RESEARCH PAPER

Green Synthesis And Uv Characterization Of Copper Nanoparticles Using *Terminalia Chebula* Linn Extract And Their Antibacterial And Dye Degradation Application

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Abstract
Copper nanoparticles (CuNPs) were successfully synthesized using *Terminalia chebula* (Kadukai) fruit extract, with its rich phytochemical content acting as natural reducing and stabilizing agents. The synthesized CuNPs demonstrated significant photocatalytic activity by effectively degrading methylene blue, as well as broad-spectrum antibacterial activity against both Gram-negative and Gram-positive bacteria. The observed antimicrobial effects were dose-dependent, likely enhanced by the synergistic action of CuNPs and bioactive compounds in *T. chebula*. These findings suggest that Kadukai-mediated CuNPs are promising eco-friendly materials with potential applications in wastewater treatment, antimicrobial formulations, and other biotechnological uses.

Key words: Copper Nanoparticles, *Terminalia chebula*, Anti- bacterial activity, Methylene blue and Dye degradation

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1. Introduction

The rapidly increasing interest in green synthesis methodologies for nanoparticle production arises from their environmental compatibility, cost-effectiveness, and potential to mitigate the toxicity associated with conventional chemical routes (Kalakonda *et al*., 2023; Keerthika *et al*., 2022). Among various metal nanoparticles, copper-based nanoparticles, particularly copper sulphate nanoparticles, have garnered significant attention due to their diverse applications in catalysis, sensing, and biomedicine (Kalakonda *et al*., 2023). Specifically, copper oxide nanoparticles synthesized through green methods have shown promise in antibacterial applications against common pathogens like *Staphylococcus aureus* and *Escherichia coli*, as well as in photocatalytic degradation of organic dyes (Alvarez *et al*., 2024; Peddi *et al*., 2021). This potential is largely attributed to their unique optical, electronic, and structural properties, which are significantly influenced by their size, shape, and surface chemistry (Priya *et al*., 2023). The use of plant extracts, such as *Malva sylvestris* leaves or *Tribulus terrestris* seeds, in the synthesis of copper nanoparticles not only offers a sustainable alternative but also often incorporates bioactive materials that act as reducing and capping agents, enhancing stability and preventing oxidation (Zeebaree *et al*., 2021; Meena *et al*., 2023). Such biogenic approaches leverage the inherent phytochemicals present in plant extracts, including flavonoids, phenolic acids, tannins, terpenoids, and alkaloids, which facilitate the reduction of metal ions into nanoparticles while simultaneously stabilizing them (Siddiqi & Husen, 2020).

Fresh fruits of *Terminalia chebula* (commonly known as Kadukai or Haritaki), a medicinal plant widely used in Ayurveda, represent another promising source for nanoparticle synthesis. These fruits are rich in bioactive compounds such as tannins, gallic acid, ellagic acid, flavonoids, and polyphenols, which not only exhibit potent antioxidant, antimicrobial, and anti-inflammatory properties but also act as excellent reducing and stabilizing agents during nanoparticle formation. The phytochemical richness of *T. chebula* contributes to enhanced stability, uniformity, and bioactivity of the synthesized copper nanoparticles, making it a suitable candidate for biomedical and environmental applications. In addition to antimicrobial and dye-degradation activities, copper nanoparticles synthesized by green routes have demonstrated significant antioxidant, anticancer, and wound-healing properties, further broadening their biomedical relevance. Their role in water purification, environmental remediation, and as catalysts for organic transformations has also been increasingly documented in recent studies. Moreover, unlike noble metal nanoparticles such as silver and gold, copper nanoparticles are cost-effective and abundant, making them a more practical choice for large-scale applications. This has led to a surge in research exploring various plant species for their efficacy in synthesizing copper nanoparticles for applications such as dye degradation, antimicrobial activity, drug delivery, and environmental remediation (Pavithran *et al*., 2020). In this context, the present study utilizes fresh *Terminalia chebula* fruits for the green synthesis of copper nanoparticles and evaluates their antibacterial potential against pathogenic bacteria.

