# Application of Microgranule from IAA Producing-Endophytic Bacterial as Biofetilizer on Rubber Plants (Hevea brasiliences Muell. Arg.)

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#### Abstract

Rubber plant (<u>Hevea brasiliensis</u> Muell.Arg) is the main commodity of plantations, some of whose products are exported and some to meet domestic needs as raw materials for industrial purposes. Purpose on research diversity and crateristics of endophytic bacteria producing hormone IAA from rubber plants. Then the effectiveness treatment of microgranules addition and immersion of endophite bacterial suspension and rubber seed soaking suspension to spur the growth of rubber plants. This research was conducted using the factorial CRD (Completely Randomized Design) method consisting of 16 treatments with 2 replications. The first factor was immersion of endophite bacterial suspension consisting of R0 = 0 hours; R1 = 12 hours; R2 = 24 hours and R3 = 36 hours and the second factor was the addition of microgranules consisting of J0 = 0 gr; J1 = 10 gr; J2 = 15gr; J3 = 20 gr. The results of this study showed that the results on the isolation of endophite bacteria from the tea plant from the roots and stems of the tea plant yielded 3 isolates from plant roots and 2 isolates from plant stems. For IAA levels, the five isolates were positive. The use of endophytic bacteria as an alternative biofertilizer has a significant effect on the parameters observed. **Keywords:** Endophyte Bacteria, IAA, Rubber



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## INTRODUCTION

Rubber plant (*Hevea brasiliensis* Muell.Arg) is the main commodity of plantation plantations, some of whose products are exported and some to meet domestic needs as raw materials for industrial purposes (Supriadi, 2020) Technically, the cultivation of rubber plants by farmers in Indonesia has been using inorganic fertilizers to get good growth and production results. However, the continuous use of inorganic fertilizers can reduce soil fertility levels and also reduce biodiversity (Raksun et al., 2019). (Susila, 2017) argues that the solution to replacing inorganic fertilizers is the use of organic fertilizers.

In accordance with (Subekti, 2015) the advantages of organic fertilizers can increase farmers income, environmentally friendly, increase soil fertility by repairing physical damage to the soil due to excessive inorganic fertilizers. Research (Saefudin,

2017) also shows that the application of organic fertilizers has an effect on increasing the growth of rubber plants.

For now there is still little research that uses endophytic bacteria as organic fertilizer, according to the statement (Lestari et al., 2015) almost every part of the plant found the presence of endophytic bacteria, both on leaves, roots and stems. endophytic bacteria have content that can promote plant growth, as revealed (Bacon, et al., 2015) suggested that endophytic bacteria can promote plant growth by providing nutrients for plants such as nitrogen, phosphate and other minerals and producing growth hormones such as ethylene, auxin and cytokinin.

The hormone IAA is an endogenous auxin that plays a role in cell enlargement and the formation of Xylem and phloem tissue, inhibits the growth of side shoots, stimulates abscission, and affects the development and elongation of roots. In the preparation of biological fertilizer formulas, researchers try to find potential IAA hormone-producing microbes by exploring various places. IAA is a group of natural auxin phytohormones and plays a role in stimulating plant growth by increasing the process of cell elongation, cell division and differentiation in plants (Herlina et al., 2016).

Plant-bacterial associations can affect plant productivity directly and indirectly. Directly, one of them is that bacteria can act as activators or growth stimulants by synthesizing growth regulators. Endophytic bacteria may play a role in enhancing plant growth, providing nutrients, producing growth hormones and promoting plant resistance (Putri et al., 2016). It is hoped that the results of this study can be useful as a guideline for farmers who want to engage in breeding rubber plants in polybags.

#### METHOD

#### **Tools and Materials**

The tools used in this study were petri dishes, test tubes, test tube racks, measuring cups, beaker glass, Erlenmeyer, autoclave, oven, spatula, needle loops, incubator, hot plate, stir bar, analytical balance, sprayer, laminar water flow, shaker, glass bottle, aluminum foil, cotton, electron microscope, cutter knife, polybag.

While the materials used in this research were tea roots and stems, Media Nutrient Agar (NA), distilled water, alcohol 70%, chlorine solution, CaCl2, Sodium Alginate, inulin, poultry manure, top soil, rice husk charcoal, NaCl 0,9%, Crystal violet, Safranin, Acetone, alcohol, iodine, L-tryptopan, Peptone, Salkowsky.

