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Research Article

Potency of six isolates of biocontrol agents endophytic *Trichoderma* against fusarium wilt on banana

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Abstract: Fusarium wilt caused by *F. oxysporum* f.sp. *cubense* is one of very damaging banana plant diseases which can cause plant death. Disease control using intensive chemical fungicides will have negative impacts on the environment and humans. Endophytic *Trichoderma* is one of the biological control agents which can reduce the amount of inoculum of pathogens, so it can reduce disease intensity. The objectives of this study was to assess the ability of endophytic *Trichoderma* in inducing plant resistance against fusarium wilt. Endophytic *Trichoderma* was obtained from healthy roots of banana from three regencies in Yogyakarta, namely *Trichoderma harzianum*.swn-1, *T. harzianum*.swn-2, *T. harzianum*.psr-1, *T. asperrellum*, *T. gamsii*, and *T. koningiopsis*. Research on induced resintance was conducted in the greenhouse with polybag using Completely Randomized Design with 14 treatments and 3 replications. The results showed that the ability of *Trichoderma gamsii* antagonism against *F. oxysporum* f.sp. *cubense* was 60.61%. *T. asperellum and T. harzianum*.swn-2 could suppress this disease resulted in disease intensity of 8.33% which categorize as resistant. *Trichoderma harzianum.psr-1* was significantly different in stimulating plant vegetative growth. Induced resistance by using endophytic *Trichoderma* spp. against *F. oxysporum* f.sp. *cubense* showed increase in total phenolic compounds on the third and fourth weeks as well as peroxidase activity on the third, fourth and fifth weeks. Observation of lignification on the fifth week showed that lignification occurred in root xylem.

Keywords*: banana, endophytic Trichoderma, fusarium wilt, induced resistance*

Introduction

Efforts to increase the yield and quality of banana have several problems, one of which affects the growth and production of banana is fusarium wilt caused by *Fusarium oxysporum* f.sp. *cubense* (Foc). Fusarium wilt is found in almost all banana production centers, and is one of very detrimental diseases in the tropical and subtropical regions. Since the 19th century, in a period of 50 years in Central and South America more than 50.000 ha of banana plantation infected by *F. oxysporum* f.sp. *cubense*. In Indonesia, the disease was reported since 1980 and is quickly spreading in some provinces (Ploetz, 2000).

F. oxysporum f.sp. *cubens* is a soil borne pathogen that can last for decades in different types of soil as mycelium or chlamydospores without host plants. Almost all banana cultivars are highly susceptible to fusarium and cause death (Ploetz, 2006). Pathogen infects roots through root tip and lateral roots, further into the core and the vascular tissue. Direct penetration is uncommon in banana plants, unless there is a wound that will expand the infection and disease intensity. Fusarium can also penetrate through the hair root or epidermal cells near the root hood, the rear of root tip and in the root elongation zone. The spread of hyphae occur from one vascular tissue to another vascular tissue through the holes in the walls of xylem vessels (Ploetz, 2000; Agrios, 2005). Since the 2008-2013 banana production fluctuated, and in 2009-2013 the

production decreased from 6.373.533 tons to 5.359.115 tons, while there were 13.41% decreased in production between 2012 to 2013 (Anonimous., 2015).

To prevent the decline in production, the control of fusarium wilt is needed. Some control techniques that can be done such as surface sterilization using formaldehyde and copper sulfate (Moore et al., 1993), fertilization containing NO_3-N to affect soil pH (Peng et al., 1999), utilization of chemical compound phosphonat and benomyl (Davis et al., 1994), and the use of nonpathogenic Fusarium (Nel et al., 2006) . Utilization of systemic fungicide intensively will be ineffective, since it had some following negative impacts such as environmental pollution that resulted in the destruction of antagonists microorganisms, resistant of pathogen that will bring new races of the pathogen which is more virulent, and the presence of pesticide residues in plants that can affect human health (Agrios, 2005). In addition to that, fungicide price is quite expensive in the market, so that production costs will increase. Therefore, alternative such as biological control methods is needed.