2. Materials and Method

2.1 Collection and Preparation of Plant Extract

Fresh *Terminalia chebula* (Kadukai) fruits were procured from a local herbal market. The fruits were

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thoroughly washed with distilled water to remove surface impurities and shade-dried for 7– 10 days to preserve their phytochemical constituents. The dried material was ground into a coarse powder using a mechanical grinder. For extract preparation, 10 g of powdered fruit was boiled in 100 mL of distilled water at 60–80 °C for 20–30 minutes. The mixture was cooled to room temperature and filtered through Whatman No. 1 filter paper. The obtained filtrate was stored at 4 °C for further experimental use.

2.2 Preparation of Copper Sulfate Solution

A 0.01 M aqueous solution of copper(II) sulfate pentahydrate ($CuSO_4·5H_2O$) was prepared by dissolving the required amount of salt in double-distilled water.

2.3 Green Synthesis of Copper Nanoparticles (CuNPs)

The synthesis of CuNPs was carried out by mixing the *T. chebula* aqueous extract and $CuSO_4$ solution in a 1:1 (v/v) ratio under constant stirring. The reaction mixture was maintained at ambient temperature (25–30 °C) and incubated in the dark for 24 hours to prevent photoreduction. The preliminary formation of CuNPs was indicated by a distinct color change of the solution from light blue to greenish-brown, attributed to surface plasmon resonance.

2.4 Purification of Synthesized CuNPs

The reaction mixture was centrifuged at 10,000 rpm for 15 minutes. The resulting pellet containing CuNPs was washed sequentially with distilled water and ethanol (2–3 times) to remove unbound phytochemicals and impurities. The purified nanoparticles were dried in a hot air oven at 50 °C and stored in an airtight container for further analysis.

2.5 Characterization of Copper Nanoparticles

The formation and stability of the synthesized CuNPs were confirmed using UV-Visible spectroscopy.

2.6 UV-Visible Spectroscopy

The synthesized copper nanoparticles (CuNPs) were characterized using a UV–Vis spectrophotometer. A colloidal suspension of CuNPs was prepared and transferred into a clean quartz cuvette, with the solvent used for nanoparticle dispersion serving as the blank. The absorbance spectrum was recorded over a wavelength range of 300–800 nm, and the characteristic surface plasmon resonance (SPR) peak of CuNPs, typically observed around 560–580 nm, was analyzed to confirm nanoparticle formation and assess their optical properties. Measurements were repeated in triplicate to ensure reproducibility.

2.7 Photocatalytic Dye Degradation Activity

The photocatalytic activity of the synthesized copper nanoparticles was evaluated using methylene blue (MB) as a model organic dye. A known concentration of MB solution was prepared, and CuNPs were added under continuous stirring. The reaction mixture was exposed to ambient light, and aliquots were withdrawn at fixed time intervals (0, 30, 60, 90, and 120 minutes). The residual dye concentration was monitored by measuring the absorbance at the characteristic maximum wavelength (λmax = 664 nm) of MB using a UV–Visible spectrophotometer. The percentage degradation of the dye was calculated by comparing the absorbance values at different time intervals with the initial value (A_0) (Parvathalu *et al*., 2023).

2.8 Antibacterial Activity

The antibacterial efficacy of the synthesized copper nanoparticles was assessed against both Gram-negative and Gram-positive bacterial pathogens using the agar well diffusion method. Gram-negative strains included *Klebsiella* sp., *Enterobacteriaceae* sp., *Vibrio* sp., and *Salmonella* sp., while Gram-positive strains comprised *Staphylococcus* sp., *Bacillus* sp., *Corynebacterium* sp., and *Micrococcus* sp. Fresh bacterial cultures were uniformly swabbed onto nutrient agar plates, and wells were aseptically prepared. Varying concentrations of CuNPs (200, 500, and 1000 µg/mL) were loaded into the wells, with distilled water serving as the negative control. The plates were incubated at 37 °C for 24 hours, after which the antibacterial activity was determined by measuring the diameter of the inhibition zones (mm) surrounding each well.

