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Effect of Fungicidal Methanol Extract of *Kirinyuh* Leaves (*Eupatorium odoratum* L.) and Noni Leaves (*Morinda citrifolia* L.) against MOLD *Fusarium oxysporum* on Tomatoes

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Abstrak: Tanaman tomat merupakan salah satu komoditas hortikultura unggulan yang mudah terserang penyakit. Penyakit layu tanaman merupakan salah satu penyakit yang sering menyerang tanaman tomat yang disebabkan oleh *Fusarium oxysporum*. Penggunaan fungisida kimia banyak menyebabkan kerugian, sehingga perlu adanya fungisida nabati yang terbuat dari bahan alami. Daun Kirinyuh dan daun Mengkudu merupakan bahan alami yang dapat dijadikan fungisida nabati untuk mengendalikan jamur *F. oxysporum*. Percobaan dilakukan secara *in vivo* disusun menggunakan Rancangan Acak Lengkap dengan 6 perlakuan 6 ulangan. Data yang diperoleh dari hasil pengamatan akan dianalisis menggunakan uji duncan pada taraf 5%. Konsentrasi yang digunakan oleh masing-masing ekstrak yaitu 20%, 40%, 60%, dan 80%, kontrol negatif (aquadest), kontrol positif (dithane M-45). Hasil menunjukkan bahwa fungisida ekstrak daun Mengkudu dengan konsentrasi 60% memiliki pengaruh paling efektif dalam menghambat zona pertumbuhan jamur *F. oxysporum* pada buah tomat.
Kata kunci: Fungisida nabati; *Fusarium oxysporum*; kirinyuh; mengkudu; tomat.

Abstract: Tomato plants are one of the leading horticultural commodities that are easily attacked by disease. Plant wilt disease is a disease that often attacks tomato plants and is caused by *Fusarium oxysporum*. The use of chemical fungicides causes many losses, so there is a need for plant-based fungicides made from natural ingredients. Kirinyuh leaves and Noni leaves are natural ingredients that can be used as vegetable fungicides to control fungi such as *F. oxysporum*. The experiment was carried out alive and prepared using a Completely Randomized Design with six treatments and six replications. Data obtained from observations will be analyzed using the Duncan test at the 5% level. The concentrations used by each extract were 20%, 40%, 60%, and 80%, negative control (Aquadest) and positive control (distance M-45). The results showed that the Noni leaf extract fungicide with a concentration of 60% had the most effective effect in inhibiting the fungal growth zone *F. oxysporum* on tomatoes.
Keyword: Fungicides; *Fusarium oxysporum*; Kirinyuh; Noni; Tomatoes.

1. Introduction

According to the Directorate General of Horticulture, Tomato plants are among the leading domestic horticultural commodities based on their economic and strategic value [1]. So, this must be supported by increasing the number of tomato plant harvests to meet market needs. However, tomato plants are a horticultural crop that is easily attacked by disease. According to Balai Penyuluhan Pertanian (Agricultural Extension Center) Sedayu in 2017, the productivity of tomato plants decreased due to various environmental factors and Plant Pest Organism (OPT) factors such as septoria leaf spot (36.16%), green aphids (29.18%), thrips aphids (27.16%), anthracnose disease (23.20%), wilt disease Fusarium (22.77%) and fruit flies (12.90%) [2].

Fusarium diseases caused by pathogenic fungi are still a significant problem today because controlling and preventing these diseases' development has been ineffective [3]. This is due to the characteristics of the fungus, which can spread through the soil or air very quickly. One of the fungi that easily spreads in the soil is *Fusarium oxysporum*. This fungus is a pathogen with a self-defence system, namely chlamydospores, which allows it to live in the soil even though it does not have a host [4]. Mold *Fusarium oxysporum* often infects through injured plant roots [2].

Mold *Fusarium oxysporum* will enter the roots through injured roots, tissue, and lateral roots. The fungus will grow in the vascular bundles until the mycelium reaches the xylem vessels. The status of the xylem, which carries water and nutrients throughout the plant body, will also carry mycelium from fungi. After the mycelium spreads throughout the body, the plant will wilt. After that, the entire body of the tomato plant will wilt until it dies [5]. Apart from that, fusarium causes tissue necrosis [6].

