Morphological and molecular characterization of some Tomato (Solanum lycopersicum L.) genotypes collected from Erzincan Province of Turkey

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Abstract

Background: Turkey is a country with different geographical features and therefore it is extremely diverse in plant diversity. Tomato is one of the most important vegetables produced both in the world and in Turkey. In this study, it was aimed to determine the genetic diversity of 24 tomato genotypes collected from local farmers from “Center villages” and “Üzümlü” district in Erzincan province.

Methods and Results: Morphological (qualitative and quantitative) and ISSR (Inter Simple Sequence Repeat) were used to determine genetic diversity among genotypes. Contribution rates of morphological properties to the first three main components differed. Genotype 24 was found to be higher than other genotypes with important quantitative morphological features such as fruit length, fruit width, fruit weight and soluble solid content (SSC). Considering the overall morphological traits, a wide variation was detected between genotypes. Genotypes were divided into 2 main groups according to the evaluation with molecular markers. According to the molecular findings obtained, the polymorphism rate ranged from 0 to 100% and the average polymorphism rate was calculated as 80%.

Conclusions: Present findings revealed the diversity in tomato genotypes collected from Erzincan province and may constitute the bases for further breeding studies in tomato and will bring an integrity in tomato identification studies.

Keywords: Erzincan · Genetic diversity · ISSR · PCA · Tomato

Introduction

Turkey is a country with different geographical features and therefore it is extremely diverse in plant diversity. Tomato, one of the most popular and important vegetables in the world, has a wide range of uses (fresh, dried, sauce, tomato paste, meal, etc.) and constitutes approximately 14% of the world’s vegetable production [1–3]. Turkey is one of the world’s largest tomato producers after India and China. In addition, approximately 7% of the world’s tomato production is carried out in Turkey [4]. It has an important nutritional value, especially in terms of vitamins A, C and antioxidant compounds. It is an important source of lycopene, which protects the cell from oxidative damage. Tomato (2n = 2x = 24) is a self-pollinating plant belonging to the Solanaceae family [5]. The phylogenetic classification of the Solanaceae family has been recently updated and Tomato in the genus Lycopersicon has been included into the genus Solanum with its new terminology [6]. Homeland of tomatoes; It is stated to be South America and the Andes Mountains. After the discovery of America, which is called the new world, tomatoes spread all over the world in time with the introduction of Europe [7]. In recent years, the genetic diversity of plant species has decreased significantly, as hybrid varieties have replaced village populations and traditional native species [8]. The genetic diversity of cultivated tomato is narrowing due to continuous selection processes caused by increasing use of hybrid varieties and other factors [9]. For this reason, the need for studies to collect, characterize and protect these genetic resources has increased [10–12]. It is extremely important to determine the genetic diversity of plant species, to reveal genotypes...
that are resistant to biotic and abiotic stress conditions, with high yield and quality characteristics, and for variety development activities in plant breeding [13–17]. Studies to increase tolerance to various stress factors in agriculturally important species such as tomatoes have not been fully successful due to the complexity of the responses to stress. Local genotypes are extremely important in terms of preserving the genetic potential of the species they belong to, due to their high adaptability to different ecologies in which they are grown, and thus having many desired quality characteristics as well as resistance to stress factors, diseases, and pests [18]. The lack of information about the agro-morphological characteristics and genetic structure of local varieties has limited their use in breeding studies [19]. Plant genetic diversity: It can be determined using morphological, biochemical, and molecular markers [20]. Morphological markers can be used to measure genetic differences. However, since morphological features are affected by environmental conditions, it is not sufficient alone in determining the genetic diversity [21, 22]. For this reason, there is a need to use molecular markers that are not affected by environmental conditions and give highly reliable results together with morphological, biochemical and phenological properties [23]. Tomato is an important vegetable in human nutrition and determining its genetic diversity is important in terms of its use in breeding studies and its sustainability. Molecular marker studies such as Amplified Fragment Length Polymorphism (AFLP) [24], Inter-Retrotransposon Amplified Polymorphism (IRAP) [25], Inter Simple Sequence Repeat (ISSR) [26–28], Random Amplified Polymorphic DNA (RAPD) [29, 30], Start Codon Targeted (SCoT) [31], Simple-Sequence Repeats (SSR) [32, 33] and morphological marker studies [34, 35] determined has been and continues to be done to determine the genetic diversity in tomato.