3. Results and discussion

The copper nanoparticles synthesized using *Terminalia chebula* fruits in the present study demonstrated efficient formation, as indicated by the observed color change (Figure. 1) and characteristic surface plasmon resonance. The rich phytochemical content of *T. chebula*, including tannins, gallic acid, flavonoids, and polyphenols, likely acted as natural reducing and stabilizing agents, promoting the formation of uniform and stable CuNPs. The optical characterization of the synthesized copper nanoparticles was carried out using a UV-Visible spectrophotometer. The absorbance spectrum was recorded in the wavelength range of 200-800 nm to monitor nanoparticle formation. Particular attention was given to identifying the absorption maximum (λmax), as copper oxide nanoparticles typically exhibit a surface plasmon resonance (SPR) peak within 250–350 nm.

Figure 1. Schematic Diagram showing the synthesis of Coppersulfate Nanoparticles from *Terminalia Chebula* Linn Aqueous extract

3.1 Photocatalytic Degradation of Methylene Blue

The photocatalytic degradation of methylene blue (MB) using copper sulphate nanoparticles synthesized from *Terminalia chebula* (Kadukai) extract showed a progressive increase in degradation efficiency over 60 min (Figure 2). Within the first 10 min, 10.40% of MB was degraded, which increased to 28.50% at 20 min and 51.70% at 30 min, indicating rapid catalytic activity during the initial phase. Subsequently, a slower but steady increase was observed, reaching 53.20% at 40 min, 57.50% at 50 min, and a maximum of 59.06% at 60 min (Table 1). The initial sharp rise suggests efficient generation of reactive oxygen species facilitated by the bioactive compounds in *T. chebula*, which act both as reducing and capping agents. The gradual plateau phase indicates depletion of available dye molecules and partial surface passivation of the nanoparticles. Linear regression analysis of the degradation data yielded the equation $y = 0.0095x + 0.1021$ with a correlation coefficient $R2 = 0.8191$, suggesting a reasonably good linear relationship between reaction time and degradation efficiency under the given experimental conditions. These findings are consistent with previous studies on green-synthesized copper- based nanoparticles. From the previous study, copper ―nanoflowers‖ from *Ficus benghalensis* leaf extract degraded ~72% of MB in 85 min (Agarwal *et al*., 2016), while CuO nanoparticles synthesized from plant extracts achieved ~90% removal in 120 min under visible light (Mahmoud *et al*., 2021). An eco-friendly CuO system derived from citrus peel extract demonstrated >95% MB degradation over 120 min, with rate constants of 0.0255 min^{-1} (CuO) and 0.033 min^{-1} (modified CuO) (Mahmoud *et al*., 2024). Compared to these studies, the Kadukai-derived copper system exhibits good catalytic activity, achieving nearly 60% degradation within 60 min. The slightly lower efficiency may be due to factors such as larger particle size, limited active surface area, or catalyst loading, suggesting that further optimization—through particle size reduction, surface area enhancement, or heterojunction formation—could improve the photocatalytic performance.

Figure 2. Dye Degradation Activity

Table 1. Degradation Percentage at different time intervals

Time (Min)	Absorbance at 664 nm	% Degradation
0	0.298	0
10	0.267	10.40%
20	0.213	28.50%
30	0.185	51.70%
40	0.17	53.20%
50	0.127	57.50%
60	0.122	59.06%

