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ASSESSING THE EFFECT OF *TRICHODERMA* SPP. ON THE GROWTH OF WETLAND AND DRYLAND RICE CULTIVARS AT VEGETATIVE STAGE

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Abstract

The use of biological fertilizers can reduce farmer’s dependence on chemical fertilizers which are known to pollute the environment. The most widely used and commercially produced fungal group for biological fertilizers is *Trichoderma* spp. This study aims to determine the effect of the application *Trichoderma* spp. on the growth of gogo rice and paddy rice plants in the early vegetative phase. The pot study was conducted at the Biology Laboratory, State University of Padang in 2020. This research was an experimental study using completely randomized design with 8 treatments and 4 replications. Parameters observed were shoot length, root length, fresh weight, dry weight, and plant height. The rice seedlings were inoculated with the suspension *Trichoderma* 10 mL at a density of 10^7 spores/mL for 24 hours. Measurements of shoot length, root length, fresh weight and dry weight were carried out when the rice plants were two weeks old and plant height was measured after the rice was eight weeks old. Gogo rice plant response to administration *Trichoderma* isolate SB_Solok showed significant performance for root length parameter on AA75 variety, but not significant for other parameters. Paddy rice plant response to administration *Trichoderma* isolate TS_Solok only increased significantly for the wet weight parameter of the Batang Sungkai variety. *Trichoderma* isolates KRT_PasBar and SBT_PasBar only significantly increased the plant height parameter for the Batang Sungkai variety. The research results also show the application of all isolates *Trichoderma* can improve all the parameters observed in lowland rice of the Batang Sungkai variety.

Key words: Paddy rice, Gogo rice, *Trichoderma*, Growth.

Introduction

The application of biological fertilizers can reduce farmers dependence on the use of chemical fertilizers which are known to be environmentally unfriendly (Nicolopoulou-Stamati *et al*., 2016; Kumar & Singh, 2015). Biofertilizers are also biological pest control agents, which have the ability to influence physiological processes, increase growth, and make plants more tolerant of biotic and abiotic stresses (Dardanelli *et al*., 2010a, 2010b; Berendsen *et al*., 2012; Matthews *et al*., 2014). Biofertilizers consist of living organisms (bacteria and fungi) or plant extracts, and cellular products (fatty acids or pheromones) found in nature (Mehrotra *et al*., 2017).

The most widely used and commercially produced biological fertilizer from the mushroom group is *Trichoderma* spp. or free living non-pathogenic fungi colonizing the roots of many plants, as plant symbionoportunistic (Harman *et al*., 2004; Hermosa *et al*., 2013). *Trichoderma* has great variability resulting in strains having various biostimulant actions and antagonistic activities (Colla *et al*., 2015; Contreras-Cornejo *et al*., 2015; López-Bucio *et al*., 2015).

Several *Trichoderma* species have been shown to exert different enhancing effects on plants (Hohmann *et al*., 2012). Giving *Trichoderma* spp. able to increase plant growth, development, and productivity. Specifically, the fungal application *Trichoderma* in tomato plants can increase plant resistance to abiotic stress, nutrient uptake and use, and phosphorus uptake (Saba *et al*., 2012). Application *T. harzianum* in cucumber also showed a significant increase in the accumulative root length, root surface area, number of root tips, dry weight, shoot length, leaf area, and concentration of microelements (Akladious dan Abbas, 2012; Khan *et al*., 2005).

Inoculation of rice plants with *T. asperellum* SL2 markedly increased plant height, root length, fresh weight, number of leaves, and plant biomass of rice compared to control (Doni *et al*., 2017). Application *T. harzianum* can also increase root length, dry weight, and growth of rice seedlings (Akladious dan Abbas, 2012; Khan *et al*., 2005).

Although application *Trichoderma* in rice plants has a positive effect on plant growth, but there is still a lack of information about the response of rice plants grown in dry land (gogo rice) and wet land (paddy rice) to application *Trichoderma* spp. In Indonesia, efforts to increase rice production are directed at both types of agroecosystems. This study aims to determine the effect of the application *Trichoderma* on the growth of lowland rice and upland rice plants in the early vegetative phase.

