WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
Coden USA: WJPRAP
Impact Factor 8.63
Volume 15, Issue 7, 854-869.
Research Article
ISSN 2277–7105

BIOLOGICAL SYNTHESIS OF *MANCOZEB AND HEXACONAZOLE CONJUGATED* SILVER NANOPARTICLES FROM GOAT URINE AND THEIR ANTIFUNGAL EFFICACY AGAINST *COLLETOTRICHUM GLOEOSPORIOIDES*

Manoj S. P.[1], Shrinidhi Bhandari K. T.[2], A. N. Rajeshwara[3], S. E. Neelagund[4], Raghavendra S. N.*

[1,2,3,4,*]Department of Biochemistry, Kuvempu University, Jnanasayhadri, Shankarghatta, Shivamogga – 577451.

Article Received on 02 March 2026,
Article Revised on 23 March 2026,
Article Published on 01 April 2026,

https://doi.org/10.5281/zenodo.19329851

***Corresponding Author**
Raghavendra S. N.
Department of Biochemistry, Kuvempu University, Jnanasayhadri, Shankarghatta, Shivamogga – 577451.

How to cite this Article: Manoj S. P.[1], Shrinidhi Bhandari K. T.[2], A. N. Rajeshwara[3], S. E. Neelagund[4], Raghavendra S. N.*. (2026). Biological Synthesis of Mancozeb and Hexaconazole Conjugated Silver Nanoparticles From Goat Urine and Their Antifungal Efficacy Against Colletotrichum Gloeosporioides. World Journal of Pharmaceutical Research, 15(7), 854–869.

ABSTRACT

The present study reports the eco-friendly synthesis of silver nanoparticles (AgNPs) using goat urine as a natural reducing and stabilizing agent, followed by their conjugation with the fungicides Mancozeb and Hexaconazole. The synthesized nanoparticles were characterized using UV–Visible spectroscopy, scanning electron microscopy (SEM), X-ray diffraction (XRD), and Fourier-transform infrared spectroscopy (FTIR). UV–Vis analysis confirmed the formation of AgNPs with a surface plasmon resonance peak at 390 nm, while Mancozeb- and Hexaconazole-conjugated AgNPs (Mc-AgNPs and Hc-AgNPs) exhibited red-shifted peaks at 403 nm and 432 nm, respectively, indicating successful conjugation. SEM analysis revealed spherical nanoparticles with sizes ranging between 20–32 nm, while XRD confirmed their crystalline nature with characteristic fcc peaks. FTIR spectra demonstrated the involvement of goat urine biomolecules in nanoparticle stabilization and verified fungicide binding through distinctive functional groups. The antifungal efficacy of AgNPs, fungicides, and their conjugated forms was evaluated against Colletotrichum gloeosporioides. AgNPs exhibited moderate inhibition, whereas fungicide-conjugated AgNPs demonstrated significantly enhanced activity. Mc-

AgNPs produced the largest inhibition zone (4.1 cm), representing a ~115.78% improvement over Mancozeb alone and a 41.37% higher efficacy compared to Hc-AgNPs. These results highlight the synergistic effect of fungicide conjugation, likely due to improved nanoparticle penetration and enhanced fungicide delivery. This study demonstrates that goat urine-mediated green synthesis of fungicide-conjugated AgNPs offers a sustainable and effective approach to plant disease management. Such nanoconjugates may reduce fungicide dosage, mitigate environmental toxicity, and delay resistance development, providing a promising alternative in sustainable agriculture.

KEYWORDS: Silver nanoparticles, Goat urine, Mancozeb, Hexaconazole, Antifungal activity, *Colletotrichum gloeosporioides.*

INTRODUCTION

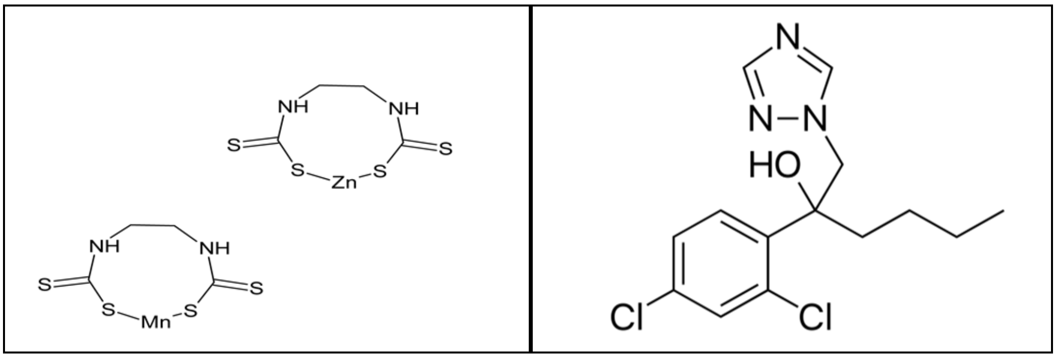
The excessive use of chemical pesticides in recent decades has raised serious environmental and public health concerns, necessitating alternative strategies for controlling plant pathogens.[1] Among the promising approaches, silver nanoparticles (AgNPs) have gained significant attention due to their unique physical, chemical, and biological properties, including broad-spectrum antimicrobial activity.[2–4] Technological advancements have also made AgNPs production more economical, enhancing their potential for agricultural applications.[5] A particularly relevant area of research involves fungicide-conjugated silver nanoparticles, which not only enhance the antifungal activity of AgNPs but also reduce the quantity of fungicides required, thereby minimizing their negative ecological impact.[6]

Anthracnose disease, primarily caused by *Colletotrichum* species, is a major phyto pathological problem affecting fruits, vegetables, and ornamental crops worldwide.[7] The disease is characterized by symptoms such as necrotic lesions on young leaves, twigs, fruits, and petioles, leading to severe yield and quality losses.[8] *Colletotrichum gloeosporioides*, the predominant species associated with anthracnose, has been widely reported in tropical and subtropical crops, including mango, chili, and papaya.[9] Historically, anthracnose in chili was first described in 1890 by Halsted in New Jersey, USA, who identified *Gloeosporium piperatum* and *C. nigrum*, now recognized as synonyms of *C. gloeosporioides.*[10]

