https://journal.uhamka.ac.id/index.php/bioeduscience/

BIOEDUSCIENCE
ISSN: 2614-1558
http://journal.uhamka.ac.id/index.php/bioeduscience

Potential of *Trichoderma* sp. The Origin of Peat Soil as a Biological Control Agent for Pathogenic Fungi Carried by Rice Seeds

Shinta Maulina Aizah [1*], Arika Purnawati [1], Endang Triwahyu Prasetyawati [1]

[1] Agrotechnology, Faculty of Agriculture, Universitas Pembangunan Nasional University "Veteran", Jl. Rungkut Madya Gunung Anyar, Surabaya, Jawa Timur, Indonesia, 602942
* Correspondence: arika_p@upnjatim.ac.id

Abstract

Background: Rice (*Oryza sativa*) is the world's second most important food crop after wheat. In Indonesia, more than 90% of the population consumes rice, and there is a very high level of dependence, so rice has essential value for the Indonesian population. However, this has not been accompanied by the availability of healthy seeds because many pathogenic fungi are carried by rice seeds, such as Rhizopus sp., Mucor sp., Aspergillus sp., Fusarium sp., Curvularia sp. Seed treatment using synthetic fungicides is an effort to control pathogenic fungi on seeds, but it hurts seeds, humans, and the environment. A safe control alternative is using biological agents, namely the fungus Trichoderma sp. isolated from peatlands and has an antibiosis mechanism. The research aimed to test the potential of the fungus Trichoderma sp. from peat soil as a biological control agent for pathogenic fungi carried by rice seeds (*Oryza sativa*). **Methods:** The test uses the incubation method on PDA media and the growing test method on sterile soil media. The research used a completely randomized design with a factor of 2 isolates of Trichoderma sp. and eight replications and data analysis using BNJ5%. **Results:** isolation results obtained two isolates of Trichoderma sp. (P1 and P2), two isolates of Trichoderma sp. (P1 and P2) were able to suppress the level of pathogenic fungal infections carried by rice seeds in the incubation method using PDA media and the growing on test method, 3.75%, two isolates of Trichoderma sp. able to increase the germination capacity of rice seeds by P1 (12.50%) and P2 (31.25%) respectively. **Conclusions:** Two isolates of Trichoderma sp. (P1 and P2) could suppress the level of pathogenic fungal infections carried by rice seeds in the incubation method using PDA media and in the growing on test method. Apart from that, two isolates of Trichoderma sp.

Keywords: Biological Control Agency; Seed Pathogens; Trichoderma sp

Check for updates

Article history
Received: 23 Jun 2024
Accepted: 20 Dec 2024
Published: 31 Dec 2024

Citation:
Aizah et al., 2024. Potential of *Trichoderma* sp. The Origin of Peat Soil as a Biological Control Agent for Pathogenic Fungi Carried by Rice Seeds. BIOEDUSCIENCE, 8(3), 352-358. doi: 10.22236/jbes/15354

Introduction

Rice (*Oryza sativa*) is the world's second most important food crop after wheat. In Indonesia, more than 90% of the population consumes rice, so rice is essential for the people of Indonesia. National rice productivity, based on data (Badan Pusat Statistik, 2022), Productivity in 2021, namely 54.42 million tonnes, decreased by 233.91 thousand tonnes or 0.43 percent compared to rice production in 2020 of 54.65 million tonnes. This decline in production occurred due to the lack of healthy and quality seeds. Seed health is one factor in seed quality; seed health is often disturbed by pathogenic fungi infecting seeds. Pathogenic fungi carried by rice seeds include Aspergillus sp., Fusarium sp.