Material and Methods

The pot research was conducted from July 2020 to November 2020 in the biology laboratory of the Faculty of Mathematics and Natural Sciences, Padang State University. This study was an experimental study using randomized completely design (CRD) with 8 treatments and 4 replications. Soil planting medium is taken in the plantation area and as much as 500 grams in each pot.

Isolates *Trichoderma* spp.: A total of eight fungal isolates previously identified as *Trichoderma* spp. with classical phenotype testing (Syahputra *et al*., 2017) was used in this study. In this research, a total of eight isolates of *Trichoderma* were isolated and identified from rice fields in West Sumatra. Morphologically, *Trichoderma* was identified based on the color and shape of its hyphae in PDA media. *Trichoderma* colonies on PDA media initially appeared white, but the hyphae gradually changed to greenish-green. The green color of *Trichoderma* colonies and the filamentous shape of its hyphae are distinctive characteristics of *Trichoderma* fungi. In addition to colony shape, *Trichoderma* was identified based on the shape of its conidiophores and conidia. *Trichoderma* possesses conidiophores with branches and conidia that are either round or elongated, along with cylindrical or hook-like fialids at the tip of the conidiophores (Abdullah *et al*. 2021). The successfully isolated morphological characteristics of *Trichoderma* are listed in (Table 1).

Inoculum preparation *Trichoderma*: Spore *Trichoderma* adult (dark green) which will be used as an inoculum, put into a test tube containing 10 mL of distilled water. The spore density is less than cells/mL. The spore density was calculated using the formula (Doni *et al*., 2017):

$$\text{Spore density} = \frac{\text{Number of conidia x 5 x dilution factor}}{\text{Volume haemocytometer}}$$

Preparation of rice seed treatment with *Trichoderma*: Isolate *Trichoderma* tested on four rice varieties (Table 2). A total of 50 seeds of each rice variety, which had been selected and sterilized, were soaked with suspension *Trichoderma* 10 mL (density 10^7 spores/mL) for 24 hours (Doni *et al*., 2018). For the control treatment, the seeds were soaked with 10 mL of distilled water for 24 hours. Seeds of paddy rice varieties were sown and transferred to pots after they were two weeks old, while upland rice seeds were immediately grown on soil media in pots. This experiment was conducted in a glasshouse at the Department of Biology, State University of Padang with natural glasshouse setting (29 ± 6°C) and normal daylight.

Table 1. Origin of isolates *Trichoderma* used in research.

No.	Isolate name	Rice varieties	Isolation place	Rice type
1.	KRT_PasBar	Ketan Rancong Tolang	Gunuang Tuleh, Pasaman Barat	Gogo Rice
2.	SL_PasBar	Silampung		
3.	SBT_PasBar	Situ Bagendit	Labuah Luruih Batang Saman, Pasaman Barat	
4.	RE_Solok	Remaja	Lembah Gumanti, Solok	Paddy rice
5.	TS_Solok	Talang Surian		
6.	SRU_Solok	Sirandah Umbilin		
7.	SB_Solok	Cisokan Balang	Gunung Talang, Solok	
8.	SRBA_Solok	Sirandah Batuampa	Talang Babungo, Solok	

Tabel 2. Gogo rice and paddy rice varieties tested.

No.	Variety	Information
1.	Mekonga	Gogo Rice
2.	AA 75	Local gogo rice
3.	Batang Sungkai	Local paddy fields
4.	Saganggam	Local paddy fields

Measurement of vegetative parameters: Parameters measured were shoot length, root length, plant height, fresh weight and dry weight of plants. The length and weight of the plants were measured after the plants were four weeks old, while the height of the plants after the plants were eight weeks old.

Data analysis

All data were analyzed statistically using tests Analysis of Variances (ANOVA). The significance of the treatment effect was determined by the DMRT follow up test (*Duncan's New Multiple Range Test*) at the level of 5%.

