

DETERMINATION OF THE EFFECT OF DIFFERENT DOSES OF JASMONIC ACID APPLICATIONS ON ALLEVIATING SALT STRESS IN TOMATO PLANTS WITH PHYSIOLOGICAL PARAMETERS

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Abstract

Salt stress is one of the most important abiotic stress factors that limit productivity in crop production due to climate change, whose effects have been felt more clearly in the world in recent years, and incorrect applications in agricultural soils. Research in plant production in saline soils, based on alleviating the effects of stress through applications to plants or soil, has gained momentum in recent years. In our study, we aimed to demonstrate that foliar jasmonic acid application to tomato plants alleviates the effects of salt stress with physiological parameters. Two tomato genotypes, sensitive to salt stress (TOM 106) and tolerant (TOM 23), determined in previous studies on abiotic stress, were used as plant materials. These two genotype selections were chosen to determine how close jasmonic acid brings the sensitive genotype to the tolerant genotype in salt stress. Salt dose is 100 mM and Control (0mM), jasmonic acid doses are; It was applied as 0, 20, 30 and 40 μ M. The experiment carried out in pots was carried out under controlled conditions. Plant growth parameters (plant fresh and dry weight, stem diameter and plant length) and physiological parameters (such as leaf water potential, leaf osmotic potential, photosynthetic rate, transpiration rate, internal CO₂, stomatal conductivity, chlorophyll) were measured at regular intervals. Jasmonic acid applications created a statistically significant difference. As a result, it was determined that 20 and 30 μ M jasmonic acid applications were more effective in reducing salt stress damage.

Keywords: *Solanum lycopersicum*, Salt stress, Jasmonic acid, Physiological parameters, Plant growth

INTRODUCTION

Tomato, which is a rich source of vitamins, lycopene, and minerals, is the most produced vegetable in many parts of the world with its nutritious and delicious characteristics. Tomato is an important greenhouse and field vegetable crop grown in the semi-arid regions of Mediterranean countries. According to FAO data, Turkey ranks 3rd after China and India in tomato production in the world with a production of 13 095 258 tons and ranks 5th in world tomato exports with 617 000 tons (FAO, 2021). Many economically important plants, such as tomatoes, are sensitive to salinity. Salinity in soil and irrigation water significantly affects yield and quality in tomato production. It is possible to group growth inhibiting factors into three groups for a plant growing in salty environments as follows; a) Decreased water intake because of low water potential in the root zone or, in other words, water stress, b) Accumulation of Na⁺ and Cl⁻ ions in the plant, which increase to a level that causes ion toxicity, c) Imbalances during the uptake

and transport of nutrients, and especially the emergence of K^+ and partly Ca^{+2} deficiency (Munns and Terster, 2008; Kamran et al., 2019; Stavi, 2021; Karanlık, 2001; Tunçer, 2007). Ion toxicity, lack of water, nutritional imbalance, and high salt in the root zone inhibit normal tomato growth and development at significant levels. Although tomato is grown in a wide climatic zone, its production is concentrated in hot and dry areas where saline soils are the limiting factor. Although these areas are optimum conditions for tomato production, salinity is a serious problem. Commercial tomato genotypes are generally sensitive to salinity during the entire development period. When the EC exceeds 2.5 dS/m in the soil solution, tomato fruit yield begins to decrease (Maas, 1990; Saranga et al., 1991). According to this threshold, this causes a decrease of approximately 10% in tomato yield with an increase in EC of 1 dS/m (Saranga et al., 1991).

Jasmonates are fatty acid-derived cyclopentanones in the plant kingdom in plants and are accepted as stimulants or signaling agents that induce the production of antimicrobial and antifungal compounds against microbial, fungal, and physical stimuli under biotic and abiotic stress conditions. MeJA (Methyl Jasmonate) is naturally found in many higher plants. It was reported that when plants face stress conditions, jasmonates also promote the synthesis of certain proteins (jasmonate-induced proteins). These proteins were found in all plants tested in previous studies conducted on jasmonates. It is widely accepted that Jasmonic Acid is a signaling molecule activating the defense mechanism. It was also reported that Jasmonic Acid stimulates the production of various defense substances such as alkaloids and phenolic substances when the plant is under stress (Redman et al., 2001). It was reported that jasmonates have effects on fruit development. It was also reported that Methyl Jasmonate has effects on fruit ripening, coloration, softening, and starch loss, similar to ethylene, and Jasmonic Acid increases anthocyanin formation in apples (Concha et al., 2013; Fan et al., 1998; Fan and Mattheis, 1999). Jasmonic Acid, which was applied externally in the preclimacteric period in tomatoes and apples, increased ethylene biosynthesis and coloration. It was reported that it was due to the increase in ACC oxidase and ACC synthase enzyme activity, which are effective in ethylene biosynthesis. In a previous study reporting that MeJA triggers the stimulation of antioxidants and secondary metabolites in plants, it was found that the application of MeJA increased APX activity in *Ricinus communis* significantly (Kim et al., 2009). Chanjirakul et al. (2006) showed that MeJA could play roles in the resistance of tissues to decay by improving antioxidant systems and free radical scavenging characteristics. Similarly, Ghasemnezhad and Javaherdashti (2008) showed that MeJA increased antioxidant power in raspberry and the highest antioxidant activity was recorded in MeJA-treated fruit.

Salinity problem is the second biggest abiotic stress factor after drought stress in the world. As the world population increases, agricultural production must increase for food supply. One of the ideas put forward for the solution of this necessity was to bring new areas into agriculture and the other was to obtain maximum yield per area. Since the first proposal is not easy to implement, the second one has come to the fore as an important opportunity. As the world's human population continues to grow, the demand for agriculture to meet future

food needs will be one of the greatest challenges facing the agricultural community. In order to overcome this challenge, either genotypes resistant to abiotic stress factors due to global warming and climate change will be used or stress-resistant practices will come to the fore in order to utilise existing genotypes in agricultural production. As examples of these applications; we can give applications such as the use of beneficial microorganisms (mycorrhiza, PGPR, etc.), salicylic acid, jasmonic acid that will support the plant in terms of resistance under stress conditions. In this sense, it has been reported in previous studies that Jasmonic Acid will increase the resistance of plants against salinity problem, which is one of the most common abiotic stresses in the world. In this study, the effects of different doses of Jasmonic Acid applied to tomato plants under salt stress on plant growth and physiological parameters were investigated. The hypothesis of the study was that Jasmonic Acid applications would increase the tolerance of tomato, the most produced vegetable in the world, to salinity stress and improve the damage caused by stress in the plant. We aimed to demonstrate this in plant growth parameters and gas exchange parameters (photosynthesis rate, transpiration rate, stomatal conductance). In addition, in a previous study on salt stress, two genotypes defined as salt tolerant and salt sensitive were used. The purpose of using two genotypes with different salt sensitivity was to determine to what extent Jasmonic acid treatment assisted the sensitive genotype in salt stress, to what extent it reduced salt stress damage and to what extent it prevented damage in the tolerant genotype. The aim of this study, the results of which are presented here, is to increase the resistance of the plant under stress conditions by applying Jasmonic Acid to the plant in tomato cultivation in soils with salt problems. The aim of this study is to minimise the yield losses due to salt in tomato, which is the most produced vegetable in greenhouse cultivation, especially in Mediterranean countries. Because greenhouse soils are fertilised more because the production season is longer and the yield is higher compared to open field cultivation. Since they are closed areas, it is not possible for the salt to go deeper with precipitation. Therefore, greenhouse soils are more salty. Although soilless greenhouse production has become widespread in recent years, there is still greenhouse production with soil in Mediterranean countries. Therefore, practices to increase resistance to salt stress are important in both open field and greenhouse tomato cultivation.

