

MOLECULAR CHARACTERIZATION OF SOME ELAZIG PEPPER (*CAPSICUM ANNUUM* L.) GENOTYPES

Hasan Pinar ^{1*}, Zeliha Duruk ¹, Aydın Uzun ¹, Emre Seker ²

¹ Erciyes University, Department of Horticulture, Kayseri, Turkiye

² United Genetics Seed Company, Bursa, Turkiye



Abstract

Pepper is widely produced and consumed in the world, but facing some threats such as loss of genetic resources. For this reason, it is very important to protect genetic resources including pepper genetic materials and try to include them into breeding programs at the same time. One of these genetic resources is the Elazig pepper, which is grown as standard in and around the province of Elazig, located in the Northeastern region of Turkey for many years and is consumed fondly by the people of the region. However, when the production areas are visited, it is observed that there are segregation due to open pollination in this variety. In this study, it was aimed to conduct the molecular characterization of some Elazig native peppers belonging to the *Capsicum annuum* species. For molecular characterization, 9 POX markers were used and total 105 band obtained using POX markers and 60 of them were polymorphic. According to the data obtained, quite important polymorphism was obtained as a result of molecular characterization in some Elazig peppers. This polymorphism shows that Elazig local pepper can be used in terms of breeding for breeding programs.

Keywords: *Capsicum annuum*, Elazig pepper, molecular characterization

1. INTRODUCTION

There are regions in Turkey with quite different characteristics in terms of climate and soil. Turkey is located in the geography where one of the eight main plant gene centers in the world is located, and is located in one of the regions where agriculture was first practiced in the world. Therefore, the geography covering Anatolia is known as the diversity center and micro gene center of many cultivated plant species. However, a high degree of plant endemism has emerged. Therefore, Anatolian geography contains around 10,000 species and about 1/3 of them are endemic. However, as in the whole world, plant genetic resources in this geography are in danger of extinction for various reasons. Conservation of diversity in plant genetic resources, especially of cultivated species, is very important for the sustainability of plant production (Tan and İnal, 2003). As reported by Tan (1992); ever-increasing land openings, the replacement of improved uniform varieties, the native varieties in the form of population, natural disasters such as fire and erosion, the construction of dams etc. for the development of the country, the construction of facilities, urbanization and applications in the areas of development, the change of agricultural systems and agricultural struggle applications, without production Due to reasons such as consumption by constantly removing from nature, herbal diversity is decreasing, however it is disappearing.

Realizing this danger, many countries have started studies for the detection, protection and storage of herbal resources. Later, the collection and characterization of genetic resources, especially in cultivated species with high economic added value, became a priority. Pepper is one of the important species cultivated in Turkey and also has an important place in world production.

Şalk et al., (2008) stated that the homeland of pepper is the tropical and subtropical countries of America, the primary gene center of *Capsicum annuum* is Mexico and the secondary gene center is Guatemala, *C. chinense* and *C. frutescens* are Amazon basin, *C. pendulum* and states that *C. pupescens* is the primary gene center of Peru and Bolivia, *C. annuum* and *C. frutescens* are common in all the Americas and Carib islands from Mexico, and the most commonly grown species in South America is *C. chinense*. Today, it is known that *C. annuum* is the most widely grown species in the world in ecologies suitable for pepper production, but other species are produced, albeit to a limited extent, in their homeland, especially in regions with subtropical and tropical climates.