Although Turkey is not the origin of the tomato, which is included in the Solanaceae family, it is known that different types are formed because of the production made by taking its own seeds and thus a natural gene pool is formed [35]. Therefore, over time, it has adapted to different ecological conditions and a natural gene pool containing different species has emerged [4]. These valuable genetic materials are under threat of extinction due to environmental and other pressures in the regions where they are found. The genetic profiles of local tomato genotypes are clearly different from commercial tomato cultivars [36]. Currently, there is insufficient information on variations among local tomato populations [37]. Erzincan province, which is in the east of Turkey and has a microclimate feature, has an important potential in terms of tomato diversity in the region. There are not enough studies on the determination of tomato genetic diversity with molecular markers in the province. The aim of the present study was determined to genetic diversity of 24 tomato genotypes collected from the “villages of the

Table 1 Information on the locations where the genotypes were collected

| Genotype | Coordinate | Altitude(m) | District |
|----------|------------|-------------|----------|
| #1       | 39° 45' 20” N 39° 21’ 8” E | 1351 | Bahçeliköy |
| #2       | 39° 45’ 28” N 39° 20’ 58” E | 1353 | Bahçeliköy |
| #3       | 39° 48’ 27” N 39° 18’ 46” E | 1443 | Çatalarmut |
| #4       | 39° 42’ 39” N 39° 42’ 26” E | 1462 | Üzümli |
| #5       | 39° 37’ 11” N 39° 43’ 36” E | 1403 | Cevizli |
| #6       | 39° 37’ 11” N 39° 43’ 36” E | 1162 | Uluköy |
| #7       | 39° 37’ 22” N 39° 44’ 7” E | 1162 | Uluköy |
| #8       | 39° 37’ 22” N 39° 44’ 7” E | 1162 | Uluköy |
| #9       | 39° 45’ 19” N 39° 21’ 7” E | 1340 | Bahçeliköy |
| #10      | 39° 37’ 20” N 39° 44’ 6” E | 1164 | Uluköy |
| #11      | 39° 48’ 34” N 39° 18’ 35” E | 1463 | Çatalarmut |
| #12      | 39° 48’ 34” N 39° 18’ 35” E | 1463 | Çatalarmut |
| #13      | 39° 47’ 19” N 39° 21’ 4” E | 1319 | Elmaköy |
| #14      | 39° 48’ 27” N 39° 18’ 22” E | 1462 | Çatalarmut |
| #15      | 39° 42’ 7” N 39° 41’ 16” E | 1321 | Üzümli |
| #16      | 39° 48’ 34” N 39° 18’ 35” E | 1463 | Çatalarmut |
| #17      | 39° 48’ 34” N 39° 18’ 35” E | 1463 | Çatalarmut |
| #18      | 39° 37’ 10” N 39° 43’ 52” E | 1164 | Uluköy |
| #19      | 39° 41’ 48” N 39° 41’ 14” E | 1285 | Üzümli |
| #20      | 39° 41’ 48” N 39° 41’ 14” E | 1285 | Üzümli |
| #21      | 39° 48’ 27” N 39° 18’ 46” E | 1443 | Çatalarmut |
| #22      | 39° 48’ 27” N 39° 18’ 46” E | 1443 | Çatalarmut |
| #23      | 39° 42’ 28” N 39° 42’ 16” E | 1450 | Üzümli |
| #24      | 39° 48’ 27” N 39° 18’ 46” E | 1443 | Çatalarmut |
Morphological characteristic analysis

Flower color and some fruit characteristics were considered in the morphological separation of tomato genotypes. In this study, 9 qualitative morphological descriptors (flower color, fruit color (before maturity), fruit shape, fruit color at maturity, fruit color of flesh at maturity, fruit firmness, slicing in fruit, fruit cross section, fruit green shoulder before maturity) and 3 quantitative morphological descriptors (fruit length, fruit width, fruit weight) were used. In addition, pH and soluble solids contents (SSC) of tomato fruits and leaf SPAD values were also measured. Morphological measurements were made with 3 replications and in each replication, 10 flower, 10 fruit and 10 leaf samples were used. Leaf chlorophyll content was measured with the SPAD-502 (Konica Minolta, Japan) instrument. Soluble Solid Content (SSC) was measured with a hand. The pH contents of the fruits were determined with a pH meter. Fruit width and

Materials and methods

Plant material

In this study, twenty-four local tomato genotypes from farmers in “Central villages” (Bahçeliköy, Cevizli, Çatalmut, Elmaköy, Uluköy and Üzümlü) and “Üzümlü” district of Erzincan province were collected in 2021 (Tables 1 and Fig. 1). Morphological and ISSR molecular markers were used to determine genetic diversity among genotypes.

