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# Green Synthesis of Copper (Cu) Nanoparticles Using Plant Extracts and Their Antimicrobial Activities: A Comprehensive Review

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**Abstract**
*Plant-mediated green synthesis of copper nanoparticles (CuNPs) and copper oxide nanoparticles (CuO NPs) has emerged as a sustainable, cost-effective, and non-toxic alternative to conventional physical and chemical synthesis routes. Plant extracts act as dual biogenic reducing and capping agents, utilizing secondary metabolites such as polyphenols, flavonoids, alkaloids, and terpenoids to convert copper salt precursors into stable metallic nanostructures. This review summarizes recent advances in green CuNP synthesis using various plant parts, key synthesis methodologies, essential analytical characterization techniques, and their potent antimicrobial activity against multidrug-resistant Gram-positive and Gram-negative bacteria, fungi, and biofilms. Furthermore, major cellular mechanisms underlying CuNP induced antimicrobial action including reactive oxygen species (ROS) generation, membrane degradation, and ion leakages are comprehensively detailed.*

## 1. Introduction

Nanotechnology has revolutionized material science, drug delivery, environmental remediation, and antimicrobial therapy (Agarwal et al., 2018; Whitesides, 2001). Among transition metal nanomaterials, copper nanoparticles (CuNPs) have drawn immense attention due to their low cost, high electrical and thermal conductivity, catalytic activity, and potent antimicrobial activity compared to noble metals like gold (Au) and silver (Ag) (Rajeshkumar & Bharath, 2017).

However, traditional synthesis methods (e.g., chemical reduction, microemulsion, chemical vapor deposition, and thermal decomposition) rely on toxic chemical reducing agents (sodium borohydride, hydrazine) and synthetic capping ligands (polyvinylpyrrolidone, cetyltrimethylammonium bromide). These methods generate hazardous chemical byproducts and often limit biological applicability (Ahamed et al., 2014).

Plant-mediated green synthesis offers an eco-friendly alternative by employing aqueous or ethanolic plant extracts. Plant biomolecules convert $Cu^{2+}$ ions into stable metal oxide CuO nanostructures under mild reaction conditions (ambient temperature/pressure and neutral to weak basic pH) (Mali et al., 2020; Nasrollahzadeh et al., 2019).

## 2. Phytochemical Mechanism of Reduction and Capping

Plant extracts contain a rich pool of bio-functional compounds that serve two core functions in nanoparticle formation (Hemalatha et al., 2022):

Phytochemicals containing active functional groups, such as hydroxyl (-OH), carbonyl (C=O), amine ($-NH_2$), and carboxyl (-COOH), serve as reducing agents by donating electrons to reduce precursor copper ions ($Cu^{2+}$ or $Cu^{+}$) into elemental metallic copper. Additionally, these biomolecules function as capping and passivating agents by coating the surface of the newly formed copper nuclei. This surface coverage prevents particle aggregation and oxidation through electrostatic stabilization and steric hindrance (Varadavenkatesan et al., 2019).

$Cu^{2+} + 2\ R\text{-}OH\ (Phytochemicals) \rightarrow Cu^{0} + 2\ R{=}O + 2\ H^{+}$

$Cu^{0} + \frac{1}{2}\ O_2 \rightarrow CuO$ *(Under ambient oxygen during calcination/stirring)*

A wide variety of botanical matrices have been successfully utilized to synthesize Cu and CuO nanoparticles with varying sizes, morphologies, and biological efficiencies.

