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MOLER DISEASE CONTROL IN SHALLOTS USING BOTANICAL PESTICIDES JENGKOL PEEL POWDER AND ITS IMPACT ON MICROBIAL BIODIVERSITY IN PEATLANDS

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A B S T R A C T

Chemical pesticides are still used to control Moler disease on shallots. The impact has the potential to kill macro species as well as non-target bacteria. Several prior researches have shown that botanical pesticides can suppress plant-disturbing organisms, but data on their impact on beneficial microbes is still limited. Hence, the impact of applying jengkol peel to control the primary disease of shallots and its impact on non-target organisms in peatlands was investigated in this study. The study was carried out in Landasan Ulin, South Kalimantan, from April to November 2021. Treatments were t0 (negative control), t1 (positive control, fungicide), t2 (0.125 kg/ha jengkol peelpowder), t3 (0.25 kg/ha jengkol peelpowder), and t4 (0.375 kg/ha jengkol peeljengkol peel powder). The parameters observed were the intensity of moler disease, the components of shallot production, species diversity, species richness, the evenness of microbial species, and the dominance index. The results showed that the application of jengko lpeel powder could suppress the attack of moler disease on shallots. Microbial diversity in shallot plantations treated with botanical pesticides and those not treated with botanical pesticides was similar, in the moderate range, as in shallot plantations treated with chemical pesticides. The species richness index, dominance index, and balance index had low-status values. The types of microbes found were Trichoderma sp., Aspergillus sp., Fusarium sp., Mucor sp., Aeromonas sp., Corynebacterium sp., Enterobacter sp. Sphingomonas sp., and Penicillium sp. The microbial population was affected by pesticide application. Plants that were not applied with botanical pesticides or chemical pesticides had fewer microbes. The application of botanical pesticides produced various impacts, the higher the dose, the lower the microbes in the onion rhizosphere.

Keywords: evenness of species, jengkol peel, microbes., species diversity, species richness.

INTRODUCTION

Shallots (*Allium ascalonicum*) are commonly consumed as a spice to add flavor to dishes, and are used as traditional medicine. In general, shallots contain nutrients and active compounds that have a preventive function, which are obtained when consumed as a cooking spice, and have a curative function when used as herbal medicine. Some of the active chemical compounds (sulfur compounds) in shallots that have pharmacological effects on health include: alliin, allisin, adenosine, diallyl-trisulfide, ajoene, prostaglandin A-1, diallyl-sulfide, phloroglucinol, kaemferol, cycloaliin, and diphenyl-amine. One of the constrain in the production of shallots is the presence of disease attacks caused by *Fusarium oxysporum* f.sp. *cepae*. Molar disease caused by the pathogen *Fusarium oxysporum* f.sp. *cepae* attacks shallots in the highlands and lowlands as well as in all phases of shallot cultivation. Affected plants will show yellowing, twisted, wilted leaves and the tubers will rot causing the plant to die. *F. oxysporum* f.sp. *cepae* can survive in the soil in the form of chlamydospores for a long time so that the pathogen inoculum is always present in the soil (Semangun, 2001, Udiarto *et al*., 2005). Plant mortality due to this disease can reach 100% (Suryabhakti *et al*., 2021). This disease attack causes plants to be unable to be harvested. Until now, farmers are still using synthetic pesticides such as Benlate & Antracol to control this shallot moler disease.

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Given the numerous negative consequences of synthetic pesticide application, including environmental degradation and ecosystem imbalances that might result in human poisoning, an environmentally benign method of controlling plant pests and diseases is required. Certain pesticides derived from plants have a detrimental effect on natural enemies and pollinators (Sharma *et al*., 2012; Lebuhn *et al*., 2012). This is because that botanical pesticides contain active ingredients similar to those found in synthetic pesticides and thus have a detrimental effect on natural enemies and non-target pollinators. Some botanical pesticides are non-specific and toxic. Several studies have found that the application of three types of botanical pesticides (kepayang fruit extract, galam leaf, and *Chromolaena odorata*) has an effect on the diversity of arthropods (Salamiah *et al*., 2021) and microbes–(Salamiah and Aidawati, 2022) in shallot plantations.—The results showed that the application of several types of botanical pesticides affected the diversity of arthropods (Salamiah *et al*., 2021) and microbes (Salamiah and Aidawati, 2022) on shallot plants.

The maximum diversity index of 2.03 was obtained when botanical pesticides were combined with 1 ml/L kepayang fruit extract.—In comparison, *Chromolaena odorata* fruit extract and galam leaf extract significantly reduced the population of microbes by 80.44% and 75.26%, respectively.——Chirinyuh increased the population by 36.60% (Salamiah and Aidawati, 2022). For example, the nicotine from tobacco plant extracts is categorized by WHO as a group Ib toxin, which is very dangerous. Rotenone from Derris and Tephrosia species is classified as Class II.—The natural rotenone and pyrethrum of chrysanthemum are highly toxic. Synthetic pesticides, also when applied, can kill other non-target organisms such as natural predators and parasites from pests and organisms that are beneficial to the health and balance of the ecosystem (McMichael, 2003; Zacharia, 2011). In Africa, pesticide use rates are very low compared to global markets (FAOSTAT, 2005).

The limited understanding of the impact of botanical pesticides on natural enemies and microorganisms makes botanical pesticides still recommended as an environmentally friendly alternative for controlling plant pests and diseases. However, there is insufficient information about the type, dose, and time interval for the proper administration of botanical pesticides. This highlights the importance of collecting information on the impacts of botanical pesticides on beneficial arthropods, particularly natural enemies and beneficial microorganisms in agriculture.

One of the natural materials that can be used as a botanical pesticides to control pests and plant diseases is jengkol peel waste. According to Nurussakinah (2010), the chemical substances identified in jengkol peel include terpenoids, saponins, phenolic acids, and alkaloids. These substances are classified as secondary metabolites and have the potential to protect plants from pests and diseases. The tannin and flavonoid components of jengkol peel are just as protective against pests and illnesses as tannins found in woody plants and herbs. Jengkol peel has the potential to be utilized as a biopesticide due to the presence of these tannins (Nurussakinah, 2010).

Therefore, this study was conducted to examine the effects of jengkol peel in suppressing primary shallot infections and their effect on non-target species (beneficial microorganisms) in shallot plantations on peatlands.

MATERIALS AND METHOD

The research was carried out from April to November 2021 at the Phytopathology Laboratory, Department of Plant Pests and Diseases, Faculty of Agriculture, Lambung Mangkurat University, Banjarbaru, and Tegal Arum Village, Landasan Ulin, Banjarbaru, South Kalimantan, Indonesia.

Research Method

The study used an environmental design, randomized block design with one factor: the administration of 3 doses of jengkol peel powder (0.125 kg/ha, 0.25 kg/ha, and 0.375 kg/ha) plus two controls (positive and negative controls). *Fusarium oxysporum* was inoculated at a density of 10^6 spora/m.The factors tested were: t0 (negative control) = *F. oxysporum* inoculation; t1 (positive control) = Inoculation of *F. oxysporum*+Fungicide (Benomil); t2 = Jengkol peel powder at the concentation of 0.125 kg/ha + *F. oxysporum* inoculation; t3 = Jengkol peel powder at the concentation of 0.25 kg/ha + *F. oxysporum* inoculation; and t4 = Jengkol peel powder at the concentration of 0.375 kg/ha + *F. oxysporum* inoculation.-The isolates of *F. oxysporum* f.sp*. cepae* was isolated from shallots attacked by the moler disease pathogen from peatland, South Kalimantan, Indonesia. The shallot plants used in the study came from shallot breeders from Tapin Regency, South Kalimantan, Indonesia. The shallot seeds

used were the Bima Brebes variety tubers planted at a spacing of 20 cm x 20 cm.

Botanical pesticides were applied three weeks after planting (WAP). Subsequent applications were made at weekly intervals for a total of seven. Maintenance of plants referred to applying basic NPK fertilizer, irrigation, and mechanical removal of nuisance plants.

The experiment was carried out with the following stages of work:

PDA media is made using standard methods of general media processing.

Preparation of *Fusarium oxysporum* isolates. For the preparation of isolates, the symptomatic parts of shallot plants were cut with small scissors and immersed in 70% alcohol solution for 15 seconds. After that, it was washed with sterile water 3 times and then dried on a sterile tissue. The dried pieces were transferred to a petri dish containing PDA media and incubated for 7 days for further purification. After that, The 7-day-old culture of *F. oxysporum* isolate was added with 10 ml of sterile distilled water and then dredged with a flat triangle over the entire surface of the mycelium. The suspension was put into a sample bottle and sterile water was added then shaken for 30 minutes and then the spore density was calculated to get a concentration of 106/ml using a haemacytometer (Rahmawati, 2019).

