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REVIEWER'S REPORT

Manuscript No.: IJAR-50680 Date:15-03-2025

Title: In vitro Effect of Copper Oxychloride Nanoparticles on Fusarium Wilt Disease Resistance in *Solanum lycopersicum* Through Seedling Root Treatment

Recommendation:
Accept as it is without modification

Rating	Excel.	Good	Fair	Poor
Originality	√			
Techn. Quality	√			
Clarity	√			
Significance		√		

Reviewer Name: Dr. Manju M **Date:** 15-03-2025

Reviewer's Comment for Publication.

1. Copper oxychloride (COC) nanoparticles (NPs) showed strong antifungal activity against *Fusarium oxysporum*, with a minimum inhibitory concentration (MIC) of 32 µg/mL, demonstrating their potential as an effective treatment for Fusarium wilt in tomatoes.
2. Seedlings treated with COC NPs and Ag-doped COC NPs showed significantly better growth in terms of root and shoot length compared to untreated seedlings, indicating that these nanoparticles promote plant health and enhance disease resistance.
3. The NP treatments, particularly at 8 mg concentration, exhibited a synergistic effect in helping tomato seedlings resist Fusarium wilt, with minimal wilting observed even after two weeks of exposure to the pathogen.
4. The study introduced a simple and rapid method for evaluating the effectiveness of nanoparticle treatments by observing the response of NP-coated plants to pathogen inoculation, offering a useful tool for screening other bioagents for their potential disease-fighting properties.

Detailed Reviewer's Report

1. **Background and Problem**:

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- Tomatoes (Solanum lycopersicum) are highly produced globally, and Fusarium wilt, caused by the fungus *Fusarium oxysporum*, is a major pathogen leading to significant yield losses in tomato crops.
- Fusarium oxysporum is a soil-borne pathogen that attacks the vascular system of the plant.

2. **Nanoparticles as a Solution**:
   - Copper (Cu) nanoparticles (NPs) and copper oxychloride (COC) NPs were previously explored for their antifungal activity against plant diseases.
   - This study investigates the use of copper oxychloride (COC) NPs and silver (Ag)-doped copper oxychloride NPs as potential solutions for managing Fusarium wilt in tomatoes.
3. **Objective of the Study**:
   - To explore and compare the effectiveness of COC NPs and Ag-doped COC NPs against *Fusarium oxysporum*, assessing their antifungal activity and the minimum inhibitory concentration (MIC).
   - To evaluate the disease resistance and growth enhancement in tomato seedlings treated with these nanoparticles.
4. **Antifungal Activity and MIC**:
   - COC NPs exhibited a minimum inhibitory concentration (MIC) of 32 µg/mL against *Fusarium oxysporum*.
   - Ag-doped COC NPs showed a MIC of 64 µg/mL, indicating that COC NPs are more effective than the Ag-doped variant at inhibiting the pathogen.
5. **Methodology: Root Dip Treatment**:
   - Tomato seedlings were treated with COC NPs and Ag-doped COC NPs using a root dip method.
   - Seedlings were coated with 8 mg of either COC NPs or Ag-doped COC NPs and then exposed to *Fusarium oxysporum*.
6. **Growth of Seedlings Post-Treatment**:
   - Seedlings treated with COC NPs and Ag-doped COC NPs exhibited excellent growth in both root and shoot length, even in the presence of the wilt pathogen.
   - Only a very small amount of wilting was observed at the 8 mg NP concentration after 2 weeks.
7. **Control Group Comparison**:
   - Control seedlings, which were not treated with any NPs, showed visible wilting within just one week of exposure to *Fusarium oxysporum*.
   - This contrasts with the NP-treated seedlings, which showed better resistance to the pathogen.

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8. **Chlorophyll and Carotenoid Estimation**:
   - Chlorophyll and carotenoid levels were measured to assess the physiological health of the seedlings.
   - The results revealed that NP-treated seedlings had higher levels of these pigments, suggesting enhanced plant health and resistance to stress.
9. **Synergistic Effect of NPs**:
   - The study showed that at the 8 mg concentration, the combined effect of COC NPs or Ag-doped COC NPs with *Fusarium oxysporum* provided a synergistic effect that promoted early-stage disease resistance.
   - The treated seedlings were more resistant to the pathogen's effects compared to untreated seedlings.
10. **Minimum Fungicidal Activity**:
    - The minimum fungicidal activity of the NP treatments was confirmed by plating the NP concentrations from 1 µg/mL to 128 µg/mL, which showed clear evidence of the NPs' ability to inhibit fungal growth at specific concentrations.
11. **Conclusion and Significance**:
    - The study concludes that COC NPs and Ag-doped COC NPs show strong potential for managing Fusarium wilt in tomatoes.
    - The NP treatments improve seedling growth, induce disease resistance, and provide a simple, rapid method for screening other bioagents for their antifungal effectiveness.
    - The use of these nanoparticles could be a promising alternative to traditional chemical treatments, reducing reliance on harmful pesticides while promoting sustainable agricultural practices.

- Copper and silver-doped nanoparticles can be used to manage Fusarium wilt and other soil-borne diseases in crops like tomatoes.
- Nanoparticle coatings can enhance early-stage disease resistance and promote healthy seedling growth.