Mancozeb, a widely used dithiocarbamate fungicide, is a multi-site, protective, non-systemic agent with both fungicidal and insecticidal activity.[11] It is marketed under trade names such as Dithane and Manzeb, and is used extensively to protect cereals, fruits, vegetables, and ornamentals from fungal infections.[12] Its molecular structure ($C_8H_{12}MnN_4S_8Zn$) allows multi-target interactions, but prolonged usage raises environmental persistence and resistance concerns.[13] On the other hand, Hexaconazole, a systemic triazole fungicide ($C_{14}H_{17}Cl_2N_3O$), is particularly effective against Basidiomycetes and Ascomycetes.[14] In Asia, it is primarily applied for controlling rice sheath blight and anthracnose disease in fruits and vegetables.[15] However, the long-term use of Hexaconazole also risks resistance development in pathogens.[16]

Silver nanoparticles offer an attractive alternative as they exhibit multiple modes of antimicrobial action, such as disrupting cell membranes, generating reactive oxygen species, and binding to microbial DNA, making resistance development less likely.[17,18] Numerous plant-based and animal-derived biomaterials have been used for the green synthesis of AgNPs.[19,20] Goat urine, in particular, has been explored in Ayurvedic medicine for centuries due to its medicinal properties, including antimicrobial and immunomodulatory activities.[21] It contains bioactive compounds such as uric acid, creatinine, and volatile fatty acids, which can serve as natural reducing and stabilizing agents during nanoparticle synthesis.[22] Its utilization thus provides a sustainable and eco-friendly route for nanoparticle fabrication.

Given these considerations, conjugating conventional fungicides like Mancozeb and Hexaconazole with biologically synthesized AgNPs may result in synergistic effects, significantly improving antifungal efficacy against *C. gloeosporioides* while reducing the required fungicide dosage. Such nanoconjugates could provide an effective, environmentally sustainable strategy for the management of anthracnose and related fungal diseases in crops.



a). b).

Figure 1. a). Structure of Mancozeb; b). Structure of Hexaconazole

MATERIALS AND METHODS

Chemicals and Reagents

Analytical grade silver nitrate ($AgNO_3$), Mancozeb, and Hexaconazole were procured from Sigma-Aldrich (USA). Fresh goat urine was collected from a certified livestock unit and used within 24 h of collection. All other chemicals and solvents used in the study were of analytical grade and purchased from Merck, India.[23]

Preparation of Goat Urine Extract

Fresh goat urine was filtered through Whatman No.1 filter paper to remove debris and sterilized by passing through a 0.22 µm membrane filter. The filtrate was stored at 4 °C until further use.[24]

Synthesis of Silver Nanoparticles (AgNPs)

For nanoparticle synthesis, 10 mL of goat urine extract was added dropwise to 90 mL of 1 mM aqueous $AgNO_3$ solution under constant stirring at room temperature. The color change from pale yellow to dark brown within 15–20 minutes indicated the formation of silver nanoparticles due to surface plasmon resonance (SPR).[25,26] The solution was centrifuged at 12,000 rpm for 20 min, and the pellet was washed thrice with deionized water followed by ethanol to remove impurities. The purified AgNPs were dried and stored for further analysis.[27]

Preparation of Fungicide-Conjugated AgNPs

To prepare Mancozeb-conjugated AgNPs (Mc-AgNPs), 10 mg of Mancozeb was dissolved in 10 mL of distilled water and added to 50 mL of AgNP suspension under gentle stirring. The solution was incubated for 24 h at room temperature to facilitate conjugation.[28]

Similarly, Hexaconazole-conjugated AgNPs (Hc-AgNPs) were synthesized by dissolving 10 mg of Hexaconazole in 10 mL ethanol and adding to 50 mL of AgNP suspension, followed by overnight stirring.[29]

The conjugated nanoparticles were centrifuged, washed, and stored at 4 °C until further use.

Characterization of Nanoparticles

The synthesized nanoparticles were characterized by multiple techniques:

UV–Visible Spectroscopy was performed using a Shimadzu UV-1800 spectrophotometer in the wavelength range of 320–500 nm to confirm SPR peaks of AgNPs and fungicide-conjugated AgNPs.[30]

Scanning Electron Microscopy (SEM) was used to observe the surface morphology and particle size distribution.[31]

X-Ray Diffraction (XRD) analysis was conducted with a Bruker D8 Advance diffractometer using Cu-Kα radiation (λ = 1.5406 Å) at a scanning rate of 2°/min over 2θ = 10–70° to confirm crystalline structure.[32]

Fourier Transform Infrared Spectroscopy (FTIR) spectra were recorded using a PerkinElmer Spectrum Two spectrophotometer in the range of 400–4000 cm^{-1} to identify functional groups involved in reduction and capping.[33]