Rhizopus sp., Curvularia sp., Pyricularia sp., Alternaria sp., Bipolaris sp., Nigospora sp. Seeds that pathogenic fungi have infected can directly affect plant growth and will become a source of infection in the field (Sobianti *et al.*, 2020). Seed treatment using synthetic fungicides is an effort to control pathogenic fungi in seeds, but it harms seeds, humans, and the environment. A safe control alternative is using biological agents, namely the fungus Trichoderma sp. isolated from peatlands and has an antibiosis mechanism. Apart from that, several studies state that the fungus Trichoderma sp. from peat soil can suppress soft rot disease in aloe vera and the fungus *Ganoderma sp*. (Supriyanto & Sulistyowati, 2011)

This research aims to test the potential of the fungus Trichoderma sp. from peat soil as a control agent for pathogenic fungi carried by rice seeds (*Oryza sativa*).

Methods

Time and Place

The research was conducted at the Plant Health Laboratory, Faculty of Agriculture, UPN "Veteran" East Java, from June to September 2023.

Tools and Instrument materials

Tools used in the research: autoclave, laminar air flow, microscope, vortex, Neubauer type hemocytometer, 250 ml Erlenmeyer, test tube, 1000 ml beaker, petri dish, bunsen, tweezers, tube needle, pipette. Materials used in the research: peat soil samples from Central Kalimantan, rice seeds, Potato Dextrose Agar (PDA) media (Himedia), Potato Sugar Extract (EKG) media, 70% alcohol, sterile distilled water.

Isolation of Trichoderma sp

A 10-gram sample of soil from the peatlands of Central Kalimantan was isolated on PDA media using the sprinkler method and then incubated at room temperature for four days. Purify growing fungal isolates and identify them macroscopically and microscopically.

Preparation of Trichoderma sp. Mushroom Suspension.

A pure isolate of the fungus Trichoderma sp. is made into a suspension used to soak rice seeds using the bio-priming method. The suspension was created using a modified technique, according to (Puspita et al., 2020), using ECG media. The steps are to prepare isolates of Trichoderma sp. seven days old and 50 ml of ECG media in an Erlenmeyer, inserting 30 pieces of 0.5 cm diameter each (for P1 and P2) into the ECG media, then homogenizing using a vortex for five minutes and counting using serial dilution techniques to obtain 106 spores/ml spore density. Spore density was calculated by dripping one ml of the suspension into a Neubauer-type hemocytometer and counted using a binocular microscope with a magnification of 400 times.

Seed Testing (Incubation Method Using PDA Media)

Rice seeds are soaked in 50 ml of *Trichoderma sp.* fungus suspension. for 24 hours (Al-Husyainiyah & Adelia, 2024). In the control treatment, rice seeds were soaked in fungicide containing the active ingredient Benomil 50% (0.02 grams of powder dissolved in 50 ml of water) for 24 hours. Dry the seeds for 5 minutes. Next, 10 seeds were planted on PDA media and incubated at 280C for seven days (Pusat Karantina Tumbuhan, 2007), and the infection, germination, and pathogens growing on the rice seeds were observed.

Seed Testing (Growing on Test Method)

The soil was sterilized using 5% formalin (12.5 ml formalin/5 kg soil) and stirred evenly, then the soil was wrapped in plastic for seven days, after the wrap was opened and transferred to a polybag and then air-dried for seven days (Sianipar et al., 2019). Next, put the soil into a 40x40 cm polybag and 10 rice seeds treated with Trichoderma sp. in seedlings, and maintain by watering every day for 14 days. Observations on the level of infection and germination were carried out 1 day after sowing (DAP) up to 14 DAP.

Data collection

Data was obtained from observations on the seventh day for infection levels, germination, and pathogens growing on rice seeds in seed testing using the PDA media incubation method. The formula used to calculate the infection rate, according to (Pusat Karantina Tumbuhan, 2007), is the following formula:

$$\text{Infection Rate} = n/N \times 100\ \%$$

Information:
n: the number of rice seeds that have colonies of pathogenic fungi growing on them
N: total number of rice seeds tested

The formula used to calculate germination capacity, according to (Pusat Karantina Tumbuhan, 2007), is as follows:

$$\text{Germination power} = JK/JC \times 100$$

Information:
JK = number of regular sprouts produced
JC = number of seed samples tested

Data Analysis

Testing was carried out using a Completely Randomized Design (CRD). The research data obtained were analyzed using R studio software with the ANOVA procedure. If there is a difference at the 5% level, it is tested using BNJ 5%.

