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Assessment of growth and productivity of five peanut cultivars and genetic diversity using RAPD markers

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Abstract

Background: This study was conducted to evaluate the genetic diversity of five peanut cultivars grown under field conditions. A field experiment was conducted using five peanut cultivars (Giza-5, Giza-6, Ismailia-1, Gregory, and R92) in a randomized complete block design with five replications during two following seasons to estimate the performance of five peanut cultivars for vegetative growth, yield, and yield component traits as well as seed quality traits. Twenty RAPD primers were used to identify a unique fingerprint for each of five cultivars.

Results: Giza-6 cultivar surpassed all the tested peanut cultivars in the most vegetative growth traits and yield and its components traits, while the lowest values were observed in Giza-5 cultivar. The dendrogram constructed from RAPD analysis showed that Gregory and Giza-5 were the most distant among five peanut cultivars.

Conclusions: RAPD markers are useful in the detection of genetic diversity of peanut. The availability of genetic diversity is important for the genetic improvement of peanut.

Keywords: Peanut, Yield components traits, Genetic diversity, RAPD-PCR

Background

Peanut (*Arachis hypogea* L.) is an important oil and protein crop, which is grown mainly in semi-arid tropic and sub-tropic areas of 109 countries around the world (Siva et al. 2014). Peanut contains about 50% oil, 25–30% protein, 20% carbohydrates, and 5% fiber and several other micronutrients and minerals (vitamin E, calcium, phosphorus, magnesium, zinc, riboflavin, and potassium) (Settaluri et al. 2012; Toomer 2018). The high-energy value, protein content, and minerals make peanut a rich source of nutrition at a comparatively low price. It has multipurpose uses of each plant part in direct consumption, cooking oil, and a rich source of protein for animal feed (Abou Kheira 2009; Akhtar et al. 2014).

The peanut crop grows in light soil and thrives in improving the characteristics of the newly reclaimed sandy soils which commonly suffer from some constraints such as poor physical properties and nutrients deficiency (Sabate 2003). Recently, peanut crop has been given great attention due to its suitability for growth in the newly reclaimed sandy areas in Egypt. More than half of the national production is consumed directly without oil extraction and the remaining exported because of its high price in the international commodity market. It is successfully cultivated in the newly reclaimed sandy soils which commonly suffer from deficiency or unavailability of most the micronutrients (Ahmad and Rahim 2007). Increasing the growing area of oil seed crops out of the Nile valley is considered as a good way to reduce the competition with other main summer crops (maize, rice, and cotton). This approach is needed in increasing seed and oil production (Shaban et al. 2009).

Despite significant morphological variation in groundnut, the lack of variability at the genetic level is often cited as one of the reasons for little progress in genetic enhancement of the crop (Norden et al. 1982). The genetic diversity of peanut is important for their efficient use in breeding programs. Molecular marker analysis is a powerful tool for grouping of genotypes based on genetic distance data and for the selection of progenitors that might constitute new breeding populations (Westman and Kresovich 1997).

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Different techniques were used to determine genetic variation in plants including isozyme electrophoresis, and morphological traits, but these techniques are not considered as accurate markers due to environmental influences on morphological traits and insufficient polymorphism resulted among closely related cultivars (Matus and Hayes 2002). Molecular marker tools are helpful for the detection of genomic fragments contributing to the genetic diversity of a character and chosen of supreme genotypes. Marker analysis provides exact genotypic data, giving accuracy lacking with phenotypic measurements due to environmental interaction and experimental error (Altinkut et al. 2003).

Molecular markers minimize the time required to develop new genotypes with desirable traits and eliminate the need for chemical analysis phenotypic evaluation in the early generation breeding program. Among the molecular markers, random amplified polymorphic DNA (RAPD) is a rapid method for developing genetic maps and to determine DNA fragments to characterize peanut cultivars (Guo et al. 2005; Azzam et al. 2007; Eskandari et al. 2013). Application of RAPD markers in peanuts aid in determining the markers associated with genes controlling important traits. These molecular techniques assist in the identification of new and various sources of diversity which may aid breeders to choose what genotypes for creating new genetic combinations and to determine which genetic resources should be retained in a collection in order to conserve maximum genetic variation in the gene bank (Al-Saghir and Abdel-Salam 2015).

The objective of this study was to evaluate five peanut cultivars for growth and productivity and estimate the genetic diversity of five cultivars using RAPD markers.

