Evaluation of Some Olive Genotypes Cultivated in Iraq using Molecular Markers

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ABSTRACT

The olive tree belonged to family *Oleaceae* is an ancient woody crop with more than 46,000 years of cultural history, and is mainly distributed around the Mediterranean area. Molecular markers have become increasingly important in plant molecular biology in its relationship to plant breeding, plant systematic and plant evolution. Eight olive cultivars grown in Iraq with 18 RAPD and 8 SSR markers have been developed during current study. The number of amplification fragments differed from cultivar to the other one with RAPD markers. The highest number was recorded to cultivar Manzenllo by 157 fragments followed by cultivars Qaysi and Labeeb by the same number 138 fragments. Also, cultivar Qaysi and Nepali showed the same amplification fragments (132 fragments) followed by cultivars Khdier and Sorani by 126 and 129 fragments, in respect. The lowest cultivar was Shami by 114 fragments. The data indicated that from 1068 amplification fragments there were 28 unique fragments were recorded to cultivars Labeeb (7 unique fragments), Shami and Manzenllo (5), Shami (3 unique fragments), Sorani and Nepali (2 unique fragments) and finally Dahkan (1 unique fragments). While for the eight microsatellites (SSR) a total of 136 specific amplification fragments were detected among the cultivars, 71 fragments were polymorphic and 65 were monomorphic. Form the previous data we can concluded that based on the RAPD and SSR markers it could be helpful for calculate the genetic diversity between the eight-olive growing in Iraq.

Key words: Olive, RAPD, SSR, Genetic diversity.

INTRODUCTION

Olive tree (*Olea europaea* L.) is valued for both its beauty and fruit. The olive was native to Asia Minor and spread from Iran, Syria and Palestine to the rest of the Mediterranean basin 6,000 years ago, it is among the oldest known cultivated trees in the world being grown before the written language was invented (Loukas and Krimbas, 1983). The olive tree (2n = 2x = 46) belonged to family Oleaceae is an ancient woody crop with more than 46,000 years of cultural history, and is mainly distributed around the Mediterranean basin. In recent decades, olive trees have been introduced and cultivated in the United States, Australia, South Africa, China and in other countries (Breton *et al*., 2006). Olives are in high demand in Iraq. In 2011, Iraq imported more than 10,000 metric tons of olives from Turkey for a value of $15.3 million ($1.50 per kilogram). This quantity represents a 35 percent increase as opposed to 2009 when only 7,500 metric tons were imported, which nearly doubled the quantity imported in 2008 (4,900 metric tons). To study the diversity in plants or genotypes molecular markers will asissified for this purpose (Bari *et al*., 2003). The request for higher food safety has raised, it was reported that RAPD-PCR technique has the capability to study the genetic variability between different species and between the individuals belong the same species; in addition, they are considered as genetic markers which have been used extensively in many different applications and in different plant species because of its simplicity (Salimath *et al*., 1995 and Ulanovsky, 2002). Moreover, RAPDs have been successfully used to study the genetic relationships among various date palm accessions and cultivars from Egypt, Morocco, Tunisia, Iraq and King Saudi Arabia (Adawy *et al.,* 2002 and Motawei *et al*., 2003).

Molecular markers have become increasingly important in plant molecular biology in its relationship to plant breeding, plant systematic and plant evolution. Most molecular marker systems are presently based on PCR technology, molecular markers of various types have helped to increase understanding and elucidate new aspects of plant evaluation (Tanksley *et al*., 1998). RAPD analysis could distinguish clearly between olive cultivars (Perri *et al*., 1999) which provides a new method for cultivar identification in olive. The use of RAPD primers (Williams *et al*., 1990) has given environment independent markers. At present, olive cultivars are identified using biochemical and molecular markers such as isoenzymes (Lumaret *et al*., 2004), RAPDs (Martins-Lopes *et al*., 2008), AFLPs (Montemurro *et al*., 2008), SSRs, ISSRs (Souza et al., 2012), SCARs (Pafundo *et al*., 2007), and SNPs (Belaj *et al*., 2012). Molecular markers are valuable tools that can be used to study olive genetics, and have been applied to the identification of cultivars. Therefore, the present study aims to study the genetic polymorphism between cultivars from based on RAPD

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and SSR markers and detect the genetic distance and similarity.

MATERIALS AND METHODS

Eight olive cultivars grown in Iraq (*Olea europea* L.) namely Sorani, Dahkan, Shami, Qaysi, Nepali, Khdier, Labeeb and Manzenllo, were obtained from the ministry of Agriculture of Iraq used for this experiment. Ten mature trees were used to represent each cultivar to study the molecular analysis.

