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INFLUENCE OF PSEUDOMONAS KOREENSIS AND PSEUDOMONAS CHLORORAPHIS STRAINS ISOLATED FROM THE RHIZOSPHERE OF HIPPOPHAE RHAMNOIDES ON THE GERMINATION AND GROWTH RATES OF TOMATO (SOLANUM LYCOPERSICUM L.) SEEDS

Mamirova Odina Abdulkhamidkhon kizi
Doctoral Candidate
E-mail: odinaxonmamirova135@gmail.com
phone: +998903094418

Artikova Rano Mamurkhanovna
b.f.n. Associate Professor
Tashkent State Agrarian University, UZ
ranoartikova898@gmail.com,
https://orcid.org/0009-0009-0570-4510

Abstract

This article examines the effect of rhizabacteria of the Chakanda- (Hippophae rhamnoides.L) plant on tomato (Solanum lycopersicum L.) seeds under various environmental conditions. During the study, Ryzobacteria were isolated from the rhizosphere of Hippophae rhamnoides L. and identified using the MALDI-TOF MS method. Based on the identification results, strains of Pseudomonas koreensis and Pseudomonas chlororaphis were identified, and their influence on the germination and initial growth indicators of tomato seeds (Solanum lycopersicum) was studied. As a result of experiments conducted under laboratory conditions, it was determined that the germination energy of seeds treated with these strains increased by 20-30 percent compared to the control. The results obtained confirm the growth-stimulating (PGPR) properties of the strains P.koreensis and P. chlororaphis and indicate the possibility of their use as promising biostimulants in agriculture.

Keyword: Hippophae rhamnoides, Pseudomonas, MALDI-TOF MS, tomato, unuvc hanlik, PGPR strains, in vitro.

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Introduction

Global climate change is intensifying the impact of abiotic stresses on plants. Vegetables are rich in vitamins, minerals, dietary fiber, and various phytochemicals; therefore, they are of great importance for human health. The growth of vegetable crops is regulated by various abiotic stress factors, which not only affect their normal growth and metabolism but also lead to a decrease in yield and quality [1.5]. Plant growth-stimulating rhicobacteria (PGPR) can alter the morphological or physiological properties of plants by inducing nitrogen fixation, phosphorus solubility, potassium solubility, siderophore production, secretion of secondary metabolites and hormones, and expression of the plant stress resistance gene. As a result, this increases the level of nutrient utilization in plants, increasing their yield, quality, and stress resistance. Today, one of the most pressing problems in agriculture is reducing various chemical substances in the soil and growing environmentally friendly products. In recent years, the use of plant growth-stimulating rhizosphere bacteria (PGPR) has been recognized as an effective strategy to replace chemical fertilizers [1,2]. Representatives of PGPR, specifically the genus Pseudomonas, are distinguished by their ability to synthesize phytohormones, mobilize phosphates, and increase resistance to various abiotic stresses in plants.

Strains belonging to the genus Pseudomonas, including P. koreensis and P. chlororaphis, exhibit high biostimulatory properties in the early stages of crop development, particularly in tomatoes (Solanum lycopersicum), due to the production of their secondary metabolites and siderophores [3]. Studies show that beneficial rhizabacteria such as Pseudomonas koreensis have the ability to increase plant biomass, improve its physiological state, and activate the defense system [4]. This determines the need for the widespread use of these microbial communities in the development of environmentally friendly and sustainable agricultural technologies. The rhizosphere microflora possesses unique characteristics depending on the plant species and its growing conditions. In this regard, the rhizosphere of sea buckthorn (Hippophae rhamnoides L.), which is considered a medicinal and resistant plant, is a rich microbiological resource, and strains isolated from it have high potential for increasing the yield of cultivated crops. Modern mass spectrometry methods, such as MALDI-TOF MS, have made it possible to accelerate microbial identification and accurately classify strains, increasing the reliability of PGPR studies [5]. However, there are still insufficient comprehensive studies on the influence of strains P. koreensis and P. chlororaphis isolated from the rosehip rhizosphere on tomato seed germination.

