Genetic Diversity of Some Tomato *Lycopersicon esculentum* Mill Varieties in Iraq Using Random Amplified Polymorphic DNA (RAPD) Markers

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Abstract

This study was conducted to evaluate the genetic diversity among 19 tomato varieties (determinate and indeterminate) cultivated in Iraq using polymerase chain reaction based DNA markers (PCR based DNA markers) ; Random Amplified Polymorphic DNA (RAPDs) .To achieve PCR reactions ,total genomic DNA was isolated from fresh leaves (2 weeks old).The average yields of DNA were in the range of 100-295 ng/µl with a purity ranging between 1.8-1.9.

RAPDs amplifications were performed for varieties fingerprinting by testing 27 Operon primers. DNA polymorphisms among varieties were scored within detectable amplified fragments (their numbers and molecular weight) after agarose gel electrophoresis and staining by ethidium bromide. These 27 primers produced 442 of main bands,out of which 312 were polymorphic bands (70.5%) and 70 were monomorphic (15.8%) across all tested varieties.

Each selected primer produced between 60 bands (OPA-14) to 290 bands (OPD-13). DNA amplification products ranged in their size from 250 bp (OPA-01, OPU-14, OPX-15,OPX-19,OPT-08) to 2755 bp (OPX-18). The highest number of polymorphic bands (21 bands) was produced by primer OPU-03 while, the lowest number of polymorphic bands (3 band) was produced by both primers OPA-14 and OPB-17.

The primers varied in their capacity in producing polymorphic amplified profiles among studied tomato varieties which individually reflected variety specific DNA profiles (fingerprints). The most important primers for this purpose were primers that produced more variety specific DNA profiles, such as OPD-13, OPT-08, OPW-04, OPA-04, OPA-15, OPB-18, OPU-03, OPC-09.

The highest value of discrimination among varieties in this study was obtained by primer OPU-03 while the lowest discrimination value was produced by both primers OPA-14 and OPB-17.The primer efficiency ranged from 0.13 in (primer OPC-09) to 0.02 in (primer OPB-17).

The lowest genetic distance was (0.2294) between varieties Oula and Shadylady, while, the highest genetic distance was (0.9459) between varieties Fotton and Special pack. Cluster analysis (phylogenetic tree) by unweighted pair-group method of arithmetic means (UPGMA) based dendrogram revealed that they were two main genetic groups (major clusters).

The first small major clusters included four (4 varieties) while the second large major cluster included (15 varieties). The overall analysis of the results show that RAPDs markers are powerful tool in fingerprinting and revealing the genetic relationships among tomato varieties.The relationship among varieties was not concern to their morphological characters and geographical origins .

Key words: Genetic diversity; *Lycopersicon esculentum*; RAPD markers; genetic distance.

الخلاصة

أجريت الدراسة الحالية لتقدير التنوع الوراثي ل 19 صنف من اصناف الطماطة (المحدودة وغير المحـــدودة النمـــو) المستزرعة في العراق باستخدام مؤشرات الدنا (DNA Markers) المعتمدة على تفاعلات البلمرة المتسلســـلة Polymerase Chain Reaction(PCR) وهي مؤشرات التفاعل التضاعفي العشوائي المتعدد الأشكال لسلسلة الدنا Random Amplified Polymorphic DNA (RAPDs) . لتنفيذ تفاعلات ال PCR تم عزل دنا المجين (Genomic DNA) من أوراق الطماطة الفتية (بعمر أسبوعين) و الحصول على كمية من الدنا تراوحت بين 100-295 نانوغرام/مايكروليتر وبنقاوة تراوحت بين 1.8-1.9.اجريت تفاعلات ال RAPDs للحصول على بصمة للاصناف المدروسة باختبار 27 بادئ والكشف عن التباينات بين القطع المتضاعفة لكل صنف (أعدادها وأحجامها الجزيئية) بعد تصبيغ و ترحيل نواتج التضاعف للعينات على هلام الأكآروز.