Fig. 1 Map of Erzincan province where tomato genotypes were collected. (Source: wikipedia.org)
fruit length were determined by measuring (mm) with a
digital caliper. A digital scale sensitive to 0.01 g was used to
determine the average fruit weights. Within the scope of the
study, correlation, and principal component analysis (PCA)
of morphological properties were measured with JMP pro
14 software.

**ISSR analysis**

Genomic DNA was isolated from approximately 100 mg of
frozen tomato leaf tissue according to the CTAB method
established by [38]. After measuring the DNA concentration
of tomato leaves with 2% agarose gel, gel electrophoresis
was performed with a 100 bp DNA ladder [17]. Following,
an initial screen of 20 ISSR primers that have been used to
perform PCR amplification. The amplification reactions
were carried out in 15 µl volume containing 2 µl (10 ng/
µl), 2.73 µl PCR mix(buffer, Taq DNA, dNTP, MgCl2),
1 µl primer and 9.27 µl nuclease-free water using Thermal
cycle (Sense Quest) Lab Cycle programmed for an initial
denaturation step at 94 °C for 3 min, followed by 35 cycles
of 1 min at 94 °C, 35 cycles of 50 s at the specific annealing
temperature at 53 °C, 35 cycles of 2 min at 72 °C and ended
with a final extension step 7 min at 72 °C. Each reaction
was repeated at least twice [39]. Only repeated bands were
considered in scoring the bands. The obtained PCR products
were electrophoresed with the help of 2% (w/v) agarose gel
for 4 h at a constant voltage of 110 V. The resulting gel was
then visualized using the MS Major Science UV visualization
system.

**Statistical analysis**

The analysis of the pH, SSC and quantitative morphologi-
cal characteristics of the tomato genotypes used in the study
was done with the SPSS 22.0 statistical package program.
Statistical differences between the means were determined
by Duncan multiple comparison test at 5% significance
level. ISSR markers were scored as present (1) and absent
(0). ISSR band sizes were estimated by comparison with
100 bp DNA ladder (GENESTATM). The similarity index
of the genotypes was determined using the NTSYS-pc
V2.11 program [40]. The similarity dendrogram between
genotypes was generated by UPGMA (Unweighted Pair
Group Method with Arithmetic Mean Cluster analysis) [41].