MATERIAL and METHOD

Tolerant (T) Tom 23 genotype and salt-sensitive (S) Tom 106 genotypes, which were selected among 41 tomato genotypes used in a previous doctorate study (Bayram, 2016), were used as plant materials in the study. The salt doses applied were 0 (control) and 100 mM and the Jasmonic Acid doses applied were 20, 30, and 40 μ M. The experiment was conducted in a climate-controlled polycarbonate greenhouse of the Department of Horticulture, Faculty of Agriculture, Malatya Turgut Ozal University. The seeds were planted in the vials on 20.09.2019 and the seedlings that reached the planting stage (4-5 true leaves) were transferred to 4-liter plastic pots that contained vermiculite as a substrate on

24.10.2019. The plants were watered with $\frac{1}{2}$ Hoagland Nutrient Solution. Jasmonic Acid application was started two weeks after the seedlings were transferred to the pots, in other words, at the same time as the salt application, and was applied by spraying on the leaves once a week until the end of the experiment. The experiment was set up according to the Split-Plot experimental design, with the main plots being salt doses (0 and 100 mM) and the subplots being Jasmonic Acid doses (20, 30, and 40 μM). A total of 240 plants were used in the trial, which was set up with 3 replications (5 plants in each replication, 120 plants from the salt-sensitive TOM 106 variety and 120 plants from the salt-tolerant TOM 23 variety). The following measurements were made on the 15th and 30th days when tomato plants transferred to 4-liter pots were subjected to salt stress.

Plant Growth Parameters and Physiological Parameters Measured in Plants

The following measurements were made on the 15th and 30th days when tomato plants transferred to 4-liter pots were subjected to salt stress. The heights, number of leaves, and stem diameters of the plants were measured on potted plants, and fresh and dry weights of green parts and fresh and dry weights of roots were obtained by removing the plants. The amount of chlorophyll was measured at 3 different points in the leaves with SPAD and averaged.

Relative Growth Rate (g/dry weight/day)

The plants were weighted in terms of total dry weight before the exposure to salt stress and after the stress period was completed, and by dividing the difference between the 2 measurements by the number of days, the growth rates of the genotypes during the stress were determined as g/dry weight/day.

Determining Leaf Water Potential (MPa)

Water potential in MPa was determined in the second leaves of the plants, starting from the growth tip by using a portable pressure circle.

Determining Leaf Osmotic Potential (MPa)

The measurements were made on the leaves with an Osmometer Device. A 1-gram sample was taken from the leaves, made up to 20 grams with 19 grams of pure water, homogenized in a porcelain mortar, 150 μl of the homogenized samples were taken and filtered, and the leaf osmotic potential was determined based on freezing temperature in the Osmometer device (Akhoundnejad and Dasgan, 2019).

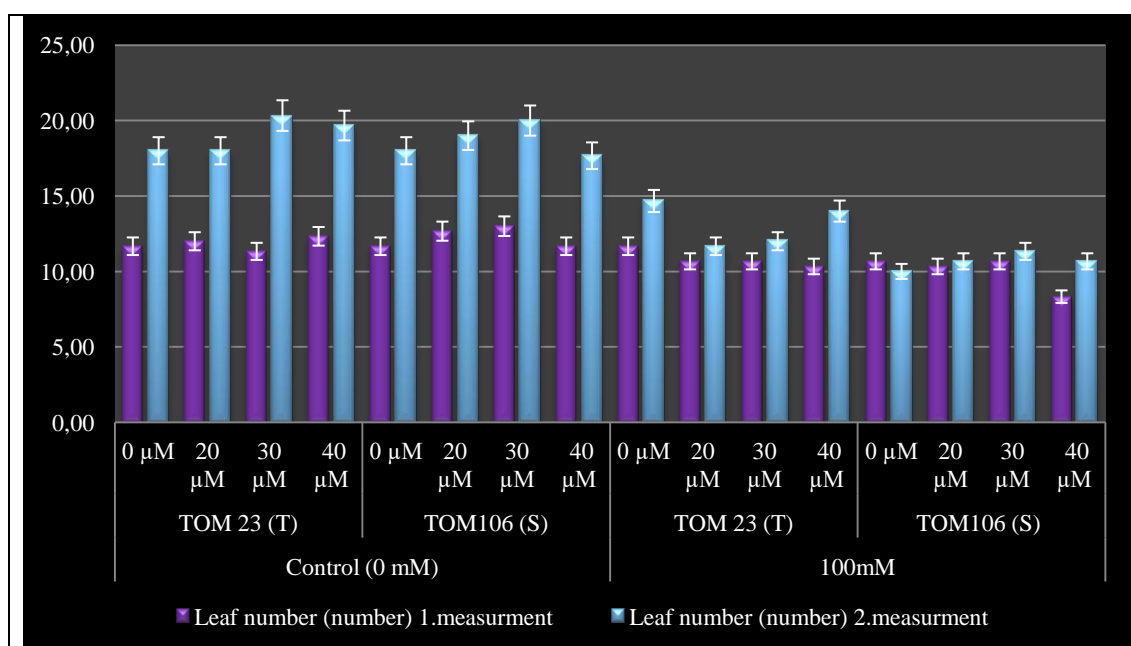
Determining photosynthesis rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), Transpiration rate ($\mu\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$), Stomatal Conductance ($\mu\text{mol m}^{-2} \text{ s}^{-1}$); and Internal CO_2

These parameters were measured in the 3rd and 4th leaves from the growth tip by using the Licor 6400x Portable Photosynthesis Device in leaf tissues.