Land Preparation. The land that is used as a place for planting shallots is peat soil. For cultivation. The lumps of soil are crushed and weeds or grass are cleaned and made beds with a size of 2.5 × 5 m. Lime is given 2 weeks before planting and manure and NPK fertilizer are given 1 week before planting. Then the soil is left for a week to dry. Liming is done if the pH of the soil is not in accordance with the needs for the growth of shallots. After the soil dries, a bed with a size of 1 x 2 m is formed and small ditches are made between the beds to separate the beds.

Preparation of Test Plants. The selection of shallot seeds used was the Bima Brebes variety. Choose seeds that are uniform and that are not attacked by pests and diseases, then the shallots are cleaned of the outermost and dried skin and the roots of the tubers that are still there. The ends of the tubers are cut with a clean knife to facilitate shoot growth, then let stand until the cut marks dry to avoid rotting on the cut marks. The seeds are ready to be planted.

Jengkol peel extraction. The extraction process begins with dry sorting of plant material, namely the separation of dirt and damaged plant parts. Then the sorted material is cut into small pieces and then dried and mashed using a blender until it becomes a powder, then sieved using interwoven gauze. After that the powder was weighed ± 600 grams. Each extracted powder was immersed in methanol for 48 hours with a ratio of 1:10 (w/v). The soaking results are then filtered using filter paper. Each filter was evaporated using a rotary evaporator at a temperature of 60-70 ^{o}C with a pressure of 400-450 mmHg to obtain a crude extract. The remaining methanol from the immersion was used to rinse the residue on the filter paper with 3-4 rinses. The extract obtained was then stored in a refrigerator with a storage temperature of ± 40 ^{o}C until the desired time.

Planting preparation. The shallot seeds used were tuber seeds planted at a spacing of 20 cm x 20 cm.

Pesticide application. Pesticides were applied when the plants were three weeks after planting. Then the application continues regularly once a week.

Plant maintenance carried out includes fertilizing with basic fertilizers, weeding growing weeds, and irrigation.

Microbial Isolation and Purification: Isolation of Non Heat Resistant Bacteria

A total of 10 g of soil was taken from the rhizosphere of shallot plantations and put into an Erlenmeyer containing 90 ml of distilled water, and then shaken for 15 minutes at 150 rpm. The solution was diluted with distilled water to 10^{-7}. A total of 0.5 ml of the 10^{-7} dilution was spread on King's B medium, incubated, and purified.

Isolastion of Heat Resistance Bacteria

Isolation of heat-resistant bacteria was carried out by shaking the soil obtained from the rhizosphere of onion plantations as much as 10 g, which had been dissolved in 90 ml of distilled water for 15 minutes, then diluted to 10^{-7}. The 10^{-7} dilution results were put into a glass bottle and heated at80^{o}C for 30 minutes, after which 0.5 ml was taken and spread on NA media. The NA medium containing the bacterial suspension was incubated at 27^{o}C27^{o}C for 48 hours and then purified (Schaad, *et al*., 2001)

Isolation of fungi from shallot rhizosphere

The soil sample in the rhizosphere of shallot plantations was weighed as much as 10 g, then suspended in 90 ml of distilled water and shaken for 15 minutes at 150 rpm. Following that, 1 ml of the suspension was added to 9 ml of distilled water and homogenized using a vortex. The dilution was carried out until 10^{-5} dilutions were

obtained. A total of 0.5 ml of the dilution results was transferred to a PDA medium, cultured, and purified.

Microbial Identification: Fungus Identification: Identification using pure fungal isolates that have been isolated from diseased shallot plants. The PDA media was cut into a rectangle using a spatula and placed on a glass slide. The isolates to be identified were taken using an ent needle and placed at the end of the rectangular piece of PDA media under the slide glass and then covered with a cover glass. The tissue under the slide glass was moistened in a petri dish using a dropper. Then the petri dish was closed and wrapped with a cling wrap. The growth of spores was observed under a microscope and identified morphologically.

Bacteria Identification: Bacterial identification was performed by evaluating the colony's shape, optical characteristics, colony color, colony size, the shape of the colony's edge/periphery, and the gram of bacteria. Gram bacteria were identified using pure bacterial isolates from healthy shallots, sterile water, and 3% potassium hydroxide (KOH). Gram testing is classified into two major groups, namely gram-positive and gram-negative. The test was conducted by staking one ose of bacterial isolate in a sterile manner and placing it on a glass slide that had been treated with one drop of 3% KOH solution. The bacterial mass was mixed, and the changes were observed. The mucus formation indicated that the bacteria were Gram-negative, whereas the mass of bacteria that did not form mucus indicated that the bacteria were Gram-positive.

Following gram staining, a test was conducted to classify the bacteria into fluorescent and non-fluorescent bacteria. The incubated media was inspected under UV light to observe the bacteria's luminescence. Colonies on each medium were observed, photographed, and identified by comparing the existing literature from books and other sources related to identification guides.

Observation:

1. Intensity of Moler Disease

The disease intensity was observed every week from the appearance of symptoms until before harvest. Based on the systemic nature of the disease, the intensity of the disease was calculated by the formula(Merah *et al*., 2009):

$$I = \frac{a}{b} \times 100\%$$

Note: I: Disease intensity

a: Number of diseased plants

b: Total number of plants

2. Types of microbes. Microbial species identification was carried out to the microbes found in the rhizosphere of shallot plantations treated with botanical pesticides.
3. Microbial Population. The number of colonies was calculated using a colony counter. The petri dish was placed upside down, or the petri dish lid was opened and placed on the colony counter. Calculations were carried out with the help of thick lines on a checkerboard patterned base. The total population was determined by counting the colonies in the top row, then in the bottom row from left to right, and so on. The formula used is as follows:

$$Cc = \frac{\text{Colony number}}{\text{FX diluttion ml suspension}}$$

4. Diversity, richness, dominance, and evenness index of species. The diversity of species was calculated based on Shannon-Wiener diversity index (H'). The richness of species was calculated based on Margalef indeks (R) (1958). The dominance was calculated was calculated based on Simpson dominance index (D). The type of evenness was calculated based on Shannon-Wiener's evenness Index Wiener's evenness Index(E) (Krebs1985). The index equation is equation is as follows:

$$\boldsymbol{D} = \sum_{i=1}^{S} (\boldsymbol{ni/N})^2 \qquad E = \frac{H'}{ln\, S} \qquad R = \frac{(S-1)}{In\, N} \qquad H' = -\sum_{i=1}^{S} (pi)(ln\, pi)$$

$$pi = \text{ ni/N} \qquad \boldsymbol{D} = \sum_{i=1}^{S} (\boldsymbol{ni/N})^2 \qquad E = \frac{H'}{ln\, S} \qquad R = \frac{(S-1)}{In\, N} \qquad H' = -\sum_{i=1}^{S} (pi)(ln\, pi) \qquad pi = \text{ ni/N}$$

Note: ni = the i-th individuals in the species

N = The total number of individuals of all types of species

S = The number of types of species

DATA ANALYSIS

Data collection and identification were processed using Microsoft Office Excel 2007. Next, the data was calculated based on the Shannon-Wiener species

diversity index (Megurran, 1988), the Margalef species richness index (1958), the Simpson species dominance index and the Pielou species evenness index (1975).

RESULTS AND DISCUSSION

Chemical and botanical pesticides had varying effects on the intensity of moler disease infections, productivity, and microorganism populations in the rhizosphere of shallot plantations (Table 1).

Table 1. The correlation between the presence of microbes, the attack of moler disease, and the production of shallot in peatland

Treatments	Species	disease intensity (%)	the yield components			
			Σbulbs/ clump	Bulbs diameter (cm)	wet weight(g)	dry weight(g)
t0	*Trichoderma* sp. *Aspergillus* sp. *Trichoderma* sp. *Aeromonas* sp *Sphingomonas* sp	87.5^{b}	6.8^{a}	1.61^{a}	25.45^{a}	15.08^{a}
t1	*Aspergillus* sp. *Fusarium* sp. *Trichoderma* sp.	56.3^{a}	55.5^{b}	2.20^{a}	719.85^{b}	336.00^{b}
t2	*Fusarium* sp. *Trichoderma* sp. *Corynebacterium* sp *Enterobacter* sp. *Sphingomonas* sp	86.3^{b}	8.8^{a}	1.64^{a}	57.13^{a}	26.98^{ab}
t3	*Aspergillus* sp. *Trichoderma* sp. *Aeromonas* sp *Corynebacterium* sp	70.0^{ab}	15.0^{a}	2.05^{a}	94.65^{a}	58.50^{a}
t4	*Aspergillus* sp. *Penicillium* sp. *Fusarium* sp. *Scopulariopsis* sp. *Curvularia* sp. *Aeromonas* sp *Corynebacterium* sp	60.0^{a}	11.8^{a}	1.73^{a}	70.78^{a}	39.88^{a}

Description: t = treatment code, t0 = Negative Control (*F. oxysporum* inoculation), t1 = Positive control (*F. oxysporum* + Benomyl fungicide), t2 = Jengkol Skin Powder 0, 125 kg/ha + *F. oxysporum* inoculation, t3 = Jengkol peel powder 0.25 kg/ha + *F. oxysporum* inoculation, t4 = Jengkol peel powder 0.375 kg/ha + *F. oxysporum* inoculation, The average value which has the same letter shows no significant difference according to the LSD test at a significant level of 5 %.

