chňapek[1] and Martin Bačkor[1]*

Address(es): doc. Ing. Martin Vivodík, PhD.
[1] Slovak University of Agriculture, Faculty of Biotechnology and Food Sciences, Institute of Biotechnology, Tr. A. Hlinku 2, 949 76 Nitra, Slovakia.
[2] Aydın Adnan Menderes University, Faculty of Agriculture, Department of Agricultural Biotechnology, South Campus, Cakmar, Aydin, Türkiye.

*Corresponding author: martin.vivodik@uniag.sk

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ABSTRACT

Castor bean (*Ricinus communis* L.) is a plant of immense importance worldwide and its main industrial use is for the production of castor oil. Knowing the diversity of castor is important in order to breed plants that will produce the most castor oil possible. In our work, we used a combination of 30 TRAP markers and 6 random primers to study genetic relatedness and similarity in a set of 111 new castor bean lines. Based on the results obtained with TRAP markers, we calculated the PIC value to determine genetic diversity. The 66 combinations of TRAP markers and 6 random primers we used together and determined 528 DNA bands that were present in all 111 castor bean lines analyzed. The lowest number of fragments (5) was determined using 7 combinations of TRAP markers and random primers (TRAP 04 $_x$ arb 1, TRAP 19 $_x$ arb 1, TRAP 40 $_x$ arb 2, TRAP 53 $_x$ arb 2, TRAP 22 $_x$ arb 3, TRAP 30 $_x$ arb 3 and TRAP 32 $_x$ arb 4) and the highest (13) was determined using 2 combinations of primers (TRAP 37 $_x$ arb 2 and TRAP 56 $_x$ arb 2). The size of the amplified bands ranged from 200 bp to 2000 bp for all analyzed castor bean genotypes. In total, 360 polymorphic bands were determined, with an average value calculated as 5.45 polymorphic bands per primer. To determine the level of polymorphism, we calculated the PIC value, which determines the level of relatedness between the genotypes analyzed. Based on our results, we found and confirmed that combinations of TRAP markers and random primers are suitable for detecting castor bean polymorphism and can be used to improve ricin quality in further ricin breeding.

Keywords: Castor bean, Dendrogram, PIC, Polymorphism, TRAP markers

INTRODUCTION

Castor bean (*Ricinus communis* L.) is an annual woody plant whose main importance is the production of seeds, which are very rich in ricinoleic acid, which has great industrial uses **(Xu *et al.*, 2019)**. The genetic makeup of castor is made up of 20 somatic chromosomes (2n = 2x = 20) and the total size of the chromosomes ranges from 1.19 to 2.12 µm and the average total length of the diploid set is 32.15 µm **(Vasconcelos *et al.*, 2010)**. Ten chromosomes make up the castor bean genome, the size of which is estimated to be ~320 Mb based on flow cytometry **(Chan *et al.*, 2010)**. Worldwide, castor oil breeding is aimed at obtaining genotypes that will contain high concentrations of castor oil in the seeds, but will also be highly resistant to pathogens and diseases, as well as to drought **(Audi *et al.*, 2005)**. **Wan *et al.* (2019)** report in their work that the castor oil content in seeds ranges from 37 to 60 percent and the best commercially grown varieties contain 48-50% castor oil and the oil yield can be up to 1400 kg of oil per hectare for the best varieties. Functional molecular markers TRAP (target region amplification polymorphism), which are a new technique for studying plant genetic diversity, are a very important tool for breeding new castor bean genotypes with high castor oil concentration **(Simões *et al.*, 2017a)**. Knowledge of genetic diversity and relatedness of genotypes is very important in the selection and breeding of new lines and genotypes of castor, which can then be used for industrial cultivation **(Thatikunta *et al.*, 2016)**. Various DNA techniques have been used worldwide to study the genetic relatedness of ricin, such as: simple sequence repeats (SSRs) **(Dharajiya et al., 2020; Memon *et al.*, 2024)**, expressed sequence tag SSRs (EST-SSRs) **(Dhingani *et al.*, 2012)**, amplified fragment length polymorphism (AFLP) **(Pecina-Quintero *et al.*, 2013)**, single nucleotide polymorphism (SNP) **(Senthilvel *et al.*, 2019; Sathishkumar *et al.*, 2025)**, random amplified polymorphic DNA (RAPD) **(Kim *et al.*, 2021)**, sequence-related amplified polymorphism (SRAP) **(Akhila *et al.*, 2022)**, target region amplifcation polymorphism (TRAP) **(Simões *et al.*, 2017a,b)**, inter-simple sequence repeats (ISSRs) **(Kim *et al.*, 2021)** and start codon targeted markers (SCoT) **(Mahdieh *et al.*, 2023)**.