Results

Plant height, stem diameter, number of leaves

A statistically significant difference was detected between the treatments in terms of plant height, stem diameter, and number of leaves, which are among the plant growth parameters given in Figure 1. When the results were examined, it was seen that 20 μ M and 30 μ M Jasmonic Acid applications of the salt-sensitive variety (TOM106) were related to each other and yielded the highest results in terms of plant height, stem diameter, and leaf number parameters in the Control Group. In salt application, the highest values in plant height, stem diameter, and number of leaves parameters were measured in the plants of the salt-tolerant (TOM23) variety without Jasmonic Acid application, and the lowest results were measured in the Jasmonic Acid applications of the salt-sensitive (TOM106) variety (especially at 40 μ M). In previous studies conducted by Kusvuran (2010) on melon, by Altuntas et al. (2016) on pepper, by Agamy et al. (2013) on tomato, and by Kaya and Dasgan (2013) on bean genotypes, it was found that the height, stem diameter and number of leaves of the plants decreased under salinity stress.



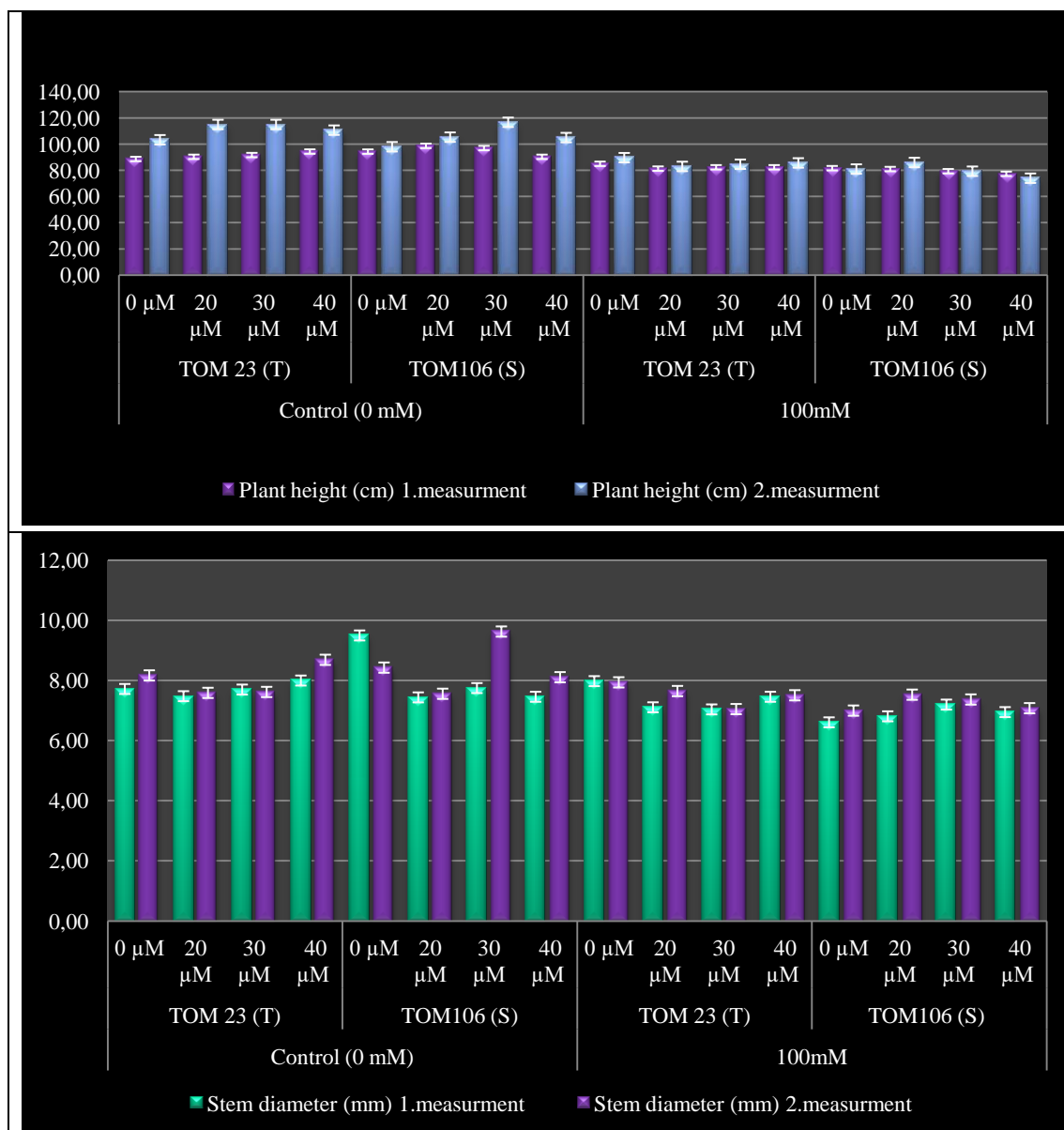


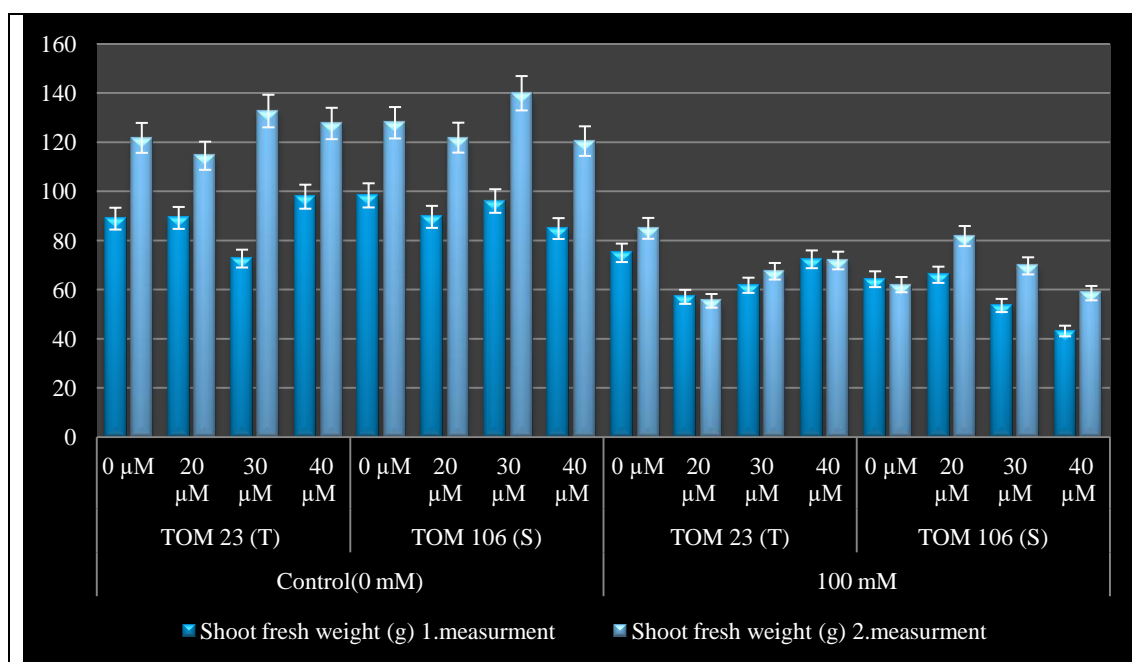
Figure 1. Effect of Jasmonic Acid applications on the plant height, stem diameter and leaf number in the salt stress

