

Assessment of Genetic Diversity in Cucumber (*Cucumis sativus* L.) Genotypes Using Morphological Characters and AFLP Analysis

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ABSTRACT

For this purpose, 18 cucumber lines were evaluated for their genetic diversity using six morphological characterizations (plant morphology, plant length, length of leaf blade, fruit length, fruit diameter and fruit stem length) and eight AFLP markers. These AFLP primer combinations amplified well and also showed polymorphism. Thus, 1975 AFLP fragments were obtained and 1468 fragments were polymorphic (75.34%). Dendrograms were drawn using UPGMA (Unweighted Pair Group Method) arithmetical averages and according to the UPGMA dendrogram, the cucumber accessions clustered into two main groups. The genetic distances of the dendrogram varied between 0.92 and 0.96. Cluster analysis based on morphological data discriminated all lines into three major clusters in UPGMA dendrogram. The similarity coefficient ranged between 0.888 and 0.982 indicating that the cucumber lines used in the study have a low level of genetic variation. Results obtained from the phylogenetic dendrogram by 8 pairs of AFLP primers were consistent with those from the UPGMA clustering analysis, which were in according with the morphological taxonomy on cucumber.

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Hıyar (*Cucumis sativus* L.) Genotiplerinde Genetik Çeşitliliğin Morfolojik Karakterler ve AFLP Analizi Kullanılarak Değerlendirilmesi

ÖZET

Bu çalışmanın amacı hıyar genotiplerinin morfolojik ve moleküler çeşitliliğini değerlendirmektir. Bu amaçla, altı adet morfolojik özellik (bitki morfolojisi, bitki boyu, yaprak ayası büyüklüğü, meyve uzunluğu, meyve çapı ve meyve sapı uzunluğu) ve sekiz AFLP markeri kullanılarak 18 hıyar hattı genetik çeşitlilik açısından değerlendirilmiştir. Kullanılan AFLP primer kombinasyonları polimorfizm göstermiştir. Çalışma sonucunda 1975 AFLP fragmanı elde edilmiş ve 1468 fragmanın polimorfik olduğu görülmüştür (%75.34). Dendrogramlar, aritmetik ortalamalar kullanılarak UPGMA (Unweighted Pair Group Method) yöntemiyle çizilmiş ve UPGMA dendrogramına göre hıyar genotipleri iki ana gruba ayrılmıştır. Dendrogramın genetik mesafeleri 0.92 ile 0.96 arasında değişmiştir. Morfolojik verilere dayanan küme analizinde ise UPGMA dendrogramı tüm hatları üç ana kümeye ayırmıştır. Çalışmada kullanılan hıyar hatlarının benzerlik katsayısının 0.888 ile 0.982 arasında değişen düşük bir genetik varyasyon seviyesine sahip olduğu belirlenmiştir. 8 çift AFLP primeri ile oluşturulan filogenetik dendogramlarla, morfolojik taksonomi ile yapılan UPGMA kümeleme analizleriyle oluşturulan dendogramlar örtüşmektedir.

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Cucumber (Cucumis sativus L.) is one of the most economically important and fresh eaten vegetables belonging to the family Cucurbitaceae. Due to its narrow genetic base, the intraspecific genetic diversity in cucumber is relatively low (3-12%) as compared to other Cucumis species as revealed in early studies with marker types such as isozymes, RFLPs (restriction fragment length polymorphisms), AFLPs (amplified fragment length polymorphisms) or RAPDs (randomly amplified polymorphic DNAs), (Knerr et al. 1989; Dijkhuizen et al. 1996; Meglic and Staub, 1996; Meglic et al. 1996; Staub et al. 1997; Cavagnaro et al. 2010). However, all methods except AFLP have been reported to have significant disadvantages for this vegetables (Knerr et al. 1989; Waugh and Power, 1992; Staub et al. 1997). As cucumber has a narrow genetic base, it is desirable to develop populations and transfer genes specific to these species using exotic gene sources that control important characters. The evolutions of genetic diversity, relationships and population structure are very important for vegetable characterization and conservation which enhance agricultural production, leading to sustainable development (El-Esawi et al. 2016).

