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Araştırma Makalesi

Türkiye Yerli Patlıcan (*Solanum melongena* L.) Populasyonlarının Moleküler ve Morfolojik Karakterizasyonu

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ÖZ

Bu çalışma, Kayseri ilinden toplanan, "Yamula Patlıcanı" olarak adlandırılan 28 yerel patlıcan genotipi ile Türkiye'de yaygın olarak yetiştirilen 3 Kemer patlıcanı ve 1 Manisa patlıcanı genotipinin moleküler ve morfolojik karakterizasyonu için yapılmıştır. 10 ISSR ve SRAP primeri ile 30 morfolojik karakter kullanılarak analizler gerçekleştirilmiştir. Morfolojik olarak birbirine en çok benzeyen genotipler ERU 3006 ve ERU 3007'dir. Moleküler çalışmalarda polimorfizm oranı ISSR tekniğinde %77,36, SRAP tekniğinde ise %73,72 olarak tespit edilmiştir. Genotiplerin morfolojik karakterizasyonu için UPOV kriterleri listesinden seçilen toplam 25 karakter kullanılmıştır. "Kaliksteki antosiyanin renklenme yoğunluğu" karakterinde herhangi bir varyasyon görülmezken, en yüksek varyasyon katsayısı "yaprak rengi" ve "yaprak kenarı şekli" karakterleri için sırasıyla %89.60 ve %86.15 olarak hesaplanmıştır. Tüm bitki kısımları gruplara ayrıldığında en yüksek varyasyonun yaprak özelliklerinde meydana geldiği anlaşılmıştır (Varyasyon katsayısı: %59.39). Bunu bitki gövde ve meyve özellikleri sırasıyla %48,14 ve %43,20 olarak izlemiştir. Mevcut bulgular, Yamula patlıcan genotipleri ile kontrol genotipleri arasında önemli farklılıklar olduğunu ve Yamula patlıcan genotiplerinde büyük bir varyasyon olduğunu ortaya koymuştur. Mevcut varyasyonlar patlıcan yetiştirme programlarında kullanılabilir. Ayrıca bölgesel genetik popülasyonların geniş bir patlıcan genetik çeşitliliğini içerdiği ve ileriki ıslah programları için iyi bir kaynak olabileceği sonucuna varılmıştır.

Anahtar kelimeler: Tanımlama, genetik çeşitlilik, ISSR, popülasyon yapısı, SRAP.

Molecular and Morphological Characterization of Turkish Local Eggplant (*Solanum melongena* L.) Populations

ABSTRACT

This study was conducted for molecular and morphological characterization of 28 local eggplant genotypes (so-called "Yamula" eggplant) collected from Kayseri province, 3 Kemer eggplant, and 1 Manisa eggplant genotype, commonly cultivated in Turkey. Molecular analyses were carried out with the use of 10 ISSR and SRAP primers and 30 morphological characteristics. Morphological analyses revealed the nearest genotypes as ERU 3006 - ERU 3007. The polymorphism ratio was identified as 77.36% and 73.72% with ISSR and SRAP markers, respectively. The 25 differenet characters selected from the UPOV description list were used for the morphological characterization of the accessions. While no variation was observed in "intensity of anthocyanin coloration in calyx", the highest variation coefficient was calculated for "leaf color" and "leaf blade margin shape" (89.60% and 86.15%, respectively). When all plant parts were divided into groups, the highest variation belonged to leaf characteristics (Variation coefficient: 59.39%), followed by the plant stem and fruit characteristics (48.14% and 43.20%, respectively). Results showed that variations exist within Yamula and between Yamula and control genotypes. Present variations could be used in eggplant breeding programs. It was also concluded that regional genetic populations inhabit a wide eggplant genetic diversity which can be a good source for further breeding programs.

Key words: Description , genetic diversity, ISSR, population structure, SRAP.

INTRODUCTION

Eggplant (*Solanum melongena* L.) belongs to Solanaceae family and is largely produced all around the world. It has been cultured in Asia for a thousand year and so called as the "king of vegetables" in India (Daunay and Janick, 2007). Eggplant is rich in vitamins and minerals and is a strong antioxidant. It is a rich source of some polyphenols (Sudheesh et al., 1999; Nisha et al., 2009). Therefore, it has a great economical value. Eggplant is originated from Asia and Africa. *S. melongena* is the most known species and largely grown all around the world. The other cultured eggplants are relatives of *S. melongena* and include red eggplant (*S. aethiopium* L.) and Gboma eggplant (*S. macrocarpon* L.) grown in Africa. Wild species are relatives of all cultured eggplants and the number of wild species is around 200 (Daunay et al., 2000a).

Eggplant is believed to be originated in India (Laumonnier, 1952). Following India, it exhibited spread to the west and entered the Europe over Spain. Zhukowsky (1958) indicated that eggplant was cultured in Europe in the 13th and 14th centuries. Eggplant entered Anatolia toward the end of the16th century or at the beginning of the17th century. It entered the America after the exploration of the American Continent. Today, eggplant is cultured over large sections of northern and the southern hemispheres (Vural et al. 2000). In terms of production quantities, eggplant comes after tomato, pepper, and cucumber. Eggplant is consumed as fresh, canned, and processed food and has a special place in Turkish cuisine.

Mutation, natural pollination, and hybridization, together with selection gave rise to genetic diversity in eggplant genotypes, decreased fruit prickles and bitterness, and altered fruit shape, size and color (Frary et al., 2007). Genetic diversity accumulated and many different heirlooms emerged in countries where it was cultivated (Prohens et al., 2003). The entrance of the eggplant into Turkey was carried out by the silk-road. The genetic diversity accumulated in producing areas and by the trade of eggplant throughout the centuries in Anatolia (Janick, 2001). Eggplant cultivation was done in the open field until the second half of the 1970s in Turkey and then protected cultivation started. Eggplant cultivation in greenhouse begun with local varieties. There is an accelerated transition in eggplant cultivation in Turkey from the production of standard cultivars into production of F1 hybrid cultivars. However, similar to some other vegetables, despite the yield increase with F1 hybrids, desired quality is not always achieved in eggplant production. Therefore, local cultivars are commonly used for standard and F1 hybrid seeds. To do this, initially local genetic resources should be collected and characterized. Then, breeding programs should be conducted to eliminate some negativities of a standard cultivar. The narrowing genetic base is one of the important problems faced in eggplant breeding programs. The genetic diversity is low among the genotypes with dark purple-black fruits (Muñoz-Falcón et al., 2009). To create variations, time-consuming and expensive methods are needed, including mutation breeding, interspecific hybridization, and biotechnological approaches. The genetic variation among the heirlooms was seen in previous studies (Demir et al., 2010; Muñoz-Falcón et al., 2008, 2009; Prohens et al., 2003, 2008, 2011).

