

Green Synthesis and Characterization of CuO Nanoparticles Using Sterculia Foetida Leaf Extract

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ABSTRACT

This study aims to provide an updated overview of the green synthesis of CuO NPs using *Sterculia foetida* leaf extract, their characterization, and antibacterial activity. CuO NPs were synthesized by reducing 1 mM CuSO₄·5H₂O with 2% (m/v) aqueous *Sterculia foetida* leaf extract under optimized conditions (pH 11). The formation of CuO NPs was initially indicated by a color change from colorless to light green and subsequently to greenish yellow. The reaction was monitored using UV–Vis spectrophotometry, which showed a strong absorption peak at 333 nm. The synthesized CuO NPs exhibited antibacterial activity against the human pathogens *Salmonella typhimurium* and *Escherichia coli*, with average inhibition zones of 12.5 mm and 8 mm, respectively.

Keywords: *Green Synthesis, Sterculia Foetida, Nanoparticles, Copper, Metal.*

I. INTRODUCTION

The rapid advancement of nanotechnology has created significant opportunities in the fields of materials science, medicine, agriculture, environmental remediation, catalysis, and electronics. Nanoparticles exhibit unique physicochemical properties, including high surface area, enhanced catalytic activity, optical characteristics, and improved reactivity, which differ considerably from their bulk counterparts. Among various metal oxide nanoparticles, copper oxide (CuO) nanoparticles have attracted considerable scientific interest because of their distinctive semiconductor properties, relatively low cost, chemical stability, antimicrobial activity, catalytic potential, and suitability for a wide range of technological applications. CuO is a p-type semiconductor with a narrow band gap and has been investigated for applications in sensors, photocatalysis, energy-storage devices, antimicrobial formulations, and environmental treatment. Conventional methods for synthesizing CuO nanoparticles commonly involve chemical reducing agents, organic solvents, high temperatures, or other processing conditions that may generate hazardous by-products and require substantial energy. Consequently, the development of environmentally friendly, economical, and sustainable

approaches for nanoparticle synthesis has become an important area of research. Green synthesis provides an alternative strategy in which biological materials such as plants, microorganisms, algae, and agricultural residues are utilized as reducing, stabilizing, and capping agents. Plant-mediated synthesis is particularly advantageous because it is generally simple, rapid, cost-effective, and does not require sophisticated biological culture conditions.

Plant extracts contain naturally occurring phytochemicals such as flavonoids, phenolic compounds, tannins, alkaloids, terpenoids, proteins, carbohydrates, and other biomolecules that can participate in the reduction and stabilization of metal ions and influence the nucleation, growth, morphology, and surface characteristics of nanoparticles. *Sterculia foetida*, commonly known as wild almond or Java olive, is a plant belonging to the family Malvaceae and contains various bioactive phytoconstituents that make its leaves a potentially useful biological resource for nanoparticle synthesis. The use of *S. foetida* leaf extract in the synthesis of CuO nanoparticles therefore represents an interesting approach toward combining plant-based resources with nanotechnology while reducing dependence on potentially hazardous synthetic chemicals. During green synthesis, phytochemicals present in the leaf extract may interact with copper ions and facilitate the formation and stabilization of CuO nanoparticles. The resulting nanoparticles require systematic characterization to confirm their formation and determine their structural, morphological, optical, and chemical properties. Techniques such as ultraviolet-visible (UV-Vis) spectroscopy, Fourier-transform infrared (FTIR) spectroscopy, X-ray diffraction (XRD), scanning electron microscopy (SEM), transmission electron microscopy (TEM), and energy-dispersive X-ray spectroscopy (EDS/EDX) can provide valuable information regarding optical absorption, functional groups, crystalline structure, particle morphology, size, elemental composition, and surface characteristics.