TRAP technique is a new, modern, highly specific technique for studying plant polymorphism, which is based on the combination of two pairs of primers - specific TRAP primers and arbitrary random primers **(Hu &Vick, 2003)**. This new technique for studying genetic similarity is capable of detecting a high degree of polymorphism, making it a suitable technique for studying the genetic origin of plants and can also be used to identify genes important for agronomic purposes. The combination of TRAP markers and random primers has been used in many studies to determine genetic similarity and relatedness between genotypes and lines, and these markers have also been involved in the analysis associated with castor oil biosynthesis and are therefore very important for the analysis of the association between a functional marker and a candidate gene **(Agarwal *et al.*, 2008)**. Sunflower was the first plant on which the authors tested the utility of TRAP markers and random primers for studying polymorphism and relatedness **(Hu and Vick, 2003)**. These highly specific primers were later used to study polymorphism in many species: fava bean (*Vicia faba* L.) **(Kwon *et al.*, 2010)**, durum wheat (*Triticum durum*) **(Al-Doss *et al.*, 2011)**, cassava (*Manihot esculenta Crantz*) **(Carmo *et al.*, 2015)**, sugarcane (*Saccharum* L. spp. Hybrids) **(Singh *et al.*, 2017)**, *Lactuca georgica* **(Beharav *et al.*, 2018)**, soybean (*Glycine max* L.) **(Kim *et al.*, 2020)**, sweet sorghum (*Sorghum bicolor* L. Moench) **(Khidr *et al.*, 2020)** and Lilium L. **(Hu *et al.*, 2020)**. DNA markers and polymerase chain reaction (PCR) are used worldwide to study polymorphism and relatedness and have been used in practice by several authors, such as **(Sevindik *et al.*, 2023; Farkasová *et al.*, 2023; Carvalho *et al.*, 2024; Chtourou *et al.*, 2024; Malani *et al.*, 2024; Nurtaza *et al.*, 2024; Han *et al.*, 2025; Delgado and Martín, 2025; Tian *et al.*, 2025; Tunç *et al.*, 2025; Máchová *et al.*, 2026; Rottová *et al.*, 2026)**.

The objective of our study was to estimate the genetic distance between 111 castor bean lines, while we used 30 TRAP markers and 6 random primers, and in total we used 66 combinations of these primers for castor bean analyses and to verify the usability of these new markers for castor bean polymorphism analysis. Our analyses are focused only on the study of polymorphism and are not aimed at studying genes related to specific metabolic or agronomic traits.

MATERIAL AND METHODS

Plant material

Together, we analyzed 111 castor lines (*Ricinus communis* L.) (Table 1) that were obtained from Zeainvent Trnava (Slovakia). Zeainvent Trnava is a breeding company that deals with the breeding of cereals, oilseeds and fodder crops. The castor lines analyzed were newly bred and had no stated origin. We grew the plants in a cultivation box on humus soil and total DNA from individual castor lines was isolated from 20-day-old plants using GeneJET Plant Genomic DNA Purification

Mini Kit according to the manufacturer's instructions. Individual castor plants were grown in a cultivation box at a temperature of 26 °C, photoperiod of 12 hours light and 12 hours dark and a humidity of 60 %. The analyzed DNA was isolated from leaves of one plant of each analyzed line. To determine the total concentration of isolated DNA from individual genotypes, we used a UV-VIS spectrophotometer and the final DNA concentration was diluted with deionized water to a concentration of 50 ng/µl. All DNA samples were frozen in a deep freezer at -20 °C.

