ISSN (Online) 2456 -1304

International Journal of Science, Engineering and Management (IJSEM)
Vol 6, Issue 5, May 2021

Sugarcane Juice Stabilized Biosynthesis of Copper Oxide Nanoparticles and Their Antibacterial Activity

[1] A.P. Angeline Mary
[1] Department of chemistry, Sacred Heart College, Tirupattur, Tamilnadu,India

Abstract: **- In this present work, sugarcane juice stabilized copper oxide nanoparticles was synthesized and characterized by various spectral techniques. The FTIR spectrum confirms the transformation of functional groups through Cu-O. The XRD analysis confirms the monoclinic crystalline structure. The SEM-EDAX confirm the nanoparticles formation and elemental compositions. Various fascinating shapes of CuO NPs obtained from the TEM images. The results revealed that the sugarcane is a good stabilizing agent which reduces the size of particles significantly at higher concentrations and altered shapes to spherical. Antibacterial activity of CuO NPs was assessed against some pathogenic bacteria.**

Key Words: Sugarcane juice, biosynthesis, CuO nanoparticles, antibacterial activity

1. INTRODUCTION

Nanomaterials are entirely differing from the bulk materials in terms of properties [1]. Metal oxide nanoparticles drawn the attention of researchers due to its wide applications in various fields. The metal oxide nanoparticles have been proved as disinfectant in the medical field, antimicrobial agent, fillers in cosmetics and as catalysts in industrial field [2]. Outstanding characteristics such as thermal stability, optical properties and electrical conductivity of copper oxide inspired the researcher to work on this material for photocatalytic activity and electrical conductivity [3]. The CuO NPs also used in antimicrobial devices. There is a relationship between the efficiency and particle size of the CuO. The morphological aspects and structure of the CuO NPs found to be improved by applying various techniques [4].

CuO has drawn the curiosity of researchers due to its biological properties. Antimicrobial and biocidal properties, which led utilize in the biomedical applications [5]. Electrical conducting, optical and electrical properties of CuO find its applications as supercapacitors, infrared filters, storage devices and sensors [6]. It is interesting to know that the size of the particles playing key role in the constructing optical, catalytic, electrical and biological properties [7]. Apart from the cosmetic and pharmacological applications, CuO NPs also used as paints and coatings [8]. Sugarcane juice is a rich source of sucrose, mineral and other biomolecules. It contains chloride, calcium and sodium ion. This juice is also well known for the presence of food supplements such as malic acid, oxalic acid, citric acid D-gluconic acid. Therefore, in this work, it is proposed to utilize sugarcane juice as stabilizing agent for the synthesis of CuO NPs.

2. Materials and methods

2.1. CuO NPs synthesis

Fresh sugarcane juice was obtained from the local vendor and the purified juice was used as stabilizing agent for the synthesis the copper oxide nanoparticles. Copper nitrate was purchased from LobaChemie, Mumbai, India.

To synthesize CuO NPs, 5 ml of juice was mixed with 50 ml of 0.1 M copper nitrate solution kept on a magnetic stirrer and stirred at 80°C for an hour. The blue color of reaction mixture turned to green by revealing the formation copper hydroxide precipitate was obtained by adding ammonium hydroxide as reducing agent. The precipitate was carefully removed from the beaker and thoroughly washed with water, followed by acetone and alcohol to remove the organic impurities. Then the precipitate was dried in air at 80°C for 8 h followed by the annealing at 500°C for 3 h in a muffle furnace.

2.3. Characterization methods

Functional groups and crystalline phase were characterized by fourier trnasfom spectrophotometer (Jasco 6300 FT-IR spectrophotometer) and X-ray diffractometer (Model: X'Pert-PRO) with Cu–Ka radiation. Formation of the CuO NPs was

ISSN (Online) 2456 -1304

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identified using scanning electron microscope with the resolution (JEOL Model JSM - 6390LV). Morphology of particles was studied using transmission electron microscope (TEM model: Jeol/JEM 2100) with resolution. Elemental composition of sample was analyzed by energy dispersive study (EDS).