Random Amplified Polymorphic DNAs analysis

18 RAPD-PCR has been developed, in which DNA is amplified by the polymerase chain reaction (PCR) using arbitrary short (10 nucleotides) primers (Williams *et al.*, 1990). RAPD has become an important technique for population genetic studies since the amplified products provide random representation of both coding and non-coding regions across the whole genome.

DNA Extraction

DNA was extracted following (Murray and Thompson, 1980) with minor amendments as follows.

The Polymerase Chain Reaction (PCR) conditions

RAPD analyze was carried out using ten oligonucleotide primers (Table 1) that were selected from the Operon Kit (Operon Technologies Inc., Alabameda, CA). The polymerase chain reaction mixture (25µl) consisted of 13µl master mix (Promega) *Taq* DNA polymerase; 2µl of genomic DNA,2µl primer, 8µl deionized water. PCR amplification was performed in a Biometra T1 gradient thermal cycler for 35 cycles after initial denaturation for 5min at 94°C. Each cycle consisted of denaturation at 94°C for 1min; annealing at 36°C for 1min; extension at 72°C for 2min and final extension at 72°C for 5min (Williames *et al.*, 1990).

Simple Sequence Repeats (SSRs)

Simple Sequence Repeats (SSRs), also known as microsatellites, are repeating sequences of nucleotides, such as (AC)n. They have been found in abundance on most eukaryotic chromosomes and are often highly polymorphic (Rafalski and Tingey, 1993). Eight SSR markers described (Kong *et al.,* 2000) were used for genotyping assays. Primers names, sequences and corresponding annealing temperatures are listed in Table (2).

PCR conditions

The PCR reaction mixture (25µl) consisted of the following: 2µl DNA genome, 13 µl master mix (Promega), 6µl deionized weter, 2µl each of forward and reverse primers The PCR profile starts with 95°C for 5 min followed by 35 cycles of denaturation at 94°C for 1 min, annealing at (50-61°C vary with primer) for 1 min extension at 72°C for 2 min. A final extension 72°C for 5 min was included.

Computer programmes and statistical analysis

Fragments scored as present/absent. Fragment scoring and lane matching performed automatically on digital images of the gels, using Phoretix 1D advanced Version 4.00 (Phoretix International, Newcastle upon Tyne, UK).

All but the faintest bands scored, where necessary scores and matches corrected manually. Clustering methods and similarity coefficients were tested usingthe procedures SIMQUAL, SAHN, and TREE from the program NTSYSpc version 2.10 (Applied Biostatistics, Setauket, New York, USA).

Table 1. Primers name and their oligonucleotide sequences used in the current study

Primer code	Primer name	Sequence
1	OPA-05	5`-AGG GGTCTTG-3`
2	OPA-10	5`-GTGATC GCAG-3`
3	OPA-15	5`-GTCGTA GCGG-3`
4	OPB-07	5`-GAAACGGGTG -3`
5	OPB-17	5`-AGG GAACGAG-3`
6	OPC-05	5`-GATGACCGCC -3`
7	OPC-06	5`-GAT GACCGCC-3`
8	OPC-09	5`-TGG ACCGGT G-3`
9	OPC-12	5`-TGTCATCCCC-3`
10	OPN-04	5`- GACCGACCCA -3`
11	OPN-10	5`-ACAACTGGGG -3`
12	OPN-13	5`-AGCGTCACTC - 3`
13	OPM-05	5`- GGGAACGTGT -3`
14	OPD-05	5`- TGAGCGGACA -3`
15	OPD-07	5`-AGC GCCATT G-3`
16	OPG-12	5`-CAGCTCACGA -3`
17	OPQ-12	5`- AGTAGGGCAC -3`
18	OPQ-14	5`-GGACGCTTCA -3