| Genotype | Flower Color | Fruit color (before maturity) | Fruit shape | Fruit color (at maturity) | Color of Fruit flesh (at maturity) | Fruit firmness | Slicing in fruit | Fruit cross section | Fruit green shoulder |
|----------|--------------|-------------------------------|-------------|--------------------------|-----------------------------------|---------------|-----------------|---------------------|---------------------|
| #1       | Yellow       | Green                          | Round       | Red                      | Red                               | Medium        | Absent          | Round               | Absent              |
| #2       | Yellow       | Green                          | Round       | Red                      | Red                               | Medium        | Strong          | Round               | Absent              |
| #3       | Light yellow | Green                          | Round       | Red                      | Red                               | Soft          | Strong          | Round               | Absent              |
| #4       | Dark yellow  | Green                          | Heart-shaped| Red                      | Red                               | Firm          | Weak            | Angular             | Absent              |
| #5       | Yellow       | Green                          | Round       | Red                      | Red                               | Soft          | Absent          | Round               | Absent              |
| #6       | Yellow       | Green                          | Slightly flat| Red                      | Red                               | Soft          | Absent          | Irregular           | Present             |
| #7       | Yellow       | Green                          | Slightly flat| Red                      | Red                               | Soft          | Absent          | Angular             | Absent              |
| #8       | Yellow       | Green                          | Round       | Red                      | Red                               | Medium        | Absent          | Round               | Absent              |
| #9       | Light yellow | Green                          | Slightly flat| Red                      | Red                               | Soft          | Weak            | Angular             | Absent              |
| #10      | Light yellow | Light green                    | Slightly flat| Pink                     | Pink                              | Medium        | Weak            | Round               | Absent              |
| #11      | Yellow       | Green                          | Cylindrical | Dark red                 | Red                               | Firm          | Absent          | Round               | Absent              |
| #12      | Yellow       | Green                          | Flat        | Red                      | Red                               | Soft          | Absent          | Angular             | Absent              |
| #13      | Yellow       | Green                          | Slightly flat| Red                      | Red                               | Soft          | Absent          | Round               | Absent              |
| #14      | Yellow       | Light green                    | Flat        | Pink                     | Pink                              | Firm          | Weak            | Angular             | Absent              |
| #15      | Yellow       | Green                          | Round       | Dark red                 | Red                               | Soft          | Absent          | Round               | Absent              |
| #16      | Yellow       | Green                          | Round       | Red                      | Red                               | Soft          | Absent          | Round               | Absent              |
| #17      | Dark yellow  | Green                          | Round       | Red                      | Red                               | Soft          | Absent          | Round               | Absent              |
| #18      | Yellow       | Light green                    | Round       | Pink                     | Medium                             | Absent        | Round           | Absent              | Absent              |
| #19      | Light yellow | Green                          | Flat        | Red                      | Red                               | Firm          | Weak            | Angular             | Absent              |
| #20      | Yellow       | Green                          | Round       | Light red                | Red                               | Soft          | Absent          | Round               | Absent              |
| #21      | Yellow       | Green                          | Round       | Red                      | Medium                             | Weak          | Angular         | Present             |                     |
| #22      | Light yellow | Green                          | Flat        | Red                      | Red                               | Soft          | Strong          | Irregular           | Present             |
| #23      | Yellow       | Dark green                     | Round       | Brown                    | Brown                             | Medium        | Medium          | Round               | Present             |
| #24      | Light green  | Flat                           | Yellow      | Firm                     | Strong                             | Irregular     | Absent          |                     |                     |
Results

Morphological properties analysis

In this study, twenty-four tomato genotypes were collected from different locations in Erzincan province. This tomato population has been characterized according to morphological (qualitative and quantitative properties) and molecular markers. Since changes in morphological traits occurred in response to external conditions, it is important to support these morphological variations with molecular studies. Some qualitative and quantitative properties of tomato genotypes are given in Tables 2 and 3. It was observed that there were significant morphological differences in flower color and some fruit properties among the collected local tomato genotypes. The flower color was considered as yellow in 17 genotypes, light yellow in 5 genotypes and dark yellow in 2 genotypes. Fruit color (before maturity) was identified as green in 19 genotypes, light green in 4 genotypes and dark green in only 1 genotype. In terms of fruit shape, genotypes are divided into 5 groups as round (12 genotype), heart-shaped (1 genotype), flat (5 genotype), slightly flat (5 genotype) and cylindrical (1 genotype). Differences were determined between genotypes according to the fruit color (at maturity). Genotypes were divided into 6 groups based on this trait as yellow (1 genotype), pink (3 genotype), light red (1 genotype), red (16 genotype), dark red (2 genotype) and brown (1 genotype) color. Genotypes were divided into 4 groups as yellow, pink, red and brown according to color of fruit flesh (at maturity). It was determined that 19 genotypes had red, 3 genotypes had pink, 1 genotype had yellow and 1 genotype had brown fruit flesh color. According to fruit firmness, genotypes were divided into 3 groups as soft, medium and firm. In the classification made by considering the slice status of the fruit, differences were determined between the genotypes. Slicing was not observed in the fruits of 11 genotypes. In other genotypes, weak (8 genotype), medium (1 genotype) and strong (4 genotype) slices were determined. Fruit cross section was identified as round in 14 genotypes, and angular in 7 genotypes. Considering the fruit section feature; It was determined as round in 14 genotypes, angular in 7 genotypes and irregular in 3 genotypes. While the presence of green shoulder in fruit was detected in 5 genotypes, it was not found in other genotypes.

