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Potential of Bacteria from Goat Rumen in Suppressing the Growth of *Ganoderma boninense*

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Abstract:- Ganoderma boninens is a soil-borne fungus that currently threatens oil palm. This disease causes stem rot in oil palms. This fungus can cause great damage to oil palm plantations. The high infection rate and rapid spread of G. boninense in the soil make control of the disease a challenge. Goat rumen bacteria are bacteria that can act as biological agents that are expected to inhibit the growth of Ganoderma boninens..The aim of this research is to suppress the growth of the Ganoderma boninens fungus both in vitro and ex vivo. The research used a completely randomized design consisting of 1 factor with 15 levels and 2 replications. The results of the research showed that the highest percentage of inhibitory power against the growth of the fungus Ganoderma boninense was sequentially found in isolates P204, P102, P104, P206 and P106 and the level of root rot disease in oil palm plants could be reduced by the application of biological agents derived from the isolates. goat rumen.

Keywords:- *Goat Rumen ; Bakteria ; Ganoderma ;Oil Palm.*

I. INTRODUCTION

Oil palm (*Elaeis guineensis* Jacq) is a very important plant for the plantation industry. In terms of economic turnover, palm oil commodities are ranked second after rice. Cultivation and processing of palm oil results in increased income of farmers and communities, increased added value in the country, export of crude palm oil to generate foreign exchange, and employment opportunities in various other subsectors [1]

Palm oil production is increasing, while the demand for palm oil is increasing worldwide. Indonesia is the world's largest producer and exporter of palm oil. The domestic market for palm oil and palm kernel oil is still very large, although export opportunities are opening up more and more. Indonesia and CPO production has decreased over the last three years (2019-2022). 47.03 million tons were produced in 2020, that is 0.3% less than 47.18 million tons in 2019; In 2021, production will be 46.89 million tons, that is 0.31% less than 47.03 million tons in 2020; and in 2022 production will be 46.73 million tons, that is 0.34% less than 46.89 million tons in 2021 [2].

In cultivating oil palm plants, there are problems that can reduce the productivity and quality of the harvest. Stem Root Rot caused by *Ganoderma boninense* Pat is a major disease of oil palm plants [3]. In well-known palm oil producing countries in Southeast Asia, especially Indonesia and Malaysia, the disease can cause significant revenue losses. *Ganoderma boninense* infection starts from the roots and can spread through spores and root-to-root contact. The consequences of *Ganoderma boninense* Pat attacks are that the color of the leaves becomes pale, stunted growth, rot on the plant stem and shortening the life of the plant [4] .

To treat Stem Root Rot disease, appropriate techniques and environmentally friendly controls are needed, such as biological control. Biological control is a method that does not harm the environment and living creatures. To control *Ganoderma boninense* Pat biologically, use microorganisms or antagonistic bacteria. Antagonistic bacterial tests can be carried out to suppress the development of Ganoderma boninense Pat. Bacteria can act as a source of biofungicide. Biofungicides are populations of microorganisms that can inhibit and even destroy pathogens [5]

In this study, goat rumen microbes are used in disease control to suppress or suppress the fungus Ganoderma boninense Pat. This antagonistic bacteria from the goat rumen is able to inhibit the formation of pathogen spores, has the ability to increase plant immunity against pathogenic fungal infections and produces antibacterial compounds that can suppress fungal growth. There were a total of 15 isolates, with 7 isolates from liquid goat rumen and 8 isolates from solid goat rumen [6] and[7]. All isolates have been tested to function as biodecomposers, biofertilizers and biological agents. They contain bacteria that damage organic matter, substances that stimulate plant growth, substances that control pests and diseases, and necessary plant nutrients [8]. This study used 15 bacterial isolates originating from liquid goat rumen and solid goat rumen, the fifteen isolates, namely P101, P102, P103, P104, P105, P106, P107, P201, P202, P203, P204, P205, P206, P207 and P208.

II. MATERIAL AND METHODS

A. Time and Location of Research

The studies are carried out in the Agronomy Laboratory of the Plantation Training Program of the State Agricultural University of Samarinda and and at PT. Sentosa Kalimantan Jaya. This study will be conducted in September-December 2023

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B. Tools and Materials

The tools used ini this this research are laminar air flow cabinet (LAFC), Erlenmayer, Petridishes, Bunsen lamp, autoclave, cover glass, oven, micropipette, hot plate knife/cutter, glass preparation, scissors, tube needle, microscope, tweezers, camera and stationery.

The materials used in the research are namely isolates P101, P102, P103, P104, P105, P106, P107, P201, P202, P203, P204, P205, P206, P207 and P208, ganoderma fungus, potatoes, agar-agar, distilled water, tissue, spirit, dextrose, label paper, aluminum foil, plastic wrap.

C. Reasrch Design

The study was conducted using a non- factorial completely randomized design (CRD) with 15 treatments and 2 replications. The tested treatments consisted of: isolate P101, P102, P103, P104, P105, P106, P107, P201, P202, P203, P204, P205, P206, P207 and P208.

