ECOPHYSIOLOGICAL RESPONSES OF *SOLANUM LYCOPERSICUM* L. TO DIFFERENT LEVELS OF SALT STRESS

GULCIN ISIK

Eskisehir Technical University, Faculty of Sciences, Department of Biology, 26470 Eskisehir, Turkey Corresponding email: glcnylmz@gmail.com

Abstract

Germination is one of the most important stage for the development of plants. One of the factors affecting germination is salt stress. All over the world due to increasing drought and semi-drought conditions most plants face salinity in soil. In this study, ecophysiological responses of *Solanum lycopersicum* "Marmande" seeds under different NaCl concentrations (% 0.5, 1, 2, 50 mM, 100 mM, 150 mM, 0.25 M, 0.45 M) were studied. Germination percentage, germination rate, total biomass, root length, green pixels and salt tolerance index (STI) were measured. The highest germination percentage and rate were observed in 0.5 % NaCl application (97.5%; 79.59) and the lowest were in 0.45 mM NaCl (2.5%; 5.88). Biomass, root length and STI reached the highest value at 50 mM NaCl application (respectively 460, 67 kg/ha, 11, 96 cm, 1,142). The biomass, root length and STI value of 2%, 0,25 M and 0,45 M NaCl applications were very low (respectively 0.001 kg/ha, 0.01, 0.001). There was no seedling development for green pixel count for 2%, 0,25 M and 0.45 M NaCl applications so the value was equal to 0, but other NaCl applications resulted in homogenic groups statistically.

Key words: Biomass, Marmande, Germination, Tomato, Tolerance.

Introduction

All over the world due to increasing drought and semi-drought conditions, most plants face salinity in soil. Salinity is one of the abiotic stress factors which adversely affects plant and agricultural productivity (Talaat & Shawky, 2012; Zhong et al., 2020). Seed germination and seedling growth are the most critical stages in agricultural activities and are also the most sensitive stages to salinity for the future of plant populations (Silva Oliveira et al., 2019). Salt stress causes a decrease in plant growth and productivity by disrupting physiological processes, especially photosynthesis (Sudhir & Murthy, 2004). Ecological factors are affected by pollution which is expected to have increasingly negative effects in the upcoming years and it is estimated that half of the existing agricultural lands will encounter salinity by 2050 (Jamil et al., 2011; Jiménez-Arias et al., 2019). Salinity lowers the water absorption, inhibits metabolic courses, and affects nutrient composition (Feng et al., 2002; Munns, 2005). Salt stress significantly affects fruit quality by causing metabolic changes in plants (Meza et al., 2020). The negative effect of salt on growth and development is the highest in the germination period (Taiz & Zeiger, 2002). The harmful effects of salt stress on agricultural plants are reduction of growth rate and sprouting and inhibition of reproductive development (Munns & Tester, 2008; Roy et al., 2014). Exposure to salinity during the seed germination phase limits water absorption and causes toxicity effects on germinating seeds (Foti et al., 2019). The integrity of cellular membranes, the activities of various enzymes, nutrient acquisition, and function of photosynthetic apparatus are all known to be prone to the toxic effects of high salt stress (Zhu, 2001). Salt stress induces TGase expression in plants, which increases transpiration, K/Na ratio, and photosynthetic system damage (Zhong et al., 2020). To decrease salt stress in plants, IAA producing bacteria inoculation to plants has a positive effect (Barra et al., 2016). It is well known that salinity elicits oxidative stress (Ostrowski *et al.*, 2016). IAA-Asp affects plant responses to salt stress by modulation of catalase and peroxidase activity (Ostrowski *et al.*, 2016). High salt concentrations in the soil increase the content of Ca, Cl and Na in the biomass ash but reduce that of K (Zhuo *et al.*, 2015).

