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Effects of Phosphate-Solubilizing Bacteria and Organic Fertilizer Types on the Vegetative Growth of Waxy Maize (*Zea mays var. ceratina*)

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Abstract

Limited soil phosphorus (P) availability can restrict the early growth of waxy maize, especially when P is fixed in forms that are not readily absorbed by roots. The combined use of phosphate-solubilizing bacteria (PSB) and organic amendments offers a potential strategy to improve nutrient availability while supporting more sustainable soil management. This study examined the individual and combined effects of PSB doses and organic fertilizer types on the vegetative growth of waxy maize. A 4 x 3 factorial randomized block design with three replications was used. The first factor was PSB dose, namely 0, 10, 20, and 30 mL ha-1, whereas the second factor was organic fertilizer type, namely no organic fertilizer, cattle manure at 15 t ha-1, and chicken manure at 15 t ha-1. Plant height, leaf number, and leaf area were recorded from 1 to 5 weeks after planting (WAP). Data were evaluated using analysis of variance, followed by Duncan's Multiple Range Test at the 5% level when treatment effects were significant. PSB application did not significantly influence any observed vegetative parameter. In contrast, organic fertilizer type produced significant differences, with chicken manure giving the highest plant height, leaf number, and leaf area. No significant PSB x organic fertilizer interaction was detected. These findings indicate that, under the conditions of this experiment, chicken manure was more influential than PSB dose in improving early waxy maize growth.

Keywords*: waxy maize, phosphate-solubilizing bacteria, organic fertilizer, vegetative growth*

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INTRODUCTION

Waxy maize (*Zea mays L. var. ceratina*) is a specialized maize variety distinguished by an exceptionally high amylopectin content of 95–100%, compared to approximately 72–75% in common maize. This characteristic confers a distinctly glutinous, sticky texture upon cooking, resulting in considerable consumer appeal and economic value. Beyond its role as a fresh food commodity and traditional culinary ingredient, waxy maize holds significant industrial potential as a raw material for specialty starch production.

With the increasing demand for food and the diversification of processed products, the development of waxy maize cultivation has become increasingly strategic. Data from the Central Statistics Agency (2024) shows that national maize production in 2023 reached 21.73 million tons. However, increased productivity, particularly for specialty maize varieties such as waxy maize, is still needed to meet growing domestic demand. Maize productivity is strongly influenced by nutrient availability, particularly phosphorus (P). Among the essential macronutrients for maize, phosphorus holds a central position because it participates in nearly all fundamental growth processes, from energy metabolism through ATP to the structural requirements of nucleic acids and cell membranes, making its availability during vegetative growth particularly consequential (Santos et al., 2020). During the vegetative phase, adequate phosphorus levels determine optimal leaf area, plant height, and root development, which in turn contribute to photosynthetic capacity and biomass formation. Phosphorus deficiency in the early growth phase can inhibit canopy and root system development, thereby reducing yield potential. The main issue in phosphorus management lies not only in the total amount present in the soil but also in its availability. Approximately 95–99% of soil phosphorus is in an unavailable form due to binding with Al and Fe in acidic soils or Ca in alkaline soils (Manzoor et al., 2022).

This condition results in relatively low efficiency of inorganic phosphate fertilizers, as most of it undergoes re-fixation after application. While intensive use of chemical fertilizers can indeed increase phosphorus availability in the short term, it has the potential to degrade soil quality, increase the risk of eutrophication, and is economically and ecologically unsustainable (Penn & Camberato, 2019; Dhaliwal et al., 2021). Therefore, alternative approaches that are more efficient and sustainable are needed to enhance phosphorus availability.

The utilization of phosphate-solubilizing bacteria (PSB) is a promising biotechnological strategy. The capacity of PSB to mobilize fixed phosphorus operates through several overlapping pathways: secretion of low molecular weight organic acids that dissolve mineral phosphate complexes, enzymatic hydrolysis of organic P via phosphatase activity, and chelation reactions that shift bound phosphate into the soil solution as

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orthophosphate accessible to plant roots (Alori et al., 2017; Nath et al., 2023). Several genera, such as Pseudomonas, Bacillus, Azotobacter, and Enterobacter, have been reported to be effective in enhancing P uptake and maize plant growth (Adnan et al., 2020; Suleman et al., 2021).