Materials and methods

Field experiment

Five peanut cultivars, namely Giza-5, Giza-6, Ismailia-1, Gregory, and R92 were used. This study was conducted at the Department of Genetics, Faculty of Agriculture, Ain Shams University, Shoubra El-Kheima, Egypt, and Field Crops Research Department of the National Research Centre, Dokki, Giza, Egypt. These five peanut cultivars were grown for two seasons in a randomized complete block design with five replications at the Research and Production Station of the National Research Centre in El-Nubaria, Al-Beheira Governorate, Egypt, to evaluate the performance of these cultivars for vegetative growth, yield, and yield components as well as seed quality traits. Protein content was calculated by $N\% \times 6.25$ according to AOAC (2000). Oil content was estimated using a Soxhlet apparatus, while oil quality with respect to oleic acid and linoleic acid contents was estimated using gas chromatography according to AOCS (2012). Oleic to linoleic acid (O/L) ratio was calculated as the ratio of oleic acid to linoleic acid.

Molecular genetic studies

Genomic DNA extraction

Total genomic DNA was extracted from dry seed of five peanut cultivars according to Yu et al. (2010). Approximately 0.2 g of dried seeds were placed into a 1.5-ml tube with a 200 μl of DNA extraction buffer [10 mM Tris-HCl (pH 7.6), 5 mM EDTA, 0.5% SDS, 0.5% NP-40, 0.5% Tween-20, 5 mg/ml PVP 40, 80 μg/ml proteinase K] and ground with a plastic pestle until a milky white solution or a paste was formed. The tube with homogenate was then incubated in a 55 °C water bath for 20 min for cell lysis and protein digestion. After digestion was completed, a 200 μl of phenol-chloroform-isoamylol (25:24:1, V/V/V) was added to the tube to remove proteinase K. After centrifugation at 9,000×*g* for 5 min, the supernatant was collected (~ 150 μl) in a sterile Eppendorf tube with an equal volume of isopropanol. The mixture was gently mixed and centrifuged at 10,000×*g* for 2 min to precipitate the DNA. The dried DNA pellets were then dissolved in a 150 μl of TE buffer.

RAPD-PCR analysis

PCR reactions were performed according to Williams et al. (1990) using 20 arbitrary 10-mer primers (Table 1). The reaction conditions were optimized and mixtures (25 μl total volume) contained 2 μl of DNA template (25 ng/μl), 2 μl of primer, 0.5 μl of dNTPs (2.5 mM), 2.5 μl of $Mgcl_2$ (2.5 mM), 2.5 μl of 10X buffer, Taq DNA polymerase (1 U/μl), and H2O up to 25 μl. Amplification was carried out in a Primus Thermocycler, which was programmed for 37 cycles as follows: denaturation at 94 °C/2 min (one cycle), annealing at 37 °C/1 min, extension at 72 °C/2 min (35 cycles), final extension at 72 °C/10 min (one cycle), and then kept at 4 °C until use. Agarose gel (1.5%) electrophoresis was used for separating the PCR products. The run was performed at 100 V for about 1 h. The DNA marker used in this study was 1 kb DNA ladder

Table 1 List of used primers and their nucleotide sequences for RAPD-PCR analysis

Primer (OP-)	Sequence (5′ → 3′)	Primer (OP-)	Sequence (5′ → 3′)
A01	CAGGCCCTTC	B01	GTTTCGCTCC
A02	TGCCGAGCTG	B03	CATCCCCCTG
A03	AGTCAGCCAC	B06	TGCTCTGCCC
A05	AGGGGTCTTG	B08	GTCCACACGG
A07	GAAACGGGTC	B10	CTGCTGGGAC
A08	GTGACGTAGG	D04	TCTGGTGAGG
A09	GGGTAACGCC	O11	GACAGGAGGT
A16	AGCCAGCGAA	O14	AGCATGGCTC
A17	GACCGCTTGT	O15	TGGCGTCCTT
A19	CAAACGTCGG	Z12	TCAACGGGAC

which consists of ten different DNA fragments (1000, 900, 800, 700, 600, 500, 400, 300, 200, and 100 bp).

Statistical analysis

The collected data for these four traits from five peanut cultivars were statistically analyzed using analysis of variance (ANOVA) procedure according to Snedecor and Cochran (1969). The differences among means were compared using Duncan's new multiple ranges test (Duncan 1955). The correlation coefficient was used to determine the relationship between oil content and protein content. All fragments resulting from RAPD gels were detected on an UV transilluminator filter and photographed under UV light with Polaroid film 667 and scanned with Bio-Rad video densitometer Model 620 at a wavelength of 577. The gel image was analyzed using the Total lab TL 120 to determine the molecular sizes of the amplified fragments. The amplified fragments were scored as present (1) or absent (0).