Research materials and methods. For scientific research, soil samples from the roots of the Hippophae rhamnoides.L plant were taken from the banks of the Zarafshan River in the Samarkand region and from the Sakson-ota forestry located in the Yukori-Chirchik district of the Tashkent region, and laboratory research was conducted at the Department of Biotechnology of the Tashkent State Agrarian University.

Preparation of soil samples from the rhizosphere of Hippophae rhamnoides L. Samples are taken sequentially, starting from the bottom layer. Immediately after excavation, the first sample is taken from the parent rock or the lower layer with a shovel. After describing the soil profile, samples are taken from the above layers. Finally, a sample is taken from the upper layer. It was collected from layers at depths of 0-10, 10-20, and 20-30 cm under sterilized conditions. (Litvinov, 1969).

Sample Taking Technique. In the middle of each layer, a gap of approximately 10 cm is marked between the layers, from which a soil sample is cut with a knife along the entire width of the front wall and placed on a sheet of thick paper. Samples are taken from humus-containing and plow-in layers along the entire thickness of the layer. If the thickness of the humus layer exceeds 20 cm, two to three samples are taken every 10 cm. On the paper label, the region, district, village, area, field and pit number, layer thickness, and depth from which the sample was taken are written in pencil, along with the date and student's surname, and the wrapped paper is wrapped around the sample. The layer from which the sample was taken, its depth, and the date are also written on the paper.

After determining the morphological characteristics of the soil and writing an explanation, a square sample is taken from each layer. Make sure the sampling location belongs to this layer. You can't take a sample from between two squares. If the sample is taken correctly, the result will be correct. Therefore, before sampling, the walls of the pit are thoroughly cleaned, and then the sampling locations are marked. One sample is taken from each soil layer, and two samples are taken when the soil is thick. The soil sample should be taken from the lowest layer. Otherwise, i.e., if taken from above, the bottom layers may be contaminated, and the sample may be damaged. The sample is taken at a distance of 0–10 and 10–20 cm from the plow layer.

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Separating microorganisms from soil samples and obtaining pure cultures. The process of diluting the soil taken as a sample is carried out to sow bacteria and isolate a pure culture. Soil dilution is carried out the very next day after the sample is taken, according to the method accepted in general microbiology and mycology (Litvinov, 1969).

To dilute the soil, 10 g of soil was dissolved in 90 ml of water in a sterilized flask for 5 minutes. Using a sterilized pipette, 1 ml of suspension was added to 9 ml of water in a sterilized test tube. This process was repeated again. The liquid from the third and fourth test tubes was sown on a platter nutrient medium (1:1000, 1:10000). For this purpose, 0.5 ml of the resulting suspension was spread evenly using a spatula over the surface of the agar nutrient medium placed in a Petri dish. This process is repeated three times. Identification is performed after obtaining pure cultures. Before identifying the rhizabacteria, I prepared a physical solution to obtain a clean colony. A 0.9% NaCl solution is used. The physical solution is poured into the test tubes, brought to a ready state, and autoclaved. Rhizobacteria for identification should not have been planted for more than 48 hours. When planting, take some bacteria from the petri dish with a hook and dip them into the fizrostvor. Shake well. The hook is sterilized and cooled, soaked in fizrostvor, and sown on peptone feed. Then the colonies will be separated.

The MALDI-TOF (Matrix-Assisted Laser Desorption/Ionization-Time of Flight) mass spectrometry method is one of the advanced technologies used for the rapid and accurate detection of microorganisms, including bacteria, fungi, and other pathogens. The Zybio EXS 2600 model was created to facilitate and accelerate the detection of microorganisms in clinical and laboratory settings.

Bacteria isolated from the rhizosphere of the Hippophae rhamnoides.L plant were grown on solid nutrient media (meat-pepton agar) and their colonial and cellular morphological characteristics were studied. Observations revealed that the majority of the isolated bacteria were Gram-positive.

