

THE INFLUENCE OF BENEFICIAL BACTERIA ON SOIL AND TOMATO ROOTS

Claudia Loredana DRAGOMIR¹, Gheorghita HOZA¹, Alexandra BECHERESCU³,
Liliana BĂDULESCU^{1,2}, Aurora DOBRIN², Mihai FRÎNCU², Dorel HOZA¹

¹University of Agronomic Sciences and Veterinary Medicine of Bucharest, Faculty of Horticulture,
59 Mărăști Blvd, 011464, District 1, Bucharest, Romania

²University of Agronomic Sciences and Veterinary Medicine of Bucharest,
Research Center for Studies of Food Quality and Agricultural Products,
59 Mărăști Blvd, 011464, Bucharest, Romania

³University of Life Sciences "King Mihai I" from Timișoara,
119 Calea Aradului, 300645, Timișoara, Romania

Corresponding author email: hozagh@yahoo.com

Abstract

The soil is one of the most important resources of mankind, being the physical support as well as providing the supply of nutrients that plants need to grow and bear fruit. It is well known from specialized research that the application of products based on microorganisms can improve the physical and chemical qualities of the soil and also the productivity of culture. The present study examined the efficacy of two products Rizobac and Bactilis on 3 tomato hybrids Kingset (red fruit), Bucanero (black fruit), Buffalosun (yellow fruit) grown in protected spaces (plastic tunnels), in the Scărișoara district of county Olt. The experiment took part in 2 years, 2022 and 2023. The culture was established by seedling (seedling maturity = 60-65 days) and after planting in the plastic tunnels, the technology specific to tomatoes was applied. The results showed that there were considerable differences regarding length (cm) and root volume (cm³) from 16% in terms of root length to 98% higher volume for plants treated with products used. Furthermore, the application of Rizobac and Bactilis improved soil apparent density, total porosity, and compaction degree. In summary, based on statistical correlation tests, it can be concluded that as the total nitrogen content decreases, the number of bacterial colonies increases proportionally.

Key words: biological, fertilizers, microorganisms, PGPR, soil.

INTRODUCTION

Tomato (*Lycopersicon esculentum* L.) is a highly nutritious and widely consumed product globally, known for its rich flavor and versatility in culinary applications, are consumed fresh in salads, cooked in various vegetable dishes, and processed into a wide array of products including ketchup, sauces, purees, syrups, juices, and more. The fruits are abundant sources of vitamins, minerals, amino acids, and pigments while being low in calories (Dinu et al., 2017; Soare et al., 2015).

These products are enjoyed on a broad scale worldwide, showcasing the versatility and popularity of tomatoes in culinary applications (Javaria et al., 2012).

Due to its high nutrient demands, tomato plants typically require substantial amounts of chemical fertilizers to meet their nutritional

needs. Despite this, tomatoes rank third in terms of global vegetable production (Helal et al., 2022; Shi & Maguer, 2000).

Tomatoes are cultivated in a wide range of soil types, from small-scale home gardens to large commercial farms, where they are grown as a profitable cash crop by vegetable growers (Pandey & Chandra, 2013). As a heavy-yielding crop, tomatoes demand substantial nutrient inputs to support their growth and maximize yield potential. Tomatoes exhibit positive responses to a variety of technological interventions, including both physical methods such as increasing the number of stems to enhance yield, as well as the application of biofertilizers containing substances like arginine and cysteine (Apahidean et al., 2019; Becherescu et al., 2021; Hoza et al., 2019).

In recent years, the concentrations of heavy metals in soils have risen significantly to

hazardous levels, primarily due to unsustainable practices such as sewage irrigation, excessive use of chemical fertilizers, and widespread pesticide application in agriculture. These practices pose serious threats to soil biological systems and overall ecosystem health (Jaishankar et al., 2014).

The demand for organic tomatoes is surging worldwide, driven by their superior nutritional quality, extended shelf-life, and sustainable yield. Similarly, there is growing momentum in restoring metal-contaminated soils and preserving soil health through the application of organic inputs and microbial inoculations. In recent years, farmers are increasingly adopting good agricultural practices, transitioning to organic farming, and optimizing water and nutrient usage (Verma et al., 2023). To achieve higher-quality production, formulations containing bacteria with biopesticidal, biostimulant, or biofertilizer properties are being utilized. These bacteria not only enhance plant productivity but also reduce plant residues, contributing to consumer safety. Additionally, these bacteria are known for their ability to produce lytic enzymes that inhibit plant pathogens and promote plant growth (Basu et al., 2021; Chojnacka, 2015).