PCR reaction and statistical analysis

For DNA analysis, we used TRAP markers developed by **Simões *et al*. (2017a)** and combined with decamer random primers (Table 2 and Table 3). The total reaction volume was 15 µl and contained the following components: 1.5 µl DNA (≈8–51 ng/µL), 7.5 µl GoTaq^R G2 Green Master Mix, 1.5 µl primer (combination of TRAP primer and random primer) and 4.5 µl deionized H_2O. Amplification was performed in a programmed Analytik Jena Biometra TOne 96 Well 0.2 ml Gradient PCR Thermal Cycler, with the following cycle: 94 °C for 2 min; 5 cycles at 94 °C for 45 s, 35 °C for 45 s and 72 °C for 1 min; followed by 30 cycles at 94 °C for 45 s, 40 °C for 45 s, 72 °C for 1 min; and a final extension at 72°C for 7 minutes **(Simões *et al.*, 2017a)**. The amplified products were electrophoresed in 1.5% agarose in 1x TBE buffer on Electrophoresis Flat Bed Unit MULTI. Ethidium bromide was used to visualize the amplification products and the gels were documented using a Bio-Rad Gel Imaging Systems. A dendrogram based on hierarchical cluster analysis using the unweighted pairwise group mean (UPGMA) method was constructed from the results with the IBM SPSS Statistics software package. The PIC (polymorphism information content) was calculated using the GENALEX 6.1 software **(Peakall and Smouse, 2012)**. Principal coordinates analysis (PCoA) was created using the PAST 4.06b program.

Table 1 Set of analyzed castor bean lines (*Ricinus communis* L.)

1.	RM-4	41.	RM-66	81.	RM-107
2.	RM-5	42.	RM-67	82.	RM-108
3.	RM-6	43.	RM-68	83.	RM-109
4.	RM-7	44.	RM-69	84.	RM-110
5.	RM-8	45.	RM-70	85.	RM-111
6.	RM-9	46.	RM-71	86.	RM-112
7.	RM-10	47.	RM-72	87.	RM-113
8.	RM-11	48.	RM-73	88.	RM-114
9.	RM-22	49.	RM-74	89.	RM-115
10.	RM-23	50.	RM-75	90.	RM-116
11.	RM-24	51.	RM-76	91.	RM-117
12.	RM-25	52.	RM-77	92.	RM-118
13.	RM-26	53.	RM-78	93.	RM-119
14.	RM-27	54.	RM-79	94.	RM-120
15.	RM-28	55.	RM-80	95.	RM-121
16.	RM-29	56.	RM-81	96.	RM-122
17.	RM-30	57.	RM-82	97.	RM-123
18.	RM-31	58.	RM-83	98.	RM-124
19.	RM-32	59.	RM-84	99.	RM-125
20.	RM-45	60.	RM-85	100.	RM-126
21.	RM-46	61.	RM-86	101.	RM-127
22.	RM-47	62.	RM-87	102.	RM-128
23.	RM-48	63.	RM-88	103.	RM-129
24.	RM-49	64.	RM-89	104.	RM-130
25.	RM-50	65.	RM-90	105.	RM-131
26.	RM-51	66.	RM-91	106.	RM-132
27.	RM-52	67.	RM-92	107.	RM-133
28.	RM-53	68.	RM-93	108.	RM-134
29.	RM-54	69.	RM-94	109.	RM-135
30.	RM-55	70.	RM-95	110.	RM-136
31.	RM-56	71.	RM-96	111.	RM-137
32.	RM-57	72.	RM-98		
33.	RM-58	73.	RM-99		
34.	RM-59	74.	RM-100		
35.	RM-60	75.	RM-101		
36.	RM-61	76.	RM-102		
37.	RM-62	77.	RM-103		
38.	RM-63	78.	RM-104		
39.	RM-64	79.	RM-105		
40.	RM-65	80.	RM-106		

