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Research Article

Mix Fertilizer "BiTrichompos" Inhibition Ability against Bacterial Wilt Disease, *Ralstonia solanacearum*, in Chili Plants (*Capsicum annuum* L.)

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ABSTRACT

Chili production is hindered by bacterial wilt disease caused by *Ralstonia solanacearum*. Application of BiTrichompos can be an alternative control method of bacterial wilt diseases. BiTrichompos is an organic fertilizer derived from the secondary metabolism of *Fusarium oxysporum* and *Trichoderma* sp. dissolved in rice washing water (*leri*). Secondary metabolism and *Trichoderma* sp. have been reported to be able to suppress plant disease causing pathogens. The purpose of this study is to evaluate BiTrichompos ability to manage bacterial wilt disease (*Ralstonia solanacearum*) and growth promoting effects on chili plants (*Capsicum annuum* L.). This research was conducted at Screen House Harzianum Parepare City, using a Completely Randomized Design with 4 treatments, namely A0 (control), A1 (5 mL of BiTrichompos/plant), A2 (10 mL BiTrichompos/plant), A3 (15 mL BiTrichompos/plant), and each treatment was repeated 5 times. Each replicated consisted of 3 plants totaling in 60 experimental units. Observations were made in the first week after planting and continued weekly for 7 weeks. Results showed that treatment A3 resulted in lowest bacterial wilt disease intensity of 31.53%, while to control resulted in 39.15%. Highest average fruit number and weight resulted from treatment A2, where fruit numbers averaged at 6.75 and total fruit weight was 29.57 g. Highest average root length of chili plants was also shown A2 (46 cm).

Keywords: bacterial wilt disease; chili; mix fertilizer BiTrichompos

INTRODUCTION

Chili pepper (*Capsicum annuum* L.) is a horticultural crop with unique flavors that contains nutrients such as protein, dietary fiber, vitamin C and minerals (Michel *et al.*, 2021). Farmers' chilies production are often challenged by bacterial wilt disease, caused by *Ralstonia solanacearum* (Fiqri *et al.*, 2023) causing detrimental production and economic losses (Kasidal *et al.*, 2019). Accordingly, Raihanah *et al.* (2023) also stated that the economic effects of bacterial wilt disease can lead to a 90% production decrease. Thus, there are always merits effective management strategies.

Various strategies have been used to manage bacterial wilt disease, including chemical pesticides, soil disinfection, antibiotics, antimicrobial plant extracts, using resistant plants varieties, crop rotation, and applications of antagonistic bacteria (Elsayed *et al.*, 2020). However, reports have shown that results from these methods often vary causing resistance of farmer adoption. Therefore, effective and environmentally safer control strategies that can also promote plant growth are needed. BiTrichompos is the strategy being used here.

BiTrichompos is an organic fertilizer derived from the secondary metabolism of *Fusarium oxysporum* and *Trichoderma* sp. dissolved in rice washing water (*leri*). Ambar *et al.* (2015) stated that the secondary metabolism of *F. oxysporum* can increase plant resistance by forming lignin and suberin. Similar to Irawati *et al.* (2020), endophytic fungi such as *Fusarium* spp. and *Trichoderma* spp. has the ability to produce antimicrobials that can directly inhibit *R. solanacearum* through an antibiosis mechanism and trigger the growth of chili plants.

BiTrichompos have been reported by Ambar *et al.* (2021) that BiTrichompos combined with *Pseudomonas fluorescens* showed an inhibitory effect on bacterial wilt disease caused by *R. solanacearum* on tomatoes. Similarly, Aminullah *et al.* (2021), stated that the combination of BiTrichompos and Biochar had a significant effect on the growth of mustard greens on acid soil. This study aims to determine the effect of BiTrichompos on bacterial wilt disease caused by *R. solanacearum* by observing the intensity of the disease and its effect on the growth of chili plants (*Capsicum annuum* L.).

MATERIALS AND METHODS

The study was carried out between March−July 2023 at the Screen House Harzianum Parepare City and the Agrotechnology Laboratory of the Faculty of Agriculture Universitas Muhammadiyah Parepare (UMPAR).