The effectiveness of phosphate-solubilizing bacteria is highly influenced by soil environmental conditions and the availability of organic substrates as an energy source for microorganisms. In this context, organic fertilizers play a crucial role in enhancing soil fertility holistically. Beyond their role as nutrient sources, organic fertilizers contribute to a broader improvement of the soil environment, including enhanced aggregate stability, greater capacity to retain both water and cations, and a more diverse and active microbial community that supports nutrient cycling (Bulgari et al., 2021; Mehmood et al., 2022). Different types of organic fertilizers, such as chicken manure and cattle manure, have varying nutrient content, C/N ratios, and decomposition rates, so plant responses to each type of fertilizer may vary (Lazcano & Domínguez, 2021; Setyorini et al., 2022). Theoretically, the combination of phosphate-solubilizing bacteria and organic fertilizers has the potential to produce a synergistic effect.

Organic matter can serve as a source of carbon and energy for phosphate-solubilizing bacteria (PSB), thereby enhancing phosphate solubilization activity in the rhizosphere. Conversely, microbial activity can accelerate the decomposition of organic matter and the release of nutrients, including phosphorus, into the soil solution (Yadav et al., 2023). Several studies indicate that the integration of biofertilizers and organic fertilizers can enhance vegetative growth and maize productivity (Suleman et al., 2021; Hidayat et al., 2023).

However, most of these studies have focused on hybrid corn or sweet corn, while specific studies on waxy maize remain limited. Furthermore, few studies have directly compared various types of organic fertilizers in combination with P-solubilizing bacteria (PSB) regarding the vegetative growth of waxy maize. Yet, the vegetative phase is a crucial stage that determines the capacity for canopy and root system development, which serves as the foundation for yield formation. The lack of comprehensive information regarding the response of waxy maize to the combination of phosphate-solubilizing bacteria and types of organic fertilizers indicates a research gap that needs to be addressed.

Therefore, this study evaluated the effects of phosphate-solubilizing bacteria and organic fertilizers on the vegetative growth of waxy maize, as well as their interaction. The results of this study are expected to contribute scientifically to the development of an integrated bio-based and organic fertilization strategy that is more efficient, environmentally friendly, and sustainable for waxy maize cultivation.

Materials and Methods

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Time and Location

The field experiment was carried out from October 2025 to January 2026 in Pone Hamlet, Ilomangga Village, Tabongo District, Gorontalo Regency, Gorontalo Province, Indonesia. The experimental site is located at approximately 0 deg 34'43.7" S and 122 deg 56'1.7" E, with an elevation of about 10-15 m above sea level. The soil pH at the site was 6.56, indicating slightly acidic conditions.

Tools and Materials

The tools used in this study included a tractor, hoe, shovel, measuring tape, raffia rope, sprayer, and weighing scale. The materials used included waxy maize seeds, cattle manure, chicken manure, and an isolate of phosphate-solubilizing bacteria, *Pseudomonas fluorescens*, obtained from BRMP Gorontalo, the Agricultural Assembly and Modernization Agency.

Research Design

The experiment was arranged in a factorial randomized block design involving two treatment factors, namely PSB application and type of organic fertilizer. The first factor, Factor A, was the dose of PSB application, consisting of four treatment levels as follows: A0 = without PSB, A1 = 10 mL ha^{-1}, A2 = 20 mL ha^{-1}, and A3 = 30 mL ha^{-1}. The second factor, Factor B, was the type of organic fertilizer, consisting of three treatment levels as follows: B0 = without organic fertilizer, B1 = cattle manure at 15 t ha^{-1}, and B2 = chicken manure at 15 t ha^{-1}.

The combination of Factors A and B resulted in 12 treatment combinations. Each treatment combination was replicated three times, resulting in a total of 36 experimental units. The data obtained were statistically analyzed using analysis of variance (ANOVA) according to the factorial randomized block design to determine the effects of the treatments on the growth of waxy maize. When significant effects were detected, further analysis was performed using Duncan's Multiple Range Test (DMRT).

Research Procedure

The experimental field was tilled using a tractor until the soil became loose and friable. Experimental plots measuring 3 × 2 m were prepared with a spacing of 75 cm between plots. Organic fertilizers were applied at a dose of 15 t ha^{-1} and evenly incorporated into the soil. Planting holes were then prepared according to the predetermined planting distance.