Table 2 List of TRAP markers used for the analysis of 111 castor bean lines (*Ricinus communis* L.) **(Simões *et al*., 2017a)**

	TRAP Primer	Sequence (5'-3')
1.	TRAP 01	CCACATCCAGCACCTTTTG
2.	TRAP 02	TGTGGAGCGTTGAGGATTC
3.	TRAP 03	TGCTCGCAGGCAAAGATAC
4.	TRAP 04	TGTCCCATATTTGCCAACG
5.	TRAP 15	CCGTGATTCTGGTGGTGAG
6.	TRAP 16	TTACAACTGCGGCATCTCC
7.	TRAP 10	CGGGTGGCATCAGTTACAG
8.	TRAP 11	GGCGGATGCTATCTGTGAA
9.	TRAP 22	CACTCGCCTGTTCAGCACT
10.	TRAP 23	AGCAAGCCGCACCTAAGAT
11.	TRAP 24	GTCCAAGCAAAAGCCACCT
12.	TRAP 25	CCACCAATCCAACGCATAG
13.	TRAP 19	AATGCCAGCACCTACACCA
14.	TRAP 30	CTTCTCAGTTGCCCGTTCA
15.	TRAP 31	CCACCAATGAACCAACTGC
16.	TRAP 32	TGCCGACTTCTCCTTTCCT
17.	TRAP 35	CCTCATCATCGTTGCTGCT
18.	TRAP 27	CGAAATCCTCCTGCTCCTC
19.	TRAP 28	GCCACCATCTTCACCACAG
20.	TRAP 37	GCTCACGCACTGGACTCAT
21.	TRAP 39	GCACCCGAAATCTTCCACT
22.	TRAP 40	CCACTCAACACCGTTCCAC
23.	TRAP 44	CGTCCACCCACACTTTCAC
24.	TRAP 46	CCAGTCACCGTTTGTTGCT
25.	TRAP 49	TCCTGTCCAATGCTGAACC
26.	TRAP 51	CCACCGAGAGAGCATACCA
27.	TRAP 52	GTGGCAAATGCTCACAGGT
28.	TRAP 53	TACAACTTCGGGTGGTGGA
29.	TRAP 55	TGATGGAAACCCTTGTGGA
30.	TRAP 56	CTTGTGCCCTACCAACTGC

Table 3 Arbitrary primers used to genotype the castor lines (*Ricinus communis* L.) **(Simões *et al*., 2017a)**

	Arbitrary primers	Nucleotide sequence (3'-5')
1.	arb 1	GACTGCGTACGAATTGAC
2.	arb 2	GACTGCGTACGAATTTGA
3.	arb 3	GACTGCGTACGAATTGCA
4.	arb 4	GACTGCGTACGAATTAATT
5.	arb 5	GACTGCGTACGAATTTGCC
6.	arb 6	GACTGCGTACGAATTGACC