Table 2. Sequences of the SSR loci and annealing temperature for PCR reaction

Locus	Repeat motif	Primer sequence (5'–3')	*Annealing Tem.*
OeUA-DCA04	$(GA)_{16}$	F: CTTAACTTTGTGCTTCTCCATATCC R: AGTGACAAAAGCAAAAGACTAAAG C	55°C
OeUA-DCA05	$(GA)_{15}$	F: AACAAATCCCATACGAACTGCC R: CGTGTTGCTGTGAAGAAAATCG	50°C
OeUA-DCA07	$(AG)_{19}$	F: GG CATAAAACATAGAGTGCTGGGG R: AGGGTAGTCCAACTGCTAATAGACG	60°C
OeUA-DCA08	$(GA)_{18}$	F: ACAATTCAACCTCACCCCCATACCC R: TCACGTCAACTGTGCCACTGAACTG	55°C
OeUA-DCA10	$(TA)_{14}$ $(GA)_{17}$	F: CGT GAC CAC CTA AAT CCG CCC C R: CTG TCC AGA GCT AAA GGT TTC G	50°C
OeUA-DCA13	$(GA)_{15}$	F: GATCAGATTAATGAAGATTTG G R: AACTGAACCTGTGTATCTTGCATC C	55°C
OeUA-DCA16	$(GT)_{13}(GA)_{29}$	F: TTAGGTGGGATTCTGTAGATGGTTG R: TTTTAGGTGAGTTCATAGAATTAGC	50°C
OeUA-DCA17	$(GT)_9(AT)_7$AGA TA$(GA)_{38}$	F: GATCAAATTCTACCAAAAATATA R: TAATTTTTGGCACGTAGTATTGG	50°C

The clustering methods UPGMA, WPGMA, Complete-link, and Single-link were applied in all possible combinations with the similarity coefficients Dice, Jaccard and simple matching (Rohlf, 2000) describes clustering methods and similarity coefficients.

RERSULTS AND DISCUSSION

Randomly Amplified Polymorphic DNA (RAPD-PCR)

The results of primer OPA-05 are illustrated in Figure (1) and Table (3). The total fragments were ٩٢ amplification products at the fragment lengths ranged between ١٥٠ bp. to 1800 bp. 20 fragments were polymorphic and 72 were monomorphic. The percentage of the polymorphism was 27.73%. While, the results of primer OPA-10 showed total 23 amplification fragments products at the fragment lengths ranged between 300 bp. to 800 bp. fifteen fragments were polymorphic and 8 were monomorphic. The percentage of the polymorphism was 65.21%. The results of primer OPA-15 are illustrated in Figure (1) and Table (3) detect 85 amplification fragments ranged between ٣٥٠ bp. to 1500 bp. Twenty-one fragments were polymorphic and 64 were monomorphic. The percentage of the polymorphism was 24.70%. The results of primer OPB-07 are illustrated in Figure (1) and Table (3). The total fragments were 85 amplification products at the fragment lengths ranged between ٢٥٠ bp., to 1800 bp. In a total of 68 fragments 44 fragments were polymorphic and 24 were monomorphic. The percentage of the polymorphism was 64.70%.

The results of primer OPB-17 are presented Table (3) showed 79 fragments ranged between 200 bp. to 1800 bp. 39 fragments were polymorphic and 40 fragments were monomorphic. The percentage of the polymorphism was 49.36%. The primer OPC-05 detect 46 fragments in lengths ranged between 400 bp. to 850 bp. From 46 total fragments 30 fragments were polymorphic and 16 fragments were monomorphic. The percentage of the polymorphism was 65.21%. Concerning to primer OPC-06 in Figure (1) and Table (3) produced 50 fragments in lengths ranged between 350 bp. to 1200 bp. From 50 total fragments 26 fragments were polymorphic and 24 fragments were monomorphic. The percentage of the polymorphism was 52.0%. The primer OPC-09 showed 50 fragments products at the fragment lengths ranged between 200 bp. to 1200 bp. From 50 total fragments 26 fragments were polymorphic and 24 fragments were monomorphic. The percentage of the polymorphism was 52.0%. while, the primer OPC-12 detect 74 amplification products at the fragment lengths ranged between 200 bp. to 1800 bp. From 74 total fragments 50 fragments were polymorphic and 24 fragments were monomorphic. The percentage of the polymorphism was 67.56%.

For the primer OPN-04 are illustrated in Figure (1) showed 27 amplification fragments ranged between 400 bp. to 1500 bp. From 27 total fragments 11 fragments were polymorphic and 16 fragments were monomorphic. The percentage of the polymorphism was 40.74%. The results of primer OPN-10 are illustrated in Figure (1) and Table (3). The total fragments were 41 amplification products at the fragment lengths ranged between 400 bp. to 1500 bp. From 41 total fragments 25 fragments were polymorphic and 16 fragments were monomorphic. The percentage of the polymorphism was 60.97%. The present primer showed high number of amplification fragments ranged from 10 to 14 fragments. The results of primer OPN-13 are illustrated in Figure (1) and Table (3).