Yamula eggplant largely grown in and around Kayseri province, is a standard eggplant cultivar and constitutes an important genetic resource. Yamula eggplant has great potential in Central Anatolia Region (Kırıkkale, Aksaray, Niğde, Nevşehir, Kırşehir, Kayseri, Sivas and Yozgat provinces). Eggplant has a specific striped texture and firm fruit flesh. It is consumed in fresh, dried and brined fashions. Because growers use their seeds, yields are continuously decreasing and disease sensitivity, new pests and diseases limited the production. Nonuniform fruit sizes and colors are commonly encountered in markets. Unless the yield and quality were not improved in this cultivar, it will not be produced anymore in production regions. Breeding is the only way to prevent decreases in production. Increased resistance to pests and diseases may also prevent such decreases in production activities. Selection is the first step of breeding programs. Pure-line selection should be made for the most proper plants in terms of yield, quality and plant strength, then these lines should be purified through selfing, the genotypes with the greatest yield and quality should be identified and their potentials should be put forth with micro and macro yield trails.

This study was conducted for molecular and morphological characterization of Yamula eggplant genotypes locally grown in Kayseri province and selected by considering fruit and plant characteristics.

MATERIALS AND METHODS

Morphological and molecular studies were conducted at Erciyes University, Kayseri, Turkey in 2015-2016. In total, 32 plant materials were used in the study and 4 of them were control genotypes (3 Kemer eggplant and 1 Manisa eggplant) to compare with Yamula eggplant (Table 1).

Table 1. Eggplant genotypes used in the study.

No	Label of the genotype	Name of genotype	Providing method	Place/company of origin
1	ERÜ-3004	Yamula Eggplant	Survey in the field	Grower
2	ERÜ-3005	Yamula Eggplant	Survey in the field	Grower
3	ERÜ-3006	Yamula Eggplant	Survey in the field	Grower
4	ERÜ-3007	Yamula Eggplant	Survey in the field	Grower
5	ERÜ-3008	Yamula Eggplant	Survey in the field	Grower
6	ERÜ-3009	Yamula Eggplant	Survey in the field	Grower
7	ERÜ-3010	Yamula Eggplant	Survey in the field	Grower
8	ERÜ-3011	Yamula Eggplant	Survey in the field	Grower
9	ERÜ-3012	Yamula Eggplant	Survey in the field	Grower
10	ERÜ-3013	Yamula Eggplant	Survey in the field	Grower
11	ERÜ-3014	Yamula Eggplant	Survey in the field	Grower
12	ERÜ-3015	Yamula Eggplant	Survey in the field	Grower
13	ERÜ-3016	Yamula Eggplant	Survey in the field	Grower
14	ERÜ-3017	Yamula Eggplant	Survey in the field	Grower
15	ERÜ-3018	Yamula Eggplant	Survey in the field	Grower
16	ERÜ-949	Yamula Eggplant	Survey in the field	Grower
17	ERÜ-950	Yamula Eggplant	Purchased	Seed Sales Office
18	ERÜ-951	Yamula Eggplant	Survey in the field	Grower
19	ERÜ-952	Yamula Eggplant	Survey in the field	Grower
20	ERÜ-953	Yamula Eggplant	Survey in the field	Grower
21	ERÜ-954	Yamula Eggplant	Survey in the field	Grower
22	ERÜ-955	Yamula Eggplant	Survey in the field	Grower
23	ERÜ-956	Yamula Eggplant	Survey in the field	Grower
24	ERÜ-957	Yamula Eggplant	Survey in the field	Grower
25	ERÜ-961	Yamula Eggplant	Survey in the field	Grower
26	ERÜ-964	Manisa Eggplant	Purchased	Seed Sales Office
27	ERÜ-1255	Kemer Eggplant	Purchased	Seed Sales Office
28	ERÜ-1256	Kemer Eggplant	Purchased	Seed Sales Office
29	ERÜ-3000	Yamula Eggplant	Survey in the field	Grower
30	ERÜ-3001	Yamula Eggplant	Survey in the field	Grower
31	ERÜ-3002	Yamula Eggplant	Survey in the field	Grower
32	ERÜ-3003	Yamula Eggplant	Survey in the field	Grower

Survey

The survey studies were performed in Kayseri province in September 2015 and 2016. In total, 28 materials were collected from different farms. The locations of collected eggplants in Kayseri province are presented in (Figure 1) and their geographic coordinates were defined (38° 53′ 11.2956" and 35° 15′ 41.9832").

Figure 1. Sampling locations.

Morphological characterization

For each accession, the seeds were sown in seedling trays containing peat moss. Seedlings were transplanted into the greenhouse when they reached 4-5 leaf stages. Fifteen plants were planted for each genotype. Morphological observations were performed according to 30 descriptors chosen by the International Board for Plant Genetic Resources Institute (IBPGRI), the International Union for the Protection of New Varieties of Plants (UPOV), plant feature criteria and some criteria of the breeders. Descriptors include plant, leaf, flower and fruit trait observations and measurements (Table 2).

Table 2. Descriptors used for characterization and evaluation of eggplant accessions used in the study.

