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Screening of Eggplant F_3 Segregating Population for Salt Tolerance

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ABSTRACT

Utilizing salt-tolerant varieties in affected lands is the most prominent environmentally friendly solution. Wild relatives of eggplant have tolerance to some abiotic stresses. The aim of the study was to assess salinity tolerance in the third filial segregating population of eggplant lines that were previously associated with salt tolerance, then they could be used in breeding programs. The 50 F_3 families resulting from crossings of the inbred line BATEMTDC47 (*Solanum melongena* L.) and *Solanum incanum* L. were screened under 150 mM NACl stress. A total of fourteen seedlings at the four-five leaves stage from each of the 50 F_3 lines, accompanied by seedlings of two parents, were examined beside, four seedlings per line served as controls. All stressed seedlings were assessed comparatively with their controls by 0-5 visual scale, on the 12th day following the final salt treatment. Additionally, malondialdehyde (MDA) and proline levels in stressed fresh leaf samples were analyzed. The most tolerant four plantlets from each line were selected and transferred to the greenhouse to generate F_4 seeds. During the greenhouse cultivation period, 13 morphological traits including plant and fruit features, such as plant height, stem diameter, anthocyanin presence, fruit color, and fruit shape etc., were studied. Following the observations, F_3 plants were self-pollinated to produce F_4 generation. Except for a few outliers, the visual scale and proline accumulations showed concurrent increases and reductions. Overall, the results also demonstrate that enhancement of salt tolerance of *Solanum melongena* can be improved using *Solanum incanum* as a donor of alleles.

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ÖZET

Toprak tuzluluğundan etkilenen alanlarda tuza dayanıklı çeşitlerin kullanılması çevre dostu bir çözümdür. Patlıcanın yabani akrabaları bazı abiyotik streslere karşı toleransa sahiptir. Bu çalışmanın amacı, patlıcanda türler arası melez programından geliştirilen F_3 popülasyonunun tuza tolerans durumunu belirlemektir. Arzu edilen özelliklere sahip bir kültür patlıcanı saf hattı BATEM-TDC47 (*Solanum melongena* L.) ve *Solanum incanum* L. arasındaki melezlemelerden geliştirilen 50 F_3 hattı ve ebeveynlerinden 4-5 yapraklı büyüme aşamasındaki on dörder bitki 150 mM NaCl stresi altında test edilmiştir. Her bir hat ve ebeveyn için dörder adet bitki de kontrol olarak kullanılmıştır. Son tuz uygulamasından sonraki 12. günde bitkiler 0-5 görsel skalası kullanılarak değerlendirilmiştir. Ayrıca, stres altındaki bitkilerden alınan taze yaprak örneklerinde malondialdehit (MDA) ve prolin düzeyleri analiz edilmiştir. Yapılan gözlemler sonucu her hattan tuza en dayanıklı dört bitki seçilerek F_4 jenerasyonunu üretmek için seraya transfer edilmiştir. Serada normal koşullarda yürütülen yetiştirme dönemi boyunca bitki boyu, gövde çapı, meyve rengi, meyve şekli ve antosiyanin varlığı gibi bitki ve meyve özellikleri de dâhil olmak üzere patlıcan için önemli 13 morfolojik özellik incelenmiş ve bu esnada

Bahçe Bitkileri

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F_3 bitkileri kendilenerek F_4 nesli üretilmiştir. Araştırma sonuçları değerlendirildiğinde, görsel skala ve prolin birikimlerinin bazı istisnalar dışında paralel artışlar ve azalışlar gösterdiği belirlenmiş olup, *S. melongena*' nın tuz toleransının arttırılması için *S. incanum*' un polen donörü olarak kullanılabileceği açıklanmıştır.

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INTRODUCTION

Sustainable agriculture is under threat of salinity increase in soil, this turned into a serious problem on a global scale and approximately one billion hectares' arable land has been affected by salinity (Ivushkin et al., 2019). Generally, salt stress restricts the water and nutrient uptake by roots from soil, thus plant growth reduces and eventually plants die under constant exposure to stress. The majority of plants are sensitive to salt stress, and they can't remain alive under high stress or their yield decreased (Dasgan et al., 2002). In particularly, vegetables are more sensitive to salinity compared to the other major crops (Chinnusamy et al., 2005) and they are classified as glycophytes (Hannachi and Van Labeke, 2018; Brenes et al., 2020a). Eggplant is known as moderately sensitive to salt stress however for sufficient fruit yield, salinity threshold value in soil should be under 1.5 dS m^{-1} (Ünlükara et al., 2010). A salt concentration in soil, equivalent to 100–200 mM NaCl causes in glycophytes to exhibit total inhibition of growth (Tang et al., 2015; Brenes et al., 2020a). Halophytes can sustain their life at level of 200 mM NaCl and above soil salinities (Himabindu et al., 2016). Salinity effects on plants may vary depend on various factors; however, tolerance to salt stress, mainly in control of genotype (Sumalan et al., 2020). Utilizing tolerant varieties on affected lands is the most prominent environmentally friendly solution. Tolerance or resistance of plants can be improved by using several techniques such as interspecific hybridization (Plazas et al., 2019; Cebeci et al., 2024), with the interspecific hybridization, useful genes from wild relatives could flow to the gene pools to improve salt tolerance levels of crops (Sekara et al., 2007).