One of endophytic fungi that have been found and act as biological control agents is *Trichoderma* spp. (Harman et al., 1989; Bailey et al., 2006; 2008). This fungus can suppress plant diseases especially soil borne pathogen through parasitism, competism and antibiosis which can directly enhance plant growth as well as immune response against disease (Holmes et al., 2004; Samuels, 2006; Hanada et al., 2008).

The objective of this study was to assess the ability of isolates of endophytic *Trichoderma* against fusarium wilt disease and to learn control mechanism using endophytic *Trichoderma* as biological control agents.

Materials and Methods

Plant materials, endophytic Trichoderma isolate and Foc inoculum

The plant material used in this study was 2 month old tissue culture cultivar of the Ambon Kuning banana, planted in (30 x 35 cm) polybag in sterile soil with the ratio of manure 1:1 (v/v). Seven day old of six isolates of endophytic *Trichoderma* that had been molecularly identified, namely *T. harzianum.swn-1, T. harzianum.swn-2, T. harzianum.psr-1, T. asperellum, T. gamsii,* and *T. koningiopsis* in PDA medium, were propagated on corn, whereas isolates of *Fusarium oxysporum* f.sp. *cubense* (isolates A-13), is a collection of Laboratory of Clinical Plant Pathology, Faculty of Agriculture, Gadjah Mada University, propagated in the PDB (Potato Dextrose Broth) medium with a density of 10^6 conidium/mL sterile distilled water.

Screening of endophytic Trichoderma that antagonistic to Foc

Antagonistic ability of endophytic *Trichoderma* in vitro was tested using *Fusarium oxysporum* f.sp. *cubense* (Foc) A-13 with dual technique culture. Parameter observed was the percentage of inhibition of radial growth on PDA medium. *Trichoderma* isolates of 4 days old in PDA was marked with a cork drill (5 mm), taken using a needle preparations and placed 2 cm from the edge of the petri dish containing PDA medium. Foc of the same size were placed 2 cm from the edge of the same petri dish in contrary of the antagonist. As a control Foc was grown on PDA medium without *Trichoderma.* Antagonistic activity was calculated after 2 days of incubation (Rahman et al., 2009; Ahmed and Lee, 2008).

Percentage Inhibition of Foc Radial Growth was calculated using following formula developed by Skidmore Rahman et al., (2009):

$$\text{PIRG} = \frac{R_1 - R_2}{R_1} \text{ x } 100\%$$

where :

PIRG = Percentage Inhibition of radial growth

R_1 = Radial colony of Foc in the control treatment

R_2 = Radial colony of Foc on treatment with *Trichoderma*

Induced resistance

This research was conducted in the greenhouse of Faculty of Agriculture, University of Gadjah Mada, using a completely randomized design (CRD), with the following treatments: A = endophytic *Trichoderma* isolates, F = *F. oxysporum* f.sp. *cubense*, with 14 levels of treatments and three replications of each, namely A0F0: control ; A0F1: without *Trichoderma* isolates and Foc inoculum A1F1: *T. harzianum.* swn-1 isolate with Foc inoculum; A2F1: *T. harzianum*. swn-2 isolate with Foc inoculum; A3F1: T. asperellum isolate with Foc inoculum; A4F1: *T. harzianum* isolate with Foc inoculum ; A5F1: *T. gamsii* isolate with Foc inoculum; A6F1: *T. koningiopsis* isolate with Foc inoculum.

Disease intensity

Observation of Disease Intensity was done from first symptoms by looking at the wilting

symptoms appear on the leaves. The diseases intensity was calculated using the formula from Wibowo (2002) which has been modified with wilting index (Table 1).

Table 1.Index of wilt banana with modification

Score	Note
0	No yellow leaf/healthy plant
1	1-2 yellow leaves (wilt)
2	3 yellow leaves (wilt)
3	4 yellow leaves (wilt)
4	More than 4 yellow leaves (wilt) ordie plant

Disease Intensity (DI), calculated using the following formula:

$$DI = \frac{\sum(n \times v)}{N \times Z} \times 100\%$$

where :

DI	:	Disease Intensity
n	:	number of plant with v score
v	:	certain score
N	:	number of plant tested
Z	:	the highest score

Plant resistance was determined based on disease intensity scoring refer to rules of Wibowo (2002).