Trait No	Traits	Description
1	Plant growth habit:	(3) Erect, (5) Semi-erect, (7) Horizontal
2	Plant height:	(3) Short, (5) Medium, (7) Tall
3	Plant stem length:	(3) Short, (5) Medium, (7) Long
4	Stem, anthocyanin coloration:	(1) Absent, (2) Present
5	Stem, intensity of anthocyanin coloration:	(3) Weak, (5) Medium, (7) Strong
6	Stem, pubescence:	(3) Weak, (5) Medium, (7) Strong
7	Branch, internode distance:	(3) Short, (5) Medium, (7) Long
8	Leaf blade size:	(3) Small, (5) medium, (7) Large
9	Leaf blade margin shape:	(1) Whole, (2) Serrated, (3) Wavey
10	Leaf blade margin sinuation:	(3) Weak, (5) Medium, (7) Strong
11	Leaf blade, blistering:	(1) Absent, (2) Present
12	Leaf prickliness:	(1) Absent, (3) Weak, (5) Medium, (7) Strong, (9) Very strong
13	Leaf color:	(1) Green, (2) Bluish green, (3) Violet green
14	Hypocotyl, anthocyanin coloration:	(1) Absent, (2) Present
15	Hypocotyl, intensity of anthocyanin coloration	(3) Weak, (5) Medium, (7) Strong
16	Flower, intensity of purple color	(3) Light, (5) Medium, (7) Dark
17	Fruit, general shape:	(1) Pear, (2) Ovoid, (3) Globular, (4) Cylindrical
18	Fruit, apex shape:	(3) Pointed, (5) Semi-pointed, (7) Round
19	Fruit curvature:	(3) Absent, (5) Medium, (7) Strong
20	Fruit, skin color at harvest maturity:	(3) Lilac, (5) Purple, (7) Black
21	Fruit, stripes:	(1) Present, (2) Absent
22	Fruit, intensity of stripes:	(3) Sparce, (5) Medium, (7) Dense
23	Fruit, venation:	(1) Absent, (2) Present
24	Fruit, intensity of anthocyanin coloration of calyx:	(3) Weak, (5) Medium, (7) Strong
25	Fruit, flesh color:	(1) Whitish, (2) Greenish
26	Fruit, seediness:	(3) Weak, (5) medium, (7) Strong
27	Fruit length	The average measurement of ten fruits
28	Fruit diameter	The average measurement of ten fruits
29	Fruit weight	The average measurement of ten fruits
30	Fruit flesh firmness	The average measurement of ten fruits

Molecular characterization

Ten ISSR primers and ten SRAP primer combinations were selected to detect polymorphisms and identify genetic relationships of the eggplant genotypes (Table 3 and Table 4). DNA extractions were performed from young leaves in accordance with the modified Doyle and Doyle (1990) method by using CTAB protocol. ISSR analysis was performed according to Demir et al. (2010). In ISSR PCR process, for 25 µl optimum PCR mixture in each tube, as stock solution, 20 ng DNA, 50 mM 10X buffer, 25 mM MgCl2, 1.25 mM dNTP, 20 µM primer and 5U/ µl Taq DNA polymerase were used. In Cycler (BIO-RAD) Thermal device, samples were subjected to one cycle at 94 °C for 3 minutes, 35 cycles at 94 °C for seconds, 38- 59 °C for 45 seconds, 72 °C for 1 minute, followed by 72 °C for 10 minutes, program was terminated at 4 °C.

PCR reaction for SRAP was performed in 15 µl volume containing 1.5 µl templates DNA, 7.5 µl PCR master mix (2x), 1.5 µl diluted primer and 3.5 µl nuclease-free water. The polymerase chain reactions (PCR) was carried out with the following thermal profile: an initial denaturalization at 94 °C for 5 min; followed by five cycles of 1 min at 94 °C (denaturation), 1 min at 35 °C (annealing), and 1 min at 72 °C (extension); followed by 35 cycles of 1 min at 94 °C (denaturation), 1 min at 50 °C (annealing), and 1 min at 72 °C (extension); followed by a final extension at 72 °C for 10 min. Following the PCR procedure, amplified DNAs were run in 2% agarose gel (containing Ethidium bromide) with the use of 1X TBE buffer at 120for 120 minutes. Then, amplified bands were visualized and imaged under UV light.

Table 3. ISSR primers used in molecular characterization and their expressions.

Order	Primer name	Primer expression 5'-3'
1	VHVGTG7	HVVGTGTGTGTGTGTGTG
2	CAC3GC	CACCACCACGC
3	CA6AC	CACACACACACAAC
4	TCC5RY	TCCTCCTCCTCCTCCRY
5	HVHTCC7	HVHTCCTCCTCCTCCTCCTCCTCC
6	GAA6	GAAGAAGAAGAAGAAGAA
7	GACA4	GACAGACAGACAGACA
8	BDBCA7C	BDBCACACACACACACAC
9	AG8T	AGAGAGAGAGAGAGAGT
10	GT8YA	GTGTGTGTGTGTGTGTYA

Table 4. SRAP marker pairs used in molecular characterization and their expressions.

Order	Primer name	Primer expression 5'-3'
1	Em2-Me4	GACTGCGTACGAATTTGC – TGAGTCCAAACCGGACC
2	Em2-Me5	GACTGCGTACGAATTTGC – TGAGTCCAAACCGGAAG
3	Em2-Me11	GACTGCGTACGAATTTGC – TGAGTCCAAACCGGAAC
4	Em2-Me12	GACTGCGTACGAATTTGC – TGAGTCCAAACCGGAGA
5	Em3-Me2	GACTGCGTACGAATTGAC – TGAGTCCAAACCGGAGC
6	Em6-Me2	GACTGCGTACGAATTGCA – TGAGTCCAAACCGGAGC
7	Em6-Me4	GACTGCGTACGAATTGCA – TGAGTCCAAACCGGACC
8	Em8-Me1	GACTGCGTACGAATTCAC – TGAGTCCAAACCGGATA
9	Em8-Me2	GACTGCGTACGAATTCAC – TGAGTCCAAACCGGAGC
10	Em14-Me4	GACTGCGTACGAATTCTT – TGAGTCCAAACCGGACC

Statistical analysis

Values were assigned to the characteristics investigated for morphological characterization as defined above (Table 2). In molecular characterizations, all gel images were scored as (1) for the existence of bands and (0) for the absence of bands. Data from each marker system were analyzed and assessed separately. Resultant data were analyzed fusing NTSYS (Numerical Taxonomy Multivariate Analysis System, NTSYS-pc version 2.11, Exeter Software, Setauket, N.Y., USA, Rohlf, 2000) software. The similarity index was calculated by the use of DICE (1945) method and a dendrogram was generated byUPGMA (Unweighted Pair-Group Method with Arithmetic Average) method. UPGMA method was proved to be more successful in the identification of the relationships among the genotypes than the other methods.