There is interspecies variability in terms of salinity resistance in agriculturally important plant species, and some cultivars are known to be more resistant to NaCl than others (Benazzouk et al., 2020). Plants have different tolerance abilities against boron and salinity; those which are not tolerant to boron can tolerate salinity (Grieve et al., 2012; Smith et al., 2013). Integration of high value vegetable crops into the schedule of rotations with more salt-tolerant agronomic crops has obvious economic advantages (Smith et al., 2013). Solanum lycopersicum L. (Solanaceae) is one of the most important vegetable plants in the world and originated in America (Kimura & Sinha, 2008). Because of its economic value, tomato (S. lycopersicum) has been widely used as a research material (Kimura&, Sinha 2008). Solanum lycopersicum fruit is rich in protein, fiber, carbohydrates, various vitamins and numerous minerals (Tanveer et al., 2020).

This study aimed to examine the germination percentage, germination rate, biomass, root length, green pixel quantities and salt tolerance indices of *Solanum lycopersicum* "Marmande" seeds at different concentrations (%, mM and M) in saline solutions. Besides determining ecophysiological responses of tomato against salt stress, methods of deciding salt stress on model plant species are evaluated. This study provides an answer to which ecophysiological parameters the scientists should examine.

Material and Methods

Research was conducted on tomato seeds (cv. Marmande). For each application, 10 seeds were sowed in seedbeds, which include sterile petri dishes and double layer filter papers. Experiments were carried out in four replicates at concentrations of % 0.5, 1, 2, 0.25 M, 0.45 M, 50 mM, 100 mM, 150 mM NaCl. Distilled water was

used as control group. The prepared seedbeds were labeled and placed in the climate chamber ($22^{\circ}C \pm 1$, Sanyo, MLR 350). During the experiment, the seeds were observed on daily basis and when a radicle touched the seedbed, the seed was accepted as germinated. The total period of experiments was 21 days.

The germination percentage is an estimate of the viability the seed will likely develop under favorable conditions, while the germination rate is a measure of the seed germination time (Saupe, 2009). So, at the end of the experiments, germination percentage (GP) and germination rate (GR) were calculated by the following formulae (Raun *et al.*, 2002):

$$GP = \frac{\text{Number of total germinated seeds}}{\text{Total number of seeds tested}} X 100$$

GR = (Number of germinated seeds/ Day of the first count) + (/) + (Number of germinated seeds/Day of final count) (Vibhuti*et al.*, 2015).

Root length of each seed was measured by caliper as cm. For biomass calculation, seedlings were weighed as fresh, then dried at 70°C for 48 hours. Dried seedlings were measured and biomass was calculated as follows (Isik & Caliseki, 2017):

$$B=W_f-W_w/S$$

The photographs were taken and green pixels were counted using Adobe Photoshop CS6 in accordance with Kovacs *et al.*, (2015). STI value was calculated according to Turner & Marshall (1972). The data obtained from experiments were tested statistically with the SPSS Statistics 20 package program, One-Way ANOVA Test, with a sensitivity of p = 0.05.

Results

The highest germination percentage and rate (97.5%; 79.59) were observed in 0.5% NaCl application and the lowest (2.5%; 5.88) were in 0.45 mM NaCl (Table 1) (F=69.707; df=8.27; p<0.05). As the salt concentration increased, germination percentage decreased. Biomass was observed to be higher in the 50 M NaCl application than in the control group (F=24.796; df=8.27; p<0.05).

 Table 1. Germination percentage, rate, biomass, seedling growth, photosynthesis and salt tolerance index (STI) data of Solanum lycopersicum under salt stress conditions.

Treatment	% Germination	Germination rate	Biomass (h/kg)	Root length (cm)	Mean green pixel no	STI
Control	95	58.46	196.51	8.72	199.52	1.000
0.5 % NaCl	97.5	79.59	384.67	5.88	201.88	0.710
1 % NaCl	30	25.53	103.02	0.69	201.69	0.064
2 % NaCl	10	15.38	0.001	0.01	0.00	0.001
50 mM NaCl	90	32.73	460.67	11.96	199.69	1.142
100 mM NaCl	87.5	26.92	357.53	5.88	202.10	0.594
150mM NaCl	72.5	19.33	221.12	1.41	201.06	0.181
0.25 M NaCl	10	10.81	0.001	0.01	0.00	0.001
0.45 M NaCl	2.5	5.88	0.001	0.01	0.00	0.001