Described as moderately sensitive to salinity (Maas, 1990; Ünlükara et al., 2010), it is urgent to improve resilient varieties to sustain of eggplant production on agricultural lands under salinization threat. As well as local genotypes are valuable gene sources to enhancement of new tolerant varieties (Ayranci and Bagci, 2020), eggplant has significant wild relatives such as S. insanum, S. incanum, S. linnaeanum and S. sisymbriifolium which are known have more tolerance to abiotic stresses and it can be crossed with many of its relatives (Plazas et al., 2019). Successful crossings of S. melongena with S. incanum, S. aethiopicum and S. torvum have been reported previously (Daunay et al., 1991, Nee, 1999, Kameswara Rao, 2011; Cebeci et al., 2024). Breeding programs for tolerance to salt stress have been newly started in eggplant (Brenes et al., 2020b; Ortega-Albero et al., 2023). Successful results have been reported for almond (Dejampour et al., 2012), cotton (Tiwari et al., 2013), rice (Kilian et al., 2021) and Egyptian clover (Dwivedi et al., 2022). In a study conducted by Ortega-Albero et al., (2023), compared the responses of cultivated eggplant, S. insanum L., and their interspecific hybrid, under 200 and 400 mM NaCl stress by determining some growth parameters and the levels of biochemical stress markers. At the end of the study researchers stated that, the hybrid S. melongena x S. insanum presented an obvious heterotic effect in terms of plant growth, and it was found as being more salt-tolerant than two parents. Phylogenetic analysis by Lester and Hasan (1991) was revealed very close relationship between S. incanum and S. melongena, thus, interspecific hybridization between them expected to exhibit better heterosis. Although several studies have identified desirable tolerance features of S. incanum L. (Gramazio et al., 2016), its potential under salt stress was reported for the first time by Cebeci et al. (2024).

Under stress conditions, each plant species can continue their growth depending on the efficiency of the stress response mechanisms. There are two main types of tolerance mechanisms in plants; one of them is protection for ion toxicity and osmotic stress and the other is osmolyte biosynthesis and protection for oxidative stress (Ortega-Albero et al., 2023). A commonly used indicator of lipid peroxidation in plant tissue that elevates in response to oxidative stress is the level of malondialdehyde (MDA). In general, salinity-sensitive plants have a greater MDA content than salinity-tolerant ones (Yasar, 2003). Proline, which is an amino acid, plays a major role to protect plants from various stresses. Higher proline accumulation in plants results higher tolerance to stress conditions. Also, it facilitates the plants to recover from stress in a quick way (Hayat et al., 2012). The success of plant breeding programs is dependent on establishing a strong modelling basis which is constitute pyramid the different traits constantly measured at different times. Breeding objectives often concern improving multiple quantitative traits simultaneously, such as yield, quality, and resistance to both biotic and abiotic stress factors. These traits are monitored by observations or tests made on lines or segregating populations obtained at different stages of breeding programs. (Araus and Cairns, 2014; Vieira et al., 2025). Researchers employed several techniques to determine genetic diversity of a germplasm such as principal component analysis (PCA) which is a multivariate statistical method has capability to convert many similar correlated factors into fewer factors named principal components (Ziegel, 2002).

This study aimed to assess of possibilities of successful introgressions of salt tolerance traits from S. incanum to the cultivated eggplant. The plant materials identified as tolerant (50 F3 lines) will assist in further improvement of salt tolerant eggplant lines and varieties.

MATERIAL and METHOD

Plant material

This research is a part of a comprehensive project titled "Development of Tolerant Inbred Lines to Salt and Drought Stresses in Eggplant through Interspecific Hybridization". Previous studies by Cebeci et al., (2024), F_2 segregating population (256 seedlings) and parents; high yielding eggplant inbred line (BATEM-TDC47) and salt tolerant wild relative *Solanum incanum* L., were tested under salt stress. In terms of growth parameters and biochemical analysis, 50 individuals were determined as salt tolerant. In this study, these 50 F_3 lines were tested to reveal their salt tolerance potential within the scope of the mentioned project.

Stress Treatment

A total of 30 seeds per line were sown in viols (15x10) on August 10, 2022. Uniform seedlings with 2-3 true leaves were transferred to the 1 litre pots. After two weeks, seedlings with 4-5 true leaves in 1 litre pots were subjected to salt stress in 100 m^2 semi-controlled compartment conditions. During the experiment, mean temperature and humidity were recorded as 30 ^oC and 70% with HOBO data logger. For the test, 14 seedlings in the 4-5 leaves growth stage per F_3 lines and parents, a totally of 728 seedlings, were evaluated under severe salt stress conditions generated by applying 150 mM NaCl solution. The salt solution was administered in daily aliquots of 50 mM. In addition, 4 seedlings per F_3 lines and parents, 228 seedlings, were employed as controls and irrigated with similar amount of nonsaline water. On the 12^{th} day of the salt treatment on October 17, four seedlings were selected as salt tolerant, considering symptom severity using 0-5 visual damage scale (Kiran, 2015; Cebeci et al., 2024) (Table 1) and transferred to the 650 m^2 greenhouse to produce F_4 generation. During this period, some of their morphological features were noted.