#### **Research Methods**

This research was conducted using the factorial CRD (Completely Randomized Design) method consisting of 16 treatments with 2 replications. The first factor was immersion of endophite bacterial suspension consisting of R0 = 0 hours; R1 = 12 hours; R2 = 24 hours and R3 = 36 hours and the second factor was the addition of microgranules consisting of J0 = 0 gr; J1 = 10 gr; J2 = 15 gr; J3 = 20 gr. The application of microgranule was carried out 4 weeks after planting. The data obtained were analyzed using ANOVA. The results of the analysis of variance were continued with Duncan's multiple range test.

## **Isolation of Endophite Bacteria**

Endophite bacteria was isolated from tea roots and stems. As for the types of types. Isolation of endophite bacteria using the method (Singh et al., 2022) modified. Prior to isolation, the surface of the roots and stems of tea was sterilized.

### Measurement of IAA from Endophite Bacteria

Potential test of endophite bacteria in IAA producing used streak plate method. Isolates were inoculated on flat Nutrient Agar media supplemented with tryptophan at a concentration of 100 ppm and incubated at room temperature for 48 hours. The Salkowski reagent is dripped onto the endophite bacterial colonies. Colonies that has been dripped with Salkowski reagent was stored in a dark room for 30 minutes. A positive result is indicated by a change in the color of the colonies became red (Herlina et al., 2017).

### Preparation and Sterilization of Planting Media

The planting media used in this study were top soil that had been cleaned of weeds, broiler manure and rice husk charcoal. With a ratio (1:  $\frac{1}{2}$  :  $\frac{1}{2}$ ), top soil: 50%, broiler manure: 25% and rice husk charcoal: 25%. Sterilization was carried out at 150°C for 10 hours.

### Immersion Seeds with Suspension of Endophite Bacteria

Collection of endophite bacteria solution was carried out by adding 10 ml of NaCl 0,9% solution in 1 petri, and stirring using a triangular stir bar. Then the rubber seeds were immersion at a ratio of 12 hours, 24 hours and 36 hours in a container covered with aluminum foil to keep it sterilized.

### Producing of Microgranule From Endophite Bacteria As Biofertilizers

alginate and steril inulin containing 100 ml of endophytic bacterial suspension is included in 300 grams of tapioca starch. The addition of starch is useful as an adhesive in the manufacture of fertilizers but should not exceed 30%. The less amount of adhesive is added, the granule fertilizer is expected to be easily damaged and quickly damaged when packaged (Lisa et al., 2015). The extraction method chosen due to easy operation and cheap, provide high usability for processing, and do not experience a variety of possibilities as when using spray-drying techniques (Suryani et al., 2019) at the time of addition of endophytic bacterial suspension solution

### **Observation Parameters**

The parameters observed in this research are characteristic endophyte bacteria, IAA measurment of endophite bacteria, plant height (cm), amount of leaves (strands), leaf area (cm<sup>2</sup>).

### **RESULTS AND DISCUSSION** Isolation of Endophite Bacteria from Roots and Stems of Rubber Plants

The isolation Results of IAA-producing endophite bacteria from the roots and stems of the rubber plant (*Hevea brasiliensis* L.) can be seen in Table 1, was obtained as 5 isolates. Consists of 3 isolates from plant roots and 2 isolates from plant stems, these 5 isolates showed varying characteristics, both morphology and coloring properties.

 Table 1. Isolation of Endophite Bacteria from Roots and Stems of (Hevea brasiliensis L.)

Character of Morphology							
Isolate	Coloni				Gram		
	Color	Shape	Edge	Elevation	Shape	Setup	
AA1 BK	White	Lobate	Irreguler	Convex	Bacili	Diplobacili	+
AA2 BK	White	Irregular	Irreguler	Rised	Bacili	Diplobacili	+
AA1 AK	White	Lobate	Irreguler	Flat	Bacili	Diplobacili	+
AA2 AK	White	Filamentous	Irreguler	Flat	Bacili	Streptobacili	+
AA3 AK	White	Smooth	Smooth	Planteau	Bacili	Streptobacili	+

## The ability of root and stem endophite bacteria in tea plants to produce IAA hormones

Bacteria originating from the roots and stems of the tea plant can produce auxin hormone which is indicated by the formation of transparent pink and pink colors and the results of auxin rate in the six isolates produce positive auxin rate. The results of auxin hormone rate from root and stem endophite bacteria can be seen in table 1 below.