Death of tomato plants will result in significant losses for farmers. So farmers carry out fungus eradication of *Fusarium oxysporum* using chemical fungicides. Using chemical fungicides is the quickest and easiest way to eradicate various fungi and diseases. Chemical fungicides pollute the

environment, causing plants, microbes, and animals around them to die [5]. Apart from that, the fruit obtained will also carry residue from fungicides, so it will not have a good impact on human health. Therefore, other alternatives are needed to replace chemical fungicides, namely bio fungicides derived from plants and other natural ingredients.

Bio fungicides will be safer to use to kill fungi. This is because vegetable fungicides will only kill the target fungus and no other organisms [7]. Biofungicides will reduce the residue produced after using fungicides, and they are easily decomposed so that the surrounding environment will not be polluted easily. The materials used in making fungicides are also easier to find in the environment. Therefore, plant extracts have the potential to be used as alternative fungicides [8].

Kirinyuh leaves and Noni leaves are examples of natural ingredients that have been widely studied because they have beneficial properties. Kirinyuh contains secondary metabolite compounds such as phenols, triterpenoids, flavonoids, and alkaloids, which can be used as antifungals. Likewise, the Noni plant contains various substances, such as anthraquinones, which function as antimicrobials, and flavonoids, which can inhibit the growth of conidia in fungi [9]. Kirinyuh leaves and Noni leaves can be converted into vegetable fungicides using several techniques. Therefore, in research, The antifungal activity of the methanol extract of Kirinyuh and Noni leaves was tested with concentrations of 20%, 40%, 60%, and 80% against fungi. *Fusarium oxysporum* on tomato esalive.

2. Research Method

This research was carried out in March 2024. The study was conducted at the Biology Learning Laboratory, Faculty of Teacher Training and Education, Ahmad Dahlan University, Yogyakarta. This research was carried out randomly alive. The treatments were arranged using the RAL (Completely Randomized Design) method with six treatments and six replications. Treatment consisted of (P1) positive control, namely Dithane M-45, (P2) negative control, namely distilled water, (P3) extract with a concentration of 20%, (P4) extract with a concentration of 40%, (P5) extract with a concentration

of 60%, (P6) extract concentration 80%. Data analysis used the Duncan test with a level of 5%.

a. Making Simplicia of Kirinyuh Leaves and Noni leaves

Kirinyuh leaves and Noni leaves are taken from parts that don't have insect bite marks. Kirinyuh leaves and Noni leaves are washed in running water, drained, and wiped with a clean cloth. Next, the leaves are placed at room temperature and left to dry. The leaves are cut into smaller pieces and blended until the leaves are smaller.

b. Making Kirinyuh and Noni Leaf Extracts

The blended leaves were then weighed 200 g using an analytical balance. Next, the weighted leaves are put into a jar and macerated using 3000 ml of 80% methanol for 3x24 hours and stirred every 1x24 hours. Once the process is complete, it is filtered to obtain a filtrate. Next, distillation is carried out by inserting the filtrate into a three-neck flask that was previously assembled and ensuring that it is tight. Next, the electric stove and the water flow are turned on. Once the distillate droplets decrease, the electric stove can be turned off.

Varying extract concentrations were made using dilution with distilled water. Concentration consists of 20%, 40%, 60%, 80%. Concentration variations are made using the formula P1xV1=P2xV2. The negative control used Aquades (without extract), and the positive concentration used dithane M-45 with a concentration of 5 g/l.

c. Fungal Infection *Fusarium oxysporum* on Tomatoes

The tomatoes are washed using clean running water and then dried. Next, the tomatoes were sprayed using Bayclin 1% and dried at room temperature. Next, the tomatoes are labeled according to the treatment; on the right side, they are sprayed with Noni leaf extract, and on the left, they are sprayed with 1 ml of Kirinyuh leaf extract. Wait until the treatment solution has been sprayed on the tomatoes; then, the treatment solution has absorbed the tomatoes, and the surface is dry. After the tomatoes are dry, the mushrooms are injected with a *Fusarium oxysporum* of 0.5 ml with a needle depth of 1 mm.

d. Observation of fungal growth zones *Fusarium oxysporum* on tomatoes

Observations were carried out twice, namely at three days and six days. Observation of fungal growth zones *Fusarium oxysporum* Measure directly using a ruler on the part of the tomato that grows white hyphae on the injured side.