II. REVIEW OF LITERATURE

Sukhadiya, Madhuri. (2025) Using nontargeted gas-chromatography mass-spectrometry (GC-MS), this research examined eight separate sources of *Sterculia fetida* leaf samples from the south Gujarat area for the presence of volatile phytochemicals. The leaves contained a total of 38 volatile phytochemical substances. With area percentages of 17.16, 11.39, 11.35, 10.12, and 8.11 respectively, Lupeol, Squalene, dl- α -Tocopherol, β -Amyrin, and D-Friedoolean-14-en-3-ol topped the list of compounds based on the relative area *S. fetida* %. Next came several more significant ones, including Phytol, Sitosterol, Neophytadiene, 24-Noroleana3,12-diene, and Stigmasterol. Additionally, molecules such as β -Tocopherol, Campesterol, -Tocopherol, and d-Tocopherol were found, which have significant γ biological activity. Antioxidant, antibacterial, anti-inflammatory, and antimicrobial were the most often reported biological capabilities by 14, 9, 8, and 6 substances, respectively, according to the meta-analysis for biological activity confirmation. Even though there were fewer compounds overall, they exhibited a wide range of biological activities, including anti-diabetic, antifungal, analgesic, antidepressant, anti-allergic, anti-pyretic, anti-ulcer, anti-cancer, anti-tumour, antispasmodic, anti-aging, anti-HIV-1, and other properties. Therefore, the leaf contains a wealth of biologically active chemical components including *S. fetida* derivatives.

Bhuiyan, Imrul Hasan et al., (2025) Since it provides affordable and environmentally friendly methods to produce nanoparticles, the advancement of green technology has attracted a lot of interest from researchers. In this paper, we detail the process of creating CuO Nanoparticles (CuO NPs) including *Mesua ferrea* extract using an alkaline hydrolysis method. Characterisation of the synthesised CuO NPs was carried out using FTIR, FESEM, XRD, zeta potential, DLS, EDX, and UV-vis studies. Based on the JCPDS standard card no: 48-1548, the XRD method confirmed that the CuO NPs exhibited a monoclinic crystalline phase, with an average crystallite size of 32.31 nm for calcined CuO NPs and 18.57 nm for non-calcined CuO NPs. The FESEM pictures demonstrate the spherical nanoparticle shape and validate the creation of nanoparticles with diameters ranging from 10 to 60 nm. The presence of linkages in the 400 to 700 cm^{-1} region of the FTIR spectra demonstrated the production of CuO NPs. A look at the synthetic CuO NPs' thermal behaviour is provided by the TGA and DTG curves. The synthesised CuO NPs had a zeta potential of -33.4 mV and -61.9 mV, correspondingly. The UV-vis spectrophotometer recorded the highest absorbance between 250 and 350 nm, and the band gap energy of the synthesised CuO NPs was determined to be 2.502 and 1.99 eV, respectively, according to the Tauc plot. These values were favourable for the authorised direct transition mode. In the DPPH test, the highest radical inhibition values for calcined and non-calcined CuO NPs were 61.64% and 67.51%, respectively. In the ABTs assay, the values were 52.37% and 79.43%. The antibacterial effectiveness of the CuO NPs that were created was examined against a fungus, two gram-positive bacteria, and two gram-negative bacteria. The CuO NPs not only show heightened sensitivity to the gram-negative bacteria *S. abony* and *Escherichia coli*, but they also have a bigger inhibition zone (16.8 ± 0.76 mm) against the gram-positive bacterium *Staphylococcus aureus*.

Yadav, H.J. et al., (2024) Using *Mimosa pudica* leaf extract as a fuel source, we present here the production of copper oxide nanoparticles (NPs) by low temperature burning. The synthetic particles have a highly crystalline structure, with an average crystallite size ranging from 27 to 40 nanometres. Copper oxide nanoparticles are agglomerated and have a loosely bonded shape. Additionally, harmful bacterial strains such as Gram-negative *Escherichia coli* and Gram-positive *Staphylococcus aureus* are effectively inhibited by the antibacterial activity of as-formed CuO NPs.

