Synthesis and characterization of Cu-doped TiO\(_2\) (Cu/TiO\(_2\)) nanoparticle as antifungal **phytophthora palmivora**

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**Abstract.** Preparation of Cu-doped TiO\(_2\) nanoparticles (Cu/TiO\(_2\)) as antifungal Phytophthora palmivora has been carried out. In this study, we did a synthesis of Cu/TiO\(_2\) nanoparticles by the sol-gel method and determine the performance of the modification of Cu/TiO\(_2\) nanoparticles as antifungal agents P. palmivora. The stages of this research include synthesis of Cu/TiO\(_2\) nanoparticles using the sol-gel method, characterization of Cu/TiO\(_2\) nanoparticles using UV-Vis DRS and characterization of Cu/TiO\(_2\) nanoparticles using SEM-EDX, and P. palmivora antifungal activity test. TiO\(_2\) is known to be used as an antifungal agent and to increase photocatalytic activity, TiO\(_2\) is doped with Cu metal ions to be active in visible light. The results showed that Cu/TiO\(_2\) has a band gap energy of 3.12 eV. Based on the results of SEM-EDX analysis of TiO\(_2\)-Cu nanoparticles confirmed the presence of elements Cu (2.16%), Ti (28.74%) and O (69.10%) with morphological shapes such as irregular spheres. Cu/TiO\(_2\) nanoparticles have a strong ability at concentrations of 0.05% and 2.5% in inhibiting the growth of the fungus P. palmivora. Percent inhibition respectively 60.41% and 75% and at concentrations>3% have activities that can kill P. palmivora fungus.

1. **Introduction**

The productivity of cocoa yield in several countries such as Ivory Coast, Ghana, Nigeria, Cameroon, Brazil, Ecuador, Malaysia, and Indonesia has been reported to have decreased due to Phytophthora palmivora fungus attacks that cause rot on cocoa plants\(^{[1,2]}\). Various attempts have been made including the use of spraying with chemical fungicides. But the use of chemical fungicides is also a major source of heavy metal pollution on plantation soils. Aside from being a source of pollutants, the use of chemical fungicides has not shown optimal performance due to the nature of compounds that are unstable and resistant to fungi. In addition, the use of chemical fungicides is also very expensive, for that we need an alternative material that can be used to replace chemical fungicides\(^{[3]}\).

The development of anti-fungus based on TiO\(_2\) nanoparticles was reported as an interesting study to overcome various pests and plant diseases. Titanium dioxide is a semiconductor material used as photocatalysts, solar cells, biological sensors, and antimicrobial agents\(^{[4–7]}\). Among the existing semiconductors, TiO\(_2\) is a good semiconductor used because it is non-toxic, has high thermal stability, corrosion resistance and abundant availability in nature, so the price is relatively cheap\(^{[8–10]}\). From the results of several studies conducted, TiO\(_2\) nanoparticles showed excellent performance in inhibiting the growth of fungi such as Fusarium oxysporum, Helminthosporium maydis, Asperillus niger, Fusarium graminearum, Hypocrella xii, and Mucor circinelloidei\(^{[11]}\).

To increase antifungal activity, the surface of TiO\(_2\) can be modified through metal or non-metal element insertion. Doping TiO\(_2\) with nonmetal atoms such as carbon (C) \(^{[12]}\), sulfur (S) \(^{[13]}\), fluorine
(F) [14], phosphorus (P) [15], nitrogen (N) [16] and boron (B) [17] have been shown to cause TiO$_2$ to absorb visible light in the wavelength range ($\lambda$) 450-600 nm. In addition to non-metals, some metal ions are also used as dopants in TiO$_2$ semiconductors including using dopants Fe, Cu and Ce have been shown to be able to shift photocatalytic uptake in the direction of visible light Popa et al. [18]. In this work, we have successfully synthesized Cu/TiO$_2$ composites by the sol-gel method. Cu is also reported to have high toxicity to microorganisms and is able to widen the working area of TiO$_2$ to visible light so that it is widely used as a material modification. Cu/TiO$_2$ composites will be applied as an anti-fungal agent P. palmivora in overcoming cocoa plants.