Result and Discussion

Application *Trichoderma* spp. on the growth of gogo rice and paddy rice in the early vegetative phase showed varying performance. In general application *Trichoderma* spp., on gogo rice seeds did not significantly increase plant growth in the early vegetative phase. Only three isolates increased one of the observed parameters. RE_Solok, SB_Solok, and SRU_Solok isolates respectively increasing shoot length of gogo rice variety Mekonga, and root length and plant height of upland rice variety AA75. Specifically, application *Trichoderma* sp., in paddy rice can only increase plant height in the Batang Sungkai variety. The three influential isolates are KRT_PasBar, SBT_PasBar, and SRBA_Solok. Effect of isolating *Trichoderma* spp., on the growth of gogo rice and paddy rice at the beginning of the vegetative phase can be seen in (Tables 3 & 4).

This study also looked at the effect of the isolate application *Trichoderma* on the wet weight and dry weight of rice plants. The results showed that there were differences in the response of each rice variety to the administration *Trichoderma* spp. The results also showed that only the lowland rice of the Batang Sungkai variety showed an increase in wet weight with application treatment *Trichoderma* (Fig. 1). Besides that, the isolate application *Trichoderma* also increased the dry weight of the Batang Sungkai variety compared to the control (Fig. 2). Isolate application *Trichoderma* increased the wet weight and dry weight of the tested rice varieties although with different levels.

According to Korolev *et al*., (2008), application *Trichoderma* inconsistent effect on plant growth. This is possible because plants and microorganisms integrate with each other through various signaling mechanisms (Cai *et al*., 2013). Signaling molecules can have a positive or negative influence on interactions. The success of the interaction depends on signals that initiate the process, then signals that maintain and control the interaction (Halverson and Stacey, 1986; Contreras-Cornejo *et al*., 2016).

Table 3. Influence of giving *Trichoderma* spp., to shoot length (PT), length root (PA), and plant height (TT) of gogo rice varieties Mekonga and AA75 pot experiment.

No.	Isolate treatment *Trichoderma*	Shoot length (cm)	Root length (cm)	Plant height (cm)
	Mekonga variety			
1.	Control	21,80 ± 1,91^{a}	2,95 ± 1.24^{a}	62,18 ± 3.02^{ab}
2.	KRT_PasBar*	21,15 ± 1,91^{a}	3,25 ± 1.24^{a}	66,60 ± 3.02^{ab}
3.	SL_PasBar*	15,70 ± 1,91^{a}	6,78 ± 1.24^{a}	58,48 ± 3.02^{a}
4.	SBT_PasBar *	21,63 ± 1.91^{a}	3,83 ± 1.24^{a}	61,33 ± 3.02^{ab}
5.	RE_Solok**	20,95 ± 1.91^{a}	4,88 ± 1.24^{a}	67,75 ± 3.02^{b}
6.	SB_Solok**	21,10 ± 1.91^{a}	4,23 ± 1.24^{a}	64,05 ± 3.02^{ab}
7.	SRBA_Solok**	19,28 ± 1.91^{a}	5,55 ± 1.24^{a}	63,30 ± 3.02^{ab}
8.	TS_Solok**	21,10 ± 1.91^{a}	5,63 ± 1.24^{a}	61,88 ± 3.02^{ab}
9.	SRU_Solok**	21,03 ± 1.91^{a}	5,25 ± 1.24^{a}	59,63 ± 3.02^{ab}
	AA75 variety			
1.	Control	31,55 ± 1.40^{a}	10,00 ± 1.08^{a}	100,35 ± 5.51^{a}
2.	KRT_PasBar*	35,85 ± 1.40^{a}	11,00 ± 1.08^{a}	115,85 ± 5.51^{c}
3.	SL_PasBar*	34,50 ± 1.40^{a}	11,25 ± 1.08^{a}	104,88 ± 5.51^{abc}
4.	SBT_PasBar *	35,73 ± 1.40^{a}	11,55 ± 1.08^{a}	113,58 ± 5.51^{bc}
5.	RE_Solok**	35,25 ± 1.40^{a}	10,03 ± 1.08^{a}	112,38 ± 5.51^{abc}
6.	SB_Solok**	35,38 ± 1.40^{a}	13,10 ± 1.08^{a}	105,03 ± 5.51^{abc}
7.	SRBA_Solok**	34,25 ± 1.40^{a}	12,25 ± 1.08^{a}	101,45 ± 5.51^{bc}
8.	TS_Solok**	33,05 ± 1.40^{a}	12,08 ± 1.08^{a}	109,83 ± 5.51^{abc}
9.	SRU_Solok**	34,98 ± 1.40^{a}	10,25 ± 1.08^{a}	104,90 ± 5.51^{abc}

*) Isolated from gogo rice rhizosphere
**) Isolated from paddy rice rhizosphere
Aligned numbers followed by the same letter are not significantly different according to the DMRT test at the 5% level

Table 4. Influence of giving *Trichoderma* spp., to shoot length (PT), length root (PA), and plant height (TT) of paddy rice varieties of Saganggam and Batang Sungkai. Pot experiment.