The following equation was used to determine the polymorphism rate of the primers:

Polymorphism ratio = $\frac{Number\ of\ polymorphic\ bands}{\text{Total number of bands}}$x100

Table 5. Descriptor scale and feature means, standard deviations and Variation Coefficient of eggplant accessions used in the study.

Descriptor	Scale	Mean	Standard Deviation	Variation Coefficient (%)
Plant growth habit	Erect, semi-erect, horizontal (1-3-5)	2.375	1.385	58.33
Plant Height	Short, Medium, Tall (1-3-5)	2.250	1.414	62.85
Plant Stem Length	Short, Medium, Long (1-3-5)	2.125	1.237	58.25
Stem, anthocyanin coloration	Absent, Present (1-3)	4.750	0.983	20.71
Stem, intensity of anthocyanin coloration	Light, Medium, Strong (1-3-5)	4.437	1.162	26.19
Stem, pubescence	Weak, Medium, Strong (1-3-5)	1.937	1.014	52.34
Branch, internode distance	Short, Medium, Long (1-3-5)	2.375	1.385	58.33
Leaf blade size	Small, Medium, Large (1-3-5)	2.125	1.338	62.97
Leaf blade margin shape	Whole, Serrated, Wavy (1-3-5)	1.375	1.184	86.15
Leaf blade margin situation	Weak, Medium, Strong (1-3-5)	2.406	1.562	64.95
Leaf Color	Green, Bluish Green, Violet Green (1-3-5)	1.500	1.344	89.60
Hypocotyl, anthocyanin coloration	Absent, Present (1-3)	4.750	0.983	20.71
Hypocotyl, intensity of anthocyanin coloration	Weak, Medium, Strong (1-3-5)	2.6870	0.859	31.96
Fruit Length	Value	129.2740	33.271	25.74
Fruit Diameter	Value	50.3610	4.685	9.30
Fruit general shape	Pear, Ovoid, Globular, Cylindrical (1-3-5)	2.125	1.827	85.99
Fruit apex shape	Pointed, Semi-Pointed, Round (1-3-5)	2.625	1.288	49.10
Fruit, skin color at harvest maturity	Lilac, Purple, Black (1-3-5)	2.1870	1.330	60.82
Fruit stripes	No, Yes (1-3-5)	4.625	1.184	25.61
Fruit intensity of stripes	Sparce, Medium, Dense (1-3-5)	2.9060	1.593	54.83
Fruit, intensity of anthocyanin coloration of calyx	Weak, Medium, Strong (1-3-5)	1,00	0	0.00
Fruit flesh color	Light, Medium, Much (1-3-5)	1.375	1.184	86.15
Fruit weight	Value	136.7610	34.376	25.14
Fruit flesh firmness	Value	4.021	0.634	15.79
Fruit seediness	Pointed, Semi-Pointed, Round (1-3-5)	2.1870	1.7494	79.97

RESULTS AND DISCUSSION

Results on plant characteristics

Plant characteristics of the genotypes used in this study are provided in (Table 5). In terms of plant growth habit, 4 genotypes had erected, 14 genotypes had semi-erect and 14 genotypes had horizontal growth. In terms of plant height, 16 genotypes were identified as short, 12 genotypes as medium height and 4 genotypes as tall. In terms of plant stem length, 16 genotypes had short, 14 genotypes had medium and 2 genotypes had a long stem. Stem anthocyanin coloration was not observed in 2 genotypes and was observed in 30 genotypes. Of these 30 genotypes, 25 had dense anthocyanin coloration and 5 genotypes had weak coloration. In terms of stem pubescence, 15 genotypes had medium and 17 genotypes had weak pubescence.

Internode distance was short in 14 genotypes, long in 4 genotypes and medium in 14 genotypes. Of present genotypes, 3 had large leaves, 17 genotypes had small leaves and 12 genotypes had medium-sized leaves. Leaf margins had a serrated structure in 3 genotypes and a wavy structure in the rest. Leaf blade margin sinuation was strong in 5 genotypes, weak in 10 genotypes and medium in 14 genotypes. Leaf blistering and prickliness were not observed in any genotypes. In terms of leaf color, 4 genotypes had green and the rest had bluish-green color. Hypocotyl anthocyanin coloration was not observed only in 2 genotypes. In coloration-observed genotypes, 2 had weak and the rest had medium coloration. In terms of flower intensity of purple color, 28 genotypes had light purple and 4 genotypes had medium purple color.

Results on fruit characteristics

Fruit characteristics of the genotypes are provided in (Table 5) and (Figure 2). Fruit lengths varied between 79.44 - 243.77 mm and fruit diameters varied between 38.01 - 59.67 mm. In terms of fruit shape, all genotypes had cylindrical fruit. In terms of fruit apex shape, 23 genotypes had pointed and 9 genotypes had oval apex shapes. While fruit curvature was not observed in 10 genotypes, medium curvature was observed in 18 genotypes and strong curvature was observed in 4 genotypes. In terms of fruit skin color at harvest maturity, 3 genotypes had blocked, 16 genotypes had lilac and 13 genotypes had purple color. Of the present genotypes, 3 did not have stripes on fruit surface and the rest had stripped structure. Of stripped genotypes, 16 had medium density, 5 genotypes had sparce and 8 genotypes had dense stripes. Fruit venation was not observed in any genotypes. In terms of anthocyanin coloration of the calyx, all genotypes had weak coloration. In terms of fruit flesh color, two colors were dominant. While 3 genotypes had greenish flesh color, the rest had whitish flesh color. Fruit weights of the genotypes varied between 68.08 - 211.26 g and fruit flesh firmness values varied between 2.96 - 5.52 kg/cm2. Fruit seediness was weak in 21 genotypes, strong in 8 genotypes and medium in 3 genotypes.