Disease Intensity (%)	Resistance Criteria
≤25	Resistant
25≤IP≥50	Moderate
≥50 or die	Susceptible

Data collected` was analyzed with ANOVA and the significantly different treatment was tested using DMRT (Duncan Multiple Range Test) at 95% level of significant.

The role of endophytic Trichoderma as plant growth promoting fungi

The following parameters were observed on plant vegetative growth, i.e. plant height, stem diameter, number of leaf, plant fresh weight, root fresh weight and root length.

Detection of induced resistance

Total phenolic analysis

The roots of plants that have been induced resistance by endophytic *Trichoderma* isolates were analyzed for total phenol compounds, using methods Aberouman and Deokule (2008). The roots were taken 1,2, 3,4, and 5 weeks after treatment. Five gram of roots were taken randomly and grounded with a mortar sterile. The extract dissolved in 10 mL of 80% methanol, and homogenized with vortex. Banana root extract was made twice and centrifuged at 1500 rpm for 10 minutes and the supernatant was taken. 0.5 mL supernatant was put into a 10 mL test tube, then 2.5 mL of Folin-Ciocalteu reagent and 2 mL of 7.5% sodium carbonate were added. After completely mixed, absorption value was observed at a wavelength of 765 nm using a UV-Vis spectrophotometer PC 1650, and as a blank used 80% methanol. Standard curve was measured using tannin with the following concentrations 100, 200, 300, 400, 500, 600, 700, 800, 900, and 1000 ppm in 30% methanol. Total phenolic compounds calculated from the regression equation of the relationship between the absorption (absorbance) at a concentration of standard solution.

Peroxidase activity analysis

Peroxidase activity was analyzed using the roots of induced resistance plant. Roots of plants were taken 1,2, 3, 4, and 5 weeks after treatments. Five g banana roots added with 25 mL of sodium phosphate buffer (0:01 M, pH = 6) and smoothed with mortar sterilized at a temperature of 4ºC, filtered through Wathman 40 paper. 15 mL tissue extracts centrifuged at 5000 rpm for 5 min at 4ºC. The supernatant was stored at a temperature of -20ºC. Colorimetric test for enzymatic activity performed using a spectrophotometer. Reagent consists of 10 mL pirogallol 0.5 M and 12.5 mL of 0.sodium phosphate buffer 0.066 M in 100 ml of distilled water. To start the reaction 0.5 mL of 1% H_2O_2 was added into 1.5 mL reagent and 0.2 mL of the supernatant was added into the sample cuvette and the absorbance read for 3 minutes and measured using a spectrophotometer (Spectronic 21) at 420 nm, reading from zero. Peroxidase enzyme activity (units per gram) was shown in the change of absorbance of a mixture of roots per gram per minute in each sample (Saravan et al., 2004).

Observation of lignification

Roots of induce resistance plant were taken to observe lignification. The roots of banana used were 5 weeks after treatments. The roots were cleaned with water, and cut 1 cm, fixed in FAA solution (Formalin Acetic Acid Alcohol) for 24 hours. The thin crosswise preparate was made using microtome. Preparates were painted with a solution of 0.1 g of phloroglucinol in 10 mL of 95% ethanol, and then drops of HCl 25%, after 15 minutes, the preparations were observed under a microscope. Root epidermal tissue containing lignin will be purplish red (absorbs phloroglucinol-HCl) (Clark, 1981).

Result and Discussion

Screening of endophytic Trichoderma antagonist to Foc

The inhibition ability of endophytic *Trichoderma* on the growth of *F. oxysporum* f.sp. *cubense* (Foc) (Fig.1) showed that colony of *T. asperillum* (isolates Ksn) and *T. harzianum*.psr-1 (isolate Psr-1) where meet with Foc, and showed inhibition zones (arrow). Colony growth of both species of *Trichoderma* clearly suppress and go over the limit of Foc. This proved that *Trichoderma* was able to suppress growth of Foc (Bae et al., 2011; Mulaw et al., 2013).