Like biomass, root length (F=45,755; df=8, 27; p<0.05) and STI have reached the highest value (Fig. 1) at 50 mM NaCl application (respectively 460, 67 kg/ha, 11,96 cm, 1,142). The biomass, root length and STI value of 2%, 0.25 M and 0.45 M NaCl applications were very low (respectively 0,001 kg/ha, 0,01, 0,001). For green pixel count, there was no seedling development for 2%, 0,25 M and 0,45 M NaCl applications so the value is equal to 0, but other NaCl applications resulted in homogenic groups (F=23130,628; df=8, 27; p<0.05).

According to One-Way ANOVA test, sigmoid value of all experimental groups was lower than 0.05 which means they were not homogenic groups statistically.

Discussion

Solanum lycopersicum fruit is one of the most important agricultural crops after *S. tuberosum*, and number one in productivity among other products (Meza *et al.*, 2020). Therefore, effects of increasing salinity problem of soil on important agricultural crops must be uncovered by scientific researches.

Cyanobacteria need low levels of NaCl for better photosynthesis, especially Na-ions are important in cyclic

electron transport (Sudhir & Murthy, 2004). Even though tomato is a higher plant, it demanded lower NaCl values to overcome photosynthesis reactions, like Cyanobacteria do. Smith et al., (2013) found that broccoli growth and appearance (color and vigor) were similar for salinity and boron treatments. Broccoli plants had a slightly darker green-blue color in saline treatments compared to nonsaline treatments (Smith et al., 2013). In this study, color/darkness difference between experimental groups was measured by counting green pixels, so this parameter was converted into numeric data. Because there was no seedling development (no green organ), for 2%, 0.25 M and 0.45 M NaCl applications, the green pixel number observed was equal to zero. Parida & Das (2005) presented that salinity tolerance ability of plants is determined by multiple biochemical pathways such as protective function of the chloroplast. According to the findings of Parida & Das (2005), green pixel data showed that all applications provide similar pixel numbers, excluding high level salinity applications. The increase in salinity level has an inverse relationship with leaf area for Jatropha curcas (Campos et al., 2012). Similarly, green pixel numbers were dramatically decreased when salinity levels were increased up to 2% and M degrees (Campos et al., 2012).

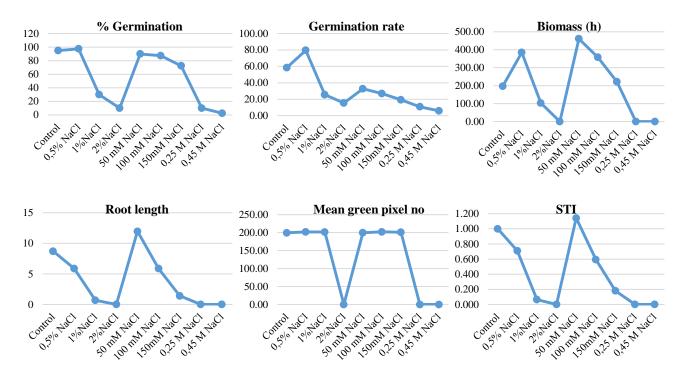


Fig. 1. Graphical demonstration of germination percentage, rate, biomass, seedling growth, photosynthesis and salt tolerance index (STI) data of *Solanum lycopersicum* under different NaCl stress conditions.

Dogan *et al.*, (2008) found that in tomato varieties, the maximum salt concentration in tolerant genotypes was 125-150 mM NaCl, while in sensitive genotypes 50-75 mM NaCl. Dogan *et al.*, (2008) found out the germination percentages of different cultivars against salinity stress, but in this study, it was observed that germination percentage or rate is not enough to determine the salt stress level in tomato seedlings, because biomass, root length, STI and photosynthesis values should be considered to evaluate plant stress.