Morphological markers are not widely used because they can be affected by environmental conditions and biochemical markers are not used much due to their number in determining limited the genetic relationships between plant species and varieties. Genetic diversity studies are currently supported by molecular methods, such as molecular markers (Li et al. 2019). AFLP is an effective method allowing the identification of genotypes, the construction of a high saturation genetic map and gene cloning (Vos et al. 1995; Scott et al. 2000). Sequence information is not needed in the AFLP technique and a high rate of polymorphism is obtained. AFLP could explore variation throughout the entire genome, including both coding and non-coding regions of DNA and therefore genome-wide variation was allowed (Wu et al. 2019). The advantage of the system is also its high reproducibility (Witkowicz et al. 2003).

The determination of the genetic distance between genotypes is very important for the breeders in terms of the control of genetic resources and genetic diversity and the selection of genotypes for crossing. In this study, the morphological, phenological, yield and quality characteristics of selected cucumber genotypes that are valuable for agricultural production and the differences between genotypes were determined by using AFLP molecular markers.

MATERIALS and METHODS

Plant material and DNA extraction

In the study, 18 cucumber genotypes (Table 1) that

were estimated to be genetically and morphologically different, were determined and numbered, and some of them were planted and grown in the greenhouse (Aybak and Kaygısız, 2004). Measurements and observations were taken on 10 plants from each genotype and the phenological, morphological, yield and fruit properties were determined based on UPOV criteria. Some seeds were planted in small vials, and leaves were collected from the seedlings and stored at -80°C for DNA isolation. For this purpose, DNA was isolated from 100 mg of leaf material using the CTAB extraction protocol (Weising et al. 1991). DNA quality and concentration were controlled by running each sample on 1% agarose gel electrophoresis and read on a spectrophotometer at 260 to 280 nm wavelengths.

Table 1. The samples used in the study

No.	<u>1. Çalışmada kull</u> Genotypes	Collection site
	Genotipler	Toplandığı yer
1	147 GY	Turkey, Antalya
2	$159 \mathrm{MO}$	Turkey, Antalya
3	523 MO	Turkey, Antalya
4	$529~\mathrm{GY}$	Turkey, Antalya
5	$224~\mathrm{GY}$	Turkey, Antalya
6	225 MO	Turkey, Antalya
7	1102 MO	Turkey, Antalya
8	1103 MO	Turkey, Antalya
9	1140 GY	Turkey, Antalya
10	$315~\mathrm{GY}$	Turkey, Antalya
11	316 MO	Turkey, Antalya
12	1082 GY	Turkey, Antalya
13	1085 MO	Turkey, Antalya
14	1095 MO	Turkey, Antalya
15	$309~\mathrm{GY}$	Turkey, Antalya
16	1008 MO	Turkey, Antalya
17	10222 MO	Turkey, Antalya
18	$10226~{ m GY}$	Turkey, Antalya

AFLP analysis

AFLP reactions were performed with DNA samples obtained from plants using a commercial kit (INVITROGEN) as described (Vos et al. 1995; Roldán-Ruiz et al. 2000). Eight AFLP primer sets were used to analyze polymorphisms. Total genomic DNA was digested using the two restrictive enzymes EcoRI (New England Biolabs Ltd, NEB) and MseI (NEB). DNA fragments were ligated with EcoRI and MseI adapters using T4 DNA ligase (NEB) for PCR amplification. After the adapters were ligated to the DNA, they were pre-selected to amplify the existing DNA fragments and eliminate the components that were not ligated to the adapter. In the pre-amplification step, genomic DNA was amplified with AFLP pre-amplification primers EcoRI (5'-GACTGCGTACCAATTCA-3') and MseI (5'-GATGAGTCCTGAGTAAC-3'). The preselective amplification reactions were prepared in a 25 µl reaction mixture, containing 3 µl of DNA sample, 1.25 μ L of each preselective primers (50 ng/ μ l), 2.5 μ L reaction buffer (10x), 0.5 µL dNTP mixtures, 2 units of Taq DNA polymerase (Thermo Scientific) and 16.5 µL sterile-double distilled water. The PCR reactions were conducted for 15 cycles of 94 °C for 3 min, 94 °C for 1 min, 65 °C for 1 min, 70°C for 1 min; and then 20 cycles of 94 °C for 1 min, 55°C for 1 min, 72°C for 30 s, plus a final elongation step of 72°C for 7 min. The samples were diluted for selective amplification and PCR reactions were performed with a touch-down cycle as follows: 12 cycles of 94 °C for 45 s, 65 °C for 30 s (a decrease of 0.7 °C per cycle) and 72 °C for 45 s, and then 25 cycles of 94 °C for 45 s, 55 °C for 30s, 72 °C for 45s, and 72 °C for 5 min for a final elongation step. The PCR products were separated by 8% (W/V)polyacrylamide gel electrophoresis.