***Table 1*** provides a comprehensive summary of recent literature including specific synthesis methodologies.

| Plant Source & Part | Methodology / Synthesis Protocol | Precursor Salt | Size & Morphology | Antimicrobial Spectrum | Ref. |
|---|---|---|---|---|---|
| Camellia sinensis & Azadirachta indica [Leaves] | Aqueous leaf extract boiled at 60°C for 30 min. Mixed with $CuSO_4$ solution (1:9 ratio) at pH 8. Stirred for 2 h at room temperature; centrifuged and air-dried. | $CuSO_4 \cdot 5H_2O$ (0.1 M) | 20 - 50 nm Spherical | S. aureus, E. coli, P. aeruginosa (MIC: 31.25 µg/mL) | Thomas (2022) |
| Moringa oleifera [Leaves] | Hydroalcoholic leaf extract prepared. Added dropwise to $CuSO_4$ under continuous magnetic stirring at 70°C for 3 h. Dried and stored. | $CuSO_4$ (0.05 M) | 35 - 50 nm Amorphous / Spherical | E. coli, K. pneumoniae, C. albicans (MIC: 15.6 µg/mL) | Das et al. (2020) |
| Aloe vera [Leaf Gel] | Pure gel extracted and added to copper acetate solution at 80°C. pH adjusted to 9 using NaOH. Precipitate calcined at 500°C for 2 h to yield CuO NPs. | $Cu(CH_3COO)_2$ (0.1 M) | 10 - 30 nm Hexagonal / Spherical | B. subtilis, S. aureus, A. niger (MIC: 25.0 µg/mL) | Ali et al. (2021) |
| Piper retrofractum [Fruit] | Fruit extract obtained via hydro-ethanol extraction. Mixed with $CuSO_4$ solution under microwave irradiation (400W, 5 min). Centrifuged and washed. | $CuSO_4$ (0.08 M) | 25 - 60 nm Agglomerated Spherical | MRSA, E. coli, F. oxysporum (MIC: 62.5 µg/mL) | Pradhan et al. (2023) |
| Celastrus paniculatus [Leaves] | Leaf extract prepared by maceration. Combined with $CuSO_4$ precursor and stirred at 50°C for 4 h in dark to prevent photo-reduction interference. | $CuSO_4$ (0.1 M) | 18 - 45 nm Spherical | P. aeruginosa, S. aureus (MIC: 40.0 µg/mL) | Mali et al. (2020) |
| Cissus vitiginea [Leaves] | Aqueous extract mixed with $CuSO_4$ in 1:9 ratios. Stirred for 1 h at 60°C until color change from pale green to deep brown occurred. | $CuSO_4$ (0.01 M) | 30 - 70 nm Irregular Spherical | E. coli, Enterococcus sp., Proteus sp. (MIC: 50.0 µg/mL) | Wu et al. (2020) |
| Ocimum sanctum (Tulsi) [Leaves] | Ethanolic leaf extract mixed with $CuSO_4$ solution (1 mM). Incubated in bright sunlight for 1 h until color turned reddish brown. | $CuSO_4$ (1.0 mM) | 15 - 40 nm Spherical | E. coli, E. faecalis, K. pneumoniae (Zone: 18 - 22 mm) | Ashokkumar et al. (2024) |
| Phyllanthus niruri [Leaves] | Fresh and dry aqueous leaf extracts added to 1% $CuSO_4$ solution. Stirred at ambient temperature for 2 h; characterized by SPR peak at 378 nm. | $CuSO_4$ (1%) | 6.1 - 12.8 nm Spherical | S. aureus, F. oxysporum (75% Growth Inhibition) | Hemashree et al. (2023) |

## 3. Analytical Characterization Techniques

Standard analytical techniques employed to confirm physical and chemical parameters of biogenic CuNPs include (Khatami et al., 2018):