Luo *et al.*, (2015) observed the inhibitory effect of high salinity on the biomass with cell plasmolysis and loss of metabolic activity. In this research it was observed that 2% and M applications of NaCl caused nearly zero h/kg biomass for seedlings. Souza *et al.*, (2012) found out that fresh and dry biomass of halophyte *Atriplex nummularia* were sensitive to soil moisture. Salinity tolerance of tomato is not as high as *Atriplex*, but 50-150 mM NaCl applications showed higher biomass level than control application in the present research.

Wang *et al.*, (2017) suggested that SINAC11 gene might play significant roles in the abiotic stress response for tomato. Salt sensitivity is the most apparent during germination and seedling period in tomato (Kaveh *et al.*, 2011; Wang *et al.*, 2017). Wang *et al.*, (2017) compared differences of NaCl stress on seed germination between transgenic and wild type tomato, and experimental data showed that germination percentage of transgenic lines was lower than wild type. Cultivation of wild tomato types or determining salinity tolerance of known tomato varieties is very important for further studies and agricultural treatments. With this study, ecophysiological responses of cv. Marmande is presented under three different (%, mM and M) salinity levels and it has been revealed that molar NaCl applications (0.25-0.45) were

resulted in the lowest values. However, NaCl (mM) applications showed slightly higher results than NaCl (%) treatments for Marmande cultivar of tomato.

Salinity decreases fresh weight and dry weight of seedlings (Mansour & Ali, 2017; Kaur & Gupta, 2018). Kaur & Gupta (2018) reported that, for tomato, salinity affects different growth stages and seed germination. In this study, similarly to Kaur & Gupta's (2018) findings, increased salinity applications caused the drop of development steps and germination percentages.

Silva Oliveira *et al.*, (2019) highlighted that melon seeds should not be used without pretreatment in agricultural areas suffering from salinity because they have a low germination rate and decline in seedling growth like the findings of present study about tomato cv. Marmande.

Jiménez-Arias *et al.*, (2019) applied 100 and 150 mM NaCl in their study using Robin cultivar of tomato plant as an experimental material and they obtained results which are similar to those of this research, but they did not apply molarity and percentage levels of NaCl. As in this study, the ecophysiological responses of the living creature are sharper when molarity and percentage levels of NaCl applications are applied in addition to the millimolar application; therefore, while similar studies are carried out, it will be beneficial to obtain more accurate results with molarity and percentage applications.

Exposure to salinity in plants converts stored polysaccharides into monosaccharides, resulting in inhibition of germination, problematic or incomplete seedling formation and subsequent poor development of plant populations (Foti *et al.*, 2019). The population size of a taxon can have both economic and ecological importance in the life cycle of plant species, especially in agriculturally important plants such as tomatoes used in

this study. For this reason, it is essential to define the salinity tolerance of the monocultured plant species together with other ecophysiological characteristics.

Salinity tolerance is too complex to be easily influenced by improvement through selection as a trait in itself (Roy *et al.*, 2014). It is important to find tolerance response of economic plants in terms of ecophysiology for the future of agriculture. In this research, obtained data gave rise to the thought that germination rate, biomass, root length and STI parameters were the most valuable for ecophysiological studies of salt stress in tomato plant.

Conclusion

Solanum lycopersicum is a worldwide-consumed plant species and salinity is an environmental problem that causes dramatic reduction of agriculture for any economical plant excluding extreme halophytes. Finding out ecophysiological responses of nutritionally significant plants has become globally important. With this study, responses of Marmande variety of tomato to different salinity treatments were revealed and tolerance intervals were evaluated.