## Plant Height (cm)

Plant height was observed on the 1,2,3,4 and 5 week after planting (WAP). Based on the results of observations and analysis of variance, it is known that the immersion treatment affected the height growth of rubber plants (*Hevea brasiliensis* L.) had a significantly different effect on plant height (cm) at 1 WAP, 2 WAP, 3 WAP, 4 WAP and had a highly significant different effect at 5 WAP. Treatment microgranule addition at 1 WAP, 2 WAP, 3 WAP the effect was not significantly. However, 4 WAP and 5 WAP had a highly significant different. Duncan distance test results can be seen in Table 2 below.

Treatment	Average Plant Height					
Treatment	Week 1	Week 2	Week 3	Week 4	Week 5	
Immersion (R)						
R0 = 0 Jam	16,07 <sup>cB</sup>	20,31°C	26,11 <sup>cC</sup>	$34,47^{dD}$	$41,25^{dD}$	
R1 = 12 jam	16,67 <sup>cB</sup>	21,58 <sup>cC</sup>	26,90 <sup>cC</sup>	36,23 <sup>cC</sup>	44,25 <sup>bC</sup>	
R2 = 24 jam	$18,68^{abA}$	24,05 <sup>bB</sup>	$31^{bB}$	40,61 <sup>bB</sup>	$49,87^{\text{bB}}$	
R3 = 36 jam	17,98 <sup>aA</sup>	23,22 <sup>aA</sup>	28,88 <sup>aA</sup>	37,66 <sup>aA</sup>	45,02 <sup>aA</sup>	

 Table 2. The average hight of rubber plant (*Hevea brasiliensis* L.) to the immersion treatment and microgranule addition.

Microgranule A	ddition (J)				
J0 = 0 gr	17,06 <sup>aA</sup>	21,91 <sup>aA</sup>	27,23 <sup>aA</sup>	36,58 <sup>aA</sup>	43,85 <sup>cC</sup>
J1 = 10  gr	17,66 <sup>aA</sup>	22,57 <sup>aA</sup>	28,16 <sup>aA</sup>	37,02 <sup>bB</sup>	44,58 <sup>cC</sup>
J2 = 15 gr/Polibag	17, <b>32</b> ªA	22,30 <sup>abA</sup>	28,21 <sup>aA</sup>	36,95 <sup>Bb</sup>	45,03 <sup>bB</sup>
J3 = 20 gr/Polibag	17,37ªA	22,38 <sup>aA</sup>	29,28 <sup>aA</sup>	38,42 <sup>Bb</sup>	46,77ªA

The Interaction effect of immersion variation and microgranules giving highly significant effect on plant height measurement data (cm) on the growth of rubber plants (*Hevea brasiliensis* L.). Duncan distance test results can be seen in Table 3.

Treatment -	Average Plant Heigt				
	Week 3	Week 4	Week 5		
R0J0	25,15 <sup>iH</sup>	34,20 <sup>jK</sup>	$40,05^{\text{Gg}}$		
R0J1	$26,55^{\mathrm{ghFG}}$	34,20 <sup>jK</sup>	$40,40^{\text{Gg}}$		
R0J2	$26,40^{hG}$	34,35 <sup>ijHIK</sup>	$41,40^{\mathrm{fFG}}$		
R0J3	$26,35^{hG}$	$35,15^{iHI}$	$43,105^{\text{efEF}}$		
R1J0	$26,50^{hG}$	$35,65^{\mathrm{hiHI}}$	$43,75^{\text{efEF}}$		
R1J1	$27,10^{\mathrm{fghFG}}$	$36,45^{\text{ghiGH}}$	$44,10^{\text{defDEF}}$		
R1J2	$26,45^{hG}$	$35,80^{\mathrm{ghi}\mathrm{GH}}$	$43,20^{\text{efEF}}$		
R1J3	$27,55^{\text{efghEFG}}$	$37,05^{\mathrm{fgFG}}$	$45,35^{\text{cdeCDEF}}$		
R2J0	$28,45^{\text{efghDEFG}}$	$38,80^{\text{cdCD}}$	$47,75^{\text{bcdBCD}}$		
R2J1	$30,35^{bcdBCD}$	$39,30^{bcC}$	$48,1^{bcdBCD}$		
R2J2	$31,55^{abcABC}$	$40,7^{abcAB}$	$50,05^{abcABC}$		
R2J3	33,65 <sup>abAB</sup>	43,65 <sup>abAB</sup>	53,60 <sup>abAB</sup>		
R3J0	$28,85^{\text{defDEA}}$	$37,70^{\text{efEF}}$	$43,805^{\text{efEF}}$		
R3J1	$28,65^{efDE}$	$38,15^{deDE}$	$45,75^{\text{cdeCDE}}$		
R3J2	$28,45^{efghDEFG}$	$36,95^{\mathrm{ghFG}}$	$45,5^{cdeCDEF}$		
R3J3	$29,60^{\text{cdefCDEF}}$	$37,85^{eDEF}$	45,00 <sup>cdeCDEF</sup>		