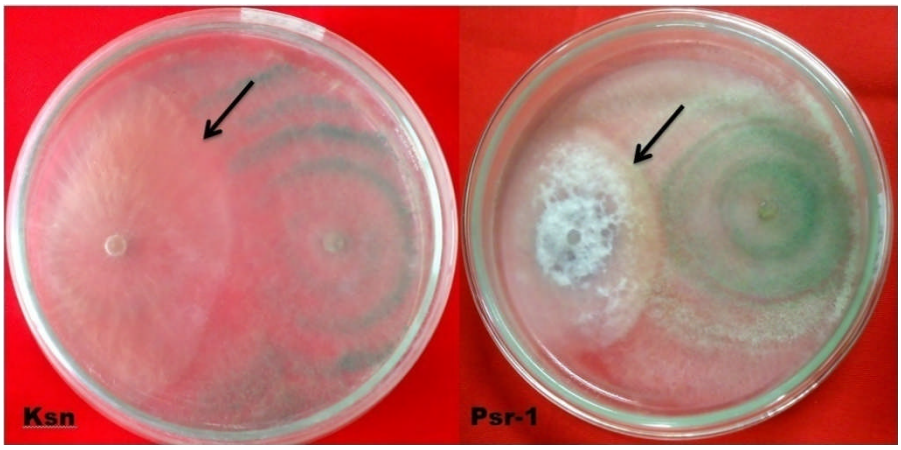

Figure 1. Antagonisms between endophytic *Trichoderma* and *Fusarium oxysporum* f.sp. *cubense*
Note: Meeting zone between *T. asperellum* (Ksn), *T. harzianum*.psr-1 with *F. oxysporum* f.sp. *cubense* (arrow)

Testing of inhibition ability of endophytic *Trichoderma* against Foc was shown in Figure 2. The average of inhibition was 49.67 to 60.61%. The highest percentage growth inhibition was of *T. gamsii* (60.61%), followed by *T. harzianum*.psr-1 (59.08%), *T. harzianum*.swn-1 (55.80%), *T. koningiopsis* (55.58%), *T. harzianum*.swn-2 (54.05%), and *T. asperellum* (49.67%).

Inhibition abilities of *T. harzianum*.swn-1, *T. harzianum*.swn-2, and *T. koningiopsis* were similar, while that of *T. harzianum*.psr-1 was closely to that of *T. gamsii*. This showed, that each species of *Trichoderma* have different ability to inhibit Foc (Harman et al., 2004; Holmes et al., 2004).

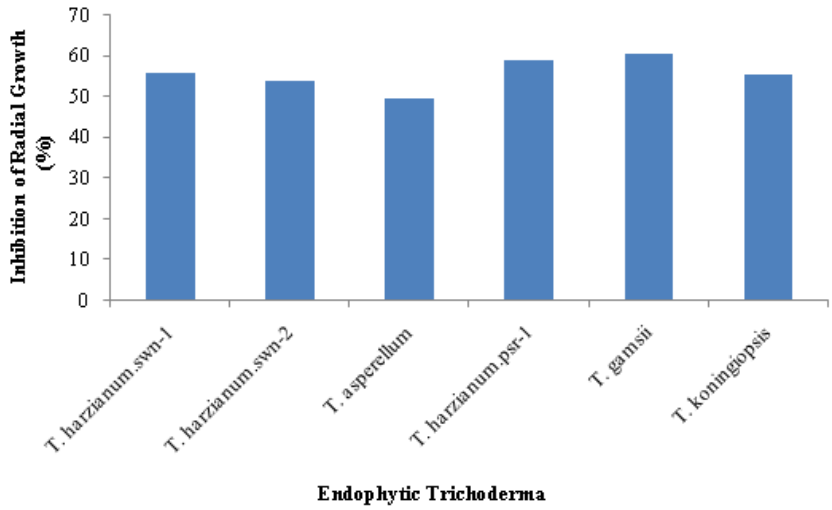

Figure 2. Growth inhibition percentage of endophytic *Trichoderma* on colony of *Fusarium oxysporum* f.sp. *cubense*