The identification, collection and protection of agricultural biodiversity is of vital importance for the sustainability of plant diversity (Tan et al., 2004). National plant genetic resources have been enriched day by day with the addition of genetic materials brought from other countries to the populations that have emerged as a result of natural pollination seen in vegetable cultivation in Turkey for many years and selections made by people in terms of features of economic importance. Great agricultural biodiversity is reported in terms of important vegetable species, among which Turkey is homeland. However, this biodiversity is subject to genetic erosion for various reasons. One of these species is the pepper, which is consumed locally in different regions of Turkey with love by the people of the region and is among the vegetable species with significant genetic diversity. Pepper is grown in wide areas in every region of Turkey, and it is among the important species with commercial potential in terms of export by being processed in the industry or for table consumption as well as for fresh consumption. Pepper used in the food industry; It is evaluated in different ways in tomato paste, powder-chilli powder, frozen, pickles, hot sauce, ketchup, paint and pharmaceutical industry (Aybak, 2002). Pepper, which has great variation in terms of morphological-agronomic characteristics of fruit and plant, is consumed in different ways according to fruit structure and shape (Bozokalfa and Eşiyok, 2006). Selections and open pollination applied to the local populations grown by the producers in terms of consumer demands and yield, in the pepper grown one-year-old in Turkey conditions, caused variations in plant and fruit structure, thus increasing the number of genotypes in the country and plant genetic resources day by day. Today, the variety dynamics in vegetable species are quite high in line with consumer preferences. A large number of varieties are developed every year in the main vegetable species such as tomatoes, peppers and eggplants. The first step of variety breeding is the richness of plant genetic resources. Plant genetic resources consist of local genotypes, their wild relatives, old cultivars and lines with well-defined genetic characteristics. Due to its importance in plant breeding and new variety development studies, it is of great importance to define the local populations and breeding lines of the investigated plant species both at the morphological and molecular level and to reveal the degree of relatedness based on these data, especially in terms of hybrid variety development. Turkey is very rich in terms of local pepper genetic resources, most of it is produced commercially, it has export potential as well as being consumed regionally. However, some pepper genotypes cause losses in productivity and quality over time, as the producers obtain their own seeds from open pollinated production areas for many years and continue the production in this way.

Elazig pepper is the local pepper variety, which has been grown locally in the province of Elazig and surrounding provinces for many years and is consumed commonly by the people of the region.

However, when the production fields were observed, it is seen that there are wide variations because of open pollination feature of this variety. From this point of view, the molecular characterization of 26 pepper genotypes collected from Elazig province was aimed in this study.

2. MATERIALS AND METHODS

In this study, 26 pepper genotypes collected from the province of Elazig in 2020 and Pin Yavuz Acı (K1), Yalova Corbaci (K2), Sera Demre (K3), Sweet Kandil Dolma (K4), which are grown in different regions in our country as a standard, were used in this study. A total of 32 pepper genotypes, Cırgalan (K5) and Şanlıuفا pepper (K6), were used as material. The data obtained as a result of the molecular analyzes were evaluated and NTSYS (Numerical Taxonomy Multivariate Analysis System) 2.1 computer package program was used for the analysis of the obtained data., The level of genetic relationship was determined by the UPGMA (Unweighted Pair Group Method Arithmetic Average) grouping of the similarity matrix obtained according to the Rohlf method. (Rohlf, 1998), Similarity indexes were calculated according to Dice. (Dice, 1945).

3. RESULTS AND DISCUSSIONS

In present study, 21 pepper genotypes collected from Elazig province and a total of 32 pepper genotypes, namely Pin Yavuz Acı (K1), Sweet Kandil Dolma (K4) and Şanlıuفا pepper (K6) grown in different regions in Turkey as standard, were used as material and 9 POX marker combinations were used. Molecular characterization was performed using a combination of markers. A total of 105 scoreable bands were obtained from 9 POX marker combinations, and 60 of these bands were determined as polymorphic. The mean polymorphism was 57%. The highest number of bands was obtained from primer combinations POX6F-POX6R, POX8F-POX8R, POX10Fa-POX10C, POX10Fa-POX10Rd and POX12Fa-POX12Ra (12 pieces), while the lowest number of bands was obtained from primer combination POX1F-POX1R. The highest polymorphism POX1F-POX1R (88%) was obtained, while the lowest popliorphism POX10Fa-POX10Rd (17%) was obtained (Table 1).

Table 1. Total, polymorphic number of monomorphic bands and polymorphism ratios as result of amplification of POX primers (%)

Marker combination	Base	Total	Polymorphic	Monomorphic	% Polymorphism
POX1F-POX1R	100-580	8	7	1	88
POX5R-POX5R	100-610	10	2	8	20
POX6F-POX6R	100-790	12	7	5	58
POX8F-POX8R	100-690	12	8	4	67
POX10F-POX10R	100-1350	16	10	6	63
POX10F-POX10C	100-950	12	11	1	92
POX10F-POX10R	100-870	12	2	10	17
POX11F-POX11R	100-850	11	4	7	36
POX12F-POX12R	100-760	12	9	3	75
	Total	105	60	45	57

The dendrogram obtained as a result of the characterization study performed on 24 pepper genotypes with 9 POX marker combinations is given in Figure 1. According to the dendrogram obtained, 2 main groups were formed and Elazig pepper genotypes E1 and E9 were in the 1st main group, while the control and other Elazig pepper genotypes were in the 2nd main group. In addition, Pin Yavuz bitter (K1) and Sweet Kandil Stuffed (K4) control genotypes were included in the subgroup of the 2nd main group, apart from the Elazig pepper genotypes. On the other hand, while genetic similarity was determined between 0.81-0.97 in dendrogram, genotypes E20 and E21 were determined to be similar to each other.