Moroda, Mekides et al., (2024) The rise of green technology has piqued the curiosity of scientists looking for sustainable, cost-effective ways to synthesise nanoparticles (Nps). The researchers here used a green synthesis method to create copper oxide nanoparticles (CuO Nps), with the help of a reducing agent and a capping agent derived from the leaves of the *Rosmarinus officinalis* plant. The materials were examined using a variety of analytical tools, including scanning electron microscopy (SEM), ultraviolet-visible spectra (UV-VIS), X-ray diffraction (XRD), and Fourier transform infrared (FTIR). The UV-Visible spectra revealed a red shift with increasing CuO Nps weight fraction and substantial absorption for the green synthesised CuO Nps characteristic band. The presence of CuO Nps, with their characteristic monoclinic structure and crystalline size varying between 18 and 26 nm, was verified by the XRD examination. According to the FTIR spectrum analysis, the bioactive compounds' (O-H) functional groups, which serve as reducing and capping agents in the plant extract's CuO Nps synthesis, are responsible for the strong and wide peak seen at

3384 cm^{-1} . The green synthesised CuO NPs showed up in scanning electron microscopy (SEM) pictures looking aggregated and almost spherical. We tested CuO Nps against gram-positive (*S. aureus* and *B. cereus*), gram-negative (*E. coli*), and fungal pathogen (*C. albicans*) microorganisms. The findings showed that the antibacterial activity of CuO Nps was greatest at a 50% weight fraction against *E. coli*, *S. aureus*, *C. albicans*, and *B. cereus*, with zones of inhibition of 26.0 ± 0.10 , 25.0 ± 0.15 , 18.1 ± 0.32 , and 17.0 ± 0.0 mm, respectively. Additionally, the MIC of CuO Nps for *E. coli* was determined to be 1.56 mg/mL, whereas for *S. aureus* and *Candida albicans* it was 3.12 mg/mL.

III. MATERIALS AND METHODS

All chemicals used were of analytical grade and obtained from reputed Indian suppliers. Fresh leaves of *Sterculia foetida* were collected, authenticated, and used as plant material. $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, FeCl_3 , HCl, H_2SO_4 , ethanol, methanol, KBr, NaOH, ammonia, Benedict's solution, MgCl_2 , iodine, KI, chloroform, agar, and gentamicin were procured from standard laboratory chemical suppliers in India. Deionized water was used throughout the study.

Preparation of *Sterculia Foetida* Leaf Extract

Fresh leaves were washed thoroughly with tap and distilled water, cut into small pieces, and homogenized. Two grams of leaf material were mixed with 100 mL deionized water, stirred for 2–3 min, and heated at 50–60 °C for 2 min. The mixture was cooled and filtered through qualitative followed by quantitative filter paper. The filtrate was stored at 4 °C until use.

Phytochemical Screening

The leaf extract was screened qualitatively for alkaloids, glycosides, tannins, flavonoids, saponins, steroids, phenols, and carbohydrates using standard chemical tests, including Wagner's, ferric chloride, Salkowski, Benedict's, and iodine tests. Positive results were identified based on characteristic color changes or precipitate/foam formation.

Synthesis of CuO Nanoparticles

For synthesis, 45 mL of 1 mM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ solution was mixed with 2 mL of 2% (m/v) *Sterculia foetida* leaf extract and stirred at room temperature until the color changed from light blue to light green. The mixture was heated at 50–60 °C for 6 min and cooled. Subsequently, 1 M NaOH was added dropwise with continuous stirring until pH 11 was reached. The resulting CuO nanoparticles were characterized and evaluated for antibacterial activity.

Characterization of CuO Nanoparticles

The synthesized CuO nanoparticles were characterized by UV–Visible spectroscopy, and FT-IR spectroscopy. UV–Visible spectra were recorded from 200–700 nm at 1 nm resolution to monitor nanoparticle formation and Cu^{2+} conversion. For FT-IR analysis, the leaf extract and synthesized nanoparticles were mixed with KBr and scanned from 400–4500 cm^{-1} at 8 cm^{-1} resolution. The spectra were compared to identify functional groups involved in nanoparticle synthesis and stabilization.

IV. RESULTS AND DISCUSSION

Phytochemical Test

The aqueous leaf extract of *Sterculia foetida* was subjected to qualitative phytochemical screening. The extract showed the presence of phenols, flavonoids, carbohydrates, tannins, alkaloids, and other phytochemicals, as shown in Figure 1. These bioactive compounds, particularly phenolic and flavonoid compounds, may act as reducing, capping, and stabilizing agents during the green synthesis of CuO nanoparticles, thereby contributing to their formation and stability.