3. Results and Discussion

Based on research carried out and analyzed by Duncan, it can be seen that the most effective ingredients used in inhibiting the fungal growth of Fusarium oxysporum are Noni leaves, which can be seen in Table 1.

Table 1. Average area of fungal growth zone *Fusarium oxysporum* for 3 DAI and 6 DAI with different material treatments

Materials	Installment-Installment (cm)		Increasing the Area of Mushrooms
	3 DAI	6 DAI	
Methanol-Noni	0,9375[a]	1,9042[a]	0,9667
Methanol-Kirinyuh	0,8917[a]	2,0250[a]	1,1333
Dithane M-45 (K. Positive)	0,8667[a]	2,7583[ab]	1,8916
Aquades (K. Negative)	0,9583[a]	3,4500[b]	2,4917

Information: The same letter notation indicates that the data are not significantly different in the Duncan test with α 0.05

Based on the results of the Duncan test, it can be seen that the methanol extract treatment of Kirinyuh leaves and Mengkudu leaves was significantly different from the adverse control treatment (*Aquadest*). This shows that Noni and Kirinyuh leaves' methanol extract can inhibit the fungal growth of *Fusarium oxysporum*. The most significant increase in the area of the fungal growth zone with the adverse control treatment using distilled water showed a result of 2.4917 cm. Noni leaf extract treatment showed the most minor results, namely 0.9667 cm. Growth *Fusarium oxysporum influenced* by the fungicide used.

Noni leaf extract and Kirinyuh leaf extract can inhibit the fungal growth of Fusarium oxysporum because they contain secondary metabolites. Noni leaf extract analysis results were positive for containing alkaloid, flavonoid, and terpenoid compounds as antifungals. Kirinyuh leaves also contain bioactive compounds, including saponins, tannins, alkaloids, phenolics, and flavonoids, which can be fungicides [10].

Flavonoids and phenolics have antimicrobial activity by destroying proteins, causing cell walls to become brittle so bioactive compounds can easily penetrate them [11]. Alkaloids are compounds that can inhibit the biosynthesis

of fungal nucleic acids so that fungal cells die because they cannot develop [12]. Terpenoids, which are fungistatic, can hinder the work of specific enzymes, which results in the disruption of fungal cell metabolism so that the elongation process of fungal hyphae is hampered and causes fungal cells to be unable to reproduce within a specific time [13].

Saponin can cause microbial cell lysis by disrupting the stability of the cell membrane. As a polar surfactant, Saponin will reduce the surface tension of the sterol membrane of the fungal cell wall, thereby causing disruption of membrane permeability, which results in the entry of necessary materials or substances being disrupted and, ultimately, the cells swelling and bursting [14]. Tannin compounds also have antifungal activity, which can shrink fungal cell walls, as a result of which the permeability of the fungal cell walls will be disrupted so that the fungal cell walls will not be able to carry out cell metabolic activities [15].

The following observation is the average area of the fungal growth zone *Fusarium oxysporum* with different concentrations and material treatments, as shown in Table 2.

Table 2. Average area of fungal growth zone *Fusarium oxysporum* for 3 DAI and 6 DAI with various concentrations and different ingredients

Combination of Concentration and Ingredients	Installment-Installment (cm)		Increase in Mushroom Area (cm)
	3 DAI	6 DAI	
M 20%	0,9167[a]	2,0333[ab]	1,1166
M 40%	0,9500[a]	1,7167[a]	0,7667
M 60%	0,7500[a]	1,1167[a]	0,3667
M 80%	1,1333[a]	2,7500[ab]	1,6167
K 20%	0,9167[a]	2,2500[ab]	1,3333
K 40%	0,9000[a]	2,0000[ab]	1,1
K 60%	0,7167[a]	2,0000[ab]	1,2833
K 80%	1,0333[a]	1,8500[ab]	0,8167
Positive (Dithane M-45)	0,8667[a]	2,7583[ab]	1,8916
Negative (*Aquadest*)	0,9583[a]	3,3500[b]	2,3917

Information: The same letter notation indicates that the data are not significantly different in the Duncan test with α 0.05

The area of the fungal growth zone *Fusarium oxysporum* for 3 DAI and 6 DAI with various concentrations and different ingredients can also be seen in Figure 1 and Figure 2.