Figure 2. Fruit characteristics of the Yamula genotypes

Analysis of morphological characterization data and assessment of dendrograms

A total of 25 characters selected from the UPOV description list were used for the characterization of accessions (Table 5). While no variation was observed in “anthocyanin coloration of calyx” character, the highest variation coefficient was calculated for “leaf color” and “leaf blade margin shape” (89.60% and 86.15%, respectively). When all plant parts were divided into groups, it was understood that the highest variation occurred in leaf characteristics (Variation coefficient: 59.39%). This was followed by the plant stem characters and fruit characteristics (48.14% and 43.20%, respectively).

Cluster analysis was conducted for similarities among the genotypes using the DICE coefficient, and a dendrogram representing the relationships among the genotypes was obtained using these coefficients. The cluster analysis grouped 32 eggplants accessions into 2 main groups using 25 morphological characters (Figure 3). The first main group (Cluster-I) consisted of four accessions: ERÜ-3014, ERÜ-3016, ERÜ-3005, ERÜ-3015. The similarity coefficient was measured as 0.710. Group-II included 27 accessions. This group was divided into 2 sub-groups. The largest sub-group is Group-II / A and consists of 20 accessions. The accessions ERÜ-3006 and ERÜ-954 within this sub-group had the highest morphological similarity coefficient (0.960).

The projections of 32 accessions and 25 characters in a 2-D graph are presented in (Figure 4). The first (PC1) and the second (PC2) principal components explained 48.107% of the total variation. Principal component analysis revealed that 7 PCs, and seven principal components (PC1 to PC7) with Eigenvalues of >1 accounted for 84.29 % of the total variation in eggplant accessions. The value of four characters in the first

principal component was above |0,3|. This component (PC1) had a high value (|-0. 326|) for hypocotyl anthocyanin coloration intensity, fruit stripes (|0. 310|), fruit flesh color (|0. 310|).

The genetic distance between eggplant accessions varied between 0.479 - 0.977 with an average value of 0.809. The highest genetic similarity coefficient (0.977) was found between accessions ERÜ-3006 and ERÜ-3009. The lowest coefficient was obtained between ERÜ-3008 and ERÜ-954. Based on the UPGMA cluster analysis, two main clusters were obtained (Figure 3). Clusters of studied accessions did not reveal any specific features based on any morphological character.

Topcu (2014) conducted morphological and molecular characterization of 100 eggplant lines. Observations and measurements were performed for 32 characteristics in morphological characterizations. Genotypes were divided into 17 groups. In that study, 53 lines had purple, 34 lines had dark purple and 13 lines had light purple flower color. In terms of leaf color, 73 lines had green, 13 lines had light green and 14 lines had dark green color. Observations revealed 6 different fruit shapes and 8 different fruit colors. In terms of fruit shape, 32 lines had long, 31 lines had medium, 15 lines had short, 4 lines had oval, 8 lines had pear-shaped and 10 lines had round fruit shapes. In terms of fruit color, 2 lines had pink, 9 lines had light purple, 56 lines had purple, 27 lines had black, 2 lines had striped pink, 1 line had green, 2 lines had white and 1 line had greenish purple fruit color. In terms of fruit flesh color, 18 lines had greenish, 49 lines had greenish cream, 3 lines had white, 14 lines had white cream, 3 lines had cream and 13 lines had cream flesh color. The present findings comply with the findings of that study. Differences between the two studies were mainly attributed to differences in cultivars used.

Boyaci et al. (2015) used a total of 38 eggplant genotypes, of which 32 were heirloom accessions collected from different regions of Burdur province and five were different local genotypes from the other provinces, and one was a cultivar used as a reference, to determine genetic variations among the genotypes. The phylogenetic relationships among these heirlooms were evaluated using 40 morphologic descriptors. Burdur heirloom accessions showed high genetic diversity based on morphological and molecular data. It was stated that the genetic similarity rates ranged from 0.29 to 0.91 according to the morphological data.

