

MOLECULAR MARKER BASED ESTIMATION OF APRICOT GENOTYPES COLLECTED FROM YEŞİLHISAR REGION OF KAYSERİ

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Abstract

Apricot is one of the most important stone fruits in the world. It can be evaluated as both fresh consumption and drying. Turkey is the first apricot producing country and has a rich genetic diversity. There are many apricot populations in different regions of Turkey. Some of them contain standard cultivars and some of them consist of local varieties or genotypes. Kayseri province has relatively important local apricot populations. In this study, genetic diversity in apricot populations in Yeşilhisar, Kayseri-Turkey was investigated. In the study, 32 apricot genotypes were studied with ISSR and POX markers. In apricot genotypes, 88.1% polymorphism was obtained with ISSR markers and 32.7% with POX markers. A dendrogram was created by combining ISSR and POX markers data. Accordingly, the level of similarity among genotypes varied between 0.73-1.00. Among the apricots, the genotypes 31 and 32 in particular were clearly apart from the others. In addition, genotype 21 was located alone in the dendrogram. The remaining apricots are divided into two main groups. The study revealed that Yeşilhisar region apricots have a significant variation.

Keywords: apricot, Central Anatolia, genetic diversity

1. INTRODUCTION

Apricot is a strategically important fruit for Turkey, both socially and economically. Turkey is the world leader in production and produces 22.5% of the world amount of 3.58 million with 800 thousand tons. (FAO, 2021). The apricot is a fruit tree originated in Central Asia and China, and has been long cultivated in eastern Asia. It was introduced into Europe (400 BC) from Central Asia and Asia Minor. Apricot has been widely cultivated throughout Turkey since ancient times for its edible fruit, but mostly in Eastern Anatolia (Uzun et al., 2010). On the other hand, table apricot production is concentrated in the Mediterranean and Aegean regions. Türkiye has significant apricot genetic resources in different geographical regions. Some of these genetic resources consist of trees that originated from seeds.

Apricot has species and varieties that can adapt to different climatic and soil conditions. It can be grown in many different conditions, from the subtropical regions of North Africa to the deserts of Central Asia, from the coldest parts of Siberia to the humid regions of Japan and East China (Asma, 2000). In addition, it is reported that the apricot's need for chilling, which varies in a large interval between 100 and 1600 hours, allows to expand the cultivation areas and to make cultivation in still unused areas (Mehlanbacher et al., 1991).

There are different problems encountered in apricot production. In particular, late spring frosts, diseases and pests, self-incompatibility, yield and quality problems are priority. Especially in

European countries, different breeding studies are carried out to find solutions to these problems (Batnini et al., 2016; Krska, 2018). Under these conditions, the development of competitive new varieties has become a necessity. For the breeding of new varieties, first of all, a broadly defined gene pool is needed. It is common practice to use molecular markers to identify genetic resources. In this way, the genetic variation of the population is revealed. DNA screening is an indispensable tool to assess genetic diversity, discriminate varieties, identify possible synonyms and homonyms, and genetically trace plant varieties in food chains (Corrado et al., 2021).

Different peroxidase isoenzymes are found in many higher plants (Yoshida et al. 2003). They can oxidize compounds when peroxide (H_2O_2) or oxygen (O_2) is present in the environment. They contain three conserved motifs, namely the distal heme binding domain, the central domain of unknown function, and the proximal heme binding domain (Hiraga et al. 2001). Peroxidases are involved in many events that occur under stress in the life of the plant (infection, tolerance to insects, senescence, etc.) (Passardi et al., 2005; Gulsen et al., 2010). Peroxidase gene-based (POX) markers have previously been used for genetic diversity in fruit species such as apple (Gulsen et al., 2010), citrus (Uzun et al., 2014), and almond (Pinar et al., 2016). On the other hand, ISSR markers are widely used for genetic studies in fruit species. Numerous studies have been conducted with ISSR markers in *Prunus* species (Shahi-Gharahlar et al., 2011; Yilmaz et al., 2012; Sheikh et al., 2021; Zargar et al., 2022).

Kayseri in Central Anatolia has important local apricot gene resources. Production in the region for many years has led to the formation of a seed-based population over time. Determining the genetic diversity level of this population is the first step in the conservation and evaluation of these materials. In this context, in this study, local apricot populations in Yeşilhisar district of Kayseri province were studied with molecular markers. With the study, genetic identification of the apricot population in the region was provided.

2. MATERIALS AND METHODS

Thirty-two local apricot genotypes grown in Kayseri-Yeşilhisar used as material in this study. Young leaves at the shoot tip of the plants were used for DNA isolation. DNA isolations were performed according to the CTAB protocol modified from the method of Doyle and Doyle (1990). DNA concentrations were measured using the Nanodrop ND 1000 spectrophotometer and 10 ng/ μ L DNA solutions were prepared using TE (10-mM Tris-HCl, 0.1-mM ethylene diamine tetra acetic acid).

