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Research Paper

Growth Promoting Effects of Endophytic Bacteria from *Vanda* sp. on Tissue-Cultured Seedlings

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ABSTRACT

Endophytic bacteria live and associate with plant tissues during their life in nature without causing disease and have the potential as plant growth promoters (PGPR). However, in tissue culture techniques, the sterilization process is known to kill a number of endophytic bacteria in plants resulting from tissue culture. This study aims to explore the potential of endophytic bacteria from the epiphytic orchid Vanda sp. as a growth-stimulating agent in tissue-cultured orchid seedlings. The method used in this study is through a series of experiments including the process of isolation, characterization, and testing the potential of endophytic bacteria as Plant Growth Promoting (PGP) on the growth of roots and new leaves and amylum content in the root tissue of plants resulting from tissue culture. A total of 29 isolates of endophytic bacteria were successfully isolated. All isolates showed potential as PGPR through IAA production tests, dissolution of phosphate, and nitrogen fixation. Isolate EA4(5) was selected as a superior isolate with the highest IAA production (15.16 µg/mL), high phosphate solubilization index (1.143 cm) and able to grow on nitrogen-free media. Inoculation of selected isolates on orchid Dendrobium sylvanum seedlings from tissue culture showed that soaking the roots for 2 hours was able to give the best results with a growth percentage of new roots by 70% and new leaves by 10%. Amylum content in the root tissue also increased in plants that had been inoculated with endophytic bacteria.

Introduction

Indonesia is known as one of the countries with an abundant wealth of orchid species. Orchids belong to the Orchidaceae family and have high economic value as ornamental plants due to their shape, color, and flower durability. In addition to their role in the horticultural industry, orchids are also utilized in international trade, tourism, traditional medicinal plants, and as raw materials for the cosmetics industry (Ayuningtyas et al., 2021). This biodiversity offers great potential for developing superior orchid varieties through the utilization of local genetic resources. However, generative orchid cultivation often faces obstacles, primarily because the seeds lack an endosperm, thus incapable of providing nutrient reserves during germination. As a result, germination is slow and highly dependent on symbiosis with mycorrhizal fungi. To overcome these limitations, vegetative orchid propagation through tissue culture techniques has become a commonly used method because it can produce large numbers of seedlings in a relatively short time (Andriani & Heriansyah, 2021).

Tissue culture techniques require strict aseptic conditions to avoid microbial contamination. However, the sterilization process carried out during tissue culture has the potential to eliminate endophytic microorganisms, which actually play a crucial role in supporting seedling growth and survival in the external environment (Dita et al., 2014). Endophytic bacteria are a group of bacteria that can live symbiotically within plant tissue without causing disease symptoms and are known to produce secondary metabolites that play a role in plant physiology (Linggi et al., 2023). The presence of these bacteria not only helps plants absorb nutrients but also increases tolerance to environmental stresses through the production of phytohormones such as IAA, gibberellin, and abscisic acid, as well as the ability to solubilize phosphate and fix nitrogen (Gaiero et al., 2013; Pan et al., 2020). As plant growth-promoting rhizobacteria (PGPR), endophytic bacteria show advantages over other biological agents because their colonization in plant tissue provides higher stability in responding to biotic and abiotic stresses (Resti et al., 2013).

Therefore, reintroducing appropriate endophytic bacteria into tissue cultured plants is crucial for restoring their natural symbiotic function. This study aims to isolate and characterize endophytic bacteria from Vanda sp. orchid roots and evaluate the potential of these isolates to support the growth of tissue cultured orchid seedlings through the mechanisms of IAA production, phosphate solubilization, and nitrogen fixation. This effort is expected to contribute to the development of endophytic microbial technology to improve production efficiency and orchid plant quality. Orchids (family Orchidaceae) represent one of the largest and most diverse plant families, with exceptional ecological, ornamental, and economic value. Among them, *Vanda* sp. is highly prized for its attractive flowers, long flowering period, and commercial importance in the global ornamental plant market. In many tropical countries, particularly Indonesia, *Vanda* orchids are widely cultivated and hold strong potential for export-oriented horticulture. However, large-scale production of high-quality orchid seedlings remains a major challenge due to slow growth rates and specific ecological requirements during early developmental stages.

Tissue culture has become a crucial propagation technique for orchids, enabling rapid and mass production of uniform, disease-free seedlings. This method overcomes the limitations of conventional propagation, such as low germination rates and long juvenile phases. Despite these advantages, tissue-cultured seedlings often experience physiological stress during in vitro growth and subsequent acclimatization to ex vitro conditions. One major limitation of tissue culture is the absence of beneficial microbial communities that naturally associate with plant tissues in their native environments. The strict sterilization procedures applied during in vitro culture eliminate not only pathogenic microorganisms but also beneficial endophytic microbes that play essential roles in plant growth and development. Endophytic bacteria are microorganisms that reside within plant tissues without causing disease symptoms and form mutualistic relationships with their host plants. These bacteria have been widely reported to enhance plant growth by producing phytohormones such as indole-3-acetic acid (IAA), gibberellins, and cytokinins, as well as by facilitating nutrient acquisition through phosphate solubilization and biological nitrogen fixation. In addition, endophytic bacteria can improve plant tolerance to abiotic stresses, such as drought and nutrient deficiency, and protect plants against pathogens through the production of antimicrobial compounds and competition for resources.

