Frontiers in Health Informatics
ISSN-Online: 2676-7104
www.healthinformaticsjournal.com
2024; Vol 13: Issue 3
Open Access

Co-inoculation Effect of Multi-functional Rhizobacteria on Productivity of *S. tuberosum*

[1]Verinder Virk, Rishita Srivastava*
[1]Assistant Professor, Department of Microbiology, , Kanya Gurukula Campus, Gurukula Kangri, Haridwar - 249404, Uttarakhand, India.
Email: virender.wahla@gkv.ac.in
*PhD Scholar, Department of Microbiology, Kanya Gurukula Campus, Gurukula Kangri, Haridwar - 249404, Uttarakhand, India.
Corresponding author email: Srivastavarishita111@gmail.com

Cite this paper as: Verinder Virk, Rishita Srivastava (2024) Co-inoculation Effect of Multi-functional Rhizobacteria on Productivity of S. tuberosum. *Frontiers in Health Informatics, 13* (3), 11244-11255

ABSTRACT

The simultaneous introduction of nitrogen-fixing bacteria and plant growth-promoting rhizobacteria (PGPR) into crops has been shown to significantly enhance plant growth. *S. tuberosum* (commonly known as potato) is a key dietary source of high-quality protein for human consumption. PGPR are known to improve various aspects of plant development, including overall growth, nodule formation, and nitrogen fixation capacity. This study aimed to investigate whether the co-inoculation of potato plants (*Solanum tuberosum*) with *Pseudomonas putida* and *Bradyrhizobium japonicum* could enhance plant growth, productivity, and nutrient uptake. The findings demonstrated that the combined inoculation of *B. japonicum* and *P. putida* resulted in significantly greater growth and yield benefits compared to plants inoculated with *B. japonicum* alone or the uninoculated control group. Specifically, co-inoculation with *P. putida* and *B. japonicum* increased the nutrient content of potato plants: nitrogen (N) by 45%, phosphorus (P) by 35%, potassium (K) by 42%, magnesium (Mg) by 20%, sodium (Na) by 84%, and calcium (Ca) by 42% relative to the control. In contrast, inoculation with *B. japonicum* alone resulted in a smaller but still significant increase in nutrient content, raising N by 17%, P by 19%, and K by 17% compared to *P. putida* inoculation or the control. The highest values for N, P, and K were observed with co-inoculation treatments, which also enhanced soil nutrient levels. Under normal conditions, co-inoculation with *P. putida* and *B. japonicum* produced a marked improvement in both crop yield and soil nutrient composition compared to other treatments, underscoring the synergistic potential of these microbial inoculants.

Keywords: Inoculation; Biofertilizer; Plant Growth-Promoting Rhizobacteria; Micro-organisms.

INTRODUCTION

Potatoes (*S. tuberosum*) are consumed by over a billion individuals and rank among the most prominent non-cereal agricultural products. This crop is distinguished by its high nutritional value and superior yield-to-soil occupancy ratio compared to alternative crops. It is the fourth most cultivated and consumed crop worldwide, following maize, wheat, and rice (Aloo *et al*., 2019). Reports highlight that potatoes provide the greatest nutritional vitality and economic value per unit area of cultivation, thriving across a wide range of climatic conditions, elevations, and latitudes (Wu *et al*., 2013). Furthermore, potatoes are recognized for their ability to produce nutrient-dense food more efficiently, on less land, and under harsher conditions than most other crops. Approximately 85% of the potato is edible, in contrast to only 50% in cereals, and it contains the highest protein

content among tuber crops, with 2.1% of its fresh weight comprising protein (Fageria *et al*., 1997).

As of 2014, potatoes were cultivated in 82% of nations globally, with a total production of 382 million tons. Consequently, potatoes are indisputably vital for both commercial and nutritional purposes on a global scale. This importance is amplified by the increasing global population and diminishing availability of arable land, underscoring the crop's pivotal role in global food security systems (Aloo *et al*., 2019). However, potatoes are fertilizer-intensive, requiring up to 150 kg of phosphorus (P) and 250 kg of nitrogen per hectare (Hochmuth and Hanlon, 2000). To meet these demands, synthetic fertilizers and pesticides are frequently employed to enhance yield and combat pathogens, though these practices often come with significant environmental and economic costs (Mohammadi and Sohrabi, 2012).