أعطى 27 بادئ نواتج تضاعف متباينة بين الأصناف المدروسة بلغت 312 حزمة متباينة Polymorphic bands(70.5%) و 70 حزمة وحيدة الشكل monomorphic bands (15.8%) من أصل 442 حزمة رئيسية main band. تباينـــت اعـــداد الحـــزم

المتضاعفة ما بين 60 حزمة (OPA-14) إلى 290 حزمة (OPD-13). وتراوح حجم النواتج المتضاعفة للدنا بين 250 زوج قاعدي OPX-19,OPT-08 (OPA-01, OPU-14, OPX-15) إلى 2755 زوج قاعدي (OPX-18). أكبر عدد من الحزم المتباينة (21 حزمة) نتجت بواسطة البادئ OPU-03 في حين ظهر أن أقل عدد من الحزم المتباينة (3 حزم) كان للبادئين OPB-17وOPA-14 . وقد اختلفت البادئات في قدرتها على إيجاد أنواع مختلفة من النسق بين الأصناف المدروسة polymorphic RAPD profile أمكن من خلالها إيجاد البصمة الوراثية للأصناف قيد الدراسة من الطماطة والتي تعكس ملامح متنوعة فردية محددة الحمض النووي (البصمات). والبادئات الأكثر أهمية لهذا الغرض هي تلك التي تنتج المزيد من DNA profiles متنوعة محددة، مثل OPD-13، OPT-08، OPW-04، OPA-04, OPA-15, OPB-18 , OPU-03 و-OPC. 09 ووجد في هذه الدراسة بأن البادئ OPU-03 كان له أعلى قيمة في القدرة على التمييز ،بينما اظهر كل من البادئين (OPB-17 وOPA-14) اقل قيمة كما و تراوحت كفاءة البوادئ بين 0.13 للبادئ (OPC-09) إلى 0.02 في البادئ (OPB- 17).وكان أقل بعد وراثي هو (0.2294) بين صنفي الطماطة Oula و Shadylady ، بينما اعلى بعد وراثي هو (0.9459) بين الصنفين Fotton وSpecial pack وكشف التحليل التجميعي (شجرة النشوء والتطور) من خلال طريقة unweighted pair-group method of arithmetic means (UPGMA) أي المعتمد على شجرة العلاقات التطورية إلى اثنين من المجموعات الوراثية الرئيسية.الاولى صغيرة ضمت 4 اصناف والثانية كبيرة ضمت 15 صنف .أظهر التحليل العام للنتائج أن كل من المؤشر الوراثي المعروف بال RAPD يعد اداة قوية لتحديد البصمة والكشف عن العلاقات الوراثية بين أصناف الطماطة . لم تظهر العلاقات بين الاصناف المدروسة ارتباطا بالموقع الجغرافي والصفات المظهرية.

الكلمات الدالة : التنوع الوراثي الطماطة , مؤشرات التفاعل التضاعفي العشوائي المتعدد الأشكال لسلسلة الدنا RAPD ,البعدالوراثي

Introduction

Tomato(*Lycopersicon esculentum*) is one of the most important vegetable crop grown throughout world it is second important vegetable after potato because of its wider adaptability,high yielding potential and multipurpose uses (Sekhar *et al.*,2008).It is amember of family Solanaceae and significant vegetable crop of special economic importance in the horticultural industry worldwide (He*etal*.,2003;Wang *et al.*,2005).

Tomato also used as model plant species to study the physiology and biochemistry of seed development, germination and dormancy (Suhartanto, 2002),therefore, tomato is an excellent tool to improve knowledge on horticultural crops (Taylor, 1986; Kinet and Peet, 1997).

Tomato breeding projects have improved characteristics such as disease resistance ,fruit abscission, soluble solids, fruit size, texture, flavor, pigmentation, and storage ability,thus, the

An improvement in yield and quality in self pollinated crops like tomato is normally achieved by selecting the genotypes with desirable character combinations existing in nature or by hybridization .The success of hybridization programme depends upon selection of suitable parents of diverse origin(Sekhar *et al.*, 2008) .