Their internal colonization allows for more stable and long-lasting interactions compared to rhizosphere-associated microorganisms. In natural ecosystems, orchids maintain close associations with diverse microbial communities, including fungi and bacteria, which contribute significantly to their growth and survival. For epiphytic orchids such as *Vanda* sp., these associations are particularly important due to limited nutrient availability in their growing environments. Endophytic bacteria associated with orchid roots, stems, and leaves are thought to compensate for nutrient scarcity by enhancing nutrient uptake efficiency and regulating plant hormonal balance. However, these beneficial associations are disrupted during tissue culture, leading to reduced physiological performance and lower survival rates during acclimatization. Recent studies have highlighted the potential of reintroducing beneficial endophytic bacteria into tissue-cultured plants as a sustainable strategy to improve seedling quality and establishment. The application of endophytic bacteria as plant growth-promoting agents offers an environmentally friendly alternative to chemical fertilizers and growth regulators. By restoring natural plant–microbe interactions, endophytic inoculation can enhance root development, leaf formation, and overall plant vigor, which are critical factors for successful acclimatization of tissue-cultured seedlings.

Despite increasing interest in plant growth-promoting endophytic bacteria, research focusing on orchids, particularly *Vanda* sp., remains limited. Most studies on plant growth-promoting rhizobacteria (PGPR) have concentrated on agricultural crops, while ornamental and epiphytic plants have received less attention. Moreover, the specific mechanisms by which endophytic bacteria influence the growth of tissue-cultured orchid seedlings are not yet fully understood. There is a need for systematic investigations that isolate and characterize endophytic bacteria from orchids and evaluate their growth-promoting effects under controlled conditions. Therefore, this study aims to investigate the growth-promoting effects of endophytic bacteria isolated from *Vanda* sp. on tissue-cultured seedlings. The research focuses on evaluating key plant growth-promoting traits, including phytohormone production, nutrient mobilization, and their influence on seedling growth performance. The findings of this study are expected to contribute to a deeper understanding of orchid–microbe interactions and provide a scientific basis for the application of endophytic bacteria in orchid tissue culture and sustainable ornamental plant production systems.

Orchids, particularly *Vanda* sp., are economically valuable ornamental plants whose large-scale propagation relies heavily on tissue culture techniques; however, tissue-cultured seedlings often experience suboptimal growth and low acclimatization success due to the absence of beneficial microorganisms eliminated during sterilization processes. Endophytic bacteria, which naturally inhabit plant tissues without causing disease, play a crucial role in promoting plant growth through the production of phytohormones, enhancement of nutrient availability, and improvement of stress tolerance. In natural conditions, *Vanda* orchids maintain close associations with such endophytic microbes that support their growth in nutrient-limited environments, but these symbiotic relationships are disrupted under in vitro culture. This study aims to evaluate the growth-promoting effects of endophytic bacteria from *Vanda* sp. on tissue-cultured seedlings, providing insights into their potential application in sustainable orchid propagation systems.

Method

Isolation of Endophytic Bacteria

2 cm long Vanda sp. orchid root explants were taken from various root positions, including near the rhizosphere, attached to the pot, on the surface of the media, and associated with young 'Giant Cavendish' bananas. Before sterilization, the roots were washed with running water and tween 20 for 20 minutes. Sterilization was carried out in stages with 75% alcohol (1 minute), 3% NaOCl (10 minutes), 90% alcohol (30 seconds), then rinsed with dH_2O sterile three times for 1 minute each, and drained onto sterile tissue. The samples were then grown on NA medium and incubated at 37°C for 24–48 hours. The resulting colonies were then purified (Shah et al., 2021).

Characterization Test of Endophytic Bacteria

Phosphate solubilization characteristic test, endophytic bacterial isolates were inoculated on Pikovskaya medium containing tricalcium phosphate (5 g/L) and incubated for 7 days at 28±1°C. Phosphate solubilization activity was indicated by a clear zone around the colony.Bacteria were inoculated into 5 mL of TSB with L-tryptophan (1 mg/mL, pH 7) and incubated for 48 hours at room temperature. The culture was centrifuged (13,000 rpm, 15 minutes), then 1 mL of the supernatant was mixed with Salkowski's reagent (1:2) and incubated for 30 minutes. A pink color indicates the presence of IAA. IAA concentration was measured spectrophotometrically at λ 530 nm using a standard curve (De Fretes et al., 2021).