MDA and Proline Determination

On the 12^{th} day after the last salt treatment, fresh leaf samples from each line and parent were collected to MDA and proline analysis. Measurement of malondialdehyde (MDA) was conducted on fresh leaf extracts by the trichloroacetic/thiobarbituric acid method as described by Hodges et al. (1999). The 50 mg ground fresh leaf samples were homogenized in 80% (v/v) methanol, and the extract was centrifuged at 3000xg for 10 minutes at +4°C. The supernatant was divided into two parts, and A and B mixtures were prepared with them. The mixtures A and B were prepared as follows: A: 1 ml extract, 1 ml 20% TCA (Trichloroacetic Acid), B: 1 ml extract + 1 ml 20% TCA (Trichloroacetic Acid) + 0.65% TBA. These samples were incubated at 95°C for 20 minutes, then cooled on ice and centrifuged at 12,000 g at 4°C for 10 minutes. After stopping the reaction on ice, the absorbance of the supernatants was measured at 532 nm. The nonspecific absorbance at 600 and 440 nm was subtracted, and malondialdehyde concentration was determined using the equations by Hodges et al. (1999) and expressed as µmol g^{-1} FW. Proline concentration was detected in fresh plant material by using the ninhydrin-acetic acid method according to Bates et al. (1973). Proline was extracted in 3% aqueous sulfosalicylic acid, the extract was mixed with acid ninhydrin solution, incubated for 1 h at 95 °C, cooled on ice, and then extracted with two volumes of toluene. The absorbance of the supernatant was read at 520 nm, using toluene as a blank. Proline concentration was expressed as µmol g^{-1} FW.

Morphological measurements

The salt-treated seedlings in pots were evaluated using 0-5 visual damage scale as presented in Table 1 (Kiran et al., 2015; Cebeci et al., 2024), and the most tolerant four seedlings were selected per F_3 lines. These selected salt-tolerant lines were transferred to planting sites in the greenhouse to produce F_4 generation following stress treatments in pots. Morphological observations were performed on plants grown under normal conditions, including some phenotypic traits. During the greenhouse period, under normal irrigation conditions, some of their fruit and plant features, such as plant height, stem diameter, fruit color, fruit shape, leaf size, anthocyanin presence, were studied (Table 1) to determine the features of salt-tolerant lines. A total of 13 phenotypic features, regarding plant and fruit characteristics derived from the International Union for the protection of new varieties of plants (UPOV, 1992), were visually observed once 50% of the plants in a genotype had started to fruit set.

Table 1. The visual rating scale for the morphologic features of eggplant used in the study*
*Çizelge 1. Denemede kullanılan morfolojik değişimleri gösteren parametreler**

No	*Features (Özellikler)*	*Ratings (Puanlama)*
1	Plant height	Short: 1; Medium:3; Long:5
2	Stem diameter	Narrow:1; Medium:3; Wide:5
3	Growth habit	Closed:1; Open:3
4	Fruit colour	White:1; Purple:3; Green:5
5	Seconder fruit colour	White:1; Purple:3; Green:5
6	Fruit colour distribution	Plain:1; Mottled:3; Netted:5
7	Fruit shape	Round:1; Oval:3; Long:5
8	Prickliness	Presence:1; Absence:3
9	Hairiness	Less:1; Medium:3; Very:5
10	Leaf size	Small:1; Medium:3; Large:5
11	Anthocyanins	Absence:1; Very:3; Less:5; Medium:7
12	Fruit curvature	Presence:1; Absence:3
13	Sepal size	Small:1; Large:3
14	Salt stress symptoms	0:no effect; 1:local yellowing and curling of leaves with slow growth; 2; necrosis and chlorosis on 25% of the leaf; 3: necrotic spots on the leaves and defoliation by 25-50%; 4: necrosis by 50-75% and death of several plants; 5: severe necrosis on leaves by 75-100% and/or predominant plant deaths

*: The first 13 observations were noted during greenhouse period and the last one was noted from seedlings in pots.
*: İlk 13 gözlem seraya aktarılmış bitkilerden, en son gözlem ise saksıdaki fidelerden alınmıştır.

Data evaluation

Scale results were used to drawn pivot charts (Figure 2) in Microsoft Office Excel program (version 2016). For the clarification of the results, correlation graphic was created using MDA, proline and 0-5 visual scale data. Whole data derived from the measured parameters in greenhouse were subjected to multivariate analysis through a Principal component analysis (PCA) which was explained which parameter is more effective in explaining the variation, before running the program all data were standardized. Additionally, whole data was used to Hierarchical clustering of the 50 F_3 eggplant lines and parents using Ward's method. Data were analyzed using the software program PAST version 4.03.