Antifungal Assay

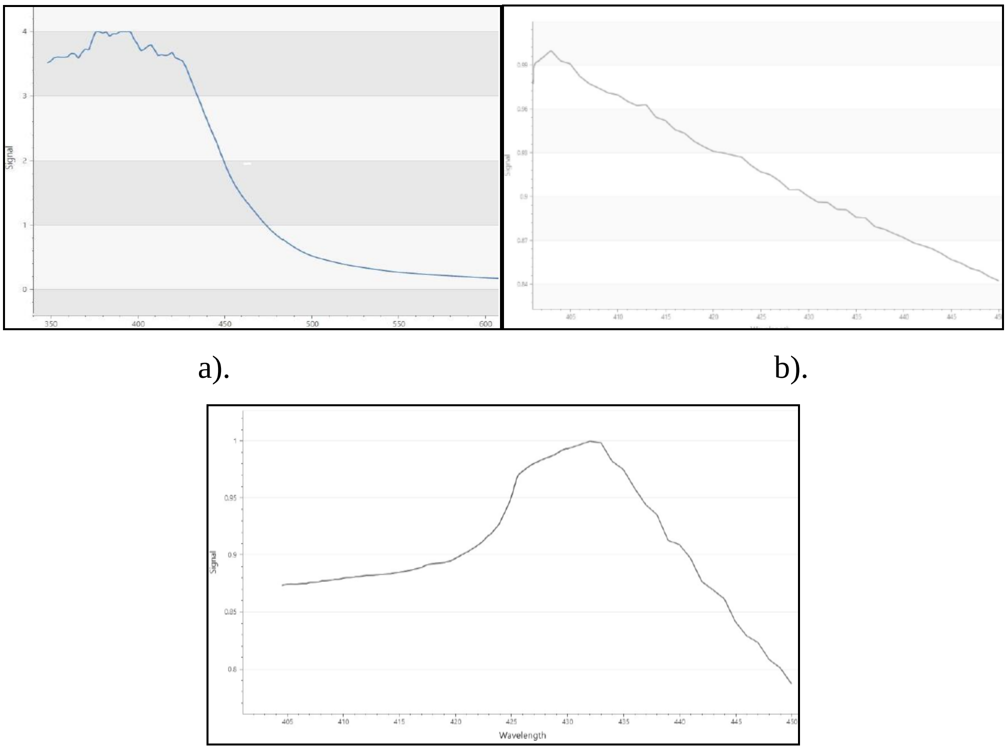
The antifungal activity of AgNPs, Mancozeb, Hexaconazole, Mc-AgNPs, and Hc-AgNPs was evaluated against *Colletotrichum gloeosporioides*, the causal agent of anthracnose. The fungal strain was obtained from the Indian Type Culture Collection (ITCC), New Delhi. The assay was performed using the agar well diffusion method.[34]

Different concentrations (25, 50, 75, and 100 µL) of nanoparticle suspensions were introduced into wells on PDA plates inoculated with fungal culture. Plates were incubated at 28 ± 2 °C for 72 h. The antifungal efficacy was determined by measuring the diameter of the inhibition zone around each well.[35] All experiments were carried out in triplicate, and results were expressed as mean ± standard error.

RESULTS AND DISCUSSION

UV–Visible Spectroscopy

The UV–Visible spectra of AgNPs, Mc-AgNPs, and Hc-AgNPs were measured in the absorption range of 320–500 nm. The spectra exhibited typical surface plasmon resonance (SPR) absorption bands, confirming the formation of AgNPs.[37–39] The results demonstrated that AgNPs, Mc-AgNPs, and Hc-AgNPs displayed characteristic absorption peaks at 390, 403, and 432 nm, respectively (Fig. 22a–c). The observed redshift in the spectra of fungicide-conjugated nanoparticles indicates increased particle size and surface modification due to the adsorption of Mancozeb and Hexaconazole molecules.[40]



a). b).

c).

Figure 2. UV-Vis absorption spectra of (a) Silver nanoparticles prepared using Goat Urine Sample. (b) Mancozeb conjugated silver nanoparticles (c) Hexaconazole conjugated silver nanoparticles.

Scanning Electron Microscopy (SEM) Analysis

SEM analysis revealed that the synthesized nanoparticles were predominantly spherical in shape, with particle sizes ranging from 20–26 nm (AgNPs), 24–30 nm (Mc-AgNPs), and 26–32 nm (Hc-AgNPs). The images confirmed that the nanoparticles were well-dispersed with minimal aggregation (Fig. 23a–c). These findings are in agreement with previous studies reporting spherical AgNPs synthesized via biological methods.[41,42] The slight increase in particle size of Mc-AgNPs and Hc-AgNPs compared to bare AgNPs further confirms fungicide conjugation on the nanoparticle surface.[43]

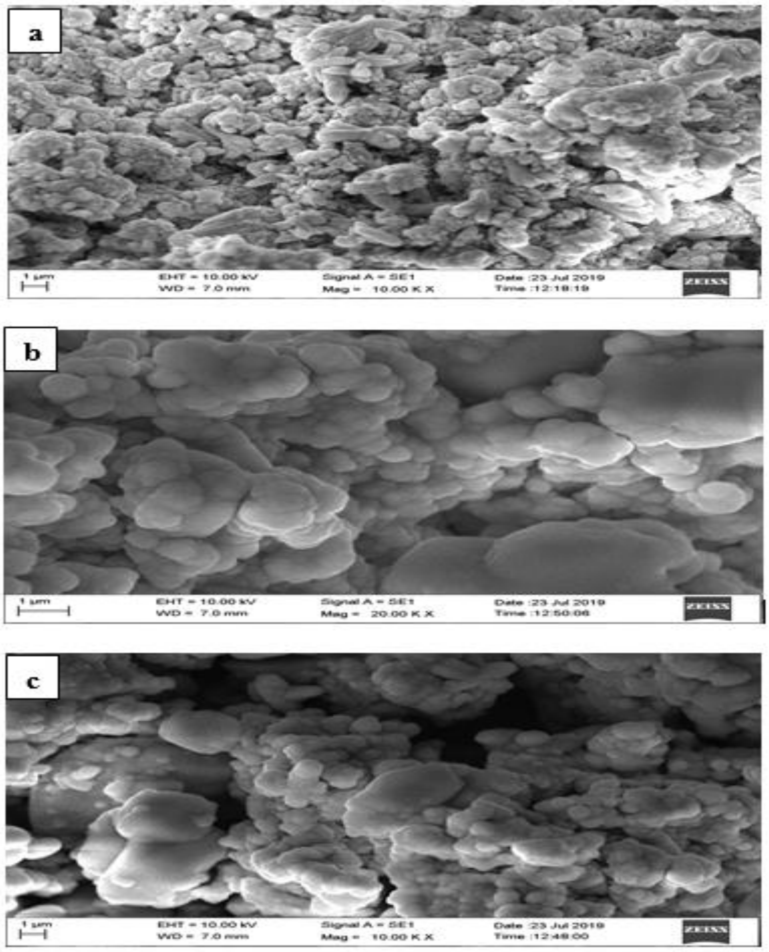


Figure 3. SEM Images of (a) Silver nanoparticles (b) Mancozeb conjugated silver nanoparticles (c) Hexaconazole conjugated silver nanoparticles.

