Effectiveness Of Rhizosphere Bacteria Microcapsules On The Growth Of Arabica Coffee Leaves (*Coffea Arabica* L.)

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INTRODUCTION

As one of the largest coffee producing countries in the world after Colombia, Brazil and Vietnam, coffee production in Indonesia reaches 600,000 tons per year, where more than 80% is produced from plantations managed by the community. Coffee ranks fourth after wood, rubber and palm oil as a foreign exchange earner from high agricultural commodity exports in the Indonesian economy. Coffee can be used as a raw material for the food, beverage and cosmetic industries (Pertiwi and Ardian, 2016).

Coffee is one of the sources of foreign exchange for Indonesia and plays an important role in the development of the plantation industry. In the span of 20 years, the area and production of coffee plantations in Indonesia, especially people's coffee plantations, have experienced very significant development. In 1980, the area and production of people's coffee plantations were each amounting to 663 thousand hectares and 276 thousand tons, and in 2009 there was an increase in the area and production of 1,241 million hectares and 676 thousand tons respectively (Directorate General of Plantations, 2010).

According to Najiyati and Danarti (1997), there are three types of coffee groups known in Indonesia (namely Arabica coffee, Robusta coffee and Liberica coffee . The coffee groups known to have economic value and are traded commercially are Arabica and Robusta coffee. Robusta coffee (Coffea canephora pierre) is currently the type of coffee that dominates coffee plantations in Indonesia because it has important factors that other types of coffee do not have. These factors include resistant to leaf rust disease, its production is higher than other types of coffee and the price of robusta coffee is not much different from Arabica coffee on the market (Yaninta Ginting et al., 2024)

Nutrients are very important for coffee plants. However, acidic conditions with a pH below 5.5 cause nutrients such as P, K, N, Ca, S, Mg to be unavailable in the soil. (Wasito et al., 2023). Phosphate (P) is one of the important nutrients needed in large quantities for the growth and development of Arabica coffee plants (Coffea arabica L.). However, the availability of phosphate in the soil is often limited (trapped) by the metal nutrient Iron (Fe) due to the acidic pH which causes Phosphate (P) not to be fully absorbed by plants (Supriadi et al., 2018)

Evaluation of soil fertility and quality can be measured using microbes as natural bioindicators. Microbes that have the highest quantity in the soil are bacteria (Suharjono & Yuliatin, 2022), so the presence of bacteria can be used as a bioindicator of soil health. Rhizosphere bacteria that are around plant roots contribute significantly to increasing soil fertility and plant productivity. The use of rhizosphere bacteria can be used as an alternative to increase the number of soil bacterial colonies so that the mineralization and decomposition processes of soil organic matter are maximized and plants can be optimally nourished, known as PGPR (Moncada et et al., 2021).

The association of PGPR bacteria in plants can produce phytohormones and

secondary metabolites needed for plant growth and protection. The phytohormones produced are Indole Acetic Acid (IAA) by rhizosphere bacteria is found in almost all types of plants (Suharjono & Yuliatin, 2022). The estimated rhizosphere bacteria produce 80% of IAA, almost 98% of which comes from the PGPR strain (Arruda et al., 2013). The presence of PGPR in plant roots causes PGPR to live in colonies near or on the surface of plant roots (Youssef , 2016). With this potential, PGPR bacteria can be developed as a solution to overcome the decline in coffee productivity in UBF (Sajar et al., 2024).

RESEARCH METHODS

This study was conducted using the factorial CRD (Complete Randomized Design) method consisting of 2 factors, 16 treatments, 2 replications. The treatment of seed immersion time with the addition of endophytic bacterial suspension consisted of: B0: without immersion, B1: 6.5 hours; B1: 7.5 hours; B3: 8.5 hours. The addition of endophytic bacterial microcapsules consisted of I0: 0 grams; I1: 5 grams; I2: 10 grams; I3: 15 grams. The data obtained were analyzed using analysis of variance. The results of the analysis of variance were continued with Duncan's multiple range test.