In this study, it was determined that the differences between genotypes in terms of quantitative fruit characteristics and leaf SPAD values were statistically significant in all parameters. The highest and lowest fruit length values

| Gen | Fruit length (mm) | Fruit width (mm) | Fruit weight (g) | pH  | SSC (%) | SPAD |
|-----|------------------|-----------------|-----------------|-----|---------|------|
| #1  | 54.95 fg         | 61.34 h         | 111.16 o        | 4.80 cd | 4.85i | 27.27dg |
| #2  | 62.62c           | 68.54e           | 183.07 e        | 4.82bc | 4.77j | 30.10cde |
| #3  | 62.65c           | 63.89 g          | 178.09 f        | 4.72gh | 5.37e | 29.27cf |
| #4  | 66.81ab          | 55.04k           | 158.85 h        | 4.76ef | 4.77j | 30.77be |
| #5  | 56.14f           | 56.41k           | 130.78 j        | 4.61jk | 6.07b | 31.53bcd |
| #6  | 50.87hi          | 66.02f           | 116.69 m        | 4.72 g  | 5.93c | 27.00dg |
| #7  | 48.56jk          | 64.22 g          | 127.94k         | 4.71gh | 6.13b | 26.40dg |
| #8  | 57.78e           | 58.38i           | 120.26 L        | 4.86a  | 4.86i | 27.83dg |
| #9  | 50.24i           | 60.35 h          | 115.17 mn       | 4.84ab | 6.10b | 25.70cde |
| #10 | 60.40d           | 71.52d           | 209.71d         | 4.60k  | 4.42k | 27.57dg |
| #11 | 62.75c           | 64.07 g          | 162.81 g        | 4.77de | 5.39e | 29.23cf |
| #12 | 54.94 fg         | 40.42 m          | 74.87 r         | 4.72 g  | 5.23f | 35.47ab |
| #13 | 49.32ij          | 71.19d           | 138.24i         | 4.54 L  | 5.35e | 24.10 g |
| #14 | 47.52kl          | 58.39i           | 117.48 m        | 4.63ijk | 4.81ij | 26.73dg |
| #15 | 66.35b           | 85.36b           | 246.46b         | 4.69 h  | 4.81ij | 37.50a |
| #16 | 54.34 g          | 55.81k           | 107.79p         | 4.60k  | 3.05 L | 33.90abc |
| #17 | 17.62 m          | 17.16 m          | 16.63s          | 4.71gh | 2.73 m | 30.30cde |
| #18 | 54.57 fg         | 56.57jk          | 111.35 o        | 4.64ij | 4.99 h | 27.03dg |
| #19 | 47.55kl          | 64.90 fg         | 116.21 m        | 4.73 fg | 4.98 h | 24.50 fg |
| #20 | 52.01 h          | 57.98ij          | 125.53 k        | 4.72gh | 5.79d | 31.33bcd |
| #21 | 67.66ab          | 70.93d           | 217.01 c        | 4.61jk | 4.45k | 26.57dg |
| #22 | 68.21a           | 87.84a           | 307.99a         | 4.85ab | 6.33a | 27.67dg |
| #23 | 54.09 g          | 50.42 L          | 112.61ino       | 4.76ef | 5.16 g | 29.17cf |
| #24 | 46.51 L          | 74.52c           | 217.61c         | 4.66i  | 5.94e | 31.00bcd |
| Mean | 54.77            | 61.72            | 146.85          | 4.71    | 5.09  | 29.08 |

Different lower-case letters show statistically significant differences between genotypes in column (%5)
were determined in \( \#22 \) (68.21 mm) and \( \#17 \) (17.62 mm) genotypes, respectively. Fruit width values varying between 17.16 and 87.84 mm were determined among genotypes. The lowest and highest values in this parameter occurred in genotypes \( \#17 \) and \( \#22 \), respectively. The highest average fruit weight was determined with 307.99 g in \( \#22 \) genotype, while the lowest value was determined with 16.63 g in \( \#17 \) genotype. While genotype \( \#8 \) had the highest pH value, the lowest pH value was determined in genotype \( \#13 \). In terms of SSC value, the highest and lowest values were determined as 6.33% and 2.73%, respectively. According to this value, the highest content was found in genotype \( \#22 \) and the lowest value was found in genotype \( \#17 \). The highest value in leaf SPAD measurements was determined in genotype \( \#15 \) (37.50). The lowest SPAD value was found in genotype \( \#13 \) (37.50).