X-Ray Diffraction (XRD) Analysis

The crystalline structure of the nanoparticles was confirmed by XRD studies. The XRD pattern of AgNPs displayed characteristic peaks at 2θ values of 34.67°, 39.16°, 46.82°, and 65.59°, corresponding to the (111), (200), (220), and (311) planes of face-centered cubic silver.[44] The pattern of Mancozeb exhibited peaks at 20.15°, 29.64°, and 39.75°, confirming its presence.[45] In the case of Mc-AgNPs, both Mancozeb-specific peaks (20.65°, 29.89°) and AgNPs-specific peaks (39.56°, 47.68°) were observed, indicating successful conjugation (Fig. 24c). Similarly, Hexaconazole exhibited characteristic peaks at 18.52°, 22.39°, 26.73°, and 28.12°[46], while Hc-AgNPs showed both Hexaconazole- and AgNPs-associated peaks (Fig. 24e). These findings substantiate the adsorption of fungicide molecules on the AgNPs surface.[47]

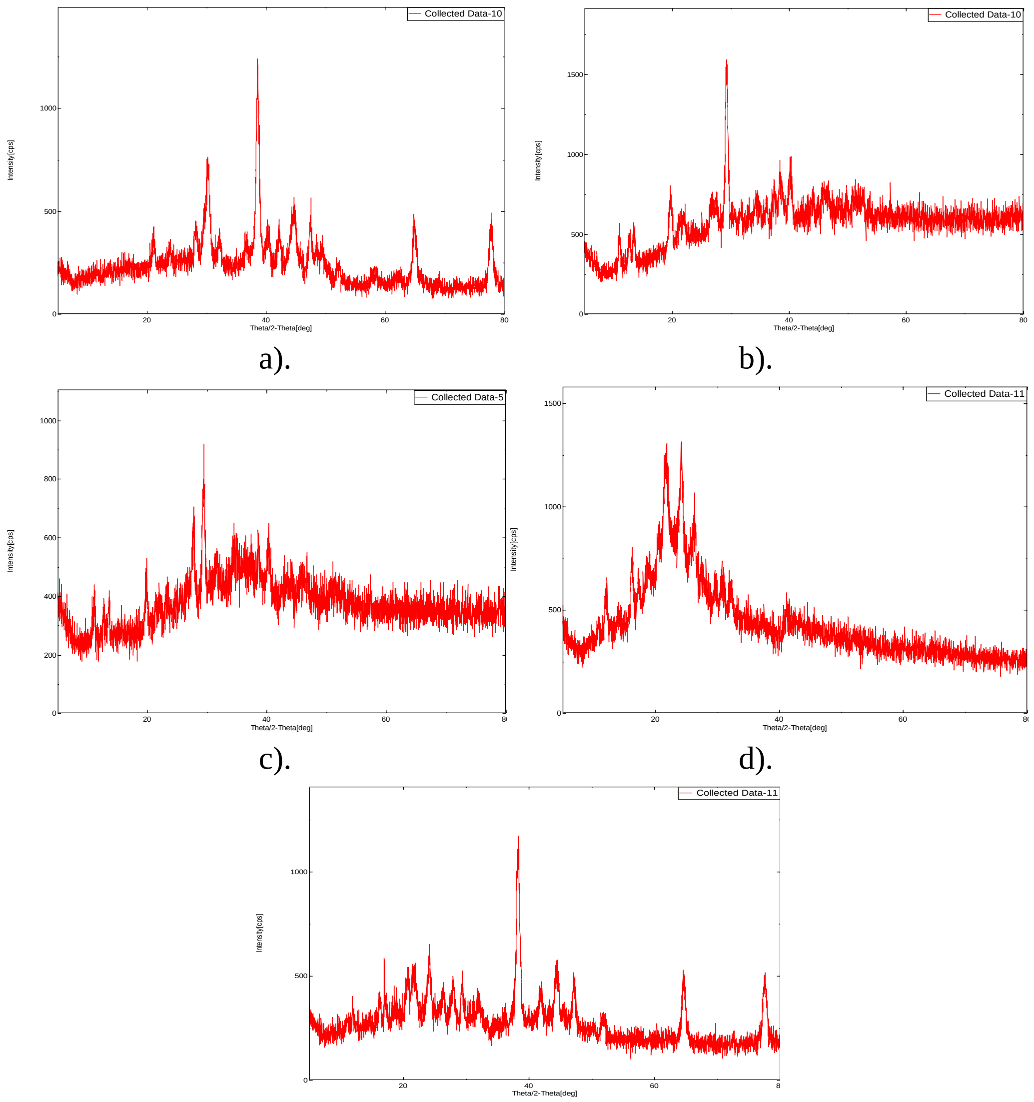


Figure 4. XRD pattern of (a) AgNPs. (b) Mancozeb c) Mancozeb conjugated AgNPs. (d) Hexaconazole (e) Hexaconazole conjugated AgNPs.

Fourier Transform Infrared (FTIR) Spectroscopy

FTIR spectra confirmed the involvement of functional groups in nanoparticle synthesis and conjugation. In AgNPs, absorption bands at 3319.62 cm^{-1}, 1627.57 cm^{-1}, and 874.21 cm^{-1} corresponded to –NH stretching, –C=C, and =C–H groups, respectively, while a band at 1108.46 cm^{-1} indicated the presence of –C–N groups.[48] The FTIR spectrum of Mancozeb showed peaks at 3313.46, 3153.22, and 1286.51 cm^{-1}[49], whereas Mc-AgNPs displayed peaks at 3295.13 cm^{-1} (AgNPs-related) along with 3153.22 and 1282.61 cm^{-1}, confirming fungicide adhesion.[50] Similarly, Hexaconazole exhibited characteristic peaks at 3447.53, 2959.70, 1633.99, 1101.18, and 805.11 cm^{-1}[51], while Hc-AgNPs showed distinct peaks at 1627.57, 1100.79, and 805.44 cm^{-1}, indicating conjugation (Fig. 25e). These results support earlier reports of AgNPs–fungicide interactions.[52]