The difference between the means shown with different letters in the same column is statistically significant ($p \leq 0.05$)

Fresh and dry weights of the shoot (Stem and Leaves)

The fresh and dry weights of shoot showed statistically significant differences between applications among the plant growth parameters given in Figure 2. In the first measurement of shoot fresh and dry weight values, the measurement results of the 40 μM Jasmonic Acid application of the salt-tolerant (TOM23) variety in the Control Group and the plants of the salt-sensitive (TOM106) variety without Jasmonic Acid application were in the same group in

statistical terms and were obtained the highest result. The Jasmonic Acid application to the salt-tolerant (TOM23) genotype in the Control Group had a positive effect on the shoot weight in the second measurement, and it increased the foliage fresh weight by 8.9%, especially in the plants treated with 30 μM Jasmonic Acid, and by 4.8% in the plants treated with 40 μM Jasmonic Acid. In the second measurement made on plants in the salt-sensitive (TOM106) genotype, it was found that Jasmonic Acid applications increased the fresh weight of shoots by 9.3% at a maximum dose of 30 μM . In the second measurement of the shoot fresh weight in the salt-treated group, although no positive effects were detected in the application of Jasmonic Acid doses to the salt-tolerant (TOM23) genotype, it was determined that the application of maximum 20 μM Jasmonic Acid to two genotypes (salt-sensitive (TOM106)) increased the fresh weight of shoots by 31.8%, and 31.8%, followed by 40 μM Jasmonic Acid that increased the fresh weight of shoots by 12.2% (Table 2). Mohammad et al. (1998), Dasgan et al. (2002), and Agamy et al. (2013) conducted studies on tomatoes, Altuntas et al. (2018) on pepper, Kusvuran (2010) in melon, and Süyüm (2011) on watermelon genotypes, and reported that salt stress caused significant losses in fresh and dry weights of shoots.



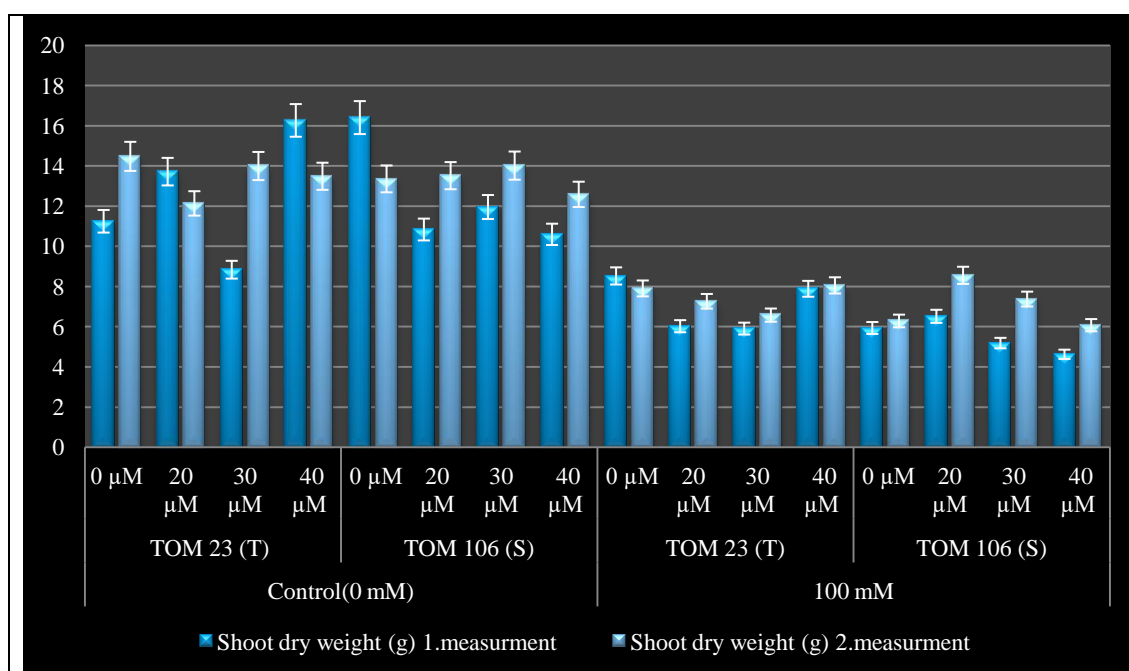


Figure 2. Effect of Jasmonic Acid applications under salt stress on the fresh and dry weights of shoot

The difference between the means shown with different letters in the same column is statistically significant ($p \leq 0.05$)

The root fresh and dry weights

A statistically significant difference was detected between the applications in terms of root fresh and dry weights, which are among the plant growth parameters (Figure 3). In the first measurement of root fresh and dry weight parameters, the highest results were measured in the salt-tolerant (TOM 23) variety with 40 μM Jasmonic Acid application and the salt-sensitive (TOM106) varieties that were not treated with Jasmonic Acid in the Control Group. In the second measurement of root fresh and dry weights, the highest values were measured in plants of the salt-tolerant (TOM 23) variety that was not treated with Jasmonic Acid. In salt application, the lowest values of these parameters were detected in the salt-sensitive (TOM106) variety in the plants without Jasmonic Acid application (Figure 3). In the second measurement of root fresh weight, it was found that the Jasmonic Acid doses applied to the salt-tolerant (TOM23) genotype in the Control Group and the salt-treated group did not have positive effects. It was also found that the 1000 mg dose increased the dose by 33.6%. On the second measurement date, it was determined that the salt-sensitive (TOM106) genotype was increased the root fresh weight by 33.6% compared to the control dose of 20 μM of jasmonic acid treatment. It was determined in the measurements that the root dry weights were directly proportional to the fresh weights of the roots.