Potato agriculture is further challenged by its vulnerability to various pests and diseases, which can lead to substantial financial losses and increased cultivation expenses (Hill and Lazarovits, 2005). Addressing these issues, the use of beneficial rhizosphere bacteria has emerged as a promising strategy in organic farming globally (Hungria *et al*., 2013; Naqqash *et al*., 2016). The physiological traits of plants, as well as nutrient availability, significantly influence their growth, development, and productivity (Makbul *et al*., 2011; Anjum *et al*., 2011). Plant growth-promoting rhizobacteria (PGPR) are now widely accepted for both biocontrol and growth enhancement (Sureshbabu *et al*., 2016). These microorganisms enhance nutrient uptake, improve nutrient availability, and stimulate plant development, qualifying them as effective biofertilizers.

Biofertilizers are increasingly recognized as viable alternatives or supplements to chemical fertilizers for boosting crop yield in low-input agricultural systems (Atieno *et al*., 2020). Certain PGPR strains exhibit the ability to fix nitrogen, solubilize mineral nutrients, and mineralize organic molecules. Extensive studies on biofertilizers have predominantly focused on nitrogen fixation and the efficient utilization of insoluble phosphorus compounds (Martínez-Viveros *et al*., 2010). PGPR also play a critical role as rhizosphere microorganisms, fostering plant proliferation when growing alongside host plants. Their efficacy in colonizing soil ecosystems is attributed to their adaptability to diverse environmental conditions, rapid growth rates, and the metabolic versatility to process a wide array of natural and xenobiotic compounds (Kloepper *et al*., 1980).

However, the inability of PGPR to successfully colonize plant roots has often been cited as a primary limitation to their effectiveness in field applications (Bloemberg and Lugtenberg, 2001). Despite this challenge, PGPR colonize root surfaces or interiors and exert significant positive effects on plant growth and development (Gerhardt *et al*., 2009). These attributes make PGPR an essential component of sustainable agricultural practices aimed at reducing reliance on chemical inputs.

MATERIALS AND METHODS

Characterization of the Site

The investigation was conducted in a greenhouse at the University of Gurukul Kangri Deemed to be University Haridwar in 2022. The field station is located in agro-ecological zone III at an altitude of 1800 m above sea level (ASL). The typical climate of the region is subhumid, with mean annual temperatures ranging from 12°C to 23°C and total annual precipitation of 1200-1800 mm. Rainfall is distributed bimodally, with prolonged rains occurring from March to May and shorter rains from October to November.

A. **Geographical distribution of samples: (A.(a))**- Healthy plant samples were collected from Haridwar regions of Roorkee, Bahadarabad, Shivalik nagar areas of Uttarakhand India.

Preparation of the Soil and Experimental Design

To prepare the soil, large clods and debris were removed by passing the soil through a 5 mm mesh sieve. The soil was solarized by covering it with a plastic sheet and exposing it to sunlight for one month (Pokharel, 2011). A mixture of soil and sterilized coco peat was prepared in a 1:1 ratio to serve as the growing medium in the greenhouse. Well-sprouted potato seeds were procured and planted in bread boxes lined with polythene bags. Each crate was filled with 20 kg of the soil-coco peat mixture, which was thoroughly mixed with 10 ml of bacterial inoculum. The mixture was watered regularly and allowed to stabilize for two weeks before planting four healthy potato seeds per crate.

Rhizobacteria Culture and Inoculum Preparation

The bacterial strains *B. japonicum* and *P. putida* were collected from authorized sources. Potato tubers (*S. tuberosum*) were supplied by the Agricultural Department. The bacterial strains were cultured for 48 hours in nutrient broth and yeast extract mannitol broth under controlled conditions (30°C, 120 rpm). The prepared inoculum was used for introducing bacteria to the tubers.

Inoculation of Rhizobacteria

At sowing, bacterial inoculants were applied in 4 rows, each at a depth of 6 cm. Treatments included *B. japonicum*, *P. putida*, a combination of both (*B. japonicum* + *P. putida*), and a control group with uninoculated compost. Approximately 5 µl (10^8 cfu/ml) of each rhizobacterium strain was applied according to the respective treatments. Each treatment was repeated three times using a factorial design within established bakery crates containing four plants. Parameters such as plant variety, microbial inoculants, and organic amendments were accounted for in the experimental layout.

