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Role of Beneficial Microbes in Enhancing Potato Vegetative Stage in Sindhupalchwok District

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ABSTRACT

The study titled "Role of Beneficial Microbial Inoculants in Enhancing Potato Vegetation Growth in Sindhupalchok District" was conducted at the Potato Crop Development Centre, Nigale, from February 21 to July 11, 2025. A Randomized Complete Block Design (RCBD) with five treatments- Control, Trichoderma, Pseudomonas, Phosphate-Solubilizing Bacteria (PSB), and a 1:1:1 mixture was used, each replicated four times. Data on vegetative growth were recorded and analyzed statistically using R Studio. At 45 days after planting, plant emergence was highest under PSB (58.33%), while plant height at 105 DAP was greatest in Trichoderma (83.9 cm). Although several variables showed no statistically significant differences ($p \leq 0.05$), consistent numerical improvements occurred in Trichoderma and mixture treatments, indicating that beneficial microbial inoculants can enhance vegetative growth and quality in the mid-hill regions of Nepal.

Keywords: Biofertilizer, microbial inoculants, sustainable, treatment, yield

INTRODUCTION

One of the primary goals of Nepal's Agriculture Development Strategy (ADS) is to enhance agricultural productivity while ensuring long-term sustainability and food security. In line with this objective, potatoes have been identified as a priority crop under the Prime Minister Agriculture Modernization Project (PMAMP) because of their significant contribution to national food systems, nutrition, and rural livelihoods. Potato cultivation area and production in Nepal have steadily increased since 2016/17, with national productivity rising from 13.94 t/ha to 17.11

t/ha over the last decade until 2022/23 (MoALD, 2079). Despite this progress, Nepal's potato productivity remains substantially lower than the global average. Countries such as the United States and New Zealand achieve average potato yields of 51.45 t/ha and 50.88 t/ha, respectively (FAO, 2025), which are more than three times higher than Nepal's current productivity level. This considerable yield gap highlights the urgent need for sustainable and efficient production technologies suitable for Nepalese agroecological conditions.

Recent advances in sustainable agriculture have emphasized the importance of microbial inoculants as eco-friendly alternatives to excessive chemical fertilizer use. Continuous and imbalanced application of synthetic fertilizers and pesticides has accelerated soil degradation, reduced beneficial microbial activity, and negatively affected agroecosystem health. Soil microorganisms play a crucial role in nutrient cycling, organic matter decomposition, soil aggregation, and plant growth promotion; however, intensive chemical-based farming systems often suppress these beneficial microbial populations (Vessey, 2003). The adverse effects of agrochemical dependency can be observed in areas such as Panchkhal Municipality, once widely recognized for commercial vegetable production. Excessive pesticide use has resulted in pesticide-resistant pests such as "Daduwa," increased production costs, phytotoxicity, soil quality deterioration, and chemical residues in vegetables (Karmacharya, 2012). Farmers are increasingly trapped in a cycle of chemical dependency, where crop performance declines without repeated applications of fertilizers and pesticides.

In this context, beneficial microbial inoculants such as *Trichoderma*, phosphate-solubilizing bacteria (PSB), and *Pseudomonas* spp. have gained increasing attention as sustainable biofertilizer options. These microorganisms play significant roles in improving soil fertility, nutrient availability, plant growth, and disease suppression. Microbial inoculants enhance crop productivity through several mechanisms, including nutrient solubilization, phytohormone production, induced systemic resistance, and antagonistic activity against soil-borne pathogens (Harman et al., 2004; Vessey, 2003). Their application has become particularly important in potato cultivation because of the crop's high nutrient demand and susceptibility to soil-borne diseases.

Trichoderma species are widely recognized for their biocontrol potential and plant growth-promoting abilities. They suppress pathogenic fungi through mechanisms such as mycoparasitism, competition, antibiosis, and induction of plant defense responses. In addition, *Trichoderma* improves root development and nutrient uptake, thereby enhancing overall plant vigor and yield (Harman, 2006). Similarly, phosphate-solubilizing bacteria (PSB) convert insoluble forms of phosphorus into plant-available forms through the secretion of organic acids and phosphatase enzymes. Since phosphorus deficiency is a common limitation in agricultural soils, PSB can significantly improve phosphorus use efficiency and reduce dependency on synthetic phosphate fertilizers (Rodríguez and Fraga, 1999). Likewise, plant growth-promoting *Pseudomonas* spp. contribute to crop performance through nitrogen fixation, siderophore production, phytohormone synthesis, and suppression of pathogenic microorganisms. Fluorescent *Pseudomonas* strains are particularly important in enhancing rhizosphere activity and promoting healthier root systems, ultimately improving crop growth and productivity (Weller, 2007).