Induced resistance against fusarium wilt

Disease Intensity

There were no symptoms of disease on the first and second weeks after treatments with endophytic *Trichoderma* (Fig. 3). The lowest disease intensity (8.33%) was shown on the third week of the following treatments: A2F1 (*T. harzianum*.swn-2) and A3F1 (*T. asperellum*) categorize as resistant. The highest disease intensity (16.67%) were found on the following treatments A1F1 (*T. harzianum*.swn-1), A4F1 (*T. harzianum*.psr.1), A5F1 (*T. gamsii*), and A6F1 (*T. koningiopsis*) categorize as resistant. Disease intensity of 16.67% at the fourth week were the same for all treatments except that of A5F1 (*T. gamsii*) (25%), but still in the same criteria are resistant. The lowest disease intensity on the fifth week (16.67%) were that of A1F1 (*T. harzianum*.swn-1), A2F1 (*T. harzianum*.swn-2), A3F1 (*T. asperellum*), and A4F1 (*T. harzianum*.psr-1) 16.67%, followed by treatment A5F1 (*T. gamsii*) of 25%, categorize as resistant, while the highest disease intensity (33.33%) was of A6F1 (*T. koningiopsis*) categorize as moderate.

When comparing with the positive control (A0F0) as well as negative control (A0F1), it indicates that all species of endophytic *Trichoderma* tested had ability to suppress growth of wilt fusarium pathogen. The endophytic Trichoderma then penetrates the root system of plants, and survives in plant tissue, thus protecting the plants against the entry of pathogen (Evan et al., 2003; Harman et al., 2004; Bailey et al., 2008). Endophytic *Trichoderma* isolated from cacao leaves also shown to have the ability to protect the cocoa plants against *Phytophthora* sp. by induction resistance mechanisms, and the yield loss caused by broom disease (Samuels, 2006; Arnold et al., 2007).

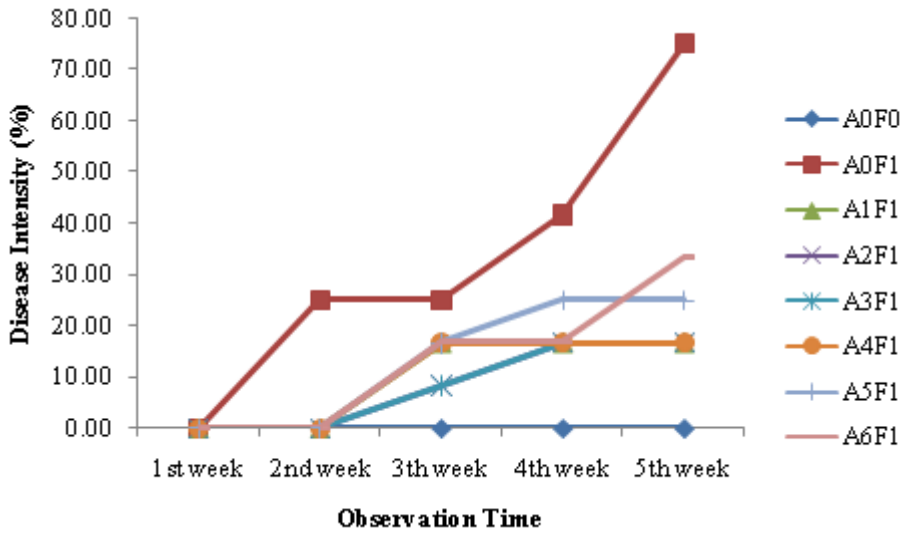

Figure 3. Disease intensity of fusarium wilt on induced resistance treatment with isolate of endophytic *Trichoderma*

Note: A0F0: Control; A0F1: Without isolate + Foc inoculate; A1F1: Swn-1 isolate + Foc isolate; A2F1: Swn-2 isolate + Foc inoculate; A3F1: Ksn isolate + Foc inoculate; A4F1: Psr-1 isolate + Foc inoculate; A5F1: Psr-2 isolate + Foc inoculate; A6F1: Psr-3 isolate + Foc inoculate.