Morphological Data Analysis

Overall, 50 days after planting, plant morphology, plant length, the length of leaf blade, fruit length, fruit diameter and fruit stem length were determinned. For each of the 6 morphological characters, the mean and standard deviation values were calculated. These morphological features were determined in 10 randomly chosen plants based on the UPOV criteria (UPOV, 2019).

Data analysis

AFLP data from eight primers were transformed into a binary matrix, scored as present "1", absent "0", for further analyses. The total number of fragments, the number of polymorphic fragments, the percentage of polymorphic loci (%) and the polymorphic information content (PIC) were calculated using the software GenAlEx 6.5 (Peakall and Smouse, 2006). The UPGMA dendrogram was constructed using (unweighted pair group method with the arithmetic average) based on Nei's genetic distance (Nei, 1972) and the NTSYS ver 2.10 software (Staub et al. 2005). Similarity indices and pairwise genetic distance values were calculated from AFLP data using the UPGMA method and NTSYS software. Clustering analysis was performed using SPSS22.0 software. To examine the correlation between six morphological characters, the pearson correlation coefficient was calculated using IBM SPSS Statistics 22.0 program.

RESULTS and DISCUSSION

Morphological characterization

Morphological plant characters including plant length, length of leaf blade, fruit length, fruit diameter and fruit stem length were analyzed in 18 selected cucumber genotypes (Fig. 1).



Figure. 1. General growth forms, leaves and flowers (A) and fruit morphology (B) of Cucumber genotypes used for diversity analysis.

Şekil 1. Çeşitlilik analizi için kullanılan hıyar genotiplerinin genel büyüme şekilleri, yaprak ve çiçekleri (A) ile meyve morfolojisi The CV (coefficient of variation) values for three of the morphological characters including internodes length, fruit length and fruit stem length indicated a high level of variation (i.e.>10%), (Table 2). The mean value of plant height was 110.9 ± 7.66 cm, varied from $69.6 \pm$ 8.1 cm (147 GY) to 161.1 ± 5.0 cm (316 MO) among lines. The mean value of internodes length was $7.7 \pm$ 0.79 cm and varied from 5.3 ± 0.7 cm to 9.2 ± 0.8 cm in 147 GY and 1085 MO lines, respectively. The average value of leaf blade length was 21.9 ± 1.54 and varied from 17.5 ± 1.2 (523 MO, and 529 GY) to 30.6 ± 1.2 (1102 MO). The mean value of fruit length was $14.8 \pm$ 1.57 cm, varied from 11.4 ± 1.0 cm (315 GY) to $17.7 \pm$ 2.4 cm (10222 MO) lines. The mean value of fruit diameter was 11.8 ± 1.14 mm and varied from 9.4 ± 0.5 mm to 14.3 ± 0.8 cm in 1095 MO and 224 GY lines, respectively. The average value of fruit stem length was 2.1 ± 0.55 and varied from 1.2 ± 0.4 (224 GY, and 225 MO) to 4.2 ± 0.6 (10226 GY) (Table 2).

Based on Pair-wise comparisons among the 18 genotypes, an Euclidean distance matrix was obtained and an unweighted pair-group method using mean (UPGMA) dendrogram arithmetic was constructed. At an Euclidean distance 19.1, three major clusters were obtained based on the morphological characters (Fig. 2), (plant morphology, plant length, length of leaf blade, fruit length, fruit diameter and fruit stem length). A large number of genotypes (12 genotypes) grouped together in cluster I. Cluster II grouped two genotypes. Four genotypes grouped together in cluster III (Fig. 2). At the 0.05 level of confidence, the results from Pearson correlation analysis appeared that plant height was positively correlated with internodes length, and leaf blade length. Internodes length was positively correlated with leaf blade length but negatively correlated with fruit diameter and fruit stem length (Table 3).