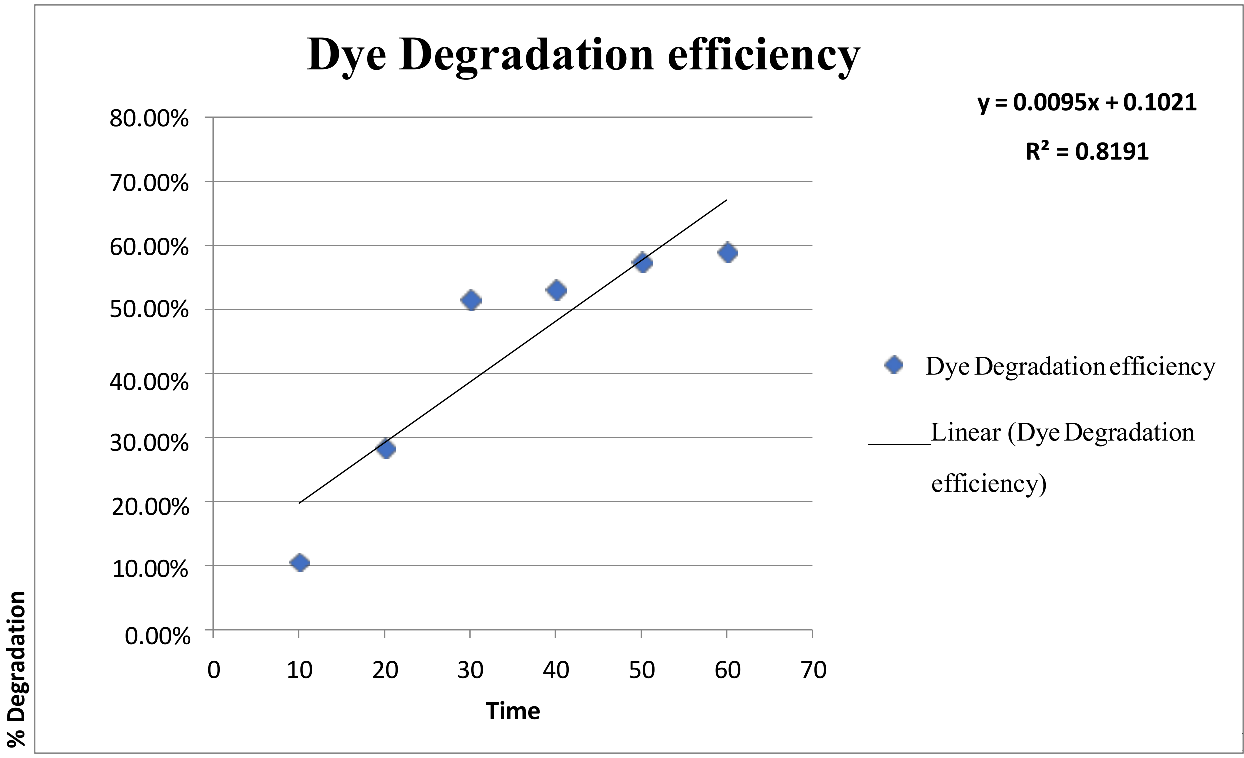

Figure 3. Dye Degradation Efficiency

3.2 Anti - bacterial activity

The antibacterial activity of copper nanoparticles (CuNPs) synthesized using *Terminalia chebula* fruits was evaluated against selected Gram-negative (*Klebsiella sp., Enterobacteriaceae sp., Vibrio sp., Salmonella sp.*) and Gram-positive (*Staphylococcus sp., Bacillus sp., Corynebacterium sp., Micrococcus sp.*) pathogens at concentrations of 200, 500, and 1000 µg/mL (Tables 2 and 3). Antibacterial effect was observed for all tested microorganisms. For Gram- negative pathogens, inhibition zones ranged from 10–11 mm at 200 µg/mL and increased progressively to 13–14 mm at 1000 µg/mL, with *Salmonella sp.* showing the highest susceptibility. *Enterobacteriaceae sp.* and *Vibrio sp.* exhibited intermediate sensitivity, while *Klebsiella sp.* showed comparatively lower inhibition. Among Gram-positive strains, *Bacillus sp.* and *Micrococcus sp.* demonstrated the largest inhibition zones (14 mm) at the highest concentration, whereas *Staphylococcus sp.* and *Corynebacterium sp.* displayed moderate A clear dose- dependent susceptibility, suggesting possible intrinsic resistance mechanisms or reduced interaction with the nanoparticles. No inhibitory effect was observed in the negative control, confirming that the antibacterial activity was specifically due to the CuNPs (Figure 4).