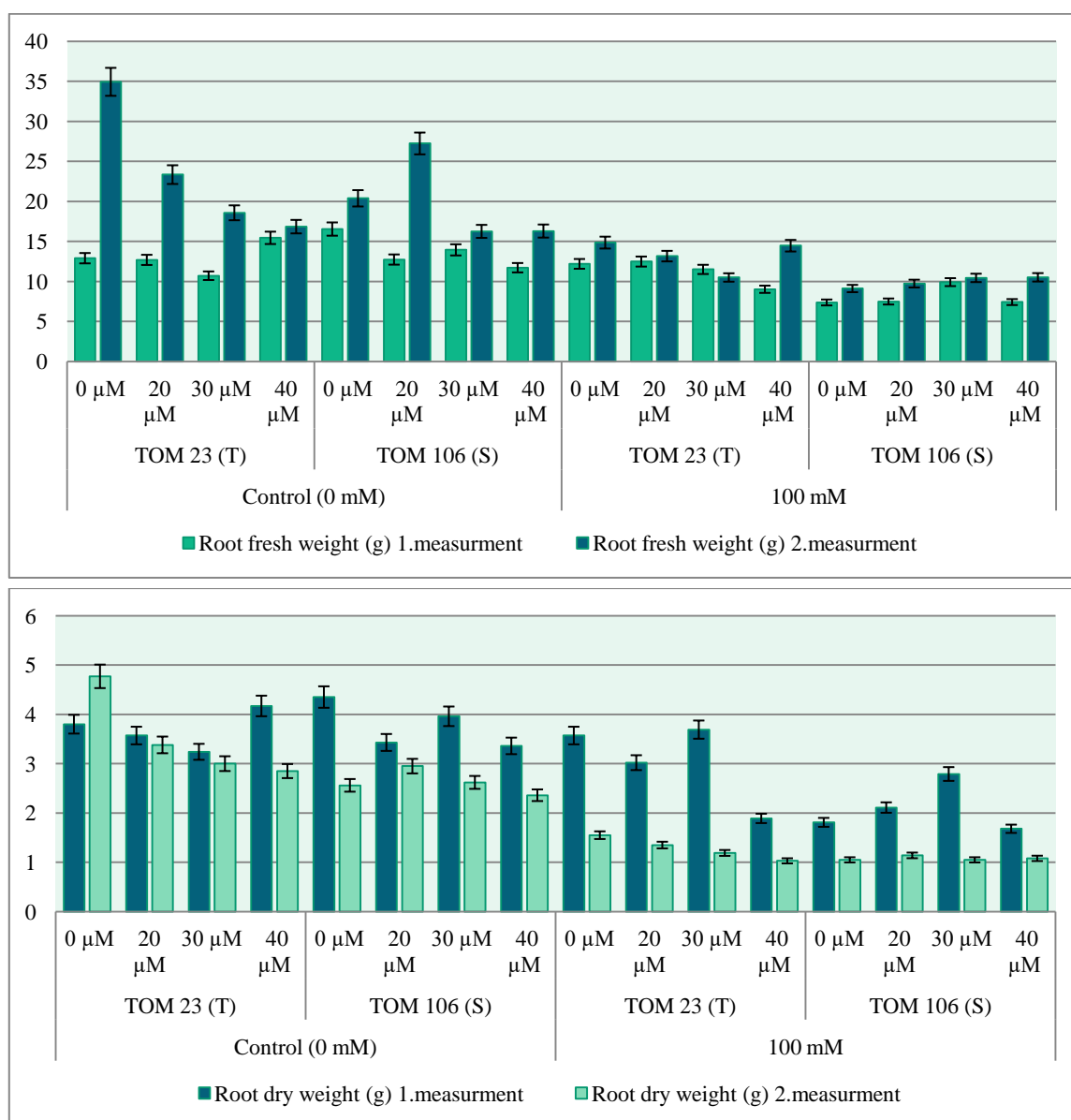


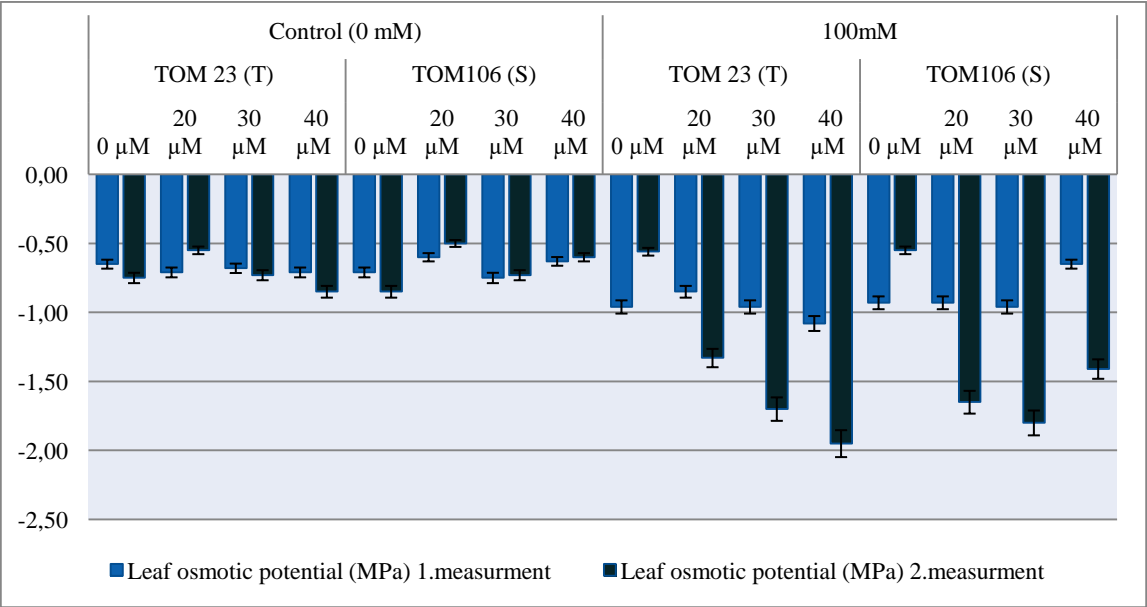
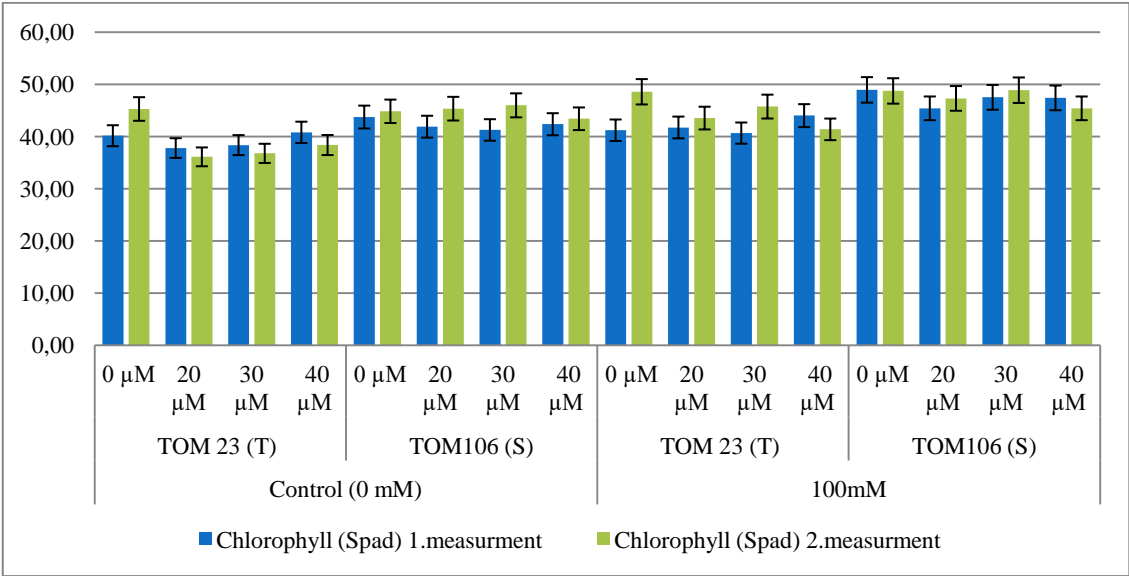
Figure 3. Effect of Jasmonic Acid applications under salt stress on the plant root fresh and dry weights