Isolation Bacteria Rhizosphere

Soil taken from area rhizosphere plant around the slopes of Mount Sinabung.. Soil taken on depth 5-10 cm then put in bottle has been sterilized and brought to laboratory. As many as 1 gram sample soil entered into the 9 ml solution NaCl 0.85%. Sample vortex during 10 minutes furthermore dilution is carried out glow until dilution 10-6. Each as much as $100 \,\mu$ l the diluted suspension is grown with Pour Plate Method using NA media. Bacterial cultivation was carried out in duplicate. The results of the pour plate was incubated for 24 hours.

Preparation and Sterilization of Planting Media

The planting media uses topsoil, rice husks and chicken manure with ratio 50%:25%:25%. All media are mixed and put into polybags. Polybags are taped first before being put into the autoclave, then sterilized for 10 hours. The sterilized planting media is immediately taken to a greenhouse that has been sterilized by spraying 0.4% formalin.

Seed Immersion with Endophytic Bacteria Suspension

Endophytic bacteria solution was done by adding 10 ml of 0.9% NaCl solution in 1 petri dish, stirred using a triangular stirring rod. Coffee beans were soaked for 6.5 hours, 7.5 hours and 8.5 hours in a container lined with aluminum foil to keep it sterile.

Microcapsules Producing From Rhizosphere Bacteria As Biofertilizer

A total of 14.7 g of CaCl2 was dissolved in a measuring flask with 1000 ml of distilled water and stirred homogeneously. Sterilize the solution using an autoclave at 121°C for 15 minutes. Insert a sterile alginate solution containing endophytic bacterial suspension into the needle and insert it into a 0.1M CaCl2 solution. The formed microcapsules were left for 1 hour. To remove CaCl2 residues, the microcapsules were filtered and rinsed with distilled water (Panichikkal et al., 2021).

Observation Parameters

The parameters observed in this study were the number of leaves (strands) and leaf area (cm²). Observation data were taken once a month.

RESULTS AND DISCUSSION

Total Leaves (Strands)

Observation of the number of leaves was carried out at the age of 2, 3, 4, 5, months after planting (MAP). Based on the results of observations and analysis of variance, it is known that the treatment of immersion with endophytic bacterial suspension on total leaves of coffee plants (Coffea arabica L.) did not have a significant effect on total leaves (strands),

In the treatment, there was a significant effect on the treatment of endophytic bacterial microcapsules on. The effect of interaction between variations in the immersion of endophytic bacterial suspensions and the provision of endophytic bacterial microcapsules had a significant effect on 3 MAP and 4 MAP. The results of the Duncan distance test are shown in following table.

Treatment	Average Total Leaves (strands)			
Immersion Treatment (B)	2 nd MAP	3 rd MAP	4 th MAP	5 th MAP
B0 = 0 Hours	2.75 ^{dD}	5.50 °C	6.50 ^{dD}	7.75 ^{a A}

Table 1. Total leaves of Coffea arabica L. by microcapsules addition

B1 = 6.5 Hours	4.00 ^{aA}	6.50 bcBC	11.00 ^{a A}	12.88 ^{a A}
B2 = 7.5 Hours	4.13 abAB	7.50 abAB	9.13 ^{abA}	10.50 ^{a A}
B3 = 8.5 Hours	5.25 ^{bAB}	8.50 ^{a A}	10.25 bcB	8.75 ^{a A}
Microcapsule Addition (I)				
I0 = 0 grams	4.25 ^{a A}	7.00 ^{a A}	8.50 ^{a A}	9.00 ^{a A}
I1 = 10 grams	4.38 ^{a A}	7.25 ^{a A}	9.38 ^{a A}	9.75 ^{abA}
I2 = 15 grams	3.50 ^{a A}	6.75 ^{a A}	9.50 ^{a A}	10.00 ^{abA}
13 = 20 gr	4.00 ^{a A}	7.00 ^{a A}	9.50 ^{a A}	11.13 ^{a A}

Leaf Area (cm²)

Leaf area was observed at 3 months after planting (MAP). Based on the results of observations and variance analysis, it is known that the treatment of soaking coffee leaf area (Coffea arabica L.) gave a significantly different effect on leaf area at 5 MAP and gave a significantly different effect on the microcapsule addition treatment at 5 MAP. The results of the Duncan distance test are shown in following table.