Phosphate-solubilizing bacteria (PSB) were applied by pouring the bacterial

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suspension into the planting holes according to the treatment doses (A1, A2, and A3), while the control treatment (A0) received no PSB. After application, the planting holes were left for seven days before seed planting to allow bacterial adaptation and development in the soil.

Waxy maize was planted at a spacing of 60 × 40 cm, with two seeds per planting hole and a total of 25 plants per plot. Plant maintenance included watering, weeding, and pest and disease control. Observations were conducted during the vegetative phase, from one week to five weeks after planting (WAP), including growth parameters such as plant height, number of leaves, and leaf area.

RESULTS AND DISCUSSION

Plant Height (cm)

Plant height is one of the important parameters used to evaluate the vegetative growth of waxy maize. The results of plant height observations under various treatments are presented in the following table.

Table 1. Mean Plant Height of Waxy Maize in Various Treatments (cm)

Treatment	Plant Height (cm)				
	1 WAP	2 WAP	3 WAP	4 WAP	5 WAP
Factor A					
A0	12.33	29.23	50.02	89.27	120.17
A1	13.20	31.10	51.17	89.92	119.46
A2	12.93	32.28	53.72	100.33	134.13
A3	11.98	29.60	49.89	92.02	121.59
Factor B					
B0	12.57 ab	28.84 a	46.35 ab	79.74 a	105.38 a
B1	11.90 a	26.14 a	41.51 a	77.57 a	103.58 a
B2	13.36 b	36.67 b	65.75 b	121.35 b	162.55 b

Note: Means within the same column followed by different letters are significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level.

The analysis of variance showed that the application of phosphate-solubilizing bacteria (PSB) at different doses had no significant effect on the plant height of waxy maize across all observation periods from 1 to 5 WAP. The mean plant height across the four levels of Factor A ranged from 11.98 to 13.20 cm at 1 WAP and from 119.46 to 134.13 cm at 5 WAP. Treatment A2, corresponding to a dose of 20 mL ha^{-1}, consistently produced the

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highest plant height compared with the other treatments; however, the differences were not statistically significant. The absence of significant differences among PSB doses in affecting plant height was presumably due to the relatively narrow and low dose range of 10–30 mL ha^{-1}, resulting in bacterial populations that were not sufficiently different to induce a measurable growth response.

In addition, plant height is a character influenced simultaneously by various factors, including genetic traits, nitrogen availability, and microclimatic conditions. Therefore, the effect of phosphate-solubilizing bacteria, which primarily function to increase phosphate availability, may not have been strong enough to significantly influence plant height without support from optimal growing-medium conditions (Kalayu, 2019).

For Factor B, the type of organic fertilizer significantly affected plant height from 2 to 5 WAP. Treatment B2, namely chicken manure at 15 t ha^{-1}, consistently produced the greatest plant height and differed significantly from B0 and B1 across all observation periods. At 5 WAP, the mean plant height under B2 reached 162.55 cm, which was 54.31% higher than B0 at 105.38 cm and 56.91% higher than B1 at 103.58 cm. Meanwhile, B0 and B1 did not differ significantly. The superiority of chicken manure in treatment B2 was associated with the initial soil condition, which had low total N content of 0.19% and low K_2O content of 19.63 mg 100 g^{-1}.

Soils with low nutrient content tend to respond more positively to chicken manure application because this fertilizer contains higher nitrogen, approximately 2.0–3.5%, and has a lower C/N ratio of 8–12 compared with cattle manure, which has a C/N ratio of 15–25. These characteristics allow faster mineralization and nitrogen release (Sims & Wolf, 1994). This result is in line with Ipinmoroti et al. (2025), who showed that the use of chicken manure significantly increased maize plant height, number of leaves, and leaf area by improving soil chemical properties, particularly total N and available phosphorus. In contrast, the absence of significant differences between B0 and B1 indicates that cattle manure at 15 t ha^{-1}, with its higher C/N ratio, was not yet able to supply sufficient nutrients during the study period.

Number of Leaves

The number of leaves is an indicator of vegetative growth that is closely related to the plant's capacity for photosynthesis. The results of leaf number observations under various treatments are presented in Table 2.