The enhanced activity at higher concentrations may be attributed to increased interaction of nanoparticles with bacterial cell walls, leading to membrane disruption, leakage of intracellular components, and generation of reactive oxygen species (ROS). The presence of phytochemicals from *T. chebula*, such as tannins, flavonoids, and polyphenols, likely acted synergistically with CuNPs, enhancing the antibacterial efficacy through additional mechanisms such as protein denaturation and enzyme inactivation. These findings are consistent with previous reports on plant- mediated copper nanoparticles, which have demonstrated broad-spectrum antimicrobial potential against both Gram-negative and Gram-positive bacteria (Munusamy *et al*., 2023; Keerthika *et al*., 2021, Anbalagan *et al*., 2024, Cao *et al*., 2021)

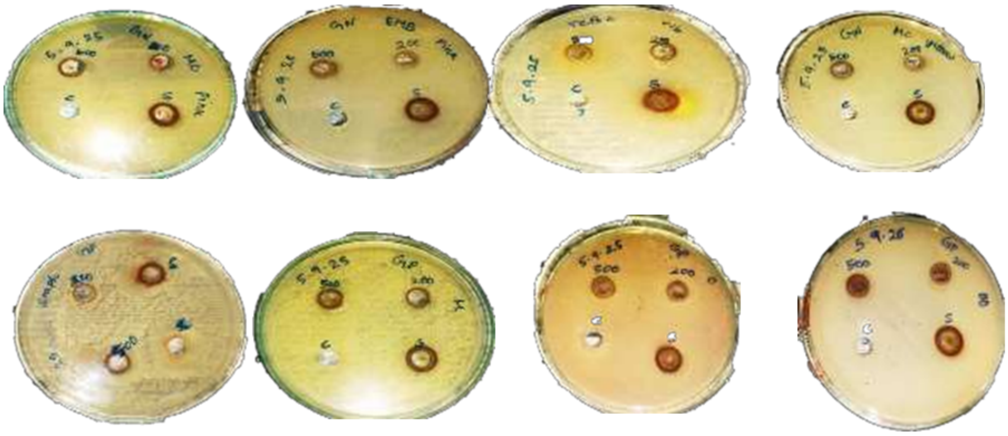

Figure 4. Antibacterial activity

Table 2. Gram Negative pathogens

Sample	Concentration (µg/mL)	Zone of Inhibition (mm) – *Klebsiella sp*	*Enterobacteriaceae sp*	*Vibrio sp*	*Salmonella sp*
Terminalia chebula CU NP	500	11 mm	13 mm	11 mm	14 mm
	200	10 mm	11 mm	11 mm	11 mm
	1000	13 mm	14 mm	14 mm	14 mm
	NC	-	-	-	-

Table 3. Gram Positive pathogens

Sample	Concentration (µg/mL)	Zone of Inhibition (mm) – *Staphylococcus sp*	*Bacillus sp*	*Corynebacterium sp*	*Micrococcus sp*
Terminalia chebula CU NP	**500**	11 mm	12 mm	11 mm	12 mm
	200	10 mm	11 mm	11 mm	11 mm
	1000	12 mm	14 mm	12 mm	14 mm
	NC	-	-	-	-

Table 4. Results and Interpretation

Zone size (mm)	Interpretation of activity
≤ 6 mm	No activity (or negligible)
7–10 mm	Weak activity
11–15 mm	Moderate activity
16–20 mm	Strong activity
> 20 mm	Very strong activity

Overall, the results indicate that *T. chebula*-derived CuNPs are effective antibacterial agents, with potential applications in therapeutic formulations, water disinfection, and as antimicrobial coatings. Their dose-dependent activity and broad-spectrum efficacy suggest that optimization of concentration and formulation could further enhance their practical applicability.

Conclusion

The study demonstrates that *Terminalia chebula*-derived copper nanoparticles possess multifunctional properties, combining eco-friendly synthesis with practical applications. Their effective photocatalytic performance and antibacterial activity highlight their potential as sustainable alternatives for environmental remediation and antimicrobial solutions. The integration of plant-based bioactive compounds not only stabilizes the nanoparticles but may also enhance their interaction with pollutants and microorganisms. These findings open avenues for further research on optimizing particle characteristics and exploring diverse applications in water treatment, antimicrobial coatings, and biotechnological innovations.

CRediT authorship contribution statement

R. Sumathi: Investigation, Writing- original draft preparation, Data curation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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