Table 3. Average immersion treatment and immersion of microgranules addition to the	
plant height rubber ( <i>Hevea brasiliensis</i> L.)	

### Amount of Leaves (strands)

Observation Data on the number of plant leaves observed in the 1, 2, 3, 4, and 5 weeks after planting (WAP). Based on the observation and analysis of sidik ragam it is known that the immersion treatment to the growth of the number of leaves of rubber plants (*Hevea brasiliensis* L.) nd had no significant effect on the number of plant leaves (strands) in the observation of the 1 WAP, 2 WAP, 3 WAP, and 4 WAP. However, Week 5 data gives had a highly significant different effect. While the treatment of microgranules obtained that in the 1 WAP, 2 WAP, 3 WAP, 4 WAP, and 5 WAP have had no significant effect on the number of leaves (strands). Duncan distance test results can be seen in table 4.

Treatment –	Average Number					
Treatment –	Week 1	Week 2	Week 3	Week 4	Week 5	
Immersion Treatmen	it (R)					
R0 = 0 Jam	8,75 <sup>aA</sup>	12 <sup>aA</sup>	14,75 <sup>aA</sup>	17,87 <sup>aA</sup>	20,5 <sup>aA</sup>	
R1 = 12 jam	9 <sup>aA</sup>	12 <sup>aA</sup>	15 <sup>aA</sup>	$18^{aA}$	21 <sup>aA</sup>	
R2 = 24 jam	9 <sup>aA</sup>	$12,37^{aA}$	12 <sup>aA</sup>	14,25 <sup>Aa</sup>	21,3 <sup>bB</sup>	
R3 = 36 jam	9 <sup>aA</sup>	12 <sup>aA</sup>	$14,75^{aA}$	17,75 <sup>aA</sup>	20,75 <sup>aA</sup>	
Mikrogranule addition	on (J)					
J0 = 0 gr/Polibag	8,87 <sup>aA</sup>	12 <sup>aA</sup>	$14,75^{aA}$	17,87 <sup>aA</sup>	20,5 <sup>aA</sup>	
J1 = 10 gr/Polibag	8,87 <sup>aA</sup>	12 <sup>aA</sup>	14,87 <sup>aA</sup>	17,87 <sup>aA</sup>	20,87 <sup>aA</sup>	
J2 = 15 gr/Polibag	9 <sup>aA</sup>	12 <sup>aA</sup>	11,12 <sup>aA</sup>	13,37 <sup>aA</sup>	21,20 <sup>aA</sup>	
J3 =20 gr/Polibag	9 <sup>aA</sup>	$12,375^{aA}$	15,75 <sup>aA</sup>	18,75 <sup>aA</sup>	21,75ªA	

**Table 4.** Average amount of plant leaves rubber (*Hevea brasiliensis* L.) to the immersion treatment and administration of microgranule

The interaction effect of variations in immersion and microgranules had a significant effect on 5 WAP. Duncan's distance test results can be seen in table 5 below.