Results

Field experiment

Vegetative growth traits

Analysis of variance for most vegetative growth traits revealed significant variations among five peanut cultivars. Their means are summarized in Table 2, except the leaf area index (LAI) trait did not record significant differences in both seasons. The number of branches/plant trait revealed significant variations in the first season only. On the other hand, crop growth rate (CGR) grams/day trait revealed significant variations in the second season only.

These results indicated that the Giza-6 cultivar surpassed all the tested peanut cultivars in most of the vegetative growth traits, except the plant height and leaf area traits. This may be due to the increase in the efficiency of Giza-6 cultivar to photosynthate more metabolites as well as increasing cell division and absorption of more water and minerals from soil. This reflected on increasing the production of more sizable organs.

Yield and yield components traits

Analysis of variance for yield and most yield components traits revealed significant variations among five peanut cultivars (Table 3), except the number of seeds/pod and migration coefficient traits did not record significant differences in both seasons.

Seed quality determination

Protein content (percentage)

Analysis of variance for protein content trait revealed significant variations among five peanut cultivars (Table 4). Means of protein content trait values were significantly varied among five peanut cultivars, which ranged from 28.5 and 28.6% for Giza-5 cultivar to 21.5 and 22.9% for Gregory cultivar in both seasons, respectively. This means that the Giza-5 cultivar is the highest one for protein content.

Oil content (percentage)

Analysis of variance for oil content trait revealed significant variations among five peanut cultivars which are summarized in Table 4.

The means of oil content trait values for all cultivars in the first season were higher than the second season. The mean of oil content trait values was significantly varied among five peanut cultivars, which ranged from 53.3 and 52.8% for Gregory cultivar to 45.2 and 45.1% for Giza-5 cultivar in the two seasons, respectively. This result showed that Gregory cultivar contained the highest oil content.

Fatty acids composition (percentage)

Oil quality trait is determined by its fatty acids composition. Analysis of variance for fatty acids composition revealed some significant variations among five peanut cultivars which summarized in Table 4. There were no significant variations among five peanut cultivars for myristic acid, arachidic acid, and TS/TU ratio in the second season and palmitoleic acid in the first season.

These results confirmed that oleic acid was the major component (40.2–57.7%) of total fatty acids, followed by linoleic acid (22.6–32.9%). The total saturated fatty acids ranged from 18.9 for Giza-5 cultivar to 21.4 for Giza-6 cultivar, while the total unsaturated fatty acids ranged from 68.1 for R92 cultivar to 84.59 for Gregory cultivar. It was clear that the highest values of oleic acid content (57.7, 56.3%) and O/L ratio (2.6, 2.1) were recorded by the Gregory cultivar in the two seasons, although this cultivar gave the lowest linoleic acid content (22.6, 27.1 %) in the two seasons. Gregory cultivar was more suitable for oil quality as compared with the other cultivars. By contrast, Giza-6 cultivar showed a high level of linoleic acid (32.2, 32.9%), and Gregory cultivar was more suitable for cooking.

RAPD-PCR analysis

DNA isolated from five peanut cultivars was tested against 20 primers. Out of these primers, nine primers did not reveal any polymorphism, while 11 primers revealed a polymorphism with five peanut cultivars as shown in Fig. 1.

Eleven RAPD primers OP-A02, OPA-05, OP-A16, OP-A17, OP-A19, OP-B03, OP-B06, OP-B10, OP-O14, OP-O15, and OP-Z12 produced different banding patterns for all cultivars. While nine primers OP-A01, OP-A03, OP-A07, OP-A08, OP-A09, OP-B01, OP-B08, OP-D04, and OP-O11 detected no polymorphism, although they did successfully amplify a range of monomorphic bands.

Table 2 The vegetative growth traits of five peanut cultivars during two seasons at 100 days for five replicates at each season