In the nitrogen fixation test, isolates were cultivated on Jensen's medium and incubated for 5 days at room temperature. Nitrogen fixation activity was indicated by the formation of a clear zone around the colonies.Healthy, uniform orchid seedlings from tissue culture (approximately 1 month old) were inoculated using a sterile syringe into only one leaf. Treatments included selected endophytic bacteria and a pathogen as a positive control. Plants were kept in a sterile culture chamber for 7 days for visual observation.

Bacterization of Selected Endophytes on Orchid Seedlings

Four-week-old Dendrobium sylvanum orchid seedlings from tissue culture with a minimum root length of 3 cm and 2–4 leaves were used as test material. The seedlings were removed aseptically from the culture bottle, cleaned with sterile water to remove residual media, dried on sterile tissue, and sorted by size. Selected endophytic bacterial isolates were cultured in 5 mL of liquid NB media, incubated for 24 hours at 37°C with a shaker at 150 rpm, then 2 mL of the culture was diluted with sterile water to reach a concentration of approximately 10^8 CFU/mL. The seedling roots were immersed in the bacterial suspension for 30 minutes, 1 hour, and 2 hours, as well as a control without immersion; each treatment consisted of two replications with five seedlings per replication. After immersion, the seedlings were dried on sterile tissue and planted in a closed transparent plastic container filled with sterile moss, then incubated at room temperature (±25–27°C) without additional nutrients. Observations are carried out periodically based on the growth of new roots and leaves.

Starch Content Test in Orchid Root Tissue

After 21 days of cultivation, starch content was analyzed in orchid root tissue inoculated with endophytic bacteria using iodine (Lugol) staining. Roots approximately 1 cm long were taken from the elongation zone, rinsed with sterile distilled water, and sliced longitudinally into thin sections. The sections were placed on a glass slide, dripped with iodine solution, and observed under a light microscope at 400x magnification. Starch accumulation was indicated by dark blue, round granules in the root cells (Hardoim et al., 2015).

Results and Discussion

Isolation and Characterization of Endophytic Bacteria

A total of 29 endophytic bacterial isolates were successfully obtained from the roots of Vanda sp. orchids. All isolates were then analyzed to evaluate their potential as plant growth promoting agents (PGPR), through testing of indole-3-acetic acid (IAA) production, nitrogen-fixing activity, and phosphate-solubilizing ability.

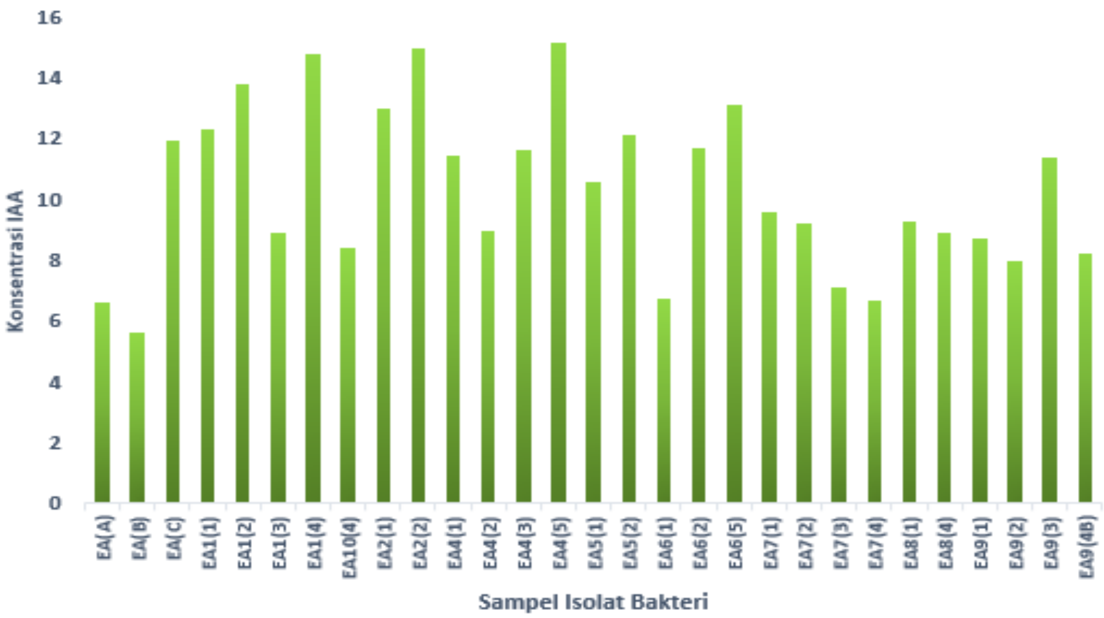


Figure 1. Graph Of IAA Concentration Produced by Endophytic Bacteria Isolates of Vanda Sp. Orchids

Of the 29 endophytic bacterial isolates of the Vanda sp. orchid obtained, all were capable of producing IAA. Based on Figure 3, the isolates with the highest IAA production were EA1(2), EA1(4), EA2(2), and EA4(5), with the highest value in EA4(5) at 15.16 μg/mL. This value was obtained through a colorimetric test using Salkowski's reagent after incubation in TSB medium supplemented with 1 mg/mL L-tryptophan. This finding is in line with De Fretes et al. (2021), who reported that endophytic isolates from orchids showed a range of IAA production between 1–28 μg/mL depending on the strain. Differences in IAA concentrations between isolates are caused by genetic diversity, especially in the IAA biosynthesis pathway, where the expression of genes such as ipdC, iaaM, and iaaH can affect IAA accumulation. In addition, growth rate also contributes to the concentration of IAA produced because greater biomass allows for higher metabolite production (Duca et al., 2014).