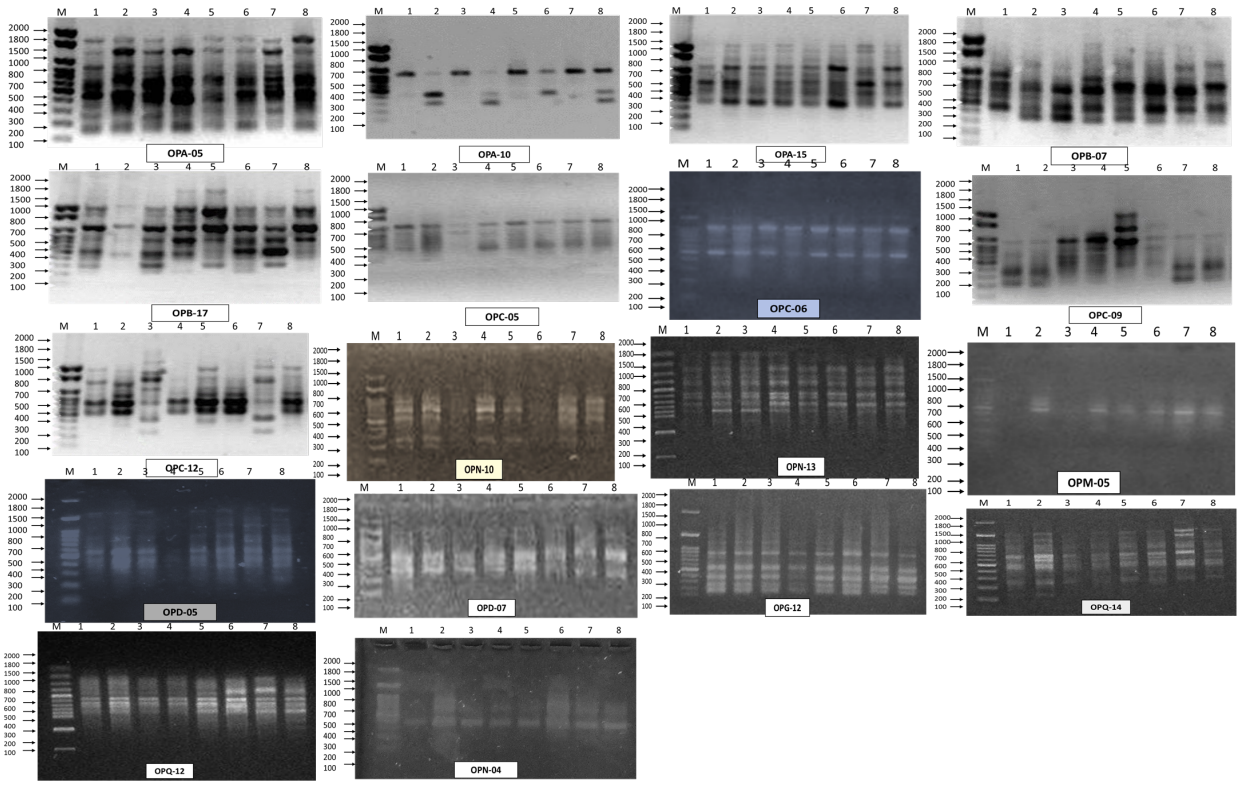

Figure 1. DNA polymorphism of eight olive cultivars growing in Iraqi with different RAPD primers

Table 3. Markers, cultivars, and amplification fragments of eight olive cultivars growing in Iraqi using RAPD-PCR

	Cultivars and amplification fragments							
Markers	**Sorani**	**Dahkan**	**Shami**	**Qaysi**	**Nepali**	**Khdier**	**Labeeb**	**Manzenllo**
OPA-05	12	11	10	14	11	12	12	10
OPA-10	2	3	1	3	4	3	3	4
OPA-15	6	10	10	11	13	10	11	14
OPB-07	8	7	8	10	8	11	8	8
OPB-17	10	7	10	13	10	10	7	12
OPC-05	6	7	4	5	6	6	5	7
OPC-06	6	8	6	5	5	5	7	8
OPC-09	7	6	6	8	7	5	6	5
OPC-12	8	10	9	7	11	7	12	10
OPN-04	3	5	2	3	2	4	4	4
OPN-10	7	6	2	6	5	1	6	6
OPN-13	11	12	13	14	10	10	12	14
OPM-05	0	4	1	3	2	2	4	3
OPD-05	7	8	7	3	6	8	10	12
OPD-07	5	3	3	5	3	2	2	5
OPG-12	12	10	9	7	9	9	8	11
OPQ-12	8	8	5	5	8	8	8	10
OPQ-14	11	13	8	10	12	13	13	14
Total	129	138	114	132	132	126	138	157

The total fragments were 96 amplification products at the fragment lengths ranged between 400 bp. to 1800 bp. From 96 total fragments 64 fragments were polymorphic and 32 fragments were monomorphic. The percentage of the polymorphism was 68.08%.