A mong molecular markers, RAPDs were the first PCR-based molecular markers to be employed in genetic variation analyses (Welsh and McClelland,1990). The standard RAPD utilizes short synthetic oligonucleotides decamer (10 bases long) (Miesfeld, 1999). This technique has been widely used in diversity studies because, in addition to its low cost, it allows polymorphism to bedetected in a simple and rapid manner (Abu ali *e t al.*, 2011).

As improvement of the tomato crop would enhance agricultural productivity and facilitate food security (Fehmida andAhmad,2007), furthermore , characterization of varieties and hybrids which are of wider acceptance by farming community need to be studied in order to regulate their genetic purity during their multiplication and seed quality evaluation (Vishwanath *et al.*, 2010)

Materials and Methods

Acollection of tomato varieties with different growth habit(determinate and indeterminate)and certified sources,such as:GSN,Sanam,Helam,Oula, Kenanh,Douna, Shady lady,Dalal,Bushra,Warda,Fotton, Super regina,Carioca,Special pack,Mongal, Super marimond,SuperQueen, Shahirah,Tamara.

DNA Isolation:

The Genomic DNA Mini Kit (Geneaid Biotech. Ltd; Taiwan Company) provides a quick and easy method for purifying total DNA (including genomic DNA, mitochondrial and chloroplast DNA) from plant tissue. DNA was isolated from leaves according to the method protocol.

PCR Amplification of RAPD-Primers:

According to the Experimental Protocol of AccuPower® TLA PCR PreMix, the PCR reaction mixture was prepared as follows: **1.** 5µl template DNA and 2 µl of primer (10 pmole/µl), were added to each AccuPower® TLA PCR PreMix tube. **2.** Sterilized deionized distilled water was added to AccuPower® TLA PCR PreMix tubes to the final volume of 20 µl. **3.** The tubes were mixed with vortex to dissolve the lyophilized blue pellet, and briefly spine down (all these steps were done in ice). A sequence was amplified individually using oligonucleotide primer (listed in table 1).
Amplification were performed in thermocycler programmed according to annealing temperatures as follows:1. one cycle of 5 min at 94C°, for 40cycle of each 1 min at 94C°, 2 min at 40C° and 2 min at 72C°, with a final extension for one cycle of 5 min at72C° (OPA-03,OPC-19,OPD-13,OPT-08,OPW-04,OPX-17,OPA-01,OPA-02,OPA-04,OPA-10,OPX-01,OPX-03,PX-04,OPX-15,OPX-18,OPN-06,OPX-19). 2. one cycle of 3 min at 94C°, for 45 cycle of each 20 sec at 94C°,20 min at 37C° and 40 sec at 72C°, with a final extension for one cycle of 10 min at 72C° (OPA-14, OPG-17,OPA-15, OPU-03,OPB-17, OPU-14,OPB-18,OPV-19,OPC-08,OPC-09).

Table 1: Operon primers and their sequences.

Primer	Sequence (5'→ 3')	Primer	Sequence (5'→ 3')
OPA-01	CAGGCCCTTC	OPX-01	CTGGGCACGA
OPA-02	TGCCGAGCTG	OPX-03	TGGCGCAGTG
OPA-04	AATCGGGCTG	OPX-04	CCGCTACCGA
OPA-10	GTGATCGCAG	OPX-15	CAGACAAGCC
OPA-14	TCTGTGCTGG	OPX-18	GACTAGGTGG
OPA-15	TTCCGAACCC	OPX-19	TGGCAAGGCA
OPB-17	AGGGAACGAG	OPA-03	AGTCAGCCAC
OPB-18	CCACAGCAGT	OPC-19	GTTGCCAGCC
OPC-08	TGGACCGGTG	OPD-13	GGGGTGACGA
OPC-09	CTCACCGTCC	OPN-06	GAGACGCACA
OPG-17	ACGACCGACA	OPT-08	AACGGCGACA
OPU-03	CTATGCCGAC	OPW-04	CAGAAGCGGA
OPU-14	TGGGTCCCTC	OPX-17	GACACGGACC
OPV-19	GGGTGTGCAG		

Then amplified DNA were separated by electrophoresis in 1.2 % agarose gels (stained with ethidium bromide) (3-4 hr, 70V) .