Result

Fungal isolate Trichoderma sp.

The results of the isolation of the fungus Trichoderma sp. Two isolates were obtained from the peatlands of Central Kalimantan (Figure 1). The first isolate had light green colonies in the middle and white at the edges. The second isolate showed an even green color throughout the colony. Microscopic observation revealed that the two isolates had several differences. The hyphae of the first isolate were insulated, while those of the second isolate were not insulated (Figure 2).

Infection Rate

The infection rate in the PDA media incubation method showed that the treatment of isolate types did not provide a significant difference in results. Treatment P2 had results that were not significantly different from P1, so the infection levels arising from treatments P2 and P1 were statistically the same (Figure 2). The infection rate using the growing-on test method showed that the treatment of the isolate type did not produce any significant differences in results. Treatment P2 had results that were not significantly different from P1, so the infection rates arising from treatments P2 and P1 were statistically the same (Figures 3 and 4).

Germination Power

Germination power in the PDA media incubation method and the growing on test method showed that the P2 treatment had significantly different results from P1. Germination power from treatment P2 was 31.25%, and from treatment P1 was 12.50%, so isolate Trichoderma sp. 2 (P2) is a better treatment because it has a higher percentage of germination than *Trichoderma sp.*isolates—1 (P1) (Figure 5).

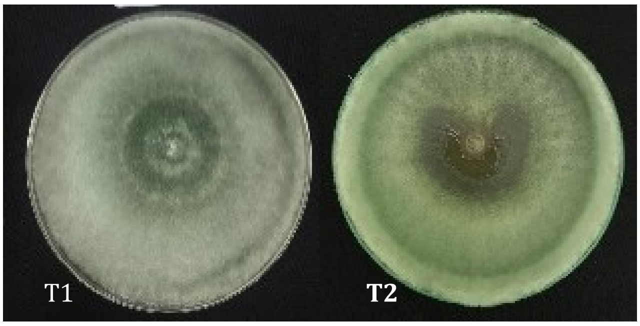

Figure 1. Isolat jamur *Trichoderma* sp.
T1 : isolat *Trichoderma* sp. 1, T2 : isolat *Trichoderma* sp. 2

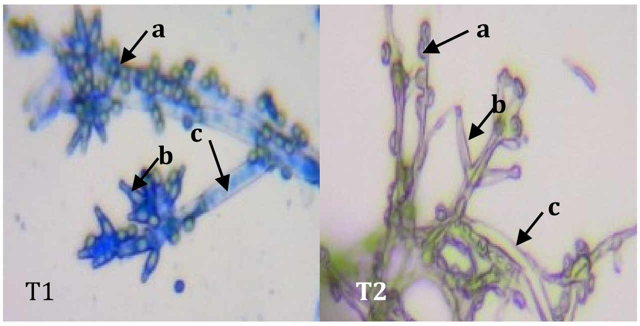

Figure 2. Mikroskopis jamur *Trichoderma* sp. (p. 10x10)
T1 : isolat *Trichoderma* sp. 1, T2 : isolat *Trichoderma* sp. 2 akonidia, b. fialida, c. hifa)