The results of primer OPM-05 are illustrated in Figure 16 and Table 6. The total fragments were 19 amplification products at the fragment lengths ranged between 600 bp. to 1500 bp. From 19 total fragments 11 fragments were polymorphic and 8 fragments were monomorphic. The percentage of the polymorphism was 57.89%.

The results of primer OPD-05 are illustrated in Figure (1) and Table (6). The total fragments were 61 amplification products at the fragment lengths ranged between 200 bp. to 1600 bp. From 61 total fragments 37 fragments were polymorphic and 21 fragments were monomorphic. The percentage of the polymorphism was 60.65%.

The results of primer OPD-07 are illustrated in Figure (18) and Table (3). The total fragments were 61 amplification products at the fragment lengths ranged between 350 bp. to 1000 bp. From 28 total fragments 12 fragments were polymorphic and 16 fragments were monomorphic. The percentage of the polymorphism was 42.85%.

The results of primer OPG-12 are illustrated in Figure (1) and Table (3). The total fragments were 75 amplification products at the fragment lengths ranged between 200 bp. to 1500 bp. From 75 total fragments 35 fragments were polymorphic and 40 fragments were monomorphic. The percentage of the polymorphism was 46.66%. The results of primer OPQ-12 are illustrated in Figure (1) and Table (6). The total fragments were 60 amplification products at the fragment lengths ranged between 300 bp. to 1500 bp. From 60 total fragments 20 fragments were polymorphic and 40 fragments were monomorphic. The percentage of the polymorphism was 33.33%. The results of primer OPQ-14 are illustrated in Figure (1) and Table (3). The total fragments were 94 amplification products at the fragment lengths ranged between 200 bp. to 2000 bp. From 94 total fragments 46 fragments were polymorphic and 48 fragments were monomorphic. The percentage of the polymorphism was 48.93%.

Results in Table (4) showed that thirteen detected bands were showed for cultivars Dahkan, Khdier and Labeeb also results indicated that cultivars Qaysi, Sorani and Nepali detected the descending number of amplification fragments (10, 11 and 12 fragments, in respect), while, cultivars Manzenllo showed the highest number of DNA fragments (14 fragments). Two unique fragment was detected for cultivars Labeeb and Manzenllo (1500 bp).

Table 4. Number of Scorable bands, Polymorphic, Monomorphic bands Percentage of polymorphism of eight olive cultivars growing in Iraqi using RAPD-PCR

Markers	Scorable bands	Polymorphic bands	Monomorphic bands	Percentage of polymorphism (%)
OPA-05	92	20	72	21.73 %
OPA-10	23	15	8	65.21 %
OPA-15	85	21	64	24.70 %
OPB-07	68	44	24	64.70 %
OPB-17	79	39	40	49.36%
OPC-05	46	30	16	65.21%
OPC-06	50	26	24	52.0%
OPC-09	50	26	24	52.0%
OPC-12	74	50	24	67.56%
OPN-04	27	11	16	40.74%
OPN-10	41	25	16	60.97%
OPN-13	96	64	32	68.08%
OPM-05	19	11	8	57.89%
OPD-05	61	37	24	60.65%
OPD-07	28	12	16	42.85%
OPG-12	75	35	40	46.66%
OPQ-12	60	20	40	33.33%
OPQ-14	94	46	48	48.93%
Total	1068	532	536	54.86%

In a total of 1068 amplification fragments 532 fragments were polymorphic and 536 fragments were monomorphic with 54.86% genetic polymorphism. The number of amplification fragments differed from cultivar to the other one. The highest number was recorded to cultivar Manzenllo by 157 fragments followed by cultivars Qaysi and Labeeb by the same number 138 fragments. Also, cultivar Qaysi and Nepali showed the same amplification fragments (132 fragments) followed by cultivars Khdier and Sorani by 126 and 129 fragments, in respect. The lowest cultivar was Shami by 114 fragments (Figure 2). The data indicated that from 1068 amplification fragments there were 28 unique fragments were recorded to cultivars Labeeb (7 unique fragments), Shami and Manzenllo (5), Shami (3 unique fragments), Sorani and Nepali (2 unique fragments) and finally Dahkan (one unique fragments) (Table 5).

