



Research Article

Potential of *Trichoderma harzianum* OC12 as Biological Control Against Fusarium Wilt of *Capsicum frutescens* L.

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Abstract: Fusarium wilt caused by *Fusarium oxysporum* f. sp. *capsici* is a major constraint to cayenne pepper production in Indonesia. Excessive reliance on synthetic fungicides has led to environmental degradation and reduced soil microbial diversity, highlighting the need for sustainable alternatives. This study evaluated the effectiveness of different doses of *Trichoderma harzianum* OC12 as a biological control agent against Fusarium wilt in cayenne pepper (*Capsicum frutescens* L.). The experiment was conducted using a completely randomized design with five treatments: control (without *T. harzianum* OC12) and four doses of *T. harzianum* OC12 formulated on corn-rice solid medium (12, 16, 20, and 24 g per polybag), each replicated four times. Disease incidence, incubation period, plant height, number of leaves, and root length were recorded. Results showed that *T. harzianum* OC12 significantly reduced Fusarium wilt incidence and enhanced plant growth parameters. The highest dose (24 g/polybag or 24 g in 2 Kg topsoil) resulted in the lowest disease incidence (16.5%) compared with 100% disease incidence in the control. Additionally, this treatment significantly increased plant height, leaf number, and root length. These findings indicate that *T. harzianum* OC12, particularly at 24 g/polybag, is an effective and environmentally friendly biological control agent for managing Fusarium wilt in cayenne pepper cultivation.

Keywords: biological control; biopesticide; cayenne pepper; Fusarium wilt; plant growth-promoting fungi.

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Introduction

Cayenne pepper (*Capsicum frutescens* L.) is an economically important horticultural crop in Indonesia, widely used for household consumption and food processing industries [1,2]. Despite increasing demand, productivity remains unstable due to biotic stresses, particularly soil-borne diseases. Fusarium wilt caused by *Fusarium oxysporum* f. sp. *capsici* is among the most destructive diseases affecting cayenne pepper, leading to severe yield losses from the vegetative to generative growth stages [3,4].

The pathogen infects plants through the root system and colonizes the xylem vessels, disrupting water and nutrient transport, which results in leaf chlorosis, wilting, vascular discoloration, and eventually plant death [5-7]. Conventional disease management relies heavily on synthetic fungicides; however, their long-term use poses risks to human health, soil ecosystems, and environmental sustainability [8,9].

Biological control using antagonistic fungi such as *Trichoderma* sp. has gained attention as a sustainable alternative [10]. *Trichoderma* species are known for their rapid

growth, competitive ability, production of antifungal metabolites, mycoparasitic activity, and capacity to induce systemic resistance in plants [10–13]. In addition, *Trichoderma* acts as a plant growth-promoting fungus (PGPF), enhancing nutrient uptake and root development [14].

Although numerous studies have reported the antagonistic potential of *Trichoderma* sp., especially *T. harzianum* strain OC12, information on optimal application doses under in vivo conditions remains limited. Therefore, this study aimed to evaluate the effect of different doses of *T. harzianum* strain OC12 on Fusarium wilt suppression and growth performance of cayenne pepper plants.

Materials and Methods

Experimental Site

The experiment was conducted at the Integrated Laboratory and Experimental Field, Department of Agricultural Science and Technology, Faculty of Agriculture, Universitas Trunodjoyo Madura, Bangkalan, East Java, Indonesia. The experiment was performed under greenhouse conditions with average temperatures of 28–32 °C and relative humidity of 65–85%.

Plant Material

Cayenne pepper seeds of a commonly cultivated local variety were surface-sterilized using 1% sodium hypochlorite for 2 minutes, rinsed three times with sterile distilled water, and germinated in sterilized seed trays. Uniform seedlings aged 20 days were selected for transplantation.

Pathogen and Antagonist Preparation

The pathogenic fungus *F. oxysporum* f. sp. *capsici* was obtained from the East Java Assessment Institute for Agricultural Technology (BPTP) and re-cultured on potato dextrose agar (PDA) at 27 ± 1 °C for 7 days. The antagonistic fungus *T. harzianum* OC12 was laboratory collection. The isolate was sub-cultured on PDA and incubated for 5–7 days until sporulation.

Mass Propagation of Fungal Inoculum

Both fungi were mass-produced using a solid substrate consisting of sterilized corn and rice (1:1, w/w). The substrate was autoclaved at 121 °C for 30 minutes on two consecutive days. After cooling, each substrate was inoculated with 5 mm mycelial plugs and incubated for 10–14 days until fully colonized.