RESULTS and DISCUSSION

One of the permanent solutions to mitigate detrimental effects of salt stress is selection and breeding of tolerant varieties which can grow under abiotic stress conditions (Dasgan et al., 2002). Therefore, this study aimed to reveal salt tolerance potential of F3 segregating population. For this purpose, seedlings of 50 F3 lines were exposed to 150 mM NaCl stress in pots during the four-five leaves stage. Previous year F2 population were tested under similar conditions by Cebeci et al. (2024), in terms of acquired results, in the present study 0-5 scale and MDA, proline accumulations were considered to find differences. Visual observation of the plants after 12 days of treatment with increasing salinity proved that salt stress inhibited growth of S. incanum, BATEM TDC47 and F3 lines as compared to their non-stressed controls. Reductions in growth observed among the F3 lines highly variable. Although both species and F3 lines were affected negatively under salt application, the detrimental effects of salt stress were more obvious in BATEM TDÇ47 than in S. incanum. As other wild relatives, S. incanum has species specific vegetative developmental stages and generally wild relatives initially presents slow growth when compared to the cultivated ones, however this slow growth in the beginning mostly is an evident of strong plant which has some tolerances in the further vegetative stages.

Because of evaluation is less laborious, cheaper, and less time-consuming, screening studies for salinity tolerance are generally conducted on germination, seedling, and young plant stages (Akinci et al., 2002; Dasgan et al., 2002). The identification of salt-tolerant plants generally depends on classic phenotyping, visually assessing of symptoms and measuring of agronomic and physiologic traits. For visually assessing, mostly damage scales were effectively used by the researchers for screening in some vegetable crops like melon (Kuşvuran et al., 2007), eggplant (Kiran, 2015; Cebeci et al., 2024), tomato (Dasgan et al., 2002), basil etc. (Bekhradi et al., 2015). In this study, at the 12th day of the last salt treatment, all seedlings were evaluated considering 0-5 visual damage scale, and results for segregating population were ranked between 1.14 – 2.93. The visible symptoms of salt stress in sensitive plantlets under salt stress were presented in Figure 1.

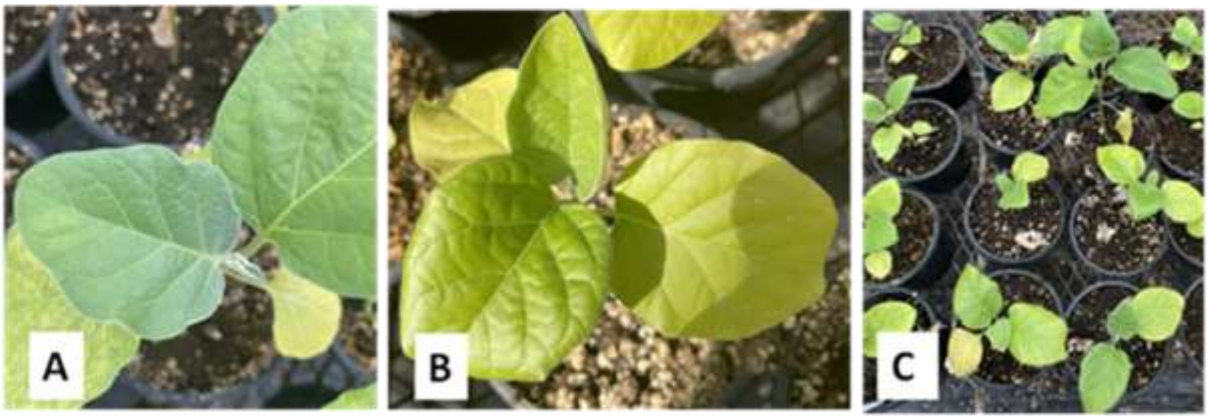

Figure 1. Responses to salt stress of F3 lines; A: putative salt tolerant, B: sensitive, C: distribution of F3 lines in terms of salt tolerance

Şekil 1. F3 hatlarının tuz stresine verdiği cevaplar; A: tuza tolerant, B: hassas, C: tuz toleranslılık bakımından F3 hatlarının dağılımı

Solanum incanum exhibited the least damage and got 1.14 point from scale evaluation and BATEM TDC47 had 1.71 scale point. Distribution of F3 lines according to their visual symptoms under salt stress were depicted in Figure 2. While the scale value of 13 F3 lines varies between 1 and 1.49, most of the lines were placed among the range of 1.5-1.99. However, 11 F3 lines had higher scale values compared to the female parent. Solanum melongena has been described as moderately sensitive or tolerant to salt stress, the response largely depends on the genotype (Plazas et al., 2019). Ortega-Albero et al. (2019) reported in their study on comparison the behavior of an eggplant cultivar (MEL), the wild relative S. insanum (INS) and their hybrid (HYB) that both parents, closely related genetically, would not differ in the type of responses to salt stress but could differ in the magnitude or efficiency of those responses. In the present study, it was seen that the female parent was moderate tolerant to salt stress. However, some of the F3 lines demonstrated sensitivity compared to the female parent. Although a study on the responses to salt stress in wheat at the seedling stage was stated that several genotypes outperformed their parents as revealing positive mid-parent transgressive segregations (Dadshani et al., 2019). In this study negative transgressive segregations were observed during evaluations of visual symptoms on eggplant seedlings. Similarly, in a study conducted on durum wheat, it was reported that no strong genetic correlation was determined on tolerance levels and the measured parameters among the segregating population (Genc et al., 2010).