Simple Sequence Repeat (SSR)

Eight microsatellites (SSR) were used in the current study. A total of 136 specific amplification fragments were detected among the cultivars as shown in Table 7. 71 fragments were polymorphic and 65 were monomorphic. The total scored fragments ranged from one to four fragments and the molecular weight ranged from 100 to 250 pb. The results of primer DcA-04 are illustrated in Figure (2) and Table (6). Results in showed that four detected fragments from 19 fragments in a total were showed for cultivars Qaysi also results indicated that other cultivars detected two amplification fragments, while, cultivars Nepali showed three fragments. The genetic polymorphism was 42.85% between thee tested cultivars using the previous marker (Table 6). The results of primer DcA-05 showed that from 10 fragments two detected fragments were showed for cultivars Dahkan and Nepali also results indicated that other cultivars detected one amplification fragment. The genetic polymorphism was 46.66% between thee tested cultivars using DcA-05 marker (Table 6). For the marker DcA-07 Results showed that from 19 fragments three detected fragments were showed for cultivars Dahkan, Labeeb and Manzenllo; also, results indicated that other cultivars detected two amplification fragments. The genetic polymorphism was 100% between thee tested cultivars (Table 6). The results of primer DcA-08 showed 19 amplification fragments: three detected fragments were showed for cultivars Sorani, Dahkan, 3, Qaysi, Khdier and Manzenllo. While cultivars Nepali and Labeeb showed two fragments, in respect. The lowest genetic polymorphism was 15.78% between thee tested cultivars using DcA-08 marker (Table 6).

The results of primer DcA-10 showed 11 amplification fragments and cultivar Qaysi showed the highest number (3 fragments), while he others showed one amplification fragment except cultivars Nepali gave one fragment. The moderated genetic polymorphism was 50.0% between thee tested cultivars using the previous marker (Table 6).

Table 5. Number of Scorable bands, Unique band and sample number of eight olive cultivars growing in Iraqi using RAPD-PCR

Markers	Number of scorable bands	Unique band	Sample number
OPA-05	92	3	300 (1), 800 (3,4) bp
OPA-10	23	0	--
OPA-15	85	2	450 (1), 550 (7)bp
OPB-07	68	2	980 (7, 8) bp
OPB-17	79	3	1800 (4, 7, 8) bp
OPC-05	46	١	٧٥٠ (٣) bp
OPC-06	50	1	850 (4) bp
OPC-09	50	3	800, 1200 (5), 800 (6) bp
OPC-12	74	4	250, 1500, 1700 (3), 250 (7) bp
OPN-04	27	1	1500 (2) bp
OPN-10	41	0	--
OPN-13	96	0	--
OPM-05	19	1	250 (8) bp
OPD-05	61	2	1500 (7, 8) bp
OPD-07	28	0	--
OPG-12	75	3	850 (6, 7), 900 (8) bp
OPQ-12	60	0	--
OPQ-14	94	2	1500 (7, 8) bp
Total	1068	28	--

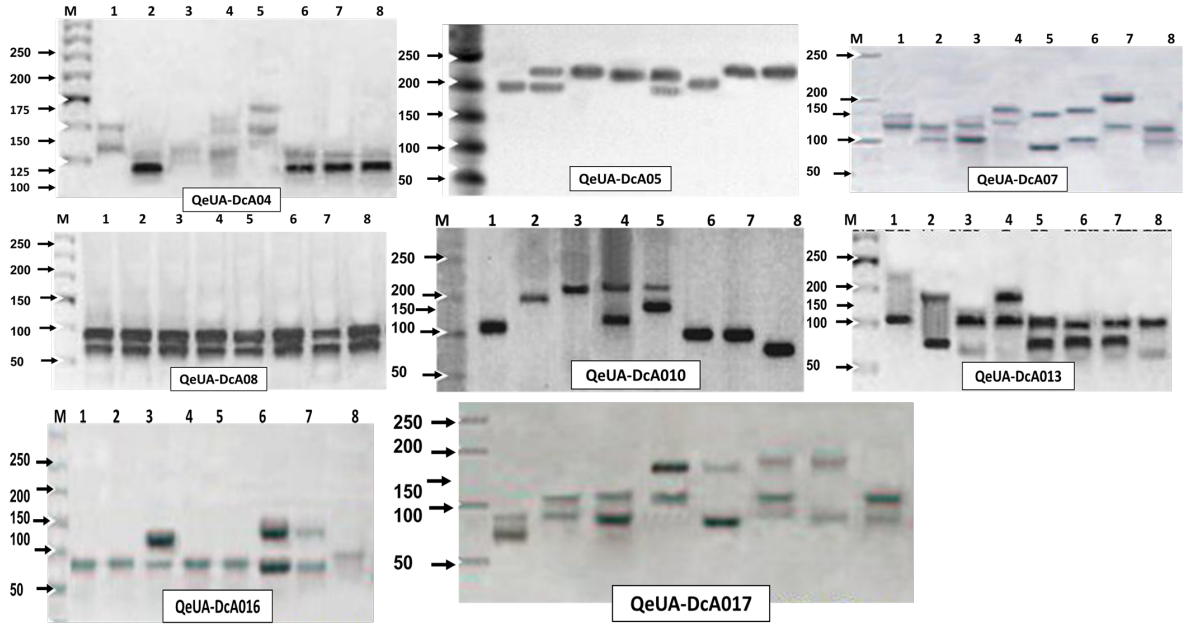

Figure 2. DNA polymorphism of eight olive cultivars growing in Iraqi using eight simple sequence repeat primers