Research Design

This study used a Complete Randomized Design with 4 treatments, namely: A0 (control); A1 (5 mL BiTrichompos/plant); A2 (10 mL BiTrichompos/plant); A3 (15 mL BiTrichompos/plant). Each treatment was repeated 5 times. Three plant units were used in each replication, resulting in 60 experimental units.

Planting Medium Preparation

Planting medium used a combination of soil+husk and placed in plastic bag, sealed, and sterilized using pressurized hot steam (autoclave: 121 °C; 15 psi) for 30 minutes. The sterile soil was mixed with roasted husks in a 1:1 ratio and 9 kg of planting media was placed into fully-filled 30×40 cm polybags.

Preparation of Isolates of *Ralstonia solanacearum* Bacteria

Ralstonia solanacearum isolates were obtained from the Indonesian Plant Protection Center, Maros Regency. The isolates were placed into NB medium and cultured for 24 hours shaken at a speed 120 rpm. The population used for virulence the *R. solanacearum* was 10^8 CFU/ml, which is achieved by mixing the suspension with sterile distilled water in 1:7 to the *R. solanacearum* suspension in NB medium (Asril *et al.*, 2020).

Seeding and Planting

Before planting, chili seeds were soaked for five minutes in warm water of 50 °C (Sultana *et al.*, 2021). This was done to produce pathogen free seeds. After that the seeds were drained for five minutes on filter paper. Seeds were planted in seedling polybags (10×10 cm) which have been filled with mixture of sterile soil and compost (1:1). The seeds were planted in each seedling polybag. The seeds were watered daily using a hand spray. The 25 day-old plants were transplanted into polybags for subsequently tests. Seedlings roots were washed with water and dried for 15 minutes. Approximately 1 cm of root tips were trimmed and immersed in BiTrichompos for 15 minutes. One chili seeds were planted in each 40×40 cm polybag. Two-week-old plants were infected with *R. solanacearum* of 10^8 CFU/mL and a solution of 5 mL/plant was applicated using a syringe (Fiqri *et al.*, 2023).

Aplication of BiTrichompos

BiTrichompos application was carried out on 3-week old seedlings. Application was done by pouring the BiTrichompos solution according in the morning (07.00). The BiTrichompos application was carried out weekly for 7 weeks.

Observation Parameters

Disease severity was measured by counting number of wilt leaves compare to all of leaves. These measurements were done weekly for 7 weeks, with using the following formula (Irawati *et al.*, 2020):

$$P = \sum \frac{ni \times vi}{V \times Z} \times 100\%$$

P = Disease severity (%); *ni* = Number of infected leaves to-i; *vi* = Scale value for each infected category to-i; V = Total number of leaves observed; Z = The highest category scale.

The category scale is: 0 (healthy plant); 1 (1−20% wilting leaves); 2 (21−40% wilting leaves); 3 (41−60% wilting leaves); 4 (61−80% wilting leaves); 5 (> 80% wilting leaves until dead).

The number of fruits were counted regardless of their infection status. Observations were conducted on a weekly basis for a period of 7 weeks. Fruits that had fallen on the ground were not included. The fruit weight, determined at the end of the observation period by weighing the entire fruit on all plants. Root length was measured at the end of the observation, by measuring from the base of the stem to the tip of the root.

Data Analysis

The ANOVA analysis was used to tests of the bacterial wilt disease intensity and plant growth. The Duncan test is used to determine the average difference between treatments at a significance level of 5%.

RESULTS AND DISCUSSION

Bacterial wilt disease symptoms occured on almost all leaves. Occasionally, while the leaves were still green, they fell off. These symptoms spread to the fruit, causing them to become yellow and rot before finally falling off. When stems were splits, brown blotches appeared on the plant tissue (Figure 1).

This disease can easily spread to other plants. Plants infected by *R. solanacearum* have wilting leaves. If the entire plant starts to wilt, especially during the day (high temperature) or heavy rain, it will suddenly die. This is an early indicator of bacterial wilt disease (Thakur *et al.*, 2021). At the peak of the bloom, adventitious roots will also start to grow on the plant's stem; this symptom is caused by *R. solanacearum* infection. The fruit becomes yellow and starts to rot when fruits are infected (Figure 1b). Damage to the xylem and phloem tissue, which is indicated by a brownish hue in the roots and stems, is as well a symptoms (Wijaya *et al.*, 2017).