Although biofertilizers and microbial inoculants have demonstrated promising results in several crops globally, limited field-based research has been conducted under Nepalese agroecological conditions, particularly in potato cultivation systems. Most previous studies have focused on laboratory-scale experiments or individual microbial strains, creating a significant research gap regarding the combined application of *Trichoderma*, PSB, and *Pseudomonas* in enhancing potato productivity and soil health under local farming conditions. Furthermore, the interaction of these inoculants with the soils and climatic conditions of Nepal, especially in the Sindhupalchowk district, remains insufficiently understood. Therefore, this study hypothesizes that the application of microbial inoculants, individually or in combination, will significantly improve potato growth, nutrient uptake, soil health, and yield compared to non-inoculated treatments.

MATERIALS AND METHODS

The study was conducted from February to July 2025 at the Potato Crop Development Center (PCDC), located at an elevation of approximately 2,500 meters above sea level in Nigale, Lisankhupakhar Rural Municipality, Sindhupalchok District, Nepal. The site lies within a moderate temperate climatic zone suitable for potato cultivation. PCDC was purposively selected because it is a national government institution engaged in potato research, varietal evaluation, seed production, and technology development in Nepal. During the experimental period, the crop was grown under prevailing field conditions of the site.

Before field preparation and treatment application, representative soil samples were collected from the experimental area at a depth of 0–20 cm using a standard zigzag sampling method. Several subsamples were taken across the field, thoroughly mixed to form a composite sample, air-dried, and analyzed for major physicochemical properties such as soil texture, pH, organic matter, available nitrogen, available phosphorus, and available potassium. These baseline soil characteristics were used to describe the fertility status of the experimental site and to support the interpretation of the treatment effects.

The experiment was laid out as a Randomized Complete Block Design. A total of five treatments were implemented, each replicated four times, resulting in 20 plots across the experimental field. The entire plot area covered 80 m², with each plot measuring 2 m × 2 m (4 m²). Each plot contained 21 potato tubers, making a total of 420 tubers across all plots. The **Rosita** potato variety, which is popular and well-suited to the agro-climatic conditions of Nigale, was selected for planting due to its high yield potential and local adaptability. Row-to-row spacing was maintained at 70 cm, while plant-to-plant spacing within rows was 25 cm to ensure proper growth and minimize competition among plants. Fertilizer was applied at the recommended N:P: rate of 100:60:60 kg/ha. Nitrogen was applied in split doses to enhance nutrient uptake and reduce potential losses through leaching or volatilization.

The treatments included PSB, *Trichoderma*, *Pseudomonas*, Control, and Mixture. Three products bearing the brand names Ferti P, Psumo, and Tree Devi were used as liquid biofertilizers from Yarsa Agro Research Pvt. Ltd., located in Lalitpur, Nepal. Ferti P is a phosphate-solubilizing biofertilizer containing *Phosphate-Solubilizing Bacteria (PSB)* at a concentration of 2×10^8 CFU/ml. Psumo is a biocontrol agent containing *Pseudomonas fluorescens* (2×10^8 CFU/ml), and Tree Devi is a biofungicide containing *Trichoderma viride* at 2×10^9 CFU/ml.

One liter of water was combined with five milliliters of each biofertilizer for the application. For the entire plot, five liters of solution were made. One of the five treatments was a combination formulation, in which five milliliters of each of the three biofertilizers were combined with five liters of water in a 1:1:1 ratio. First, seed tubers were steeped in the biofertilizer mixture for half an hour before planting. On April 25, right before flowering, a second foliar spray application was carried out. The crop was then sprayed with biofertilizer every 15 days until it reached physiological maturity, which happened 105 days after planting (DAP).