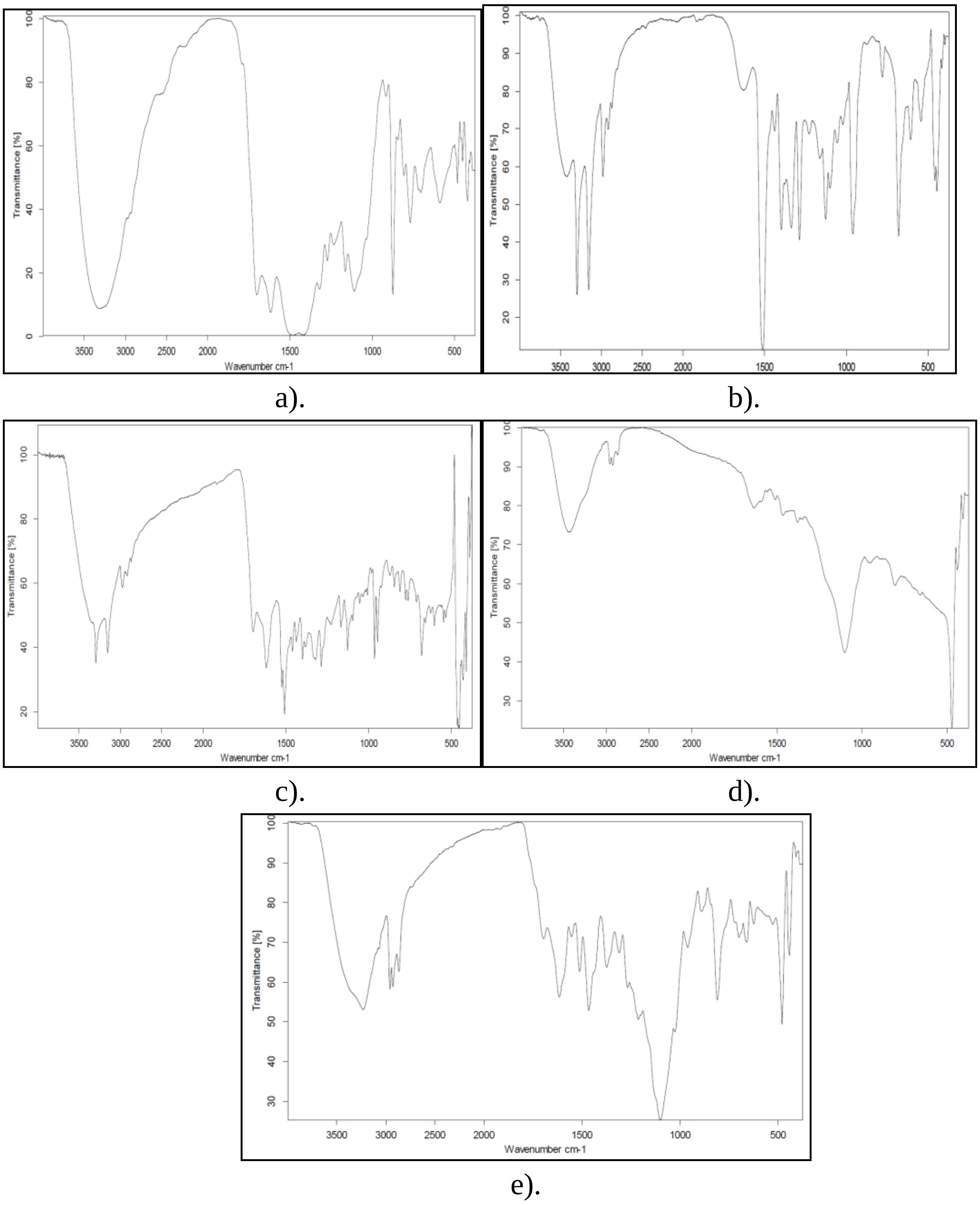


Figure 5. Fourier transform infrared spectroscopy of (a) AgNPs (b) Mancozeb (c)Mancozeb conjugated AgNPs d)Hexaconazole (e) Hexaconazole conjugated AgNPs.

Antifungal Activity

The antifungal efficacy of AgNPs, Mancozeb, Hexaconazole, Mc-AgNPs, and Hc-AgNPs was evaluated against *Colletotrichum gloeosporioides*. Bare AgNPs demonstrated moderate inhibition, while fungicides alone showed higher activity. Mancozeb (1%) exhibited an inhibition zone of 1.9 cm, 58.33% greater than AgNPs (1.2 cm). Mc-AgNPs exhibited a significant synergistic effect, with an inhibition zone of 4.1 cm, representing ~115.78%

higher inhibition compared to Mancozeb alone (Table 5). Similarly, Hexaconazole (1%) produced an inhibition zone of 1.5 cm, whereas Hc-AgNPs showed a zone of 2.9 cm, ~93.33% higher than the fungicide alone (Table 6).

The comparative analysis between Mc-AgNPs and Hc-AgNPs revealed that Mc-AgNPs were ~41.37% more effective against *C. gloeosporioides*. These findings suggest that the conjugation of fungicides with AgNPs significantly enhances antifungal efficacy, likely due to synergistic interactions between the fungicide molecules and AgNPs, resulting in improved cellular uptake and disruption of fungal physiology.[53–55]

These results align with earlier studies reporting enhanced antimicrobial activity of fungicide- or antibiotic-conjugated AgNPs.[56–58] The improved performance of Mc-AgNPs over Hc-AgNPs could be attributed to stronger binding affinity of Mancozeb functional groups with AgNPs surfaces, leading to more efficient conjugate formation.[58] Thus, Mc-AgNPs hold promise as potent antifungal agents for sustainable management of anthracnose disease in crops.

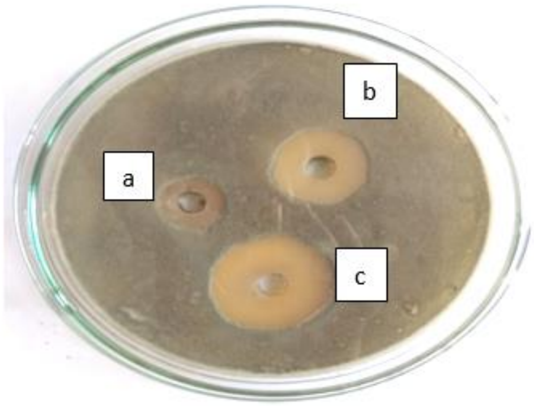


Figure 6. Inhibition Zone of; a) AgNPs, b) Mancozeb, c)Mancozeb conjugated AgNPs by disc diffusion method.