Scoring Data of RAPD Products:

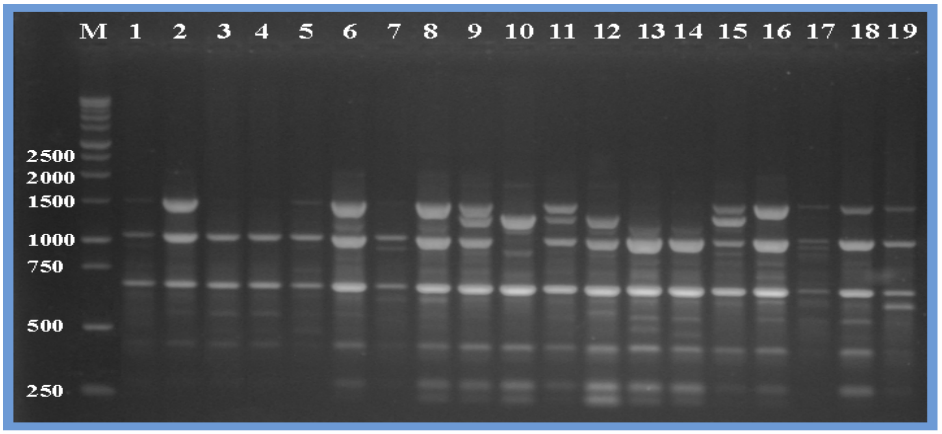
Presence of a product was identified as (1) and absence was identified as (0). By this way, data were scored for all genotypes, their amplification product and primers. The data then entered into NTSYS-PC(Numerical Taxonomy and multivariate Analysis System),Version 1.8 (Applied Biostatistics) program (Rohlf, 1993) using the

program editor.The data were analyzed using SIMQUAL(Similarity for Qualitative Data) routine to generate genetic similarity index (NeiandLi, 1979):GS=2Nij (Ni+Nj). Nij is the number of RAPD bands in common between genotypesIand j, and Ni and Nj are the total number of RAPD bands observed for genotypes Iand j.

Results and Discussion

Across all genomes tested higher number of main bands was generated by both OPV-19 and OPU-03 and higher number of amplified bands obtained in primer OPD-13While lower number of both main and amplified bands obtained by using primer OPA-14 Table 2. The results of both primers OPD-13 and OPU-03 indicate their usefulness in future since they give distinctive fingerprint for all nineteen tested tomato varieties (Figure 1 and 2).

Figure 1: The amplification results obtained with primer OPD-13, lane M: DNA ladder and lanes 1-19: tomato varieties.