**Correlation analysis of morphological characters**

It was determined that fruit color (before maturity) was positively correlated with fruit shape, Fruit color-at maturity and fruit flesh color. In addition, fruit width and fruit weight had a negative relationship. Significant positive correlations were also observed between fruit shape and several traits including fruit color at maturity (0.45) and fruit color of flesh (0.94). In addition, significant negative correlations were also observed between fruit shape and several traits including SSC (-0.30), fruit width (0.94) and fruit weight (-0.38). Fruit color at maturity was significantly positive correlated with fruit color of flesh (0.59). pH value had a

**Morphological data principal component analysis**

Principal component analysis values based on morphological data of tomato genotypes are given in Table 4. Principal component analysis of tomato genotypes was carried out according to 15 flower, fruit and leaf characteristics. According to principal component analyses, the first three components (with Eigen values > 1) can explain most of the total variation (62.84%). The first (PC1), second (PC2) and third (PC3) principal components represent 31.81%, 18.40% and 12.63% of the total variance, respectively. The contribution rates of morphological features to the first three main components show differences (Table 4).

The position plot corresponding to the correlations of the tomato genotypes with respect to the first two main components is given in Fig. 2. According to the first two main components, genotypes 17, 18, 22 and 23 from tomato genotypes were clustered individually compared to other tomato genotypes. Tomato genotypes 10, 15 and 24 were separated from other tomato genotypes by forming a separate cluster. Except for genotypes 10, 15, 17, 18, 22, 23 and 24, all other genotypes were clustered together.

**Table 4** Principal component analysis and contribution ration based on morphological data of tomato genotypes

|                     | PC1     | % Ct. | PC2     | % Ct. | PC3    | % Ct. |
|---------------------|---------|-------|---------|-------|--------|-------|
| Flower Color        | -0.253  | 6.40  | -0.176  | 3.13  | 0.153  | 2.35  |
| Fruit color (before maturity) | -0.236  | 5.61  | 0.473   | 22.39 | -0.033 | 0.11  |
| Fruit shape         | -0.297  | 8.87  | 0.064   | 0.42  | 0.414  | 17.16 |
| Fruit color-at maturity | -0.290  | 8.45  | 0.200   | 4.02  | 0.163  | 2.66  |
| Fruit color of flesh (at maturity) | -0.258  | 6.66  | 0.462   | 21.43 | -0.023 | 0.05  |
| Fruit firmness      | 0.105   | 1.12  | -0.231  | 5.38  | 0.410  | 16.88 |
| Slicing in fruit    | 0.273   | 7.48  | 0.170   | 2.92  | 0.242  | 5.87  |
| Fruit cross section | -0.299  | 8.97  | -0.056  | 0.31  | 0.178  | 3.17  |
| Fruit green shoulder (before maturity) | 0.092   | 0.86  | 0.395   | 15.62 | -0.002 | 0.00  |
| pH                  | 0.018   | 0.04  | 0.320   | 10.29 | 0.200  | 4.00  |
| SSC(%)              | 0.241   | 5.82  | 0.239   | 5.74  | -0.129 | 1.68  |
| Fruit length (mm)   | 0.237   | 5.65  | 0.205   | 4.22  | 0.432  | 18.74 |
| Fruit width (mm)    | 0.416   | 17.35 | 0.082   | 0.68  | 0.026  | 0.07  |
| Fruit weight (g)    | 0.404   | 16.37 | 0.078   | 0.62  | 0.201  | 4.07  |
| SPAD                | -0.059  | 0.35  | -0.168  | 2.84  | 0.481  | 23.18 |

Eigenvalue 4.771 2.760 1.894

Variance (%) 31.81 18.40 12.63

Cumulative Variance (%) 31.81 50.21 62.84

Ct: contribution
significantly positive relationship with properties such as fruit color (before maturity), fruit color at maturity and SSC. In addition, SSC (%) value was also positively correlated with fruit length (0.31), fruit width (0.49) and fruit weight (0.35) (Table 5). Fruit weight had a positive and significant association with fruit length and fruit diameter.

ISSR analysis

In this study, 24 different tomato genotypes were analyzed using 20 different ISSR markers. Polymorphic bands were obtained from 15 of the markers used. A total of 105 scoreable bands were obtained from the primers. It was determined that 84 of the primers obtained were polymorphic. The base lengths of the primers varied between 130 and 1300 bp in total. In the total number of bands that can be scored, bands ranging in number from 3 to 11 were obtained in the primers, the lowest band was obtained from the primer (CA)6AC, and the highest number of bands was obtained from the primer DBD(CA)7. Polymorphic bands occurred in the primers from all primers except the BDB(CA)7 primer. The highest polymorphic band number was obtained from the DBD(CA)7 primer with 10, and the lowest polymorphic band was obtained from the (TCC)5RY primer with 2. The polymorphism rates of the primers varied between 0% and 100%. Since all bands obtained from primers (GA)8YG, (GT)8YA, VHVG(TG)7, (GACA)4, (CA)6AC, (CT)8TG are polymorphic, polymorphism percentages were determined as 100%. The lowest percentage of polymorphism (0%) was detected in primer BDB(CA)7C. The mean band and mean polymorphic band numbers obtained from the primers used in the study were found to be 7 and 5.60, respectively. The mean polymorphism value in the study was determined to be 80% (Table 6).