Soil salinization has emerged as a significant challenge, severely impacting agricultural productivity and posing a threat to agricultural development worldwide (Litalien & Zeeb, 2020; Singh, 2021). To combat this issue, harnessing the potential of plant growth-promoting rhizobacteria is paramount. By leveraging these beneficial microorganisms, it can effectively restore degraded soils and sustain soil health, thus fostering sustainable agricultural practices worldwide (Kumar et al., 2023).

Utilizing organic sources of manures and fertilizers, along with proper management practices, can significantly reduce the excessive reliance on chemical fertilizers. This approach enables smallholder farmers to save costs for subsequent cropping seasons. Furthermore, the market demand for inorganic fertilizers surpasses that of organic ones due to their availability and concentrated nutrient content. In chemically fertilized soils, essential nutrients are quickly utilized and often leached out by various meteorological factors. In contrast, organic fertilizers and manures degrade slowly, allowing

for the retention of nutrients over an extended period. This results in a continuous supply of soil nutrients, promoting sustainable soil health and agricultural productivity (Praharaj, 2007). The metabolism of bacterial communities has shown significant benefits by enhancing existing soil microorganisms and improving nutrient assimilation. These positive outcomes can have notable implications for tomato growth, potentially resulting in a remarkable increase in tomato yield by up to 36.82%. These practices hold promise for improving agricultural productivity and sustainability (Dragomir & Hoza, 2022.; Wang et al., 2024).

MATERIALS AND METHODS

The experiment occurred in Scărișoara, Olt county, spanning 2022 and 2023. It followed a randomized block design, with three replicates for each experimental variant and six plants per replicate, covering an area of 200 m². Two variable factors were examined: soil characteristics and effects on tomato hybrids.

The primary objective of the experiment was to assess soil characteristics and the performance of hybrids when cultivated alongside radicular biostimulating products: Bactilis 5 L/ha and Rizobac 10 L/ha. The control group did not receive any biostimulant application.

Bactilis is a microbial inoculant which enhance root development, overall plant growth, and vigor. Additionally, these metabolites enhance the resilience of the root system against stressful conditions induced by a variety of biotic and environmental factors (Retrieved from <https://www.humofert.gr/en/product/2015-05-29-12-29-20/bactilis-detail.html>).

Rizobac is a microbial inoculant abundant in nutrients and beneficial microorganisms, designed to enhance rooting and facilitate the rapid establishment of transplanted crops in soil (Retrieved from <https://www.humofert.gr/en/product/2015-05-29-12-29-20/biostimulants-1/rooting/rizobac-1-detail.html>).

The biological material utilized consisted of three F1 hybrids: Bucanero, Buffalosun, and Kingset.

By combining the 2 factors, 9 variants resulted, each variant having 3 repetitions and 6 plants per repetition, as follows:

V1 - Kingset F1 Unfertilized; V2 - Bucanero F1 Unfertilized; V3 - Buffalosun F1 Unfertilized; V4 - Bucanero F1 + Rizobac; V5 - Buffalosun F1 + Rizobac; V6 - Kingset F1 + Rizobac; V7 - Buffalosun F1 + Bactilis; V8 - Kingset F1 + Bactilis; V9 - Bucanero F1 + Bactilis.

In order to produce seedlings, the seeds were sown in the last decade of January, followed by transplanting after 14-16 days from germination. Greenhouse planting occurred in the last decade of April, with spacing set at 0.8 m by 0.4 m, resulting in a density of 3.2 plants per square meter and 32,000 plants per hectare. The seedlings were approximately 60-65 days old at the time of planting. Throughout the vegetation period, specific care practices were implemented.

Soil assessments were conducted, samples were taken from 0-20 cm depth. Soil samples have been collected in plastic bags, then dried in the laboratory at room temperature for determination of Electrical Conductivity (EC) was analyzed according with FAO 2008 potentiometric method for pH in aqueous suspension at soil /water ratio of 1/5, pH level analyzed according with FAO 2008 potentiometric method for pH in aqueous suspension at soil/water ratio of 1/2.5., apparent density, total porosity, compaction degree analysed, according to the ICPA methodology, 1987 volume I, which is our national standard. Total nitrogen (N) was determined by using the modified Kjeldahl method as well as the count of bacterial colonies before and after inoculation using the method of dilutions up to 10^{-6} and after sowing by the "in the lawn" method (Benson 1990). Additionally, root measurements included weight (in grams) with PS R2 balances, length (in centimeters) with roulette, and the volume of the root system of the plants, using a 1 L Class A graduated cylinder (in cubic centimeters).