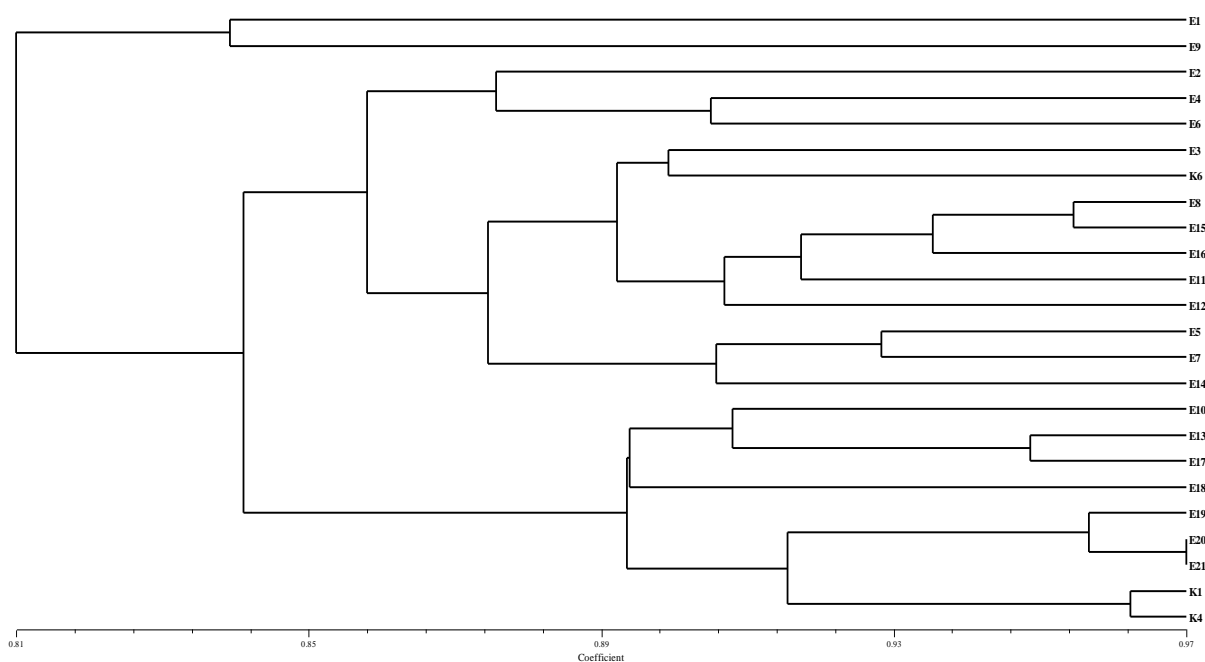


Figure 1. Dendrogram from POX molecular markers characterization using Elazig pepper genotypes

As reported by Alan (1984), a total of 176 different pepper populations were collected from the provinces covering the Southeast, Northwest and Northeast regions as a result of the genetic resources collection studies carried out between 1978-1984. In the characterization studies on these materials, a grouping was formed in terms of fruit shape, fruit size, fruit position, ripe fruit color, fruit bitterness characteristics, and the main collection of pepper genetic resources that pepper breeders can use in breeding programs was created. On the other hand, morphological characterization of 116 hot pepper types in the Sudan Agricultural Research Center (ARC) pepper germplasm collection has been reported. In this study, plant posture, stem characteristics, fruit position, fruit shape and size, fruit color and bitterness were investigated. More than 75% of the investigated pepper genotypes had green stem colour, 50% had conical fruit shapes, 39% had pointed shaped, 92% fruit lengths ranged from 1.0 to 7.5 cm. On the other hand, it has been reported that the majority of the types are green in color, and only a few of them are light or dark green. Besides, it has been observed that the ripe fruit color is distributed in dark red, magenta red, red and orange color tones. Fruit bitterness was determined as slightly bitter, bitter and high bitter. (El Tahir, 1994).