Severity of Bacteria Wild Disease (*Ralstonia solanacearum*)

BiTrichompos had positive impact on suppressing bacterial wilt severity. All BiTrichompos treatments showed low disease severity compared to the control from 1−7 week. In particular, at week 4 and week 7, statistical analysis showed a significant effect on disease severity, especially in A3 treatment, respectively 16.23% and 31.13%, compared to the control treatment of 27.42% and 39.35% (Figure 2).

The low percentage of disease in A3 implied that BiTrichompos could suppresse bacterial wilt development. BiTrichompos contained secondary metabolites of *F. oxysporum* and *T. harzianum*. These two components were effective in preventing and reducing the development of disease in plants (Ambar *et al.*, 2021), by forming physical and chemical resistance system (Ambar *et al.*, 2019).

Ambar *et al.* (2015) stated that *F. oxysporum* produced secondary metabolite of fusaric acid that can induce physical resistance in plants leading it to suppress bacterial wilt in chili plants, especially in the A3 treatment. Ambar *et al.* (2015) stated that secondary metabolism produced by *F. oxysporum* could induce lignin and suberin, as well as the research results of Nurcahyani *et al.* (2023) that can also induce chemical resistance by producing peroxidase enzymes.

BiTrichompos showed suppressive effects against bacterial wilt, especially in the A3. BiTrichompos solution may contain more *T. harzianum* because its volume is assumed to be larger than the other solutions. According to Islam *et al.* (2021), *T. harzianum*

A B C

Figure 1. Symptoms of bacterial wilt disease on leaves (A), fruit (B), and stem (C) of chili plant

could inhibit *R. solanacearum* because it produces chitinase and had secondary metabolite antibacterial effect. Similarly, Pico Rosado *et al.* (2024) stated that 3 species of *Trichoderma* sp. showed potential inhibition against *R. solanacearum*. Gomes *et al.* (2023), stated that *Trichoderma* sp. had the ability to induce resistance, for example, they colonize the root surfaces causing changes in plant metabolism to stimulate defense mechanisms, such as the production of phenols and phytoalexins, and increase nutrient availability and stress tolerance.

Quality and Weight of Fruits

One of the parameters used to evaluate the effects of treatment on plant growth was the quantity of fruit and fruit weight. In comparison to the control treatment, A2 treatment indicated higher fruit number and weight (6.75 pieces and 29.59 g) (Figures 3 and 4).

BiTrichompos had the potential to increase plant growth through increasing quantity fruit and fruit weight, especially in A2 treatment, although it was not significantly different between treatments. The South Sulawesi Agricultural Technology Assessment

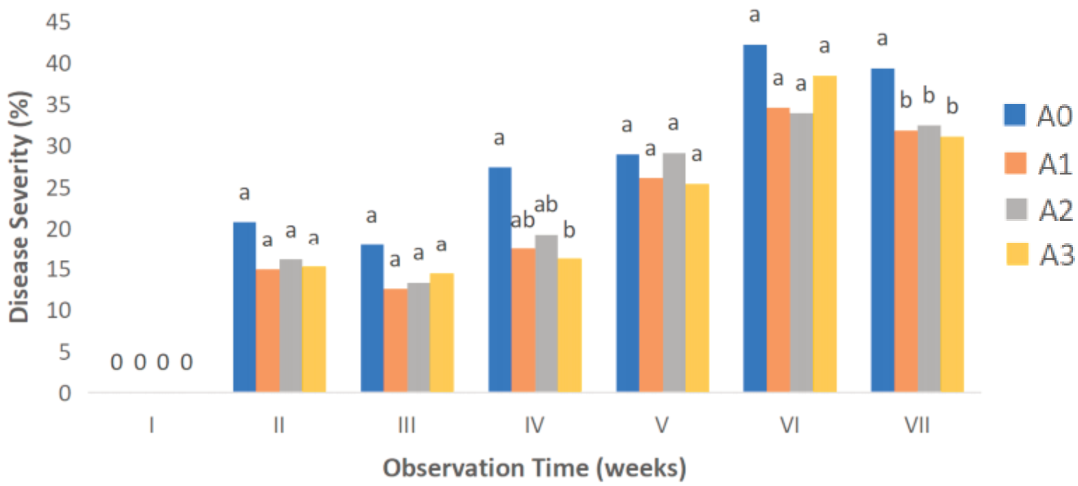

Figure 2. Disease severity of bacterial wilt (%) caused by *Ralstonia solanacearum* after BiTrichompos application during seven weeks of observation (A0 = control, A1 = 5 mL BiTrichompos/plant, A2 = 10 mL BiTrichompos/plant, A3 = 15 mL BiTrichompos/plant). Numbers followed by same letter are not significantly different.