Treatment	Average
R0J0	19 <sup>cC</sup>
R0J1	21 <sup>cC</sup>
R0J2	21 <sup>cBC</sup>
R0J3	21 <sup>cBC</sup>
R1J0	21 <sup>cBC</sup>
R1J1	21 <sup>cBC</sup>
R1J2	21 <sup>cBC</sup>
R1J3	21 <sup>cBC</sup>
R2J0	21 <sup>cBC</sup>
R2J1	21 <sup>bcBC</sup>
R2J2	$22^{abcABC}$
R2J3	24 <sup>abAB</sup>
R3J0	21 <sup>cBC</sup>
R3J1	$20,5^{\mathrm{cBC}}$
R3J2	$20,5^{\text{cBC}}$
R3J3	21 <sup>cBC</sup>

 Table 5. Average immersion treatment and immersion of microgranule to the amount of plant leaves rubber (*Hevea brasiliensis* L.) at 5 WAP

Based on the observations from Table 5 it shows that the interaction of the mean amount of leaves from the immersion treatment and addition of endophitec bacterial microgranule had a significantly different effect at 5 WAP after being tested using Duncan's Range Test.

## Leaf Area

Leaf area was observed at 5 week after planting (WAP). Based on the results of observations and analysis of variance, it is known that the immersion treatment and microgranule addition of the leaf area of the rubber plant (*Hevea brasiliensis* L.) have a highly significantly different effect on the number of leaves (strands) at 5 WAP.

 Table 6. Average leaf area of plants rubber (*Hevea brasiliensis* L.) to the immersion treatment and addition of microgranule.

Treatment	Average Leaf Area
Immersion (R)	
R0 = 0 hour	48,61 <sup>cB</sup>
R1 = 12 hour	$49,48^{bcB}$
R2 = 24 hour	53,19 <sup>aAB</sup>
R3 = 36 hour	50,43 <sup>bAB</sup>
Microgranule Addition (J)	
J0 = 0  gr/Polibag	49,20 <sup>bB</sup>
J1 = 10 gr/Polibag	49,81 <sup>bB</sup>
J2 = 15 gr/Polibag	50,47 <sup>bAB</sup>
J3 = 20 gr/Polibag	52,23 <sup>aAB</sup>

The interaction effect of variations in immersion and microgranules had a significant effect on 5 WAP. Duncan's distance test results can be seen in table 7.

Treatment	Average
R0J0	$48,13^{efgDEF}$
R0J1	$47,80^{ m gF}$
R0J2	$48,82^{\text{defgCDEF}}$
R0J3	$49,68^{\text{defgBCDEF}}$
R1J0	$47,88^{ m fgEF}$
R1J1	50,36 <sup>cdeBCDEF</sup>
R1J2	50,08 <sup>cdeBCDEF</sup>
R1J3	$49,61^{\text{defgCDEF}}$
R2J0	$50,24^{\text{cdeBCDEF}}$
R2J1	$51,50^{\text{bcdeBCDEF}}$
R2J2	$52,44^{\text{abcdeABCDEF}}$
R2J3	58,59 <sup>abAB</sup>
R3J0	$50,55^{\text{cdeBCDEF}}$
R3J1	$49,61^{defgCDEF}$
R3J2	$50,55^{bcdeBCDEF}$
R3J3	$51,03^{bcdeBCDEF}$

 Table 7. Average immersion treatment and microgranule addition to plant leaf area rubber (*Hevea brasiliensis* L.) at 5 WAP.

Based on the observations from Table 7 it shows that the interaction of the mean number of leaves from the immersion treatment and the addition of endophite bacterial

microgranule had a highly significant different effect at 5 WAP after being tested using Duncan's Range Test.

### Discussion

### Isolation of Endophite Bacteria from Roots and Stems of Rubber Plants

Based on the results of this study, the isolation of root endophytic bacteria and stems of rubber plants there are 5 isolates, 2 samples of isolates from rubber stems with labels AA1 BK, AA2 BK and there are 3 samples of rubber root isolates with labels Aa1 AK, Aa2 AK, Aa3 AK. On the results of previous research that has been done (Lestari. 2017) obtained 5 potential isolates of endophytic bacteria WL01, WL02, WL03, WL04, and WL05 from the roots of rubber plants. According to (Wilson et al., 2017) IAA hormone-producing bacteria are characterized by Colony morphology, namely colony shape, elevation, edge and color, and cell morphology through bacterial coloring, namely cell shape and gram nature of bacteria. The results obtained in isolation and characterization showed that AA1 BK samples have Colony shape: lobate, elevation: convex, edge: irregular, color: white, cell shape: bacillus, cell arrangement: diplobacillus, and gram staining get positive results (+). AA2 BK sample has colony shape: irregular, elevation: risen, edge: irregular, color: white, cell shape: bacillus, cell arrangement: diplobacillus, and gram staining get positive results (+). AA1 AK samples have Colony shape: lobate, elevation: flat, edge: irregular, color: white, cell shape: bacillus, cell arrangement: diplobacillus, and gram staining get positive results (+). AA2 AK samples have Colony shape: lobate, elevation: convex, edge: irregular, color: white, cell shape: bacillus, cell arrangement: streptobacillus, and gram staining get positive results (+). AA3 AK samples have Colony shape: smooth, elevation: planteau, edge: smooth, color: white, cell shape: bacillus, cell arrangement: streptobacillus, and gram staining get positive results (+). Endophytic bacteria are classified as gram negative or positive bacteria (Pulungan & Tumangger, 2018). Gram staining that has been done on the 5 isolates consisting of stems and roots of rubber plants produce gram positive. Endophytic bacteria from one plant will produce different species from the endophytic bacteria of other plants. This is supported by the statement (Afifah et al., 2018), endophytic bacterial diversity can be influenced by nutrient availability within plant tissues. The diversity of endophytic bacteriaalso affects the secondary metabolites produced.