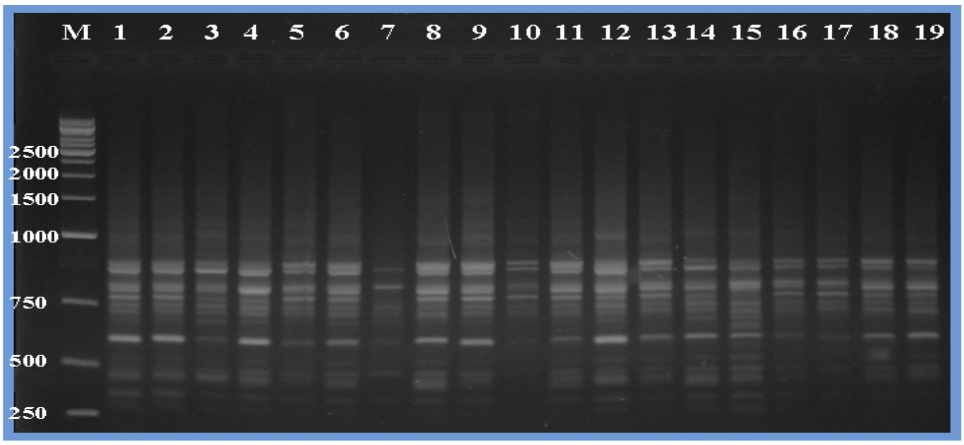


Figure 2: The amplification results obtained with primer OPU-03, lane M: DNA ladder and lanes 1-19: tomato varieties.

Data in Table 2 show that the size of scored bands ranged from 250 bp to 2755 bp,this was nearly close to data obtained by (Huh *et al.*, 2011). Across all genomes tested higher number of main bands was generated by both OPV-19 and OPU-03 and higher number of amplified bands obtained in primer OPD-13While lower number of both main and amplified bands obtained by using primer OPA-14. These variation mainly due to that some primers recognize a high number of annealing site, which is more useful than primers recognizing lower number of annealing sites. In this case the

number of amplified bands will be higher, thus giving a better chance for detecting DNA polymorphisms among individuals (Williams *etal*., 1990).

Amplification profile show the presence of monomorphic bands 70 band out of 442 main band (15.8%) which reveal that genotypes that belong to one species share some genome sequences and differ in others (Russel, *etal*.,1997; Al-Judy, 2004 and AL-Badeiry,2013). These identical sequences are constant in genome and commonly refer to as conserved sequence(Al-Judy, 2004) .

The higher number of polymorphic bands generated in primers OPU-03, OPV-19 and OPC-19 they were higher than values obtained by other reports using the same primers (Abd El Hady *et al*., 2010 and Ezekiel *et al.*, 2011). Study suggests that the primer which produces high polymorphic bands can be further used as polymorphic marker which will prove promising in identification and genetic purity testing in case of tomato (Pal and Singh, 2013). The lower values observed in both primers OPB-17 and OPA-14.

Table 2: The fragment size range (bp), no. of main bands, no. of amplified bands, no. of monomorphic bands, no. of polymorphic bands, no. of unique bands, polymorphism, primer efficiency and discrimination value of each RAPD primer in this study.

No.	Primer	Fragment size range (bp)	No. of main bands	No. of amplified bands	No. of monomorphic bands	No. of polymorphic bands	No. of unique bands	Polymorphism (%)	Primer efficiency	Discriminatory value (%)
1	OPC-08	286-860	10	93	3	6	1	60	0.06	1.92
2	OPA-01	250-2214	19	180	1	18	0	95	0.1	5.8
3	OPU-14	250-2490	18	196	3	13	2	72	0.07	4.17
4	OPX-04	254-1268	17	111	1	11	5	65	0.01	3.53
5	OPA-02	269-1515	14	145	1	12	1	86	0.08	3.85
6	OPX-03	349-2588	14	159	4	8	2	57	0.05	2.6
7	OPC-09	347-1709	22	126	0	17	5	77	0.13	5.45
8	OPX-01	265-2000	13	154	4	9	0	69	0.06	2.9
9	OPV-19	283-1271	28	271	4	19	5	68	0.07	6.09
10	OPU-03	261-2073	26	176	2	21	3	81	0.12	6.73
11	OPB-18	256-1295	13	134	1	10	2	77	0.07	3.21
12	OPB-17	298-1369	9	143	6	3	0	33	0.02	1
13	OPG-17	347-2217	14	156	5	8	1	57	0.05	2.6
14	OPX-18	260-2755	16	127	0	13	3	81	0.1	4.17
15	OPX-15	250-1547	12	127	4	6	2	50	0.05	1.92
16	OPX-19	250-1858	12	141	4	7	1	58	0.05	2.24
17	OPA-14	624-1610	5	60	2	3	0	60	0.05	1
18	OPA-15	280-2172	20	137	3	11	6	55	0.08	3.52
19	OPA-10	255-1616	13	130	3	9	1	69	0.07	2.9
20	OPA-04	338-2057	25	207	3	18	4	72	0.09	5.77
21	OPW-04	308-1920	22	198	1	15	6	68	0.08	4.81
22	OPC-19	268-1131	20	166	0	19	1	95	0.11	6.09
23	OPT-08	250-1813	19	151	3	13	3	68	0.09	4.17
24	OPA-03	327-1279	13	185	5	8	0	62	0.04	2.56
25	OPN-06	391-1172	11	135	1	8	2	73	0.06	2.56
26	OPX-17	309-2297	15	186	0	14	1	93	0.08	4.49
27	OPD-13	301-1672	22	290	6	13	3	59	0.05	4.17
Total no. of bands			442	4284	70	312	60	-	-	-
Average bands per primer			16.4	158.7	2.6	11.6	2.22	-	-	-
Average per primer %			-	-	-	-	-	69.7	0.07	4.5