The cucumber has a very narrow gene pool that limits the development of new cucumber varieties (Innark et al. 2013). Genetic diversity data in cucumber genotypes are used in cucumber breeding programs to help determine parental lines. For morphological data, most characters showed an extremely narrow range, indicating that the cucumber genotypes used in the study have a low level of genetic variation. From the correlation analysis of six morphological characters, plant height was positively correlated with internodes length, and leaf blade length. This result was logical because internodes length and leaf blade length were considered as the yield components affecting the plant height. The UPGMA cluster analysis of morphological measurements were effective in distinguishing 18 cucumber ecotypes. The results from this study were consistent with the previous results reported by Innark et al. (2013), which evaluated the morphological data correlation in switchgrass (Cortese et al. 2010) and Chrysanthemum morifolium (Shao et al. 2010).

Table 2. The morphological characteristics of eighteen cucumber lines *Tablo 2. Onsekiz hyar hattının morfolojik özellikleri*

No.	Genotypes <i>Genotipler</i>	Plant height (cm) <i>Bitki boyu</i> <i>(cm)</i>	Internodes length (cm) <i>Boğum arası</i> <i>uzunluğu</i> (cm)	Leaf blade length (cm) <i>Yaprak ayası</i> genişliği (cm)	Fruit length (cm) <i>Meyve</i> <i>uzunluğu</i> (cm)	Fruit diameter (mm) Meyve çapı (mm)	Fruit stem length (cm) <i>Meyve sap</i> <i>uzunluğu</i> (cm)
1	$147~\mathrm{GY}$	69.6 ± 8.1	5.3 ± 0.7	16.3 ± 0.9	14.7 ± 1.6	$12.0{\pm}1.6$	2.5 ± 0.5
2	159 MO	83.1 ± 8.1	8.0 ± 0.8	20.8 ± 1.8	16.6 ± 1.1	11.4 ± 0.5	$1.4{\pm}0.5$
3	523 MO	73.9 ± 9.2	8.1 ± 0.7	17.5 ± 1.2	14.0 ± 1.9	11.2 ± 0.6	1.6 ± 0.5
4	$529~\mathrm{GY}$	89.5 ± 8.9	5.8 ± 0.8	17.5 ± 1.2	13.6 ± 1.6	10.9 ± 1.1	2.5 ± 0.5
5	$224~\mathrm{GY}$	132.7 ± 7.4	7.4 ± 0.8	22.2 ± 1.4	14.7 ± 0.8	14.3 ± 0.8	1.2 ± 0.4
6	225 MO	134.2 ± 7.3	7.3 ± 0.7	21.6 ± 0.8	14.8 ± 1.2	13.0 ± 1.3	1.2 ± 0.4
7	1102 MO	117.8 ± 9.4	$7.7{\pm}0.9$	30.6 ± 1.2	13.5 ± 1.2	11.8 ± 1.0	2.5 ± 0.5
8	1103 MO	113.8 ± 6.7	8.1 ± 0.6	$27.0{\pm}1.7$	12.3 ± 0.7	11.3 ± 1.0	$1.4{\pm}0.5$
9	1140 GY	120.7 ± 9.3	9.1 ± 1.2	22.3 ± 2.3	14.7 ± 3.4	12.8 ± 2.0	$3.4{\pm}0.5$
10	$315~\mathrm{GY}$	117.4 ± 8.5	8.3 ± 0.7	17.7 ± 2.3	11.4 ± 1.0	11.7 ± 1.2	$1.7{\pm}0.5$
11	316 MO	161.1 ± 5.0	8.1 ± 1.1	21.1 ± 0.6	15.7 ± 1.5	12.5 ± 1.7	1.5 ± 0.5
12	$1082 \mathrm{~GY}$	122.1 ± 8.5	$8.4{\pm}0.7$	23.7 ± 1.9	17.4 ± 1.3	11.3 ± 1.2	$3.9{\pm}0.7$
13	1085 MO	142.8 ± 9.0	$9.2{\pm}0.8$	24.5 ± 1.5	$17.0{\pm}1.4$	$11.0{\pm}1.7$	1.5 ± 0.7
14	1095 MO	114.0 ± 8.9	8.5 ± 0.7	21.4 ± 2.0	13.2 ± 1.5	$9.4{\pm}0.5$	1.8 ± 0.9
15	$309~\mathrm{GY}$	78.7 ± 6.9	6.6 ± 0.7	18.1 ± 1.4	14.2 ± 1.7	11.7 ± 1.2	$2.4{\pm}0.5$
16	1008 MO	136.3 ± 8.2	$8.0{\pm}1.2$	25.4 ± 2.8	15.0 ± 2.2	11.5 ± 1.0	1.6 ± 0.5
17	10222 MO	98.8 ± 4.6	8.6 ± 0.5	27.4 ± 1.3	17.7 ± 2.4	12.5 ± 1.8	1.6 ± 0.7
18	10226 GY	89.2±3.8	6.3 ± 0.7	19.1 ± 1.4	16.4 ± 1.7	12.7 ± 1.3	4.2±0.6
	CV (%)	6.90	10.30	7.03	10.57	9.62	26.12