The determinations were carried out in the laboratories of the Research Center for Studies of Food Quality and Agricultural Products at the University of Agronomic Sciences and Veterinary Medicine of Bucharest.

Results were interpreted statistical F-test ($p \leq 0.05$) and T-test ($p \leq 0.05$).

Data values were collected from six replicates and subjected to an F test to determine whether variances were equal ($P > 0.05$) or unequal ($P <$

0.05). Statistical significance of differences was established using the T test, with significance levels interpreted as follows: $p > 0.05$: not significant, $p < 0.05$: weakly significant, $p < 0.01$: moderately significant, $p < 0.005$: highly significant, $p < 0.001$: very strongly significant, $p < 0.0001$: extremely significant and Correlation Coefficients.

RESULTS AND DISCUSSIONS

The application of biostimulating products on the three analyzed hybrids notably improved the EC (Table 1).

Table 1. The impact of biostimulating products on pH and EC

	Before treatment application		Control		Rizobac		Bactilis	
	EC ($\mu\text{S/cm}$)	pH	EC ($\mu\text{S/cm}$)	pH	EC ($\mu\text{S/cm}$)	pH	EC ($\mu\text{S/cm}$)	pH
Kingset F1	4.19	7.82	2.33	7.95	2.20	7.93	2.22	7.97
					$p=0.465$	$p=0.722$	$p=0.614$	$p=0.616$
Bucanero F1	4.19	7.82	3.38	7.91	1.98	7.91	2.57	8.04
					$p=0.023$	$p=1$	$p=0.142$	$p=0.053$
Buffalosun F1	4.19	7.82	2.18	7.98	1.99	7.83	2.05	8.08
					$p=0.001$	$p=0.081$	$p=0.643$	$p=0.064$

$p > 0.05$: not significant, $p < 0.05$: weakly significant, $p < 0.01$: moderately significant, $p < 0.005$: highly significant, $p < 0.001$: very strongly significant, $p < 0.0001$: extremely significant.

Before the application of treatments, the following values were recorded for EC 4.19 ($\mu\text{S/cm}$) and pH 7.82. After applying the Rizobac treatment, the EC values decreased significant for Bucanero F1 ($p=0.023$) and Buffalosun F1 ($p=0.001$) with values of 1.98 dS m^{-1} and 1.99 ($\mu\text{S/cm}$) would be optimal to create a more conducive environment for microbial activity during tomato cultivation (Maltas et al., 2022). EC remained stable for Kingset F1 compared to the control group.

pH levels generally remained stable or increased slightly for all hybrids.

Regarding Bactilis treatment, the EC values range from 2.05 to 2.57 ($\mu\text{S/cm}$), very similar with control value. The pH values range from 7.97 to 8.08.

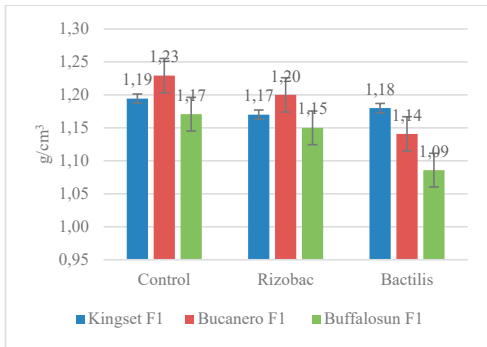


Figure 1. The influence of treatments on the apparent density of soil, g/cm³

For the Kingset F1 hybrid, it can be observed that the application of both Rizobac and Bactilis treatments resulted in an improvement in apparent soil density compared to the control group (Figure 1.). The substantial decrease in apparent soil density with the Bactilis treatment for the Bucanero F1 hybrid suggests that this treatment had a particularly positive effect on soil structure.

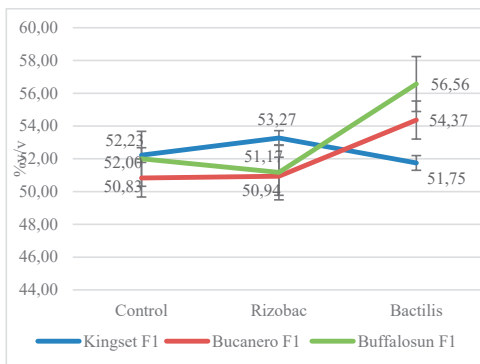


Figure 2. The influence of treatments on the total porosity, % v/v

The application of products had a significant impact on the porosity of the soil, particularly with the Rhizobac product, as evidenced by a significant increase ($p=0.029$) observed specifically in the Kingset hybrid (Figure 2.) Bucanero and Buffalosun hybrids exhibited notable positive responses to the application of the Bactilis product, showing values that were a few percentage points higher compared to control.

