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Zeta Potential Effect on CuO Nanoparticles Synthesized from *Sterculia foetida* Leaf Extract with Application

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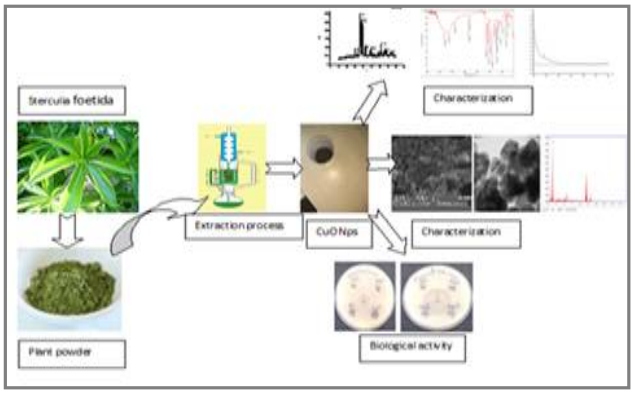
ABSTRACT

The Copper oxide nanoparticles have unique optical property and excellent antibacterial activity. The nanoparticles, nanocrystals and nanoclusters are called as zero dimensional because of the movement of charge carriers is confined in all the three directions. The present study aims for the synthesis of Copper oxide nanoparticles (CuO NPs) using Sterculia foetida leaf extract as fuel by solution combustion method. The synthesized CuO NPs was confirmed by UV-Visible spectroscopy with the peak at 340 nm and at the temperature of 400± 10℃, FT-IR wavelength observed between 1018 to 502 cm^{-1}. At 1018 cm^{-1} C-O stretching and at 502 cm^{-1}M-O-M bending found where (M=Cu). Crystalline structure and the formation of monoclinic phase revealed by PXRD pattern, the percentage of copper and oxygen of CuO NPs was confirmed with EDAX analysis. The SEM and TEM images indicate spherical in shape of CuO NPs with the average size of 5-51 nm. Staphylococcus aureus (Gram positive) and Escherichia coli (Gram negative) bacteria's were taken to study about antibacterial activity against the green synthesized CuO NPs. The CuO nanoparticles have pharmaceutical and other biomedical applications. The stability of CuO nanoparticles confirmed through zeta potential values.

Graphical Abstract



Keywords: Leaf extract, *Sterculia foetida*; CuO NPs, Antibacterial activity.

INTRODUCTION

Nanoparticles were predominantly used in the field of imaging [1], sensing [2], targeted drug delivery [3], batteries, solar cells and space. In pesticide formulations copper-based compounds shows efficient biocide properties [4]. CuO NPs have unique semiconductor and optical properties [5-8]. CuO nanostructures were prepared by sol-gel [9], microwave irradiations [10], electrochemical methods [11] and solid-state reaction method [12]. Among all Green route method of nanoparticles synthesis using solution combustion method is very cost effective and eco-friendly and reduces the usage of harmful chemicals [13]. Many metal oxide nanoparticles were reported from *Aloe barbadensis* miller [14], *Carica papaya* [15], Lemon grass [16] and tamarind [17] by various methods. Strontium aluminates doped with different amounts of europium rare earth ions ($Sr_4Al_{14}O_{25}:Eu^{2+}$) were synthesized by solution combustion method, because it is a good method to produce homogeneous, pure powders with fine particle size [18]. The solution combustion method is employed for the CuO nanoparticle synthesis using *Rauvolfia serpentine* leaf extract as fuel and XRD indexed to be monoclinic structure [19]. At present study CuO NPs were prepared using aqueous leaf extract of *Sterculia foetida* as a fuel by solution combustion method. *Sterculia foetida* belongs to Malvaceae family. It has been found in India, Taiwan, Indochina, Australia and Philippines. The formation of CuO NPs WERE confirmed with the aid of PXRD, EDAX, UV-Vis, FTIR, SEM and TEM. The antibacterial activity was tested with *Staphylococcus* (Gram+ve) and *Escherichia coli* (Gram-ve) using agar well diffusion method.

MATERIALS AND METHODS

Preparation of leaf extract: *Sterculia foetida* leaves were collected from NHCE Campus, Marathalli, Bangalore, Karnataka, India. The leaves were washed with water to remove dust particles and dried in the absence of sun light at room temperature. Then the leaves were powdered mechanically using mixer grinder, sieved and subjected to extraction through Soxhlet apparatus by the use of de-ionized water for 68 h at the temperature of 50-60°°C. The obtained aqueous solution is subjected to concentration by the use of rotary flash evaporator at 40±5°C under reduced pressure(Buchi, Flawil, Switzerland), then it is dried in hot air oven at 55-60°C, from the dried crude extract small amount is used for the nanoparticle synthesis.