Figure. 2. Dendrogram constructed by UPGMA clustering method based on morphological attributes of eighteen cucumber genotypes

Şekil 2. Onsekiz hıyar genotipinin morfolojik özelliklerine dayanan UPGMA kümeleme yöntemiyle oluşturulan dendogram

Table 3. Pearson correlation analysis of six morphological characters
Tablo 3. Altı morfolojik karakterin Pearson korelasyon analizi

	Plant	Internodes	Leaf blade	Fruit length	Fruit	Fruit stem
	height	length	length	Meyve	diameter	length
	Bitki	Boğum arası	Yaprak ayası	uzunluğu	Meyve çapı	Meyve sap
	boyu	uzunluğu	genişliği			uzunluğu
Plant height	****					
Internodes length	0.544^{*}	****				
	0.020					
Leaf blade length	0.478^{*}	0.526^{*}	****			
	0.045	0.025				
Fruit length	0.081	0.153	0.192	****		
	0.750	0.546	0.445			
Fruit diameter	0.212	-0.156	0.041	0.223	****	
	0.398	0.536	0.872	0.374		
Fruit stem length	-0.269	-0.249	-0.132	0.226	0.012	****
-	0.281	0.320	0.602	0.367	0.962	

Upper number is Pearson correlation coefficient and lower number is P value.

Polymorphism analysis of AFLP data

Eight AFLP primer combinations were utilized on eighteen cucumbers (Cucumis sativus L.) genotypes. A total of 1975 scorable fragments were determined, of which 1468 (75.34%) were polymorphic. This percentage of polymorphic loci (75.34%) was higher than that reported by Xixiang et al. (2004) (66%). They reported that eight pairs of AFLP primers generated 425 scorable bands in total and 66% of which were polymorphic. In the current study, the number of bands ranged from 116 to 372 with an average of 246.8 (Table 4) and the number of polymorphic fragments for each primer pair varied from 21 to 368 with an average of 183.5. However, the percentage of polymorphic loci ranged from 30.43 to 99.14% (Table 4). In another study, 92 Turkish cucumbers (*Cucumis sativus* L. var. sativus) accessions were characterized by using SRAP markers and 153 SRAP fragments were obtained and 138 fragments were polymorphic (90.2%). The level of polymorphism observed herein is similar to that reported by Kong et al. (2006) but lower than that detected by genomic SSRs (Watcharawongpaiboon and Chunwongse, 2008). The observed PIC values of AFLP markers in this study varied from 0.27 to 0.58, however, the PIC values were estimated for 45 SSR primer pairs as ranging from 0.11 to 0.78, with an average of 0.47(Watcharawongpaiboon and Chunwongse, 2008). Similar results were observed from the cucumber germplasm collection containing a range of ecotypes distributed in China sources by Hu et al. (2010). In their study, PIC values ranged from 0.185 to 0.642 with an average of 0.374.

Primer sets Primer setleri	Total number of fragments <i>Toplam bant</i> sayısı	Number of polymorphic fragments <i>Polimorfik batların</i> sayısı	Percentage of polymorphic loci (%) <i>Polimorfik lokusların</i> <i>yüzdesi</i>	Polymorphic information content (PIC) Polimorfik bilgi içeriği (PBİ)
EcoRI-ACA/ MseI-CAT	371	203	54.72	0.48
EcoRI-ACT/ MseI-CTA	169	21	30.43	0.58
EcoRI-AGC/ MseI-CTT	231	123	53.25	0.44
EcoRI-ACA/ MseI-CAA	289	266	92.04	0.36
EcoRI-ACT/MseI-CAG	372	368	98.92	0.45
EcoRI-AGC/MseI-CAT	242	211	87.19	0.38
EcoRI-ACA/ MseI-CTA	185	161	87.02	0.33
EcoRI-ACT/MseI-CTT	116	115	99.14	0.27
Mean	246.87	183.5	75.34	0.41