Table 1 showed that the given dose of botanical pesticides had an effect on the microbes in the rhizosphere of shallot plantations. The types of microbes found varied, but no single species dominated in each experimental plot.

The provision of botanical pesticides could reduce the intensity of moler disease. The higher the dose, the lower the moler disease attack. Applying 0.375 kg/ha of botanical pesticides resulted in the lowest moler attack intensity of 60.0% (40% inhibition). Meanwhile, chemical pesticide application resulted in low intensity of moler attack, 56.3%. It indicated that the secondary metabolites present in jengkol peel powder were equivalent to chemical pesticides. The phytochemical test conducted by Nurussakinah (2010) found that jengkol peel powder contains terpenoids, saponins, phenolic acids, and alkaloids.

Siswandi *et al*., (2020) discovered that jengkol bark extract at a concentration of 90% inhibited *Fusarium oxysporum* at a rate of 78.43%. Compared to the 10% jengkol peel powder treatment and negative control, it had a very significant effect. It was discovered that

jengkol bark extract was more effective than jengkol peel extract. However, fruit peel powder is better than bark extract because fruit peelis a waste while the bark is not a waste. If excessive exploration is carried out, it will damage plant stems.

Shallot plants that were given botanical pesticides at a dose of 0.125 kg/ha and 0.25 kg/ha did not give a significantly different effect from plants that were not given pesticides (control plants). The types of microbes explored in the rhizosphere of shallot plantations were not positively correlated with a decrease in moler disease intensity and shallots production (Table 1). The application of botanical pesticides derived from jengkol peel extract, in addition to affecting the intensity of moler disease, also affects the yield components of shallots on the number of bulbs per clump, wet weight, and dry weight of shallots.

Soil is inhabited by various microscopic life forms such as bacteria, fungi, actinomycetes, protozoa, and algae. Bacteria are the most prevalent of these microbes, accounting for 95% of them. It has long been recognized that soil has a high concentration of bacteria, ranging from approximately 10^8 to 10^9 cells per gram of soil (Schoenborn *et al*., 2004). The number and types of bacteria found in different soils are influenced by soil conditions, including temperature, humidity, presence of salts and other chemicals, and the number and types of plants found in the soil. Additionally, bacteria are not equally distributed throughout the soil. It means that the concentration of bacteria found around plant roots (rhizosphere) is usually much higher than in other parts of the soil.It is due to the presence of nutrients including sugars, amino acids, organic acids, and other small molecules from plant root exudates which can account for up to one-third of the carbon fixed by plants (Glick, 2012) .

Most fungi-type microbes function as decomposers in peat soil because the microbes secrete extracellular enzymes such as cellulases, hemicellulases, and lignocellulose that can decompose organic matter. Microbial activity that decomposes organic matter determines the maturity level of peat because the saprophytic microbial properties depend on the environment and the organic matter of the substrate. Rosita *et al*. (2014) found that on fibrous peatlands, six pure microbial isolates were found, namely *Aspergillus niger*, Fusarium sp., Paecilomyces sp., *Penicillium variabile*, Pialophora sp., and Verticillium sp. In hemic peat, 10 pure isolates were found: *A. niger*, Fusarium sp., Mortierella sp., Paecilomyces sp., Paecilomyces sp., Penicillium sp., Penicillium sp., Penicillium sp., Rhizopus sp., and *T. harzianum.* Twelve pure isolates were discovered in sapric peat*: Fusarium* sp, *Fusarium* sp*., Paecilomyces* sp*., Paecilomyces* sp*., Penicilliums*p*., Penicillium* sp*., Penicillium* sp*., Penicillium* sp*., Penicillium* sp*., Penicillium* sp *., Pythium* sp*.,* and *Trichoderma* sp *Trichoderma* sp.

The correlation between jengkol peel powder and different types of microorganisms, as well as the decline and growth in microorganism populations in shallot plantations on peatlands

The application of jengkol peel powder had had various effects on the population of microorganisms in the soil (Table 2).

Table 2. Total microorganisms before and after application of jengkol peel powder on shallot plantations in peatlands, South Kalimantan

Treatments	beforeapplicationBefore ($\times 10^{10}$)	after application ($\times 10^{10}$)	increase (+) or decrease (-) of microorganisms application effect (%)
t0	360	128	-47,54
t1	575	664	7,18
t2	154	455	49,43
t3	450	41	-83,30
t4	565	220	-43,95

Description: t = treatment code, t0 = Negative Control (*F. oxysporum* inoculation), t1 = Positive control (*F. oxysporum* + Benomyl fungicide), t2 = Jengkol Skin Powder 0, 125 kg/ha + *F. oxysporum* inoculation, t3 = Jengkol peel powder 0.25 kg/ha + *F. oxysporum* inoculation, t4 = Jengkol peel powder 0.375 kg/ha + *F. oxysporum* inoculation

There was a decrease in microbes' population by 47.54% in shallot plantations without botanical pesticides. While the application of chemical pesticides increased the microbial population by 7.18%, this increase was not as high as the administration of botanical pesticides with a dose of 0.125 kg/ha. When the dose of botanical pesticides was increased to 0.25 kg/ha and 0.375 kg/ha, the microbial population again experienced a considerable decline by 83.30% and 43.95%, respectively.

This research has succeeded in collecting two groups of microbes, namely bacteria and fungi. Five types of bacteria were found: *Sphinggomonas* sp., Enterobacteria sp., Corynebacterium sp., Bacillus sp., and Aeromonas sp. (Table3); Table 3 five types of fungi, namely *Trichoderma* were found sp., *Fusarium* spp., Mucor spp., *Aspergillus* sp., and *Penicillium* sp. from all research plots.

Table 3. Population and types of bacteria isolated in shallot plantations applied with jengkol peeljengkol peel powder

Treatments	Total population	species	Gram test
t0	127 × 10^{10}*cfu*	*Sphingomonas* sp.	Positive
t1	584 × 10^{10}*cfu*	*Bacillus* sp. *Aeromonas* sp.	Positive
t2	438 × 10^{10}*cfu*	*Bacillus* sp. *Aeromonas* sp.	Positive
t3	37 × 10^{10}*cfu*	*Corynebacterium* sp. *Enterobacteria* sp.	Positive
t4	216 × 10^{10}*cfu*	*Bacillus* sp.	Positive

Description: t = treatment code, t0 = Negative Control (*F. oxysporum* inoculation), t1 = Positive control (*F. oxysporum* + Benomyl fungicide), t2 = Jengkol Skin Powder 0, 125 kg/ha + *F. oxysporum* inoculation, t3 = Jengkol peel powder 0.25 kg/ha + *F. oxysporum* inoculation, t4 = Jengkol peel powder 0.375 kg/ha + *F. oxysporum* inoculation

Table 4 below summarizes the characteristics of each discovered bacterial genus.

Table 4. Characteristics of five types of bacteria in each research plot.

No.	Isolat code	Colony shape	optical properties	colony color	texture	Genus
1	T1A	spherical	opaque	white	rough	*Aeromonas* sp.
3	T3A	spherical	opaque	white	rough	*Aeromonas* sp.
4	T40A	spherical	slightly transparent	slightly yellowish	smooth	*Corynebacterium* sp.
5	T35A	spherical	slightly transparent	milky white	smooth	*Enterobacter* sp.
6	T421	spherical	translucent	milky white	smooth	*Enterobacter* sp.
7	T010A	spherical	slightly transparent	slightly yellowish	smooth	*Sphingomonas* sp.
8	T411	spherical	opaque	orange pink	smooth	*Bacillus* sp.
9	T231	spherical	opaque	white	dry surface wrinkle	*Bacillus* sp.
10	BT25A	spherical	opaque	white	rough	*Bacillus* sp.
11	BT210A	spherical	opaque	white	rough	*Bacillus* sp.
12	BT410F	spherical	transparent	yellow	smooth	*Bacillus* sp.
13	T32	spherical	opaque	cream	smooth	*Bacillus* sp.
14	T0B	spherical	opaque	cream	smooth	*Bacillus* sp.
15	T1D	spherical	opaque	white	rough	*Bacillus* sp.
16	T2B	spherical	opaque	white	rough	*Bacillus* sp.