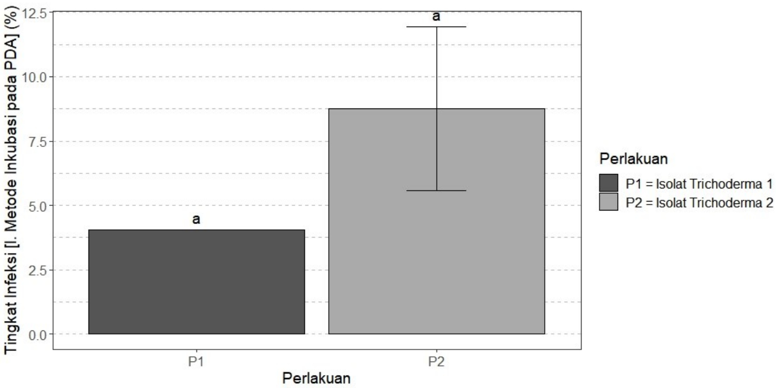

Figure 3. Tingkat infeksi metode inkubasi menggunakan media PDA

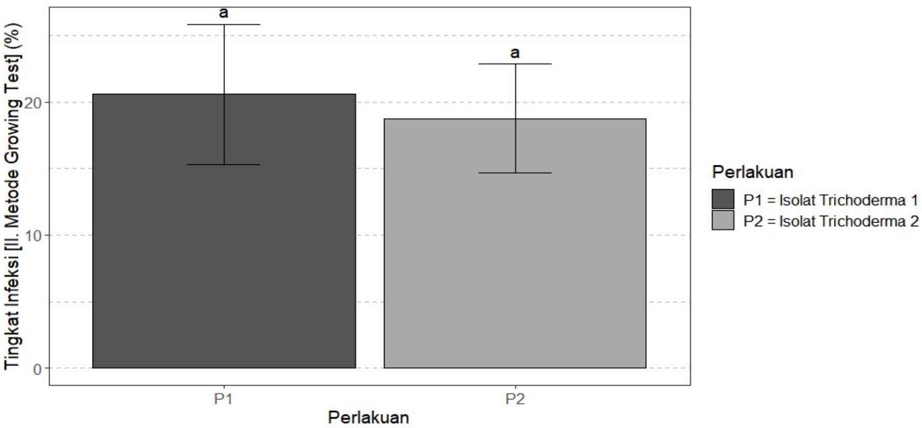

Figure 4. Tingkat infeksi metode growing on test

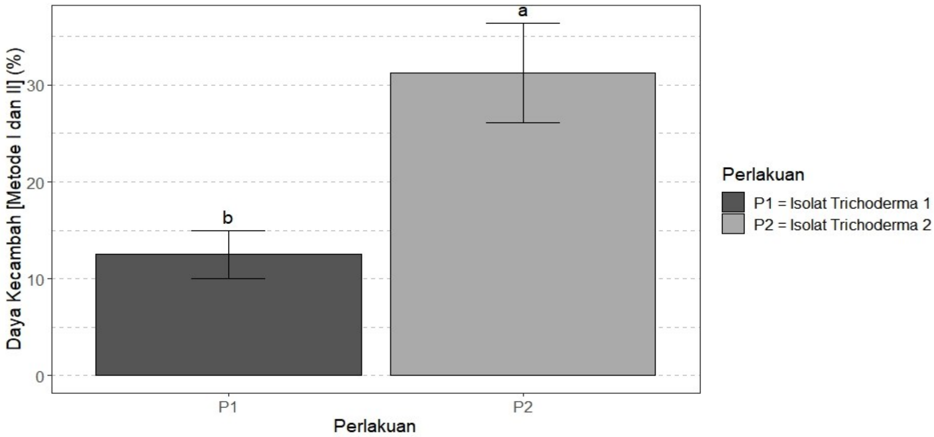

Figure 5. Daya kecambah benih padi pada metode I dan II

Discussion

Fungal isolate Trichoderma sp.

The results of the isolation of the fungus Trichoderma sp. Two isolates were obtained from the peatlands of Central Kalimantan (Figure 1). The first isolate had light green colonies in the middle and white at the edges. The second isolate showed an even green color throughout the colony. According to (Gusnawaty et al., 2014), the period of the color change of *Trichoderma sp.* colonies. Various species of Trichoderma sp. change color from white to green on the seventh day, and some species are green from the age of two days. Although there are variations in color changes, both have predominantly green colonies. These characteristics align with the macroscopic characteristics of the fungal colony Trichoderma sp., identified by (Nadhifah et al., 2016) as green. The two isolates also showed similarities in the smooth, cotton-like colony texture and concentric circular growth pattern. These results are based on research (Suanda, 2016), which noted that isolates of the fungus Trichoderma sp. have fibrous colonies and grow in a circular pattern with clear boundaries.