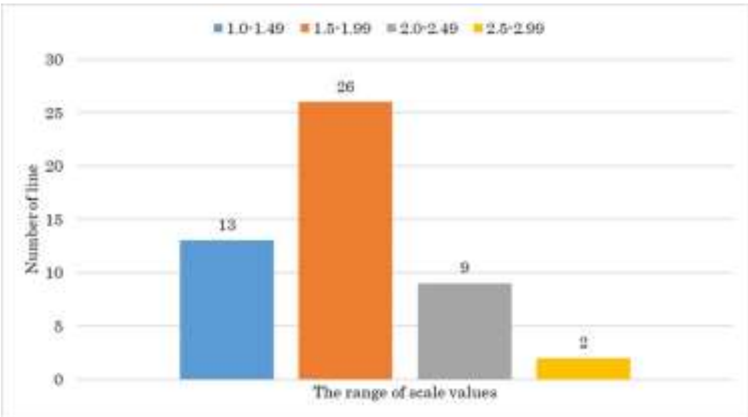

Figure 2. Distribution of F3 lines according to 0–5 rating scale generated from visual symptoms of plantlets

Şekil 2. F3 hatlarının fidelerinde oluşan görsel semptomların 0-5 görsel skalasına göre dağılımı

In a study, BC_1 population derived from backcrosses between *Lycopersicum esculentum* and *Lycopersicum pennellii* in tomato, interactions between tolerance and family were generally found insignificant and low heritability was reported (Saranga et al., 1992). This might be due to the involvement of many gene families in salt tolerance response of crops. In fact, in the mapping study conducted in cotton, 128 genes from 3 gene families associated with salt tolerance were identified. It was explained that such abnormalities in the estimated frequencies of Mendelian ratios in a segregating population were segregation distortion (SDR). In their study, SDRs were detected in salt stress-related genes segregating in the genetic map of the F_2 generation derived from interspecific crosses (Shehzad et al., 2021). Prohens et al. (2012) emphasized that distribution range and accompanying variance for some traits evaluated in the segregating generations (F2 and BC1P2) were generally greater than those observed in the non-segregating generations (P1, P2 and F1) in eggplant. In this study, it was observed that the variance for salt stress response was quite high among the F_3 family. In a study conducted by Brenes et al. (2020) young plants of *S. melongena* and *Solanum torvum* subjected to salt stress under 0-100-200 and 300 mM NaCl doses, their results indicated that *S. torvum*, which is a common rootstock plant for eggplant was found more tolerant than *S. melongena* at high salt concentrations. Consistent with this study, wild relative *S. incanum* presented better resilience under salt stress than cultivated eggplant; additionally, performance of some F_3 lines was found similar to be *S. incanum*. Adaptation capability of the lines is important, although female parent of the study, called as a sensitive parent it is not sensitive as the regular lines, it has good adaptation skills and have fine marketable fruits, male parent of the study can be cultivated on a broad ecology from continental Africa to Southwestern Europe, China or continental America. Therefore, it is possible to develop salinity tolerant eggplant lines among these F_3s suitable for different ecologies.

After visual evaluation by 0-5 scale, leaf samples from all stressed young plants and parents were collected for MDA and proline analysis. Proline and MDA values of segregating population was ranking between 5.02- 14.32 µmol g^{-1} FW and 3.48-22.26 µmol g^{-1} FW respectively. Although increments in proline level means that plant has some tolerances (Hayat et al., 2012) to the stress conditions and increments in MDA level means sensitivity of the plants to the stressed conditions (Yaşar, 2003). Therefore, as expected, *S. incanum*'s MDA level was found among one of the lowest values (7.89 µmol g^{-1} FW) conversely, its proline amount was found as the highest (12.98 µmol g^{-1} FW) after one of the F_3 genotype (14.32 µmol g^{-1} FW) (Figure 3).

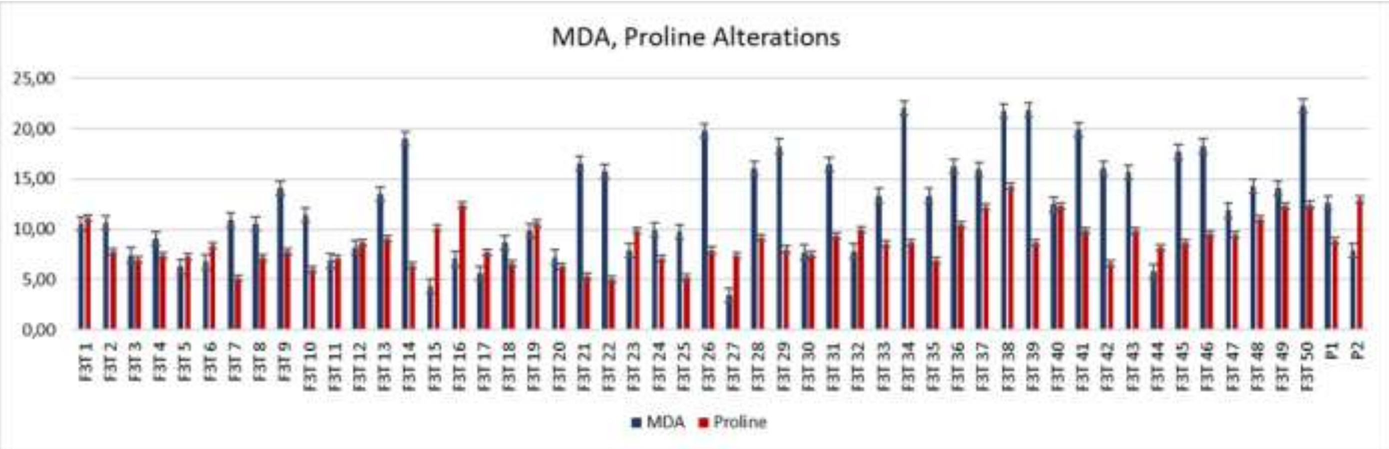

Figure 3. MDA (µmol g^{-1} FW) and proline (µmol g^{-1} FW) alterations of F_3 population, *S. incanum* L. (P2) and Batem TDC47 (P1)