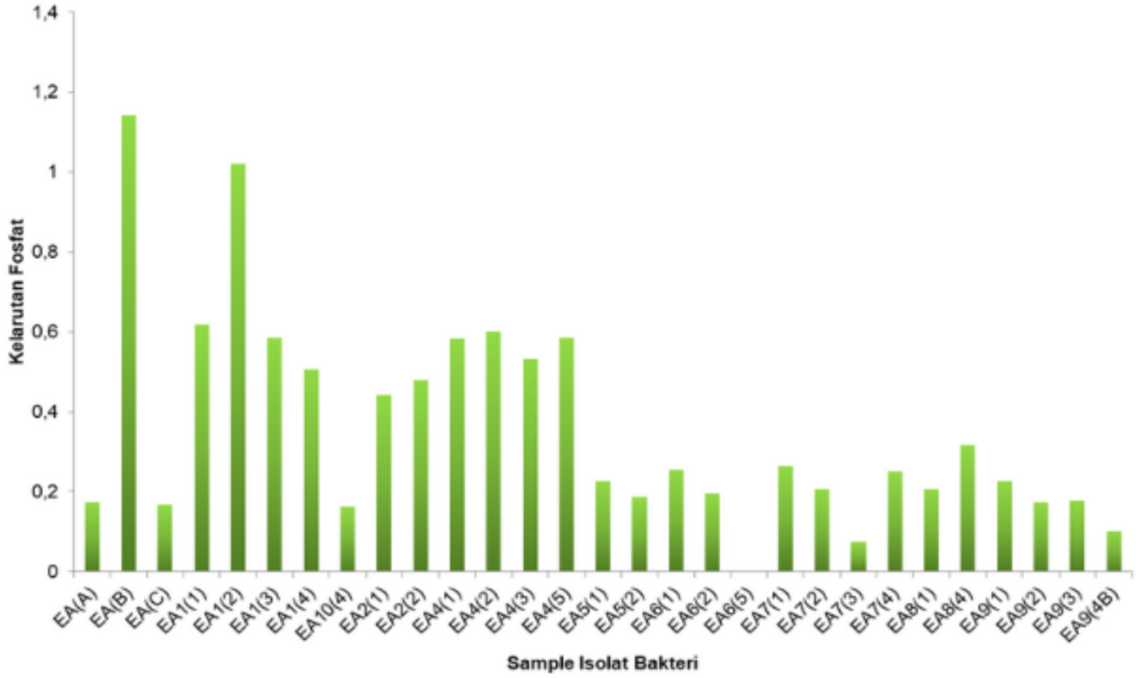


Figure 2. Phosphate Solubility Index Graph of Endophytic Bacteria Isolates of Vanda Sp. Orchids.

The results showed that of the 29 endophytic isolates of Vanda sp. orchids, almost all showed a clear zone around the colony, except for isolate EA6(5), which indicated phosphate solubilization activity. The highest solubilization index was obtained from isolate EA(B) at 1.143 cm and the lowest from EA7(3) at 0.075 cm. This variation may be caused by differences in the expression of organic acid-producing genes and the efficiency of carbon metabolism between isolates (Kalayu, 2019). This activity involves the work of phosphatase enzymes or the secretion of organic acids such as gluconate and citrate which lower pH and solubilize phosphate. Phosphorus from solubilization plays an important role in plant metabolism, including the synthesis of ATP, nucleotides, and phospholipids that support cell division and differentiation, as well as the growth of lateral roots and young leaves (Sharma et al., 2013). Therefore, isolates with a high solubilization index have the potential to be PGPR agents in increasing phosphorus availability in the root zone.

Table 1.Growth of Endophytic Bacterial Isolates Vanda Sp. on Jensen Media as an Indicator of Nitrogen Fixation Ability