Table 6. Markers, cultivars, amplification fragments of eight olive cultivars growing in Iraqi using SSR markers, (1) Sorani, (2) Dahkan, (3) Shami, (4) Qaysi, (5) Nepali, (6) Khdier, (7) Labeeb and (8) Manzenllo

	Cultivars and amplification allele							
	Sorani	**Dahkan**	**Shami**	**Qaysi**	**Nepali**	**Khdier**	**Labeeb**	**Manzenllo**
DCA-04	2	2	2	4	3	2	2	2
DCA-05	1	2	1	1	2	1	1	1
DCA-07	2	2	3	2	2	2	3	3
DCA-08	3	3	3	3	2	3	2	3
DCA-10	1	1	1	3	2	1	1	1
DCA-13	2	2	3	3	2	2	3	3
DCA-16	1	1	3	1	1	3	2	2
DCA-17	2	2	3	2	2	3	2	3
Total	14	15	19	19	16	17	16	18

The results of primer DcA-13 are illustrated in Figure (2) and Table (6). Results in Table 6 showed that from 20 fragments three detected fragments were showed for cultivars Shami, Qaysi and Labeeb also results indicated that other cultivars detected two amplification fragments. The results of primer DcA-16 are illustrated in Figure (2) and Table (6). Results in Table 6 showed that from 14 fragments three detected fragments were showed for cultivar Khdier. The genetic polymorphism was 55.5% between thee tested cultivars using DcA-16 marker (Table 6). Finally, the results of primer DcA-17 are illustrated in Figure 2 and Table 6. Results in Table 6 showed that from 19 fragments three detected fragments were showed for cultivars Shami, Khdier and Manzenllo also results indicated that other cultivars detected two amplification fragment and the genetic polymorphism was 52.63% (Table 6). The highest amplification fragments were recorded to cultivars Shami and Qaysi by 19 fragments, followed by cultivar Manzenllo by 18 fragments. While the lowest one was cultivar Sorani by 14 fragments followed by cultivars Dahkan, Nepali and Labeeb by 15 and 16 fragments, respectively (Figure 2). Results in Table (7) detected eight specific alleles between the tested olive cultivars. Nepali detected one specific allele with 175 bp, also, Qaysi detected one specific allele with 200 bp, while Labeeb detected two specific alleles with 150 and 250 bp, Sorani detected one specific allele with 250 bp. Shami and Khdier detected one specific allele with 150 bp. Cluster analysis in Figure (3) showed the genetic diversity and similarity between the eight Iraqi olive collected from different location in Iraq. The cluster

divided to two main group with 82% similarity, the first group (A) includes just one sup-cluster (A1) with genetic similarity 88% (Shami and Khdier). While the second main group (B) includes two sub-cluster B1 (90%) with (Qaysi and Nepali); B2 (85%) divided to another sub sub-cluster (B3=93%) AND (B4=85%). The last one, B4 have Sorani in separate one and Dahkan, Labeeb and Manzenllo together with 93% genetic distance.

RAPD and SSR techniques revealed a high level of polymorphism. In RAPDs the percentage of polymorphism observed was 54.86. Slightly higher values were registered for OPN-13 (68.08%) for THESE results are agreed with those (Korbin *et al*., 2002 and Galvan *et al*., 2003). These values were at the same level as the polymorphism reported within 12 clones of 3 Italian olive tree cultivars by AFLP (Sensi *et al*., 2003). The high polymorphism for RAPDs observed in comparison with other studies (Korbin *et al*., 2002 and Galvan *et al*., 2003). The high level of polymorphism observed in this study was agree with (Gemas *et al*., 2004, Lopes *et al*., 2004, Fabbri *et al*., 1995, Belaj *et al*., 2003 and Terzopoulos *et al*., 2005) indicated that olive is a highly polymorphic species. The high diversity found between olive cultivars is probably due to a diverse germplasm origin that presumably results from crosses between wild and cultivated olives resulting in new cultivars in di□erent parts of the Mediterranean, and low breeding pressures (Belaj *et al*., 2003).