During the experiment, 25 bacterial isolates were isolated from the rhizosphere of the Hippophae rhamnoides.L plant. These isolates were identified using the MALDI-TOF mass spectrometry method. Based on the results, it was established that the bacteria belong to 8 species (Fig. 1). The most common bacterial species is Bacillus cereus, which was recorded in a total of 13 samples. Species Bacillus thuringiensis, Pseudomonas chlororaphis, Pseudomonas koreensis, Micrococcus sp., Microbacterium foliorum, and Kocuria rosea were also identified.

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Identification scores ranged from 1.74 to 2.30, with most of them exceeding 2.00. This means that bacteria have been reliably identified at the species level. The results are shown in the table below:

Figure 1. Results of Rhyobacterium Identification.

Spot	Sample ID	Patient ID	Organism	Score
A8	T-7		*Bacillus thuringiensis*	1.75
A9	S17		*Bacillus cereus*	1.87
A10	T-6		*Micrococcus sp.*	1.74
A11	T-5		*Bacillus cereus*	2.18
A12	T-1		*Bacillus cereus*	1.75
B4	T-9		*Bacillus cereus*	2.15
B5	T-8		*Bacillus cereus*	2.30
B6	S-1		*Microbacterium foliorum*	2.17
B7	S4		*Kocuria rosea*	2.21
B8	S-6		*Bacillus cereus*	2.17
B9	S-9		*Bacillus cereus*	1.80
B11	S-12		*Microbacterium foliorum*	2.29
B12	S-15		*Bacillus cereus*	2.11
C1	S-16		*Bacillus cereus*	2.06

Spot	Sample ID	Patient ID	Organism	Score
A10	2		*Bacillus cereus*	2.11
A11	3		*Bacillus cereus*	1.84
A12	4		*Bacillus thuringiensis*	1.81
B1	5		*Kocuria rosea*	2.04
B2	6		*Pseudomonas chlororaphis*	2.13
B3	7		*Bacillus cereus*	2.26
B4	8		*Pseudomonas koreensis*	2.20
B5	9		*Bacillus cereus*	2.25
B6	10		*Bacillus thuringiensis*	2.18
B7	11		*Bacillus cereus*	2.09
B9	13		*Microbacterium foliorum*	2.21

Growth and biostimulatory properties. This methodology aims to determine the viability and germination energy of seeds in a sterile and controlled environment.
Necessary equipment and materials
1. Containers: Petri dishes (bottle or plastic).
2. Filter paper: Sterile moistened paper filters or a layer of cotton wool.
3. Sterilizers: 70% ethyl alcohol or 1% sodium hypochlorite (NaOCl).
4. Equipment: Thermostat (with light and temperature control function).

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5. Water: Distilled water.

Stages of the experiment.

A) Surface sterilization of seeds: Seeds are kept in 70% alcohol for 1 minute to remove microflora from the surface, then in a 1% sodium hypochlorite solution for 3-5 minutes. Then, the seeds are thoroughly washed 3-4 times in distilled water.

B) Sowing process: 2 layers of filter paper are placed at the bottom of Petri dishes and moistened with distilled water (or a bacterial suspension depending on the experimental variant). Depending on the size of the seeds, 25 or 50 seeds are placed in each cup so that they do not touch each other. The experiment is carried out at least 3-4 times.

C) Incubation conditions: The cups are placed in a thermostat. Temperature: Depends on the type of crop (for example, from +25°C to +28°C for tomatoes and cucumbers). Humidity: Constantly monitored (filter paper must not dry out).

Calculation of indicators. Results are determined using the following formulas:

Germination energy E: The percentage of seeds that germinated rapidly within a specific period (e.g., 3-4 days) after sowing.

$$\frac{n}{N} E = \times 100\%$$

(Here, n is the number of seeds that germinated within the specified period, and N is the total number of seeds).