Table 4. AFLP primer sets and the number of amplified AFLP fragments Tablo 4. AFLP primer setleri ve çoğaltılmış AFLP bantlarının sayısı

Similarity indices and genetic distance

Similarity coefficients between cucumber genotypes ranged from 0.888 to 0.982 (Table 5), which was higher than those of reported by Hu et al. (2010) for EST-SSR markers from the cucumber. Similarity coefficients calculated from EST-SSR data varied from 0.542 to 0.941 with a mean value of 0.792 (Hu et al. 2010). The differences in all of these data could be attributed to the differences in the EST analysis. The highest degree of similarity indices (0.982) and the lowest genetic distance (0.018) was observed between 10226 GY and 10222 MO, while the lowest degree of similarity (0.888) and the highest genetic distance (0.112) was observed between 1095 MO and 529 GY. The second highest similarity indices (0.964) were between 1085 MO and 224 GY, while the second lowest similarity indices (0.893) were between 1082 GY and 1102 MO. Most genotypes evaluated in this study were shown to be very closely related and shared a high degree of genetic similarity. Relatively large genetic distances were observed by RAPD profiling in cucumber (between 0.01 and 0.58) (Horejsi and Staub, 1999) suggesting that these genotypes were more unrelated and RAPD analysis was useful in genotypic differentiation for cucumber. On the contrary, the study performed by Park et al. (2000) determined that AFLP analyses created more polymorphisms than either RFLPs or RAPDs. The selected 37 primer combinations produced approximately 3000 bands, of which 339 bands (11%) were polymorphic more than RFLPs or RAPDs. Furthermore, they suggested converting the AFLP marker to a dual-primer PCR based marker to enhance its usefulness in cucumber breeding. All these results indicate that both AFLP markers and genotypes used are the most important determinants of the similarity indices and genetic distances.

Cluster analysis

The similarity matrix for genotypes in cucumber was calculated by using the Dice coefficient method with

the NTSYS program. According to the dendrograms, the minimum genetic similarity was 92% while the maximum similarity between cucumber accessions was 96%. The UPGMA analysis of the marker data resulted in two main groups. The first group included 147GY, 1140 GY, 529 GY, 225 MO, 159 MO, 1103 MO, 1102 MO, 315 GY, 316 MO, 1022 MO in which, 315 GY, 316 MO and 1022 MO clustered as a subgroup, whereas 147GY, 1140 GY, 529 GY, 225 MO, 159 MO, 1103 MO and 1102 MO clustered as another subgroup. The second group consisted of 523MO, 224 GY, 1085 MO, 1008 MO, 1082 GY, 1095 MO, 309 GY in which 1082 GY, 1095 MO and 309 GY grouped into one subgroup, and 523MO, 224 GY, 1085 MO and 1008 MO grouped into another subgroup. The cluster analysis of AFLP data showed that the cucumber genotypes were closely related to each other (Figure 3).

CONCLUSIONS

In this study, we selected eight pairs of AFLP informative primers to assess the genetic diversity and relationships among cucumber genotypes collection. AFLP fragments generated by the 8 AFLP primer pairs assayed in this study were 1975 of which 1468 (75.34%) were polymorphic. The overall mean similarity index calculated based on AFLP fragments amplified using Nei's similarity index ranged from 0.888 to 0.982 with an average of 0.936. There are only a few studies related the characterization of genetic diversity of cucumber accessions by AFLP. The study revealed a low molecular diversity among the cucumber accessions. Morphological traits of the cucumber accessions were used in conjunction with molecular data to determine germplasm collections. These results may help in the selection of accessions as breeding materials for the development of new cultivars.