Microscopic observation showed that the two isolates had several differences. The hyphae of the first isolate were insulated, while those of the second isolate were not insulated (Figure 2). The field shape of the first isolate was short and fat, while the second isolate was elongated and cylindrical. According to (Jumadi *et al.*, 2021), the field shape is a characteristic of the parts of a fungus, especially Trichoderma sp. The field type in terms of shape can be compared between two kinds of *Trichoderma sp.* species. Namely, *Trichoderma pachybasium* has a fat and short fialid shape. Meanwhile, the field shape of *Trichoderma longibrachiatum* is elongated and cylindrical. The conidia shape of the first isolate was round in clusters, while the second isolate was single oval.

Infection Rate

Two isolates of Trichoderma sp. (P1 and P2) had results that did not differ significantly in the level of infection that occurred for the incubation method using PDA media and the growing on test method. Trichoderma sp. has several mechanisms: antibiosis, parasitism, and competition for space and nutrients in vitro and in vivo. Trichoderma sp. can release compounds that are toxic to pathogens such as trihotoxin, gliotoxin, and viridian (Ela et al., 2016)*;* apart from that, it can suppress pathogen attacks through several mechanisms such as antibiosis and parasitism as well as competition for space and nutrients both in vivo and in vitro (Mahabbah et al., 2014). The ability of Trichoderma sp. to inhibit the growth of plant pathogens occurs through mechanisms including microparasites, namely parasitizing the mycelium of other fungi by penetrating the cell wall and entering the cell to take nutrients from the cell so that it dies, antibiosis, competition for nutrients, induction, and plant resistance, dissolving inorganic nutrients and inactivating enzymes. Pathogen (Harman, 2006). Inayati et al. (2019) also stated that Trichoderma sp. produces metabolites that function as antifungals.

Germination Power

Germination power in the incubation method and growing on test method showed that the P2 treatment had significantly different results from P1. Germination power from treatment P2 was 31.25%, and treatment P1 was 12.50%, so isolate Trichoderma sp. 2 (P2) is a better treatment because it has a higher percentage of germination than *Trichoderma sp.*isolates. 1 (P1). This is because the suspended spores of Trichoderma sp. 2 (P2) attach more easily to rice seeds, and seed germination generally begins with water imbibition events, including suspension of Trichoderma sp. In the seeds, the activity of gibberellin acid will break dormancy and stimulate germination. It is also suspected that Trichoderma sp. 2 (P2) plays a more active role in stimulating plant growth hormones and stimulates plant growth by secreting the growth hormones IAA and cytokinins (Sucahyono *et al.*, 2013).

Conclusions

The research results concluded that two isolates of the fungus Trichoderma sp. were obtained from the peatlands of Central Kalimantan. (P1 and P2). Two isolates of Trichoderma sp. (P1 and P2) were able to suppress the level of pathogenic fungal infections carried by rice seeds in the incubation method using PDA media and in the growing on test method, apart from that, two isolates of Trichoderma sp. Also, it can increase rice seeds' germination capacity by P2 (31.25%) and P1 (12.50%), respectively.

Declaration statement

The authors reported no potential conflict of interest.

References

Al-Husyainiyah, T.F., Adelia, E. (2024). Pengujian mutu benih dengan perendaman pgpr terhadap daya berkecambah padi varietas inpari 32 di uptd bpsb-p ntb. *UTS STUDENT CONFERENCE*, *2*(4), 1–8.

Badan Pusat Statistik. (2022). Luas Panen dan Produksi Padi di Indonesia 2022. *Badan Pusat Statistik*, 1–329.