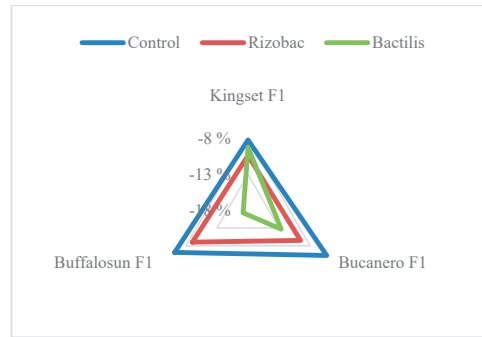


Figure 3. The influence of treatments on the compaction degree of soil, %

The application of the products led to a shift in soil settlement classes from Small, indicating slight loose soil with values between -9 and 1 (Mihalache et al., 2013), values between which the control falls -8.22 to -5.33 (Figure 3), to Very Small, indicating moderately loose soil with values between -17 and -10, observed for the application of Bactilis and Rizobac for the hybrids utilized.

To emphasize the relationship between the total nitrogen content and the number of bacterial colonies, it was conducted a "Correlation Coefficients" test (Figure 4) illustrates a robust, inversely proportional relationship.

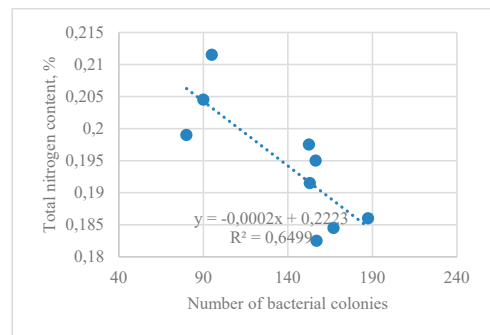


Figure 4. The correlation between the total nitrogen content and the number of bacterial colonies

In summary, based on the statistical correlation test, it conclude that the relationship between the total nitrogen content and the number of bacterial colonies (Figure 5) is as follows: as the total nitrogen content decreases, the number of bacterial colonies increases proportionally.

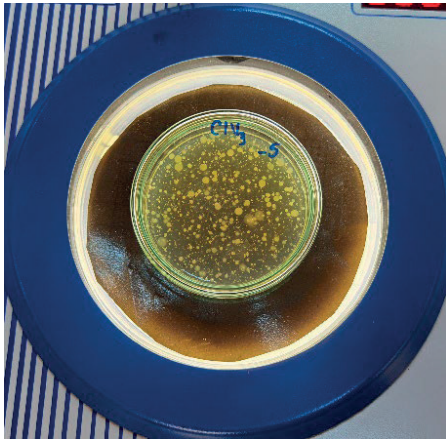


Figure 5. Bacterial colonies

By correlating roots weight with the number of bacterial colonies, a beneficial effect was observed. The correlation coefficients suggest a moderate to strong positive correlation between root weight and the number of bacterial colonies, indicating that as root weight increases, the number of bacterial colonies tends to increase as well (Figure 6).

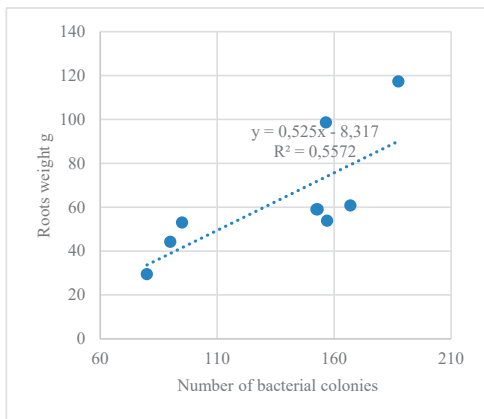


Figure 6. The correlation between roots weight and the number of bacterial colonies

Regarding root growth parameters, Rizobac fertilizer notably demonstrates positive and statistically significant effects on both analyzed parameters: root length and root volume. Specifically, the root length is significantly greater for the Bucanero F1 hybrid by approximately 5 cm compared to the non-fertilized version, with statistical significance ($p=0.001$). Additionally, it resulted in the

longest roots observed in the experiment, measuring 25.42 cm.