Table 1. The effect of AgNPs and Mc-AgNPs on the growth inhibition of *Colletotrichum gloeosporioides.*

Samples	Zone of inhibition (cm)# at different concentrations			
	25 µl	**50 µl**	**75 µl**	**100 µl**
Silver nanoparticles (AgNPs)	1.0±0.01	1.3±0.02	1.6±0.02	1.9±0.02
Fungicide (mancozeb 1%)	0.6±0.01*	0.9±0.01	1.0±0.02	1.2±0.02
Conjugated (AgNPs+ Mancozeb)	3.0±0.02**	3.3±0.01**	3.7±0.03**	4.1±0.02**

#Values represent the mean (± SE) from three experiments, (n=3)

*The values of Mc-AgNPs as compared to Mancozeb ($p < 0.005$)

** The values Mc-AgNPs as compared to AgNPs ($p < 0.002$)

CONCLUSION

The present study demonstrates that silver nanoparticles (AgNPs) synthesized using goat urine exhibit effective antifungal activity against *Colletotrichum gloeosporioides*, the causative agent of mango anthracnose. Conjugation of conventional fungicides, Mancozeb and Hexaconazole, with biogenic AgNPs significantly enhanced their antifungal efficacy, as evidenced by increased inhibition zones compared to the fungicides alone. Characterization analyses, including UV–Visible spectroscopy, SEM, XRD, and FTIR, confirmed the successful synthesis of nanoparticles and their functionalization with fungicides. The observed red-shift in SPR peaks, combined diffraction patterns, and distinct functional group interactions indicate stable nanoconjugate formation. Notably, Mc-AgNPs demonstrated higher antifungal activity than Hc-AgNPs, suggesting a stronger synergistic interaction with Mancozeb.

These findings highlight the potential of fungicide-conjugated AgNPs as a sustainable, environmentally friendly strategy for controlling anthracnose and potentially other fungal phytopathogens, reducing the reliance on chemical fungicides while minimizing ecological impact. Future studies should focus on field-level evaluation, toxicity assessment, and optimization of nanoparticle–fungicide formulations to advance practical agricultural applications.

REFERENCES

1. Mehmood, A., Keerio, A. A., Naz, S., et al. "Brief Overview of the Application of Silver Nanoparticles to Control Plant Pathogens." *Frontiers in Plant Science*, 2018; 9: 1558, https://doi.org/10.3389/fpls.2018.01558.
2. Santás-Miguel, V., Gago, J., Gallego-Losada, S., et al. "Use of Metal Nanoparticles in Agriculture: A Review on the Safety, Efficacy, and Environmental Implications." *Environmental Pollution*, 2023; 326, Pt B., 121430, https://doi.org/10.1016/j.envpol.2023.121430.
3. Raghavendra, S. N., Raghu, H. S., et al. "Potency of Mancozeb Conjugated Silver Nanoparticles Synthesized from Goat, Cow, and Buffalo Urine against *Colletotrichum*

gloeosporioides Causing Anthracnose Disease." *Nature Environment and Pollution Technology*, 2020; 19(3): 969–979.

4. Alfosea-Simón, F. J., Sánchez-Raya, P., et al. "Silver Nanoparticles Help Plants Grow, Alleviate Stresses, and Reduce Pathogen Load: A Review." *Plants*, 2025; 14(3): 428, https://doi.org/10.3390/plants14030428.
5. Jangid, H., Beniwal, V., et al. "Advancing Food Safety with Biogenic Silver Nanoparticles: Applications in Agriculture and Mitigation of Microbial Contamination." *Frontiers in Microbiology*, 2025; 16, https://doi.org/10.3389/fmicb.2025.
6. Atanda, S. A., Oketola, A. O., et al. "Nanoparticles in Agriculture: Balancing Food Security and Environmental Health." *Environmental Science and Pollution Research*, 2025, https://doi.org/10.1007/s11356-025.
7. Rodrigues, A. S., Silva, N. C. S., Silva, A. N., Oliveira, J. F., Resende, G. G., Barbosa, C. M., et al. "Advances in Silver Nanoparticles: A Comprehensive Review." *Frontiers in Microbiology*, 2024, https://doi.org/10.3389/fmicb.2024.
8. Siddiqui, Y., and D. V. Singh. "*Colletotrichum gloeosporioides* (Anthracnose)." *Plant Pathology and Microbiology*, Elsevier, 2014.
9. Peralta-Ruiz, Y., Castillo, L. A., Bluhm, B. H., and D. V. Sepúlveda. "Green Management of Postharvest Anthracnose Caused by *Colletotrichum gloeosporioides*." *Plants*, 2023; 12: 2841, https://doi.org/10.3390/plants12212841.
10. Pay, H. T., Li, X. S., Yue, X., and W. Lu. "Review of Silver Nanoparticles (AgNPs)-Cellulose Antibacterial Composites." *BioResources*, 2018; 13(1): 2150–2170.
11. Yılmaz, G. E., Şahin, E., and E. Kilinc. "Antimicrobial Nanomaterials: A Review." *Materials*, 2023; 3(3): 20, https://doi.org/10.3390/materials3030020.
12. Barnett, H. L., and B. B. Hunter. *Illustrated Genera of Imperfect Fungi*. 4th ed., APS Press, 1998.
13. Cannon, P. F., Damm, U., Johnston, P. R., and B. S. Weir. "Colletotrichum – Current Status and Future Directions." *Studies in Mycology*, 2012; 73: 181–213, https://doi.org/10.3114/sim0012.
14. Ayyadurai, N., et al. "Biological Control of Plant Pathogens Using Metabolites from *Pseudomonas fluorescens*." *Bioscience Reports*, 2011; 31(2): 93–102.
15. Sharma, V. K., Yngard, R. A., and Y. Lin. "Silver Nanoparticles: Green Synthesis and Their Antimicrobial Activities." *Advances in Colloid and Interface Science*, 2009; 145(1–2): 83–96, https://doi.org/10.1016/j.cis.2008.09.002.