Ela, N., Hakim, L., & Ulim, A. (2016). Efektivitas Beberapa Agen Antagonis dan Cara Aplikasinya Untuk Menekan Pertumbuhan Sclerotium rolfsii pada Tanaman Kedelai (Glycine max L. Merrill). *Jurnal Ilmiah Mahasiswa Pertanian*, *1*(1), 155–167. https://doi.org/10.17969/jimfp.v1i1.1012

Febrianty Sianipar, H., Sijabat, A., & Pane, E. P. (2019). Pengaruh Pemberian Berbagai Tingkat Mikoriza Arbuskula Pada Tanah Terakumulasi Logam Pb terhadap Pertumbuhan Tanaman Belimbing Wuluh (Averrhoa Bilimbi). *Jurnal Biosains*, *5*(2), 53–58. https://doi.org/10.24114/jbio.v5i2.13918

Gusnawaty, H., Taufik, M., Triana, L., & Asniah. (2014). Karakterisasi Morfologis Trichoderma spp. Indigenus Sulawesi Tenggara. *Jurnal Agroteknos*, *4*(2), 88–94.

Harman, G. E. (2006). Overview of mechanisms and uses of Trichoderma spp. *Phytopathology*, *96*(2), 190–194. https://doi.org/10.1094/PHYTO-96-0190

Inayati, A., Sulistyowati, L., Aini, L. Q., & Yusnawan, E. (2019). Antifungal activity of volatile organic compounds from Trichoderma virens. *AIP Conference Proceedings*, *2120*. https://dio.org/10.1063/1.5115750

Jumadi, O., Junda, M., Caronge, M. W., & Syafruddin. (2021). Trichoderma dan pemanfaatan. In *Penerbit Jurusan Biologi FMIPA UNM*.

Mahabbah, A. F., Aeny, T. N., & Maryono, T. (2014). Pengaruh Trichoderma Sp. dan Fungisida Sintetis Terhadap Pertumbuhan Sclerotium Rolfsii dan Keterjadian Penyakit Rebah Kecambah Kacang Tanah. *Jurnal Agrotek Tropika*, *2*(2), 208–214. https://doi.org/10.23960/jat.v2i2.2086

Nadhifah, Y., Hastuti, U., & Syamsuri, I. (2016). Mikoflora Dari Rizosfer Tanah Pertanian Tebu (Saccharum Officinarum L .) Sebagai Bahan Ajar Kingdom Fungi Untuk. *Jurnal Pendidikan*, *1*(1974), 2023–2030.

Pusat Karantina Tumbuhan. (2007). Pedoman Diagnosis Organisme Pengganggu Tumbuhan Karantina Golongan Cendawan. In *Badan Karantina Pertanian*. Pusat Karantina Tumbuhan.

Puspita, F., Ali, M., & Supriyadi, S. (2020). Kompatibilitas dan Daya Hambat Konsorsium Trichoderma spp. Endofit terhadap Penyakit Busuk Buah Kakao Phytophthora palmivora. *Agrikultura*, *31*(2), 126. https://doi.org/10.24198/agrikultura.v31i2.26063

Sobianti, S., Soesanto, L., & Hadi, S. (2020). Inventarisasi Jamur Patogen Tular-Benih pada Lima Varietas Padi. *Agro Bali : Agricultural Journal*, *3*(1), 1–15. https://doi.org/10.37637/ab.v3i1.416

Sucahyono, D., Sari, M., Surahman, M., Satriyas Ilyas, D., & Penelitian Tanaman Kacang-kacangan dan Umbi-umbian Jl Raya Kendal Payak Km, B. (2013).) 126 Pengaruh Perlakuan Invigorasi pada Benih Kedelai Hitam (Glycine soja) terhadap Vigor Benih, Pertumbuhan Tanaman, dan Hasil Effect of Invigoration Applied on Black Soybean (Glycine soja) Seed on Seed Vigor, Plant Growth, and Yield. *J. Agron. Indonesia*, *41*(2), 126–132. https://doi.org/10.24831/jai.v41i2.7517

Supriyanto, & Sulistyowati, H. (2011). Pengembangan PGPF Menjadi Pupuk dan Pestisida Hayati. *Jurnal Perkebunan Dan Lahan Tropika*, *1*, 19–27. https://doi.org/10.26418/plt.v1i1.28.