Table 2. Leaf area of *Coffea arabica* L. by microcapsules addition

<i>50 0</i>	1
Treatment	Leaf Area
B0I0	18.32 ^{abA}
B0I1	20.28 ^{a A}
B0I2	21.19 ^{bcB}
B0I3	20.52 hG
B1I0	19.65 ^{hG}
B1I1	19.61 ^G
B1I2	22.66 gF
B1I3	24.52 ^{fgEF}
B2I0	25.35 efDEF
B2I1	23.03 deCDE
B2I2	22.10 deCDE
B2I3	21 cdeBCDE
B3I0	20.09 cdeBCD
B3I1	24.67 bcdeBCD

B3I2	26.21 bcdBCD
B3I3	24.79 bcBC
B0I0	18.32 ^{a A}
B0I1	20.28 ^{bB}
B0I2	21.19 ^{a A}
B0I3	20.52 ^{a A}
B1I0	19.65 ^{a A}
B1I1	19.61 ^{aA}
B1I2	22.66 ^{dD}
B1I3	24.52 ^{aAbBcC}
B2I0	25.35 bcB

DISCUSSION

Total Leaves (Strands)

Observations on the number of leaves show the total growth of coffee plant leaves (Coffea arabica L.) in the immersion treatment had the highest data at 5 MAP in the B1 treatment (12.88 strands) and the lowest in the total leaves of the coffee plant in the B0 treatment (7.75 strands). The treatment of adding microcapsules had the highest data at 5 MAP in the I3 treatment (11.13 strands) and the lowest in the I0 treatment (9.00 strands). These results are better compared to other studies using organic fertilizer with green tea liquid waste of 3.92 strands and 3.23 strands. strands without treatment (Muningsih & Ciptadi, 2019). Another study used coffee fluff and Azotobacter sp. compost. the total leaf yield was 3.10 strands in the F1 treatment (70% topsoil + 30% tea fluff compost) and at 3 ml Azotobacter sp. the total leaf yield was 2.47 strands at 16 MST (Dewi & Wulansari, 2023). In previous research (Afiati & Purnamasari, 2019), the number of leaves of purple egg plants aged 28 days after plant showed that rhizospher bacteria with a dose of 40 ml were higher than rhizosphere with a smaller dose. The increase in the number of leaves is greatly influenced by the nitrogen nutrient element which plays a role in the composition of chlorophyll and cell turgidity as well as the increase in the number of leaves (Lubis et al., 2020)

Leaf Area (cm2)

Observations of the leaf area of tea plants at the age of 90 days after plant show the leaf area of coffee (Coffea arabica L.) in the immersion treatment had the highest leaf area at the age of 5 BST in the B3I3 treatment (25.35 cm2) and the lowest leaf area in the B1I3 treatment (11.74 cm2). The results of the microcapsule addition treatment , the highest coffee leaf area at the age of 5 MAP was in the I3 treatment (20.74 cm2) and the lowest in the I0 treatment (16.75 cm2). Research (Afiati & Purnamasari, 2019) on purple egg plants with the addition of 40 ml of rhizosphere suspension had a higher leaf area at all observation ages. It could be because the provision of rhizosphere bacteria will ensure that nutrient needs are met, such as N, especially in the leaves. Nitrogen is a primary macro element which is the main component of various compounds in plant tissue. Plants that grow must contain nitrogen in the formation of new cells. Research (Tangapo, 2020) reported that endophytic bacteria can increase plant growth by providing nutrients for plants such as nitrogen, phosphate and other minerals and producing growth hormones such as ethylene , auxin and cytokinin. (Setiawan et al., 2023).

Among the treatments tested, inoculation with a mixture of BF4 (Bacillus atrophaeus RC36, Paenibacillus polymyxa 28/3, Pseudomonas fluorescens 51/2) and BF6 (Bacillus subtilis 39/3, Bacillus subtilis RC63, Pseudomonas fluorescens 53/6) bioformulations increased the leaf area of coffee plants, chlorophyll and anthocyanin (ACI) content of coffee plants significantly different compared to the control (Cakmakci et et al., 2018).