Şekil 3. F_3 populasyonu, S. incanum L. (P2) and Batem TDC47 (P1)'de MDA (µmol g^{-1} FW) ve prolin (µmol g^{-1} FW) oranlarındaki değişimler

According to graphic, presented in Figure 3 generally, when MDA values increased, proline values were decreased under salt stress application. One of the significant results of the study, MDA increment in *S. incanum* (P2) lagged behind the proline increment, and individuals such as F3T1, F3T5, F3T6, F3T27 and F3T44 also presented similar results as their male parent in salt-stressed conditions (Figure 2). Different from the present study, Brenes et al. (2020b) reported that increasing of both MDA and proline in *S. torvum* higher than the cultivar eggplant in their salt tolerance study. In other study conducted by Brenes et al. (2020a), similarly, MDA and proline in *S. insanum* showed higher increases than the eggplant by the salt application.

In addition to this, a correlation graphic (Figure 4) was created using MDA, proline, and visual scale results. According to Figure 4, although there were a few outliers, the visual scale and proline accumulations showed concurrent increases and reductions. These outliers observed on visual scale results or MDA and proline analyses may be due to experimental conditions. Therefore, it was clear from the study, during the selection stage of salt-tolerant plants, it should be considered that higher proline, lower MDA amounts, and the lowest scale points.

Diversity among the populations can be calculated by many methods. Evaluation of the morphological parameters provides a basic method for quantifying genetic diversity under various conditions (Hanci and Cebeci, 2019) like salinity stress. In this study, besides fourteen morphologic parameters, MDA and proline analysis results were also used to evaluate F_3 genotypes by principal component analysis. According to results, the first principal component presented the highest variance among the other sixteen components. Additionally, the first six principal components with Eigenvalues >1 contributed 69.99% of the variation among the genotypes (Table 2).

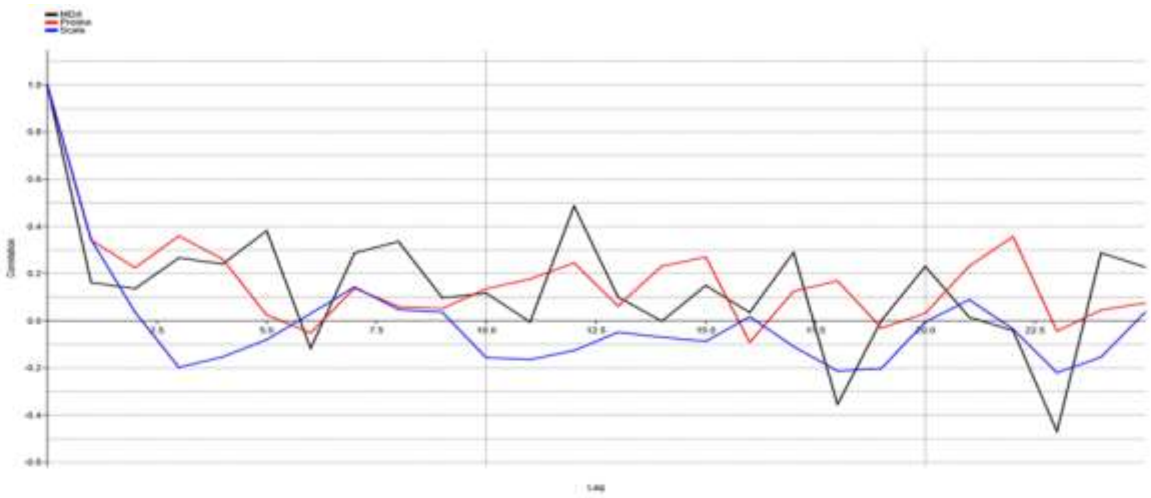

Figure 4. Correlation graphic drawn by MDA, proline and scale results
Şekil 4. MDA, prolin ve skala sonuçlarına göre çizdirilen korelasyon grafiği

Table 2. The first six Eigen values and percentage of variation for each principal component
Tablo 2. İlk 6 Eigen değeri ve herbir temel bileşenin varyasyon yüzdesi ile kümülatif varyasyon derecesi

Principal Component (Temel Bileşen)	*Eigen Values (Eigen değerleri)*	*Variance % (Varyasyon %)*	*Cumulative variance % (Kümülatif varyasyon %)*
1	3.05	19.08	19.08
2	2.06	12.90	31.99
3	1.92	11.97	43.96
4	1.72	10.78	54.74
5	1.34	8.39	63.12
6	1.09	6.87	69.99