RESULTS AND DISCUSSION

Thirty TRAP markers were used to analyze 111 lines of castor bean, which were combined with 6 random primers. To determine the utility of TRAP markers for studying castor bean polymorphism, the PIC value was calculated (Table 4). The 66 combinations of TRAP markers and random primers together produced 528 DNA fragments that could be evaluated in all 111 castor lines (Table 4, Figure 1). The lowest number of fragments (5) was determined using 7 combinations of TRAP primers and random primers (TRAP 04 $_x$ arb 1, TRAP 19 $_x$ arb 1, TRAP 40 $_x$ arb 2, TRAP 53 $_x$ arb 2, TRAP 22 $_x$ arb 3, TRAP 30 $_x$ arb 3 and TRAP 32 $_x$ arb 4) and the highest number of fragments (13) was obtained by combining 2 pairs of primers (TRAP 37 $_x$ arb 2 and TRAP 56 $_x$ arb 2). Overall, the size of the amplified products ranged from 300 to 2100 bp. Using 66 combinations of TRAP markers and random primers, we detected a total of 528 fragments, of which 360 were detected as polymorphic, with 5.45 polymorphic bands per primer. The percentage of fragment polymorphism ranged from 40% (TRAP 04 $_x$ arb 1) to 100% (TRAP 35 $_x$ arb 2, TRAP 44 $_x$ arb 2, TRAP 30 $_x$ arb 3) with an average value of 68.14%. Based on the obtained results of the analysis of 111 castor bean lines using TRAP markers, we can state the idea that a high degree of polymorphism occurs in the analyzed set of castor bean lines and these genotypes can be further used in the breeding of new, highly resistant castor bean genotypes. Polymorphic information content (PIC) is index used to determine the level of relatedness between the genotypes analyzed. We also used it to determine the degree of relatedness between the analyzed castor lines. After calculating the PIC value, we found that the lowest value was found for the primer combination TRAP 30 $_x$ arb 1 (0.08) and conversely, the highest PIC value (0.34) was determined for the primer combination TRAP 44 $_x$ arb 2 with an average value for all primer combinations of 0.23 (Table 4).From all the results obtained, we constructed a dendrogram, which was constructed from a genetic distance matrix based on the profiles of 30 TRAP markers and 6 random primers using the unweighted pairwise group arithmetic mean (UPGMA) method. The obtained dendrogram was divided into two main clusters, which were further divided into further subclusters (Figure 2 and Figure 3). Since we do not know the origin of the analyzed castor breeders, we cannot accurately analyze their distribution in the dendrogram based on their origin. Cluster 2 contained 3 unique lines (RM-27, RM-8 and RM-4) and cluster 1 was divided into 2 subclusters (1A and 1B). Subcluster 1A contained 1unique lines

(RM-54) of castor and subcluster 1B is divided into 2 subclusters (1BA and 1BB). Subcluster 1BA contained 1 castor lines (RM-67) and subcluster 1BB contains the other 106 lines, which are further divided into smaller subclusters (Figure 2 and Figure 3). Based on our results and the obtained dendrogram, we can conclude that for further breeding of new castor bean genotypes, we can use those genotypes that are the furthest genetically distant from each other. In our case, these are 2 lines from cluster 1 (RM-54 and RM-67) which would be suitable for further crossing with lines from cluster 2 (RM-27, RM-8 and RM-4). We also do not recommend most genotypes from the 1BB cluster for further breeding, because it is easy to see from the dendrogram that they have genetic relationships with each other and therefore it is not appropriate to cross them with each other. By selecting the most genetically distant parental lines from the obtained dendrogram, we can obtain very high variability between the newly obtained lines in further breeding, which may result in obtaining new varieties of castor oil that will, for example, have a high content of castor oil in the seeds. These results and the obtained dendrogram will be used by the breeding company Zeainvent Trnava (Slovakia) for further breeding of new, high-yielding castor bean genotypes. Based on the results we obtained using 66 combinations of TRAP markers and random primers, we can conclude that TRAP markers are suitable for studying polymorphism and relatedness between castor lines. TRAP markers have a high distinguishing ability and we can use the obtained results in the breeding of new castor bean genotypes.