References

- Barra, P.J., N.G. Inostroza, J.J. Acuña, L. Mora, D.E. Crowley and M.A. Jorquera. 2016. Formulation of bacterial consortia from avocado (*Persea americana* Mill.) and their effect on growth, biomass and superoxide dismutase activity of wheat seedlings under salt stress. *App. Soil Ecol.*, 102: 80-91.
- Benazzouk, S., P.I. Dobrev, Z.-E. Djazouli, V. Motyka and S. Lutts. 2020. Positive impact of vermicompost leachate on salt stress resistance in tomato (*Solanum lycopersicum* L.) at the seedling stage: A phytohormonal approach. *Plant Soil*, 446: 145-162.
- Campos, M.L.O., B.S. Hsie, J.A.A. Granja, R.M. Correia, J.S. Almeida-Cortez and M.F. Pompelli. 2012. Photosynthesis and antioxidant activity in *Jatropha curcas* L. under salt stress. *Braz. J. Plant Physiol.*, 24(1): 55-67.
- Dogan, M., A. Avu, E.N. Can and A. Aktan. 2008. Farkli Domates Tohumlarinin Cimlenmesi Uzerine Tuz Stresinin Etkisi. *SDU Fen-Ede. Fak, Fen Dergisi (E-Dergi)*, 3(2): 174-182.
- Feng, G, F.S. Zhang, X.L. Li, C.Y. Tian, C. Tang and Z. Rengel. 2002. Improved tolerance of maize plants to salt stress by arbuscular mycorrhiza is related to higher accumulation of soluble sugars in roots. *Mycorrhiza*, 12: 185-190.
- Foti C., E.M. Khah and O.I. Pavli. 2019. Germination profiling of lentil genotypes subjected to salinity stress. *Plant Biol.*, 21: 480-486.
- Grieve, C.M., S.R. Grattan and E.V. Maas. 2012. Plant salt tolerance. In: (Eds.): Wallender, W.W. and K.K. Tanji. Agricultural Salinity Assessment and Management (2nd edition).
- Isik, G. and M. Caliseki. 2017. Ecophysiological Effects of Porsuk River's Water and Boron Mine Wastewater on *Cucumis* sativus L. Seeds. Acta Physiol. Pol. A, 132: 746-748.
- Jamil, A., S. Riaz, M. Ashraf, M.R. Foolad. 2011. Gene expression profiling of plants under salt stress. *Crit. Rev. Plant Sci.*, 30: 435-458.
- Jiménez-Arias, D., J.F. García-Machado, S. Morales-Sierra, E. Suárez, J.A. Pérez, J.C. Luis, C. Garrido-Orduña, A.J.

Herrera, F. Valdés, L.M. Sandalio and A.A. Borges. 2019. Menadione sodium bisulphite (MSB): Beyond seed-soaking. Root T pretreatment with MSB primes salt stress tolerance in tomato plants. *Environ. & Exp. Bot.*, 157: 161-170.