It can also be observed that all the hybrids react very well to the application of treatments, showing positive values, when Rizobac and Bactilis were applied (Figure 7).

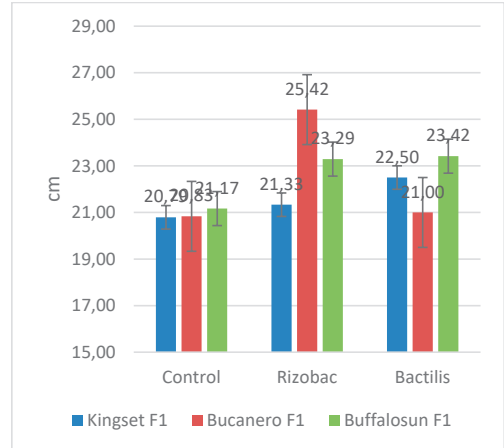


Figure 7. The impact of treatments root length, cm

The volume of the roots was significantly higher when applying the Rizobac biofertilizer compared to the non-fertilized version for all the hybrids from 35% to 98% (Table 2).

Table 2. The impact of biostimulating products on roots volume

	Roots volume, cm ³		
	Control	Rizobac	Bactilis
Kingset F1	32.33	52.83 $p=0.006$	37.50 $p=0.248$
Bucanero F1	51.25	101.30 $p=0.039$	76.60 $p=0.123$
Buffalosun F1	46.00	62.00 $p=0.027$	66.60 $p=0.036$

$p > 0.05$: not significant, $p < 0.05$: weakly significant, $p < 0.01$: moderately significant, $p < 0.005$: highly significant, $p < 0.001$: very strongly significant, $p < 0.0001$: extremely significant

Application of Bactilis lead also to a bigger volume for Buffalosun hybrid with 45%.

CONCLUSIONS

The present research highlights the significant influence of genotype on various traits, particularly evident in traits related to pH and EC. Fertilization with Rizobac yielded the best results for the Bucanero and Buffalosun hybrids

compared to the control, maintaining an ideal level of 2.0 ($\mu\text{S}/\text{cm}$) for tomato plants.

Furthermore, the application of Rizobac and Bactilis improved soil apparent density, total porosity, and compaction degree. In summary, based on statistical correlation tests, it can be concluded that as the total nitrogen content decreases, the number of bacterial colonies increases proportionally. Additionally, there is a moderate to strong positive correlation between root weight and the number of bacterial colonies, indicating that as root weight increases, the number of bacterial colonies tends to increase as well.

Regarding root growth parameters, Rizobac biofertilizer significantly and positively influenced all analyzed parameters, including root length and root volume. These findings demonstrate that the application of biofertilizers on tomato crops serve as a sustainable and organic technology, potentially reducing or even eliminating the need for conventional chemical applications.