Similar results to those we have observed in our work have been published in other studies by other authors. In contrast, in the present study, we did not perform phenotypic analysis of genotypes, but focused exclusively on polymorphism and relatedness analysis using TRAP markers. Pioneers in this area were **Simões *et al.* (2017a)**, who were the first to develop functional TRAP markers and random primers for ricin genotype analysis, which were subsequently successfully tested in practice. In a follow-up study, **Simões *et al.* (2017b)** analyzed a set of 40 Brazilian ricin lines using a combination of TRAP markers and random primers. They identified a similar number of loci as in our work (ranging from 2 to 15 for the TRAP22+ARB6 combination), but with a lower average value (1.22). In terms of fragment length, the combinations TRAP46+ARB4, TRAP46+ARB5, TRAP46+ARB6, TRAP49+ARB1, TRAP49+ARB2, TRAP51+ARB6 and TRAP52+ARB1 amplified 50 bp regions, while the other combinations produced fragments of larger lengths. The PIC values for the Brazilian lines ranged from 0.03 (TRAP1 x ARB1) to 0.33 (TRAP5 x ARB6) with a mean of 0.24. This corresponds almost exactly to our results, where the PIC values ranged from 0.08 to 0.34 with a mean of 0.23.The consistency with our findings is also confirmed by **Carmo *et al.* (2015)**, who, when studying 46 Cassava genotypes (*Manihot esculenta* Crantz) using 69 primer combinations, determined the average PIC value to be 0.23 (range 0.03–0.38). These results again demonstrate the high utility of TRAP markers in the study of plant polymorphism. In another study, **Vivodík *et al.* (2020)** focused on verifying the relatedness of castor bean genotypes from 12 regions of Tunisia. Using 30 primer combinations, they detected a total of 490 fragments (in the range of 100–1600 bp) in 56 genotypes. The number of amplified regions per combination ranged from 3 (e.g. TRAP 22 x ARB 3) to 13 (TRAP 56 x ARB 2). Of the total, 377 fragments were polymorphic, representing an average of 5.71 per primer. However, the average PIC value in this study (0.770) was significantly higher than in our case, with a maximum of 0.961 for the TRAP 44 x ARB 2 combination. In addition to ricin, the effectiveness of the combination of TRAP markers and random primers has also been confirmed for other taxa, such as the *Saccharum* **(Creste *et al.*, 2010)**, *Diospyros* **(Luo *et al.*, 2013)**, *Dendrobium* **(Feng *et al.*, 2015)** and *Paullinia* **(Da Silva *et al.*, 2016)**.

Table 4 Results obtained from the analysis of 111 castor bean (*Ricinus communis* L.) lines