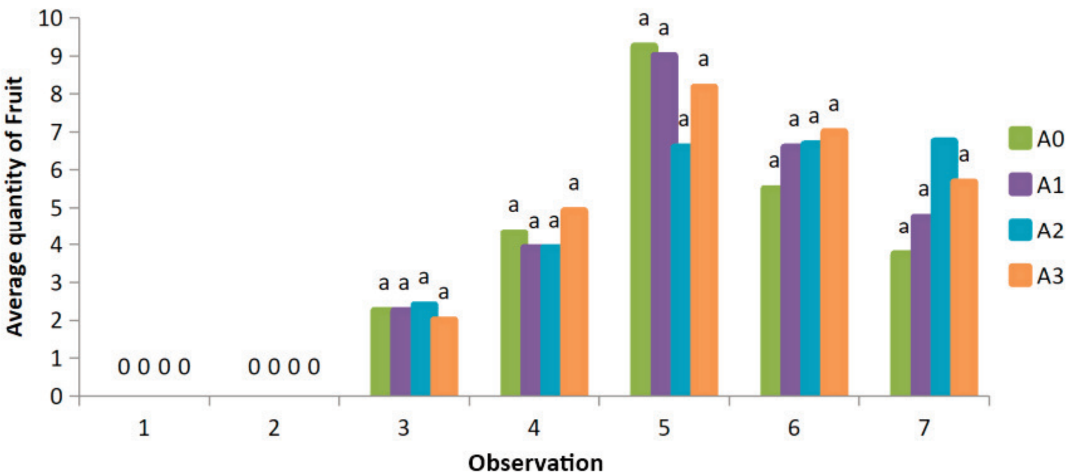

Figure 3. Average quantity of chili fruit after application of BiTrichompos in various treatments during 7 weeks of observation (A0 = control, A1 = 5 mL BiTrichompos/plant, A2 = 10 mL BiTrichompos/plant, A3 = 15 mL BiTrichompos/plant). Numbers followed by same letter are not significantly different.

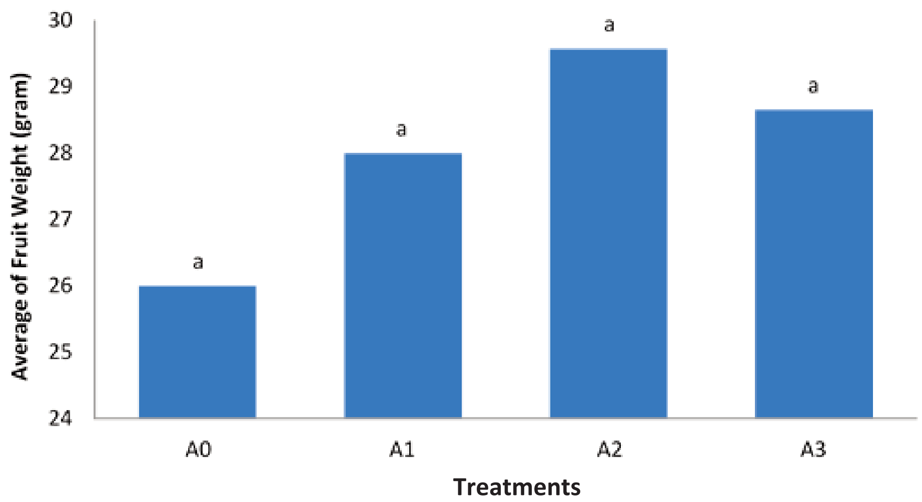

Figure 4. Average weight of chili (g) after application of BiTrichompos on various treatments (A0 = control, A1 = 5 mL BiTrichompos/plant, A2 = 10 mL BiTrichompos/plant, A3 = 15 mL BiTrichompos/plant). Treatment by same letter are not significantly different.