As shown in Table 1, Table 3, and Table 4, this research has succeeded in collecting five types of bacteria, namely *Sphinggomonas* sp., *Enterobacteria*, *Corynebacterium* sp., *Bacillus* sp., and *Aeromonas* and *Aeromonas* sp. with the following properties.

Bacillus sp. *Bacillus* sp. isolated from shallot plantations in this study showed spherical colonies, opaque optical properties, and slight variations in colony color; some were cream, and some were white. The cream-colored colonies had a smooth texture, while the white colonies had a rough texture.

Bacillus sp. is a PGPR (Plant Growth Promoting Rhizobacteria) that has the ability to stimulate plant growth and production.*Bacillus* can fixation N_2, dissolve phosphate, and synthesize phytohormones IAA (Indole 3- Acetic Acid). *Bacillus* sp. as PGPR can increase the availability of low nitrogen and phosphate nutrients in paddy fields. Loss of nitrogen nutrients generally occurs due to leaching and runoff in flooded soils and low availability of phosphate due to complex binding to Al^{2+} and Fe^{2+} elements. Nitrogen and phosphate availability in paddy fields can be improved by applying *Bacillus* sp. biofertilizers.

The nitrogenase test using the Acetylene Reduction

Assay (ARA) method found that *Bacillus* had a nitrogenase activity of 0.05685 m ml^{-1} $hour^{-1}$ and dissolving phosphate from a $Ca_3(PO_4)^2$ with a Dissolution Index (IP) of phosphate 2.6.Phosphate solubility is caused by acetic acid, oxalic acid, lactic acid, and malic acid produced by *Bacillus* sp. Biological fertilizers *Bacillus* sp. can reduce 25% usage of NPK fertilizer in lowland rice production (Husna *et al*., 2019).

Bacillus sp. was one of the microorganisms discovered in the peatlands where the research was conducted. Based on the relationship between the types of microbes found and the intensity of disease attacks on shallot plantations (Table 1), it can be concluded that the presence of *Bacillus*sp . does not play its function as a PGPR, namely as a bioprotectant, because the intensity of molar attack remains relatively high. The role of bacteria as PGPR will increase if it is in a consortium with several other antagonistic bacteria. Four bacterial consortia, each containing three bacterial strains that had complete or complementary phenotypic properties. Not only can PGPR promote plant growth, but it also possesses the ability to biologically manage plant-disturbing organisms (Kanjanasopa *et al*., 2021).

Sphingomonas sp. Characteristics of *Sphingomonas* sp. isolated from shallot plantations showed round colonies, slightly transparent optical properties, and a slightly yellowish color with a smooth texture.

Sphingomonas have been isolated from various anthropogenic contaminated environments—including terrestrial (subsurface) soil (Mueller *et al*., 1990; Adkins, 1999; Momma *et al*., 1999; Bastiaens *et al*., 2000; Pinyakong *et al*., 2000; Sørensen *et al*., 2001; Cassidy *et al*., 1999) and were found to have a unique ability to degrade various pollutants, including azo dyes (Stolz, 1999), chlorinated phenols (Cassidy *et al*., 1999; Crawford and Ederer, 1999), dibenzofurans (Keim *et al*., 1999; Wittich *et al*., 1999), insecticides (Nagata *et al*., 1999), and herbicides (Adkins, 1999; Kohler, 1999). In addition, *Sphingomonas* strains are frequently isolated as decomposers of polycyclic aromatic hydrocarbons (PAHs) from contaminated soil (Mueller *et al*., 1990; Khan *et al*., 1996; Bastiaens *et al*., 2000; Pinyakong *et al*., 2000). PAHs are highly hydrophobic toxic chemicals with low solubility in water, making them less available for natural bacterial degradation. Due to their ubiquitous distribution and diverse catabolic ability against persistent organic pollutants, *Sphingomonas* strains can be considered important biocatalysts for soil bioremediation.

Enterobacteria sp. *Enterobacteriaceae* sp. isolated from shallot plantations showed spherical colonies, slightly transparent and translucent optical properties, milky white colony color, and smooth texture.

Enterobacteriaceae are facultative aerobic microbes (Kersters *et al*., 2006) that anaerobically degrade sugars into acetate, CO_2, H_2, formate, lactate, succinate, and ethanol (White, 2007), supplemented by various carbon sources(Kusel *et al*., 2008). These findings suggest that (Daniela *et al*., 2009) facultative aerobes compete for dissolved organic carbon in the anoxic microzone of aerated soils (Daniela *et al*., 2009) under anoxic conditions similar to those formed in anoxic slurries.Both facultative aerobes and obligate anaerobes from methanogenic soils mainly assimilate [13C-U]-glucose under anoxic conditions .(Hamberger *et al*., 2008)

Corynebacterium sp. *Corynebacterium* sp. isolated from shallot plantations showed round colonies, slightly transparent optical properties, slightly yellowish colony color, and smooth texture.

Corynebacterium is a genus of Gram-positive and mostly aerobic rod-shaped bacteria. One of the *Corynebacterium* species is *C. glutamicum* which is gram-positive, a non-motile bacterium belonging to the phylum Actinobacteria. *C. glutamicum* is a glutamate-producing bacterium (Kinoshita *et al*., 2004). Due to its great glutamate and lysine production capacity, it has become a widely used organism in biotechnology (Kelle *et al*., 2013; Kumagai *et al*., 2000; Kimura *et al*., 2012). Apart from glucose, it can utilize various other carbon sources such as fructose, sucrose, gluconate, acetate, propionate, pyruvate, L-lactate, ethanol, glutamic amino acids, and serine (Kalinowski *et al*., 2003; Netzer *et al*., 2004; Cocaign *et al*., 1993; Claes *et al*., 2002). *C. glutamicum* has *glutamicum* has a potential gene that allows it to catabolize Neu5Ac as the sole carbon source (Holder *et al*. 2011).

Aeromonas sp. *Aeromonas* sp. isolated from shallot plantations showed spherical colonies, opaque optical properties, white colony color, and rough texture.

Today, the genus *Aeromonas* contains 24 legally published species. Bergey's Manual of Systematic Bacteriology, Second Edition (Bergey's) permits far less (Martin-Carnahan and Joseph, 2005.). *Aeromonassp* is halophilic, does not form spores, gram-negative, and is widely distributed in soil, food, and aquatic

environments. The three main pathogenic species of this genus are *Aeromonas hydrophila*, *A. caviae*, and *A . sobria* (Tomas). These biochemically distinct species have now been further subdivided into DNA hybridization groups (Igbinosa *et al*. 2012). The characteristics of each genus of fungi found in this study were presented in Table 5.

Table 5. The results of macroscopic and microscopic identification of fungi on shallot plantations applied with botanical pesticides

No	Isolat code	Macroscopis		Microscopis			Genus
		Colony color	Colony shape andtextureandtexture	Conidiophores/hyphae	Conidia/spore	Fialid	
1.	CT431	green	spherical	branched	round	Erect,short	*Trichoderma* spp.
2.	CT410 B CT45	dark green	rounded, circularsmoothcircularsmooth and embossed surface	cylindrical	round	erect	*Aspergillus* spp.
3.	CT231 CT311	white	Round, smooth and slippery surface, cotton-like surface	Branched, insulated	Ovoid, crescent	single	*Fusarium* spp.
4.	CT0	pale white to gray	Grew upward, thinly fibrous	unbranched, not insulated, transparent	round, transparent	single	*Mucor* spp.
5.	CT35A	greenish-white then turned bottle green	Irregular	Branched, hyaline	round	Erect	*Penicillium* sp.

The presence of microbes is influenced by environmental factors in which they are found. Environmental elements affecting peat soil decomposition include soil acidity (pH), soil temperature, soil moisture, vegetation, and the level of decomposition of peat soil. Soil acidity is a factor controlling the type of microbial diversity. The acidity of the environment (pH of the substrate) is critical for microbial development since certain enzymes are only active at a specific pH. The measurement showed that the soil pH was in the range of 3.55-4.11. Generally, microbes can grow at pH below 7 (Gandjar *et al*., 2006). Similar results were also reported by (Waluyo, 2004), that microbes are aerobic organisms and have a wide pH range, ranging from 2.0 to 8.5.

As shown in Table 1 and Table 5, this research succeeded in collecting five types of fungi, namely *Trichoderma* sp., *Fusarium* sp., *Aspergillus* sp., *Scopulariopsis* sp.and*Penicillium* sp. with the following properties:

Trichoderma sp. has green colonies, spherical shape, branched conidiophores, spherical conidia with erect phialides position and short size, belonging to the ascomycetes class, antagonistic to plant diseases because they have antifungal activity. *Trichoderma* occurs naturally in forest soils, agricultural fields, and on woody substrates.