Soil Nutrient Analysis

Soil samples (10 g) from the roots of each test pot were air-dried and agitated with 100 mL of 0.5 M ammonium acetate buffer for 30 minutes to extract minerals and nutrients. Carbon, nitrogen (N), phosphorus (P), and potassium (K) levels in the soil were measured using the method described by (Sims 2009). For this, 1.0 g of soil was mixed with 20 mL of concentrated H_2SO_4 and 10 mL of 1 N $K_2Cr_2O_7$. The suspension was blended, diluted with 200 mL distilled water, and treated with 10 mL each of sodium fluoride and H_3PO_4. Blank samples (without soil) were used as controls (Jabborova *et al*., 2021).

Plant Nutrient Analysis

One gram of pulverized plant tissue was placed in a phosphate buffer solution (pH 6.7) to determine the levels of essential plant nutrients, including phosphorus, potassium, nitrogen, magnesium, calcium, and sodium. The nutrient concentrations were analyzed using a spectrophotometer (Sharma and Kumawat, 2011). For nitrogen analysis, 1 g of dry leaf biomass was digested with 10 mL of concentrated H_2SO_4 and 5 g of a catalyst mixture. The nitrogen content was determined by titrating the cooled digest with H_2SO_4. Blank mixtures (without plant tissue) were used as controls (Daramola *et al*., 1994). Phosphorus content was extracted from the plant biomass using 0.5 N $NaHCO_3$ buffer (pH 8.5) and quantified at 540 nm following ascorbic acid treatment. Potassium levels were measured by mixing 5.0 g of plant biomass with 25 mL ammonium acetate, agitating for 5 minutes, and filtering the mixture (Sims, 2009). For magnesium, sodium, and calcium analysis, 1 g of plant extract was combined with 80 mL of 0.5 N HCl, incubated at 25°C for 5 minutes, and filtered. The filtrate was used to estimate the respective nutrient concentrations (Sahrawat, 1987).

Statistical Analysis

Each experiment was conducted in triplicate, and the means of these replicates were analyzed. Statistical analyses were performed using one-way ANOVA and the Tukey test with GraphPad Prism 5. The significance of treatment effects on plant growth, plant nutrient levels, and soil nutrient levels was assessed using p-values ($p < 0.05$).

RESULTS

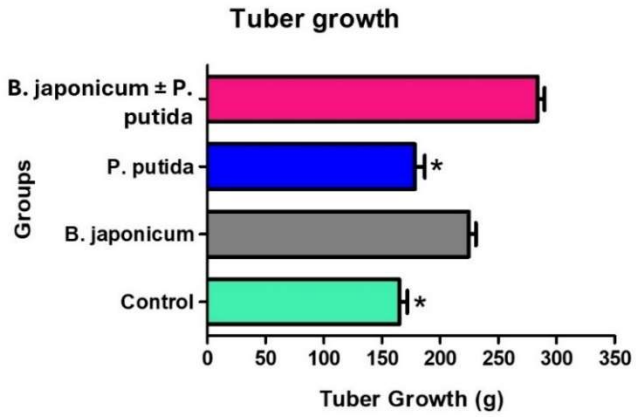

Figure 1 | Effect of co-inoculation on tuber growth in *S. tuberosum*. Graphical representation of tuber growth (g) under different treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

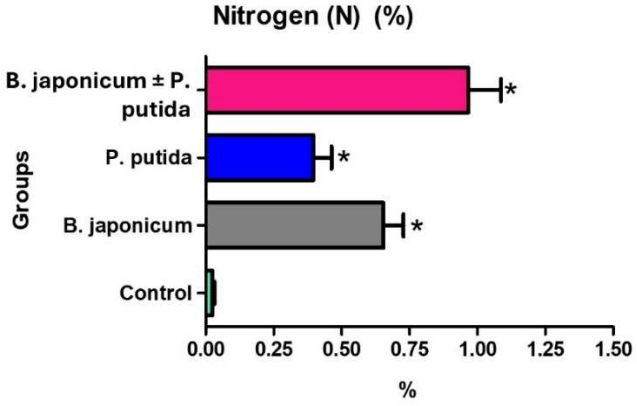

Figure 2 | Nitrogen (N) content in *S. tuberosum* under different inoculation treatments. Bar graph showing nitrogen content (%) in potato plants under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