D. Research Prosedure

➢ *The Media Making & Propagation of Ganoderma boninense Pat.*

Ganoderma boninense propagation media Pat. using PDA (Potato Dextrose Agar) media. Making PDA media uses a mixture of potato extract, dextrose and agar. The potatoes were peeled and cut into pieces measuring ± 2 x 2 x 2 cm and weighed 200 g. Next, the potatoes are boiled in 500 ml of distilled water until soft using a hotplate. Next, the potato extract is cooled. Then weigh 15 g of agar and weigh 15 g of dextrose. Next, the potato extract is poured into the agar and dextrose mixture and distilled water is added until it reaches a volume of 1000 ml. The mixture of ingredients is then cooked until it boils using a hotplate. After boiling, the PDA media is put into an Erlenmayer and then covered with plastic wrapping. The media was then sterilized in an autoclave for approximately 15 minutes at a temperature of 121 ^{o}C at a pressure of 1.25 atm. Once sterile, the PDA media can be poured directly into a cup aseptically in the LAFC until it solidifies. Bacterial multiplication is carried out by scratching and incubation for 28 hours.

➢ *Isolation of Ganoderma boninense Pat.*

Ganoderma Boninese mushrooms are obtained from the Berau area, precisely at PT. Sentosa Klimantan Jaya in East Kalimantan on the affected annual crop plantations, the ganoderma fungus was taken to the laboratory for isolation. Before isolating the ganoderma fungus, it was cleaned first using 70% alcohol and then washed with distilled water. After the ganoderma fungus is clean, then cut it using a catheter to a size of around 0.5 cm, inoculate it into a cup containing new PDA media aseptically in the LAFC.

➢ *Test in Vitro the Antagonism ability of Biological Agents against Ganoderma Fungi.*

To determine the effectiveness of 5 bacterial isolates from the rumen of goats against the fungal pathogen G. boninense, an in vitro antagonism test was performed using a two-culture assay. In a 9 cm Petri dish, all bacterial isolates were collected and placed on one side of the agar medium, while the fungal pathogens were placed on the opposite side. As a negative control, G. boninense was grown alone without antagonist isolates on potato dextrose agar (PDA) medium. The culture was then incubated at 26°C for 5 and 7 days. To determine the inhibitory effect of antagonistic isolates against G. boninense, the diameter of fungal colonies growing on agar Petri dishes was measured. The percentage inhibition is calculated using the following formula:.

$$I = \frac{R1 - R2}{R1} \text{ X } 100\%$$

Where :
I = Percentage of inhibition
R1 = G. boninense colon radius in control petri dishes.
R2 = Coloni radius of G. boninense on test plate

➢ *Test the Severity of Disease in the Field*

The biological agent derived from five bacterial isolates from the goat rumen which have been multiplied in the laboratory will be tested for effectiveness in the field by diluting the biological agent in a ratio of 1: 10 liters of water. Before controlling with 5 isolates of bacteria from the rumen of goats, oil palm plants that were infected with the disease, data collection was carried out on sample plants of 16 trees. Biological agents derived from five isolates of bacteria from the rumen of goats which have been propagated in the laboratory will be tested for their effectiveness in the field by diluting the biological agents. in a ratio of 1: 10 liters of water. Before control was carried out with 5 bacterial isolates from the goat rumen of diseased oil palm plants, data were collected on 16 sample plants. then carry out control by spraying a bioactivator solution derived from 5 isolates of bacteria from goat rumen on oil palm plants resulting in 200 ml of disease caused by the Ganoderma fungus per tree and leaving it for 6 weeks before observing again to test the effectiveness of the biological agent. to the severity of the disease. The level of severity can be seen from the score as follows:

Score 1 = Three or more spear leaves do not open when the tree does not lack water
Score 2 =Three or more spear leaves do not open when the spear leaves are not short of water, accompanied by natural wilting of the lower midrib
Score 3 = Three or more spear leaves do not open when there is no shortage of water, natural wilt at the lower midrib, and Ganoderma fruit bodies are found
Score 4 = Three or more spear leaves do not open when there is no shortage of water, natural wilting of the lower midrib, Ganoderma fruit bodies are found, and all midribs have wilted except the spear leaves
Score 5 = Three or more spear leaves do not open when the spear does not lack water, natural wilting at the lower midrib, body.

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III. RESULT AND DISCUSSION

A. *Inhibitiom Test in The Laboratory*

Graphic image of the inhibitory power of fifteen types of isolates of bacteria from the goat rumen at 3 and 7 days after isolation on the growth of the fungus Ganoderma boninense can be seen in Figure 1 below.