Synthesis of CuO nanoparticles: The CuO nanoparticles were prepared by solution combustion method using aqueous leaf extract of *Sterculia foetida* as a fuel. In this process 0.1 g of dried crude aqueous leaf extract of *Sterculia foetida* and stoichiometric amount of copper nitrate trihydrate was dissolved in 10 mL of distilled water and constantly stirred for 10 min to get homogeneous mixture. This reaction mixture kept in a pre-heated muffle furnace maintained at 400±10°C for 3-4 min. The material was removed from muffle furnace and the obtained black colored powder sample was stored in airtight container for further analysis [20, 21].

Antibacterial activity of CuO NPs: The CuO NPs were screened for antibacterial activity against *Staphylococcus aureus* and *Escherichia coli* bacterial strains using agar well diffusion method. The sample was prepared with 10 mg of CuO NPs using 1000 µL of distilled water. The Nutrient Agar (NA) plates were inoculated with test organisms. The plates were evenly spread out. Then wells were prepared in the plates with a cork borer, each well was loaded with 15, 30, 45 and 60 µL of different concentrations of sample and 10 mg of tetracycline dissolved in 1ml of DMSO was used as a positive control for antibacterial activity. The plates were incubated for 24 h at 37°C. The development of inhibition zone around the well was measured (diameter) and recorded [22, 23].

RESULTS AND DISCUSSION

The CuO NPs were synthesized using *Sterculia foetida* leaf extract as reducing agent with copper nitrate trihydrate precursor solution at the temperature of 400±10°C, resulting to the black color CuO

NPs [**24**]. The *Sterculia foetida* aqueous leaf extract reacts with precursor solution at room temperature and reduces the copper ions and develops CuO NPs. The strong absorption peak at 340 nm observed through UV-Visible spectrophotometer (Figure 1).This proves that the copper ion is efficiently reduced by *Sterculia foetida* leaf extract.

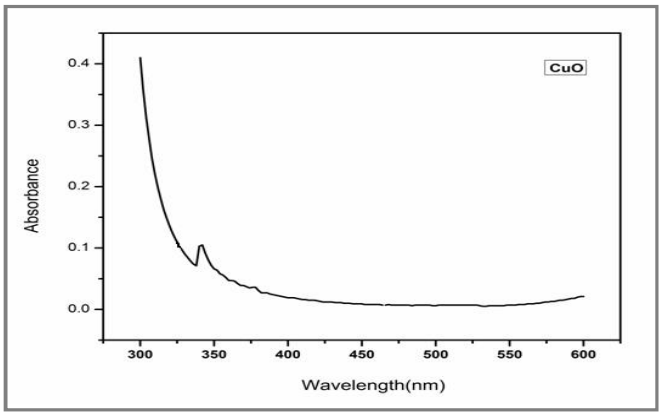


Figure 1. UV-Visible spectrum of CuO NPs using *Sterculia foetida* leaf extract.

FT-IR analysis of green synthesized CuO NPs revealed a strong band at 1100 cm^{-1}, whereas peaks at 529, 359 cm^{-1} can be attributed to vibrations of CuO NPs (Figure 2), confirming the formation of highly pure CuO NPs [**25**]. The IR Peaks were observed at various levels such as 3325 cm^{-1} due to C-H Stretching,3165 cm^{-1} bands for N-H Stretching, 2922 cm^{-1} aliphatic C-H Stretching alkanes, just below 3000 cm^{-1}, 1018 cm^{-1} -C-O Stretching, 874, 837, 808, 796, 713, 669, 517, 502 cm^{-1} vibrations M-O-M bending (M=Cu) [**26**]. The peak at 3325 cm^{-1} corresponds to O-H bond of phenols and 1654 cm^{-1} corresponds to C=O stretching vibration of primary amines.

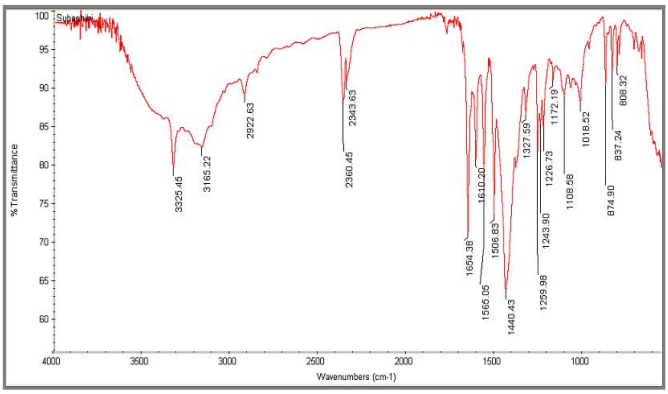


Figure 2. FT-IR of CuO NPs using *Sterculia foetida* leaf extract.