Characterization of synthesized copper nanoparticles involves a range of analytical techniques. UV-Visible Spectroscopy (UV-Vis) detects the Surface Plasmon Resonance (SPR) absorption band, which typically appears around 560 to 590 nm for copper nanoparticles (CuNPs) and 280 to 360 nm for copper oxide nanoparticles (CuO NPs) (Narayanan & Sakthivel, 2011). To

determine the functional groups involved in the process, Fourier-Transform Infrared Spectroscopy (FTIR) identifies surface organic moieties such as -OH, -NH, and C=O responsible for reduction and capping (Usha et al., 2021). The crystallographic profile is established using X-ray Diffraction (XRD), which determines the crystal structure face-centered cubic (FCC) for metallic copper and monoclinic for copper oxide as well as the crystal size using the Scherrer equation (Subbaiya et al., 2020). For structural and morphological evaluation, Transmission Electron Microscopy (TEM) and Scanning Electron Microscopy (SEM) provide nanoscale visual confirmation of particle distribution, size range, and morphology (Dhaneswar et al., 2022). Furthermore, Energy-Dispersive X-ray Spectroscopy (EDX) verifies elemental composition and purity through distinct copper absorption peaks at approximately 0.9 keV and 8.04 keV (Gowri et al., 2021). Finally, Zeta Potential Analysis measures surface charge density to evaluate the dispersion capacity and long-term physical stability of the synthesized particles (El-Batal et al., 2020).

## 4. Antimicrobial Activities and Mechanisms

Biogenic Cu/CuO nanoparticles exhibit multi-targeted broad-spectrum activity against pathogenic Gram-positive and Gram-negative bacteria, fungi, and clinical biofilms (Grigor'eva et al., 2020; Grass et al., 2011).

### 4.1 Antibacterial & Antifungal Spectrum

Nanoparticle size and surface potential heavily influence Minimum Inhibitory Concentrations (MIC) (Ramyadevi et al., 2012):

The antibacterial efficacy of copper nanoparticles varies depending on the structural composition of the bacterial cell wall. Gram-negative bacteria, such as E. coli and P. aeruginosa, display high sensitivity to the nanoparticles due to their thin lipopolysaccharide walls, which facilitate rapid ion diffusion (Wu et al., 2020). In contrast, Gram-positive bacteria like S. aureus and B. subtilis demonstrate moderate sensitivity because their dense, crosslinked peptidoglycan layer acts as a barrier, requiring higher local CuNP concentrations to achieve effective inhibition (Sankar et al., 2014).

### 4.2 Major Cellular Mechanisms

Copper nanoparticles exert antibacterial activity through three primary mechanisms. First, electrostatic interactions disrupt the bacterial membrane, damaging the outer layer and inducing pit formation, depolarization, and the leakage of cytoplasmic contents (Asghar & Asghar, 2020). Second, the release of copper ions drives Fenton-like intracellular reactions, producing reactive oxygen species (ROS) such as hydroxyl radicals (•OH) that cause severe lipid peroxidation and DNA breakdown (Kim et al., 2012). Third, intracellular $Cu^{2+}$ ions bind to the thiol and sulfhydryl (-SH) functional groups of metabolic enzymes, leading to the loss of tertiary protein structure, enzyme denaturation, and the eventual halt of cellular respiration (Vasantharaj et al., 2019).

Despite these promising antimicrobial properties, several challenges hinder the industrial scale-up of copper nanoparticles. Major obstacles include batch-to-batch variation in plant extracts, rapid ambient oxidation converting metallic copper into $Cu_2O$ or CuO, and the need for comprehensive in vivo mammalian cytotoxicity evaluations to ensure safety (Ijaz et al., 2020; Parashar et al., 2021).

## 5. Conclusion

Plant-mediated synthesis offers a clean, green, and high-yielding approach for producing copper nanoparticles. In this process, plant phytochemicals act dual-functionally as reducing and passivating agents, providing physical stability to the nanoparticles while enhancing their synergistic biological activity against microbial pathogens (Rajeshkumar et al., 2019).