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Table 2. Mean Number of Waxy Maize Leaves in Various Treatments

Treatment	Number of Leaves				
	1 WAP	2 WAP	3 WAP	4 WAP	5 WAP
Factor A					
A0	2.37	3.87	5.08	5.83	7.19
A1	2.52	3.94	5.25	6.13	7.52
A2	2.40	3.97	5.30	6.37	7.56
A3	2.32	3.76	5.03	6.02	7.10
Factor B					
B0	2.51	3.85 a	4.87 a	5.39 a	6.50 a
B1	2.27	3.51 a	4.69 a	5.57 a	6.67 a
B2	2.42	4.30 b	5.94 b	7.29 b	8.86 b

Note: Means within the same column followed by different letters are significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level

The inoculation of phosphate-solubilizing bacteria at different doses did not significantly affect the number of leaves of waxy maize throughout the observation period. Mean values under Factor A ranged from 2.32 to 2.52 leaves at 1 WAP and from 7.10 to 7.56 leaves at 5 WAP. Treatment A2, at a dose of 20 mL ha-1, showed a slight numerical advantage over the other treatments, but this tendency did not reach statistical significance.

The absence of a PSB effect on leaf number is consistent with the genetically determined nature of leaf initiation in maize. Research by Thompson et al. (2014) established that the shoot apical meristem in maize is programmed during embryogenesis to produce leaf primordia at biologically fixed intervals, a process that is largely insensitive to moderate changes in soil nutrient supply. Because PSB primarily influences P availability rather than the genetic regulation of meristem activity, its effect on leaf count was not expected to be substantial unless P deficiency was critically severe. The slight increase observed up to A2 followed by a decline at A3 nevertheless suggests the possibility of an optimum dose, which warrants investigation using a wider range in future studies.

Organic fertilizer type significantly affected the number of leaves from 2 to 5 WAP. Chicken manure (B2) consistently produced the highest leaf count throughout the observation period, reaching 8.86 leaves at 5 WAP. This was approximately 36.3% higher than B0 at 6.50 leaves and 32.8% higher than B1 at 6.67 leaves, while B0 and B1 did not differ significantly from each other.

The increase in leaf number under B2 can be understood through the interaction between nitrogen supply and meristematic activity. Chicken manure, with a C/N ratio of 8-

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12, releases mineral nitrogen into the soil solution more rapidly than cattle manure, and this nitrogen directly supports cell division and expansion at the shoot apex. Combined with the higher phosphorus availability from chicken manure, which supplies ATP and nucleic acid precursors required for primordium initiation, the conditions for accelerated leaf emergence were more fully met under B2 than under B0 or B1 (Vance et al., 2003; Sims and Wolf, 1994).

The absence of a meaningful difference between B0 and B1 suggests that cattle manure at 15 t ha-1 had not yet completed sufficient decomposition within the five-week observation window to release nutrients at levels comparable to chicken manure. Under the soil organic matter content of 2.38% and microbial population of 1.8×10^6 CFU g^{-1} recorded at the study site, the higher C/N ratio of cattle manure likely slowed decomposition to a rate that could not meaningfully elevate nutrient availability above the unfertilized control during the early vegetative period.

Leaf Area (cm^2)

Leaf area reflects the photosynthetic capacity of plants in utilizing sunlight. The results of leaf area observations under various treatments are presented in Table 3.

Table 3. Mean Leaf Area of waxy maize in Various Treatments (cm^2)

Treatment	Leaf Area				
	1 WAP	2 WAP	3 WAP	4 WAP	5 WAP
Factor A					
A0	21.66	36.30	108.65	175.78	214.29
A1	23.23	41.07	101.48	185.26	230.89
A2	22.13	43.03	118.76	214.45	278.85
A3	21.93	35.82	99.35	212.02	261.50
Factor B					
B0	29.45	33.04 a	85.67 a	150.08 a	193.48 a
B1	27.35	28.23 a	65.56 a	142.04 a	197.00 a
B2	32.16	55.89 b	169.95 b	298.52 b	348.66 b

Note: Means within the same column followed by different letters are significantly different according to Duncan's Multiple Range Test (DMRT) at the 5% significance level

The inoculation of phosphate-solubilizing bacteria had no significant effect on the leaf area of waxy maize across all observation periods. Treatment A2, at a dose of 20 mL ha^{-1}, produced the highest mean leaf area of 278.85 cm^2 at 5 WAP, although it did not differ

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statistically from the other treatments. Singh et al. (2024) reported that Pseudomonas putida significantly increased maize leaf area by improving phosphate bioavailability and activating P transporters in plant tissues. However, this effect became consistent only when soil available P was below 10 ppm. At the present study site, with Olsen P_2O_5 of 13.02 ppm, the level of P deficiency was not sufficiently critical to trigger a plant response to different PSB doses.