Mass Propagation of Fungal Inoculum

The experiment employed a Completely Randomized Design (CRD) with five treatments and four replications. Each polybag contained 2 kg of sterilized topsoil. Each replication consisted of three plants. Treatments were as follows:

P0 = Control (without *T. harzianum* OC12)

P1 = *T. harzianum* OC12 at 12 g/polybag

P2 = *T. harzianum* OC12 at 16 g/polybag

P3 = *T. harzianum* OC12 at 20 g/polybag

P4 = *T. harzianum* OC12 at 24 g/polybag

Application of *T. harzianum* OC12 and Pathogen Inoculation

T. harzianum OC12 was applied by thoroughly mixing the appropriate dose into the planting medium seven days before transplanting to allow establishment in the rhizosphere. Inoculation with *F. oxysporum* f. sp. *capsici* was performed seven days after transplanting by placing 10 g of colonized substrate around the root zone at a depth of 3–5 cm.

Observed Parameters

The following parameters were recorded. Incubation period (days) or time from pathogen inoculation to the appearance of initial wilt symptoms. Plant height (cm) was measured weekly from soil surface to the apical meristem. Number of leaves was counted weekly. Root length (cm) was measured at final harvest (30 days after transplanting). Disease incidence (%) was calculated using Formula 1.

$$\text{Disease incidence} = \frac{\text{Number of infected plants}}{\text{Total plants observed}} \times 100 \quad (1)$$

Statistical Analysis

All data were subjected to analysis of variance (ANOVA) using a 5% significance level. When significant differences were detected, Duncan's Multiple Range Test (DMRT) was applied for mean separation.

Results and Discussion

Effect of *T. harzianum* OC12 on Fusarium Wilt Development

Application of *T. harzianum* OC12 significantly delayed the incubation period and reduced Fusarium wilt incidence compared with the untreated control. Control plants exhibited initial wilt symptoms as early as 7 days after inoculation, whereas plants treated with 24 g/polybag showed delayed symptom expression up to 21 days. Disease incidence reached 100% in the control treatment, confirming the high virulence of *F. oxysporum* f. sp. *capsici*. In contrast, the highest *T. harzianum* OC12 dose (P4) reduced disease incidence to 16.5%. This strong suppressive effect indicates successful rhizosphere colonization and antagonistic activity of *Trichoderma* sp. (Figure 1 & Table 1).

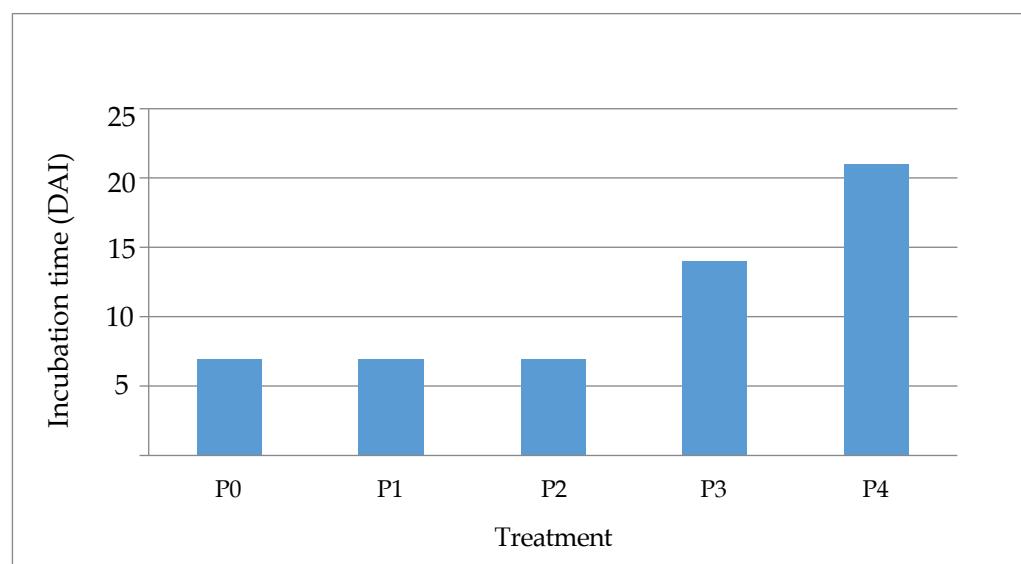


Figure 1. Effect *T. harzianum* OC12 treatment on the incubation period of Fusarium wilt disease.

The suppression of Fusarium wilt was strongly dose-dependent. Disease incidence reached 100% in the untreated control, confirming the high virulence of *F. oxysporum* in the absence of biological protection. In contrast, the lowest disease incidence (16.5%) was recorded in plants treated with 24 g of *T. harzianum* OC12. This reduction is closely associated with the superior competitive ability of *Trichoderma* sp., which rapidly

colonizes the rhizosphere and utilizes available nutrients more efficiently than the pathogen, thereby delaying pathogen contact and penetration into plant tissues [15].

Beyond competition, *Trichoderma* sp. suppresses soil-borne pathogens through antibiosis and enzymatic degradation. The fungus produces antifungal metabolites such as gliotoxin, glyoviridin, and trichodermin, which inhibit pathogen growth and viability. Additionally, extracellular enzymes including β -1,3-glucanase and chitinase degrade fungal cell walls, leading to pathogen lysis [16]. These combined mechanisms explain the substantial reduction in disease incidence observed at higher *Trichoderma* doses [17,18].