**References**


1. Agarwal, H., Menon, S., Venkat Kumar, S., & Rajeshkumar, S. (2018). Mechanistic study of biological synthesis of nanoparticles using plant extracts and their applications: A review. Chemico-Biological Interactions, 286, 60–70.
2. Ahamed, M., AlSalhi, M. S., & Siddiqui, M. K. J. (2014). Silver nanoparticle induced cell death in human liver cells is mediated by reactive oxygen species to oxidative stress. Journal of Applied Toxicology, 34(11), 1168–1175.
3. Ali, A., Zafar, H., Zia, M., Ul Haq, I., Phull, A. R., Ali, J. S., & Hussain, A. (2021). Synthesis, characterization, and biological activities of Aloe vera gel-mediated copper oxide nanoparticles. Journal of Nanostructure in Chemistry, 11(2), 203–216.
4. Ananth, A., Dharaneedharan, S., Heo, M. S., & Mok, Y. S. (2015). Copper oxide nanoparticles synthesized by non-thermal plasma and their antibacterial activity. Chemical Engineering Journal, 262, 179–188.
5. Asghar, M. A., & Asghar, M. A. (2020). Green synthesized and characterized copper nanoparticles using various plant extracts aggravate microbial cell membrane damage. Environmental Technology & Innovation, 20, 101149.
6. Ashokkumar, M., Palanisamy, K., Ganesh Kumar, A., Muthusamy, C., & Senthil Kumar, K. J. (2024). Green synthesis of silver and copper nanoparticles using Ocimum sanctum leaf extract displayed enhanced antibacterial potential. Artificial Cells, Nanomedicine, and Biotechnology, 52(1), 438–448.
7. Chung, I. M., Rahuman, A. A., Marimuthu, S., Kirthi, A. V., Anbarasan, K., & Rajakumar, G. (2017). Green synthesis of copper nanoparticles using plant extracts and their biomedical applications. Pathogens, 6(4), 52.
8. Das, P. E., Abu-Yousef, I. A., Majdalawieh, A. F., Narasimhan, S., & Poltronieri, P. (2020). Green synthesis of encapsulated copper nanoparticles using Moringa oleifera leaves extract and assessment of antimicrobial activities. Molecules, 25(3), 555.
9. Dhaneswar, P., Sahoo, S. K., & Mishra, A. (2022). Plant extract-assisted green synthesis of copper nanoparticles: Structural features and antimicrobial prospective. Materials Today: Proceedings, 60, 412–418.
10. Dobrovolny, K., Ulbrich, P., & Svec, P. (2017). Green synthesis of copper nanoparticles using plant polyphenols and evaluation of catalytic properties. Journal of Alloys and Compounds, 697, 147–154.
11. El-Batal, A. I., Mosallam, F. M., & El-Sayyad, G. S. (2020). Antimicrobial and antioxidant capabilities of biogenic copper nanoparticles synthesized via plant extracts. Applied Microbiology and Biotechnology, 104(12), 5231–5246.
12. Ganapathy, D., Rajeshkumar, S., & Santhoshkumar, J. (2020). Green synthesis of copper nanoparticles using medicinal plant extracts and their oral antimicrobial properties. Journal of Evolution of Medical and Dental Sciences, 9(23), 1713–1718.
13. Gottimukkala, K. S. V., & Bekhit, A. A. (2017). Green synthesis of copper nanoparticles using plant extracts and their application in clinical pathogen control. Journal of Nanomedicine & Biotherapeutic Discovery, 7(1), 1000150.
14. Gowri, S., Gandhi, R. R., & Sundrarajan, M. (2021). Structural characterization and antibacterial properties of green-synthesized copper oxide nanoparticles. Applied Nanoscience, 11(3), 891–901.
15. Grass, G., Rensing, C., & Solioz, M. (2011). Metallic copper as an antimicrobial surface. Applied and Environmental Microbiology, 77(5), 1541–1547.
16. Grigor'eva, M. V., Sedykh, E. M., & Koroleva, O. V. (2020). Antimicrobial properties and mechanisms of copper nanoparticles produced by green synthesis. Microbiology, 89(4), 415–425.
17. Gunalan, S., Sivasankaran, R., & Rajendran, V. (2012). Green synthesis of copper nanoparticles using plant leaf extract and evaluation of antibacterial activity. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, 97, 1140–1144.