2. Experimental Methods
Preparation of Cu/TiO$_2$ composite
Synthesis of Cu/TiO$_2$ composite was conducted by mixing of 15 mL ethanol, 2 mL Aquades, 1 mL acetic acid (0.5 M) into a reflux flask which is containing 4 mL of Titanium tetra-isopropoxide (TTIP), 0.5 mL acetylacetone, and 15 mL ethanol. The mixture was then refluxed for 3 hours at temperature 50 °C. Sol TiO$_2$ was stirred using a magnetic stirrer for 1 hour at 50 °C followed by adding 2 mL of CuSO$_4$.5H$_2$O as a dopant source. It evaporates at room temperature for 48 h to form a gel and heated at 80°C for 30 min. To obtain the Cu/TiO$_2$ powder, we calcinate the sol-gel Cu/TiO$_2$ composite at temperature 500°C for 1 h to form Cu/TiO$_2$ anatase phase and characterized by using UV-Vis Diffuse Reflectance Spectroscopy (UV-DRS) and Scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM/EDX).

2.1. Antibacterial activity test
The antibacterial activity test was carried out using Cu/TiO$_2$ composites by the dilution method. Cu / TiO$_2$ composites were prepared with various concentrations as shown in Table 1. As for control (-) using P. palmivora fungus inoculated on solid PDA media without Cu/TiO$_2$ composites and control (+) using the Dithane M-45 fungicide.

Table 1. Variation concentration and mass of Cu/TiO$_2$ composites as antifungal P. palmivora

| No. | Concentration of Cu/TiO$_2$(%) | Mass of Cu/TiO$_2$ (g) | PDA media volume (mL) |
|-----|-------------------------------|------------------------|-----------------------|
| 1.  | 0.05                          | 0.005                  | 10                    |
| 2.  | 2.5                           | 0.25                   | 10                    |
| 3.  | 3                             | 0.30                   | 10                    |
| 4.  | 3.5                           | 0.35                   | 10                    |
| 5.  | 4                             | 0.40                   | 10                    |
| 6.  | 4.5                           | 0.45                   | 10                    |
| 7.  | 5                             | 0.5                    | 10                    |

3. Results and Discussion

3.1. Preparation of Cu/TiO$_2$ composite
In this study, we used an alkoxide source as a TTIP precursor serving the distribution media for dopant ions to form TiO$_2$ and Cu/TiO$_2$ composites. The addition of ethanol to the sol-gel process serves as an inhibitor of the hydrolysis of the precursors so as to maintain the alkoxy metal reactivity for the formation of more stable soles. So that the hydrolysis and condensation processes for the formation of Ti-O-Ti polymers are increasingly optimal. While the addition of acetyl acetate functions as a ligand to chelate titanium so the solution is yellow and to form mesostructure TiO$_2$anatase[19–22].

Furthermore, TTIP, acetyl acetate, ethanol, glacial acetic acid, and distilled water were refluxed for three hours at 50°C to increase interactions between compounds [23–25]. The TiO$_2$ sol is stirred using a magnetic stirrer for 1 hour at 50°C followed by the addition of 2 mL CuSO$_4$.5H$_2$O as a copper dopant source Figure 1a.
The sol which contains Cu then evaporates the solvent to form a gel. The use of CuSO₄·5H₂O as a dopant based on research conducted by Yadav et al. [26] proved that CuSO₄·5H₂O effectively acted as a source of Cu to reduce the value of the TiO₂ bandgap. The Cu/TiO₂ composite obtained was then calcined at 500°C for 3 hours to remove solvents such as water and ethanol. The results obtained after the calcination look as shown in Figure 1b.

![Figure 1. Sol-gel Cu/TiO₂ composite (a), Powder of Cu/TiO₂ composite (b)](image)

3.2. **UV-Vis Diffuse Reflectance Spectroscopy (UV-Vis DRS)**

Measurement using DRS to determine the absorption character in areas with both UV and visible wavelengths with a wavelength range of 200-800 nm and determine the bandgap of TiO₂ and Cu/ TiO₂ composites. The results of the characterization using UV-Vis DRS can be seen in Figure 2.

![Figure 2. Bandgap spectra of TiO₂ compared with Cu/TiO₂ composite](image)

Based on Figure 2 it can be seen the value of the energy bandgap on TiO₂ of 3.27 eV, appropriate to the reported by Zhang et al. [27]. TiO₂ which was doped with Cu obtained a bandgap energy value of 3.12 eV. These results indicate that Cu dopant can reduce the bandgap of TiO₂ energy. Cu/TiO₂ composite bandgap values obtained in this study are in line with research conducted by Yadav et al. [26] and Ambrozova et al. [28] in his research has successfully synthesized doped Cu TiO₂ in the wavelength range of 300-700 nm and obtained bandgap energy of 2.95 eV and 3.06 eV, respectively. Copper doping has been proven to narrow the bandgap energy, increase the hydrophilicity of TiO₂ and increase the photocatalytic activity of TiO₂[29–32].