Season									
Traits cultivars	Plant height (cm)	Number of branches/plant	Number of leaves/plant	Dry weight of leaves/plant (g)	Dry weight of branches/plant(g)	Dry weight of plant (g)	Leaf area/plant (cm^2)	Leaf area index	Crop growth rate (g/day)
First season									
Giza-5	41.4^{d}	7.2^{bc}	161.2^{d}	18.4^{b}	16.3^{d}	71.6^{c}	131.6^{d}	0.12^{a}	5.3^{ab}
Giza-6	46.8^{ab}	8.6^{a}	212.2^{a}	23.2^{a}	25.1^{a}	87.6^{a}	157.9^{a}	0.13^{a}	5.9^{a}
Gregory	42.6^{cd}	7.6^{abc}	175.0^{c}	19.7^{ab}	19.2^{c}	85.8^{a}	143.1^{b}	0.12^{a}	5.7^{a}
Ismailia-1	43.6^{c}	6.8^{c}	172.2^{c}	18.9^{b}	18.1^{cd}	77.9^{b}	139.6^{bc}	0.12^{a}	5.4^{ab}
R92	47.6^{a}	8.0^{ab}	204.4^{b}	22.2^{a}	22.9^{b}	86.3^{a}	159.6^{a}	0.13^{a}	5.8^{a}
Second season									
Giza-5	41.6^{d}	7.6^{a}	183.6^{e}	19.0^{c}	20.7^{c}	75.6^{c}	143.1^{d}	0.12^{a}	5.8^{c}
Giza-6	47.4^{a}	8.8^{a}	240.8^{a}	25.1^{a}	27.1^{a}	92.3^{a}	169.5^{b}	0.13^{a}	6.6^{a}
Gregory	43.2^{c}	7.8^{a}	220.6^{c}	23.1^{b}	25.9^{ab}	88.4^{ab}	158.3^{c}	0.13^{a}	6.0^{bc}
Ismailia-1	46.8^{ab}	7.4^{a}	195.4^{d}	20.1^{c}	23.8^{bc}	79.9^{b}	154.8^{c}	0.13^{a}	6.5^{a}
R92	47.8^{a}	8.4^{a}	235.2^{b}	23.7^{b}	26.6^{ab}	91.8^{a}	175.6^{a}	0.13^{a}	6.3^{ab}

Means with the same letter(s) in the column are not significantly different by Duncan's new multiple range test ($P < 0.05$)

Table 3 Yield and yield components traits for five peanut cultivars during two seasons for five replicates at each season

Season														
Traits cultivars	No. of pods/plant	D. W. of pods/ plant (g)	Pods yield/ fed (kg)	Pods yield/ fed (ard.)	No. of seeds/pod	No. of seeds/plant	D.W. of seeds/ plant (g)	D. W. of 100 seeds (g)	Seeds yield/ fed (kg)	Shelling (%)	Harvest index	Crop index	Migration coefficient	Oil yield/ fed (kg)
First season														
Giza-5	37.2^{c}	58.8^{d}	1402.5^{d}	18.7^{d}	2 .0^{a}	46.8^{d}	41.0^{c}	80.1^{bc}	976.2^{d}	69.7^{b}	0.28^{d}	0.32^{c}	0.4^{a}	440.6^{d}
Giza-6	44.6^{a}	88.5^{a}	1819.5^{a}	24.3^{a}	2.6^{a}	86.4^{a}	58.8^{a}	90.1^{a}	1228.8^{a}	67.9^{c}	0.37^{a}	0.44^{a}	0.5^{a}	602.7 ^{a}
Gregory	40.4^{b}	70.1^{c}	1585.4bc	21.14^{bc}	2.2^{a}	67.8^{c}	47.6^{bc}	76.3^{d}	1120.5^{b}	66.4^{b}	0.30^{c}	0.40^{ab}	0.5^{a}	596.8^{a}
Ismailia-1	38.6^{bc}	80.8^{b}	1545.1c	20.6^{c}	2.2^{a}	72.6^{c}	52.8^{ab}	81.3^{bc}	1029.9^{c}	69.0^{a}	0.29^{cd}	0.39^{b}	0.4^{a}	501.8^{bc}
R92	39.8^{bc}	74.0^{c}	1687.5b	22.5^{b}	2.4^{a}	82.4^{b}	55.4^{a}	82.4^{bc}	1123.9^{b}	71.4^{a}	0.34^{b}	0.43^{a}	0.5^{a}	549.6^{b}
Second season														
Giza-5	39.6^{c}	57.8^{d}	1497.6^{cd}	19.9^{c}	2.3^{ab}	47.2^{d}	42.3^{d}	78.6^{bc}	1022.9^{bc}	69.7^{b}	0.30^{c}	0.34^{c}	0.4^{a}	461.3^{b}
Giza-6	51.2^{a}	90.0^{a}	1812.0^{a}	24. 6^{a}	2.6^{a}	85.8^{a}	59.4^{a}	91.4^{a}	1273.1^{a}	66.0^{bc}	0.47^{a}	0.49^{a}	0.5^{a}	625.6^{a}
Gregory	49.6^{a}	76.0^{c}	1627.5^{b}	21.7^{b}	2.3^{ab}	73.8^{c}	47.1^{d}	74.0^{cb}	1131 .1^{b}	62.0^{c}	0.34^{c}	0.44^{ab}	0.5^{a}	598.4^{a}
Ismailia-1	43.2^{b}	79.8^{c}	1530.3^{c}	20.4^{bc}	2.4^{a}	71.4^{c}	55.6^{c}	81.4^{b}	1048. 4^{c}	68.3^{b}	0.37^{bc}	0.45^{ab}	0.5^{a}	512.0^{ab}
R92	44.4^{b}	84.8^{b}	1695.0^{b}	22.6^{b}	2.5^{a}	80.4^{b}	57.9^{ab}	83.6^{b}	1126.1^{b}	73.2^{a}	0.41^{ab}	0.48^{a}	0.5^{a}	543.9^{ab}