The role of endophytic Trichoderma as on promoting plant growth

The role of endophytic *Trichoderma* as on Promoting Plant Growth was shown in Table 2. Analysis of plat height showed significantly different at A1F1 treatment (*T. harzianum*.swn-1) with A0F1 (positive control), A3F1 (*T. Asperellum*) and A6F1 (*T. koningiopsis*), but not significantly different with A0F0 (negative control), A2F1 (*T. harzianum*.swn-2), A4F1 (*T. harzianum*.psr-1) and A5F1 (*T. gamsii*). Analysis of the stem diameter showed significant differences in treatment of A1F1 (*T. harzianum*.swn-1) with A0F0 (negative control), A0F1 (positive control), A3F1 (*T. asperellum*), A5F1 (*T. gamsii*), and A6F1 (*T. koningiopsis*), but not significantly different to the treatment of A2F1 (*T. harzianum*.swn-2), and A4F1 (*T. harzianum*.psr-1) (Table 2). Analysis of the

number of leaves showed significant differences in treatment A1F1 (*T. harzianum*.swn-1) to all treatments except with A0F0 (negative control) and A4F1 (*T. harzianum*.psr-1). Analysis showed the treatment plant fresh weight A1F1 (*T. harzianum*.swn-1), A2F1 (*T. harzianum*.swn-2), and A4F1 (*T. harzianum*.psr-1) was not significantly different from one another, but significantly different against A0F0 (negative control), A0F1 (positive control), A3F1 (*T. asperellum*), A5F1 (*T. gamsii*) and A6F1 (*T. koningiopsis*). Analysis of root wet weight showed no significant differences between treatment A2F1 (*T. harzianum*.swn-2) to the treatment A1F1, A4F1 and to A0F1, but significantly different to the treatment A3F1 (*T. Asperellum*), A5F1 (*T. gamsii*) and A6F1 (*T. koningiopsis*), also A0F0 (positive control). Analysis of root length showed significant differences in treatment A4F1 to the treatment A5F1 (*T. gamsii*) and A6F1 (*T. koningiopsis*), but there was no difference between other treatments of A1F1, A2F1, A3F1 and A0F0 (positive control) or A0F1 (control negative). It proved that *Trichoderma* could stimulate banana seedling growth.

Table 2. The effect of endophytic *Trichoderma* on plant height, stem diameter, number of leaf, plant fresh weight, root fresh weight and root length on the fifth week after induced resistance

Treat-ment	Plant Height (cm)	Stem Diameter (cm)	Number of Leaf (Stalk)	Plant Fresh Weight (g)	Root Fresh Weight (g)	Root Length (cm)
A0F0	42.67 abc	6.67cde	10.00 abcd	51.33 cd	8.09 cd	30.00 bcd
A0F1	36.43 c	6.33 de	6.00 d	57.33 cd	12.00 bc	34.33 abc
A1F1	58.33 a	9.33 a	14.00 a	132.33 ab	19.89 bc	36.00 abc
A2F1	54.33 ab	8.67 abc	8.33 bcd	123.00 ab	22.33 ab	37.67 abc
A3F1	42.33 bc	7.00 bcde	7.83 d c	47.33 cd	4.93 cd	26.67 bcd
A4F1	53.00 ab	9.00 ab	11.33 abc	78.00 bc	14.48 bcd	40.17 ab
A5F1	46.67 abc	6.33 de	8.00 cd	33.33 cd	4.47 cd	22.33 cd
A6F1	35.00 c	6.00 e	6.57 cd	12.13 d	0.53 d	16.33 d

Note: Number followed by same letter in the same column is not significantly different with Duncan Multiple range test with 5% error. A0F0: Control; A0F1: without *Trichoderma* isolate + Foc inoculate; A1F1: *T. harzianum*.swn-1 + Foc inoculate; A2F1: *T. harzianum*.swn-2 + inoculation Foc inoculate ; A3F1: *T. asperellum* + Foc inoculate; A4F1: *T. harzianum*.psr-1 + Foc inoculate; A5F1: *T. gamsii* + Foc inoculate; A6F1: *T. koningiopsis* + Foc. Inoculate

Other study proved that *Thrichoderma* spp. produced antimicrobial compounds that could be used in biological control as an antifungal which was viridin (Lumsdem et al., 1992; Lumsdem and Walter, 1996). Enzymatically viridin could turn into viridiol. This compound could promote plant and root growth (Inbar et al., 1994). Accumulation of viridiol in soil reached its pick on five to six days after inoculation and directly generated in the root area (Howel and Stipanovic, 1984).