The PXRD reports of CuO NPs in (Figure 3) by solution combustion method indexed to be monoclinic crystalline structure with lattice constant a=4.6965A°, b=3.4324A° and c=5.1329A° and β=99.5287°. The Debye-Scherrer's formula used to calculate the average crystalline size of CuO NPs [**27**]. $D=k\lambda/\beta\cos\theta$ Where k is a constant equal to 0.94, λ is the wave length (1.5406A°) of X-ray radiation, β is the full-width at half maximum(FWHM) of the peak(in radians) and 2θ is the Bragg angle(degree). The average crystalline size was found to be 8-22nm.The diffraction peaks at 2θ values of 29.4, 35.5, 36.6, 39.1, 42.49, 43.53, 48.9, 61.4, 66.32, 68.1 which were assigned to (110), (110), (-110), (111), (201), (-202), (202), (-113), (311) and (004) planes respectively (with reference to JCPDS 05-0661). The EDAX report shown in (Figure 4) reveals the percentage of copper and oxygen present in the CuO nanoparticles synthesized from *Sterculia foetida* leaf extract.

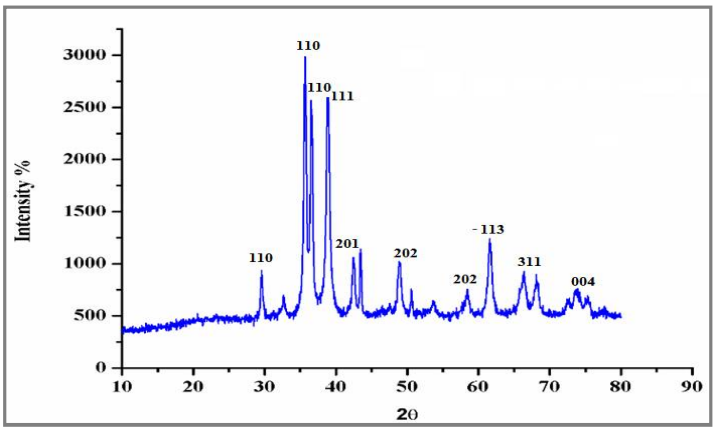


Figure 3. PXRD pattern of CuO NPs using *Sterculia foetida* leaf extract.

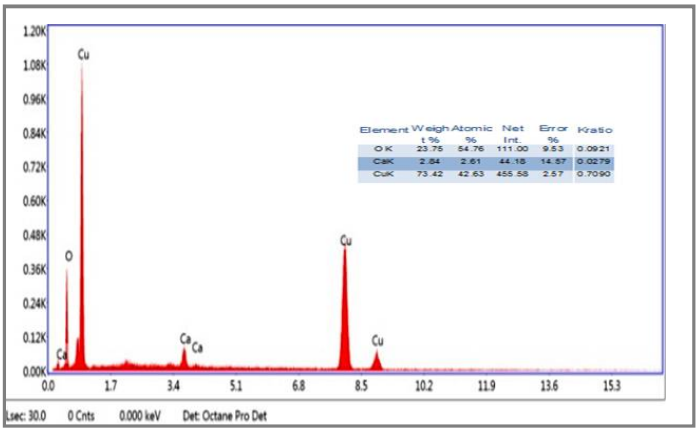


Figure 4. EDAX spectrum of CuO using*Sterculia foetida* leaf extract.

The properties of synthesized CuO NPs are based on the size and structure of the nanoparticles. The SEM image (Figure 5a) of CuO NPs reveals that the particles are almost spherical in nature with free agglomeration. The crystalline characteristics of CuO NPs observed in TEM image (Figure 5b). From the image the particles were found to be spherical in shape with the particle size range from 5-51 nm.