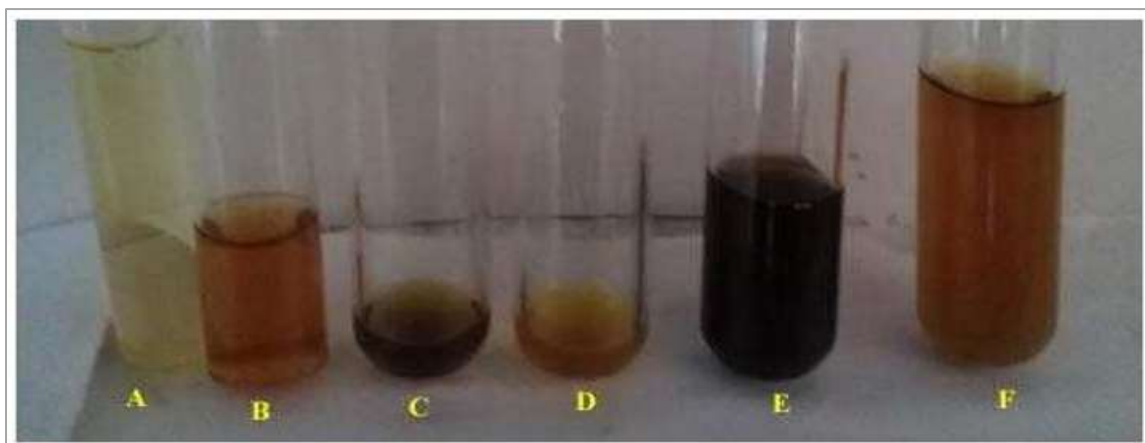


Figure 1: Qualitative Phytochemical Screening of *Sterculia foetida* Leaf Extract
(A) Alkaloids, (B) Glycosides, (C) Tannins, (D) Flavonoids, (E) Phenols, and (F) Carbohydrates

The observed color changes in the phytochemical tests indicate reactions between the plant constituents and specific reagents. For instance, alkaloids interact with their respective reagents, while ferric chloride forms colored complexes with phenolic compounds. These characteristic color changes were used as preliminary indicators for identifying phytochemicals in the *Sterculia foetida* leaf extract.

Characterization of CuO Nanoparticles

Due to the unavailability of advanced instruments, the nanoparticles were characterized using UV–Visible spectroscopy, and FT-IR spectroscopy. These methods provided preliminary evidence of nanoparticle formation and information on their optical properties and functional groups.

• UV–Visible Analysis

UV–Visible spectroscopy was used to study the optical properties of the synthesized CuO nanoparticles. The CuO nanoparticles showed a strong absorption band at approximately 333 nm, while the *Sterculia foetida* leaf extract showed an absorption band at approximately 294 nm (Figure 2). The shift from 294 to 333 nm indicates changes in the optical properties associated with CuO nanoparticle formation.

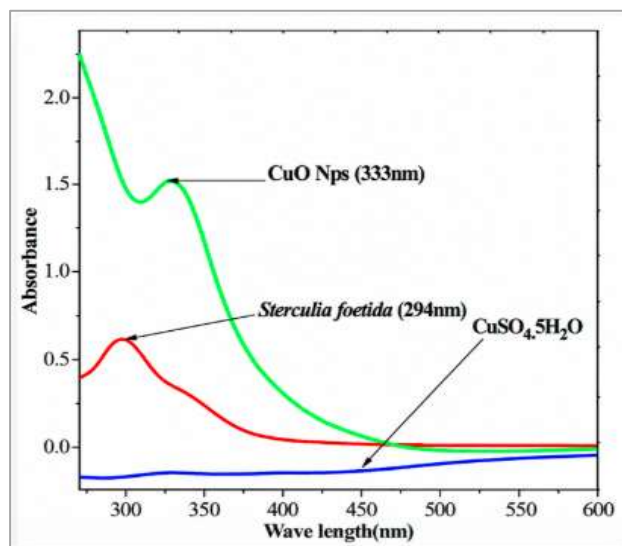


Figure 2: UV-Vis Spectra of CuO NPs and Precursors

Optimization of Precursor Concentration

Precursor concentration was optimized by varying the $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ concentration without adjusting the pH. The synthesized CuO nanoparticles showed absorption peaks close to that of the *Sterculia foetida* leaf extract (294 nm), with a slight shift toward longer wavelengths. The maximum absorption of approximately 296.5 nm was observed at 1–3 mM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ (Figure 3). These spectral changes suggest that precursor concentration influences the optical properties and formation of CuO nanoparticles and was therefore considered during further optimization.