The difference between the means shown with different letters in the same column is statistically significant ($p \leq 0.05$)

Chlorophyll, leaf osmotic potential, leaf water potential

Statistically significant differences were detected between the applications in terms of chlorophyll, leaf osmotic potential, and leaf water potential (Figure 4). The highest chlorophyll was measured in the plants of the salt-sensitive (TOM106) genotype without Jasmonic Acid application in the first measurement. In the second measurement, the highest chlorophyll was measured in the application of 30 μM Jasmonic acid of the salt-sensitive (TOM106) variety. The lowest values of this parameter were determined in the Control Group, in both measurements, in the 20 μM Jasmonic acid application of the salt-tolerant (TOM

23) genotype. In the first measurement of leaf water potential in the salt-applied group, a positive effect of all doses was observed in Jasmonic acid applications on the salt-tolerant (TOM23) genotype (20 μ M application rate was 42.9%, 40 μ M dose was 28.8%, and 30 μ M application was 100% and was found to increase by 14.6 percent). However, it was also found with the measurements that the dose applications of Jasmonic acid did not have a positive effect on the salt-sensitive (TOM106) genotype. The leaf osmotic potential was obtained the highest results in both measurements in the 40 μ M Jasmonic acid application of the salt-tolerant (TOM 23) genotype. In the studies conducted by Karipcin (2009) and Süyüm (2011) on watermelon, Akyol (2010) on pepper, Akhoundnejad (2011) on tomato, and Altuntas et al. (2018) on pepper, it was found that leaf water potential decreased with an increase in stress conditions. Although the first measurement average of the leaf osmotic potential of the salt-tolerant (TOM23) genotype in the Control Group of the experiment was -0.65 MPa, the first measurement average of the leaf osmotic potential of the plants to which salt was applied decreased to -0.96 MPa. Although the first measurement average of the leaf osmotic potential in the plants in the Control Group of the salt-sensitive (TOM106) genotype was found to be -0.71 MPa, the leaf osmotic potential decreased to -0.93 MPa in the salt-treated group. In other words, the average leaf osmotic potential of salt-tolerant and salt-sensitive genotypes grown in saline conditions seems to be less than that in the Control Group. In a salinity study, it was reported that decreasing the osmotic potential of the soil solution reduced the turgor pressure of the cells and prevented plant development (Ashraf 1994). Salinity can cause serious yield losses by having both osmotic and ion effects on plants. To avoid these harmful effects of salts, plants either reduce their ion intake by making the necessary osmotic adjustments or allow ion intake and store the ions they receive in their roots, stems, and leaves (Kantar and Elkoca, 1998). According to Ashraf (1994), salinity reduces the osmotic potential of the soil solution, reduces the turgor pressure of the cells, and prevents plant development. Salinity can cause serious yield losses by having both osmotic and ion effects on plants. To avoid these harmful effects of salts, plants either reduce their ion uptake by making the necessary osmotic adjustments or allow ion intake and store the ions they receive in their roots, stems, and leaves (Kantar and Elkoca, 1998).



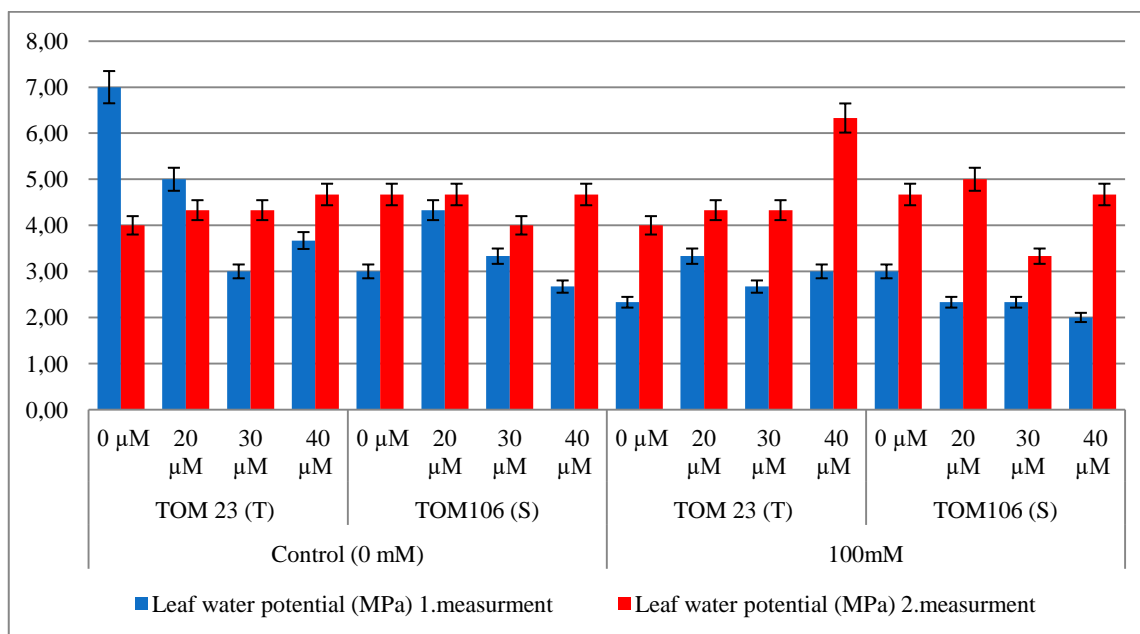


Figure 4. Effect of Jasmonic Acid applications on the chlorophyll, leaf osmotic potential and leaf water potential in tomato leaves under salt stress

The difference between the means shown with different letters in the same column is statistically significant ($p \leq 0.05$)

