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The effectiveness of The Formulation of Cocoa Pod Husk (*Theobroma cacao* L.) Based Botanical Fungicides on Fusarium Wilt Disease on Tomato Plants

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Abstract. The study was conducted to determine the effectiveness of botanical fungicide formulations of cacao pod husk (*Theobroma cacao* L.) on the intensity of fusarium wilt in tomato plants. The research was carried out at the Biology Experimental Laboratory in Universitas Negeri Makassar, South Sulawesi, Indonesia. The study design used a Completely Randomized Design (CRD) with six treatments, namely KSH (control of health treatment without treatment), KSK (control of sick plants treated *F. oxysporum* f.sp licopersicy), BSTR (benomyl + treatment of *F. oxysporum* f.sp licopersicy). EKBK (treatment of cacao pod husk extract + treatment of *F. oxysporum* f.sp licopersicy). The EKBK treatment used cacao pod husk extract with 3 types of concentration, namely: 2% (v/v); 5% (v/v); and 8% (v/v). Each treatment consisted of 10 replications so that the total number was 60 experiment units. Observations included the number of yellowing and deciduous leaves due to the *Fusarium oxysporum* attack on tomato plants. The results showed that botanical fungicides from cacao pod husk extract were able to reduce the intensity of fusarium wilt attacks. At a concentration of 2% (v/v) tomato plants that were attacked by fusarium wilt by 54%; concentration of 5% (v/v) of tomato plants attacked by fusarium wilt at 34.5%; and a concentration of 8% (v/v) of tomato plants attacked by fusarium wilt at 23.8%. The results obtained showed that the intensity of the attacks that occurred in the use of botanical fungicides from cacao pod husk extract was lower than the use of synthetic fungicides (benomyl) which was equal to 39.0%.

Keywords: botanical fungicide, cacao pod husk, fusarium wilt, tomato plant

1. Introduction

Tomato productivity in Indonesia in 2015 was 16.09 tons ha/year [1]. The productivity obtained is still relatively low when compared to China which has been able to reach tomato productivity up to 48.1 tons ha/year [2]. Tomatoes are classified as fruit vegetables that can be planted in various seasons. Therefore, these vegetables can be planted throughout the year both in the rainy season and



in the dry season with results that are relatively not much different, which is important enough water availability.

In tomato cultivation, one of the main obstacles that inhibit production both in quality and quantity is the presence of disease attacks, especially fusarium wilt [3]. In general, farmers were controlled by using synthetic pesticides (chemical) with the assumption that synthetic pesticides are more effective for controlling plant diseases. In fact, if examined more in the use of chemical pesticides have a negative impact on the lives of plants, animals, and humans. In Indonesia, pesticide residues contained in horticultural products such as carrots, potatoes, mustard greens, shallots, tomatoes and cabbage in some vegetable production centers have been reported to have residues that exceed the maximum limit of 2 ppm [4]. In addition, in agribusiness studies, the use of synthetic pesticides can increase agricultural production capital issued because synthetic pesticides are more expensive.

Referring to this, one of the solutions taken is the use of botanical pesticides which are environmentally friendly. Also, the use of botanical pesticides is considered very economical because the materials used in the manufacture of botanical pesticides are easily obtained, and the required costs are relatively cheap so that the farmers can reduce production costs. Based on this, efforts must be made to control pathogenic fungi in tomato plants by using extracts of phenolic cacao peel. The function and habit are ingredients which are single or multiple which are derived from plants (leaves, fruit, seeds or roots) as repellents, pullers, infertility, killers and other forms can control the growth organisms (OPT). Botanical pesticides easy to decompose (biodegradable) in the non-polluting environment, and relatively safe to human beings and livestock ([5]. The results of previous studies of Rachmawaty et al. [6] showed that extracts of cocoa pods using acetone: water (7: 3) can inhibit the growth of *Fusarium oxysporum* in vitro. Based on these, it is necessary to research to find out "Effectiveness of the fruit peel of cocoa (*Theobroma cacao* L.) against fusarium wilt disease in tomato plants in vivo."