Total pheholic analysis

Observation of total phenolic compounds in the roots of banana was preceded by conducting measurements using a standard curve dilution of tannin. Based on tannin diluted factors of 100, 200, 300, 400, 500, 600, 700, 800, 900 and 1000 ppm, the regression obtained was: $Y = 0.004\ x + 0.128$. Fig. 4 showed the presence of phenolic compounds in plants treated with *Trichoderma* on the first to fifth weeks. There were no differences in average increase in total phenolic compounds in the first and second week between all treatments, except for that of *T. asperillum* (A3F1) on the second week. On the third week there was an increase of phenol in all treatments, whereas at the fourth and fifth week there was a decrease, except in treatment with *T. harzianum*.psr-1 (A4F1). It could be said that there was a positive effect of induced resistance using endophytic *Trichoderma* against *F. oxysporum* f.sp. *cubense.*

Increased metabolism of phenolic compounds was a common response of plants against pathogen. Biosynthesis of phenolic compounds from phenylalanine was one of the biochemical changes that always occur on infected plants. The compound could inhibit the enzyme hydrolysis, including pechtolycis enzymes produced by pathogens. Phenol (hydroxy acid bensoat, coumarin, flavonoids, and lignin), isoprin (terpenoids and steroids) and enzyme oxidizing phenols included in secondary metabolites, so that the infected plants would have a higher pnenol compounds than that in healthy plants (Goodman et al. 1986; Agrios, 2005).

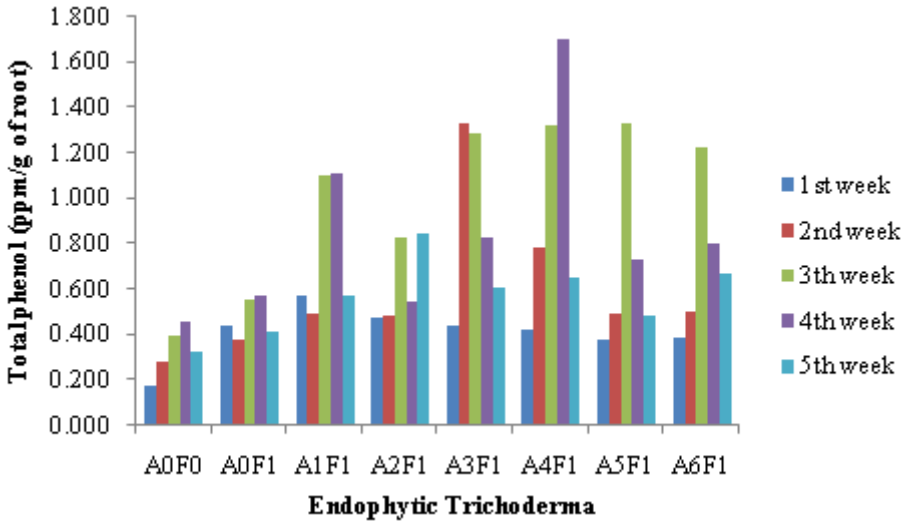

Figure 4. The effect of endophytic *Trichoderma* isolates on total phenol on banana root after induced resistance

Note: A0F0: Control; A0F1: Without isolate + Foc inoculate; A1F1: Swn-1 isolate + Foc isolate; A2F1: Swn-2 isolate + Foc inoculate; A3F1: Ksn isolate + Foc inoculate; A4F1: Psr-1 isolate + Foc inoculate; A5F1: Psr-2 isolate + Foc inoculate; A6F1: Psr-3 isolate + Foc inoculate.

Observation on peroxidase activity

Increased activity of peroxidase was highest in the third week until the fourth week and decreased in five week (Fig.5). The highest peroxidase activity in treatment A4F1 that when associated with the treatment lignification A4F1 and intensity of the disease, then isolates *T. harzianum*.psr-1 can be used as a biological control agent fusarium wilt. Increased in peroxidase` activity on the third week related to the formation of lignin in plants as a barrier when the plants were infected by pathogens, so it could reduce the disease intensity. When pathogen was in contact with the plant, the plant reacted by increasing peroxidase in plants (Anderson et al., 2004).