Isolate Code	Growth at Jensen Media	Indications of Nitrogen Fixation
EA(A)	+	Positive N Fixation
EA(B)	+	Positive N Fixation
EA(C)	+	Positive N Fixation
EA1(1)	+	Positive N Fixation
EA1(2)	+	Positive N Fixation
EA1(3)	+	Positive N Fixation
EA1(4)	+	Positive N Fixation
EA10(4)	+	Positive N Fixation
EA2(1)	+	Positive N Fixation
EA2(2)	+	Positive N Fixation
EA4(1)	+	Positive N Fixation
EA4(2)	+	Positive N Fixation
EA4(3)	+	Positive N Fixation
EA4(5)	+	Positive N Fixation
EA5(1)	+	Positive N Fixation
EA5(2)	+	Positive N Fixation
EA6(1)	+	Positive N Fixation
EA6(2)	+	Positive N Fixation
EA6(5)	+	Positive N Fixation
EA7(1)	+	Positive N Fixation
EA7(2)	+	Positive N Fixation
EA7(3)	+	Positive N Fixation
EA7(4)	+	Positive N Fixation
EA8(1)	+	Positive N Fixation
EA8(4)	+	Positive N Fixation
EA9(1)	+	Positive N Fixation
EA9(2)	+	Positive N Fixation
EA9(3)	+	Positive N Fixation
EA9(4B)	+	Positive N Fixation

Information :
(+) : bacterial isolates can grow on nitrogen-free Jensen media

Based on Table 1, all endophytic bacterial isolates from Vanda sp. orchid roots showed growth on Jensen medium, indicating nitrogen fixation ability through nitrogenase enzyme activity. This ability classifies the isolates as diazotrophic bacteria (Mus et al., 2011), which have the potential to support nitrogen availability for host plants. Previous research by Gontijo et al. (2018) showed that diazotrophic isolates such as Herbaspirillum frisingense and Stenotrophomonas maltophilia were able to increase nitrogen accumulation by up to 68% and plant biomass by increasing dry weight by 29%. Similar findings in Rhynchostylis retusa through nifH gene and nitrogenase activity tests (Yadaf et al., 2023) further strengthen the potential of orchids as a source of nitrogen-fixing endophytic bacteria. Nitrogen is an essential macronutrient that plays a role in the synthesis of proteins, nucleic acids, and other important metabolites for plant cell growth (Bhattacharyya & Jha, 2012). Therefore, isolates from Vanda sp. orchids are considered suitable for use in the treatment of endophytic nitrogen-fixing bacteria. has potential as a plant growth promoting biological agent (PGPR), especially in tissue culture systems that are generally poor in nitrogen. The combination of nitrogen fixation, IAA production, and phosphate solubilization activities demonstrates the multifunctional character of the isolate as a natural PGPR candidate in orchid cultivation.

Plant Growth Promoting (PGP) Potential of Endophytic Bacterial Isolates

Endophytic bacterial isolates possess significant plant growth-promoting (PGP) potential due to their ability to live and multiply within plant tissues without causing disease, thereby forming stable and beneficial associations with their host plants. Unlike rhizosphere bacteria, endophytic bacteria directly colonize internal plant tissues, enabling more efficient interaction with plant physiological processes and long-term functional activity. This close association allows endophytic bacteria to influence plant growth at both cellular and systemic levels. One of the primary mechanisms underlying the PGP potential of endophytic bacterial isolates is the production of phytohormones, particularly indole-3-acetic acid (IAA). IAA plays a critical role in regulating root elongation, lateral root formation, and cell differentiation, leading to enhanced root system architecture and improved nutrient and water uptake. Many endophytic bacteria also produce other growth-regulating substances, such as gibberellins and cytokinins, which further stimulate shoot development and overall plant vigor.

In addition to hormonal regulation, endophytic bacterial isolates contribute to improved plant nutrition through phosphate solubilization and biological nitrogen fixation. Phosphate-solubilizing endophytes convert insoluble forms of phosphorus into plant-available forms, increasing nutrient uptake efficiency. Nitrogen-fixing endophytic bacteria supply plants with biologically available nitrogen, reducing dependence on external fertilizer inputs and supporting sustainable plant growth, particularly in nutrient-limited environments. Endophytic bacteria also enhance plant resilience to biotic and abiotic stresses. They can induce systemic resistance against plant pathogens through the production of antimicrobial compounds, competition for space and nutrients, and activation of plant defense responses. Furthermore, endophytic bacteria improve plant tolerance to environmental stresses such as drought, salinity, and temperature fluctuations by modulating stress-related hormones and enhancing antioxidant activity. Overall, the plant growth-promoting potential of endophytic bacterial isolates is derived from their multifaceted mechanisms, including hormone production, nutrient mobilization, stress mitigation, and stable colonization within plant tissues. These characteristics make endophytic bacteria promising biological agents for improving plant growth performance, enhancing seedling quality, and supporting sustainable agricultural and horticultural production systems, particularly in tissue-cultured plants where natural microbial associations are often absent.

Endophytic bacterial isolates exhibit strong plant growth-promoting (PGP) potential because of their multifunctional roles in enhancing plant physiological performance and developmental processes. Through their ability to colonize internal plant tissues, these bacteria establish stable symbiotic interactions that directly support plant growth by improving root and shoot development, nutrient uptake efficiency, and metabolic activity. The production of growth-regulating substances, coupled with nutrient mobilization and stress-alleviating mechanisms, enables endophytic bacteria to optimize plant growth under both controlled and natural conditions. As a result, endophytic bacterial isolates represent promising biological resources for improving plant vigor and productivity, particularly in tissue-cultured seedlings where the reintroduction of beneficial microbes is essential for successful growth and acclimatization.