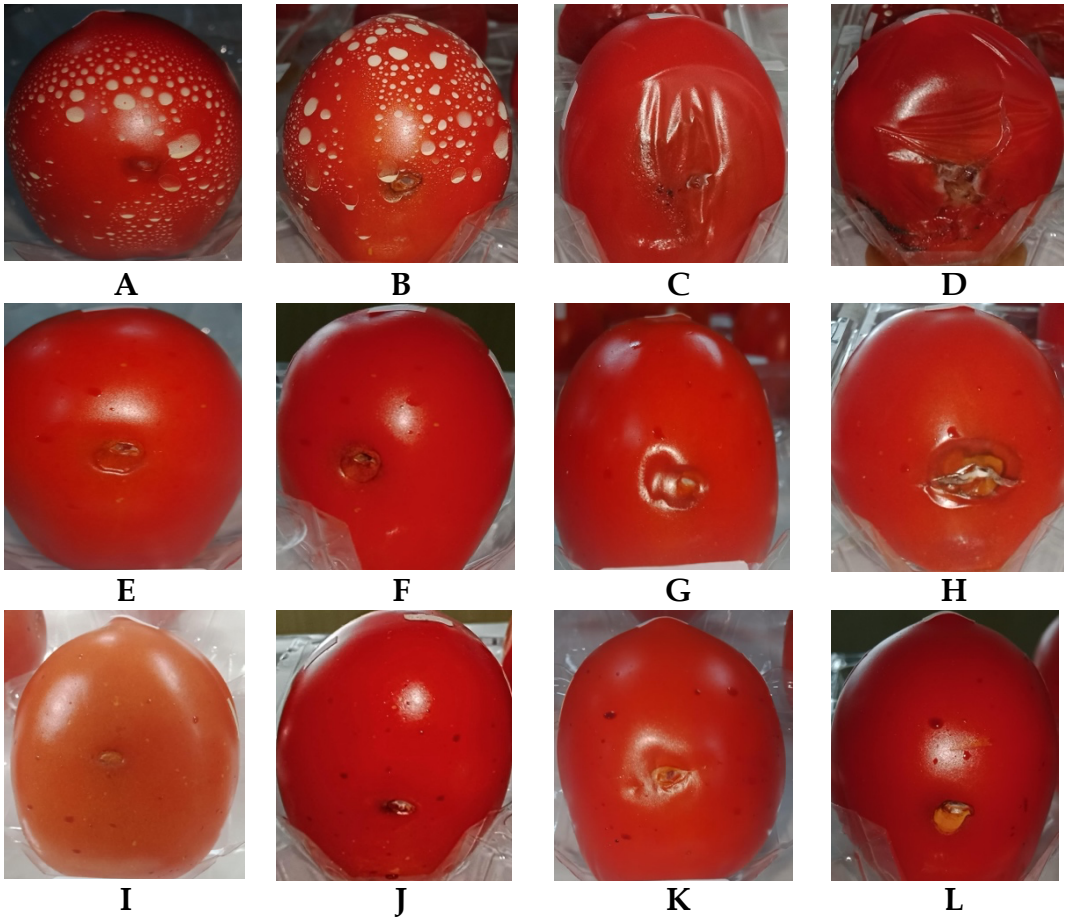
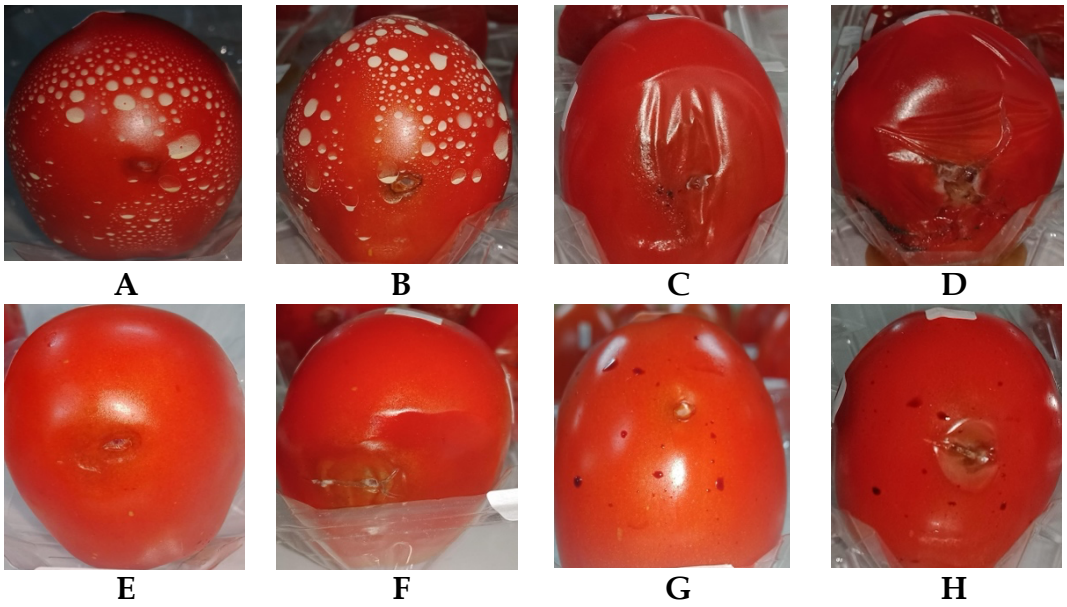