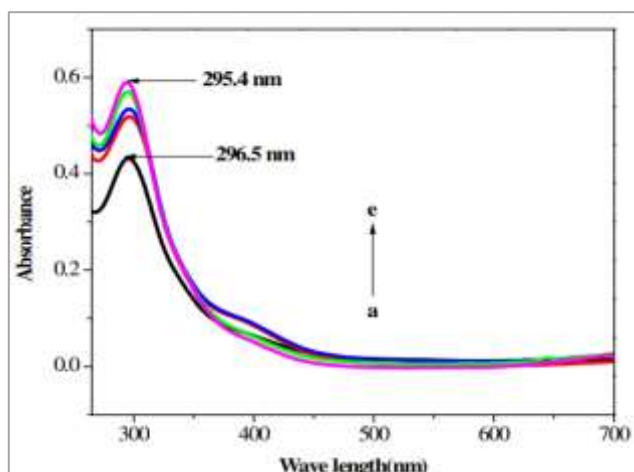


Figure 3: UV-Vis Spectra of CuO NPs at Different CuSO_4 Concentrations

At 4 and 5 mM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, the absorption peaks remained close to that of the *Sterculia foetida* leaf extract (≈ 295.4 nm), indicating less pronounced changes. The reaction mixture was therefore adjusted to pH 10 using NaOH to promote CuO nanoparticle formation. After pH adjustment, a clear shift toward longer wavelengths was observed. Among the tested concentrations, 1 mM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ showed the most intense absorption and greatest shift compared with 2–5 mM concentrations (Figure 4). Thus, 1 mM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ was selected for further optimization.

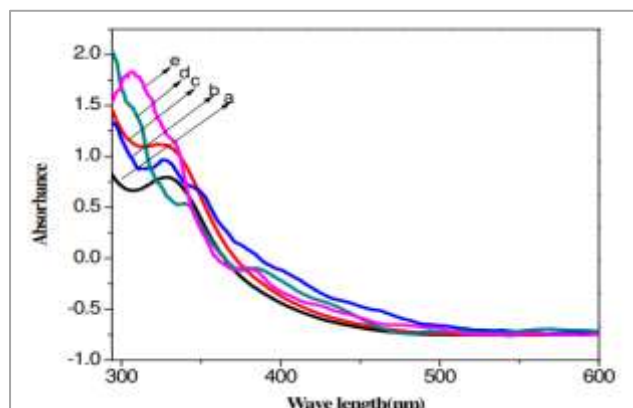


Figure 4: UV-Vis Spectra of CuO NPs at pH 10

Optimization of the Concentration of *Sterculia foetida* Leaf Extract

The concentration of *Sterculia foetida* leaf extract was optimized using the selected precursor concentration. Among the tested concentrations, 3%, 4%, 5%, and 6% (m/v) extracts showed more intense absorption peaks than 1% and 2% extracts (Figure 5). However, these spectra also showed considerable background noise. The differences may be due to variations in phytochemical concentrations involved in the reduction, complexation, capping, and stabilization of copper species during CuO nanoparticle synthesis.

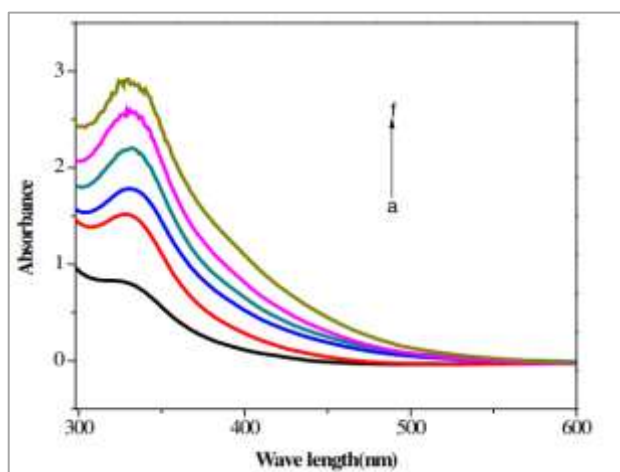


Figure 5: UV-Vis Spectra of CuO NPs at Different Extract Concentrations

Optimization of pH of the Mixture of *Sterculia foetida* Leaf Extract and Precursor