Table 2. Percentage of Plants Showing New Root and Leaf Growth After Bacteriization

Treatment (Soaking Duration)	Number of plants observed	Plants with new roots	Percentage of new roots(%)	Plants with new leaves (n)	Percentage of new leaves
Control	10	2	20%	0	0%
30 minutes	10	3	33.3%	0	0%
1 hour	10	4	40%	1	10%
2 hours	10	7	70%	1	10%

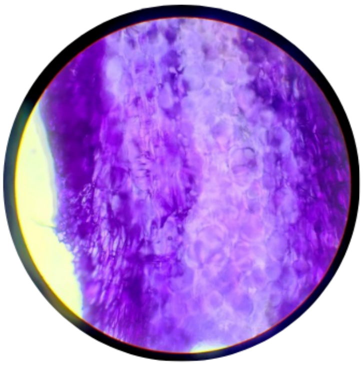
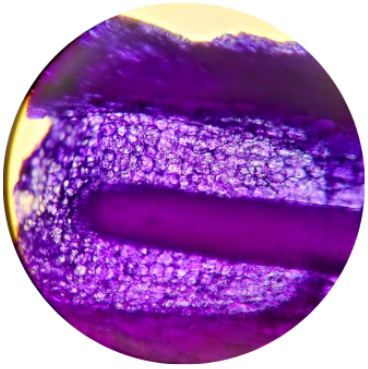
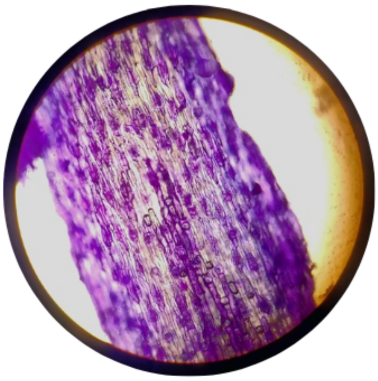
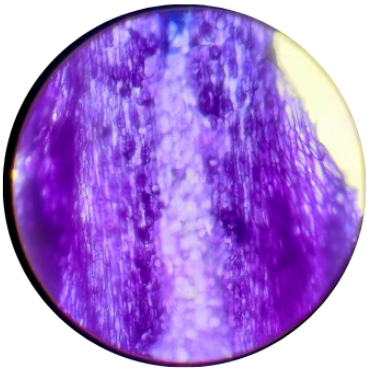
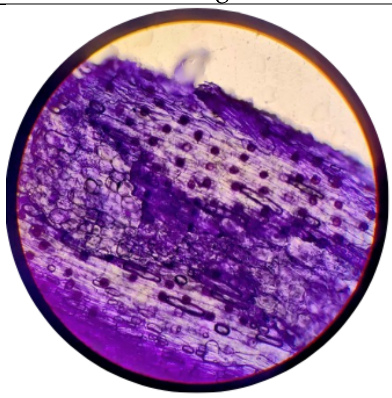
Based on Table 2, soaking tissue cultured orchid seedlings for 1–2 hours demonstrated effective endophytic bacterial colonization and optimal root growth after 21 days. This timeframe favored colonization by isolate EA4(5), which has high IAA activity, stimulating lateral root formation by up to 70%. Meanwhile, new leaf formation only reached 10%, presumably due to the longer leaf differentiation process, the need for a stable nutrient supply, and the role of cytokinin hormones. The isolate's metabolic activities, such as IAA production, nitrogen fixation, and phosphate solubilization, play a crucial role in the plant's initial adaptive response to non-aseptic environments (Suzuki et al., 2009; Mirani et al., 2017).

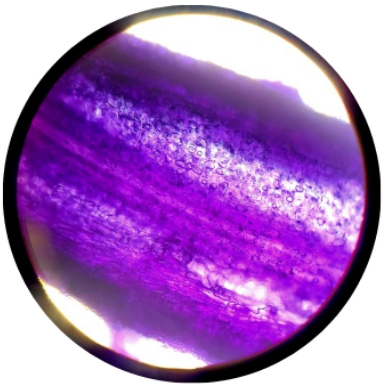
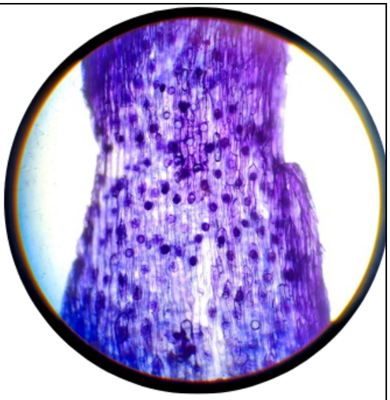
Starch Content in Root Tissue after Bacterization

Analysis of starch content in orchid roots was conducted on plants inoculated with the selected isolate EA4(5), including control and immersion treatments for 30 minutes, 1 hour, and 2 hours, using the Lugol staining method. Observations were made on days 1 and 21 post-planting to evaluate physiological changes due to bacterial colonization. The results showed higher starch accumulation on day 21, characterized by the appearance of dark purple granules in the root tissue, especially in the 1–2 hour immersion treatment. In contrast, no starch accumulation was detected on the first day, indicating that bacterial colonization and energy reserve accumulation occurred gradually.