3.3. **Scanning electron microscopy with energy dispersive X-ray spectroscopy (SEM/EDX)**

The characterization morphology of Cu/TiO₂ was carried out by using SEM/EDX. Figure 3 showed that qualitatively TiO₂ and Cu/TiO₂ are relatively different but generally consist of irregular spherical particles and microstructure in the gaps of particles. An image of Cu/TiO₂ composites shows gray particles with a more dominant microstructure. White particles indicate the presence of TiO₂ while gray particles indicate TiO₂ which has been doped with copper. The addition of copper dopants causes a decrease in particle size compared to TiO₂ without dopants. A decrease in particle size and an
increase in the surface area indicate an increase in photocatalytic activity. This indicates that photocatalyst material which has a smaller size can provide better photocatalyst activity.

![EDX analysis result](image)

**Figure 3.** Morphology and elements characterization of Cu/TiO$_2$ composite

EDX analysis is performed to find out the percentage of the constituent elements resulting from the synthesis. The results of the characterization using EDX can be seen in Figure 3. Based on the results of the characterization obtained it can be seen that the peak of Cu at 8.0 KeV, the peak of Ti at 4.5 KeV and the peak of O at 0.51 KeV. Cu peaks that are seen indicate the success of the doping process on TiO$_2$[30]. The EDX analysis of Cu/TiO$_2$ composites confirmed the presence of Cu, Ti and O elements with 69.10, 28.74 and 2.16, respectively. The peak position obtained in the EDX spectra is as reported by Segne et al. [33] and Chen et al. [34].

3.4. Antifungal Activity test of Cu/TiO$_2$ composite

Antifungal activity testing was carried out to determine the performance of Cu/TiO$_2$ composites for P. palmivora fungi on cocoa plants. Antifungal activity test of Cu/TiO$_2$ composites was made with various concentrations namely 0.05%, 2.5%, 3%, 3.5%, 4%, 4.5% and 5%. The results of the Cu/TiO$_2$ composite antifungal against P. palmivora fungus were observed in negative control for one week. Characteristics of P. palmivora colonies are generally round with uneven edges and white in color.
**Figure 4.** Test of Cu/TiO₂ composite antifungal activity against P. palmivora fungi; (a) 0.05% Cu/TiO₂; (b) 2.5% Cu/TiO₂; (c) 3% Cu/TiO₂; (d) 3.5% Cu/TiO₂; (e) 4% Cu/TiO₂; (f) 4.5% Cu/TiO₂; (g) 5% Cu/TiO₂; (h) 0.2% fungicide as positive control (+); (i) P. palmivora fungi and media as negative controls (-)

Based on Figure 4, it can be seen that the Cu/TiO₂ composite has a very good ability to inhibit P. palmivora fungus. This can be seen at a concentration of 3% - 5% which is the concentration that has the greatest inhibition and the ability is the same as the Dithane M-45 function where there is no significant colony formation on P. palmivora fungi[3,35,36]. This is in line with what was reported by Chen et al. [34] that Cu/TiO₂ has the ability as a strong antimicrobial agent. Based on measurements of growth inhibition of fungal colonies using the formula Sharma and Pandey[37] and Aulifa et al. [38] obtained the average inhibition value of P. palmivora fungal colonies as shown in Table 2.

| Sample concentration of Cu/TiO₂ (%) | Diagonal diameter (cm) | Inhibition (%) |
|-------------------------------------|------------------------|----------------|
| 0.05                               | 1.90                   | 60.41          |
| 2.5                                | 1.20                   | 75             |
| 3                                  | 0                      | 100            |
| 3.5                                | 0                      | 100            |
| 4                                  | 0                      | 100            |
| 4.5                                | 0                      | 100            |
| 5                                  | 0                      | 100            |

Table 1 shows that the anti-fungal activity at 0.05% Cu/TiO₂ concentration and 2.5% Cu/TiO₂ had strong activity against P. palmivora fungus with inhibition rates of 60.41% and 75%, respectively, and at concentrations of Cu/TiO₂ > 3% have very strong activity against P. palmivora fungi.

4. **Conclusion**

In this research, we successfully synthesized Cu/TiO₂ composites by the sol-gel method. The results showed that Cu/TiO₂ has a bandgap energy of 3.12 eV. Based on the results of the SEM-EDX analysis of Cu/TiO₂ composite confirmed the presence of elements Cu (2.16%), Ti (28.74%) and O (69.10%) with morphological shapes such as irregular spheres. Cu/TiO₂ composite have a strong ability at concentrations of 0.05% and 2.5% in inhibiting the growth of the fungus P. palmivora. Percent
inhibition respectively 60.41% and 75% and at concentrations >3% have activities that can kill P. palmivora fungus.

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