Means with the same letter(s) in the column are not significantly different by Duncan's new multiple range test ($P < 0.05$)

Table 4 Protein, oil contents, and fatty acid composition (percentage) for five peanut cultivars at two seasons for five replicates at each season

Season	Saturated fatty acid								Unsaturated fatty acid					
Traits cultivars	Protein content (%)	oil content (%)	Myristic acid	Palmitic acid	Stearic acid	Arachidic acid	Behenic acid	Lignoceric acid	Palmitoleic acid	Oleic acid	Linoleic acid	Linolenic acid	O/L ratio	TS/TU ratio
First season														
Giza-5	28.5^{a}	45.2^{c}	0.03^{b}	10.4^{b}	3.4^{b}	1.7^{bc}	2.1^{c}	1.5^{a}	0.09^{a}	42.2^{cd}	30.4^{ab}	0.08^{bc}	1.4^{b}	0.26^{abc}
Giza-6	25.7^{b}	49.1^{b}	0.06^{a}	11.6^{a}	4.4^{a}	1.8^{ab}	2.4^{b}	1.0^{b}	0.07^{b}	47.0^{b}	32.2^{a}	0.11^{b}	1.5^{b}	0.27^{abc}
Gregory	21.5^{d}	53.3^{a}	0.03^{b}	9.3^{c}	5.0^{a}	1.9^{a}	2.9^{a}	1.2^{ab}	0.09^{a}	57.7^{a}	22.6^{c}	0.19^{a}	2.6^{a}	0.24^{d}
Ismailia	24.2^{b}	48.8^{b}	0.03^{b}	11.4^{a}	3.4^{b}	1.4^{d}	2.7^{a}	1.1^{ab}	0.07^{b}	44.3^{c}	27.6^{b}	0.08^{bc}	1.6^{b}	0.28^{ab}
R92	23.4^{c}	48.9^{b}	0.02^{b}	11.4^{a}	3.1^{b}	1.6^{bc}	2.7^{a}	1.4^{a}	0.09^{a}	40.2^{d}	27.8^{b}	0.04^{d}	1.4^{b}	0.30^{a}
Second season														
Giza-5	28.6^{a}	45.1^{c}	0.03^{a}	10.3^{c}	2.7^{c}	1.5^{a}	2.7^{c}	2.1^{ab}	0.08^{bc}	42.6^{abc}	30.4^{ab}	0.18^{a}	1.4^{b}	0.26^{a}
Giza-6	24.2^{c}	49.0^{b}	0.04^{a}	11.5^{b}	3.3^{a}	1.5^{a}	3.3^{b}	1.8^{b}	0.09^{b}	46.3^{ab}	32.9^{a}	0.10^{b}	1.4^{b}	0.27^{a}
Gregory	22.9^{bc}	52.8^{a}	0.03^{a}	10.0^{c}	3.4^{a}	1.5^{a}	4.1^{a}	2.3^{a}	1.05^{a}	56.3^{a}	27.1^{c}	0.13^{b}	2.1^{a}	0.26^{a}
Ismailia	24.3^{b}	48.7^{b}	0.02^{a}	10.5^{b}	3.1^{ab}	1.5^{a}	3.6^{ab}	1.8^{b}	0.08^{bc}	45.1^{ab}	30.4^{ab}	0.11^{b}	1.5^{b}	0.27^{a}
R92	23.6^{b}	48.5^{b}	0.03^{a}	11.1^{a}	2.8^{c}	1.4^{a}	3.0^{b}	1.6^{c}	0.05^{d}	41.1^{c}	30.1^{ab}	0.07^{c}	1.4^{b}	0.28^{a}

Means with the same letter(s) in the column are not significantly different by Duncan's new multiple range test ($P < 0.05$)