Combinations	Total fragments	Polymorphic fragments	Polymorphism (%)	PIC
TRAP 01 x arb 1	10	6	60.00	0.31
TRAP 02 x arb 1	8	5	62.50	0.28
TRAP 03 x arb 1	8	6	75.00	0.33
TRAP 04 x arb 1	5	2	40.00	0.11
TRAP 15 x arb 1	11	6	54.54	0.22
TRAP 16 x arb 1	9	6	66.66	0.18
TRAP 10 x arb 1	10	8	80.00	0.23
TRAP 11 x arb 1	8	7	87.50	0.30
TRAP 22 x arb 1	6	3	50.00	0.09
TRAP 23 x arb 1	12	10	83.33	0.33
TRAP 24 x arb 1	11	7	63.63	0.28
TRAP 25 x arb 1	9	7	77.78	0.23
TRAP 19 x arb 1	5	3	60.00	0.12
TRAP 30 x arb 1	10	5	50.00	0.08
TRAP 31 x arb 1	6	5	83.33	0.30
TRAP 32 x arb 2	6	5	83.33	0.25
TRAP 35 x arb 2	7	7	100.00	0.33
TRAP 27 x arb 2	6	5	83.33	0.29
TRAP 28 x arb 2	7	4	57.14	0.15
TRAP 37 x arb 2	13	10	76.92	0.26
TRAP 39 x arb 2	12	6	50.00	0.11
TRAP 40 x arb 2	5	3	60.00	0.22
TRAP 44 x arb 2	8	8	100.00	0.35
TRAP 46 x arb 2	10	6	60.00	0.26
TRAP 49 x arb 2	9	5	55.55	0.25
TRAP 51 x arb 2	7	4	57.14	0.24
TRAP 52 x arb 2	8	7	87.50	0.31
TRAP 53 x arb 2	5	3	60.00	0.26
TRAP 55 x arb 2	10	7	70.00	0.22
TRAP 56 x arb 2	13	8	61.53	0.25
TRAP 01 x arb 3	9	5	55.55	0.23
TRAP 02 x arb 3	8	4	50.00	0.20
TRAP 03 x arb 3	6	5	83.33	0.31
TRAP 04 x arb 3	9	6	66.66	0.28
TRAP 15 x arb 3	8	6	75.00	0.19
TRAP 16 x arb 3	6	3	50.00	0.15
TRAP 10 x arb 3	8	6	75.00	0.21
TRAP 11 x arb 3	6	4	66.66	0.22
TRAP 22 x arb 3	5	4	80.00	0.28
TRAP 23 x arb 3	6	3	50.00	0.15
TRAP 24 x arb 3	7	5	71.43	0.18
TRAP 25 x arb 3	6	4	66.66	0.12
TRAP 19 x arb 3	8	7	87.50	0.15
TRAP 30 x arb 3	5	5	100.00	0.34
TRAP 31 x arb 3	6	3	50.00	0.29
TRAP 32 x arb 4	5	3	60.00	0.30
TRAP 35 x arb 4	9	6	66.66	0.31
TRAP 27 x arb 4	10	8	80.00	0.29
TRAP 28 x arb 4	7	6	85.71	0.27
TRAP 37 x arb 4	6	3	50.00	0.18
TRAP 39 x arb 4	8	5	62.50	0.16
TRAP 40 x arb 4	12	10	83.33	0.23
TRAP 44 x arb 4	8	5	62.50	0.26
TRAP 46 x arb 4	9	4	44.44	0.14
TRAP 49 x arb 4	8	6	75.00	0.17
TRAP 51 x arb 4	7	6	85.71	0.29
TRAP 52 x arb 4	10	5	50.00	0.14
TRAP 53 x arb 4	6	3	50.00	0.16
TRAP 55 x arb 4	7	4	57.14	0.21
TRAP 56 x arb 4	6	5	83.33	0.28
TRAP 01 x arb 5	7	3	42.86	0.10
TRAP 02 x arb 5	9	6	66.66	0.15
TRAP 03 x arb 5	8	6	75.00	0.17
TRAP 04 x arb 6	6	5	83.33	0.34
TRAP 15 x arb 6	12	8	66.66	0.21
TRAP 16 x arb 6	11	11	100.00	0.17
Total	**528**	**360**		**-**
Averages	**8.00**	**5.45**	**68.14**	**0.23**

PIC = polymorphism information content

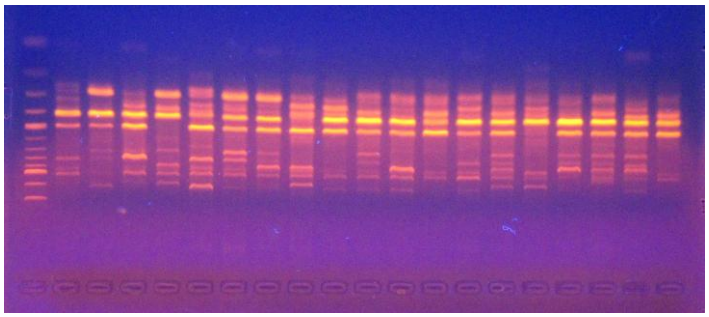

Figure 1 TRAP profile of bands in castor bean lines (TRAP 16 x arb 6). M- Quick-Load ® 2-Log DNA ladder and 1-19 are analyzed lines of castor (Table 1).