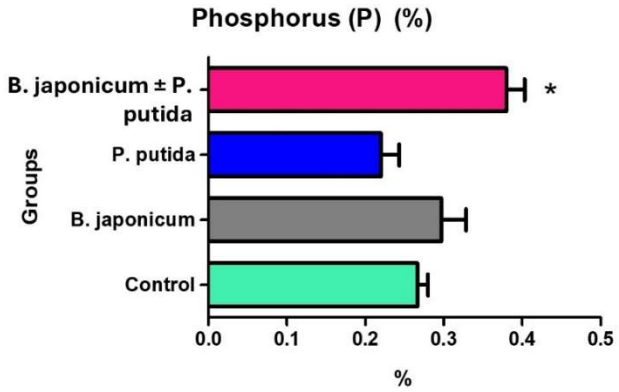

Figure 3 | Phosphorus (P) content in *S. tuberosum* under different inoculation treatments. Bar graph showing phosphorus content (%) in potato plants under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

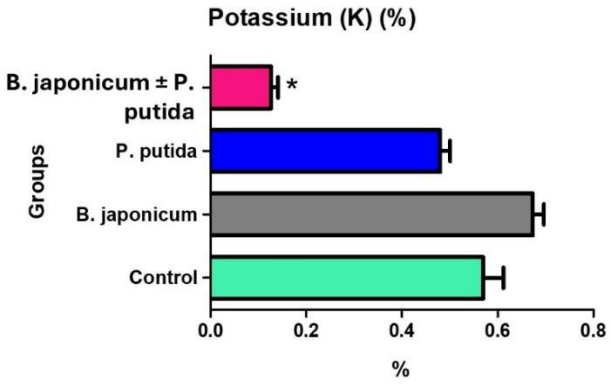

Figure 4 | Potassium (K) content in *S. tuberosum* under different inoculation treatments. Bar graph showing potassium content (%) in potato plants under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

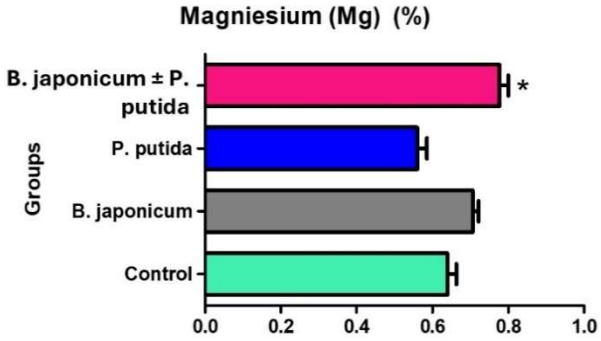

Figure 5 | Magnesium (Mg) content in *S. tuberosum* under different inoculation treatments. Bar graph showing magnesium content (%) in potato plants under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

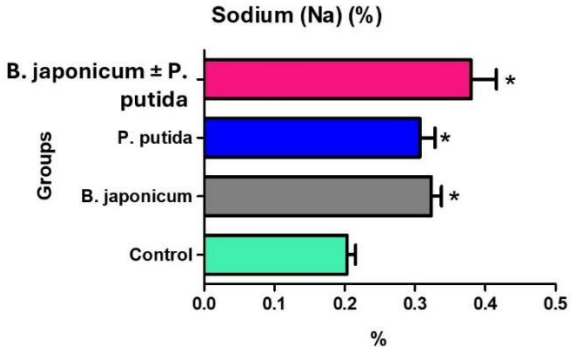

Figure 6 | Sodium (Na) content in *S. tuberosum* under different inoculation treatments. Bar graph showing sodium content (%) in potato plants under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

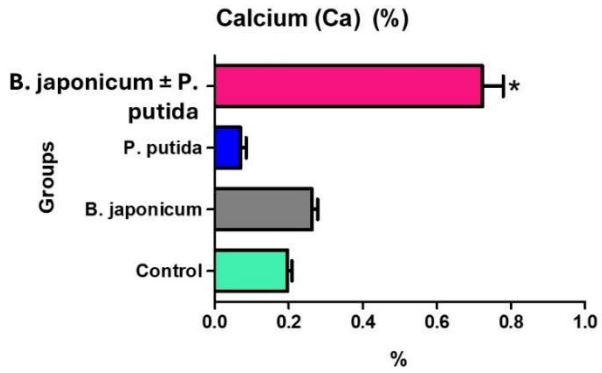

Figure 7 | Calcium (Ca) content in *S. tuberosum* under different inoculation treatments. Bar graph showing calcium content (%) in potato plants under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