it was confirmed that wild tomato varieties show 100% polymorphism compired with 36.36% in cultivated varieties (Fan-juan *et al*., 2010).

Data revealed the presence of unique bands up to 5-6 bands per primer , this indicate that every cultivar had one or more novel sequences which was not found in other cultivar. These bands can be successfully used as genetic markers for identification of these cultivars. (Vishwanath *et al*., 2010) .

Data show that out of 4284 amplified bands 312 band were polymorphic while only 70 band were monomorphic .The low degree of similarity (monomorphic bands) indicated high divergence between the genotypes evaluated (Carelli *etal*., 2006). The level of polymorphism reaches 95% the value was higher than that obtained in other reports 63.8% ,85% and 83% in discrimination studies of tomato varieties (Archak *et al*., 2002; Abd El Hady *etal*., 2010; Ezekiel *et al*., 2011) ,In contrast polymorphism could reach 100% using another set of primers and varieties of diverse origin and wider genetic base (Vishwanath *e t al*., 2010).

Evaluation of primer efficiency is of great important in reflecting ability of primer to produce high polymorphic bands according to total number of amplified bands this

indicate in high primer efficiency in both primers OPC-09 and OPU-03 compared to primer OPB-17(AL-Badeiry , 2013).

The discriminatory power of primer which increases by increasing the number of identified varieties using the selected primers (Arif *et al.*, 2010 and AL-Badeiry , 2013). The higher discriminatory power appear in primer OPU-03 which gave distinct fingureprint for all studied varieties while the lower discriminatory power shown by both primers OPB-17 and OPA-14 which failed to identify any variety .

Tomato cultivars well recognized using OPA-04 , OPB-17 and OPD-13 by their ability to produce fragement specific for particular cultivar, this approache will be useful for developing marker-assisted selection tools for genetic enhancement of the tomato plant for desirable traits(Huh *et al*., 2011).

The genetic distance value provides a useful estimate of relationship between a specific pair and a small number of genotypes. Phylogenetic analysis, however, is more appropriate for the interpretation of all possible relationships among a large group of genotypes (Lang and Hang ,2007).

To achieve the calculation of genetic distance or dissimilarity using RAPD-PCR markers at least the results of 10 primers that produce complete and well defined amplification products should be used (Brummer *e t al*., 1995).

The results in table (3)represent the genetic distance among tomato varieties. In order to calculate the genetic distances between tomato varieties, the positions of unequivocally scorable RAPD bands were transformed into a binary character matrix ("1" for the presence and "0" for the absence of a band at a particular position) .Pair wise distance matrices were acompiled by the NTSYS-PC., version 1.8 software using the Dice's and Jacquard's coefficient of similarity to produce the most logical results (Rohlf, 1993; Maguire and Sedgley, 1997).