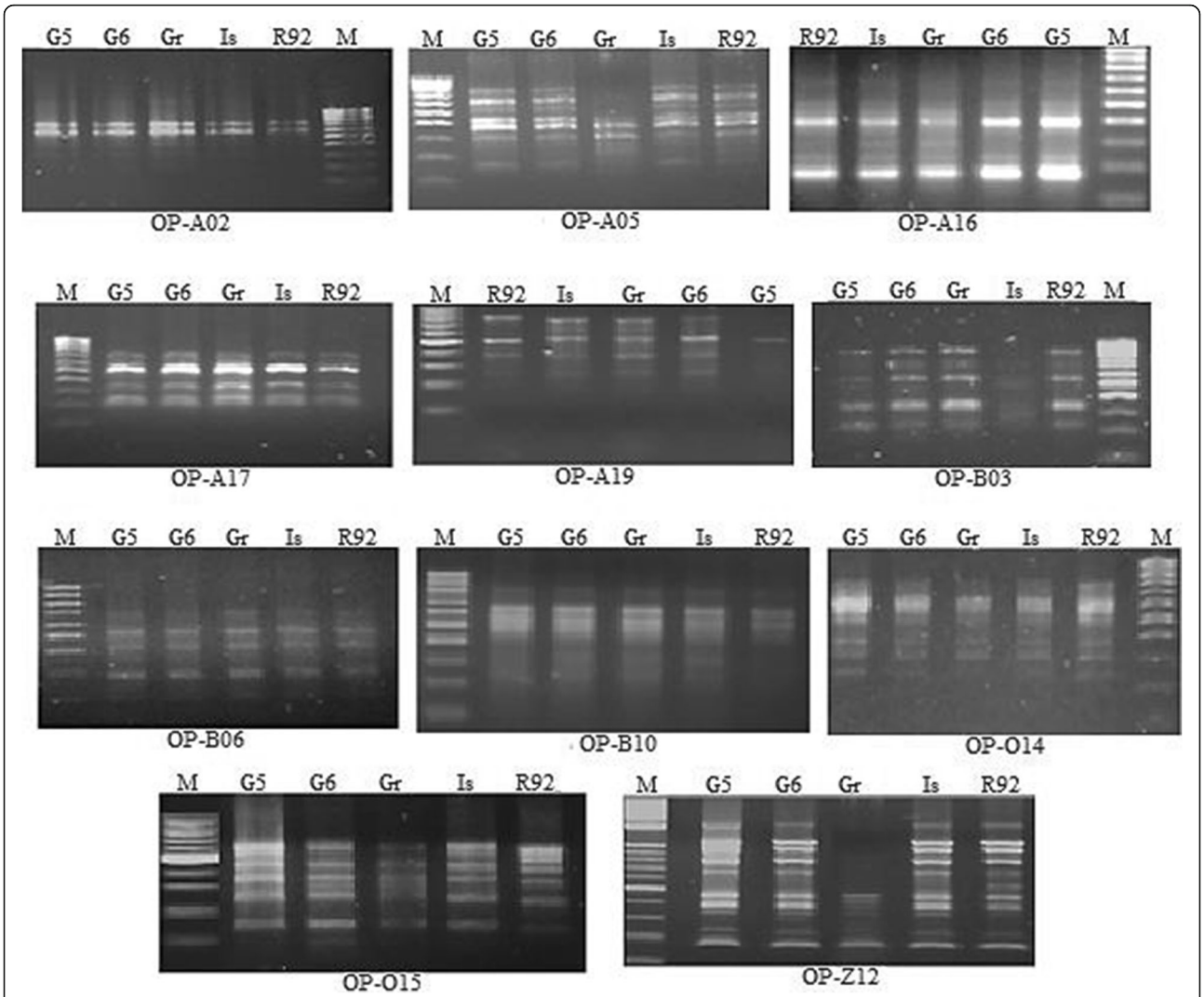

Fig. 1 RAPD-PCR fragments of the 11 primers with five peanut cultivars. M marker, G5 Giza-5, G6 Giza-6, Gr Gregory, Is Ismailia-1 and R92

The eleven primers produced a total number of 82 fragments, with an average of seven fragments per primer ranging from five fragments with primers OP-A02, OP-A17, OP-B06, and OP-B10 to17 fragments with OP-Z12 primer. However, these fragments were not presented in all cultivars. The polymorphism percentages were ranged from 20.00% to 87.50%with an average of 54.80%. The OP-A19 primer showed the highest level of polymorphism 87.50%, while the OP-A17 and OP-B06 primers showed the lowest level 20.00% (Table 5).

The genetic similarity matrix values (Table 6) based on RAPD markers were ranged from 0.91 between Ismailia-1 and R92 cultivars to 0.71 between Giza-5 and Gregory cultivars.

Based on the data of RAPD markers, the constructed dendrogram divided these cultivars into two main clusters (Fig. 2); the first one included only Giza-5 cultivar. The other cluster was divided into two main sub-clusters; the first one included only the Gregory cultivar, while the other one was divided into two sub-sub clusters, the first one included only the Giza-6 cultivar, while the second one included Ismailia1 and R92 cultivars.