Several studies have found that *Trichoderma* is one of the fungi that can act as a biocontrol agent due to its antagonistic nature toward other fungi, particularly pathogenic fungi. The antagonistic activity may include competition, parasitism, predation, or the formation of toxins such as antibiotics. This biocontrol agent can be extracted from *Trichoderma* and utilized to treat crop damage caused by pathogens for biotechnological applications.

As a biological agent, *Trichoderma* has the potential to maintain plant resistance systems, for example, from attack by pathogens such as pathogenic fungi. Because the intensity of the moler disease attack is still high in the shallot plantation under study, the presence of *Trichoderma* has not yet functioned as an antagonistic agent. This is most likely because the ability and mechanism by which *Trichoderma* inhibits pathogen growth vary between species. This ability gap is influenced by ecological factors that affect metabolite

production. *Trichoderma* produces volatile and non-volatile metabolites.

The nutritional composition of the media affects the outcomes of these metabolites. *Trichoderma* produces chitinolytic protein and chitinase enzymes when in chitin-rich conditions. This enzyme contributes to the improvement of biocontrol actions against chitin-containing diseases.

Aspergillus sp. isolates recovered from shallot plantations showed characteristic dark green colony color, rounded colony shape, circular colony texture, smooth and embossed surface, cylindrical conidiophores, spherical conidia, and erect phialides. This genus, *Aspergillus*, has septate hyphae and hyaline. It is supported by Noerfitryani and Hamzah (2017) who reported that the macroscopic characteristics of the *Aspergillus* fungus on PDA media are that its surface is light green to dark green and black, had a flour-like texture, with microscopic characteristics, namely conidia spherical in shape, with septate hyphae and hyaline. *Aspergillus* sp. is a phosphate solubilizing fungus that has been proven to dissolve phosphate from poorly soluble sources. *Aspergillus* sp. also can dissolve insoluble inorganic phosphates by secreting organic acids (Saraswati *et al*., 2007). In addition, according to Saraswati (Saraswati *et al*., 2007), *Aspergillus* sp. is capable of producing proteases that function in the transformation of organic nitrogen (in the form of protein) in the soil and other organic waste materials into inorganic N (NH_4^+), which the Kintamani Siamese citrus plant can utilize.

One of the Aspergillus species is *A. niger*. *Aspergillus niger* produces steroids (Lima *et al*., 2019). This species is a major source of citric acid and accounts for more than 99% of global citric acid production, or more than 1.4 million tonnes per year. *Aspergillus niger* is also commonly used to produce enzymes, including glucose oxidase, lysozyme, and lactase (Thom and Church, 1926).

Another *Aspergillus* species is *A. nidulans* (*Emericella nidulans*). *A. nidulans* is a pioneering organism to have its genome sequenced by researchers at the Broad Institute. In 2008, seven other Aspergillus species had their genomes sequenced: *Aspergillus niger* (two strains), industrially applicable *A. oryzae*, and *A. terreus*, and the pathogens *Aspergillus clavatus*, *Aspergillus fischerianus, Neosartoryafischeri Neosartorya*, *Aspergillus flavus*, and *A. fumigatus* (two strains). *Aspergillus fischerianus* is rarely pathogenic but is closely related to the common pathogen *A. fumigatus.* One of the reasons for sequencing the *A. fischerianus* genome was to have a better understanding of *A. fumigatus*' pathogenicity.

Fusarium sp. isolates collected from shallot plantations showed the characteristics of white colony color, round colony shape, smooth and slippery colony texture, branched, insulated cotton-like surface, ovoid-shaped conidiophores, and crescent-shaped conidia.

Fusarium is a soil saprophyte but can be pathogenic to plants*.* These molds can induce root rot and contribute to decomposition is in accordance with Saragih (2009) who found five dominant decomposers, namely *Fusarium* sp, *A. ochraceus*, *Aspergillus niger*, *Monascusrube*, and Trichoderma sp.

Most of the *Fusarium* species are economically important plant pathogens. *Fusarium* can be endophytic or saprophyte. As a pathogen, *Fusarium* causes various diseases in agricultural, horticultural, and forestry crops (Moore *et al*. 2001; Ploetz, 2001; Summerell *et al*. 2003). Over 81 commercially significant crops have been afflicted by at least one disease caused by the *Fusarium* fungus (Leslie and Surnrnerell, 2006). *Fusarium* causes pre- and post-emergence damping-off (Palmero *et al*., 2009). *Fusarium* fungi are often found as endophytes in various plants in agricultural ecosystems (Kuldau and Yates, 2000). While the *Fusarium* fungus can infect interior plant tissues without causing symptoms, it can develop disease symptoms when plants are subjected to drought or other environmental stresses.

Mucor spp. The colonies were pale white to gray. The colonies grew upward. The surface of the colonies was thinly fibrous, conidiophores unbranched, conidia not insulated, round in shape, and transparent in color.

Mucor sp. is one type of mold found in the microbial isolation carried out in this study. Mold can grow on peat soil because of the substrate produced by wood containing lignin and cellulose. According to Sennang *et al*. (2012) organic compounds such as lignin and cellulose provide energy and food for soil mould. Peat soil is a rich source of energy and food for soil molds.The deeper the peat, the lower the oxygen condition (Barchia, 2012). This is due to the low intensity of light that can penetrate the soil and the peat soil environment, which is generally always waterlogged. It is a factor that influences the presence of molds and how molds decompose peat organic materials. The more mature the peat, the more decomposing molds are discovered (Buckman and Brady, 1960). Saragih (2009) found two types of mold at the fibric peat maturity level

(*Aspergillus* sp. and *Mucor* sp.), five types of mold at the hemic maturity level (*P. chrysogenum*, Mucor sp., *P. digitatum*, Culvularia sp., and Penicillium sp.), and four types of mold at sapric maturity level (Aspergillus sp. 1, Aspergillus sp. 2, Fusarium sp., and *P. chrysogenum*).

Penicillium sp. Isolates of Penicillium sp. harvested in this study had the characteristics: at first, the colony's surface was greenish-white then turned bottle green, there were white cotton fibers, irregular colony shape and texture, branched conidiophores, hyaline conidia, and erect phialides. On the sixth day following incubation, colonies filled the Petri dish.Conidia walls are smooth. Conidiophores have smooth walls, branched conidiophores, and have metulae and phialides.

The genus *Penicillium* has septate hyphae and hyaline. It is consistent with the findings of (Anggraeni and Usman, 2015), who discovered that colonies of *Penicillium* sp. begin as white and then change turquoise, greenish-grey, olive-grey, and occasionally yellow or reddish. Meanwhile, the microscopic form of the fungus *Penicillium* sp. has hyaline hyphae, spherical unicellular conidia, and a set of phialides. According to reports, Penicillium sp. is capable of protecting plants against pathogen attack while simultaneously promoting plant development (Rozali, 2015). Additionally, *Penicillium* serves as a decomposer, contributing to soil fertility (Purwati and Hamidah, 2019). *Penicillium* sp. is a soil microorganism whose job is to deliver nutrients by converting insoluble inorganic phosphate compounds into soluble forms (H_2PO_4 and HPO_4) that plants can absorb. Microbes with a high capacity for phosphorus (P) dissolution usually have a high capacity for potassium (K) dissolution too (Wulandari *et al*., 2013).

Application of synthetic pesticides and botanical pesticidesbotanical pesticides to microbial biodiversity, species richness, dominance index, and microbial balance index in shallot plantations

The application of botanical pesticides had various effects on the diversity index, species richness index, dominance index, and microbial balance index in shallot plants (Table 6 and Table 7).

Table 6. The average diversity, species richness, dominance, and microbial balance index in shallot plants given the peel powder of jengkol fruit peel vegetable pesticide on peatlands.

Treatmens	Species Diversity Index (H′)	Species Richness Index (R)	Domination Index (D)	Microbial Balance Index (E)
t0	1.18	0.83	0.34	0.73
t1	1.23	0.94	0.43	-0.08
t2	1.65	1.32	0.23	-0.12
t3	1.22	1.67	0.45	-0.12
t4	1.67	2.23	0.27	0.91

Description: t = treatment code, t0 = Negative Control (*F. oxysporum* inoculation), t1 = Positive control (*F. oxysporum* + Benomyl fungicide), t2 = Jengkol Skin Powder 0, 125 kg/ha + *F. oxysporum* inoculation, t3 = Jengkol peel powder 0.25 kg/ha + *F. oxysporum* inoculation, t4 = Jengkol peel powder 0.375 kg/ha + *F. oxysporum* inoculation

Several studies have found that the application of three types of botanical pesticides (kepayang fruit extract, galam leaf, and *Chromolaena odorata*) has an effect on the diversity of arthropods (Meiyana *et al*., 2021) and microbes (Salamiah and Aidawati, 2022) in shallot plantations. The maximum diversity index of 2.03 was obtained when botanical pesticides were combined with 1 ml/L kepayang fruit extract. In comparison, *Chromolaena odorata* fruit extract and galam leaf extract significantly reduced the population of microbes by 80.44 % and 75.26 %, respectively. Chirinyuh increased the population by 36.60% (Salamiah and Aidawati, 2022). For example, the nicotine from tobacco plant extracts is categorized by WHO as a group Ib toxin, which is very dangerous. Rotenone from Derris and Tephrosia species is classified as Class II. The natural rotenone and pyrethrum of chrysanthemum are highly toxic (Isman, 2014).