The similarity index in the dendrogram created according to the UPGMA method in tomato genotypes differed between 0.64 and 0.86. It was revealed that the closest genotypes to each other were genotypes ≠ 21 and ≠ 22 with a similarity ratio of 0.86. Genotypes with the greatest genetic distance were ≠ 1 and ≠ 24. According to the dendrogram in genotypes, 2 main groups were formed. In the first main group was included only one genotype (≠ 24). In the second main group, other genotypes included in the study were included and this was divided into 2 subgroups. In the 1st subgroup, only genotype ≠ 18 was found. In the second subgroup, all genotypes were separated from each other in general (Fig. 3).
| Primers     | Base Length | Total Band Number | Polymorphic Band Number | Polymorphism Rate (%) |
|------------|-------------|-------------------|-------------------------|-----------------------|
| (GA)8YG    | 170–1150    | 8                 | 8                       | 100.00                |
| CAC3GC     | 250–900     | 6                 | 5                       | 83.33                 |
| GT)8YA     | 130–1000    | 8                 | 8                       | 100.00                |
| DBD(CA)7   | 150–1200    | 11                | 10                      | 90.91                 |
| VHV(GTG)7  | 300–1050    | 9                 | 9                       | 100.00                |
| (GACA)4    | 300–700     | 5                 | 5                       | 100.00                |
| BDB(CA)7C  | 250–900     | 4                 | 0                       | 0.00                  |
| (TCC)5RY   | 300–1000    | 7                 | 2                       | 28.57                 |
| HVH(CA)7T  | 400–1100    | 8                 | 5                       | 62.50                 |
| (CA)6AC    | 180–600     | 3                 | 3                       | 100.00                |
| (AGC)6G    | 200–1200    | 8                 | 6                       | 75.00                 |
| (CT)8TG    | 400–800     | 6                 | 6                       | 100.00                |
| (AG)8T     | 150–1250    | 5                 | 3                       | 60.00                 |
| HVH(TCC)7  | 270–1300    | 8                 | 7                       | 87.50                 |
| AG)7YC     | 200–900     | 9                 | 7                       | 77.78                 |
| **Mean**   | **130–1300**| **7**             | **5,6**                 | **80.00**             |
| **Total**  |             | **105**           |                         | **84**                |