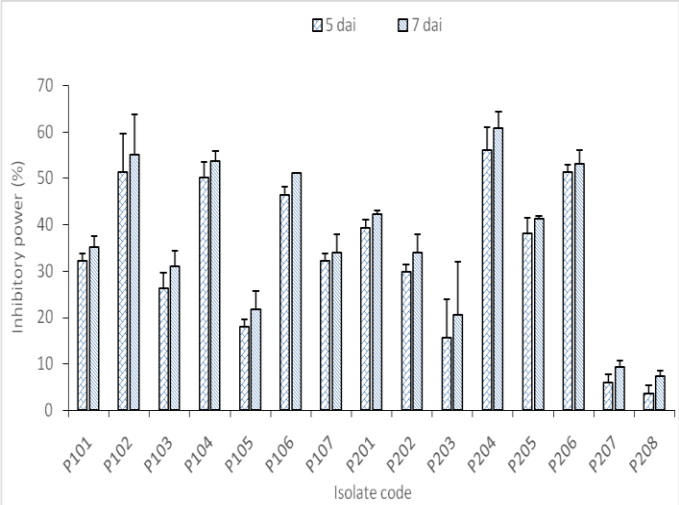

Fig 1 Graph of the inhibitory power of fifteen types of isolates of bacteria from the goat rumen at 3 and 7 days after isolation on the growth of the fungus Ganoderma boninense

Figure 1 shows that fifteen isolates originating from goat rumen were able to show different inhibitory activities on the growth of the Ganoderma boninensa fungus, including isolates P101, P102, P103, P104, P105, P106, P107, P201, P202, P203, P204, P205, P206, P207 and P208. Inhibitory activity is expressed as a percentage of growth inhibition, with a higher percentage indicating stronger inhibitory activity. All 15 isolates were able to inhibit, but the highest inhibitory activity was observed in isolates P204, P102, P104, P206 and P106. The results of this study are consistent with those of [9], who found that the five microbial isolates used could inhibit the growth of Ganoderma boninense. Suppression of plant pathogens can occur through a variety of mechanisms, including hyperparasitism or mycoparasitism, which inhibits the pathogen's growth zone. In some cases, biocontrol agents can grow faster than pathogens, as observed with goat rumen-derived biologics isolated against G. boninense.

Biologics derived from goat rumen can inhibit or inhibit the growth of Ganoderma boninense. The application of microbes from the goat rumen is very beneficial and contributes to improving the plant defense system against the *Ganoderma* b pathogen. This is thought to be because all isolates, especially the 5 isolates that have the highest inhibitory power, contain amylase and protease enzymes which are able to suppress the growth of pathogenic fungi and apart from that they also contain hormones which can stimulate growth, cellulase enzymes which can decompose organic material into compost. This claim is supported by [10]; [11] [12]; [13] where it was found that several endophytic microbes such as *Trichoderma* spp., *Bacillus* sp. and *Pseudomonas* sp. secreting both enzymes, it produces hydrolytic enzymes such as chitinase and glucanase. so that it can act as a biological control agent..

B. *Disease Severity Levels in the Field*

Based on the results of in vito tests, it was found that 5 isolates produced the highest inhibitory power on the growth of the Ganoderma boninense fungus, namely isolates P204, P102, P104, P206 and P106. These five isolates were multiplied and combined into one solution to control stem rot disease which attacks oil palm plants in the leaf growing at the PT company. Sentosa Kalimantan Jaya, Graphic image of the level of severity of attack on oil palm plants after application with biological agents originating from 5 goat rumen isolates can be seen in Figure 2 below:

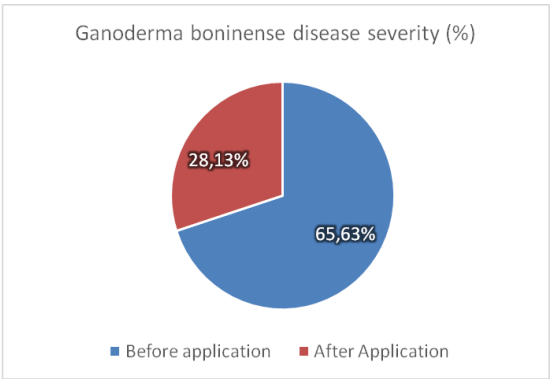

Fig 2. Graphic of the level of severity of attack on oil palm plants after application with biological agents from 5 goat rumen isolates

Based on the graph in Figure 2, it shows that the severity of the attack from the Ganoderma boninense fungus before the application of the biological agent from bacteria originating from the goat's rumen was 65.63%, while after application it fell to 28.13%, this shows that the biological agent is able to increase growth and suppressing the severity of oil palm stem rot disease from the *Ganoderma boninense* fungus. The high reduction in the rate of root rot disease attacks on oil palm plants is also due to the fact that these five isolates contain amylase and protease enzymes which are able to inhibit the growth of *Ganoderma* fungus. Apart from that, the bacteria from these five isolates have the ability to act as biofertilizers because they are able to fix the element N, Phosphate solvent, produces the hormones auxin, gibberellins and cytokinins so that the growth of oil palm plants becomes more fertile and can even produce flowers and fruit again. This opinion is supported by [14] ; by [15] ; [16]; [17] and by [18] research results show that local bacteria are able to inhibit the growth of *Ganoderma* fungi on oil palm plants.

IV. CONCLUSION

- The highest percentage of inhibition against the growth of the Ganoderma boninense fungus was sequentially found in isolates P204, P102, P104, P206 and P106
- The level of root rot disease in oil palm plants can be reduced by the application of biological agents derived from goat rumen isolates

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