The distributions of both genotypes (Figure 5-A) and parameters (Figure 5-B) were examined on the coordinate plane. The origin point on the coordinate plane is the area with the least variation, the close positioning of the parameters or genotypes indicates that there are many similarities between them. With respect to PCA analysis, the first six components accounted for 69.99% of the total variation for salt stressed F_3 genotypes and parents. Bi plot revealed obvious differences between male parent *S. incanum* (P2) and female parent (P2) and F_3 genotypes (Figure 5). Female parent BATEM TDC47 (P1) has good marketable capacity thus, it is understood from Figure 5, F_3 genotypes near the P1 may also have good marketable capacity. Moreover, parent *S. incanum* (P2) was distributed far from the other genotypes of the study which means that selected F_3 genotypes have tolerance to salt stress, besides have better morphological behavior than *S. incanum* (P2). Different plant species (wild relatives) and cultivars within a crop species exhibits great differences in their response to salt stress (Marschner, 1995) and genetic variations in a species considered as a beneficial tool for screening and breeding to tolerance to stress (Dasgan et al., 2002). In addition to this, bi-pilot also revealed strong correlation between MDA accumulation - anthocyanin presence and plant height – stem diameter.

Additionally, observed data was used to Hierarchical clustering of the 50 F_3 eggplant lines and parents using Ward's method. Dendrogram was divided into two main groups and the sub-groups. While group A was formed with 20 F_3 lines beside sensitive parent (P1) and tolerant parent (P2), the group B was formed 28 F_3 eggplant lines. Tolerant parent (P2) was formed as a separate subgroup under the group A.

CONCLUSION

In conclusion, the results of plant growth measurements and biochemical analysis indicate that *S. incanum* has more tolerance to salt stress than *S. melongena*. This is mostly because of its ability to accumulate higher concentrations of proline and lower concentrations of MDA. Additionally performance of F_3 population generally found between male (*S. incanum*) and female (BATEM TDC47) parents. Finally, *S. incanum* can contribute to the development of salt-tolerant eggplant pure lines and cultivars as a pollen donor in further breeding studies. Moreover, to sustain eggplant production under salt-treated lands, it could also be used as a rootstock.