This finding strengthens the explanation that, under soil conditions where P is not extremely low, variations in PSB dose within a narrow range do not produce statistically significant differences in leaf area (De Zutter et al., 2021). For Factor B, the type of organic fertilizer significantly affected leaf area starting from 2 WAP. Treatment B2 produced the highest leaf area, reaching 348.66 cm^2 at 5 WAP, which was 80.2% higher than B0 at 193.48 cm^2 and 77.0% higher than B1 at 197.00 cm^2.

The substantial increase in leaf area under B2 reflects an integrated response involving mesophyll and epidermal cell expansion due to improved availability of multiple nutrients from chicken manure. Ipinmoroti et al. (2025), in a two-season study, reported that chicken manure applied at 15–20 t ha^{-1} significantly increased maize leaf area by simultaneously improving total N, available P, and soil K. Higher phosphorus availability under treatment B2 is known to specifically enhance leaf expansion through its role in the synthesis of cell membrane phospholipids (Vance et al., 2003). The lack of a substantial improvement in B1 compared with B0 further indicates that cattle manure decomposed more slowly, so the nutrients released were not yet optimal within the observation period. This is consistent with its higher C/N ratio (Sims & Wolf, 1994).

Interaction Between Phosphate-Solubilizing Bacteria and Type of Organic Fertilizer

Based on the three observed parameters, the analysis of variance showed no significant interaction between the dose of phosphate-solubilizing bacteria (Factor A) and the type of organic fertilizer (Factor B) on all vegetative growth parameters, namely plant height, number of leaves, and leaf area, across all observation periods from 1 to 5 WAP. These two factors acted independently, without mutually strengthening or weakening each other.

The absence of an interaction between PSB dose and type of organic fertilizer can be explained from the perspective of rhizosphere ecophysiology. The effectiveness of PSB strongly depends on the availability of carbon substrates for bacterial metabolism. Under the conditions of this study, the moderate soil organic matter content of 2.38% and low microbial population of 1.8×10^6 CFU g^{-1} indicate that the rhizosphere environment was not sufficiently active to optimally amplify the role of PSB, even when combined with

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organic fertilizer (Elhaissoufi et al., 2022). The growth improvement observed under B2 appeared to originate mainly from the direct release of nutrients, particularly mineral N and P, rather than from enhanced PSB activity. Therefore, the effect of B2 was likely a simple additive effect and did not result in a synergistic interaction with Factor A.

From a technical perspective, the tested PSB dose range of 0–30 mL ha^{-1} was likely too narrow to produce substantial differences in cell population within the rhizosphere. Dasila et al. (2023) emphasized that standardization of Pseudomonas sp. inoculant cell density in CFU mL^{-1} is an essential prerequisite for different doses to generate measurable field responses. This explains why dose variations of 10–30 mL ha^{-1} without CFU mL^{-1} standardization did not produce a significant interaction with the type of organic fertilizer in this study.

The absence of an interaction between PSB dose and type of organic fertilizer has important practical implications for farmers. Under conditions comparable to those of this study, there was no specific combination of Pseudomonas sp. dose and organic fertilizer type that was consistently superior. However, these findings provide opportunities for further research using standardized inoculant doses of ≥10^8 CFU mL^{-1}, fully composted organic fertilizers, and direct measurement of Pseudomonas sp. populations in the rhizosphere as mechanistic evidence (Pan et al., 2023; Romero-Perdomo et al., 2023).

CONCLUSION

Based on the results of this study, the following conclusions can be drawn:

1. Phosphate-solubilizing bacteria (PSB) applied at different doses of 0, 10, 20, and 30 mL ha^{-1} had no significant effect on plant height, number of leaves, and leaf area of waxy maize (*Zea mays var. ceratina*) throughout the observation period from 1 to 5 WAP. Nevertheless, treatment A2, corresponding to 20 mL ha^{-1}, consistently produced the highest values for all growth parameters.
2. The type of organic fertilizer significantly affected the vegetative growth of waxy maize. Chicken manure applied at 15 t ha^{-1} produced the best growth performance compared with cattle manure and the treatment without organic fertilizer.
3. There was no interaction between PSB and type of organic fertilizer, indicating that the two factors acted independently.

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