Biodiversity refers to the variety of species and their distribution patterns within an organism or living natural resources and ecosystems. Biodiversity consists of two components: the total number of species per unit area and evenness (abundance, dominance, and spatial distribution of individual species). A biodiversity index is a metric that integrates these two concepts into a single number. The variables were then combined into a single value regarding the number of species, relative species abundance, homogeneity, and sample plot size. For this reason, the biodiversity index depends on the richness index, the diversity index, and evenness index (Barnes *et al*., 1997).

Table 7. Status of diversity, richness, dominance and balance of microbial populations index in shallot plantations applied with jengkol peel botanical pesticides

Treatments	Standard of Species Diversity Index (H′)	Species Diversity Index (H′)	Standard of Species Richness Index (R)	Species Richness Index (R)	Standard of Domination Index (D	Domination Index (D)	Standard of Microbial Balance Index (E)	Microbial Balance Index (E)
t0	1,0 < H'≤ 3,0 : medium	1.18	R ≤ 3,5 : low	0.83	D < 0.5: low	0.34	E > 0.5: high	0.73
t1	1,0 < H'≤ 3,0 : medium	1.23	R ≤ 3,5 : low	0.94	D < 0.5: low	0.43	E < 0.5: low	- 0.08
t2	1,0 < H'≤ 3,0 : medium	1.65	R ≤ 3,5 : low	1.32	D < 0.5: low	0.23	E < 0.5: low	- 0.12
t3	1,0 < H'≤ 3,0 : medium	1.22	R ≤ 3,5 : low	1.67	D < 0.5: low	0.45	E < 0.5: low	- 0.12
t4	1,0 < H'≤ 3,0 : medium	1.67	R ≤ 3,5 : low	2.23	D < 0.5: low	0.27	E > 0.5: high	0.91

Description: t = treatment code, t0 = Negative Control (*F. oxysporum* inoculation), t1 = Positive control (*F. oxysporum* + Benomyl fungicide), t2 = Jengkol Skin Powder 0, 125 kg/ha + *F. oxysporum* inoculation, t3 = Jengkol peel powder 0.25 kg/ha + *F. oxysporum* inoculation, t4 = Jengkol peel powder 0.375 kg/ha + *F. oxysporum* inoculation

Table 7 showed that the Shannon Wiener (H') diversity index value in shallot plantations area ranged from 1.18 to 1.67.According to Krebs (1985), this value is in the moderate diversity category because the H' value is in the range of 1-3.Diversity in the medium category indicates that the community's species are relatively diversified, and the ecosystem is stable (Odum and Barrett, 1971).

The Margalef species richness index (R') in the research location ranges from 0.83-2.23. According to Magurran (1988), a richness index of R 3 is considered low. This situation indicates that the shallot plantation contains a limited number of microbial species. The development and distribution of soil microbes are generally influenced by biotic and abiotic factors in their living environment, including soil oxygen and vegetation (Jha *et al*., 1992; Rao, 1994). Aerobic microbes require oxygen in their respiration to develop properly, which means they require a well-aerated environment (Suharni, 2008). Based on the type of land, the research location was swampland with inadequate aeration. This condition can be a limiting factor for development. Only certain microbes can adapt, resulting in a low number of microbial species found in the research location.

Vegetation is another environmental factor that affects the number of microbial species. Vegetation on the soil surface can modify microbial communities (Bezemer *et al*., 2006; Carney and Matson, 2006).Variations in the number and types of these plants affect the composition of the soil microbial community (Han *et al*., 2007; Zul *et al*., 2007; Liu *et al*., 2020). Bernadip *et al*. (2015) found that the shallot rhizosphere in several observation areas has a soil pH that tends to be acidic. This may also be a limitation of the low microbial species present in the study site.

The dominance index (D) was between 0.23 and 0.45, considered the low category. This suggests that microbial species are diversified in the shallot plantation, with no dominant species. This is in line with Odum and Barrett (1971naru) statement`s that the range of dominance index values starts from 0-1. If the value obtained is near zero, it indicates no species that completely dominate other species in the observed community structure.

Evenness Index (E) describes the distribution of species in a community, and this value is related to the Dominance index (D). The evenness index in shallot plantations ranged from -0.08 to 0.91. When viewed

from the average value of 0.26, it is included as a low category. It depicts that the distribution of species in the community is not evenly distributed. A species dominates, but its dominance is not extreme if associated with a low dominance index value.

CONCLUSIONS

1. The application of jengkol peel powder suppressed the attack of moler disease on shallots. The lowest attack intensity was achieved using botanical pesticides at 0.375 kg/ha, or 60%.
2. Biodiversity in shallot plantations was quite good due to a moderate diversity index and the absence of dominant species.
3. Application of botanical pesticides at a dose of 0.125 kg/ha increased the microbial population in the soil, but the intensity of moler disease on shallots was still high (86.3%).
4. The type of found microbe was*Trichoderma* sp., *Aspergillus* sp., *Fusarium* sp., *Penicillium* sp., *Mucor* sp., *Aeromonas* sp., *Corynebacterium* sp., *Enterobacter* sp., *Sphingomonas* sp., and*Bacillus* sp..
5. Microbial population is affected by pesticide application. Plants that were not treated with botanical pesticides or chemical pesticides had a drop in the number of microbes, whereas chemical pesticides increased the number of microbes in the rhizosphere.
6. The application of botanical pesticides had a varied impact as the dose increased, and then it affected the decrease in microbes in the shallot rhizosphere.

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REFERENCES

Adkins, A. 1999. Degradation of the phenoxy acid herbicide diclofop-methyl by *Sphingomonas paucimobilis* isolated from a Canadian prairie soil. Journal of Industrial Microbiology and Biotechnology, 23: 332-335.

Anggraeni, D.N. and M. Usman. 2015. Activity Test and Identification of Rhizosphere Fungi on Root Soil of Banana Plants (*Musa paradisiaca*) against Fusarium Fungus. BioLink, 1: 89–98.

Barchia, M. F., G. Agroekosistem and T. Karbon. 2012. Gadjah Mada University Press. Yogyakarta.

Barnes, B.V., R.Z. Donald, R.D. Shirley and H.S. Stephen. 1997. Forest Ecology. New York, John Wiley and Sons, pp. 349–588.

Bastiaens, L., D. Springael, P. Wattiau, H. Harms, R. deWachter, H. Verachtert and L. Diels. 2000. Isolation of adherent polycyclic aromatic hydrocarbon (PAH)-degrading bacteria using PAH-sorbing carriers. Applied and environmental microbiology, 66: 1834-1843.

Bernadip, B. R., H. Hadiwiyono and S. Sudadi. 2015. Diversity of Fungi and Bacteria of Shallot Rizosphere Against Moler Pathogen. Journal of Soil Science and Agroclimatology, 11: 52-58.

Bezemer, T. M., C. S. Lawson, K. Hedlund, A. R. Edwards, A. J. Brook, J. M. Igual, S. R. Mortimer and W. H. Van Der Putten. 2006. Plant species and functional group effects on abiotic and microbial soil properties and plant-soil feedback responses in two grasslands. Journal of Ecology, 94: 893-904.

Buckman, H. O. and N. C. Brady. 1960. The Nature and Properties of Soils. Soil Science, 90: 212-220.

Carney, K. M. and P. A. Matson. 2006. The Influence of Tropical Plant Diversity and Composition on Soil Microbial Communities. Microbial Ecology, 52: 226-238.

Cassidy, M. B., H. Lee, J. T. Trevors and R. B. Zablotowicz. 1999. Chlorophenol and nitrophenol metabolism by Sphingomonas sp UG30. Journal of Industrial Microbiology and Biotechnology, 23: 232-241.

Claes, W. A., A. Pühler and J. Kalinowski. 2002. Identification of two prpDBC gene clusters in *Corynebacterium glutamicum* and their involvement in propionate degradation via the 2-methylcitrate cycle. Journal of Bacteriology, 184: 2728-2739.