Table 7. Number of scorable bands, Polymorphic, Monomorphic bands Percentage of polymorphism of eight olive cultivars growing in Iraqi using SSR markers

Markers	Scorable bands	Polymorphic bands	Monomorphic bands	Percentage of polymorphism (%)	Unique allele	Sample number
DCA-04	14	6	8	42.85 %	1	175 (5) bp
DCA-05	15	7	8	46.66 %	0	--
DCA-07	19	19	0	100.0 %	2	200 (4), 250 (7) bp
DCA-08	19	3	16	15.78 %	0	--
DCA-10	16	8	8	50.0%	0	--
DCA-13	16	8	8	50.0%	1	250 (1) bp
DCA-16	18	10	8	55.5%	3	150 (3, 6, 7) bp
DCA-17	19	10	9	52.63%	1	75 (1) bp
Total	136	71	65	51.20%	8	--

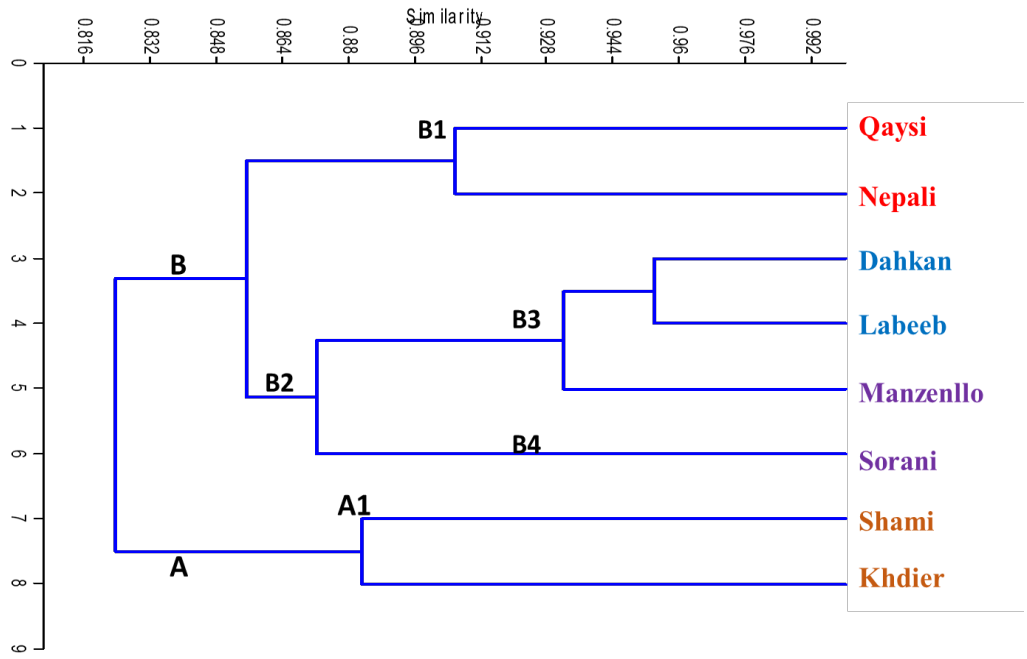

Figure 3. Genetic distance and similarity of eight olive cultivars growing in Iraqi based on RAPD and SSR markers

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الملخص العربي

تقييم بعض التراكيب الوراثية من الزيتون المنزرع فى العراق باستخدام المعلمات الجزيئية

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أجريت هذة الدراسة بكلية الزراعة سابا باشا جامعة الاسكندرية خلال العام ٢٠١٦ بغرض تقييم بعض أصناف الزيتون المنزرع بالعراق باستخدام المعلمات الوراثية العشوائية والمتخصصة. إستخدم فى هذة الدراسة ثمانية انواع من الزيتون بالاضافة الى ١٨ معلم وراثى عشوائى وثمانية معلم أخر متخصص يتبع تكرار التراكيب البسيطة وتم إستخلاص المادة الوراثية من الأصناف موضع الدراسة. أوضحت النتائج بالنسبة لتكنيك القطع العشوائية انة إجمالى القطع المتحصل عليها كان ١٠٦٨ من بينهم ٢٨ قطعة متخصصة وأعطى الصنف منزينولو ١٥٧ قطعة تالها الصنف قيسى ولبيب ١٣٨ قطعة بينما كان أقل الأصناف هو الشامى ب ١١٤ قطعة. بينما عند إستخدام تكرار التراكيب الوراثية اعطى اجمالى ١٣٦ اليل متخصص وكانت منهم ٧١ أليل متعدد الشكل الظاهرى و٦٥ أليل لا يوجد بينهم تعدد فى الشكل المظهرى. وبناء على تلك النتائج أمكن تقسيم تلك الاصناف الى مجموعات مختلفة من خلالها يمكن البدء فى برامج التربية المختلفة.