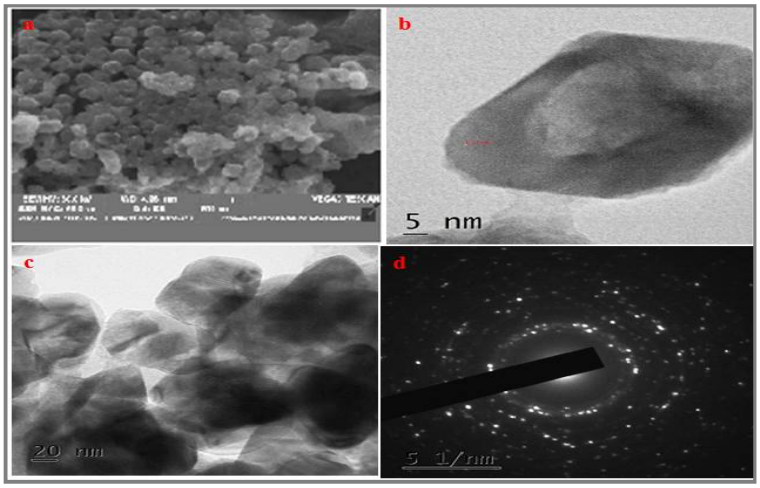


Figure 5a). SEM image, b and c). TEM images of CuO NPs *Sterculia foetida* leaf extract, d). TEM SAED of 51 nm.

Zeta Potential: The average size of the CuO nanoparticles can be measured through particle size analyser. The measurements are based on the size of the particle core, particle concentration and the type of the ion in the mixture [28]. The zeta potential of CuO nanoparticle was -13.2 mV, zeta potential between 30 mV to -30 mV indicates the stable system. The aggregation between the CuO nanoparticles prevented due to negative charge, which leads to strong repulsive force among particles. Hence CuO nanoparticles are said to be highly stable [29]. The zeta potential peak is shown in (Figure 6).

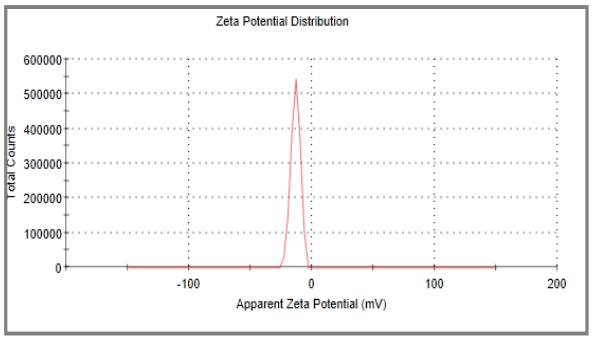


Figure 6. Zeta potential graph of CuO NPs synthesized from *Sterculia foetida* leaf extract.

APPLICATION

Antibacterial activity: The antibacterial activity of green synthesized CuO NPs were studied against *Staphylococcus aureus* (Gram +ve) and *Escherichia coli* (Gram –ve) by agar well diffusion method. The antibacterial activity checked with various concentrations (150, 300, 450 and 600 µg mL^{-1}). The more effects of zone of inhibition showed against *Staphylococcus aureus* (13 mm) and *Escherichia coli* (11 mm) due to per-oxidation. The zone of inhibition was satisfactory at higher concentrations (450-600 µg mL^{-1}) (Figure 7). The copper ions from CuO NPs attached with bacterial cell wall and rupture it, which leads to protein denaturation and cell death [30]. The CuO NPs synthesized from *Sterculia foetida* leaves were in spherical shape due to the presence of phytochemical constituents such as phenols and proteins [31]. The capping and stabilization of nanoparticles depends on the above phytochemical constituents (Table 1).

Figure 7. Antibacterial activity of CuO NPs.

Table 1. Antibacterial activity of the plant extract

Microorganisms	Concentration (µg)	Zone of Inhibition(mm)	
		Sample	Control (5 µg)
Staphylococcus aureus	150	Nil	35
	300	Nil	
	450	12	
	600	13	
Escherichia coli	150	Nil	31
	300	Nil	
	450	9	
	600	11	

CONCLUSION

The CuO NPs were successfully synthesized using *Sterculia foetida* leaf extract as a fuel by solution combustion method. The formed CuO NPs shows the UV-Spectrum peak at 340 nm. The IR vibrations from 1018 cm^{-1} to 502 cm^{-1} indicates M-O-M bending (M=Cu). The PXRD report reveals that the formed CuO NPs were in monoclinic crystalline structure. EDAX spectrum reveals the copper and oxygen weight percentage of 73.42%, 23.75% and confirms CuO NPs. The Spherical shape of the CuO NPs reported in SEM analysis. Through TEM image the overall nanoparticle size was found to be 5-51 nm. The antibacterial activity of the formed CuO Nps were carried out against *Staphylococcus* (Gram +ve), and *Escherichia coli* (Gram -ve) bacterial strains through agar well diffusion method. The zeta potential value of CuO nanoparticles observed was -13.2 mV. From the above reports the applicability of CuO NPs as reducing, stabilizing agents has been proved.

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