### The ability of root and stem endophite bacteria in tea plants to produce IAA hormones

The results obtained in this study showed that endophytic bacteria are able to spur plant growth. This is in line with the statement (Saridewi et al., 2020) suggested that endophytic bacteria are able to spur growth related to their ability to produce phytohormones, such as indole acetic acid-3 (IAA), increase phosphate availability, produce various enzymes, such as amylase, sellluase, and protease. In addition, the IAA hormone has an important role in various aspects of development and growth in plants, there fore its production by endophytic bacteria promotes plant growth (Herlina et al., 2016).

## Plant height

Based on the data obtained by Duncan testing showed that the best immersion and microgranule treatment in the last week of plant height observation is R2 treatment 24 hours with an average yield of 49.87 cm and J3 treatment 20 gr/polybag: 46.77 cm. And for the lowest plant in treatment R0 (control) with a plant height of 41,25 cm and treatment J0 (control) with a plant height of 43,85 cm. The results of this study are better compared with research conducted (Dongoran & Sularno, 2019) high rubber plant seedlings with P3 treatment of liquid organic fertilizer application of coconut water with a dose of 3.222 ml has a higher yield (44.70 cm) than other treatments at 9 MST. From these results, endophytic bacteria are able to provide effectiveness against rubber plant, this is in line with research (Munif et al., 2016) The effect of endophite bacteria on the length of the rice plant canopy showed that almost all the isolates tested were able to stimulate the growth of the rice plant canopy compared to the control. In research (Khaeruni et al., 2020) the treatment of endophite bacterial isolates significantly affected the height of the cocoa plants and was significantly different compared to the controls. Arabidopsis plants treated with B. aryabhattai showed increased growth compared to control plants. The analysis showed that bacterial inoculation significantly increased the size of the Arabidopsis plants (4.55 cm) compared to the control plants (3.10 cm). Plant growth of N. tabacum. The height of N. tabacum inoculated by plants was significantly greater (4.05 cm) than that of non-inoculated plants (2.25 cm) (Xu et al., 2022).

The result of the interaction of the two treatments showed that the treatment of R2J3 gives the best value with an average of 53.60 cm at the height of rubber plants. (Suhandono et al., 2016) interactions between plants and endophytic bacteria are mutually beneficial. Plants provide nutrients to bacteria while bacteria will protect plants from pathogens, aid in the synthesis of phytohormones, and also help increase mineral absorption. In addition, endophytic bacteria in plants are also able to spur plant growth. While the average result of the interaction of the two treatments with the lowest value obtained in the treatment R0J0: 40.05 cm.

## Amount of Leaves

The results of observing the amount of tea plant leaves showed that the growth in the amount of rubber plant leaves (*Hevea brasiliensis* L.) in the immersion treatment had the highest yield at 5 WAP in R2 treatment in 24 hours of immersion with an average amount of leaves 21.3 (strands). And for the lowest amount of rubber plant leaves in treatment R0 (control) with an average amount of leaves 20.05 (strands). And for the treatment of giving granule the highest yield was at 5 WAP in treatment J3 with 20 g/polybag with an average amount of plant leaves of 21.75 (strands) and for the lowest yield in treatment J0 (control) with an average amount of plant leaves 20.05 (strands). The results of this study are better compared with research conducted (Dongoran & Sularno, 2019) The number of leaves of rubber plant seedlings treatment time interval of giving coconut water once every 4 days with a dose of 3.222 ml has a higher result (21.20 strands) than other treatments at 9 MST. This is a comparison that the use of endophytic bacteria other than that based on research (Dwiati 2016) can be seen that the administration of auxin and cytokinin in Phalaenopsis affect the growth of roots and leaves of the plantThen the interaction of the average number of leaves at 5 WAP with

the immersion treatment with endophite bacteria and the treatment of endophite bacterial microcapsules had the highest data in the R2J3 treatments with an average number of leaves of 24 (strands).