Table 5. Similarity indices (below diagonal) and the genetic distance values (above diagonal) calculated from AFLP data of 18 cucumbergenot, Tablo 5. 18 Hiyar genotipinin AFLP verilerinden hesaplanan benzerlik indeksleri (diagramın altında) ve genetik uzaklık değerleri (diyagı üzerinde)	ucumbergenotypes	eğerleri (diyagramın	
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	$147~{\rm GY}_{\rm MO}^{159}$	$ m Y_{MO}^{159}$	523 MO	$529~{ m G}^{\circ}$	$529~{ m GY}224~{ m GY}^{22}_{ m MC}$	$ m _{MO}^{225}$	1102 MO	1103 MO	1140 GY	$^{315}_{ m GY}$	316 MO	1082 GY	1085 MO	1095 MO	$309\mathrm{GY}_{\mathrm{MO}}^{1008}$,1008 MO	10222 MO	10226 GY
147 GY	* * * *	0.07	0.051	0.067	0.069	0.058	0.059	0.061	0.048	0.070	0.074	0.078	0.069	0.060	0.090	0.075	0.062	0.089
159 MO	0.931	****	0.069	0.069	0.065	0.077	0.062	0.051	0.068	0.074	0.071	0.066	0.080	0.086	0.070	0.071	0.086	0.072
523 MO	0.950	0.931	* * * *	0.073	0.041	0.064	0.066	0.053	0.054	0.092	0.094	0.056	0.061	0.068	0.073	0.074	0.059	0.081
$529~{ m GY}$	0.935	0.931	0.928	* * * *	0.047	0.051	0.066	0.054	0.062	0.056	0.073	0.099	0.063	0.112	0.103	0.088	0.085	0.062
$224~{ m GY}$	0.932	0.935	0.960	0.953	****	0.061	0.092	0.058	0.073	0.081	0.091	0.066	0.037	0.092	0.084	0.071	0.064	0.073
225 MO	0.943	0.924	0.936	0.950	0.939	***	0.066	0.067	0.062	0.077	0.065	0.077	0.076	0.081	0.102	0.103	0.073	0.052
1102 MO	0.942	0.938	0.935	0.934	0.909	0.935	****	0.055	0.062	0.086	0.082	0.107	0.106	060.0	0.096	0.068	0.054	0.069
1103 MO	0.940	0.950	0.947	0.947	0.943	0.933	0.946	****	0.057	0.066	0.084	0.081	0.086	0.091	060.0	0.076	0.083	0.057
1140 GY	0.953	0.934	0.946	0.938	0.927	0.939	0.938	0.943	****	0.075	0.055	0.075	0.080	0.071	0.063	0.093	0.047	0.086
$315~{ m GY}$	0.931	0.926	0.909	0.945	0.920	0.924	0.915	0.935	0.926	****	0.048	0.068	0.066	0.071	0.086	0.079	0.051	0.050
316 MO	0.927	0.930	0.906	0.927	0.909	0.935	0.919	0.917	0.945	0.952	****	0.055	0.077	0.061	0.059	0.083	0.063	0.054
$1082~{ m GY}$	0.924	0.934	0.945	0.901	0.934	0.924	0.893	0.921	0.926	0.933	0.945	****	0.067	0.049	0.057	0.087	0.068	0.065
1085 MO	0.932	0.920	0.939	0.939	0.964	0.925	0.894	0.915	0.920	0.934	0.924	0.934	****	0.071	0.062	0.055	0.089	0.071
1095 MO	0.941	0.914	0.934	0.888	0.908	0.919	0.910	0.909	0.929	0.929	0.940	0.951	0.930	****	090.0	0.068	0.081	0.085
$309~{ m GY}$	0.912	0.930	0.927	0.897	0.916	0.898	0.904	0.910	0.937	0.914	0.941	0.944	0.938	0.940	****	0.067	0.080	0.084
1008 MO	0.927	0.930	0.927	0.912	0.930	0.898	0.933	0.924	0.908	0.922	0.918	0.914	0.945	0.932	0.933	****	0.081	0.069
10222 MO	0.939	0.914	0.942	0.917	0.936	0.927	0.947	0.917	0.953	0.949	0.938	0.932	0.912	0.920	0.920	0.920	****	0.018
10226 GY	0.912	0.928	0.919	0.939	0.929	0.949	0.932	0.944	0.914	0.950	0.946	0.935	0.929	0.916	0.917	0.931	0.982	***
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Figure 3. Phylogenetic dendrogram based on Nei's genetic identity of eighteen *Cucumis sativus* genotypes Şekil 3. *Onsekiz Cucumis sativus genotipinin Nei'nin genetik tanımlamasına dayalı filogenetik dendogramı*

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Authors have declared no conflict of interest.

Author's Contributions

The contribution of the authors is equal.

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