According to RAPD analysis, these results indicated that the most closely related cultivars were Ismailia1 and R92, which were located in the same sub-sub cluster, while the most dissimilar cultivars were Giza-5 and Gregory, which located in the two different main clusters.

Discussion

Field experiment

From the previous results (Tables 2 and 3), it is clear that Giza-6 cultivar surpassed all the tested peanut cultivars in vegetative growth, yield, and its most components traits. The obtained results were in a good line with those obtained by El-Saady et al. (2014), Meena et al. (2014), Mahrous et al. (2015), and Sarkees (2015).

Table 5 The number of total amplified fragments, polymorphic fragments, polymorphism percentage, and specific markers using RAPD analysis

Primer no.	Primer name	TAF	PF	MF	UF	P %	Band no.	MS (bp)	Cultivars
1	OP-A02	5	3	2	0	60.00	–	–	–
2	OP-A05	9	5	4	1	55.56	8	241	Gregory
3	OP-A16	6	2	4	0	33.33	–	–	–
4	OP-A17	5	1	4	0	20.00	–	–	–
5	OP-A19	8	7	1	1	87.50	5	593	R92
6	OP-B03	7	4	3	2	57.14	4	451	Ismailia-1
							6	235	Ismailia-1
7	OP-B06	5	1	4	0	20.00	–	–	–
8	OP-B10	5	2	3	0	40.00	–	–	–
9	OP-O14	6	2	4	0	33.33	–	–	–
10	OP-O15	9	4	5	2	44.44	6	293	Giza-6
							9	105	Giza-6
11	OP-Z12	17	9	8	4	52.94	2	1464	Giza-6
							8	588	R92
							11	413	Gregory Giza-6
							15	265	
Total		82	40	42	10				

TAF total amplified fragments, *PF* polymorphic fragments, *MF* monomorphic fragments, *UF* unique fragments, *MS* molecular size, *P%* polymorphism percentage

On the other hand, these results were not agreed with Abdalla et al. (2009) who found that Giza-5 cultivar was superior to Giza-6 cultivar in plant height, number of branches/plant, number of pods/plant, and a 100-pod weight trait.

Also, these results are confirmed by El-Saady et al. (2014) who showed that Giza-6 cultivar surpassed Giza-5 cultivar in most of the yield and its components traits. The superiority of Giza-6 cultivars may be due to its high ability to grow under El-Nubaria conditions. This means that the Giza-6 cultivar was the best one for the production of seeds and pods yield kilograms/fed. Moreover, differences among five peanut cultivars may be due to the differences in their genetic make-up and their response to the environmental conditions.

Table 6 The genetic similarity matrix of five peanut cultivars based on RAPD markers

Cultivars	Giza-5	Giza-6	Gregory	Ismailia-1	R92
Giza-5	1.00				
Giza-6	0.81	1.00			
Gregory	0.71	0.82	1.00		
Ismailia-1	0.76	0.86	0.85	1.00	
R92	0.79	0.84	0.81	0.91	1.00

Seed quality determination

The previous results (Table 4) are confirmed by Newase et al. (1990) and Tomar et al. (1995) who found differences among four peanut cultivars in protein content of seed was significant. Abdalla et al. (2009) found that Giza-5 cultivar was superior to Giza-6 cultivar in protein content trait. Mahrous et al. (2015) found that Giza-5 cultivar was superior to Gregory in protein content trait. As discussed by Migawer et al. (2001), El-Saady et al. (2014) found significant differences among some tested cultivars concerning seed oil and protein contents traits. Similar results were obtained by Mahrous et al. (2015) who found that Gregory cultivar showed the highest seed oil content (52.22 %). Also, Gulluoglu et al. (2016) found that oil content of peanut cultivars varied between 46.96 and 51.55 % based on dry weight of seeds and the highest oil content value (51.55%) was reported in Georgia Green cultivar, while the lowest one (46.96%) was noticed in Flower-22 cultivar.