The lowest genetic distance was (0.2294) between varieties Oula and shadylady which means that the presence of similarity between these two varieties is high degree using RAPD markers, despite the fact they were introduced from different region of geographical origin (China and Holland respectively) and on the basis of morphological features and traits, it was found that these two varieties have diverse characters. Uddin and Boerner, (2008) found that the most closely related two varieties originated from different collection sites.

Ezekiel *etal*.,(2011) reported that varieties may introduced from one locality to the other and assigned anew name.

The impression that the varieties of tomato from aparticular geographical zone are genetically similar may not be true; though, the phenotypic expressions and possible local trade names given by traders or may be the same. Thus, the phenotypic variations exhibited by closely related genotypes may be attributed to response to environmental influences (Falconer, 1989).

The highest genetic distance was (0.9459) between varieties Fotton and Special pack which means that the presence of similarity between them are very low. This concerned with their different geographical origins (China and Holland respectively) and diverse morphological characteristic .

The genetic similarity values ranging from (0.0541 to 0.7706) depending up on the genetic distance values ranging from (0.9459 to 0.2294), which indicate the substantial diversity (94% to 22%) among the varieties used for this study. The genetic distance that relies on nucleotides sequence of DNA giving an evidence for potential genetic similarity between two groups or among individuals within the same species or between species of same genus (Zaid *et al*., 1999).

Table 3: The genetic distance values between tomato varieties studied in RAPD analysis.

Varity	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
1	0.0000																		
2	0.6833	0.0000																	
3	0.6489	0.6895	0.0000																
4	0.9177	0.6763	0.6489	0.0000															
5	0.7609	0.6440	0.7639	0.6149	0.0000														
6	0.3244	0.6855	0.7255	0.9156	0.7719	0.0000													
7	0.8785	0.6421	0.6149	**0.2294**	0.5610	0.8782	0.0000												
8	0.8272	0.7947	0.6164	0.5111	0.6436	0.8352	0.5671	0.0000											
9	0.8555	0.6167	0.7255	0.4619	0.6155	0.8564	0.5144	0.7609	0.0000										
10	0.7947	0.6883	0.7330	0.5722	0.5129	0.7911	0.5226	0.6069	0.5603	0.0000									
11	0.9443	0.7939	0.6856	0.3971	0.5622	0.8864	0.4588	0.4512	0.6054	0.6129	0.0000								
12	0.7912	0.6018	0.7947	0.5620	0.6069	0.7935	0.6055	0.7608	0.6488	0.7254	0.6853	0.0000							
13	0.7630	0.5622	0.6069	0.6069	0.6421	0.7608	0.6488	0.5619	0.7608	0.6139	0.6489	0.7634	0.0000						
14	0.3244	0.6773	0.7340	0.9143	0.8272	0.3244	0.8745	0.8715	0.7947	0.7947	**0.9459**	0.7947	0.7616	0.0000					
15	0.8584	0.6843	0.7334	0.8619	0.6848	0.8621	0.5129	0.6119	0.6488	0.6517	0.6369	0.5645	0.7909	0.8584	0.0000				
16	0.7650	0.5619	0.6051	0.5129	0.6489	0.7630	0.5623	0.6488	0.6880	0.6862	0.6489	0.6009	0.4588	0.7608	0.6853	0.0000			
17	0.8473	0.6603	0.6488	0.4319	0.8831	0.8584	0.5121	0.5236	0.6480	0.5622	0.6214	0.7254	0.6201	0.8587	0.5632	0.6783	0.0000		
18	0.8745	0.6451	0.6719	0.4054	0.5615	0.8749	0.4581	0.4588	0.6229	0.5129	0.4588	0.6883	0.4568	0.8855	0.6069	0.4588	0.5129	0.0000	
19	0.3974	0.7241	0.6872	0.9430	0.7245	0.3977	0.9176	0.8574	0.8271	0.7608	0.8584	0.8272	0.7254	0.3973	0.8885	0.7947	0.8274	0.8434	0.0000

Presence of some common morphological characters among varieties (inflorescence type, fruit shape and colour) increase the present of genetic similarity between studied varieties using RAPD marker(Huh *et al*.,2011) and this agrees with Bai *et al.*, (1997) who found that the degree of genetic similarity between medical plant variety (ginseng) by applying RAPD marker is high when he selected long plants only compared with genetic similarity between random samples of the same species.