RESULTS AND DISCUSSION

Germination percentage

At 45DAP, the data revealed that the highest number of plants emerged was recorded in T_4(PSB) with 58.33%, followed by T_2(Trichoderma), 54.76%, T_5(Mixture), 40.48%, and T_3(Pseudomonas), 35.71%, with the least germination on T_1(control), 32.14%. The ANOVA results for germination percentage show that neither the replication nor the treatment effects were statistically significant. The F-value for replication was 1.85 ($p = 0.192$), and for treatment, it was 1.38 ($p = 0.299$), both of which exceed the 5% significance level. This indicates that neither block-to-block variation nor the applied treatments had a statistically meaningful influence on the germination rate of the seeds. The mean square error (Ms error) was 393.62, and the overall mean germination rate was 44.29%. However, the experiment exhibited a high coefficient of variation (CV) of 44.80%, and the least significant difference (LSD) at the 5% level was 30.57%. This high LSD suggests that only differences above 30.57 percentage points between treatment means would be statistically detectable.

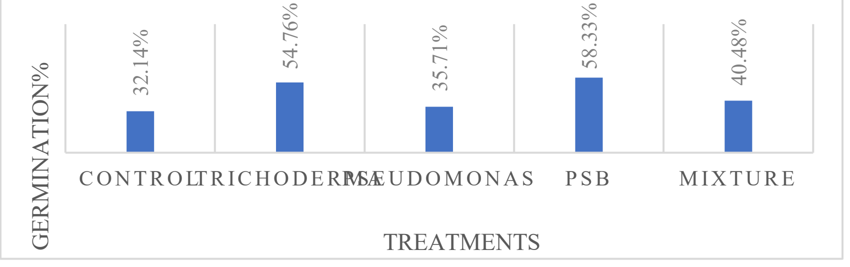

Figure 1. Graph illustrating the germination percentage

The high CV indicates substantial variability in germination performance, possibly due to environmental factors such as uneven moisture content, seed vigor differences, or inconsistencies in seedbed preparation. According to Joshi *et al.* (2020), variability in seed germination across treatments is common in field conditions, especially when seed quality and micro-environmental conditions are not uniform. The non-significant treatment effect implies that the germination differences observed among the treatments are likely due to random variation rather than the treatments themselves. This aligns with the findings of Adhikari *et al*. (2017), who reported non-significant differences in germination among tomato varieties under similar treatment conditions, attributing it to genetic uniformity and uniform pre-germination conditions.

Plant height

The analysis of plant height across different treatments from 60 to 105 Days After Planting (DAP) reveals a consistent pattern where T_2 exhibited superior vegetative growth, particularly in the later stages. At 60DAP, although T_5 recorded the highest mean plant height (12.75 cm), it was closely followed by T_4 (12.7 cm) and T_2 (12.45 cm), with no significant differences among treatments (p = 0.3187). The coefficient of variation (CV%) was relatively high at 45.04%, indicating considerable variability in early growth. According to Sharma et al. (2016), early plant height can often be influenced by seedling vigor and initial soil moisture conditions, which may not necessarily predict final plant performance.

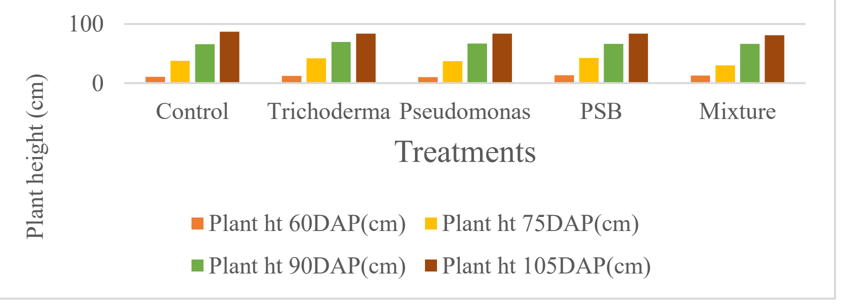

Figure 2. Graph illustrating plant height

At 75DAP, T_4 achieved the highest mean height (42.7 cm), slightly ahead of T_2 (41.85 cm), suggesting that both treatments promoted better vegetative growth than the others. The growth patterns align with findings by Adhikari *et al. (*2020), who noted that mid-vegetative stages are critical in determining overall plant performance, particularly under varied agronomic treatments. However, the differences remained statistically non-significant (p = 0.1244), indicating that the variability within treatment groups overlapped.