Laboratory germination G: The total percentage of seeds that have fully germinated at the end of the experiment (e.g., 7-10 days).

$$\frac{n_{final}}{N} G = \times 100\%$$

Morphometric indicators of grasses:

The length of the root part of the grass (mm).

Length of the hypocotyledon part of the grass (mm).

Calculation of biostimulation. Experiment and control are compared. Calculated as a percentage or index

$$\frac{T-C}{C} \times 100 SI =$$

Laboratory germination of seeds was determined in Petri dishes in accordance with the requirements of GOST 12038-84 (GOST 12038-84.,1985). Before sowing, the seeds were sterilized in a 1% NaOCl solution. The experiment was conducted in a thermostat at a temperature of +25°C with 3 repetitions. Germination energy was calculated on the 3rd day, and laboratory germination on

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the 7th day. The obtained results were subjected to statistical analysis. During the research, experiments were conducted under laboratory conditions to determine the germination of tomato seeds. When selecting seeds, attention was paid to the absence of foreign impurities in the quality of the seeds. Attempts were made to use local varieties, and 3 local varieties of Solanum lycopersicum L. were used.

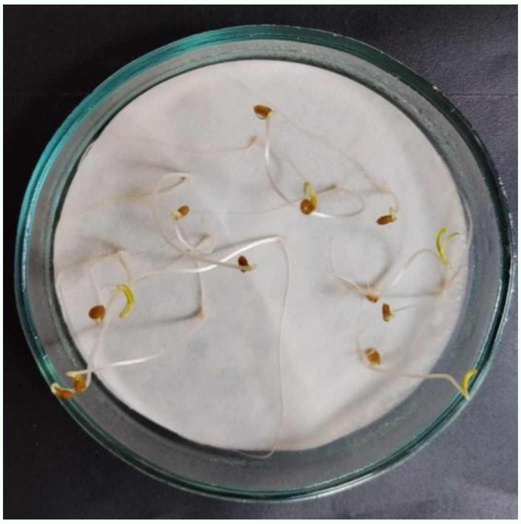
Table 1Varieties of Solanum lycopersicum L.selected for experiment

	Solanum lycopersicum L
Local	Volga-Girat
Local	Yusupsky
Local	TMK-22

Table 2. Required equipment

	Solanum lycopersicum L
Petri dish	32
Seed	320
Filter paper	62
Measuring pipette	1
flask	2
Distilled water	500ml
Liquid bacterial solution	10ml
Glass	4 items

A. Control variant; B.1/10; C. 1/100; D. 1/100 concentrations



Control 1/10

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1/100 1/1000

Figure 2. Tomato seed germination (with Pseudomonas chlororaphis)

Conclusion

As a result of the conducted research, it was proven that rhizobacteria isolated from the specific agro-ecological conditions of the Tashkent and Samarkand regions are important biological agents in ensuring the stable growth of Hippophae rhamnoides.L plants. Based on the research results, the following conclusions were drawn:

1.Microbiological diversity and adaptation: Bacillus cereus, Bacillus thuringiensis, Micrococcus sp., Microbacterium foliorum and Kocuria rosea strains isolated from the cycads rhizosphere are highly adapted to local soil and climatic conditions. This indicates that bacteria actively colonize the rhizosphere, which allows them to enter into long-term symbiotic or associative relationships with the plant.

2. Effectiveness of PGPR mechanisms: While the ability of isolates to synthesize indole-3-acetic acid (IAA) accelerates the development of the plant's root system, the ability to mobilize phosphates ensures the transition of poorly soluble compounds in the soil into a form that the plant can absorb. This, in turn, allows the plant's need for nutrients to be met naturally.

3. Ecological and practical significance: Biohumus and biological fertilizers created on the basis of identified PGPR strains serve to reduce the consumption of chemical fertilizers in agriculture, prevent soil degradation, and grow

environmentally friendly products. These bacteria are an important resource for improving the cultivation technology of sea buckthorn, especially in saline and low-fertility soils. Given the specific degree of salinity and drought in the soils of the Zarafshan River basin and the Tashkent region, the resistance of isolated PGPR bacteria to osmotic stress is an important indicator. By increasing the amount of proline in the plant and activating the system of antioxidant enzymes, these microorganisms help the chamomile accumulate high biomass even in unfavorable environmental conditions.