## Leaf Area

The results of observing the leaf area of the tea plant at 5 WAP showed that the leaf area of the rubber plant (Hevea brasiliensis L.) in the immersion treatment had the highest yield at 5 WAP in the R2 treatment within 24 hours with an average leaf area of 53.19 cm<sup>2</sup>. And for the lowest leaf area in treatment R0 (control) with an average plant leaf area of 48.61 cm<sup>2</sup>. And for the treatment of microgranule addition, the highest yield of tea plant leaf area at 5 WAP was in treatment J3 with 20 g/polybag with an average plant leaf area of 52.23 cm<sup>2</sup> and for the lowest yield in treatment J0 (control) with an average plant leaf area of 49.20 cm<sup>2</sup>. The results of this study are better compared with research conducted (Dwiyanti & Jati, 2019) in the observation of the leaf area of rubber plants obtained a larger leaf area in the treatment of B4 (planting media 20% compost waste tofu dregs and 80% soil), which reached 24.49 cm<sup>2</sup> and very significantly different from the treatment of planting media B1, B2 and B3, respectively, the extent of 0.93 cm<sup>2</sup>, 4.39 cm<sup>2</sup>. In line with research (Murthi et al., 2015) states that endophite bacteria can enhance plant growth by providing nutrients for plants such as nitrogen, phosphate and other minerals as well as producing growth hormones such as ethylene, auxin and cytokinins.

Based on the observations from table 7, it shows that the interaction of the average leaf area with the immersion treatment and microgranul addition with endophite bacteria had the highest data in the R2J3 treatment with an average leaf area of 58.59 cm<sup>2</sup> and the lowest data in the R0J1 treatment with an average leaf area of 47.80 cm<sup>2</sup>.

### The effectiveness of Microgranules on Rubber Plants (Hevea brasiliensis L.)

The material for making microgranules using tapioca starch, alginate, inulin, and suspension solution of endophytic bacteria, where endophytic bacteria have a role in enhancing the growth of the host plant (Santoyo et al., 2016). The study (Nasrun & Nurmansyah, 2015) using strains of Bacillus and Pseudomonas isolation results from rubber rhizosphere able to inhibit R.microporus of 72.69-90.94% using less inoculum R.microporus. according to (Sebola et al., 2019) states that several strains of bacilus have been used in agriculture as plant growth promoters and biological control agents. The results obtained from the parameters observed: plant height, number of leaves, and leaf area provide the best growth in R2 treatment with 15 gr/polybag.

## CONCLUSION

The results of this study showed that the results on the isolation of endophite bacteria from the tea plant from the roots and stems of the tea plant yielded 3 isolates from plant roots and 2 isolates from plant stems. For IAA levels, the five isolates were positive. The use of endophytic bacteria as an alternative biofertilizer has a significant effect on the parameters observed. Where found, (1) The plant height parameter showed the highest

data was in treatment R2 with an average yield of 49.87 cm and treatment J3: 46.77 cm. In the parameter of the amount of leaves the highest data was in treatment R2 with an average yield of 21.3 (strands) and treatment J3: 21.75 (strands). (2) In the leaf area parameter, the highest data was in treatment R2 with an average yield of 53.19 cm<sup>2</sup> and treatment J3: 52.23 cm<sup>2</sup>. Lowest data plant in treatment R0 (control) with a plant height of 41,25 cm and treatment J0 (control) with a plant height of 43,85 cm. (3) The amount of leaves parameter found the lowest data was in treatment R0 with an average yield of 20.05 (strands) and treatment J0: 21.75 (strands). In the parameter of leaf area the lowest data was in treatment R0 with an average yield of 48.61 cm<sup>2</sup> and treatment J0: 49.20 cm<sup>2</sup>.

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