Cocaign, M., C. Monnet and N. Lindley. 1993. Batch kinetics of *Corynebacterium glutamicum* during growth on various carbon substrates: use of substrate mixtures to localise metabolic bottlenecks. Applied Microbiology and Biotechnology, 40: 312-321.

Crawford, R. L. and M. M. Ederer. 1999. Phylogeny of Sphingomonas species that degrade pentachlorophenol. Journal of Industrial Microbiology and Biotechnology, 23: 320-325.

Daniela, M., S.K. Degelmann, M. Dumont, J.C. Murrell and H.L. Drake. 2009. Enterobacteriaceae facilitate the anaerobic degradation of glucose by a forest soil. Federation of European Microbiological Societies. Published by Blackwell Publishing Ltd.

FAOSTAT. 2005. The State of Food and Agriculture 2005. The State of Food and Agriculture. UN.

Gandjar, I., W. Samsuridjal and A. Oetari. 2006. Basic and Applied Mycology, Yayasan Obor Indonesia.

Glick, B. R. 2012. Plant growth-promoting bacteria: mechanisms and applications. Scientifica, 2012: 963401-963401.

Hamberger, A., M. A. Horn, M. G. Dumont, J. C. Murrell and H. L. Drake. 2008. Anaerobic consumers of monosaccharides in a moderately acidic fen. Applied and environmental microbiology, 74: 3112-3120.

Han, X., R.Q. Wang, J. Liu, M.C. Wang, J. Zhou and W.H. Guo. 2007. Effects of vegetation type on soil microbial community structure and catabolic diversity assessed by polyphasic methods in North China. Journal of Environmental Sciences, 19: 1228-1234.

Holder, J. W., J. C. Ulrich, A. C. DeBono, P. A. Godfrey, C. A. Desjardins, J. Zucker, Q. Zeng, A. L. B. Leach, I. Ghiviriga, C. Dancel, T. Abeel, D. Gevers, C. D. Kodira, B. Desany, J. P. Affourtit, B. W. Birren and A. J. Sinskey. 2011. Comparative and functional genomics of *Rhodococcus opacus* PD630 for biofuels development. PLoS Genet, 7: e1002219-e1002219.

Husna, M. 2019. The Role of Bacillus sp. in the Provision of Nutrients and Growth Regulatory Substances in the Production of Rice Fields. Journal of Soil and Climate, 43: 117-125.

Igbinosa, I. H., E. U. Igumbor, F. Aghdasi, M. Tom and A. I. Okoh. 2012. Emerging Aeromonas species infections and their significance in public health. Scientific World Journal, 2012: 625023-625023.

Isman, M. B. 2014. Botanical insecticides: a global perspective, Biopesticides: State of the art and future Opportunities. ACS Publications, pp. 21-30.

Jha, D. K., G. D. Sharma and R. R. Mishra. 1992. Ecology of soil microflora and mycorrhizal symbionts in degraded forests at two altitudes. Biology and Fertility of Soils, 12: 272-278.

Kalinowski, J., B. Bathe, D. Bartels, N. Bischoff, M. Bott, A. Burkovski, N. Dusch, L. Eggeling, B. J. Eikmanns and L. Gaigalat. 2003. The complete *Corynebacterium glutamicum* ATCC 13032 genome sequence and its impact on the production of L-aspartate-derived amino acids and vitamins. Journal of biotechnology, 104: 5-25.

Kanjanasopa, D., W. Aiedhet, S. Thitithanakul and C. Paungfoo-Lonhienne. 2021. Plant Growth Promoting Rhizobacteria as Biological Control Agent in Rice. Agricultural Sciences, 12: 1-8.

Keim, T., W. Francke, S. Schmidt and P. Fortnagel. 1999. Catabolism of 2,7-dichloro- and 2,4,8-trichlorodibenzofuran by Sphingomonas sp strain RW1. Journal of Industrial Microbiology and Biotechnology, 23: 359-363.

Kelle, R., T.H. Bathe, B.S.F. Lkdb and K. Ha. 2013. Handbook of *Corynebacterium glutamicum*, 147, 465–488.

Kersters, K., P. Lisdiyanti, K. Komagata and J. Swings. 2006. The Family Acetobacteraceae: The Genera Acetobacter, Acidomonas, Asaia, Gluconacetobacter, Gluconobacter, and Kozakia. The Prokaryotes. Springer New York, pp. 163-200.

Khan, A. A., R. F. Wang, W. W. Cao, W. Franklin and C. E. Cerniglia. 1996. Reclassification of a Polycyclic Aromatic Hydrocarbon-Metabolizing Bacterium, Beijerinckia sp. Strain B1, as *Sphingomonas yanoikuyae* by Fatty Acid Analysis, Protein Pattern Analysis, DNA-DNA Hybridization, and 16S Ribosomal DNA Sequencing. International Journal of Systematic Bacteriology, 46: 466-469.

Kimura, M., H. Araoka and A. Yoneyama. 2012. *Aeromonas caviae* is the most frequent pathogen amongst cases of *Aeromonas bacteremia* in Japan. Scandinavian Journal of Infectious Diseases, 45: 304-309.

Kinoshita, S., S. Udaka and M. Shimono. 1957. Studies on the amino acid fermentation. The Journal of General and Applied Microbiology, 3: 193-205.

Kohler, H. P. E. 1999. *Sphingomonas herbicidovorans* MH: a versatile phenoxyalkanoic acid herbicide degrader. Journal of Industrial Microbiology and Biotechnology, 23: 336-340.

Krebs, C.J. 1985. Ecology The Experiment Analysis of Distribution and Abundance. United States, Harper Collins Publisher, 765ppp.

Kuldau, G.A. and I.E. Yates. 2000. Evidence for Fusarium endophytes in cultivated and wild plants. In: E.C.W. Bacon and J.F. White (Editors) Microbial

Endophytes. Marcel Dekker, New York, USA, 85–117.

Kumagai, H. 2000. Microbial Production of Amino Acids in Japan. History of Modern Biotechnology I. Springer Berlin Heidelberg, pp. 71-85.

Küsel, K., M. Blöthe, D. Schulz, M. Reiche and H. L. Drake. 2008. Microbial reduction of iron and porewater biogeochemistry in acidic peatlands. Biogeosciences, 5: 1537-1549.

Lebuhn, G., S. Droege, E. F. Connor, B. Gemmill-Herren, S. G. Potts, R. L. Minckley, T. Griswold, R. Jean, E. Kula, D. W. Roubik, J. Cane, K. W. Wright, G. Frankie and F. Parker. 2012. Detecting Insect Pollinator Declines on Regional and Global Scales. Conservation Biology, 27: 113-120.

Leslie, J.F. and B.A. Surnrnerell. 2006. Fusariurn Laboratory Manual. Iowa, USA, Blackwell Publishing.

Lima, M. A., M. d. C. de Oliveira, A. Pimenta and P. Uchôa. 2019. *Aspergillus niger*: A Hundred Years of Contribution to the Natural Products Chemistry. Journal of the Brazilian Chemical Society, 30: 2029-2059.

Liu, L., K. Zhu, N. Wurzburger and J. Zhang. 2020. Relationships between plant diversity and soil microbial diversity vary across taxonomic groups and spatial scales. Ecosphere, 11: e02999.

Magurran, A. E. 1988. Ecological Diversity and Its Measurement. Springer Netherlands.

Martin-Carnahan, A. and S. W. Joseph. 2005. Aeromonadales ord. nov. Bergey's Manual® of Systematic Bacteriology. Springer US, pp. 556-587.

McMichael, A. 2003. Global climate change and health: an old story writ large, Climate change and human health: Risks and responses. World Health Organization (WHO Press).

Meiyana, R.Y., S. Salamiah, S. Soedijo and M.I. Pramudi. 2021. Diversity of Soil Surface Arthropodes on Shallot Plants (*Allium ascalonicum* L.) Applied by Several Botanical Pesticides in Peatlands. International Journal of Biosciences, 19, 73–82.

Merah, T. T. K. B., S. Wiyatiningsih, W. Arit and E. TP. 2009. Tanggapan tujuh kultivar bawang merah terhadap infeksi *Fusarium oxysporum* f. sp. *cepae* penyebab penyakit moler Sri Wiyatiningsihl), Arif Wibowo2) dan Endang Triwahyu Pi). Jurnal Pertanian MAPETA, 12: 1-71.

Momma, K., W. Hashimoto, O. Miyake, H. J. Yoon, S. Kawai, Y. Mishima, B. Mikami and K. Murata. 1999. Special cell surface structure, and novel macromolecule transport/depolymerization system of Sphingomonas sp A1. Journal of Industrial Microbiology and Biotechnology, 23: 425-435.