In previous studies Fan-juan *etal.*,(2010)detected the lower genetic variation in the cultivated tomato species, the similarity coefficients d etected by RAPD ranged from 0.72 to 1, with an average of 0.95, therefore, the cultivated tomato varieties had narrow genetic background,which was also reported by Hiroaki *et al.*, (2000).

study the genetic relationships among nineteen tomato genotypes varieties based on RAPD can be seen in dendrogram Figur 3, which shows how closely these varieties are related to each other. The aim of producing a dendrogram is to visualize the best representation of the phenetic (overall similarity) or phylogenetic (evolutionary history) relationships among a group varieties, individuals, cultivars, populations, or species. According to this dendrogram it was possible to distinguish two main genetic groups (major clusters). The first small major clusters included four varieties (Douna,GSN,Special pack,Tamara) originate from(Peru,France,and Holland respectively), the first three varieties run and meet at 0.3244, this refers to the genetic distance between them and the genetic similarity is 68%. (varieties that meet at the same genetic distance usually form independent group).

The second large major cluster included (15 varieties) divided in two sub groups.

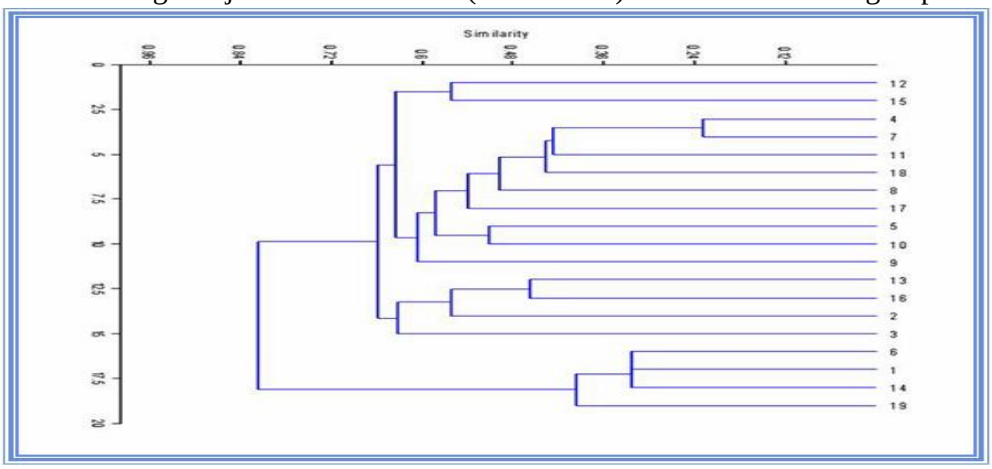


Figure 3: UPGMA dendrogram illustrating the trees of genetic relationship between tomato varieties using RAPD markers.

The distribution of the tomato hybrid varieties into different sub clusters may be an indication of the diversity of the parental species used to generate these population. The phyllogram can be used in the formulation of breeding plans.for example,crosses between closely related genotypes are less likely to produce heterosis(Lang and Hang,2007).

Genetic relationships will help plant breeders to prevent gene erosion within varieties by selecting a large number of different clones of each variety (Ruhl *et al*.,2000).

The importance of finding the genetic distance between varieties studied to help plant breeders in making the right decision by choosing appropriate parents to form new genetic consensus, especially when developing plants in terms of increasing resistance to pathogens and unfavorable environmental conditions (Weeden *et al*., 1992),Thus, the results of the present study have produced the first informative DNA-based markers for common tomato genotypes identification of Iraq and could have strong implications for breeding programs for development of tomato variety as a commercially important crop and would be helpful for future programs regarding tomato varieties genetic improvements, building a genetic map for the local tomato varieties.

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