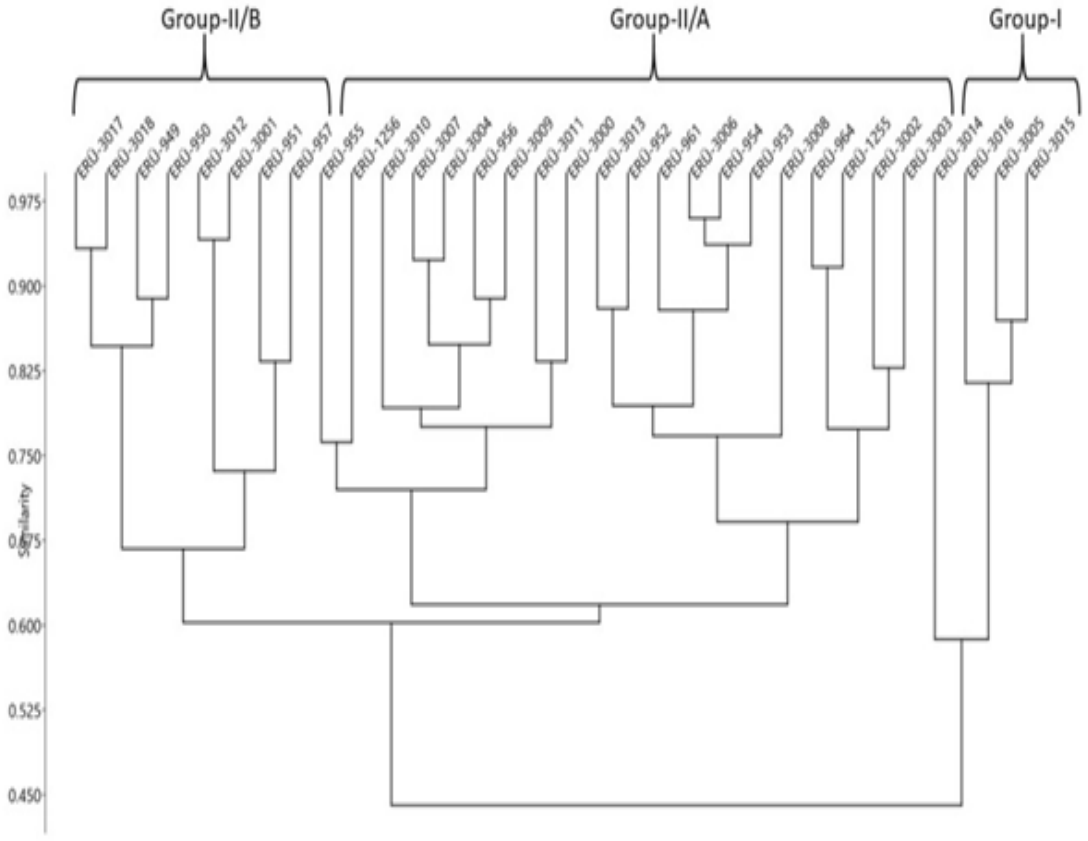

Figure 3. Cluster analysis for plant and fruit characteristics of eggplant genotypes used in the present study.

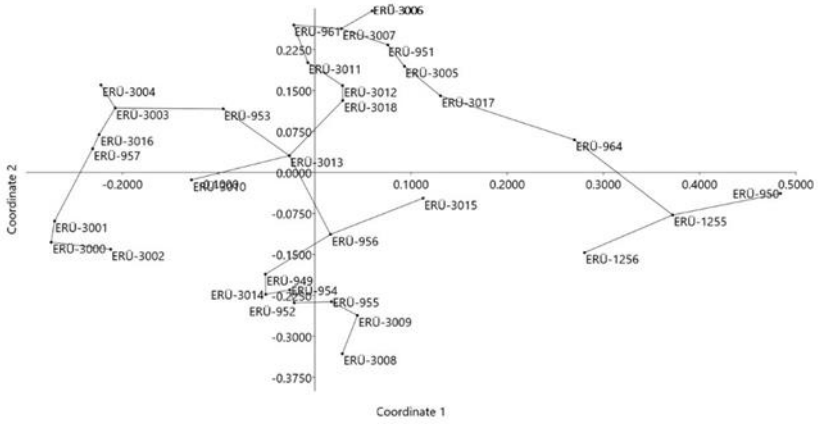

Figure 4. Distribution of eggplant genotypes based on the first and the second components.

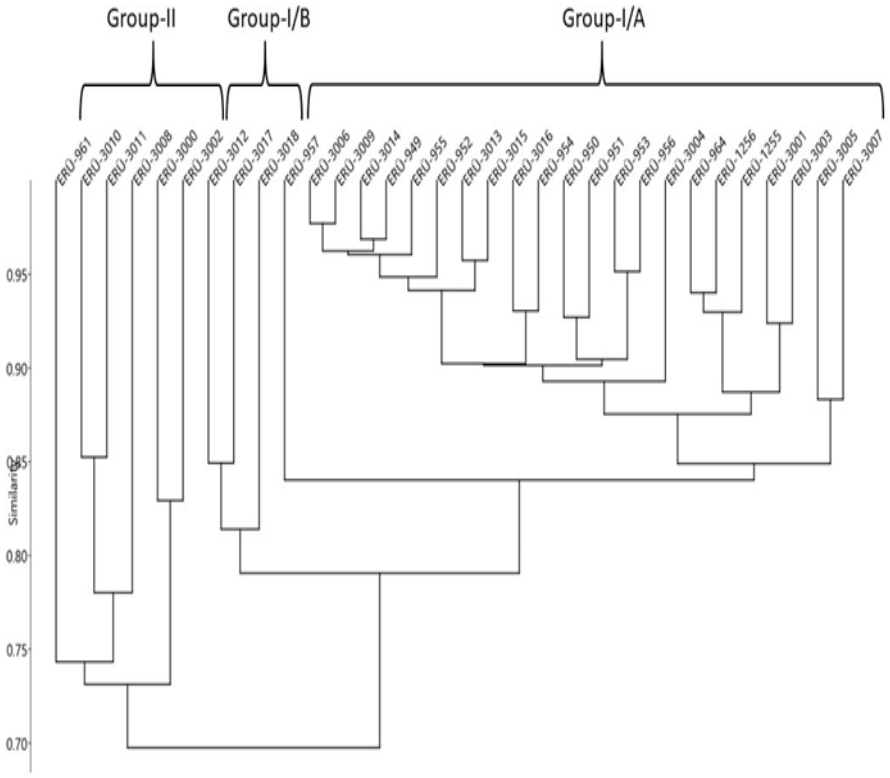

Figure 5. Cluster analysis for plant and fruit characteristics of eggplant genotypes used in the present study.

Molecular characterization results

ISSR Marker analysis

In ISSR analysis conducted to determine genetic relationships among the genotypes, 16 primers were used. Of these primers, 10 yielded a band image. Polymorphic band lengths varied between 190 - 1100 bp. Totally 72 bands were obtained from the primers and the number of bands per primer was 7.2. Of these bands, 56 were polymorphic and the number of polymorphic bands per primer was 5.6. The greatest polymorphism ratio (100%) was observed in CAC3GC and the lowest (60%) in TCC5RY primers (Table 6).

According to the dendrogram generated as a result of ISSR marker analysis, genetic similarity among the genotypes varied between 0.66 - 0.99. There were two main groups in the dendrogram. Both groups had a different number of sub-groups. Genotypes 5, 29, 31 (ERÜ-3008, ERÜ-3000, ERÜ-3002) were placed into the first group and the rest were placed into the second group. The closest genotypes were identified as 3-6 (ERÜ 3006-ERÜ 3009), 17-18 (ERÜ 950-ERÜ 951) and 22-23 (ERÜ 955-ERÜ 956) (Figure 5).

Table 6. Polymorphism table obtained after amplification of ISSR primers.