Photosynthetic rate and stomatal conductance in leaves

According to the results in Figure 5, photosynthetic rate and stomatal conductance values showed statistical differences between applications. In the first measurement date, the photosynthetic rate in the leaves was measured the highest result in the 40 μM Jasmonic acid application of the salt-sensitive (TOM106) genotype. In the second measurement date, it was determined that the 30 μM Jasmonic acid application of the salt-sensitive genotype (TOM106) belonging to the Control Group the highest results. In the stomatal conductance, in the first measurement, from the salt-sensitive genotype (TOM106) in the Control Group to which Jasmonic acid was not applied was obtained the highest results. In the second measurement date, it was determined that the application of 40 μM Jasmonic acid to the salt-tolerant (TOM23) genotype in the Control Group the highest results. The lowest result in photosynthetic rate was obtained in the 20 μM Jasmonic acid application of the salt-sensitive (TOM106) genotype. The lowest result in stomatal conductance was detected in the 20 μM Jasmonic acid application of the salt-tolerant (TOM23) genotype. In the study conducted by Avcu et al. (2013) on tomatoes, it was reported that stomatal conductance under salt stress was 69% lower on average than in control plants. In the study conducted by Kusvuran (2012) on melon genotypes, it was reported that stomatal conductance decreased under salt and drought conditions. A decrease of 40% to 56% occurred in tolerant genotypes. It has been reported that this change varies between 66% and 81% in sensitive melon genotypes. Altuntas et al. (2018) reported that salt stress caused a decrease in stomatal conductance in pepper. Xu et al. (1994) and Parveen and Ashraf (2010) reported that the decrease in

photosynthetic rate was partly due to the decrease in leaf water potential and that photosynthetic function in salt stress depends on sufficient leaf water potential and maintenance of sufficient turgor pressure.