For PCR studies, different ISSR and POX primers were tested and studies were carried out with 3 ISSR and 3 POX primers with successful results. PCR components and PCR programs was designed according to Uzun et al. (2009) and Gulsen et al. (2010). After the PCR process, the PCR products obtained from the PCR studies were loaded onto a 2% agarose gel and run for 3 hours under 110 V. 1X TBE buffer was used in the preparation of the agarose gel and 25 μ l (0.5 mg/ml) ethidium bromide solution was added into it. 100 bp DNA Ladder was loaded as standard in each electrophoresis procedure. After this process, the gels were visualized under UV in the imaging system.

The score file was obtained by evaluating the gel images as present (1) or absent (0). Data from ISSR and POX were combined and analyzed in the computer package program NTSYS (Numerical Taxonomy Multivariate Analysis System, NTSYS-pc version 2.11, Exeter Software, Setauket, N.Y., USA, Rohlf, 2000). The similarity indices were calculated according to the Dice (1945)

method and the UPGMA (Unweighted Pair-Group Method With Arithmetic Average) method was used to create the dendrogram.

3. RESULTS AND DISCUSSIONS

In ISSR analysis, a total of 28 bands were obtained, 23 of which were found to be polymorphic and the polymorphism rate was determined as 82.1%. The mean number of bands per primer was 9.3, and the mean number of polymorphic bands was 7.6. The most bands were obtained in GA₈YG (15 pieces), and the least bands were obtained in the AGC₆G primer (6 bands). The number of polymorphic bands was determined the most in the GA₈YG primer (11 bands), and the least in the AGC₆G and VHVGTG₇ primers (6 bands). The highest polymorphism rate was detected in the AGC₆G (100%) primer (Table 1).

Table 1. List of ISSR and POX primers, their numbers of total and polymorphic fragments and ratio of polymorphism range.

	TBN	PBN	PR (%)
ISSR Primers			
AGC ₆ G	6	6	100
GA ₈ YG	15	11	78.5
VHVGTG ₇	7	6	85.7
Mean	9.3	7.6	82.1
Total	28	23	-
POX Primers			
POX10Fa/10Rb	8	1	12.5
POX1F/1R	7	2	28.5
POX12Fa/12Ra	6	3	50
Mean	10.57	9.57	30.3
Total	21	6	-

A total of 21 bands were obtained in POX analyzes and 6 of them were found to be polymorphic. The rate of polymorphism was 30.3%. Polymorphism rate of POX markers was much lower than ISSR markers. The reason for this is considered to be that POX is more related to stress factors. It is likely that there is less variation within the apricot species in terms of stress factors. The highest band number was obtained from the POX10Fa/10Rb primer (8), the most polymorphism was found in POX12Fa/12Ra primer (50%).

The cophenetic correlation coefficient, which reveals the correlation between the similarity indices and the dendrogram, was found to be $r = 0.78$. From this point of view, it is seen that there is a relatively high level of correlation between the similarity indices and the dendrogram obtained. In the dendrogram, 32 apricot genotypes were divided into two main groups. The similarity ratios of the genotypes varied between 0.73-1.00. In studies conducted with RAPD markers in different apricot cultivars, the level of similarity was found to be 0.90-0.96 (Uzun et al., 2007), and 0.77-0.97 with SRAP markers (Uzun et al., 2010). On the other hand, Basille et al. (2023) determined the similarity level between different apricot genotypes between 0.60-1.00 in their study with SSR

markers in Italy. According to Zargar et al. (2022) detected a higher level of diversity with ISSR markers in 106 apricot genotypes collected from different geographical regions of India. The different levels of variation obtained in the studies may be caused by the genetic diversity and number of the material used and the different marker systems.

All genotypes except two genotypes (6 and 7) are genetically separated from each other. Genotypes 31 and 32 were separated from the other 30 genotypes as a separate group. Among the remaining 30 genotypes, genotype 21 was grouped separately. The large group (B) consisting of 29 genotypes was divided into two subgroups at a similarity level of 0.86. In the first subgroup, there were 12 genotypes (2, 3, 4, 14, 18, 16, 19, 20, 27, 29, 28, 30). In the other subgroup, there are the remaining 17 apricot genotypes. In this subgroup, genotypes 6 and 7 were found to have almost the same genetic background.