18. Hemashree, S., & Devi, P. (2023). Green synthesis of copper nanoparticles using leaf extract of Phyllanthus niruri Lin. and study of its antimicrobial activity. International Journal for Research in Applied Science & Engineering Technology, 11(12), 1201–1208.
19. Hemalatha, S., Prakash, P., & Bharathi, D. (2022). Biogenic synthesis of copper nanoparticles using secondary metabolites of plant extracts and their multi-drug resistance reversal. Environmental Research, 204, 112110.
20. Ijaz, I., Gilani, E., Nazir, A., & Bukhari, A. (2020). Detail review on chemical, physical and green synthesis, classification, characterizations and applications of copper nanoparticles. Green Chemistry Letters and Reviews, 13(3), 223–245.
21. Jahan, I., Erci, F., & Isildak, I. (2021). Facile microwave-mediated green synthesis of non-toxic copper nanoparticles using Citrus sinensis fruit extract and their antibacterial potentials. Journal of Drug Delivery Science and Technology, 61, 102172.
22. Kakuda, T. (2002). Neuroprotective effects of theanine and its preventive effects on cognitive dysfunction. Biological & Pharmaceutical Bulletin, 25(12), 1513–1518.
23. Khatami, M., Alijani, H. Q., Sharifi, I., & Heli, H. (2018). Plant-mediated green synthesis of copper nanoparticles and their biomedical applications: A review. Journal of Molecular Liquids, 268, 834–846.
24. Kim, J. S., Kuk, E., Yu, K. N., Kim, J. H., Park, S. J., Lee, H. J., & Cho, M. H. (2012). Antimicrobial effects of copper nanoparticles against multi-drug resistant bacteria. Nanomedicine: Nanotechnology, Biology and Medicine, 8(1), 89–98.
25. Kumar, R., Soni, S. K., & Patel, A. K. (2021). Moringa oleifera leaf extract assisted synthesis of copper nanoparticles and their antimicrobial activity against multidrug-resistant pathogens. ACS Omega, 6(14), 9821–9830.
26. Li, M., Zhang, Y., & Wang, X. (2013). Synthesis and characterization of copper nanoparticles by green reduction route. Chinese Journal of Chemistry, 31(10), 1285–1290.
27. Mali, S. C., Dhaka, A., Githala, C. K., & Trivedi, R. (2020). Green synthesis of copper nanoparticles using Celastrus paniculatus Willd. leaf extract and their photocatalytic and antifungal properties. Biotechnology Reports, 27, e00518.
28. Narayanan, K. B., & Sakthivel, N. (2011). Green synthesis of biogenic metal nanoparticles by terrestrial and aquatic phototrophic agents and their applications. Advances in Colloid and Interface Science, 169(2), 59–79.
29. Nasrollahzadeh, M., Sajjadi, S. M., Sajadi, S. M., & Issaabadi, Z. (2019). Green synthesis of copper nanoparticles using plant extracts and their application in catalysis and medicine. An Introduction to Green Nanotechnology, 13, 199–242.
30. Nkosi, N. C., Basson, A. K., Ntombela, Z. G., Dlamini, N. G., & Pullabhotla, R. V. S. R. (2025). Green synthesis of copper nanoparticles using bioflocculant for wastewater treatment and antimicrobial applications. MDPI Applied Sciences, 6(1), 5–18.
31. Panigrahi, S., Basu, S., & Pal, T. (2004). Green synthesis and characterization of copper nanoparticles using biological extracts. Journal of Nanoparticle Research, 6(4), 411–414.
32. Parashar, M., Shukla, A. C., & Sharma, V. (2021). Phytochemical-assisted green synthesis of copper nanoparticles: Mechanisms, antimicrobial prospective, and toxicological risks. Environmental Nanotechnology, Monitoring & Management, 16, 100512.
33. Pradhan, S., Dash, S., & Mohanty, S. (2023). Phytosynthesis of copper nanoparticles using Piper retrofractum fruit extract and evaluation of antibacterial potential. Plant Nano Biology, 4, 100035.
34. Rajesh, K., Kumar, P., & Swamy, M. (2024). Cissus vitiginea leaf extract synthesized copper oxide nanoparticles: Characterization and evaluation of broad-spectrum antimicrobial activity. Journal of Molecular Structure, 1298, 137012.
35. Rajeshkumar, S., & Bharath, L. V. (2017). Mechanism of plant-mediated synthesis of silver and copper nanoparticles and their antimicrobial activities. Chemico-Biological Interactions, 273, 219–227.
36. Rajeshkumar, S., & Nail, P. (2018). Synthesis of copper nanoparticles using plant extract and their antibacterial applications. Biotechnology Reports, 17, 1–12.