CONCLUSION

- a. Rhizosphere bacteria from the soil eruption of Mount Sinabung were effective in stimulating the growth of arabica coffee leaf (*Coffea arabica* L.)
- b. Observation of the total leaves in treatment I3 (11.13 strands). For the leaf area parameter, the highest number was in treatment B3I2 (26.21 cm²).
- c. Rhizosphere bacteria as a biofertilizer was very effective in maintaining and increasing the potential of rhizosphere bacteria when applied to plants.

REFERENCE

Afiati, I., & Purnamasari, R.T. (2019). Pengaruh pemberian bakteri endofit terhadap

pertumbuhan dan hasil tanaman terung ungu (Solanum melongena L.). J. Agroteknologi Merdeka Pasuruan, 3(1), 32–37.

- Aizar, A., & Parlina, I. (2017). Coffee Root Endophytic Bacteria and Their Potential as Controlling Agents for White Root Disease *Rigidoporus microporus*. Bioleuser Journal,1(2),54–62. <u>http://jurnal.unsyiah.ac.id/bioleuser/article/view/9073/7150</u>
- Arruda, L., Beneduzi, A., Martins, A., Lisboa, B., Lopes, C., Bertolo, F., Passaglia, L., & Vargas, L. (2013). Screening of rhizobacteria isolated from maize (*Zea mays* L.) in Rio Grande do Sul State (South Brazil) and analysis of their potential to improve plant growth. *Applied Soil Ecology*, 63, 15–22. https://doi.org/10.1016/j.apsoil.2012.09.001
- Amutha.R, Karunakaran.S, Dhanasekaran.S, Hemalatha.K, Monika.R, Shanmugapriya. P, Sornalatha.T, 2014. "Isolation And Mass Production Of Biofertilizer (Azotobacter And Phosphobacter) ", *International Journal of Latest Research in Science and Technology*. Vol. 3, Issue 1, pp 79-81,
- Cakmakci, R., Atasever, A., Erat, M., Erturk, Y., Haliloglu, K., Haznedar, A., Kotan, R., Sekban, R., Turkyilmaz, K., & Varmazyari, A. (2018). Effect of Bacteria-Based Formulation on Growth, Yield, and Enzyme Activity of Tea (*Camellia sinensis* L.). Annals of Warsaw University of Life Sciences -SGGW -Horticulture and Landscape Architecture, 38, 5–18. <u>https://doi.org/10.22630/ahla.2017.38.1</u>
- Dewi, C., & Wulansari, R. (2023). Pengaruh Aplikasi Kompos Tea Fluffdan *Azotobacter* sp. Terhadap Sifat Fisik Tanah dan Pertumbuhan Bibit Pada Persemaian Teh. Jurnal Tanah Dan Sumberdaya Lahan, 10(1), 135–142. https://doi.org/10.21776/ub.jtsl.2023.010.1.15
- Ditjenbun. 2015-2017. Percepatan perluasan dan peremajaan tanaman kopi. http://www.ditjenbun.pertanian.go.i d. Diaskes tanggal 24 Januari 2024.
- Panichikkal, J., Prathap, G., Nair, R.A., & Krishnankutty, R.E. (2021). Evaluation of the Performance of Probiotic Plants Pseudomonas sp. Packaged In Alginate Plus Salicylic Acid And Zinc Oxide Nanoparticles. International Journal of Macromolecular Biology, 166, 138–143. https://doi.org/10.1016/j.ijbiomac.2020.10.110
- Pertiwi, I. dan Ardian. 2013. Pemberian pupuk vermikompos pada bibit kopi arabika. Departement of Agrotecnology, Faculty of Agricultur, Riuan University. 8:1-2.
- Liao, N., Luo, B., Gao, J., Li, X., Zhao, Z., Zhang, Y., Ni, Y., & Tian, F. (2019). Oligosaccharides as co-encapsulating agents: effect on oral *Lactobacillus fermentumsurvival* in a simulated gastrointestinal tract. Biotechnology Letters, 41(2), 263–272. https://doi.org/10.1007/s10529-018-02634-6

Lubis, N., Agustiono, J., & Gilang Pradana, T. (2020). Effect of Red Dragon Fruit Peels

(*Hylocereus polyrhizus*) as a Natural Dye and Preservatives on Chicken Nuggets. International Journal of Research and Review (Ijrrjournal.Com), 7, 3.