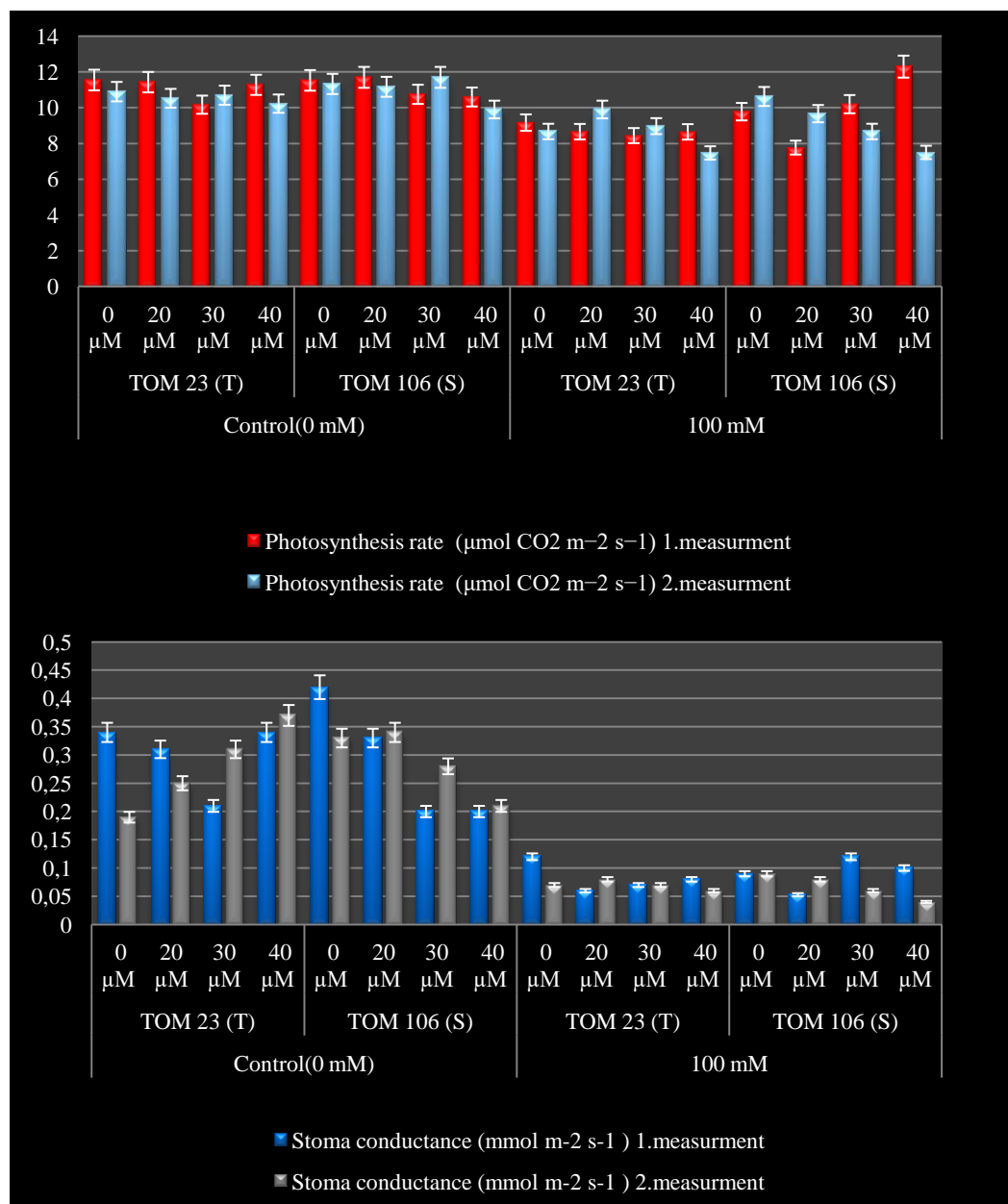


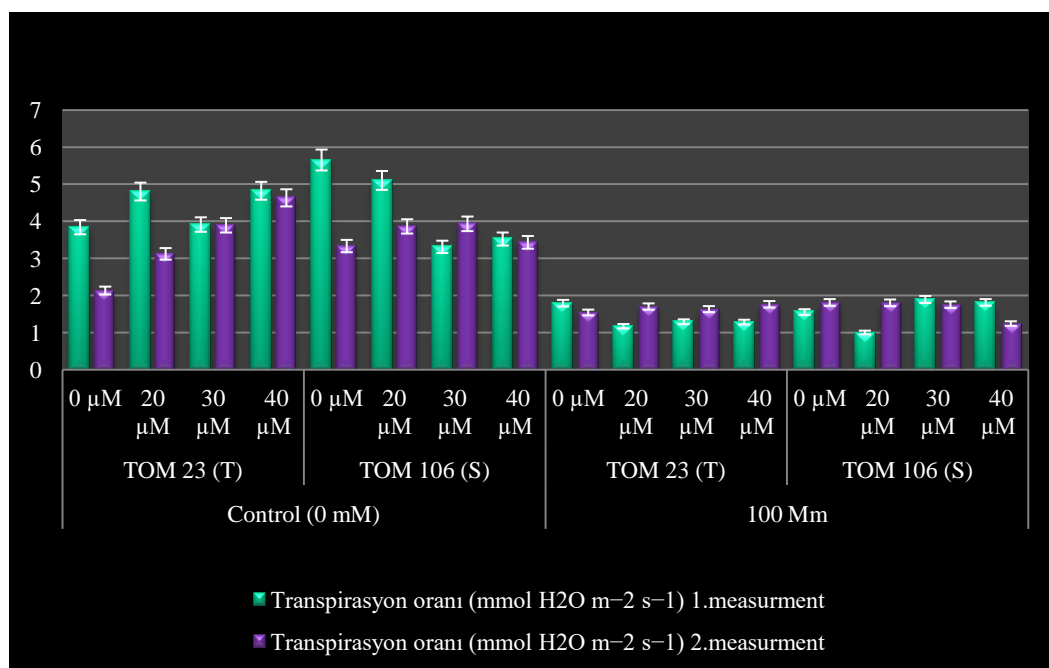
Figure 5. Effect of Jasmonic Acid applications on the photosynthetic rate and stomatal conductance in tomato leaves under salt stress.

The difference between the means shown with different letters in the same column is statistically significant ($p \leq 0.05$)

Transpiration rate and internal CO_2 in the leaves

A statistically significant difference was detected between the applications transpiration rate and internal CO_2 (Figure 6). In the first measurement date, the

transpiration rate was obtained the highest results from the Control Group's salt-sensitive (TOM106) genotype plants, to which Jasmonic Acid was not applied and 20 μM Jasmonic Acid was applied (included in the same group). In the second measurement date, the highest transpiration rate was detected in the 40 μM Jasmonic Acid application of the salt-tolerant genotype (TOM23) of the Control Group. The results in the Control Group were found to be close to each other in the first measurement date of internal CO_2 , and the lowest value was given by 20 μM Jasmonic Acid application of the salt-tolerant genotype (TOM23). Similar results were obtained in the second measurement (30 μM and 40 μM doses of Jasmonic Acid increased transpiration in the sensitive genotype compared to the non-treated one). In the second measurement of transpiration rate, Jasmonic Acid doses applied to the salt-tolerant (TOM23) genotype in the Control Group had positive effects, and it was found that the 40 μM dose increased it by 117.4%, the 30 μM dose by 82.6% and the 20 μM dose by 46.5%, respectively. It was also found through measurements that the 20 μM dose increased the transpiration rate by 15.9%, and the 30 μM dose increased it by 1.8%, among the Jasmonic Acid applications applied to the salt-sensitive (TOM106) genotype in the Control Group. In the second measurement date of transpiration rate, the 40 μM dose had a rate of 14.3%, the 20 μM dose had a rate of 10.4%, and the 30 μM dose had no death effect at a rate of 5.8% among the Jasmonic Acid doses applied to the salt-tolerant (TOM23) genotype increased the transpiration rate in the salt applied group. No positive effect of Jasmonic Acid doses on the transpiration rate was found in the second measurement date of the salt-sensitive (TOM106) genotype. Since plants face physiological drought during salt stress, they tend to close their stomata to reduce water loss. For this reason, the transpiration rate was found to be low in salt application.



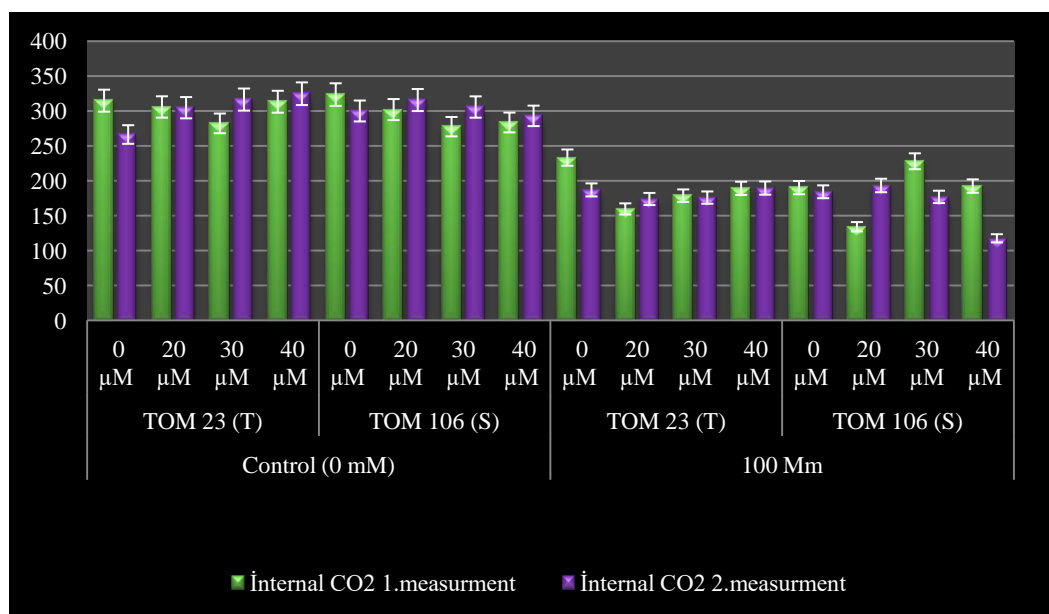


Figure 6. Effect of Jasmonic Acid applications on the transpiration rate and internal CO₂ in tomato leaves under salt stress

The difference between the means shown with different letters in the same column is statistically significant ($p \leq 0.05$)

CONCLUSION

In the present study, which was conducted to increase the resistance of the salt-sensitive genotype compared to the tolerant genotype by spraying Jasmonic Acid doses on salt-tolerant and sensitive tomato genotypes, plant growth and physiological characteristics were examined. Jasmonic Acid did not have a significant effect on plant growth parameters. However, the effect was more significant in sensitive physiological parameter measurements. The 20 μM and 30 μM doses of Jasmonic Acid yielded effective results in the salt-sensitive genotype (TOM106). When the measurements were evaluated, it was found that most parameters were close to the salt-tolerant genotype (TOM23). In conclusion, the use of Jasmonic Acid can be recommended to ensure tolerance to salt stress, especially for indoor greenhouse pepper cultivation. Since there is no study on application to plants during cultivation, the researchers also determined the dose with the present study. Doses between 20 μM and 30 μM may be tried in future studies.

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AUTHORS' CONTRIBUTIONS

All authors contributed equally. All authors read and approved the final manuscript.

Ethical Approval (for researches involving animals or humans)

Not applicable.

CONFLICT OF INTERESTS

The authors declare that there are no conflicts of interest related to this article

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