References:

1. Yang, J., Tang, M., & Zhao, H. (2025). Physiological Mechanisms of Plant Growth-Promoting Rhizabacteria in Enhancing Abiotic Stress Tolerance of Vegetable Crops: A Review. Plants, 14 (2), 239. https://doi.org/10.3390/plants14020239
2. Gupta, G., Parihar, S. S., Ahirwar, N. K., Snehi, S. K., & Singh, V. (2015). Plant Growth Promoting Rhizabacteria (PGPR): Current and Future Prospects for the Development of Sustainable Agriculture. Journal of Microbial & Biochemical Technology, 7 (2), 096-102. doi: 10.4172/1948-5948.1000188
3. Adedayo A. A., Fadiji A. E., Babalola O. O. Biochemical and Molecular Characterization of Bacterial and Fungal Isolates Associated with the Rhizosphere of Healthy and Diseased Solanum lycopersicum // International Journal of Agriculture & Biology. - 2023. Vol. 30. - No. 4. – P. 281-290.
4. Guo, Q., Sun, Y., Shi, M., Han, X., Jing, Y., Li, Y., Li, H., & Lai, H. (2021). Pseudomonas koreensis promotes tomato growth and shows potential to induce stress tolerance via auxin and polyphenol-related pathways. Plant and Soil, 462, 141–158. https://doi.org/10.1007/s11104-021-04837-9
5. Shah, A., Nazari, M., Antar, M., Msuda, L. A., Naamala, J., Liu, D., Rabileh, M., Zajonc, J., & Smith, D. L. (2021).
6. PGPR in Agriculture: A Sustainable Approach to Increasing Climate Change Resilience. Frontiers in Sustainable Food Systems, 5, 667546. </5><6>https://doi.org/10.3389/ებლაfs.2021.667546</6&gt
7. Abdullaev Z.T., Mirzaeva D.K. Microflora of saline soils and microorganisms adapted to it // Agrarian Science of Uzbekistan. - 2021. - No. 3. – P. 33-38.

8. Egamberdieva D.R. Plant growth promoting rhizabacteria under saline soil conditions: mechanisms and practical use // Microbiological Research. - 2020. Vol. 235. – P. 126447.
9. Ganie, S.A., Khan, M.A., and S.H. Role of rhizabacteria in alleviating salt stress in vegetable crops: recent advances // Frontiers in Plant Science. - 2021. Vol. 12. – Article 678673.
10. Glick B.R. Bacteria with ACC deaminase can promote plant growth and help feed the world // Microbiological Research. – 2014. Vol. 169, No. 1. – P. 30-10.
11. Spectrophotometric indicators of “Zamin-M” biopreparation immobilized with gypanflocculinate E3S Web of Conf. Volume 389, 2023 Ural Environmental Science Forum “Sustainable Development of Industrial ”https://doi.org/10.1051/e3sconf/202338903076. UESF-2023
12. Khojanazarova M, Artikova R, Khalmuminova G. etc. Proteomic and metabolomic analysis of the Eskimo-1 line on the effect of low positive temperatures E3S Web of Conf. Volume 389, 2023 Ural Environmental Science Forum “Sustainable Development of Industrial ”https://doi.org/10.1051/e3sconf/202338903076. UESF-2023
13. Artikova R., Abduraximova A. Isolation of Hippophae rhamnoides.L rhizosphere microorganisms. Web of Diskoveries:Journal of Analysis and inventions.Volume 2, Issue 7, July - 2024 ISSN(E): 2938-3773
14. Artikova R. Studying the properties of cultural liquid strains of bacteria of the genus azotobacter BIO Web of Conferences 141, 01009 (2024) . Agricultural science, 2024 https://doi.org/10.1051/ bioconf/202414101009