The superiority of T_2 became more pronounced at 90 DAP and 105 DAP, where it recorded the highest mean plant heights of 69.65 cm and 83.9 cm, respectively. Although these differences were still not statistically significant (p = 0.6285 and p = 0.8004), the consistent numerical advantage of T_2 suggests its effectiveness in supporting vegetative development in later stages. Similar observations were reported by Thapa *et al. (*2021), who emphasized that while statistical significance is important, consistent trends over time can still hold agronomic relevance, especially under field conditions where variability is common.

The lack of significant differences could be attributed to environmental variability or limited replication, as suggested by the relatively high coefficient of variation (CV%) values. Nonetheless, T_2's consistent performance supports its potential suitability for robust vegetative growth, even in the absence of statistical confirmation. This is supported by the work of Paudyal and Regmi (2018), who argue that agronomic decisions in field settings often rely on consistent trends rather than solely on statistical thresholds. T_2 demonstrated the most consistent and superior vegetative growth performance across later growth stages, highlighting its promise as a treatment option for improved plant height and vigor. Future studies with larger sample sizes and controlled environments may help confirm the statistical validity of these trends.

Number of stems

The ANOVA results for the number of stems indicate that neither replication nor treatment effects were statistically significant. The F-value for replication was 0.24 ($p = 0.869$), and for treatment, it was 1.24 ($p = 0.345$), suggesting that the variation in stem number among different treatments and across replications was not large enough to be considered significant at the 5% level. The mean square error (MS error) was 0.594, with an overall mean number of stems of 4.83 cm and a coefficient of variation (CV) of 15.96%, indicating moderate variability in the data. The least significant difference (LSD) at the 5% level was calculated to be 1.19 cm, implying that any treatment differences below this threshold are statistically indistinguishable.

The non-significant treatment effect suggests that the tested treatments did not have a notable influence on the number of stems. This may be due to environmental uniformity, limited physiological response, or the short duration of treatment exposure. Similar findings were reported by Pandey *et al*. (2016), who found no significant effect of organic and inorganic fertilizer treatments on stem number in early vegetative stages of tomato due to uniform seedling vigor and initial soil fertility levels. Additionally, the non-significant replication effect suggests consistent conditions across experimental blocks, indicating that extraneous variability such as soil texture or light exposure was well-controlled.

This strengthens the internal validity of the experiment, a feature considered critical in field trials (Gomez and Gomez, 1984).

Table 1. ANOVA for Number of Stems

Response Variable	Source	Df	Sum Sq	Mean Sq	F value	Pr (>F)	M Serror	Mean	CV (%)	LSD (5%)
Stem Length	Replication	3	0.422	0.1407	0.24	0.869	0.594	4.83	15.96	1.19
	Treatment	4	2.952	0.7380	1.24	0.345				
	Residual	12	7.128	0.5940						

Uniformity

The analysis of plant uniformity across different Days After Planting (DAP) reveals important insights into the consistency of treatment effects throughout the crop's growth period. At 60 DAP, significant differences among treatments were observed ($p < 0.001$), with T_2 and T_3 achieving the highest uniformity scores (3.75), significantly outperforming T_4 and T_5 (both 2.75), as supported by the LSD (0.43). The coefficient of variation (CV%) was 21.51%, indicating moderate variability among the experimental units. Uniformity at early growth stages is crucial, as it reflects synchronized development, which can enhance management practices and potentially contribute to better final yield and marketable quality (Gomez and Gomez, 1984). High early uniformity can also reduce intra-plot competition, contributing to more efficient resource use (Adhikari *et al.,* 2021). At 75DAP, all treatments recorded a uniformity score of 4, resulting in non-significant differences ($p = 0.4116$) and a CV% of 0%, signifying complete uniformity across treatments. This uniform growth phase may represent a developmental equilibrium, possibly influenced by favorable environmental conditions and uniform agronomic management. According to Shrestha et al. (2019), such uniformity in mid-growth stages is common in well-managed field trials and indicates physiological stability.