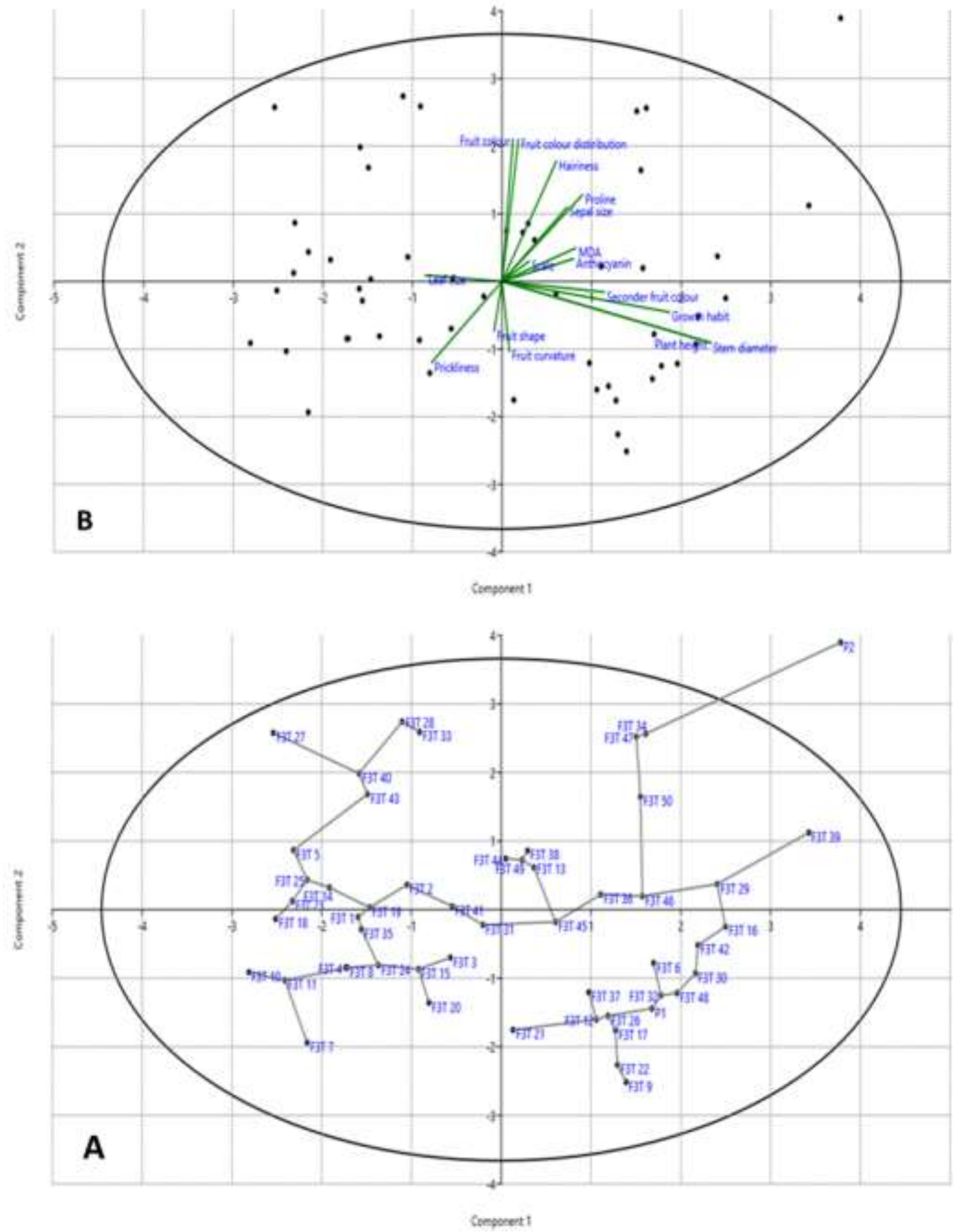

Figure 5. Distribution of F_3 genotypes, parents (A) and employed parameters (B) based on the first and second components

Şekil 5. F_3 hatları, ebeveynler (A) ve kullanılan parametrelerin (B) birinci ve ikinci temel bileşen göz önünde bulundurularak oluşturulan dağılımı

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Contribution Rate Statement Summary of Researchers

EC: Project administration, Investigation, Statistical evaluation, Writing – original draft, Writing – review & editing. HFB: Investigation, Data collection, Writing – original draft, Writing – review & editing. SK: Investigation, Data evaluation, Writing – review & editing. SSE: Data evaluation, Writing – review & editing. All authors read and confirmed the manuscript.

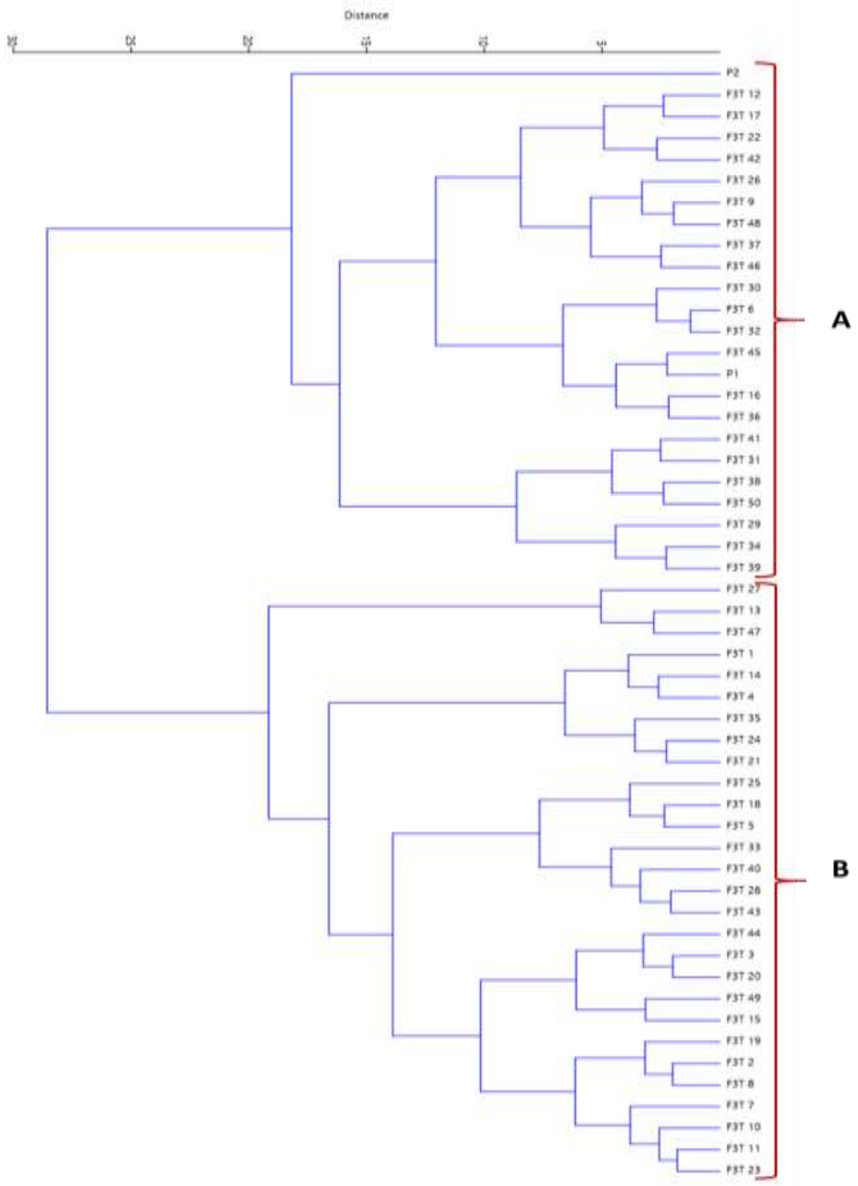

Figure 6. Hierarchical clustering of the 50 F3 eggplant lines and parents using Ward's method
Şekil 6. Ebeveynleri ile birlikte 50 F3 patlıcan hattının Ward's metodu ile hiyerarşik olarak kümelenmesi

Statement of Conflict of Interest

Authors have declared no conflict of interest.

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