Figure 1. The area of the fungal growth zone *Fusarium oxysporum* in several treatments the methanol extract concentration of Kirinyuh leaves: A=3 DAI P1; B=6 DAI P1; C=3 DAI P2; D=6 DAI P2; E=3 DAI P3; F=6 DAI P3; G=3 DAI P4; H=6 DAI P4; I=3 DAI P5; J=6 DAI P5; K=3 DAI P6; L=6 DAI P6



I J K L

Figure 2. The area of the fungal growth zone *Fusarium oxysporum* in several treatments the concentration of noni leaf methanol extract: A=3 DAI P1; B=6 DAI P1; C=3 DAI P2; D=6 DAI P2; E=3 DAI P3; F=6 DAI P3; G=3 DAI P4; H=6 DAI P4; I=3 DAI P5; J=6 DAI P5; K=3 DAI P6; L=6 DAI P6

Information:
P1: Positive control (Dithane M-45)
P2: Negative control (Aquadest)
P3: Methanol concentration 20%
P4: Methanol concentration 40%
P5: Methanol concentration 60%
P6: Methanol concentration 80%

Based on Table 2, the average area of the fungal growth zone *Fusarium oxysporum* with observations for 3 DAI showed no significant fundamental differences between treatments. Observations at 6 DAI showed that noni leaf extract with concentrations of 40% and 60% significantly differed from the negative control but not substantially different from other treatments. Increase in the fungal growth zone *Fusarium oxysporum* is the smallest was the noni leaf extract treatment with a concentration of 60%, namely 0.3667 cm. Noni leaf extract with a concentration of 60% effectively inhibits fungal growth of *Fusarium oxysporum*. Meanwhile, for Kirinyuh leaf extract, the smallest increase in fungal area was in the 80% treatment, namely 0.8167 cm.

4. Conclusion

Based on the research results, it can be concluded that Noni leaf extract fungicide with a concentration of 60% is more effective than Noni extract fungicide with concentrations of 20%, 40%, and 80%, and Kirinyuh leaf extract fungicide with concentrations of 20%, 40%, 60%, and 80%. Noni leaf extract fungicide is known to control wilt disease caused by fungi *Fusarium oxysporum* on tomatoes.

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