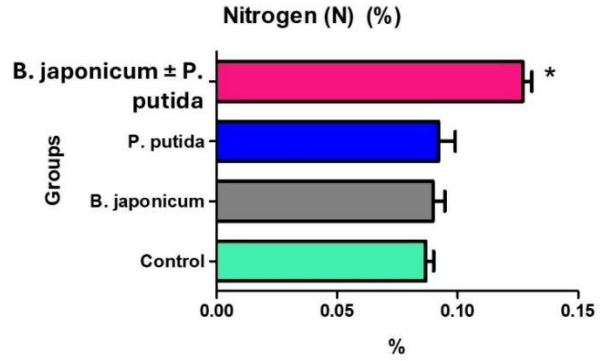

Figure 8 | Nitrogen (N) content in soil under different inoculation treatments. Bar graph showing nitrogen content (%) in soil under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

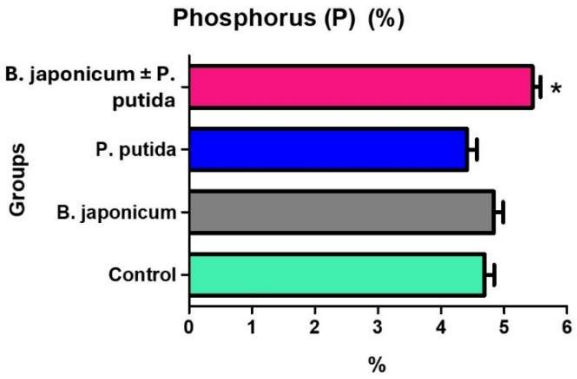

Figure 9 | Phosphorus (P) content in soil under different inoculation treatments. Bar graph showing phosphorus content (%) in soil under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

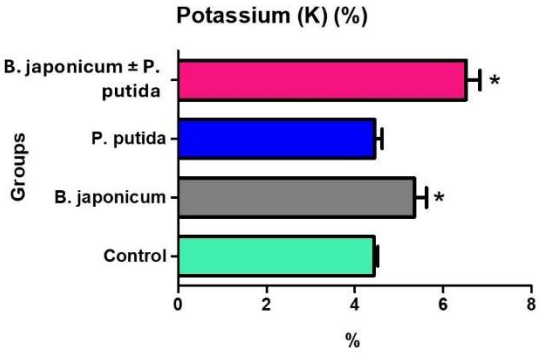

Figure 10 | Potassium (K) content in soil under different inoculation treatments. Bar graph showing potassium content (%) in soil under various treatments: control (uninoculated), inoculation with *B. japonicum*, inoculation with *P. putida*, and co-inoculation with *B. japonicum* and *P. putida*. Error bars represent the standard error of the mean (SD). ***p < 0.05**.

RESULTS AND DISCUSSION

Rhizobacteria Inoculants' Effects on Tuber Grading and Yield

Compared to the uninoculated control, tuber production was highest with the combined inoculation of *B. japonicum* and *P. putida*. Statistical analysis ($p < 0.05$) revealed that microbial inoculants and organic substances had limited effects on medium and small tuber grading **(Figure 1)**. However, the co-inoculation of both strains significantly enhanced tuber productivity compared to single-strain treatments. In contrast, plants grown without rhizobacterial inoculation (control group) showed notably lower productivity. (Zeffa *et al*. 2020) demonstrated that the simultaneous application of PGPR significantly enhances plant biomass. The results suggest that the co-inoculation technique is both a cost-effective and environmentally sustainable approach to improving plant productivity. (Berg *et al*. 2005) reported that rhizobacterial antagonists associated with potatoes

exhibit a high degree of specificity within distinct microenvironments. (Van and Van 2008) further investigated the role of plant genetics, soil conditions, and seasonal variations in shaping rhizobacterial communities associated with potatoes. Their findings highlighted that the stage of plant growth significantly influences the diversity and composition of rhizobacterial communities. These variations underscore the importance of integrating host-rhizobacteria systems to achieve effective bioprotection and enhanced crop yield (Davies *et al*., 2005). Such principles are equally applicable to potato cultivation (Klironomos and Hart, 2002), despite recommendations favoring local or native isolates for biotechnological applications (Klironomos, 2003). (Hassani *et al*., 2016) also observed in Iran that inoculating potato tubers with Pseudomonas spp. and Bacillus spp., either as independent or combined cultures, significantly improved tuberization and yield.