In recent years, local pepper populations have been collected and characterized in Turkey as well as in the world and included in both production and breeding programs. In a study conducted by

Başak, (2019), characterization studies were carried out in terms of a total of 48 agronomic and morphological characteristics determined for pepper by IPGRI and UPOV on 99 pepper genotypes collected from Kırşehir province and its surrounding villages. According to the morphological and agronomic characteristics, genotypes were divided into 15 groups in the dendrogram. As a result of the cluster analysis; It has been reported that genotypes coded S1, S2, S62, S3, S9, S67 and TR69737 are the most distant from each other in terms of agronomic and morphological characteristics. On the other hand, Karakurt et al. (2020) performed molecular characterization with SSR (Simple Sequence Repeats) markers to determine the genetic diversity of different pepper genotypes and according to their results, pepper genotypes were divided into 2 main groups in the UPGMA dendrogram obtained with SSR markers. While the first main group was divided into two subgroups, the first subgroup included Vezir, Üçburun, Acıburun, Yükselince, Anadolu, Serenad, Haifa Chile, and the second subgroup included Jalomex. On the other hand, Ergenekon and Kanyon genotypes were included in the second main group. Vezir, Üçburun and Yükselince-Anatolia cultivars are similar groups and grouped together by showing similar characteristics in terms of SSR regions studied. In the another study, which was used for the molecular characterization of a total of 30 capia, stuffed, pointed and red pepper varieties, characterization was performed using 6 SSR loci. According to the findings, it was concluded that there is no variation with regard to molecular in pepper varieties in terms of the regions, and that the varieties show similarities between 0.77-1.00 in terms of the regions (Şeker, 2018).

Okay (2019) conducted a study using 35 inbred and 3 commercial pepper varieties for the genetic characterization using 19 SRAP primer combinations. Similarity index values of pepper lines used in this study were reported to vary between 0.35 and 0.97.

On the other hand, it was conducted a study to characterization in terms of 54 morphological features determined by UPOV in 26 different pepper populations collected from Marmara, Black Sea, Eastern Anatolia and Central Anatolia Regions of Turkey and 3 different standard pepper varieties belonging to Aegean Agricultural Research Institute(Binbir and Baş, 2010) According to their results, a wide variation was observed as the populations examined included most of the pepper types.

Kar et al. (1999) characterised to determine the morphological variations of the 'Turkey F1 Hybrid Pointed Pepper' genotypes, a total of 17 characteristics including 5 vegetative traits, 2 leaf traits and 10 fruit traits belonging to 37 genotypes at the Black Sea Agricultural Research Institute. Cluster analysis was applied to the data obtained as a result of the examined features. In the dendrogram formed as a result of the cluster analysis, genotypes were divided into 7 main groups. As a result of the cluster analysis, it was determined that the types 84 and 99, 131 and 172 were quite similar to each other. In addition, it was determined that the types 170-166 and 170-173 were quite different from each other in terms of their morphological features.

In a study conducted by Kanal and Balkaya (2021) on 67 pepper genotypes belonging to the *C. baccatum* species collected from different countries of the Americas, and the morphological characterization of the pepper gene sources of the *C. baccatum* species was determined by the plant cultivar TG/76/8 specified by UPOV for pepper. Characterization was made according to the property document. According to the results, it was reported that the morphological variation value in the pepper population of the *C. baccatum* species was 75.75% according to the principal component analysis. The results show that there is a very high morphological variation among pepper genotypes of *C. baccatum* according to UPOV criteria.

Results of this study show that a wide variation was obtained between the collected Elazig pepper genotypes and the control genotypes in terms of morphological characteristics. When the previous studies mentioned, it is seen that there is variation between pepper types in terms of morphological characteristics, depending on foreign pollination and geography, and it has parallels with the aforementioned study. In addition, the variation determined among the Elazig pepper genotypes may have resulted from open pollination as in the local pepper genotypes grown in home gardens or in limited areas, due to the fact that the producer produces his own seed.

4. CONCLUSIONS

It is seen that wide variation is obtained in the characterization studies of local genotypes in Turkey. The source of this variation may have been due to the fact that the pepper had open pollination as well as self-pollination. According to the data obtained in this study, quite wide variation was obtained as a result of morphological characterization in some Elazig peppers. Elazig pepper is widely used in Elazig and surrounding provinces for drying, table, pickling, etc. purposes. The use of this variety should not be limited to the province of Elazig and surrounding provinces, and its spread to wider ecology should be ensured. Based on the data related to this study, it is thought to contribute to the country's economy in the future. From this point of view, the similarities and differences between these genotypes can be evaluated and the genotypes with appropriate characteristics can be made more quality and efficient in breeding studies. When compared with previous studies, morphological similarities and differences are observed both among themselves and with different cultivars, as in other species and cultivars. The fact that these differences are so much can be said to be open pollination, the production of their own seeds by the villagers or the people who produce in the garden, as in other wild varieties, and the adaptation to the ecological structure of the area.