Starch content in root tissue after bacterization reflects the physiological response of plants to endophytic bacterial inoculation and serves as an important indicator of enhanced metabolic activity. Increased starch accumulation in roots suggests improved photosynthate translocation from shoots to root tissues, which is commonly associated with enhanced nutrient uptake and root development. Endophytic bacteria stimulate this process by promoting phytohormone production, particularly auxins, which enhance root growth and sink strength. As a result, roots become more effective storage organs, accumulating higher starch reserves that support energy-demanding processes such as cell division, elongation, and adaptation during early growth and acclimatization stages.

Tabel 3. Results of Microscopic observations of the Starch Content of Orchid Roots Based on Treatment and Observation Time

Treatment (Soaking Duration)	Day 1	Day 21
Control	 No starch granules visible	 No starch granules visible
30 minutes	 No starch granules visible	 Few starch granules
1 hour	 No starch granules visible	 Medium starch granules

2 hours	 No starch granules visible	 Many starch granules, evenly distributed

Based on Table 3, there was a significant difference in starch accumulation in root tissue between days 1 and 21 after planting in each treatment. This accumulation is indicated by dark purple granules in the root tissue, reflecting increased photosynthetic activity and the efficiency of photosynthate distribution to the roots. The control treatment did not show starch accumulation, while the inoculation treatment, specifically with bacterial immersion for 1–2 hours, showed a significant increase in starch accumulation compared to the 30-minute immersion. This indicates that the duration of immersion affects the success of endophytic bacterial colonization, which in turn increases plant metabolic activity.

These findings are consistent with previous morphological observations (Table 2), which showed increased root and new leaf growth. Successfully colonizing endophytic bacteria are known to play a role in increasing nutrient uptake and photosynthates transport, particularly in the form of starch, from leaves to roots (Smith & Smith, 2015). Starch accumulation in roots reflects energy storage that supports growth and physiological adaptation, particularly during the acclimatization phase of tissue cultured plants (Suzuki et al., 2009). Therefore, starch accumulation can be used as an indirect indicator of successful bacterial colonization. Treatments without inoculation showed no physiological changes related to photosynthesis and photosynthetic product allocation, reinforcing the important role of endophytic bacteria in modulating plant physiological functions, particularly in increasing metabolism and accumulating energy-storing compounds.

Conclusion

A total of 29 endophytic bacterial isolates were successfully isolated from the roots of the epiphytic orchid Vanda sp., and all of them showed potential as plant growth promoting agents (PGPR) through their ability to produce IAA, solubilize phosphate, and fix nitrogen. Inoculation treatment using isolate EA4(5) with a 2-hour soaking proved to be the most effective in supporting the growth of tissue cultured orchid seedlings, with a significant increase in the formation of new roots and leaves. In addition, bacterial inoculation also increased the starch content in the root tissue, reflecting successful colonization and increased plant metabolic efficiency during the acclimatization phase.

References

Andriani, D. and Heriansyah, P., 2021. Identification of Contaminant Fungi in Various Natural Orchid Tissue Culture Explants (Bromheadia finlaysoniana (Lind.) Miq. Agro Bali: Agricultural Journal, 4(2):192-199.

Azevedo, J. L., Maccheroni, W., Pereira, J. O., & de Araújo, W. L. (2000). Endophytic microorganisms: A review on insect control and recent advances on tropical plants. *Electronic Journal of Biotechnology*, 3(1), 40–65. https://doi.org/10.2225/vol3-issue1-fulltext-4

Ayuningtyas, U., Budiman, B. and Azmi, TKK, 2021. The effect of foliar fertilizer on the growth of Dendrobium orchid seedlings Dian Agrihorti during the acclimatization stage. Journal of Precision Agriculture, 4(2):148-159.

Bhattacharyya, P. N., & Jha, D. K. (2012). Plant growth-promoting rhizobacteria (PGPR): Emergence in agriculture. World Journal of Microbiology and Biotechnology, 28, 1327–1350.

Compant, S., Clément, C., & Sessitsch, A. (2010). Plant growth-promoting bacteria in the rhizosphere, endosphere, and phyllosphere of plants: Their role, colonization, mechanisms involved, and prospects for utilization. *Soil Biology and Biochemistry*, 42(5), 669–678. https://doi.org/10.1016/j.soilbio.2009.11.024

Dobbelaere, S., Vanderleyden, J., & Okon, Y. (2003). Plant growth-promoting effects of diazotrophs in the rhizosphere. *Critical Reviews in Plant Sciences*, 22(2), 107–149. https://doi.org/10.1080/713610853

De Fretes, R., et al. (2021). Endophytic Bacteria from Orchid Roots as Plant Growth Promoting Rhizobacteria. Biodiversity, 22(4), 1853–1860.