By 90 DAP, although the F-value increased slightly (1.68), the treatment effect remained non-significant ($p = 0.1618$). The highest uniformity (4.5) was noted in T_1 and T_3, while T_2, T_4, and T_5 followed closely with a score of 4.25. The CV% of 10.87% shows mild variation, suggesting that treatment effects were beginning to diverge, but not strongly enough to reach statistical significance. As per Khadka et al. (2022), uniformity in late vegetative or early reproductive stages can be influenced by genotype-environment interactions, which may introduce variability not directly linked to treatment interventions.

At 105DAP, a highly significant treatment effect was once again observed ($p < 0.001$), with T_1, T_3, and T_5 achieving the highest uniformity (4.5), compared to T_2 and T_4 (both 4.0). The LSD at this stage was lower (0.25), and the CV% was 9.24%, suggesting a relatively stable measurement with reduced variability. This stage is critical for evaluating harvest maturity and grading potential. According to Paudel et al. (2020), high uniformity at maturity ensures better post-harvest handling, enhances the market appeal, and reduces sorting losses.

While T_2 showed the best performance at 60 DAP and maintained stable uniformity throughout the growth cycle, its performance slightly declined during the later stages compared to T_1, T_3, and T_5. However, its consistency across all four growth stages still positions it as a reliable treatment for achieving predictable plant development. As argued by Steel and Torrie (1980), stability across time often outweighs peak performance in a single stage, especially in field conditions where uniformity supports operational efficiency and production planning.

The ANOVA results show that treatment impacts on plant uniformity were temporally dynamic, statistically significant in the early and final stages, but not during mid-growth. This pattern emphasizes the importance of stage-specific evaluation in agronomic research, where early and late-stage interventions may exert the most influence. These findings align with the broader principle noted by Steel and Torrie (1980), which advocates interpreting treatment effects in light of developmental phases and environmental interactions.

Table 2. ANOVA for the uniformity

Treatments	**Uniformity (Day 60)**	**Uniformity (Day 75)**	**Uniformity (Day 90)**	**Uniformity (Day 105)**
T_1 (Control)	3	4	4.5	4.5
T_2 (Trichoderma)	3.75	4	4.25	4
T_3 (Pseudomonas)	3.75	4	4.5	4.5
T_4 (PSB)	2.75	4	4.25	4
T_5 (Mixture)	2.75	4	4.25	4.5
LSD (0.05)	0.43			0.25
F	11.08	1	1.68	9.5
p-Value	2.03e-07	0.4116	0.1618	1.67e-06
CV%	21.51	0	10.87	9.24

CONCLUSION

In this study, the treatments applied—*Trichoderma*, *Pseudomonas*, phosphate-solubilizing bacteria (PSB), and their combined mixture—did not produce statistically significant effects on major growth parameters. However, numerical differences were observed among treatments, which were largely influenced by environmental variability.

Despite the lack of strong statistical significance, several consistent agronomic trends were evident throughout the experiment. Microbial inoculant treatments contributed to improved plant uniformity at different growth stages. Significant differences in uniformity observed at 60 and 105 DAP suggest that treatment responses were stage-dependent rather than uniform throughout the crop cycle. Treatments involving *Trichoderma* and *Pseudomonas* performed better during the early stages, whereas the mixture treatment and *Pseudomonas* maintained greater uniformity at maturity.

The study further suggests that microbial inoculants can serve as sustainable and eco-friendly supplements to conventional chemical fertilizers by enhancing nutrient availability, improving rhizosphere activity, and supporting healthier crop growth. In the context of increasing soil degradation, declining microbial activity, and excessive agrochemical dependency in Nepalese agriculture, the use of biofertilizers such as *Trichoderma*, *Pseudomonas*, and PSB offers promising opportunities for sustainable potato production and long-term soil health management.

SUGGESTIONS

Future research should focus on long-term and multi-location trials with larger sample sizes, improved experimental precision, and additional assessments related to soil biological properties, nutrient dynamics, and yield performance. Such studies would help validate the observed trends and provide deeper insights into the interaction of microbial inoculants with local soil and climatic conditions.

AUTHORS' CONTRIBUTIONS

Koshila Giri: Investigation; Methodology; Conceptualization; Data curation; Software; Formal analysis; Visualization; Writing—original draft; Writing—review and editing.
Himal Bhusal: Conceptualization; Validation.

Prashant Sapkota: Conceptualization; Validation.
Rojan Karki: Writing-review and editing, Software; Formal analysis

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