REFERENCES

- Apahidean, A. I., Domocoş, D., Cărbunar, M., Bei, M., Hoza, G., & Apahidean, A. S. (2019). Cultivar and fertilization influence on production and quality of tomatoes grown in polyethylene tunnels in ecological system, *Scientific Papers. Series B, Horticulture*. Vol. LXIII, No. 1, 307-314.
- Basu, A., Prasad, P., Das, S., Kalam, S., Sayyed, R., Reddy, M., & El Enshasy, H. (2021). Plant Growth Promoting Rhizobacteria (PGPR) as Green Bioinoculants: Recent Developments, Constraints, and Prospects. *Sustainability*, 13, 1140. <https://doi.org/10.3390/su13031140>.
- Becherescu, A., Hoza, G., Dinu, M., Iordănescu, O., & Popa, D. (2021). The influence of biofertilizing and biostimulating products on the production of cornichon cucumber hybrids cultivated in heated solariums. *Scientific Papers. Series B, Horticulture*. Vol. LXV, No. 1, 375-381.
- Chojnacka, K. (2015). Innovative bio-products for agriculture. *Open Chemistry*, 13, 932-937. <https://doi.org/10.1515/chem-2015-0111>.
- Dragomir, C.L., & Hoza, D. (2022). Review on improving tomato culture technology in protected system for environmental protection and increasing productivity using PGPR. *Scientific Papers. Series B, Horticulture*. Vol. LXVI, No. 1, 444-448.
- Helal, D. S., El-khawas, H., & Elsayed, T. R. (2022). Molecular characterization of endophytic and ectophytic plant growth promoting bacteria isolated from tomato plants (*Solanum lycopersicum* L.) grown in different soil types. *Journal of Genetic Engineering and Biotechnology*, 20(1), 79. <https://doi.org/10.1186/s43141-022-00361-0>.
- Hoza, G., Jasim, M. M., Neata, G., Dinu, M., Becherescu, A., Apahidean, I. A., & Shalal, H. H. (2019). Effect of foliar spraying with Arginine and Cysteine and the number of stems on the growth and yield of cherry tomatoes grown in protected culture. *Scientific Papers. Series B, Horticulture*. Vol. LXIII, No. 1, 417-423.
- Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B. B., & Beeregowda, K. N. (2014). Toxicity, mechanism and health effects of some heavy metals. *Interdisciplinary Toxicology*, 7(2), 60-72.
- Javaria, S., Khan, M. Q., Rahman, H., & Bakhsh, I. (2012). Response of tomato yield and post harvest life to potash levels. *Sarhad J. Agric.*, 28.
- Kumar, C., Tomar, A., Pandey, S., & Prasad, M. N. V. (2023). Plant Growth Promoting Rhizobacteria Sustaining Saline and Metal Contaminated Soils. In *Agroecological Approaches for Sustainable Soil Management* (pp. 437-456). John Wiley & Sons, Ltd. <https://doi.org/10.1002/9781119911999.ch19>
- Litalien, A., & Zeeb, B. (2020). Curing the earth: A review of anthropogenic soil salinization and plant-based strategies for sustainable mitigation. *Science of The Total Environment*, 698, 134235. <https://doi.org/10.1016/j.scitotenv.2019.134235>.
- Maltas, A. S., Tavalı, I. E., Uz, I., & Kaplan, M. (2022). Monitoring the effects of pH and EC regulated drip fertigation on microbial dynamics of calcareous soil in tomato (*Solanum lycopersicum* L.) cultivation under greenhouse conditions in a Mediterranean climate. *Scientia Horticulturae*, 306, 111448. <https://doi.org/10.1016/j.scienta.2022.111448>.
- Mihalache, M., Ilie L., Marin, D., Dodociuiu, A. M. (2013). Clay minerals and potassium regime of some soils from Romania. *Current Opinion in Biotechnology*, 24(1), S141-S142.
- Dinu, M., Hoza, G., & Becherescu, A. (2017). Antioxidant capacity and mineral content of some tomatoes cultivars grown in Oltenia (Romania). *17th International Multidisciplinary Scientific GeoConference SGEM 2017, Vol. 2*, 149-156. <https://doi.org/10.5593/sgem2017B52>.
- Pandey, S., & Chandra, K. (2013). Impact of integrated nutrient management on tomato yield under farmers field conditions. *Journal of environmental biology/Academy of Environmental Biology, India*, 34, 1047-1051.
- Praharaj, C. (2007). Long term quantitative and qualitative changes in cotton and soil parameters under cultivars, cropping systems and nutrient management options. *Indian Journal of Agricultural Sciences*, 77, 280-285.
- Shi, J., & Maguer, M. L. (2000). Lycopene in Tomatoes: Chemical and Physical Properties Affected by Food Processing. *Critical Reviews in Food Science and Nutrition*, 40(1), 1-42. <https://doi.org/10.1080/10408690091189275>.
- Singh, A. (2021). Soil salinization management for sustainable development: A review. *Journal of Environmental Management*, 277, 111383. <https://doi.org/10.1016/j.jenvman.2020.111383>.

- Soare, R., Dinu, M., & Rosculete, E. (2015). *The influence of the hybrid onearly tomatoes production. XLV. Analele Universității din Craiova, Seria Agricultură, Montanologie, Cadastru (Annals of the University of Craiova - Agriculture, Montanology, Cadastre Series)*, Vol. XLV, 266-270
- Verma, S. B., Kumar, C., & Narayan, R. P. (2023). Sustaining soil health and tomato production through organic inputs and bacterial inoculations in alluvial soils detoxifying plant contaminants. *South African Journal of Botany*, 161, 404–417. <https://doi.org/10.1016/j.sajb.2023.08.017>
- Wang, J., Cui, Y., Wu, K., Wu, S., Wu, K., Li, Y., & Niu, W. (2024). Micro/nanobubble-aerated drip irrigation affects saline soil microenvironments and tomato growth by altering bacterial communities. *Soil and Tillage Research*, 239, 106034. <https://doi.org/10.1016/j.still.2024.106034>