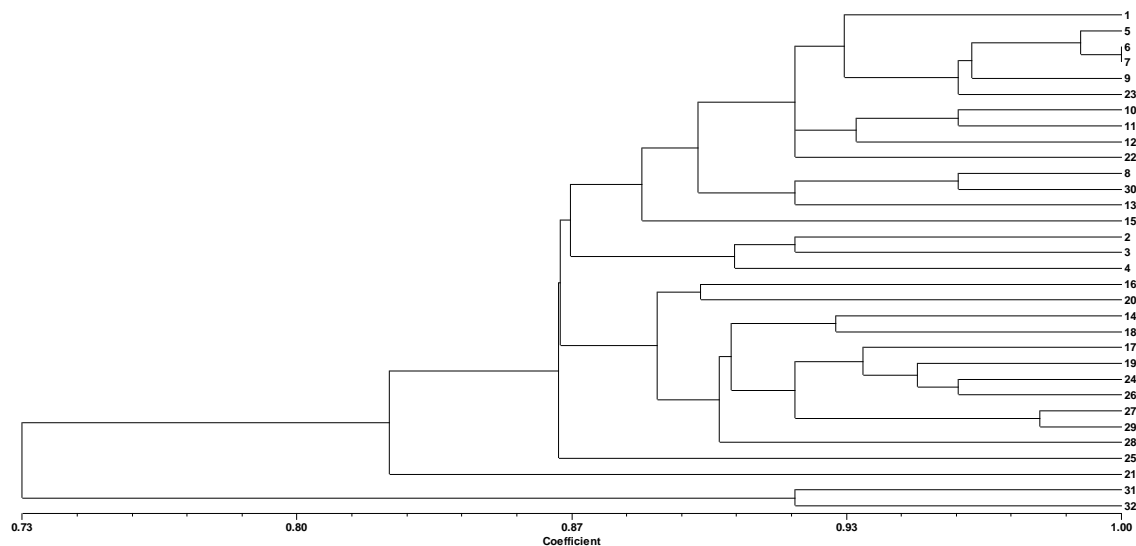


Figure 1. Dendrogram of the 32 apricot genotypes using UPGMA method obtained from ISSR and POX markers

Among the materials used in the study, 'Principal Component Analysis' (PCA) was performed and the distribution of genotypes on a two-dimensional plot was revealed (Figure 2). Principal component analysis is explained as a data reduction method and showing the total variance in a limited number of new variables in order to explain the relationship between two or more characters. Principal component analysis is considered a suitable technique for grouping individuals in the form of a distribution on the plane, where the 'eigen' values of the first two or three principal components account for most of the variation as a cumulative sum. (Mohammadi and Prasanna, 2003). According to the results of principal component analyzes in our study, the first three 'eigen' values explained 91.65% of the total variation. This shows that the variation is explained very well with the principal component analysis and the similarities/differences between the materials used in the study are well expressed with the two-dimensional distribution obtained by this analysis. In the two-dimensional plane graph, as in the dendrogram, genotypes 31 and 32 are grouped separately from the others. Likewise, the genotype number 21 was clearly differentiated. The remaining genotypes showed relatively closer grouping.

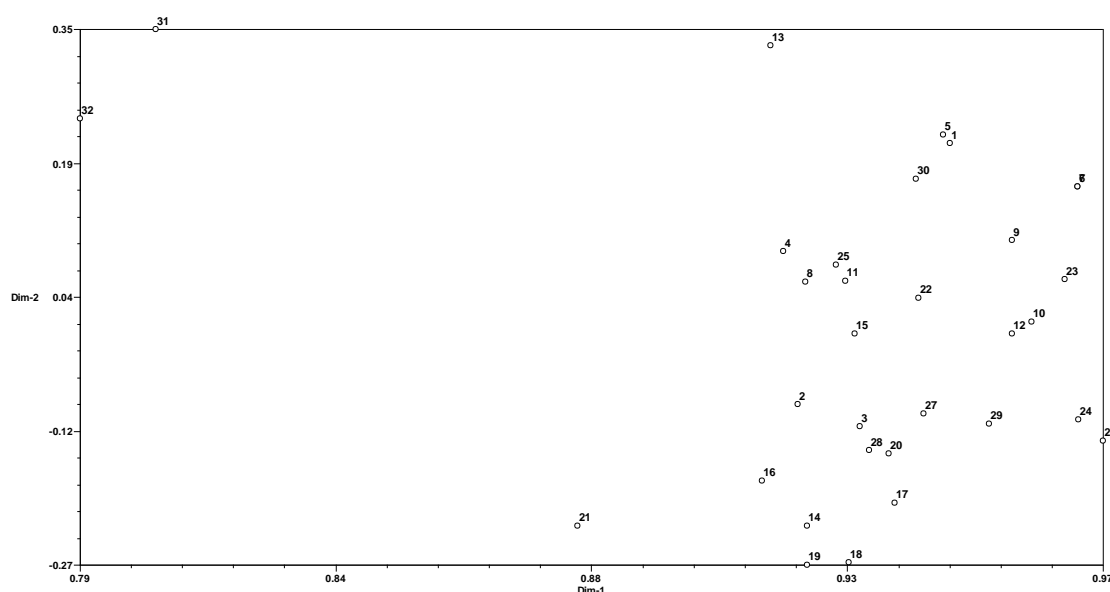


Figure 2. Two-dimensional plot of the principal components analysis of ISSR and POX data

4. CONCLUSIONS

It was emphasized that detailed genetic characterization of plants is an important step for the use of genetic resources, their successful conservation and the design of breeding programs. It has been reported that the most effective technique for genetic characterization is molecular markers (Tripathi et al., 2012; Zargar et al., 2022). The results presented in our study show that there is significant variation among local apricot genotypes in the Kayseri-Yeşilhisar region. Although this region is not a very large area, it contains valuable diversity. Breeding here for many years has created a genetic pool as a result of interactions between different cultivars and genotypes. These results may indicate the potential of the studied molecular technique to discriminate between apricot genotypes. It also guides the selection of parental genotypes to be used in crossbreeding breeding programs.

5. ACKNOWLEDGEMENTS

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