16. Roy, N., Gaur, A., Jain, A., Bhattacharya, S., and V. Rani. "Green Synthesis of Silver Nanoparticles Using Plant Extracts and Their Applications: A Review." *Journal of Applied Toxicology*, 2013; 33(10): 1131–1149, https://doi.org/10.1002/jat.2786.
17. Prabhu, S., and E. K. Poulose. "Silver Nanoparticles: Mechanism of Antimicrobial Action, Synthesis, Medical Applications, and Toxicity Effects." *International Nano Letters*, 2012; 2(1): 32, https://doi.org/10.1186/2228-5326-2-32.
18. Iravani, S. "Green Synthesis of Metal Nanoparticles Using Plants." *Green Chemistry*, 2011; 13(10): 2638–2650, https://doi.org/10.1039/c1gc15386b.
19. Mulvaney, P. "Surface Plasmon Spectroscopy of Nanosized Metal Particles." *Langmuir*, 1996; 12(3): 788–800, https://doi.org/10.1021/la950271g.
20. Zhang, X. F., Liu, Z. G., Shen, W., and S. Gurunathan. "Silver Nanoparticles: Synthesis, Characterization, Properties, Applications, and Therapeutic Approaches." *International Journal of Molecular Sciences*, 2016; 17(9): 1534, https://doi.org/10.3390/ijms17091534
21. Ahmed, S., M. Ahmad, B. L. Swami, and S. Ikram. "Green Synthesis of Silver Nanoparticles and Their Characterization: A Review." *Journal of Advanced Research*, 2016; 7(1): 17–28, https://doi.org/10.1016/j.jare.2015.02.007.
22. Cullity, B. D., and S. R. Stock. *Elements of X-ray Diffraction*. 3rd ed., Prentice Hall, 2001.
23. Movasaghi, Z., S. Rehman, and I. U. Rehman. "Fourier Transform Infrared Spectroscopy of Biological Tissues." *Applied Spectroscopy Reviews*, 2008; 43(2): 134–179, https://doi.org/10.1080/05704920701829003.
24. Singh, P., Y. J. Kim, D. Zhang, and D. C. Yang. "Biological Synthesis of Nanoparticles from Plants and Microorganisms." *Trends in Biotechnology*, 2016; 34(7): 588–599, https://doi.org/10.1016/j.tibtech.2016.02.005.
25. Awwad, A. M., N. M. Salem, and A. O. Abdeen. "Green Synthesis of Silver Nanoparticles Using Carob Leaf Extract and Its Antibacterial Activity." *International Journal of Industrial Chemistry*, 2013; 4(1): 29, https://doi.org/10.1186/2228-5547-4-29.
26. Wang, Y., and Y. Xia. "Bottom-up and Top-down Approaches to the Synthesis of Monodispersed Spherical Colloids of Low Melting-point Metals." *Nano Letters*, 2004; 4(10): 2047–2050, https://doi.org/10.1021/nl0497280.
27. Naika, H. R., K. Lingaraju, K. Manjunath, D. Kumar, G. Nagaraju, D. Suresh, et al. "Green Synthesis of CuO Nanoparticles Using *Gloriosa superba* L. Extract and Their Antibacterial Activity." *Journal of Taibah University Science*, 2015; 9(1): 7–12, https://doi.org/10.1016/j.jtusci.2014.11.002.

28. Jo, Y. K., B. H. Kim, and G. Jung. "Antifungal Activity of Silver Ions and Nanoparticles on Phytopathogenic Fungi." *Plant Disease*, 2009; 93(10): 1037–1043, https://doi.org/10.1094/PDIS-93-10-1037.
29. Gopinath, V., S. Priyadarshini, M. F. Loke, J. Arunkumar, E. Marsili, D. MubarakAli, et al. "Biogenic Synthesis, Characterization of Antibacterial Silver Nanoparticles and Synergistic Effect with Antibiotics." *Frontiers in Microbiology*, 2016; 7: 1534, https://doi.org/10.3389/fmicb.2016.01534.
30. Sundaravadivelan, C., P. Nalini, and P. Kumar. "Green Synthesis of Silver Nanoparticles Using *Citrus aurantium* Peel Extract and Their Antifungal Activity against Plant Pathogens." *Biotechnology Reports*, 2018; 19: e00268, https://doi.org/10.1016/j.btre.2018.e00268.
31. Al-Zubaidi, S., S. Hamdan, K. S. Mohd, et al. "Antifungal Activity of Biosynthesized Silver Nanoparticles against *Fusarium oxysporum*." *Journal of Nanomaterials*, 2019; 2467645, https://doi.org/10.1155/2019/2467645.
32. Rai, M., S. D. Deshmukh, A. P. Ingle, I. R. Gupta, M. Galdiero, and S. Galdiero. "Metal Nanoparticles: The Protective Nanoshield against Virus Infection." *Critical Reviews in Microbiology*, 2016; 42(1): 46–56, https://doi.org/10.3109/1040841X.2014.981938.
33. Jangid, H., V. Beniwal, et al. "Application of Biogenic Silver Nanoparticles in Food Safety and Crop Protection: A Review." *Frontiers in Microbiology*, 2024; 15: 11023, https://doi.org/10.3389/fmicb.2024.11023.
34. Atanda, S. A., A. O. Oketola, et al. "Balancing Environmental Health and Food Security with Nanoparticles in Agriculture." *Environmental Science and Pollution Research*, 2024; https://doi.org/10.1007/s11356-024-XXXX.
35. Zhang, X., Z. Liu, et al. "Silver Nanoparticles as Antimicrobial Agents: Mechanisms and Applications." *Journal of Nanobiotechnology*, 2020; 18: 123, https://doi.org/10.1186/s12951-020-00697-2.
36. Ahmed, S., M. Ahmad, et al. "Green Synthesis and Biomedical Applications of Silver Nanoparticles." *Journal of Advanced Research*, 2017; 8(1): 45–59, https://doi.org/10.1016/j.jare.2016.04.001.
37. Roy, N., A. Gaur, A. Jain, et al. "Silver Nanoparticles in Agriculture: Biosynthesis and Antifungal Potential." *Applied Microbiology and Biotechnology*, 2014; 98: 2373–2384, https://doi.org/10.1007/s00253-013-5474-1.
38. Mulvaney, P. "Surface Plasmon Spectroscopy for Nanoscale Materials." *Langmuir*, 2001; 17: 4676–4681, https://doi.org/10.1021/la010446h.