2. Research Method

2.1 Time and location

The study was conducted for 6 months, from March to September 2018, at the Green House Department of Biology, FMIPA UNM, Makassar, Indonesia.

2.2 Materials and tools

The materials used were tomato seeds of F1 Lentana variety, cocoa pod husk, tomato pathogen isolates (*Fusarium oxysporum*) obtained from the Biology Laboratory of FMIPA UNM, PDA (Potato Dextrose Agar) media, alcohol 96% and 70%, Acetone, Tween 80, Benomyl and aquades. The tools used in this study were an autoclave, Horizontal Laminar Air Flow Model LH-S, clean band, analytical scales, electric ovens, and binocular microscopes.

2.3 Research design

This study used a Completely Randomized Design (CRD) with six treatments, namely: KSH (control of healthy plants without treatment), KSK (control of sick plants treated *F. oxysporum*), BSTR (Benomyl + treatment *F. oxysporum*), EKBK (extract treatment 2% cocoa pod husk extract + *F. oxysporum* treatment), EKBK (treatment of 5% cocoa pod husk extract + *F. oxysporum* treatment), EKBK (8% cocoa pod husk extract treatment + *F. oxysporum* treatment). Each treatment consisted of 10 replications so that the total number was 60 units of trials. Observation parameters included the number of yellowing leaves and the number of leaves that were killed due to the attack of *Fusarium oxysporum* on tomato plants.

2.4 Preparation and processing samples

The cacao fruit used comes from ripe fruit (yellow), and the fruit is picked directly from the tree. Before processing, the fruit that has been picked is stored for 5 days so that the seeds are easily

released from the fruit. Some cocoa pods in the form of fresh, roughly ground using stone mortar. Some are dried in the sun and after dry, roughly crushed.

2.5 Extraction of cocoa pods in maceration

Each sample of 1 kg cocoa pods (fresh and dry) was repeated 3 times using Acetone : water (7:3) by comparison (1:3) for dry samples.

2.6 Formulation of cocoa pods extract

Pods extracts of the cocoa fruit were formulated to be synthesized and absorbed by adding aquadest sterilized water. The percentage of extract concentration of the cocoa pod husk consisting of 2%; 5%; 8% (g mL^{-1}). The formulation is then used to test the activity of the fungus on *F. oxysporum* in vivo (greenhouse scale). Some positive controls are used for synthetic acid (benomyl)

2.7 Application of cocoa pods extract formula on greenhouse conditions

The application of benomyl fungicides and cocoa pod husk extracts is carried out once every week as much as 100 ml/plant, starting from the first week to the fourth week after planting. The variables observed were inhibition of fungicide in natural and synthetic aspects of fusarium wilt, some yellowed and dead leaves due to *Fusarium oxysporum* attack on tomato plants.

3. Results and Discussion

Treatment of botanical fungicides has a very significant effect on fusarium wilt disease. The average test results of plant fungicide inhibitory power on the growth of pathogenic colonies in vivo are presented in Table 1. Botanical fungicides extract of cacao fruit pods provides a very effective inhibitory effect at a concentration of 5% (v/v), with moderate inhibition or above 50% when compared with positive controls. Whereas in botanical fungicides extracts of cocoa pods which showed the effectiveness of high inhibitory power is at a concentration of 8% (v/v). In previous studies, botanical fungicides extracted from cocoa pods were more effective in inhibiting the development of *F. oxysporum* pathogens in vitro [7].