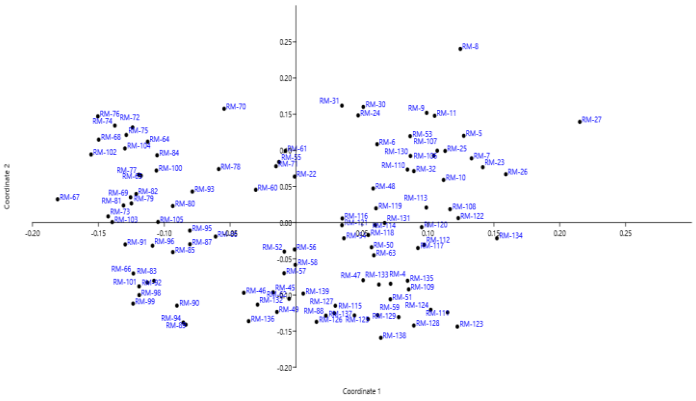

Figure 2 The principal coordinates analysis (PCoA) created from TRAP marker results

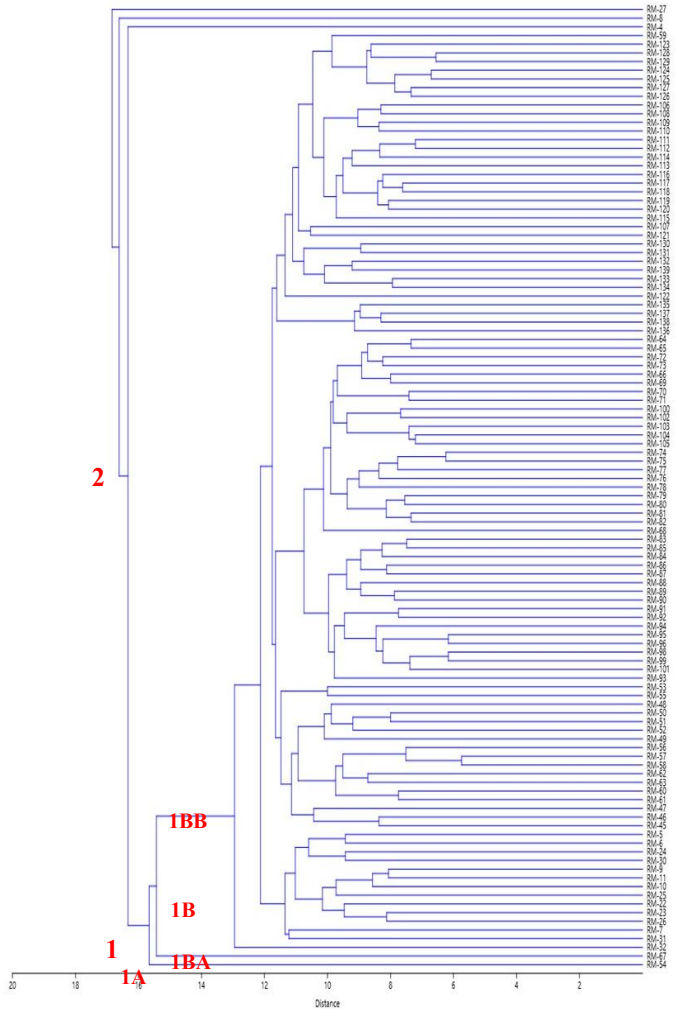

Figure 3 Dendrogram of 111 castor bean lines prepared based on TRAP markers.

CONCLUSION

Total 66 combinations of TRAP markers and random primers together produced 528 DNA fragments that could be scored in all 111 lines castor. The number of amplified fragments varied from 5 (TRAP 04 $_x$ arb 1, TRAP 19 $_x$ arb 1, TRAP 40 $_x$ arb 2, TRAP 53 $_x$ arb 2, TRAP 22 $_x$ arb 3, TRAP 30 $_x$ arb 3 and TRAP 32 $_x$ arb 4) to 13 (TRAP 37 $_x$ arb 2 and TRAP 56 $_x$ arb 2), and the amplicon size ranged from 200 to 2000 bp. Of the 528 amplified bands, 360 were polymorphic, with an average of 5.45 polymorphic bands per primer. A dendrogram was constructed from a genetic distance matrix based on profiles of the 30 TRAP markers and 6 random primers using the unweighted pairgroup method with the arithmetic average (UPGMA). According to analysis, the collection of 111 castor bean lines were clustered into four main clusters. These markers constitute another important tool for genetic improvement programs and studies about genetic variability and population genetics of castor bean.

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