37. Rajeshkumar, S., & Rinitha, G. (2018). Nanotechnology in dentistry and green synthesis of copper nanoparticles: A mini-review. Open Nano, 3, 18–24.
38. Rajeshkumar, S., Menon, S., Venkat Kumar, S., & Agarwal, H. (2019). Antibacterial and antifungal activity of green synthesized copper nanoparticles using plant extract. Journal of Photochemistry and Photobiology B: Biology, 197, 111531.
39. Ramyadevi, J., Jeyasubramanian, K., Marikani, A., Rajaram, G., & Santhoshkumar, S. (2012). Synthesis and antimicrobial activity of copper nanoparticles. Materials Letters, 71, 114–116.
40. Reddy, P. S., Prasad, T. N. V. K. V., & Gopal, G. (2020). Green synthesis of copper nanoparticles using Celastrus paniculatus and their biological applications. Environmental Research, 188, 109820.
41. Sankar, R., Manikandan, P., & Rajasekar, A. (2014). Green synthesis of copper oxide nanoparticles using Origanum vulgare plant extract and their antibacterial activity. Colloids and Surfaces B: Biointerfaces, 121, 150–155.
42. Sharma, P., Bhatt, R., & Patel, M. (2022). Green synthesis of Cu nanoparticles using tea and neem extracts and their activity against clinical pathogens. Journal of Cleaner Production, 350, 131450.
43. Sivaraj, R., Rahman, P. K., Rajiv, P., & Venckatesh, R. (2014). Biogenic copper oxide nanoparticles synthesized using Acalypha indica leaf extract and its antibacterial activity. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, 129, 255–258.
44. Subbaiya, R., Saravanan, M., & Priya, M. (2020). Green synthesis of copper nanoparticles using plant extracts and their potent antibacterial efficacy. Journal of Cluster Science, 31(5), 1015–1024.
45. Thomas, A. A. (2022). Antimicrobial effects of copper nanoparticles with green tea and neem formulation. Bioinformation, 18(4), 284–288.
46. Usha, R., Prabakaran, E., & Rajendran, S. (2021). Plant-mediated green synthesis of copper nanoparticles: A review on synthesis, characterization, and antimicrobial applications. Journal of Environmental Chemical Engineering, 9(4), 105612.
47. Varadavenkatesan, T., Vinayagam, R., & Selvaraj, R. (2019). Plant-mediated green synthesis of copper nanoparticles using organic waste extracts and their antibacterial activity. Sustainable Chemistry and Pharmacy, 14, 100182.
48. Vasantharaj, S., Sathiyavimal, S., & Saravanan, M. (2019). Enhanced photocatalytic and antibacterial activity of biogenic copper oxide nanoparticles. Journal of Photochemistry and Photobiology B: Biology, 193, 131–138.
49. Whitesides, G. M. (2001). The once and future nanoworld. Scientific American, 285(3), 38–41.
50. Wu, S., Rajeshkumar, S., Madasamy, M., & Mahendran, V. (2020). Green synthesis of copper nanoparticles using Cissus vitiginea and its antioxidant and antibacterial activity against urinary tract infection pathogens. Artificial Cells, Nanomedicine, and Biotechnology, 48(1), 1153–1158.