2.4. Antibacterial activity

Antibacterial activity of CuO NPs was studied against *Escherichia coli, Pseudomonas aeruginosa, Staphylooccus aureus* and *bacillus subtilis.* Bacterial culture (Mueller Hinton agar) 20 ml was transferred into Petri plates. The wells were punched into the agar plate. The bacteria culture was swabbed on the agar plates. CuO NPs, 100 mg/ml dispersed in DMSO were weighed down into the each well. The petri plates were incubated at 37°C. The zone of inhibition was noted after 24 h of incubation and expressed in mm dia and compared to the reference substance (Ciprofloxacin). The tests were performed in triplicates. 25 µg of Ciprofloxacin and 100 µg of CuO NPs were used to test the bactericidal activity.

3. Results and Discussion

The functional group was carried out using FTIR study. The CuO NPs synthesized using sugarcane juice is shown in Figure 1. A peak observed at 609 cm^{-1} due to stretching vibrations of Cu-O bond in CuO NPs. Two peaks shown at 799 and 855 cm^{-1} are revealed the M-O stretching vibrations of CuO NPs [9, 10]. The absorption peaks observed at 1635 and 3346 cm^{-1} contribute to the bending and stretching vibrations of moisture content present on the surface of CuO NPs. The peaks noticed near 500 to 600 cm^{-1} indicate the metal–oxygen vibration frequencies of copper and oxygen [11, 12].

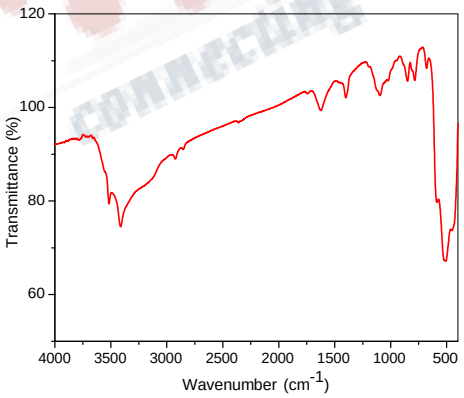

Figure 1. FTIR spectrum of CuO NPs synthesized using 2 ml sugarcane juice

The XRD spectra of the CuO NPs are shown in Figure 2. The XRD report revealed a sequence of diffraction peaks at *2θ* of X-axis at 32.11°, 35.62°, 38.87°, 48.85°, 53.49°, 58.46°, 61.71° and 66.5° matching for the (110), (111), (200), (-202), (020), (202), (-113) and (022) planes, correspondingly. The XRD spectrum proved that the CuO NPs are in the crystalline phase and it was compared with a standard XRD data card (80–1916) [11]. The average crystallite size of CuO NPs synthesized using 5 was found to be 23.61 nm.

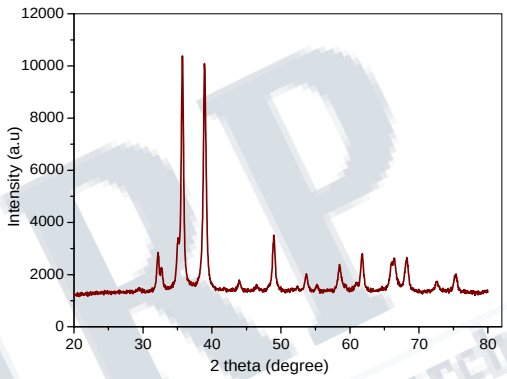

Figure 2. XRD spectrum of CuO NPs synthesized using 2 ml juice

Figure 3 shows the SEM images of Chou NPs synthesized using 5 ml sugar cane juice. SEM images exhibit lengthy particles with irregular shapes. The percentage of Cu and O confirmed by energy dispersive (EDS) study as shown in Figure 4. TEM mages of CuO NPs synthesized using sugarcane juice is shown in Figure 5. It exhibited spherical, square, cube, plate, rectangular with some irregular shapes when 2 ml of juice was used.