The pH of the reaction mixture was optimized at 6, 7, 8, 9.5, 10, 11, 12, and 13. The initial pH of the $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ solution, *Sterculia foetida* leaf extract, and reaction mixture was approximately 5.4, 5.5, and 4.15, respectively. Among the tested conditions, pH 11 produced the strongest and most distinct UV-Visible absorption response (Figure 6). Strong absorption was observed mainly between pH 9–12, with maximum intensity at pH 11. Changes in absorption with pH may be related to copper-ion speciation, hydroxide concentration, phytochemical interactions, nucleation, and nanoparticle stabilization. The alkaline medium promotes the formation of copper hydroxide intermediates, which can subsequently convert to CuO. However, excessive alkalinity may favor the

formation of $\text{Cu}(\text{OH})_2$, resulting in altered spectral behavior at higher pH values. UV–Visible spectroscopy alone cannot confirm changes in particle size or morphology; complementary techniques such as XRD, SEM, TEM, or DLS are required. Therefore, the optimized conditions were 2% (m/v) *Sterculia foetida* leaf extract, 1 mM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, and pH 11, which were used for subsequent CuO nanoparticle synthesis.

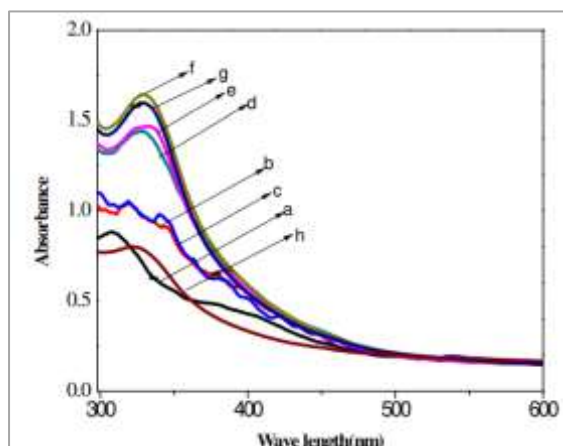


Figure 6: UV–Vis Spectra of CuO NPs at Different pH Levels (a) 6, (b) 7, (c) 8, (d) 9, (e) 10, (f) 11, (g) 12 and (h) 13

The stability of the synthesized CuO nanoparticles was evaluated by comparing their UV–Visible absorption spectra at different time intervals after synthesis. The UV–Visible spectrum recorded 24 h after synthesis showed nearly the same absorption intensity, peak broadness, and λ_{max} value as that recorded 1 h after synthesis. Furthermore, after six days of storage, no significant visible change in the absorption peak intensity or λ_{max} value was observed, as shown in Figure 7. A slight increase in absorption intensity and peak broadening was nevertheless observed after prolonged storage. These changes may be associated with minor alterations in particle aggregation, surface interactions, or colloidal stability during storage. The results indicate that the synthesized CuO nanoparticles maintained relatively stable optical characteristics over the investigated period.