Nutrient Content Measurements for Plants

Analysis of potato nutrient composition showed that inoculation with *B. japonicum* alone resulted in significant increases in nutrient levels: nitrogen (N) **(Figure 2)** and phosphorus (P) **(Figure 3)** by 30%, potassium (K) **(Figure 4)** by 33%, magnesium (Mg) **(Figure 5)** by 13%, sodium (Na) **(Figure 6)** by 51%, and calcium (Ca) **(Figure 7)** by 14% compared to the control. Similarly, inoculation with *P. putida* alone increased N by 22%, P by 15%, K by 31%, Mg by 11%, Na by 46%, and Ca by 11% over the control. Co-inoculation with *B. japonicum* and *P. putida* further amplified these effects, increasing N by 45%, P by 35%, K by 42%, Mg by 20%, Na by 84%, and Ca by 42% compared to the control group. The provided table presents mean values of three replicates, with standard deviations indicating margins of error. Nutrient levels were measured 30 days post-planting. Despite instances of disease infestation, inoculated and organic treatments enhanced tuber weight, attributed to microbial production of hormones that promote root proliferation, improving nutrient absorption and plant vigor (Gupta *et al*., 2002). Organic amendments further facilitated the proliferation of beneficial soil microorganisms that functioned as biofertilizers (Larkins, 2008). (Santiago *et al*. 2017) emphasized the critical role of strain compatibility in designing bioinoculants for plant growth promotion. Their study demonstrated the compatibility of four bioinoculant strains co-inoculated to enhance potato development. Potassium-solubilizing rhizobacteria, including *Rhizobium* spp., Bacillus spp., and Pseudomonas spp., are associated with improved plant development and higher crop yields across various agricultural contexts (Yasin *et al*., 2016). Moreover, P-solubilizing microbial species have been extensively studied for their beneficial effects on diverse crops, including potato, tomato, wheat, radish, and pulses (Bhattacharyya and Jha, 2012).

Soil Nutrient Analysis

Soil nutrient analysis revealed that rhizobacterial inoculation significantly improved N **(Figure 8)**, P **(Figure 9)**, and K **(Figure 10)** levels compared to the control. Sole inoculation with *P. putida* increased N by 14%, P by 17%, and K by 14% relative to the control. Inoculation with *B. japonicum* alone showed a higher enhancement, increasing N by 17%, P by 19%, and K by 17%. However, co-inoculation with *B. japonicum* and *P. putida* exhibited the greatest improvements in soil N, P, and K levels. The data in Table 2 represent averages of three replicates, with standard deviations as error margins. Nutrient contents were measured 30 days post-planting. Single inoculation with *B. japonicum* resulted in higher NPK values compared to *P. putida*. However, co-inoculation consistently outperformed both single treatments and the control group. Adequate phosphorus (P) nutrition is essential for potato tuber growth and maturation, as well as for maintaining high photosynthetic rates during tuber development (Wu *et al*., 2013). Additionally, phosphorus has been shown to enhance potato protein content (Beals, 2007). Photosynthetic pigments in potato crops are highly influenced by NPK levels (Mona *et al*., 2012). Fageria (2005) reported that increasing nitrogen application enhances productivity, with potato

cultivation leading to significant increases in biomass yield, stripped stalk production, and juice content. Phosphorus plays a vital role in multiple metabolic reactions, further contributing to yield improvements (El-Arquan *et al*., 2002). The yield benefits of NPK application are also linked to potassium's role as an enzymatic co-factor and its ability to maintain electro-neutrality within plant cells (Mona *et al*., 2012).

CONCLUSION

The application of PGPR has been shown to positively influence the growth and development of potato plants by improving the absorption of key nutrients such as nitrogen (N), phosphorus (P), and potassium (K) from the soil under standard conditions. Inoculation with *B. japonicum* alone significantly enhances potato growth. However, the co-inoculation of *B. japonicum* with *P. putida* leads to superior improvements in plant growth, nutrient uptake, and soil nutrient levels compared to single inoculations or uninoculated control groups.

The combined use of *B. japonicum* and *P. putida* emerges as an effective, environmentally sustainable approach to enhancing potato productivity and soil nutritional composition. This study underscores the potential of employing PGPR as biofertilizers or inoculation agents to replace or complement chemical fertilizers, promoting sustainable agricultural practices while boosting plant development and yield. Further research in this area is crucial to optimize and expand the application of PGPR in diverse cropping systems.