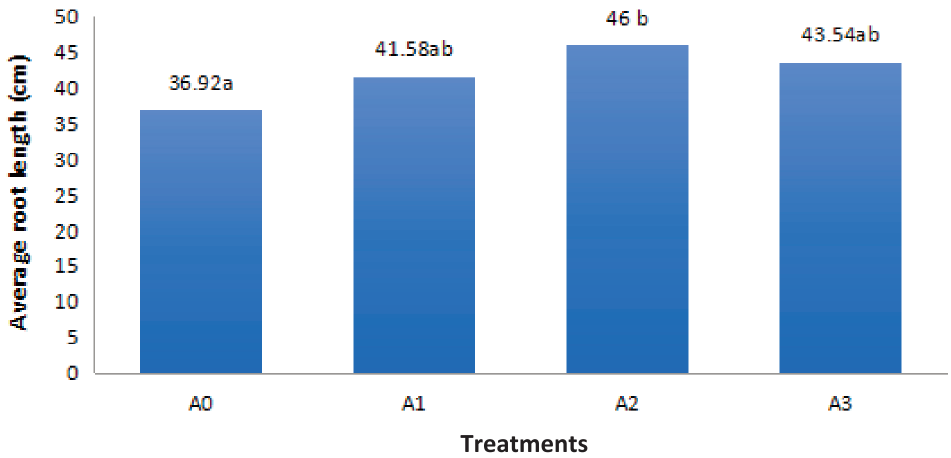

Figure 5. Average root length (cm) of chilies after BiTrichompos application in various treatments (A0 = control, A1 = 5 mL BiTrichompos/plant, A2 = 10 mL BiTrichompos/plant, A3 = 15 mL Bitrichompos/plant). Number followed by same letter are not significantly different.

Center's laboratory research revealed that the BiTrichompos products contained nitrogen, phosphate, potassium, iron, zinc, copper, calcium, and magnesium. According to Sondakh *et al.* (2020), applying Trichokompos and Nitrogen, Phosphorus and Potassium (NPK) fertilizer had significant impacts on plant development and yield, as well as fruit formation and filling.

BiTrichompos uses additional ingredients in the form of rice washing water. This rice washing water can function to stimulate plant growth. According to Nurul *et al.* (2023), rice washing water contained NPK which stimulates plant growth. In accordance with the findings of Anugrah *et al.* (2021), the complete content of rice washing water was 0.014% N, 14.452% P, 0.02% K, 3.574% Ca, 13.286% Mg, 0.005% S, 0.0698 Fe, and 0.043% vitamin B1.

Length of Root

Root length of A2 treatment was significantly longer compared to other treatment (Figure 5). BiTrichompos provided the greatest effects compared to the control indicating that *T. harzianum* can promote plant growth effectively. This is supported by the research of Samolski *et al.* (2012) that stated *T. harzianum* affected root length and number of root hairs due to *qid74*. Similarly, research conducted by Hasan *et al.* (2023) stated that plants treated with

Trichoderma spp. showed longer roots and shoots than untreated control.

Rice washing water contains organic compounds and minerals that include nitrogen, phosphorus, potassium, calcium, magnesium, carbohydrates, sulfur, iron, and thiamine. These compounds can affect root length and increase plant root growth. Vitamins such as A, C, and thiamine can be found in the white rice washing water (Nurul *et al.*, 2023). Thiamine affect plant metabolism, namely converting carbohydrates into energy used for plant activities. It could also stimulates the growth and metabolism of plant roots (Riyati *et al.*, 2022).

Root length influences plant growth and increases yields. Plant roots play an active role in absorbing nutrients and water, so the length and number of roots influence the range and amount of nutrients and water that plants can absorb.

CONCLUSION

Results showed that A3 treatment (15 mL of BiTrichompos) could reduce the severity of bacterial wilt disease. Treatment A2 (10 mL of BiTrichompos) showed that it was able to stimulate plant growth by increasing the quantity of fruit and fruit weight, while also promoting root length.

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