The delayed disease development also suggests activation of plant defense responses. *Trichoderma* sp. functions as a plant growth-promoting fungus (PGPF) capable of inducing systemic resistance, including Systemic Acquired Resistance (SAR) and Induced Systemic Resistance (ISR). These defense mechanisms enhance the plant's ability to respond more rapidly and effectively to pathogen invasion [19,20]. In treatments with lower *Trichoderma* doses, disease suppression was less pronounced, likely due to slower activation of plant defense pathways and reduced antagonist population density [21,22].

Tabel 1. Effect of *T. harzianum* OC12 treatment on the disease incidence of Fusarium wilt disease.

Treatment	Disease incidence (%) at different days after transplanting (dat)*		
	14 dat	21 dat	28 dat
P0	74.75 d	91.50 e	100 d
P1	50.00 c	41.25 d	16.50 cd
P2	41.25 b	16.50 b	8.25 b
P3	0.00 a	33.00 c	0.00 a
P4	0.00 a	0.00 a	16.50 c

*Different letters within a column indicate significant differences according to DMRT (5%).

Growth Promotion of Cayenne Pepper Plants

In addition to disease suppression, *T. harzianum* OC12 significantly enhanced vegetative growth parameters. Plants treated with 24 g/polybag exhibited the greatest plant height, leaf number, and root length compared with all other treatments (Table 2 and Table 3). *T. harzianum* OC12 significantly promoted vegetative growth of cayenne pepper plants. The highest dose (24 g/polybag) produced the greatest plant height, leaf number, and root length throughout the observation period. Improved plant growth is attributed to the role of *Trichoderma* sp. as a decomposer of organic matter, enhancing soil nutrient availability and uptake. The fungus is known to mineralize essential nutrients such as nitrogen, phosphorus, magnesium, and sulfur, which are critical for photosynthesis and vegetative development [23,24].

Enhanced root development observed in *Trichoderma*-treated plants suggests improved nutrient and water uptake efficiency. Increased root length and density improve water and nutrient absorption, contributing to overall plant vigor. This effect is partially linked to the production of auxin-like phytohormones by *Trichoderma* sp., which stimulate lateral root formation and root elongation [24,25]. The mutualistic relationship between *Trichoderma* sp. and host plants benefits both organisms: the fungus obtains carbon sources from root exudates, while the plant gains enhanced growth and disease resistance [26]. Several *Trichoderma* species are known to function as plant growth-promoting fungi (PGPF) by producing auxin-like compounds, solubilizing phosphate, and improving micronutrient availability [27,28].

Tabel 2. Effect of *T. harzianum* OC12 treatment on the plant height of cayenne pepper.

Treatment	Plant height (cm) at different days after transplanting (dat)*			
	7 dat	14 dat	21 dat	28 dat
P0	8.41 a	12.40 a	14.13 a	14.91 a
P1	8.91 ab	15.64 b	21.27 b	25.93 b
P2	8.08 a	16.70 bc	24.78 c	29.19 c
P3	8.99 b	17.94 c	27.93 d	33.44 d
P4	8.99 b	20.09 d	28.88 e	37.37 e

*Different letters within a column indicate significant differences according to DMRT (5%).

Tabel 3. Effect of *T. harzianum* OC12 treatment on the number of leaves and root length.

Treatment	Average number of leaves*				Average of root length (cm)*
	7 dat	14 dat	21 dat	28 dat	
P0	8.41 a	12.40 a	14.13 a	14.91 a	5.66 a
P1	8.91 ab	15.64 b	21.27 b	25.93 b	9.49 b
P2	8.08 a	16.70 bc	24.78 c	29.19 c	12.16 c
P3	8.99 b	17.94 c	27.93 d	33.44 d	14.74 d
P4	8.99 b	20.09 d	28.88 e	37.37 e	16.57 e

*Different letters within a column indicate significant differences according to DMRT (5%); dat=day after transplanting.

Overall, the superior performance of the highest *Trichoderma* dose highlights the importance of adequate antagonist population density for effective biological control. Rapid growth, efficient nutrient utilization, antibiosis, enzymatic activity, and induction of plant defense collectively contribute to the effectiveness of *Trichoderma* sp. in suppressing Fusarium wilt and improving plant growth [29,30].

Conclusions

T. harzianum OC12 effectively suppressed Fusarium wilt and enhanced the vegetative growth of cayenne pepper plants. The dose of 24 g/polybag was the most effective treatment, resulting in the lowest disease incidence and highest growth performance. *T. harzianum* OC12 is a promising environmentally friendly alternative to synthetic fungicides for sustainable cayenne pepper production.

Additional Section

Author Contributions: Conceptualization and methodology, S.K. and A.D.; writing—review and editing, D.M. and S.K.; formal analysis and investigation, A.G.A.; resources, data curation, writing—original draft preparation, A.G.A., A.D., C.W., E.S. and S.K. All authors have read and agreed to the published version of the manuscript.

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