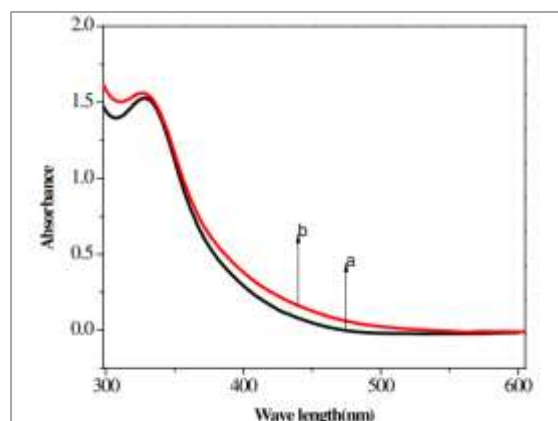


Figure 7: The UV-Vis Spectra of CuO NPs after (a) 1 Day and (b) 6 Days

• FT-IR Analysis

FT-IR spectroscopy was used to identify the functional groups present in the *Sterculia foetida* leaf extract and synthesized CuO nanoparticles and to assess their possible role in nanoparticle formation and stabilization. The leaf extract showed a strong band at 3416 cm^{-1} , mainly corresponding to O–H stretching of hydroxyl groups, with possible contributions from N–H groups. After nanoparticle synthesis, this band shifted to 3459 cm^{-1} , indicating changes in the chemical environment of these functional groups due to their interaction with copper species and the nanoparticle surface. A band around 1626 cm^{-1} may be associated with C=C and/or C=O stretching vibrations of phytochemical constituents. The absorption band near 646 cm^{-1} can be attributed to metal–oxygen vibrations, supporting the formation of CuO. Overall, the FT-IR results suggest that hydroxyl, carbonyl, amine, and other oxygen- or nitrogen-containing groups in the *Sterculia foetida* extract may contribute to the formation, capping, and stabilization of CuO nanoparticles.

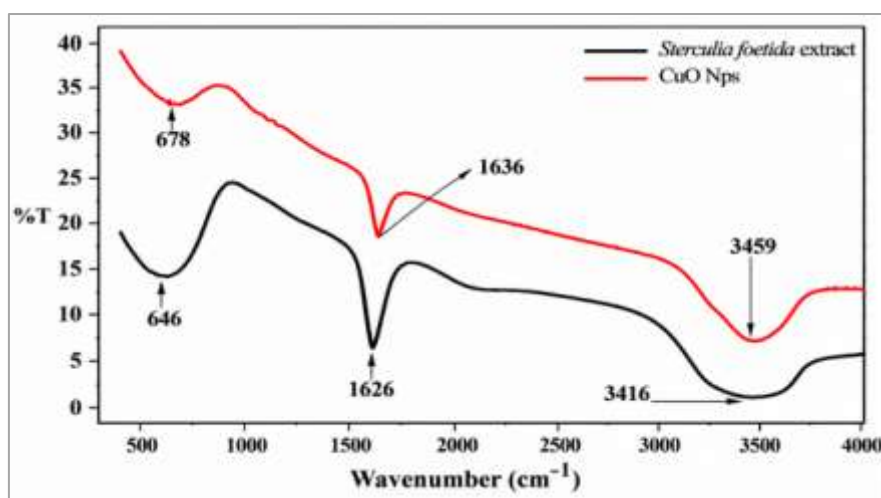


Figure 8: FT-IR Spectra of Leaf Extract of Sterculia Foetida

The synthesized product showed a major absorption band at approximately 678 cm^{-1} , which can be attributed to Cu–O stretching vibrations, providing spectroscopic evidence for CuO nanoparticle formation. The band at 1626 cm^{-1} in the *Sterculia foetida* leaf extract shifted to approximately 1636 cm^{-1} after nanoparticle synthesis (Figure 8), indicating changes in the chemical environment of oxygen- and nitrogen-containing functional groups. These shifts suggest that phytochemicals such as phenols, flavonoids, proteins, and other biomolecules may interact with the CuO nanoparticle surface and contribute to their capping and stabilization. However, FT-IR analysis alone cannot conclusively identify specific biomolecules or determine the exact mechanism of nanoparticle stabilization.

V. CONCLUSION

The present study demonstrates that *Sterculia foetida* leaf extract can be effectively used as a natural and eco-friendly agent for the synthesis of CuO nanoparticles. The simple aqueous extraction method and controlled reaction conditions enabled the formation of CuO nanoparticles without the need for harsh or complex chemical procedures. Phytochemical screening indicated the presence of various bioactive compounds in the leaf extract, which may contribute to the reduction, formation, and

stabilization of the nanoparticles. The synthesized CuO nanoparticles were characterized using UV–Visible and FT-IR spectroscopy, providing information about nanoparticle formation and the functional groups associated with their stabilization.

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