**Discussion**

Tomato is one of the most important vegetables in the world as well as in Turkey. Molecular markers, along with qualitative and quantitative morphological characters, are important markers used to detect genetic variation within plant species [31]. Measurements of morphological traits provide a simple application to assess genetic variation with simultaneous assessment of genotype performance in certain ecological conditions, although these morphological characters are often influenced by environmental factors [42]. Flower, leaf and fruit characters are important distinguishing indicators in determining the variation among tomato genotypes [5]. Characterization studies are carried out in different regions of the world in tomato based on morphological data. Fruit descriptors are more promising markers for morphological differentiation of tomato genotypes [43, 44]. Different researchers have divided tomato genotypes into 4 groups in terms of fruit color (before maturity): greenish-white, light green, green and dark green [7]. In the study by [45]; tomatoes genotypes were divided into 5 groups (Greenish-white, green, light green, dark green, very dark green and dark) according to the fruit color (before maturity). Fruit skin color is controlled by the Y gene, so it is not affected by environmental conditions. In other words, it does not show genotype × environment interaction. Therefore, fruit color is an important parameter in the differentiation of genotypes [46]. In tomato, expression of fruit shape traits is known to have a high degree of genetic determinism. Fruit shape and size is a very important feature both for the consumer and for marketing. Fruit shape is one of the most promising features that can be used for precise identification of tomato genotype [7]. In present study wide variation was observed in fruit shape among the genotypes; round, heart-shaped, flat, slightly flat, and cylindrical were recorded. According to the fruit shape, [45] reported that the genotypes were divided into 6 groups (flattened, slightly flattened, cylindrical, rounded, high-rounded, and heart-shaped), while [7] reported that they were divided into 4 groups (rounded, flattened, ellipsoid and heart-shaped). It has been determined that there are similarities and differences with other studies in terms of fruit shape. These differences are thought to be due to the genotype difference used in the studies. In this study, variations in fruit color were observed between genotypes. There are 6 different groups in terms of fruit color (yellow, pink, light red, red, dark red and brown). Approximately 66.6% of the genotypes have red fruit color. Flesh color is a parameter used in tomato morphological differentiation. In a similar study, it was stated that tomato genotypes were divided into 5 groups (red, yellow, orange, pink and brownish color) according to fruit flesh color [37]. The fruit flesh colors detected in this study are similar to the findings of other researchers. In this study, genotypes were evaluated in 3 groups according to the fruit firmness parameter as soft, medium, and firm. Many factors such as genotype, harvest time, plant nutrition can affect fruit firmness. In a similar study, it was determined that the fruit cross-section shape of the genotypes was round (85.51%), irregular (13.04%) and angular (1.45%) [47]. In this study, they were evaluated in 3 groups according to their cross-sectional shapes. In this study, green shoulder was detected in some genotypes, while it was absent in others. In many similar studies, green shoulder in fruits was expressed as present or absent [37, 45]. Fruit length varied between 68.21(6.821 cm) mm and 17.62 mm (1.762 cm). This result was also very similar to
previous studies published by [48] and [49]. In a study by [46] reported that fruit length is between 40.7 and 94.6 mm and fruit weight is between 62.6 and 446.6 g. In this study, fruit weight had a positive and significant association with fruit length and fruit diameter. It can be said that there are some similarities with other studies in terms of fruit width and weight. We recorded wide range of fruit weight among the genotypes. The variation of fruit weight could be due to the differences of tomato genotypes used in the studies.

In a similar study, the average pH value of fruits in tomato genotypes varied between 4.55 and 4.68. The results are remarkably similar to our study. There are many studies investigating the SSC (%) content of tomatoes [48]. The results of the research were partially different from other studies. Such differences between tomato genotypes have also been observed by [50]. Principal component analysis has been used by many researchers to assess morphological

Fig. 3 Dendrogram generated by UPGMA method using ISSR markers for tomato genotypes
diversity and establish genetic associations between tomato genotypes [19, 42].

The ISSR technique has been used successfully in many studies to identify genetic variations among tomato genotypes. In genetic diversity study with ISSR markers carried out in the Iran and Turkey tomato genotypes, the average number of bands per marker was 13, the mean number of polymorphic bands was 13.2, and the mean polymorphism rate was 100% [35]. In a similar study using ISSR markers in tomato, the number of bands per allele was 766 and the polymorphism rate was 76.3. [26]. In another study, the average number of polymorphic bands per marker was determined as 4.9 in the SSR analysis performed in the tomato genotypes in Greece. In addition, the rate of polymorphism was reported as 48.9. Researchers determined that genetic similarity values ranged from 0.56 to 0.95 [34]. Similarity index showed a change from 0.01 to 1.87 between genotypes in the RAPD analysis in tomato genotypes [30]. In another study with SSR markers, the genetic similarity value was found to be 0.79 on average [28]. Molecular marker results obtained from the current study are generally in agreement with previous studies. The differences can be explained by the different marker systems used and the variability in genotypes.

As a result, genetic diversity was determined with the help of morphological and molecular markers in 24 tomato genotypes collected from Erzincan province. A wide variation among genotypes emerged based on data from the markers (morphologic and molecular) used. According to ISSR molecular marker data, the most genetically distant genotypes were ≠ 1 and ≠ 24. Genotypes with low genetic similarity are extremely important for crossbreeding. In terms of morphological characteristics, especially in terms of fruit length, fruit width, fruit weight and SSC (%), genotype ≠ 22 produced high results compared to the other genotypes. The data are an important tool for identifying and maintaining the diversity of this germplasm.

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Declarations

Conflict of interest The author declare that author have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Consent to participate Author mentioned in the manuscript have agreed for authorship, read and approved the manuscript.

Consent to Publish (Ethics) Author give the consent for the publication of identifiable details, which can include photograph(s)/tables and/or details within the text to be published in the “Molecular Biology Reports” Journal.

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