REFERENCES

1. Aloo, B. N., Mbega, E. R., & Makumba, B. A., 2020. Rhizobacteria-based technology for sustainable cropping of potato (*Solanum tuberosum* L.). *Potato Research*, 63, 157-177.
2. Anjum, S. A., Xie, X., Wang, L. C., Saleem, M. F., Man, C., & Lei, W., 2011. Morphological, physiological and biochemical responses of plants to drought stress. *African Journal of Agricultural Research*, 6(9), 2026-2032.
3. Atieno, M., Herrmann, L., Nguyen, H. T., Phan, H. T., Nguyen, N. K., Srean, P., ... & Lesueur, D., 2020. Assessment of biofertilizer use for sustainable agriculture in the Great Mekong Region. *Journal of Environmental Management*, 275, 111300.
4. Beals, K. A., 2019. Potatoes, nutrition and health. *American Journal of Potato Research*, 96(2), 102-110.
5. Berg, G., 2009. Plant–microbe interactions promoting plant growth and health: perspectives for controlled use of microorganisms in agriculture. *Applied Microbiology and Biotechnology*, 84, 11-18.
6. Bhattacharyya, P. N., & Jha, D. K., 2012. Plant growth-promoting rhizobacteria (PGPR): emergence in agriculture. *World Journal of Microbiology and Biotechnology*, 28, 1327-1350.
7. Bloemberg, G. V., & Lugtenberg, B. J., 2001. Molecular basis of plant growth promotion and biocontrol by rhizobacteria. *Current Opinion in Plant Biology*, 4(4), 343-350.
8. Daramola, D. S., Danso, S. K. A., & Hardarson, G., 1994. Nodulation, N2 fixation and dry matter yield of soybean (*Glycine max* (L.) *Merrill*) inoculated with effective and ineffective *Bradyrhizobium japonicum* strains. *Soil Biology and Biochemistry*, 26(7), 883-889.
9. Davies Jr, F. T., Calderón, C. M., Huaman, Z., & Gómez, R., 2005. Influence of a flavonoid (formononetin) on mycorrhizal activity and potato crop productivity in the highlands of Peru. *Scientia Horticulturae*, 106(3), 318-329.
10. Fageria, N. K., & Baligar, V. C., 2005. Enhancing nitrogen use efficiency in crop plants. *Advances in Agronomy*, 88, 97-185.
11. Fageria, N. K., Baligar, V. C., & Jones, C. A., 1997. Growth and mineral nutrition of field crops. CRC Press.