No	Treatment	Shoot length (cm)	Root length (cm)	Plant height (cm)
	Saganggam variety			
1.	Control	41,18 ± 1.42^{a}	8,13 ± 1.19^{a}	135,50 ± 2.68^{a}
2.	KRT_PasBar*	41,98 ± 1.42^{a}	8,38 ± 1.19^{a}	134,38 ± 2.68^{a}
3.	SL_PasBar*	45,45 ± 1.42^{a}	9,75 ± 1.19^{a}	139,7± 2.683^{a}
4.	SBT_PasBar *	44,95 ± 1.42^{a}	7,25 ± 1.19^{a}	141,20 ± 2.68^{a}
5.	RE_Solok**	43,20 ± 1.42^{a}	6,08 ± 1.19^{a}	140,83 ± 2.68^{a}
6.	SB_Solok**	45,00 ± 1.42^{a}	6,13 ± 1.19^{a}	140,15 ± 2.68^{a}
7.	SRBA_Solok**	43,98 ± 1.42^{a}	7,38 ± 1.19^{a}	141,75 ± 2.68^{a}
8.	TS_Solok**	43,30 ± 1.42^{a}	6,88 ± 1.19^{a}	138,05 ± 2.68^{a}
9.	SRU_Solok**	43,68 ± 1.42^{a}	6,55 ± 1.19^{a}	136,53 ± 2.68^{a}
	Batang Sungkai variety			
1.	Control	31,55 ± 1.40^{a}	10,00 ± 1.08^{a}	100,35 ± 5.51^{a}
2.	KRT_PasBar*	35,85 ± 1.40^{a}	11,00 ± 1.08^{a}	115,85 ± 5.51^{c}
3.	SL_PasBar*	34,50 ± 1.40^{a}	11,25 ± 1.08^{a}	104,88 ± 5.51^{abc}
4.	SBT_PasBar *	35,73 ± 1.40^{a}	11,55 ± 1.08^{a}	113,58 ± 5.51^{bc}
5.	RE_Solok**	35,25 ± 1.40^{a}	10,03 ± 1.08^{a}	112,38 ± 5.51^{abc}
6.	SB_Solok**	35,38 ± 1.40^{a}	13,10 ± 1.08^{a}	105,03 ± 5.51^{abc}
7.	SRBA_Solok**	34,25 ± 1.40^{a}	12,25 ± 1.08^{a}	101,45 ± 5.51^{bc}
8.	TS_Solok**	33,05 ± 1.40^{a}	12,08 ± 1.08^{a}	109,83 ± 5.51^{abc}
9.	SRU_Solok**	34,98 ± 1.40^{a}	10,25 ± 1.08^{a}	104,90 ± 5.51^{abc}

*) Isolated from gogo rice rhizosphere
**) Isolated from paddy rice rhizosphere
Aligned numbers followed by the same letter were not significantly different according to the DMRT test at the 5% level

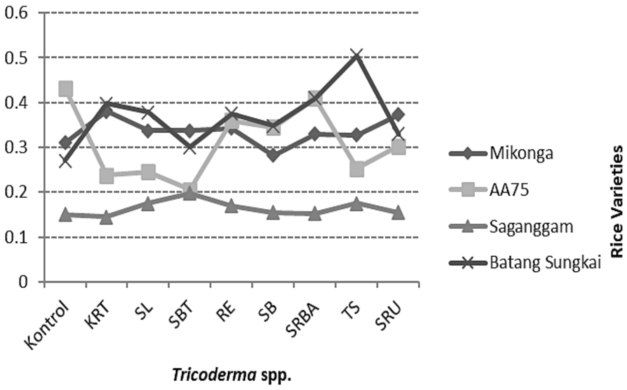

Fig. 1. Effect of giving *Trichoderma* spp., to plant wet weight (gram/pot) gogo rice and paddy rice.