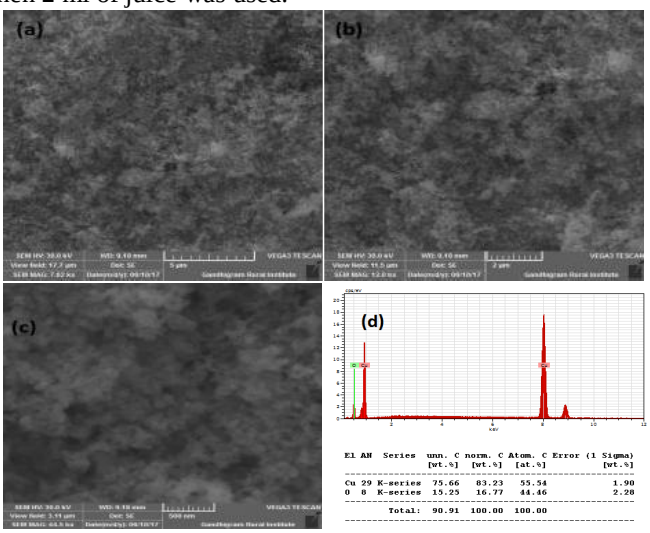

Figure 3. SEM images and EDS of CuO NPs with 2 ml juice

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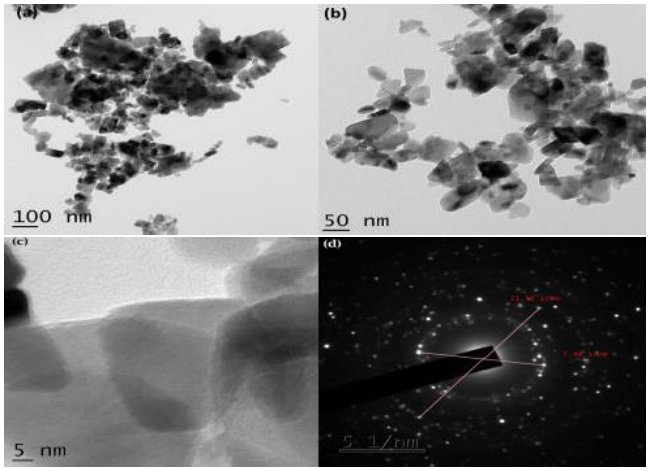

Figure 4. (a-d). TEM images and SAED pattern of CuO with 2 ml juice

The CuO NPs, 100 µg and 25 µg of ciprofloxacin were used to test the antibacterial activity. The zone of inhibition shown by CuO NPs synthesized using 2 juice against *Escherichia coli, Pseudomonas aeruginosa, Staphylooccus aureus* and *bacillus subtilis* was 5, 8 and 9 mm as shown in Figure 6. Various mechanisms have been widely proposed for the antibacterial activity of the nanoparticles. Penetration of smallest particles into the cell walls of the bacteria and binding with the nucleic acids of bacteria also causes severe toxic [13]. It is known that sugarcane juice contains 81-87 % and 3-6 % of reducing sugars and remaining constituents include minerals and amino acids [14].

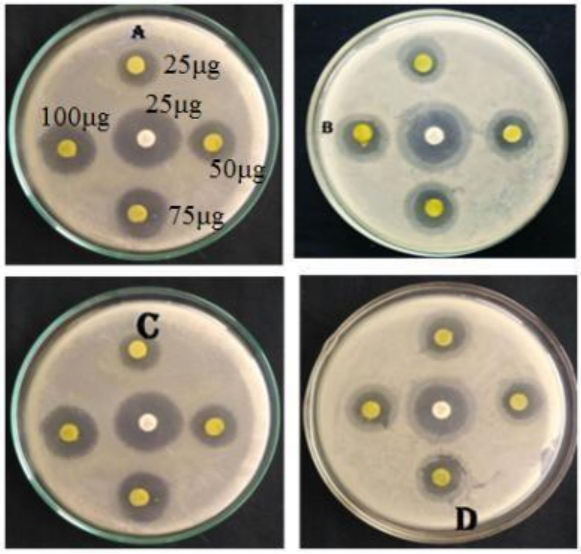

Figure 7(A-D). Antibacterial activity of CuO NPs: Zone of inhibition of (A) *Escherichia coli (B) Pseudomonas aeruginosa s(C) Staphylooccus aureus* and *(D) bacillus subtilis*

4. Conclusions

The present study report the eco-friendly and convenient greener route synthesis of Cu nanoparticles using sugar cane juice as a stabilizing agent. FTIR and XRD pattern confirmed the formation of CuO NPs and their crystalline structure. The SEM images confirmed formation of particles in nanoscale. The TEM images confirmed the spherical, square, cubic and rectangular shaped nanoparticles at 2 ml of sugarcane juice as a stabilizing agent. This work proved that the sugarcane is a suitable green stabilizing agent to produce copper oxide nanoparticles. Synthesized CuO NPs exhibit significant bactericidal activity against *Escherichia coli, Streptococcus aureus, and Pseudomonas aeruginosa* and *bacillus subtilis.*

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