5. ACKNOWLEDGMENT

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6. REFERENCES

- Alan, M.N. (1984). Collection and evaluation of pepper germplasm in Turkey, *Capsicum and Eggplant Newsletter* 3, 17-18.
- Aybak, H.Ç. (2002). Pepper cultivation, Hasad Publishing 155 s.(In Turkish)
- Başak, H. (2019). Agronomic and Morphological Characterization of Kırşehir Local Green Long Pepper (*Capsicum annuum* L. var. longum) Populations. *KSU J. Agric Nat* 22(2), 202-216, 2019
DOI: 10.18016/ksutarimdogra.vi.488204
- Binbir, S., Tülin, B. (2010). Characterization of Some Local Pepper (*Capsicum annuum* L.) Populations. *Anadolu, J. of AARI* 20 (2) 2010, 70 - 88
- Bozokalfa, M.K., Eşiyok, D. (2006). Homeland and distribution of pepper, Dünya Publishing, Food, Issue 07:92-93, Bagcilar/İstanbul. (In Turkish)
- Dice, L.R. (1945). Measures Of The Amount Of Ecologic Association Between Species, *Ecology*, 26, 297–302.
- El Tahir, I.M. (1994). Collection and Characterization of Hot Pepper Germplasm in Sudan, Horticultural Germplasm Unit, Agricultural Research Corporation, Wad Medani, Sudan, *Capsicum and Eggplant Newsletter*, 13:36-39.
- Kanal, A., Balkaya, A. (2021). The determination of morphological variation level and characterization of *Capsicum baccatum* populations, *Mustafa Kemal University Journal of Agricultural Sciences* 26 (2), 278-291.
- Kar, H., Karağaç, O., Kibar, B., Apaydın, A. (1999). An Investigation on the Collection of Domestic Green Pepper Genotypes in The BlackSea Region of Turkey and Determination of Their Morphological Traits, *Horticulture Congress in Turkey*, <https://www.researchgate.net/publication/44391745>.

- Karaağaç. (2006). Characterization and evaluation of Bafra red pepper gene sources (*Capsicum annuum* var, conoides Mill.) (Master Thesis) *Ondokuz Mayıs University Institute Of Natural And Applied Sciences*. Samsun, Turkey
- Karakurt, Y., Cesur, E., Güvercin, D. (2020). Molecular Characterization of Pepper (*Capsicum annuum* L.) Genotypes Using SSR Markers, *Ege Üniv. Ziraat Fak. Derg.*, 57 (2):185-189 DOI: 10.20289/zfdergi.614237.
- Okay, Ç.O. (2019). Molecular Characterization Of Elite Pepper Breeding Lines For Genetics And Resistance To Some Virus Diseases [Master Thesis]. *Ordu University Institute Of Natural And Applied Sciences*. Ordu, Turkey
- Rohlf, F.J., (1998) NTSYS-PC, Numerical Taxonomy And Multivariate Analysis: Version 2,02, Exeter Software, Setauket, New York.
- Şalk, A.L., Arın, M., Deveci, S., Polat. (2008). Special vegetable growing, Namık Kemal University, Faculty of Agriculture, 448 s., Tekirdağ (In Turkish)
- Şeker, A. (2018). Molecular Characterization of Some Pepper (*Capsicum annuum* L.) Cultivars with SSR Markers, (Master Thesis) *Ankara University Institute Of Natural And Applied Sciences*. Ankara, Turkey.
- Tan, A. (1992). Türkiye’de bitkisel çeşitlilik ve bitki genetik kaynakları, *Anadolu, J, of AARI*, 2 (2), 50-54.
- Tan, A., İnal, A. (2003). Aegean Agricultural Research Institute plant genetic resources studies, Aegean Agricultural Research Institute Directorate Publication Number:112, 13 s, İzmir, Turkey(In Turkish)
- Tan, A., Taşkın, T., İnal, A. (2004), Plant genetic resources studies, Aegean Agricultural Research Institute İzmir, Promotional Brochure No.3. (In Turkish)