- Muningsih, R., & Ciptadi, G. (2019). Analisis Kandungan Unsur Hara Limbah Cair Teh Hijau Sebagai Bahan Pupuk Organik Pada Bibit Teh. Mediagro, 14(01), 25–32. https://doi.org/10.31942/md.v14i01.2615
- Rahma Yenny. (2016). Isolasi dan Identifikasi Bakteri Endofit Lahan Kopi Arabika yang Terserang Nematoda *Radopholus similis*. Fakultas Keguruan dan Ilmu Pendidikan : Universitas Jember. Tesis
- Sajar, S., Setiawan, A., & Anzani, A. T. (2024). Analysis Of Liquid Organic Fertilizer Azolla SP And Chicken Manure On the Growth and Yield of Shallot Plants. Social Sciences and Technology (ICESST), 3(1), 133–143. https://doi.org/10.55606/icesst.v3i1.415.
- Setiawan, A., Sajar, S., & Proyogo, I. (2023). The effect of fertilization with various organic material compositions on the growth and yield of string bean plants (Vigna Sinensis L.) using a sustainable agricultural system. *AGRIVET*, *11*(2), 146–150.
- Suharjono, & Yuliatin, E. (2022). Bacteria communities of coffee plant rhizosphere and their potency as plant growth promoting. *Biodiversitas Journal of Biological Diversity*, *23*, 5822–5834. <u>https://doi.org/10.13057/biodiv/d231136</u>
- Stella, M., Theeba, M., & Illani, Z. I. (2019). Organic fertilizer amended with immobilized bacterial cells for extended shelf-life. Biocatalysis and Agricultural Biotechnology, 20 (September 2018), 101248. <u>https://doi.org/10.1016/j.bcab.2019.101248</u>
- Silitonga, D.M., Priyani, N., & Nurwahyuni, I. (2017). Isolasi Dan Uji Potensi Isolat Bakteri Pelarut Fosfat Dan Bakteri Penghasil Hormon IAA (*Indole Acetic Acid*) Terhadap Pertumbuhan Kedelai (*Glycine max* L.) Pada Tanah Kuning. Agrobiogen, 3(2), 66–72.
- Tangapo, AM (2020). Potensi Bakteri Endofit Ubi Jalar (*Ipomoea batatas* L.) dalam Menghasilkan Hormon Indole Acetic Acid (IAA) dengan Penambahan L-triptofan.
- Najiyati, S. dan Danarti, 1997. Budidaya Kopi dan Pengolahan Pasca Panen. Jakarta : Penebar Swadaya.
- Supriadi, H., Ferry, Y., & Ibrahim, M. S. D. (2018). Teknologi Budidaya Kopi. IAARD Press.
- Warsito, K., Asmaq, N., Irawan, I., Purba, N. S., & Heinze, J. (2023). Potential of Utilizing Arabika Coffee Dregs (*Coffea arabica* L.) as Biochar for Increasing Fertility of Plant Media. *The International Conference on Education, Social Sciences and Technology (ICESST)*, 2(2), 359–367.

https://doi.org/10.55606/icesst.v2i2.332

- Yaninta Ginting, T., Warsito, K., & Sari Br Siregar, W. (2024). PESTISIDA NABATI EKSTRAK DAUN MAHONI DAN SIRSAK UNTUK PENGENDALIAN HAMA Spodoptera exigua (Lepidoptera: Noctuidiae) PADA TANAMAN BAWANG MERAH (Allium ascalonicum L.). Penerbit Tahta Media. <u>https://tahtamedia.co.id/index.php/issj/article/view/1016</u>
- Youssef, M. (2016). Impact of Biofertilizers on Growth and Yield of Moringa oleifera Lam. Plants. 26, 127–138.
- Yuliatin, E., Ardyati, T., & Suharjono, S. (2019). Effect of Soil Physicochemical Properties on PGPR Density at A Coffee Plantation in Malang, Indonesia. *IOP Conference Series: Earth and Environmental Science*, 391, 012071. https://doi.org/10.1088/1755-1315/391/1/012071