Primer name	Primer band length	Total number of bands	Number of polymorphic bands	Polymorphism ratio (%)
VHVGTG7	250-1000	11	8	72.72
CAC3GC	300-900	7	7	100.00
CA6AC	400-950	7	5	71.42
TCC5RY	500-1000	5	3	60.00
HVHTCC7	350-1100	7	5	71.42
GAA6	190-900	9	7	77.77
GACA4	320-1100	8	7	87.50
BDBCA7C	290-850	9	7	77.77
AG8T	500-1000	4	3	75.00
GT8YA	310-750	5	4	80.00
Total	-	**72**	**56**	-
Mean	-	**7.2**	**5.6**	**77.36**

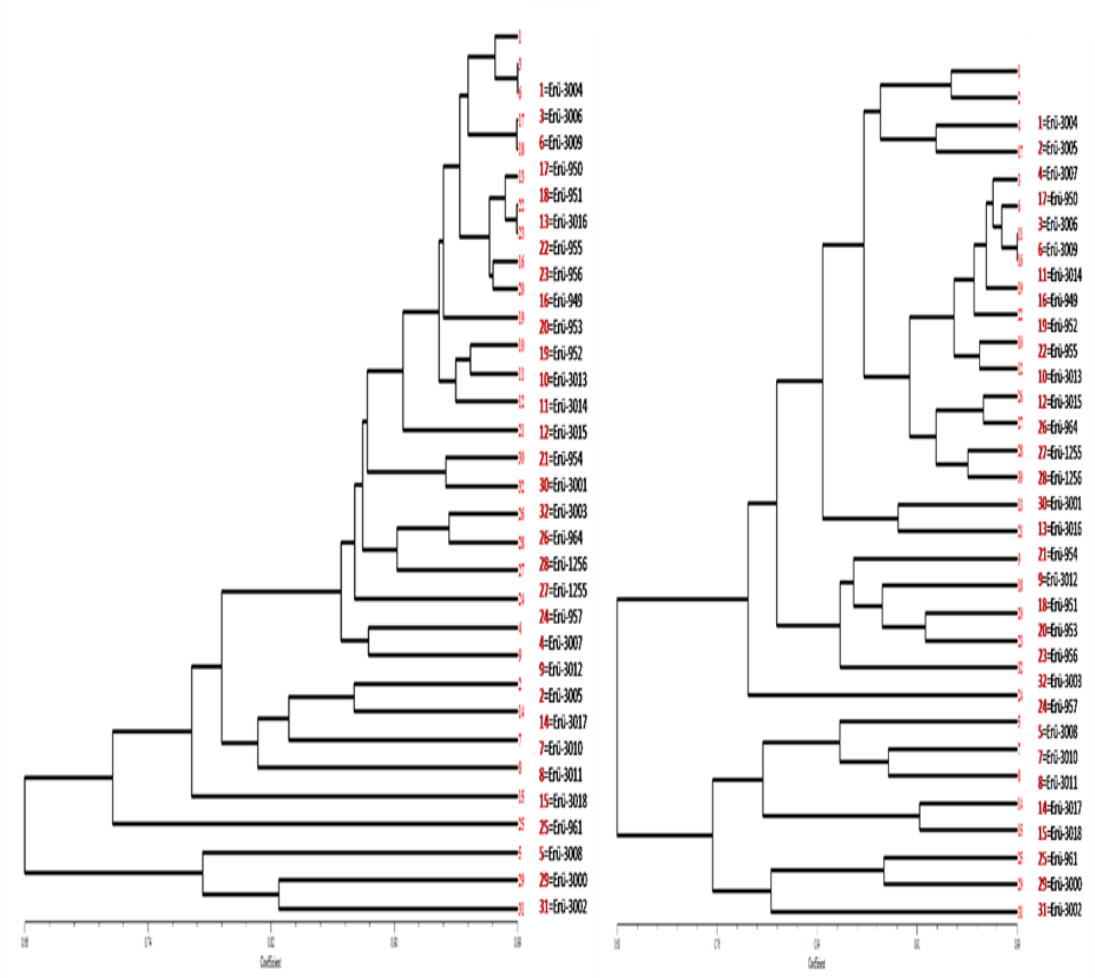

Fig. 5. Separate Cluster analysis for ISSR and SRAP marker data of eggplant genotypes used in present study.

SRAP Marker analysis

In SRAP marker analysis, 32 primers were used and 10 of these primers were studied. Polymorphic band lengths varied between 100 - 1200 bp. Totally 67 bands were obtained from these primers and number of bands per primer was 6.7. Of these bands, 51 were polymorphic and number of polymorphic bands per primer was 5.1. Polymorphism ratio was calculated as 73.72% (Table 7).

According to SRAP marker analysis, genetic similarity among the genotypes varied between 0.68 - 0.99. Resultant dendrogram had two main groups. Main groups had the different number of sub-groups. The genotypes 5, 7, 8, 14, 15, 25, 29, 31 (ERÜ-3008, ERÜ-3010, ERÜ-3011, ERÜ-3017, ERÜ-3018, ERÜ-961, ERÜ-3000, ERÜ-3002) were placed into the first group and the rest were gathered into the second group. The closest genotypes were 11-16 (ERÜ 3014-ERÜ 949).

To examine the genetic relationships among different eggplant accessions, a dendrogram was generated using molecular and morphologic data together (Figure 6). Average Dice's similarity coefficient of eggplant accessions was 0,789 based on molecular and morphological data. Eggplant accessions were clustered into two main groups. The first group included 28 accessions. The second group included only four accessions: ERÜ-3000, ERÜ-3002, ERÜ-3008, and ERÜ-961. Average Dice's similarity coefficient of this group was lower than all clusters (0,733). These accessions are located together in cluster analysis using only molecular data.