12. Gerhardt, K. E., Huang, X. D., Glick, B. R., & Greenberg, B. M., 2009. Phytoremediation and rhizoremediation of organic soil contaminants: potential and challenges. *Plant Science*, 176(1), 20-30.
13. Gupta, C., Dubey, R., & Maheshwari, D., 2002. Plant growth enhancement and suppression of *Macrophomina phaseolina* causing charcoal rot of peanut by *Fluorescent Pseudomonas*. *Biology and Fertility of Soils*, 35, 399-405.
14. Hassani, F., Asgharzadeh, A., Ardakani, M., & Hamidi, A., 2015. The impact of potato mini-tuber inoculation with plant growth-promoting rhizobacteria on tuber yield and nutrients uptake. *Journal of Crops Improvement*, 17(4).
15. Hill, J., & Lazarovits, G., 2005. A mail survey of growers to estimate potato common scab prevalence and economic loss in Canada. *Canadian Journal of Plant Pathology*, 27(1), 46-52.
16. Hochmuth, G. J., & Hanlon, E., 2000. A summary of N, P, and K research on potato in Florida. University of Florida Cooperative Extension Service, Institute of Food and Agriculture Sciences, EDIS.
17. Hungria, M., Nogueira, M. A., & Araujo, R. S., 2013. Co-inoculation of soybeans and common beans with *Rhizobia* and *Azospirilla*: strategies to improve sustainability. *Biology and Fertility of Soils*, 49, 791-801.
18. Jabborova, D., Kannepalli, A., Davranov, K., Narimanov, A., Enakiev, Y., Syed, A., ... & Gafur, A., 2021. Co-inoculation of rhizobacteria promotes growth, yield, and nutrient contents in soybean and improves soil enzymes and nutrients under drought conditions. Scientific Reports, 11(1), 22081.
19. Klironomos, J. N., 2003. Variation in plant response to native and exotic arbuscular mycorrhizal fungi. *Ecology*, 84(9), 2292-2301.
20. Klironomos, J. N., & Hart, M. M., 2002. Colonization of roots by arbuscular mycorrhizal fungi using different sources of inoculum. *Mycorrhiza*, 12, 181-184.
21. Kloepper, J. W., Schroth, M. N., & Miller, T. D., 1980. Effects of rhizosphere colonization by plant growth-promoting rhizobacteria on potato plant development and yield. *Phytopathology*, 70(11), 1078-1082.
22. Larkin, R. P., 2008. Relative effects of biological amendments and crop rotations on soil microbial communities and soilborne diseases of potato. *Soil Biology and Biochemistry*, 40(6), 1341-1351.
23. Makbul, S., Güler, N. S., Durmuş, N., & Güven, S., 2011. Changes in anatomical and physiological parameters of soybean under drought stress. *Turkish Journal of Botany*, 35(4), 369-377.
24. Martínez-Viveros, O., Jorquera, M. A., Crowley, D. E., Gajardo, G. M., & Mora, M. L., 2010. Mechanisms and practical considerations involved in plant growth promotion by rhizobacteria. *Journal of Soil Science and Plant Nutrition*, 10(3), 293-319.
25. Mohammadi, K., & Sohrabi, Y., 2012. Bacterial biofertilizers for sustainable crop production: a review. *ARPN Journal of Agricultural and Biological Science*, 7(5), 307-316.
26. Mona, E. E., Ibrahim, S. A., & Manal, F. M., 2012. Combined effect of NPK levels and foliar nutritional compounds on growth and yield parameters of potato plants (*Solanum tuberosum* L.). *African Journal of Microbiology Research*, 6(24), 5100-5109.
27. Naqqash, T., Hameed, S., Imran, A., Hanif, M. K., Majeed, A., & van Elsas, J. D., 2016. Differential response of potato toward inoculation with taxonomically diverse plant growth-promoting rhizobacteria. *Frontiers in Plant Science*, 7, 144.
28. Pokharel, R., 2011. Soil solarization, an alternative to soil fumigants. Fact sheet (Colorado State University. Extension). *Crop series*; no. 0.505.
29. Sahrawat, K. L., 1987. Determination of calcium, magnesium, zinc and manganese in plant tissue using a dilute HCl extraction method. *Communications in Soil Science and Plant Analysis*, 18(9), 947-962.
30. Santiago, C. D., Yagi, S., Ijima, M., Nashimoto, T., Sawada, M., Ikeda, S., ... & Ohwada, T., 2017. Bacterial compatibility in combined inoculations enhances the growth of potato seedlings. *Microbes and Environments*, 32(1), 14-23.

31. Sharma, M. K., & Kumawat, D. M., 2011. Co-inoculation study of *Bradyrhizobium japonicum* and *Aspergillus niger* in soybean for nitrogen fixation. *Journal of Microbiology, Biotechnology and Food Sciences*, 1(3), 383-394.
32. Sims, J. T., 2009. Soil test phosphorus: Principles and methods. Methods of Phosphorus Analysis for Soils, Sediments, Residuals and Waters, 2nd edn. *Southern Cooperative Series bulletin, 408*, 9-19.
33. Sureshbabu, K., Amaresan, N., & Kumar, K., 2016. Amazing multiple function properties of plant growth-promoting rhizobacteria in the rhizosphere soil. *International Journal of Current Microbiology and Applied Sciences*, 5(2), 661-683.
34. Van Overbee, L., & Van Elsas, J. D., 2008. Effects of plant genotype and growth stage on the structure of bacterial communities associated with potato (*Solanum tuberosum* L.). FEMS Microbiology Ecology, 64(2), 283-296.
35. Wu, F., Wang, W., Ma, Y., Liu, Y., Ma, X., An, L., & Feng, H., 2013. Prospect of beneficial microorganisms applied in potato cultivation for sustainable agriculture. *African Journal of Microbiology Research*, 7(20), 2150-2158.
36. Yasin, M., Mnir, I., & Faisal, M., 2016. Can Bacillus spp. enhance K^+ uptake in crop species? *Potassium Solubilizing Microorganisms for Sustainable Agriculture*, 163-170.
37. Zeffa, D. M., Fantin, L. H., Koltun, A., de Oliveira, A. L., Nunes, M. P., Canteri, M. G., & Gonçalves, L. S., 2020. Effects of plant growth-promoting rhizobacteria on co-inoculation with *Bradyrhizobium* in soybean crop: a meta-analysis of studies from 1987 to 2018. PeerJ, 8, e7905.