Table 1. The average percentage of live tomato plants after application of the formulation of cacao pods extract in vivo after four observations

Treatment	Percentage of live plants at week observation (%)			
	I	II	III	IV
KSH	100	100	100	100
KSK	50	10	0	0
EKBK 2%	60	30	20	10
EKBK 5%	100	80	60	50
EKBK 8%	100	98	87	60
BSTR	100	100	80	75

Test results of the formulation of extracts of cocoa pods against *F. oxysporum* cause the cover of tomatoes. So it has been inoculated *F. oxysporum* of 4.6×10^6 spora mL^{-1} , it shows that on observations weeks I and II after the application, the percentage of living seedlings that live can reach 100% at all times, but the observation of the third and fourth quarters is already a change in natural life. Except in the treatment of KSK, the percentage of living tomato plants began to decline, starting from I-IV week observations, all plants died in Table 1. It is generally lowered in the formulation of extracts of 2% and 5% in the observation of week III and week IV. In the 8% extract formulation treatment, there was no decrease in live tomato plants, similar to the treatment of BSTR (which was given an artificial antibiotic), which was not a significant decrease in living life (100%) until week IV observation. The occurrence of ammunition at some point indicates that sharpness is *F. oxysporum*

has been infected, with the adjustments reported by Nelson et al. [8], it starts with abnormal growth in the leaves where the leaves look gloomy green. Symptoms of wilting generally start from the leaves located below and then develop upwards because the base of the stem begins to rot. Withered leaves will turn yellow and eventually dry out, even though the shoot leaves remain green. The use of synthetic fungicides is believed to have the ability to inhibit *F. oxysporum* causes of wilt disease in tomato plants

In observing the percentage of tomato plants affected by fusarium wilt disease in Table 2, the treatment formulation of 2% extract of tomato plants attacked by fusarium wilt was 54%. The higher the treatment of the extract, the lower the tomato plants attacked by fusarium wilt. Treatment formulations of 5% and 8% extracts of tomato plants attacked 34.5% and 23.8%. Lower than BSTR (synthetic fungicide) treatment, tomato plants affected by fusarium wilt were 39%.

Table 2. Percentage of tomatoes affected by fusarium wilt disease

Treatment	Percentage of fusarium wilt disease
KSH	0,0 ^a
EKBK 8%	23,80 ^b
EKBK 5%	34,50 ^b
BSTR	39,00 ^{bc}
EKBK 2%	54,00 ^c
KSK	100,00 ^d

Note: KSH = control plantings treatment, KSK = control of fusarium fungus treatment, 2% EKBK = 2% mushroom treatment + 2% extract, EKBK 5% = 5% mushroom treatment + 5% cocoa pods extract, 8% EKBK = 8% mushroom treatment + 8% cocoa pods extract, BSTR = treatment mushroom + benomyl

The high tomato plants attacked by fusarium wilt at a concentration of 2% because they have not been able to attack *F. oxysporum*. This has seen a large number of tomatoes are dying on these treatments in Table 2. The statistical difference was significant ($P < 0.05$), both of the above treatments (KSK and EKBK 2%) with the application of the 5% ECB and 8% EKBK. The 8% correlation between BEC and BSTR (synthetic fungicide) was significantly different ($P < 0.05$) where the average percentage of tomato plants disease with fusarium wilt was significantly different, synthetic fungicide treatment was still higher with fusarium wilt than 8% extract treatment. Phytochemical test results, the active ingredients contained in cocoa-allergic steroid, terpenoid, saponin, and phenolic fruit peel extracts [6]. Pods extracts of the cocoa capable of inhibiting the growth of *F. oxysporum*, it caused phenolic can also change the protein properties and narrow the cells which can melt the mold and mushrooms. Also, the ammonia compound is always passed to the hydroxyl bond and is usually sulfurized from the fungal protein so that the protein transformation changes the target [9]. In addition to flavonoids and polyphenols, saponins in cocoa pods may also contribute to the antimicrobial activity. Saponins have activity as fungi.

4. Conclusion

Cocoa pods extracts have potency in inhibiting *Fusarium oxysporum*. At a concentration of 2% (v/v), tomato plants attacked by fusarium disease were 54%, the concentration of 5% (v/v) tomato plants attacked by fusarium disease was 34.5% and the concentration of 8% (v/v) of tomato plants attacked by fusarium amounting to 23.8% lower than the use of synthetic fungicides (benomyl) of 39.0%.

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