Both the seed oil and protein content traits are often influenced by the environment. Correlation coefficients between protein and oil contents (– 0.013) revealed a negative significant correlation. These results are similar to Selvaraj et al. (2009) and Sarvamangala et al. (2011) who found that the relationship between oil content was inverse to that of protein content. Moreover, Chun et al. (2014) showed a

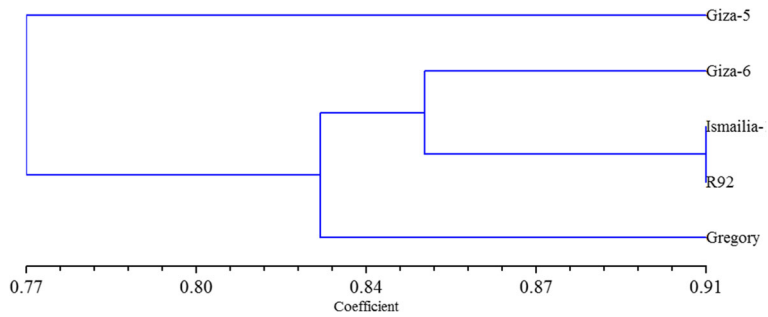

Fig. 2 Dendrogramtree of genetic distances among five peanut cultivars based on RAPD markers

significant negative correlation of the oil content with the protein content which was found in the tested cultivars.

Oils with a higher percentage of unsaturated fatty acids can be heated to high temperatures without smoking, leading to faster cooking time and absorption of less oil. High O/L in peanut is favored over low O/L because it confers health benefits and oil stability (Miller et al. 1987; Wilson et al. 2006).

Many investigators found significant variations among peanut cultivars in growth, productivity, and quality due to the variation in their genetics and their interaction with the environmental condition (Meena et al. 2014; Mahrous et al. 2015; Sarkees 2015).

RAPD-PCR analysis

This study described the genetic diversity using RAPD markers. The highest number of markers was observed in Giza-6, which recorded four unique markers at 293 bp and 105 bp of primer OP-O15 and 1464 bp and 265 of primer OP- Z12. Also Gregory has two unique markers, bands at 241 bp of primer OP-A05 and bands at 413 bp of primer OP-Z12. Ismailia-1 has two unique markers at 451 and 235 bp of primer OP-B03. R92 has two unique markers at 593 bp of primer OP-A19 and 588 bp of primer OP-Z12. On the other hand, Giza-5 did not reveal any unique marker. These results confirmed that the selected RAPD markers are dispersed in the peanut genome and may be valuable to study the genetic diversity of five peanut cultivars. These results agreed with Guo et al. (2005) and Lang and Hang (2007) who reported that the distinctive RAPD patterns generated from peanut cultivars could be used as genomic fingerprint to establish the identity of a given genotype. Similarly, Al-Saghir and Abdel-Salam (2015) observed that the technique of RAPD could be used to detect the genetic diversity in peanut and give a successful fingerprinting of peanut using these markers. Lom and Rao (2015) indicated the efficacy of RAPD markers for detecting the genetic variability in the wild *Musa acuminate*.

The low level of polymorphism might be due to the low molecular diversity among the peanut cultivars, and this is because cultivated peanut has a narrow genetic base which originated from a single and recent polyploidization event. This was in agreement with the previous findings as low level of polymorphism among cultivated groundnut (Hopkins et al. 1999; Herselman 2003; Moretzsohn et al. 2004; Mace et al. 2006). On the other hand, in our study, the polymorphism percentage (54.80%) increased compared to Dwivedi et al. (2001) who found about 18.74% of polymorphism among selected peanut cultivars using the same RAPD technique.

The genetic similarity matrix results are in harmony with those obtained by Dwivedi et al. (2001) who reported that the genetic similarity values among selected groundnut germplasm were ranged from 59.0% to 98.8 % with an average of 86.2%. NaguibNemat et al. (2011) reported that the genetic similarity among peanut cultivars ranged from 0.68 to 0.92 with an average of 0.8.

Conclusions

This study showed that Giza-6 cultivar surpassed all tested peanut cultivars in the most traits, while the lowest values were observed in Giza-5 cultivar. Gregory cultivar was more suitable for oil quality as compared with the other cultivars. The results showed that RAPD markers were distributed in the peanut genome and may be useful to detect the genetic diversity of these five peanut cultivars. The results obtained in this study may assist peanut cultivation and in peanut breeding programs.

Abbreviations

Bp: Base pair; DNA: Deoxyribonucleic acid; dNTP: Deoxy nucleoside triphosphate; EDTA: Ethylene diamine tetra acetic acid; HI: Harvest index; Mb: Mega base pairs = 1,000,000 bp; $MgCl_2$: Magnesium chloride; MS: Molecular size; MT: Marker type; O/L: Oleic to linoleic acid ratio; PCR: Polymerase chain reaction; PVP: PolyvinylpyrrolidoneRAPDRandom amplified polymorphic DNA; TAF: Total amplified fragmentsTBETris-borate-EDTA; TE: Tris-EDTA

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GS performed and analyzed this study and was a major contributor in writing the manuscript. MA was a contributor in writing and processing the manuscript. AR was a contributor in analyzing the data. All authors read and approved the final manuscript."

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