Moore, N., K. Pegg, I. Buddenhagen and S. Bentley. 2001. Fusarium wilt of banana: a diverse clonal pathogen of a domesticated clonal host. In 'Fusarium: Paul E. Nelson Memorial Symposium'.(Eds BA Summerell, JF Leslie, D Backhouse, WL Bryden, LW Burgess) pp. 212–224. APS Press: St Paul, MN.

Mueller, J. G., P. J. Chapman, B. O. Blattmann and P. H. Pritchard. 1990. Isolation and characterization of a fluoranthene-utilizing strain of *Pseudomonas paucimobilis*. Applied and environmental microbiology, 56: 1079-1086.

Nagata, Y., K. Miyauchi and M. Takagi. 1999. Complete analysis of genes and enzymes for ?-hexachlorocyclohexane degradation in *Sphingomonas paucimobilis* UT26. Journal of Industrial Microbiology and Biotechnology, 23: 380-390.

Netzer, R., P. Peters-Wendisch, L. Eggeling and H. Sahm. 2004. Cometabolism of a nongrowth substrate: L-serine utilization by *Corynebacterium glutamicum*. Applied and environmental microbiology, 70: 7148-7155.

Noerfitryani, N. and H. Hamzah. 2017. The existence of entomopathogenic fungi on rice plants rhizosphere. International Journal of Biosciences and Biotechnology, 5: 12-21.

Nurussakinah 2010. Phytochemical Screening and Antibacterial Activity Test of Peel Extract of Jengkol (*Pithecellobium Jiringa* (Jack) Prain) Plant Against *Streptococcus mutans*, *Staphylococcus aureus*, and *Eschenica coli* bacteria.Skripsi. Fakultas Farmasi USU.

Odum, E. P. and G. W. Barrett. 1971. Fundamentals of ecology. Saunders Philadelphia. Pp. 631-676.

Palmero, D., C. Iglesias, M. de Cara, T. Lomas, M. Santos and J. C. Tello. 2009. Species of *Fusarium* Isolated from River and Sea Water of Southeastern Spain and Pathogenicity on Four Plant Species. Plant Disease, 93: 377-385.

Pinyakong, O., H. Habe, N. Supaka, P. Pinpanichkarn, K. Juntongjin, T. Yoshida, K. Furihata, H. Nojiri, H.

Yamane and T. Omori. 2000. Identification of novel metabolites in the degradation of phenanthrene by *Sphingomonas* sp. strain P2. FEMS Microbiology Letters, 191: 115-121.

Ploetz, R. 2001. Significant diseases in the tropics that are caused by species of Fusarium. Fusarium: Paul E. Nelson memorial symposium. APS Press St. Paul, pp. 295-309.

Purwati, P. and H. Hamidah. 2019. Biodiversitas Mikroba Rizosfer Tanaman Jeruk Keprok Borneo Prima (*Citrus reticulata* cv Borneo Prima). Agrifarm : Jurnal Ilmu Pertanian, 7: 50-53.

Rahmawati, R., D. Hunaefi, I. Basriman, D. Saputra, W. Aozora and B. Jenie. 2019. The characteristics of indigenous yeast mold dried culture using tray dryer. IOP Conference Series: Earth and Environmental Science. IOP Publishing, p. 012036.

Rao, N.S. 1994. Mikroorganism Tanah dan Pertumbuhan Tanaman. Edisi kedua. Jakarta, Penerbit Universitas Indonesia.

Rosita, E., R. Linda and S. Khotimah. 2014. Microbes at different levels of peat maturity in the mount ambawang protection forest area, kubu raya regency. Jurnal Protobiont, 3, 10-16.

Rozali, G. 2015. Screening of Antagonist Fungus Indigenous Rhizosphere Cocoa (*Theobroma cacao* Linn.) Potentially Inhibiting Fungus Growth *Phytophthora palmivora* Butler. Skripsi. Padang, Universitas Andalas.

Salamiah and N. Aidawati. 2022. Microbial diversity of shallot plantation in peat-lands applied with three types of botanical pesticides. IOP Conference Series: Earth and Environmental Science, 976: 012032.

Salamiah, S. 2021. Diversity of Soil Surface Arthropods on Shallot Plants (*Allium ascalonicum* L.) Applied by Several Botanical Pesticides in Peatlands. Lambung Mangkurat University, Campus I Banjarmasin.

Salamiah, S. and N. Aidawati. 2021. Microbial Biodiversity of Shallot Plantation in Peat-lands Applied with Three Types of Botanical Pesticides. IOP Conference Series: ICWEB 2nd.

Saragih, S. 2009. Jenis-jenis Fungi dan Tingkat Kematangan Gambut. Fakultas Pertanian, Universitas Sumatra Utara, Medan (Skripsi).

Saraswati, R. 2007. MMetode analisis biologi tanah. Balai Besar Penelitian dan Pengembangan Sumberdaya Lahan Pertanian.

Schaad, N. W., J. B. Jones and W. Chun. 2001. Laboratory guide for the identification of plant pathogenic bacteria. American Phytopathological society (APS press).

Schoenborn, L., P. S. Yates, B. E. Grinton, P. Hugenholtz and P. H. Janssen. 2004. Liquid serial dilution is inferior to solid media for isolation of cultures representative of the phylum-level diversity of soil bacteria. Applied and environmental microbiology, 70: 4363-4366.

Semangun, H. 2001. Pengantar ilmu penyakit tumbuhan. Yogyakarta: Gadjah Mada University.

Sennang, N.R., E. Syamum and A. Dachlan. 2012. Rice Growth and Production Using Organic Fertilizers and Biological Fertilizers. Jurnal Agrovigor, 11, 61–170.

Sharma, H., A. War and K. Sahrawat. 2012. Botanical pesticides: environmental impact. Studium Press LLC, Texas, USA.

Siswandi, S., R. Astuti and M. Maimunah. 2020. Uji In-Vitro Ekstrak Kulit Jengkol (*Pithecellobium jiringa*) sebagai *Biofungisida terhadap Fusarium oxysporum*, *Colletotrichum capsici*, dan *Cercospora capsici* pada Tanaman Cabai. Jurnal Ilmiah Pertanian, 2: 144-157.

Sørensen, S. R., Z. Ronen and J. Aamand. 2001. Isolation from agricultural soil and characterization of a Sphingomonas sp. able to mineralize the phenylurea herbicide isoproturon. Applied and environmental microbiology, 67: 5403-5409.

Stolz, A. 1999. Degradation of substituted naphthalenesulfonic acids by *Sphingomonas xenophaga* BN6. Journal of Industrial Microbiology and Biotechnology, 23: 391-399.

Suharni, T.T. 2008. General Microbiology. Jakarta, Atma Jaya, pp. 36–43.

Summerell, B. A., B. Salleh and J. F. Leslie. 2003. A Utilitarian Approach to Fusarium Identification. Plant Disease, 87: 117-128.

Suryabhakti, A.K., S. Salamiah and N. Aidawati. 2021. Application of three types of *Trichoderma* sp. origin of tidal swamp land isolates in controlling moler disease caused by *Fusarium oxysporum* f.sp.*cepae* on Shallot. Thesis of the Faculty of Agriculture, Lambung Mangkurat University.

Thom, C. and M. Church. 1926. The Aspergilli. Baltimore, The Williams and Wilkins Company.

Udiarto, B. K., W. Setiawati and E. Suryaningsih. 2005. Pengenalan hama dan penyakit pada tanaman bawang merah dan pengendaliannya. Balai Penelitian Tanaman Sayuran.

Waluyo, L. 2004. General microbiology. UMM, Malang.

White, D. 2007. The Physiology and Biochemistry of Prokaryotes. New York, NY, Oxford University Press, pp. 383–403.

Wittich, R. M., C. Strmpl, E. R. B. Moore, R. Blasco and K. N. Timmis. 1999. Interaction of Sphingomonas and Pseudomonas strains in the degradation of chlorinated dibenzofurans. Journal of Industrial Microbiology and Biotechnology, 23: 353-358.

Wulandari, N. L. D., M. W. Proborini and I. K. Sundra. 2013. Eksplorasi Spasial Cendawan Tanah pada Sekitar Rhizosfer Tanaman Jambu Mete (*Anacardium occidentale* L.) di Karangasem dan Buleleng-Bali. Simbiosis: Journal of Biological Sciences, 1: 2337-7224.

Zacharia, J.T. 2011. Ecological Effects of Pesticides. INTECH Open Access Publisher

Zul, D., S. Denzel, A. Kotz and J. Overmann. 2007. Effects of plant biomass, plant diversity, and water content on bacterial communities in soil lysimeters: implications for the determinants of bacterial diversity. Applied and environmental microbiology, 73: 6916-6929.

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Salamiah, S.	:	Carry out research and write a script from the ideas that have been compiled
Rosa, H.O	:	Doing work in the laboratory and analyzing research data