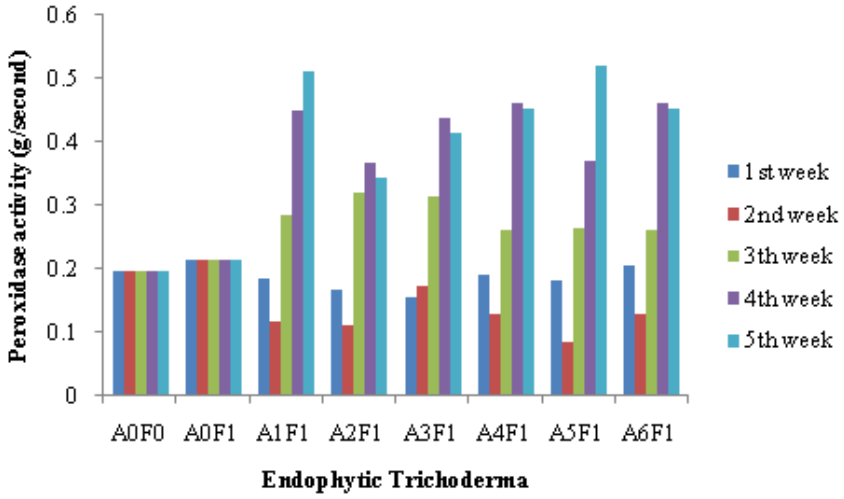

Figure 5. The Effect of endophytic *Trichoderma* isolates on peroxidase activity after induced resistance

Note: A0F0: Control; A0F1: Without isolate + Foc inoculate; A1F1: Swn-1 isolate + Foc isolate; A2F1: Swn-2 isolate + Foc inoculate; A3F1: Ksn isolate + Foc inoculate; A4F1: Psr-1 isolate + Foc inoculate; A5F1: Psr-2 isolate + Foc inoculate; A6F1: Psr-3 isolate + Foc inoculate.

e. Observation of lignification on banana root

Lignification occurred in plant tissues because it was induced with? *Trichoderma* (Table 3). In the following treatments: A1F1 (*T. harzianum.swn-1*), A2F1 (*T. harzianum.swn-2*), and A4F1 (*T. harzianum.psr-1*) lignification was occurred in every replications compared to the other 3 isolates of *Trichoderma*. Lignification influenced disease intensity (Table 2) since lignification on infected part of plant would block spreading of pathogen to healthy tissues, therefore disease intensity will be decreased.

Lignification occurred in vascular tissues of banana roots and at infected point that hard to be degrade by microorganisms. Lignin is one of structural deence mechanism that happened as` response to pathogen penetration. Beside the mechanic function, lignin also has significant function in plant protection (Agrios, 1997). The layer occurred as a response of host plant to toxic compound produced by pathogen during infection. Lignification will stop the spread of the compounds excrete by pathogen from infected plant tissue to healthy plant tissues.

Table 3. Lignification on banana root on fifth week after induced resistance with isolate of endophytic *Trichoderma*

Treatment	Lignification of banana root		
	Replication		
	1	2	3
A0F0	-	-	-
A0F1	-	-	-
A1F1	+	+	+
A2 F1	+	+	+
A3F1	+	+	-
A4F1	+	+	+
A5F1	-	-	+
A6F1	-	-	+

Note: (+) : lignification occur; (-) : No lignifications, A0F0: Control; A0F1: Without isolate + Foc inoculate; A1F1:Swn-1 isolate + Foc inoculate; A2F1: Swn-2 Isolate + Foc inoculate; A3F1: Ksn isolate + Foc inoculate; A4F1: Psr-1 isolate + Foc inoculate; A5F1: Psr-2 isolate+ Foc inoculate; A6F1: Psr-3 isolate + Foc inoculate

Conclusion

The results showed that endophytic *Trichoderma* could be used as biological control agent to suppress fusarium wilt of banana. Antagonistic test showed the ability of *Trichoderma gamsii* in suppressing 60.61% of the growth of *F. oxysporum* f.sp. *cubense*. Induced resistance using endophytic *Trichoderma* increased peroxidase activity and total phenols of banana plant from at three and four weeks after inoculation. Due to the high peroxidase activity and total phenolic compounds cause lignification which have positive effect on the severity of the disease. Endophytic *Trichoderma* as biological control agents could also stimulate vegetative growth of banana seedling.

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