Eggplant has a wide genetic diversity in the regions where it is cultivated, although they are not native to the region (Muñoz-Falcón et al., 2008). Although Turkey is not within the center of origin of eggplant, wide genetic diversity has been reported in Turkey (Demir et al., 2010; Tumbilen et al., 2011a, 2011b). It is clear in this study that Burdur, which is a small geographical region, had a rich genetic diversity. Also, wide genetic variability was determined in both Spain and Jordan local genotypes (Prohens et al., 2003). Local genotypes can contribute to enhance the gene pool used in breeding studies and to help increase heterosis (Muñoz-Falcón et al., 2009). In recent years, some factors like the cultivation of commercial varieties instead of heirlooms, the construction of buildings on agricultural lands, and innovation in cultivation methods have led to the erosion of plant genetic resources (Cericola et al., 2013). Therefore, there is a need for collection and identification of local heirlooms before they disappear (Muñoz-Falcón et al., 2008). Consistent with previous works, a higher diversity for most morphological descriptors was recorded in the collection of Kayseri local heirlooms identified in this study. Fruit color can be cream, green, red, reddish-purple, dark purple or black.

Molecular markers linked with agronomic traits are useful tools for marker-assisted selection and mapping of candidate genes in breeding programs (Nunome et al., 2009; Wang et al., 2010). Ali et al. (2011) investigated plant diversity in Chinese-originated eggplants with the use of ISSR and RAPD techniques. Researchers indicated that RAPD markers were more efficient in the identification of genetic diversity than the ISSR markers. Totally 143 eggplant genotypes were analyzed and two main groups and 8 sub-groups were obtained. Tiwari et al. (2009) worked on 19 cultivars and local genotypes with the use of 29 RAPD primers and found 2 of them sufficient in the separation of the cultivars. Researchers reported that 27.5% and 18.73% of RAPD ISSR markers, respectively, were polymorphic. Of RAPD primers, OPW 11 and OPX 07 were found to be sufficient in the separation of 19 cultivars.

Genetic relationships among 10 eggplant genotypes (*Solanum melongena* L.) were studied using ISSR molecular markers. Seven out of 20 ISSR primers were used to assay the levels of polymorphism among the Egyptian cultivars of eggplant (*Solanum melongena* L). In that study, some variations in banding patterns were observed among the 10 genotypes where there were 24 monomorphic and 47 polymorphic distinct fragments (61% of polymorphism). The results of genetic relationships showed that the genotypes were divided into two main groups. ISSR analysis and dendrograms demonstrated the relationships and revealed six cultivars (S3, S6, Moshtouhr, PIG-4, Jo=3, Black beauty) as the best ones which have a broad genetic background and must be introduced into breeding programs to produce hybrid seeds of eggplant (Mahmoud and El-Mansy, 2012).

Li et al. (2010) analyzed 56 Solanum species with the use of 55 SRAP primer combinations. Researchers obtained 635 polymorphic bands. Cluster analysis revealed 3 main groups. Genetic similarity values varied between 0.04 – 0.96 and the similarity coefficient between S. melongena accessions was calculated as 0.78. These findings revealed that SRAP could efficiently be used in the estimation of genetic diversity and to analyze phylogenetic relations among the genotypes.

The local populations are of great importance for the breeders so that they adapted well to their cultivated areas. There is a need for the collection and identification of these heirlooms before integrated into the breeding programs. In the present study, genetic variation in Yamula eggplant, grown in Kayseri province and surroundings and prominent with firm fruit flesh, was investigated to reintroduce the cultivar into the region and to prevent extinction. A high genetic diversity was determined among the present eggplant genotypes. Such differences were mainly attributed to the seed supply of growers since they mostly use their seeds. These materials can be of potential value for the breeders. It was also concluded based on present findings that both ISSR and SRAP techniques could successfully be used in genetic diversity studies.

Table 7. Polymorphism table obtained after amplification of SRAP primers.

Primer name	Primer band length	Total number of bands	Number of polymorphic bands	Polymorphism ratio (%)
Em2-Me4	120-1000	12	10	83.33
Em2-Me5	200-700	6	4	66.66
Em2-Me11	190-650	7	6	85.71
Em2-Me12	150-900	8	6	75.00
Em6-Me2	160-1200	8	7	87.50
Em6-Me4	180-500	6	4	66.66
Em14-Me4	210-900	5	3	60.00
Em3-Me2	100-500	3	2	66.66
Em8-Me1	150-610	7	6	85.71
Em8-Me2	150-610	5	3	60.00
Total	**-**	**67**	**51**	**-**
Mean	**-**	**6.7**	**5.1**	**73.72**

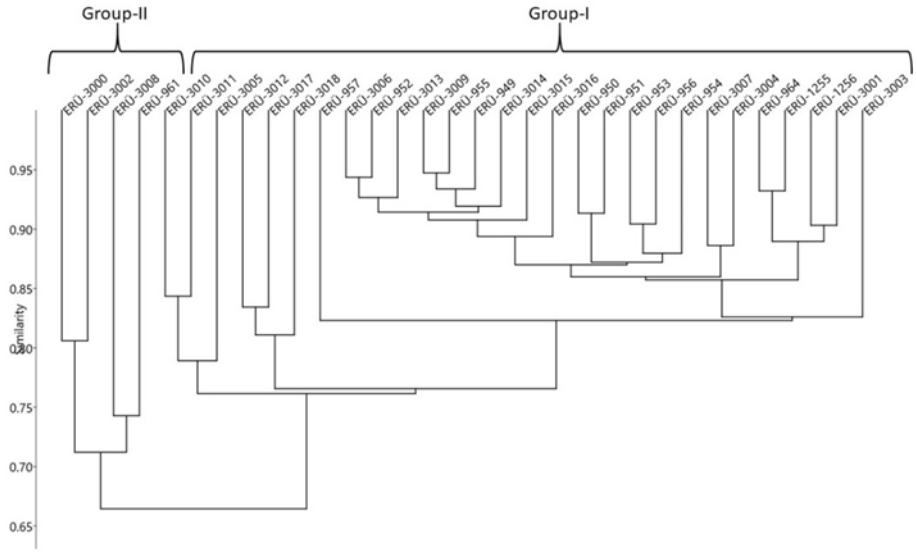

Fig. 6. Combined Cluster analysis for ISSR and SRAP marker data of eggplant genotypes used in the present study

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