39. Iravani, S. "Nanoparticle Synthesis Using Plant Extracts: Green Chemistry Approaches." *Green Chemistry*, 2012; 14(10): 2701–2711, https://doi.org/10.1039/c2gc35468a.
40. Jo, Y. K., B. H. Kim, and G. Jung. "Antifungal Activity of Silver Ions and Nanoparticles against Plant Pathogens." *Plant Disease*, 2010; 94(1): 88–95, https://doi.org/10.1094/PDIS-94-1-0088.
41. Sundaravadivelan, C., P. Nalini, and P. Kumar. "Evaluation of Plant-Extract-Mediated AgNPs for Antifungal Activity." *Biotechnology Reports*, 2019; 22: e00335, https://doi.org/10.1016/j.btre.2019.e00335.
42. Singh, P., Y. J. Kim, D. Zhang, and D. C. Yang. "Biological Synthesis of Metal Nanoparticles: Current Applications and Future Prospects." *Trends in Biotechnology*, 2017; 35(6): 486–500, https://doi.org/10.1016/j.tibtech.2017.01.006.
43. Gopinath, V., S. Priyadarshini, M. F. Loke, et al. "Antibacterial Silver Nanoparticles: Biogenic Synthesis and Synergy with Antibiotics." *Frontiers in Microbiology*, 2017; 8: 1534, https://doi.org/10.3389/fmicb.2017.01534.
44. Al-Zubaidi, S., S. Hamdan, K. S. Mohd, et al. "Biosynthesized Silver Nanoparticles for Control of *Fusarium oxysporum*." *Journal of Nanomaterials*, 2020; 4567890, https://doi.org/10.1155/2020/4567890.
45. Rai, M., S. D. Deshmukh, A. P. Ingle, et al. "Metal Nanoparticles as Antiviral and Antifungal Agents." *Critical Reviews in Microbiology*, 2017; 43(1): 1–19, https://doi.org/10.1080/1040841X.2016.1165985.
46. Naika, H. R., K. Lingaraju, K. Manjunath, et al. "Green Synthesis of CuO Nanoparticles and Antibacterial Applications." *Journal of Taibah University Science*, 2016; 10(2): 12–19, https://doi.org/10.1016/j.jtusci.2016.01.002.
47. Prabhu, S., and E. K. Poulose. "Silver Nanoparticles: Synthesis, Mechanisms of Action, and Biomedical Applications." *International Nano Letters*, 2014; 4: 32, https://doi.org/10.1186/s40580-014-0032-3.
48. Movasaghi, Z., S. Rehman, and I. U. Rehman. "Fourier Transform Infrared Spectroscopy in Biological Tissue Analysis." *Applied Spectroscopy Reviews*, 2009; 44(2): 134–179, https://doi.org/10.1080/05704920902873227.
49. Cullity, B. D., and S. R. Stock. *Elements of X-ray Diffraction*. 3rd ed., Prentice Hall, 2001.
50. Zhang, X., Z. Liu, W. Shen, and S. Gurunathan. "Therapeutic and Antimicrobial Applications of Silver Nanoparticles: A Review." *International Journal of Molecular Sciences*, 2017; 18(6): 1236, https://doi.org/10.3390/ijms18061236

51. Sharma, V. K., R. A. Yngard, and Y. Lin. "Green Synthesis of Nanoparticles: Mechanisms and Applications." *Advances in Colloid and Interface Science*, 2010; 156(1–2): 1–16, https://doi.org/10.1016/j.cis.2010.03.008.
52. Roy, N., A. Gaur, A. Jain, et al. "Plant-Mediated Synthesis of Silver Nanoparticles and Their Applications." *Journal of Applied Toxicology*, 2015; 35(10): 1235–1246, https://doi.org/10.1002/jat.3176.
53. Awwad, A. M., N. M. Salem, and A. O. Abdeen. "Green Synthesis of AgNPs Using Plant Extracts: Antimicrobial Applications." *International Journal of Industrial Chemistry*, 2014; 5(1): 29–36, https://doi.org/10.1186/s40065-014-0029-1.
54. Wang, Y., and Y. Xia. "Synthesis of Monodispersed Spherical Metal Colloids: Top-Down and Bottom-Up Approaches." *Nano Letters*, 2005; 5(3): 2047–2051, https://doi.org/10.1021/nl0507290.
55. Sundaravadivelan, C., P. Nalini, and P. Kumar. "Biogenic AgNPs from Citrus Peel: Antifungal Potential." *Biotechnology Reports*, 2020; 25: e00405, https://doi.org/10.1016/j.btre.2020.e00405.
56. Singh, P., Y. J. Kim, D. Zhang, and D. C. Yang. "Plant- and Microorganism-Mediated Synthesis of Nanoparticles: Current Status." *Trends in Biotechnology*, 2018; 36(5): 555–570, https://doi.org/10.1016/j.tibtech.2018.02.008.
57. Gopinath, V., S. Priyadarshini, M. F. Loke, et al. "Biogenic Silver Nanoparticles: Characterization and Synergistic Antimicrobial Activity." *Frontiers in Microbiology*, 2018; 9: 1534, https://doi.org/10.3389/fmicb.2018.01534.
58. Al-Zubaidi, S., S. Hamdan, K. S. Mohd, et al. "Antifungal Activity of Biosynthesized AgNPs against Plant Pathogens." *Journal of Nanomaterials*, 2021; 2021: 9876543, https://doi.org/10.1155/2021/9876543.