Duca, D., Lorv, J., Patten, C.L., Rose, D., & Glick, B.R. (2014). Indole-3-acetic acid in plant–microbe interactions. Antonie van Leeuwenhoek, 106(1), 85–125.

Gaiero JR, Mccall CA, Thompson KA , Day NJ, Best AS, Dunfield KE. 2013. Inside the root microbiome: Bacterial root endophytes and plant growth promotion. American Journal of Botany. 100(9): 1738–1750.

Gontijo, JBA, Baldotto, GVS, Baldotto, MA, & Borges Baldotto, LE (2018). Bioprospecting and selection of growth-promoting bacteria for Cymbidium sp. orchids. Scientia Agricola, 75(5).

Egamberdieva, D., Kucharova, Z., Davranov, K., Berg, G., & Makarova, N. (2011). Bacteria able to control foot and root rot and to promote growth of cucumber. *Journal of Plant Growth Regulation*, 30(4), 434–444. https://doi.org/10.1007/s00344-011-9218-x

Glick, B. R. (2012). Plant growth-promoting bacteria: Mechanisms and applications. *Scientifica*, 2012, 963401. https://doi.org/10.6064/2012/963401

Hardoim, P.R., van Overbeek, L.S., & van Elsas, J.D. (2015). Properties of bacterial endophytes and their proposed role in plant growth. Trends in Microbiology, 23(12), 749–758.

Hardoim, P. R., van Overbeek, L. S., & van Elsas, J. D. (2008). Properties of bacterial endophytes and their proposed role in plant growth. *Trends in Microbiology*, 16(10), 463–471. https://doi.org/10.1016/j.tim.2008.07.008

Hardoim, P. R., et al. (2015). The hidden world within plants: Ecological and evolutionary considerations for defining functioning of microbial endophytes. *Microbiology and Molecular Biology Reviews*, 79(3), 293–320. https://doi.org/10.1128/MMBR.00050-14

Kandel, S. L., Joubert, P. M., & Doty, S. L. (2017). Bacterial endophyte colonization and distribution within plants. *Microorganisms*, 5(4), 77. https://doi.org/10.3390/microorganisms5040077

Lugtenberg, B., & Kamilova, F. (2009). Plant-growth-promoting rhizobacteria. *Annual Review of Microbiology*, 63, 541–556. https://doi.org/10.1146/annurev.micro.62.081307.162918

Linggi, MIA, Joko, T. and Widada, J., 2023. Potential and Diversity of Endophytic Bacteria as Growth-Promoting and Biocontrol Agents for Orchids. Indonesian Journal of Agricultural Sciences, (00): 675-682.

M. Dita, A. Schilly, J. Vargas, N. Chaves, M. Guzmán, J. Sandoval, and C. Staver. 2014. Endophyte Microbiome of Banana Roots Reveals High Diversity and Potential for Agricultural Uses. Tropentag 2014 Int. Conf. Res. Food Secur. Nat. Resort. Manag. Rural Dev. N1 PF

Mirani, AA, AbulSoad, AA, & Markhand, G.S. (2017). In vitro rooting of Dendrobium nobile orchid: Multiple responses to auxin combinations. Notulae Scientia Biologicae, 9(1), 84–88.-

Mus, F., Crook, M.B., Garcia, K., et al. (2011). Symbiotic Nitrogen Fixation and the Challenges to Its Extension to Nonlegumes. Applied and Environmental Microbiology, 77(15), 5649–5656.

Resti ZT, Habazar D, Prima P, Nasrun. 2013. Screening and identification of endophytic bacterial isolates to control bacterial leaf blight disease in shallots. Journal of Tropical Plant Pests and Diseases. 13(2): 167−178.

Shah, K. Chand, B. Rekadwad, et al. 2021. A prospectus of plant growth promoting endophytic bacterium from orchid (Vanda cristata). BMC Biotechnol 21(1), 1

Sharma, SB, Sayyed, RZ, Trivedi, MH, & Gobi, TA 2013. Phosphate solubilizing microbes: Sustainable approach for managing phosphorus deficiency in agricultural soils. SpringerPlus, 2, 587

Smith, S. E., & Smith, F. A. (2015). Roles of arbuscular mycorrhizas in plant nutrition and growth: new paradigms from cellular to ecosystem scales. Annual Review of Plant Biology, 66, 227–250.

Suzuki, S., He, Y., Oyaizu, H. (2009). Indole-3-acetic acid production in bacterial strains from the rhizosphere of Arabidopsis thaliana. Microbes and Environments, 18(3), 119–123

Yadav, A., Boruah, J.L.H., Geed, S.R., & Saikia, R. (2023). Occurrence, identification and characterization of diazotrophic bacteria from aerial roots of Rhynchostylis retusa (L.) Blume for plant growth-promoting activity. Archives of Microbiology, 205, 131.