- Kaur, H. and N. Gupta. 2018. Ameliorative Effect of proline and ascorbic acid on seed germination and vigor parameters of tomato (*Solanum lycopersicum* L.) under salt stress. *Int. J. Curr. Microbiol. App. Sci.*, 7(1): 3523-3532.
- Kaveh, H., H. Nemati, M. Farsi and S.V. Jartoodeh. 2011. How salinity affect germination and emergence of tomato lines. *J. Biol. Environ. Sci.*, 5(15): 159-163.
- Kimura, S. and N. Sinha. 2008. Tomato (Solanum lycopersicum): A Model Fruit-Bearing Crop. Cold Spring Harbor Protocols, 3(11): 1-9.
- Kovács, A., M. Tóth, F. Somogyi, L. Tolner, I. Czinkota, A. Béres, T. Wilk and L. Aleksza. 2015. The effect of biodiesel by-products on germination and plant growth. *App. Ecol. & Environ. Res.*, 13(4): 1171-1181.
- Luo, W., F.I. Hai, J. Kang, W.E. Price, W. Guo and H.H. Ngo. 2015. Effects of salinity build-up on biomass characteristics and trace organic chemical removal: Implications on the development of high retention membrane bioreactors. *Biores. Tech.*, 177: 274-281.
- Mansour, M.M.F. and F.E. Ali. 2017. Evaluation of proline functions in saline conditions. *Phytochem.*, 140: 52-68.
- Munns, R. 2005. Genes and salt tolerance together: bringing them together. *New Phytol.*, 167: 645-663.
- Munns, R. and M. Tester. 2008. Mechanisms of salinity tolerance. *Ann. Rev. Plant Biol.* 59:651-681.
- Ostrowski, M., A. Ciarkowska and A. Jakubowska. 2016. The auxin conjugate indole-3-acetyl-aspartate affects responses to cadmium and salt stress in *Pisum sativum* L. *J. Plant Physiol.*, 191: 63-72.
- Parida, A.K. and A.B. Das. 2005. Salt tolerance and salinity effects on plants: a review. *Ecotox. & Environ. Safety*, 60: 324-349.
- Raun, S., Xue, Q. and K. Thlkowska. 2002. Effect of seed priming on germination and health of rice (Oryza sativa L) seeds. Seed Sci. & Tech., 30: 451-458.
- Roy, S.J., S. Negrao and M. Tester. 2014. Salt resistant crop plants. *Curr. Opin. Biotech.*, 26: 115-124.
- Saupe, S.G. 2009. Plant Physiology-Germination Rates & Percentages. College of St. Benedict/St. John's University Biology Department PENGL 335 Collegeville, MN 56321.
- Silva-Oliveira C.E., F. Steiner, A.M. Zuffo, T. Zoz, C.Z. Alves and V.C.B. Aguiar. 2019. Seed priming improves the germination and growth rate of melon seedlings under saline stress. *Ciência Rural.*, 49: 7: 1-11.
- Smith, T.E., Grattan, S.R., Grieve, C.M., Poss, J.A., Läuchli, A.E. and D.L. Suarez. 2013. pH dependent salinity-boron interactions impact yield, biomass, evapotranspiration and boron uptake in broccoli (*Brassica oleracea* L.). *Plant Soil*, 370: 541-554.
- Souza, E.R., Freire, M.B.G.S., Cunha, K.P.V., Nascimento, C.A., Ruiz, H.A. and C.M.T. Lins. 2012. Biomass, anatomical changes and osmotic potential in *Atriplex nummularia* Lindl. cultivated in sodic saline soil under water stress. *Environ. & Exp. Bot.*, 82: 20-27.
- Sudhir, P. and S.D.S. Murthy. 2004. Effects of salt stress on basic processes of photosynthesis. *Photosynthetica*, 42: 481-486.
- Taiz, L. and E. Zeiger. 2002. Plant Physiology. 3rd edition, Sinauer Associates, USA, 878 p.
- Talaat, N.B. and B.T. Shawky. 2012. 24-Epibrassinolide ameliorates the saline stress and improves the productivity of wheat (*Triticum aestivum* L.). *Environ. & Exp. Bot.*, 82: 80-88.

- Tanveer, K., S. Gilani, Z. Hussain, R. Ishaq, M. Adeel and N. Ilyas. 2020. Effect of salt stress on tomato plant and the role of calcium. *J. Plant Nutr.*, 43(1): 28-35.
- Turner, R.G. and C. Marshall. 1972. The accumulation of zinc by subcellular fractions of roots of *Agrostis tenuis* in relation to zinc tolerance. *New Phytol.*, 71(4): 671-676.
- Vibhuti, C. Shahi, K. Bargali and S.S. Bargali. 2015. Seed germination and seedling growth parameters of rice (*Oryza* sativa) varieties as affected by salt and water stress. *Ind. J.* Agri. Sci., 85(1): 102-108.
- Wang, L., Z. Hu, M. Zhu, Z. Zhu, J. Hu, G. Qanmber and G. Chen. 2017. The abiotic stress-responsive NAC

transcription factor SINAC11 is involved in drought and salt response in tomato (*Solanum lycopersicum* L.). *Plant Cell Tiss. Org. Cult.*, 129: 161-174.

- Zhong, M., R. Song, Y. Wang, S. Shu, J. Sun and S. Guo. 2020. T Gase regulates salt stress tolerance through enhancing bound polyamines-T mediated antioxidant enzymes activity in tomato. *Environ. & Exp. Bot.*, 179: 104191.
- Zhu, J.K. 2001. Plant salt tolerance. Tree In Plant Sci., 6: 2: 66-71.
- Zhuo, Y., Y. Zhang, G. Xie and S. Xiong. 2015. Effects of salt stress on biomass and ash composition of switchgrass (*Panicum virgatum*). Acta Agri. Sca., B-Soil & Plant Sci., 65(4): 300-309.

(Received for publication 3 February 2021)