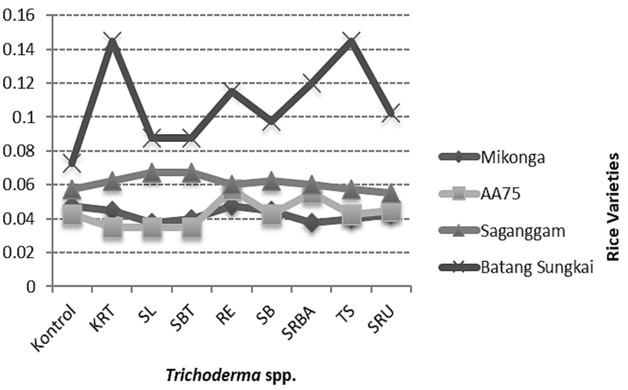

Fig. 2. Effect of giving *Trichoderma* spp., on dry weight (gram/pot) of plants gogo rice and paddy rice.

The ability to enhance plant growth may be an important character of the isolate *Trichoderma* spp. but did not occur in every species tested. Increased plant growth is evidenced by increased productivity, nutrient uptake, biomass, plant resistance to stress, and increased plant health (Azis & Rahmatullah, 2002). *Trichoderma* spp. reproduce in the rhizosphere to form symbiotic associations, thus increasing nutrition and plant growth naturally by producing phytohormones, including indole-3-acetic acid (IAA), gibberellins (GA), and cytokinins (CK) (Nusaibah & Musa, 2019). Enhancement of plant growth by IAA or auxin analogs (Vinale *et al*., 2014) usually exhibits optimal activity at low concentrations (<10 ppm) and an inhibitory effect at higher doses.

The auxin produced by *Trichoderma* can increase the total root length and number of host plant root tips (Cai *et al*., 2013). GA has not been widely studied in plantmicrobe interactions, but the results of the Guzmán-Guzmán *et al*., (2019) showed that the application of *Trichoderma* could increase plant growth with increasing GA concentrations. thereby increasing plant biomass. Cytokinins are hormones involved in plant cell differentiation, root and shoot branching, stress tolerance, and nutritional balance (Müller & Sheen, 2007; Guzmán-Guzmán *et al*., 2019).

Trichoderma has also been reported to interact with rice plants at the molecular level (Harman *et al*., 2021). This intricate process involves the release of specialized signaling molecules by *Trichoderma*, setting off a cascade of defense-related genes in the rice plant which lead to the alteration of rice resistance against harmful pathogens and a substantial improvement in the overall growth and fitness of the rice plant (Doni *et al*., 2022). These captivating molecular interactions offer tremendous potential for sustainable agriculture, presenting an eco-friendly solution to safeguard crops and boost productivity without relying on harmful agrochemical products (Cheng *et al*., 2023).

In addition, *Trichoderma* also possesses remarkable biofertilizer properties (Akbari *et al*., 2023). Acting as a biofertilizer, *Trichoderma* plays a pivotal role in promoting plant growth by enhancing nutrient uptake, especially phosphorus, and facilitating the release of nutrients bound in the soil (Doni *et al*., 2019; Abdullah *et al*., 2021). Harnessing the biofertilizer potential of *Trichoderma*, farmers can reduce their dependence on chemical fertilizers, minimize environmental impacts, and embrace sustainable agricultural practices, leading to elevated crop yields and healthier ecosystems.

Conclusion

The lowland rice of the Batang Sungkai variety showed an increase in plant wet weight and dry weight with the application treatment *Trichoderma* compared to controls with the increment more than 50%. Effect of isolate application *Trichoderma* spp. the tested varieties of gogo rice and paddy rice were not always consistent, except for paddy rice of the Batang Sungkai variety. Isolate KRT_PasBar, RE_Solok, SRBA_Solok, and TS_Solok was more able to improve the growth performance of paddy rice of the Batang Sungkai variety.

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