BIOINSPIRED GREEN SYNTHESIS AND CHARACTERISATIONS OF COPPER OXIDE NANOPARTICLES USING ARECA CATECHU LEAVES FOR ANTIMICROBIAL ACTIVITY

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ABSTRACT

Copper Oxide nanoparticles were synthesized from Copper Sulphate and Areca Catechu leaves by using green synthesis method. Utilizing X-ray diffraction, the crystalline characterizations of synthesized nanoparticles were investigated. The optical properties pertaining to bio inspired green copper oxide synthesized nanoparticles were examined, and the energy band gap was also estimated. Functional group of CuO nanoparticles were confirmed using FTIR analysis. By Scanning Electron Microscope (SEM), the structural form of copper oxide nanoparticles made by green synthesis was evaluated. Green synthesized CuO NPs have been shown to have antimicrobial activity against gram negative bacteria.

Keywords: Areca Catechu, Copper oxide nanoparticles, Antimicrobial, Energy band gap, Scanning Electron Microscope (SEM)

INTRODUCTION

For their prospective use in optoelectronics, tiny devices, nanoelectronics, nanosensors, storage of information, and catalysis, metal oxide nanoparticles (NPs) have received significant interest. Among the various metal oxide nanoparticles, CuO, the most fundamental copper compound, has drawn the greatest interest because it exhibits a variety of beneficial features, including extreme temperatures superconductivity, interaction of electrons effects, and spin dynamics. An oxide of a transition metal is cupric oxide (CuO). It is made of p-type semiconductor material and features a monoclinic structure with a small band gap. Due to its small band gap energy, it is also a desirable option for light-harvesting applications[1,2,3]. CuO compounds are extensively used as catalysts, Photovoltaic materials, electronic materials, Photonic devices, Accumulators, and junction devices like p-n diodes. They can also be used for photoconductive, photothermal, and photoelectrochemical processes[4,5,6,7,8,]. CuO NPs are being employed more frequently in a variety of uses, including, catalysis, solar energy, and gas sensors. It is a severe problem in the field of health care when different types of bacteria cause microbial contamination of the air, water, and soil. New antimicrobial copounds such tiny cationic polymers, antimicrobial peptides, and metal nanoparticles are becoming more popular as a result of the rise in antibiotic-resistant illnesses. In this research CuO nanoparticles were synthesized using Areca Catechu leaves and structural, functional, optical, scanning Electron Microscope (SEM) and Antimicrobial activity characterizations of synthesized nanoparticles were also studied.

MATERIALS AND METHOD

Preparation of Leaves Extract

Areca Catechu leaves of 50 g was collected at Mullipaadi in Dindigul district. The collected leaves are washed multiple times and then dried at room temperature. To collect leaves extract, the dried leaves were boiled at 60^0C using 100ml of double distilled water for 30 minutes. The colour of the water altered to pale green. It was then filtered using whatmann filter paper. At room temperature, the filtered leaf extract was allowed to cool and was stored in refrigerator for further experimental purpose.

Synthesis of CuO nanoparticles

Copper sulphate (Sigma Aldrich >98% gray-white) a highly soluble in water and plant extract (Areca Catechu) 0.1 M were taken in a double distilled water. Copper sulphate and Plant extract were taken in ratios of 1:1, 1:2, 3:2 and 4:1. In these different ratios concentration 1:1 ratio concentration was selected for the bulk preparation because large production was attained in this ratio than other ratios. The solution was mixed well using magnetic stirrer by rotating the pellet 800 rpm and temperature was maintained below its boiling point. The mixture turned into light green colour within 1 hr. Bulk production of synthesized CuO nanoparticle using $CuSO_4$ and Plant extract (Areca Catechu) is shown in fig.1. Twenty minutes were spent centrifuging the acquired suspension at 1,000 rpm. To get rid of contaminants, deionized water was used to wash Copper Oxide nanoparticles three to four times. The nanoparticles that had precipitated were lyophilized. For additional characterizations, lyophilized nanoparticles were kept in a cool, dry, and dark location. Synthesized CuO nanoparticles before and after lyophilization is shown in fig. 2a and 2b.

Fig. 1. Bulk production of synthesized Cu nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu).

2a)

2b)

Fig.2 a,b. Synthesized CuO nanoparticles 2a)before lyophilization 2b)after lyophilization

RESULTS AND DISCUSSION

X- Ray Diffraction (XRD)

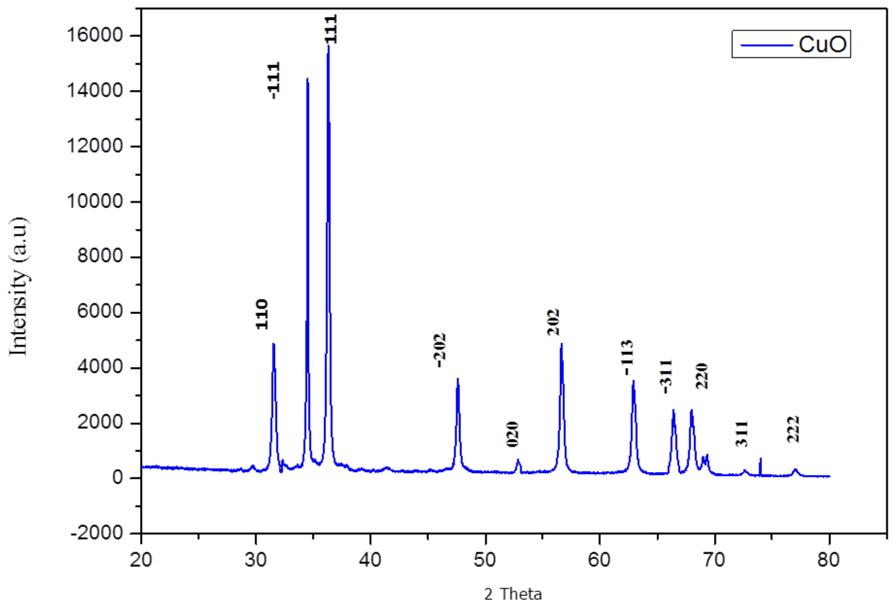

Fig. 3. X- Ray Spectra of green synthesized CuO nanoparticles using $CuSO_4$ and Plant extracts (Areca Catechu).

XPERT-PRO diffractometer utilized Cu-Kα radiation (λ=1.54 Å) to record the XRD spectra of CuO NPs. As seen in Fig. 3, the orderly arranged planes of CuO NPs is compatible with respect to JCPDS card no. 05-0661 and it supports the green synthesized materials nano crystalline character. It is understood from the XRD spectra that the sharply detected planes (110), (-111), (111), (-202), (020), (202), (-113), (-311), (220), (311), and (222)[9] contribute to the monoclinic structure of CuO NPs. The crystallite size is calculated to be 30 nm.

FTIR (Fourier Transform Infrared Spectroscopy) Analysis

FTIR spectral measurements were made between 400 and 4000 cm^{-1}. A Jasco 460 PLUS FTIR spectrometer was used to measure the FTIR spectra using the KBr pellet method at room temperature. FTIR analysis of synthesized CuO nanoparticles using $CuSO_4$ and Plant extracts (Areca Catechu) are shown in fig.4. Based on the peak value in the IR region, the functional groups of the active components were identified using FTIR Spectroscopy. The bands that are visible are lattice vibrational modes that show the functional groups of biomolecules adsorbed on artificial nanoparticles. Vibrational frequency is observed at 3430cm^{-1} due to symmetric and asymmetric stretching.

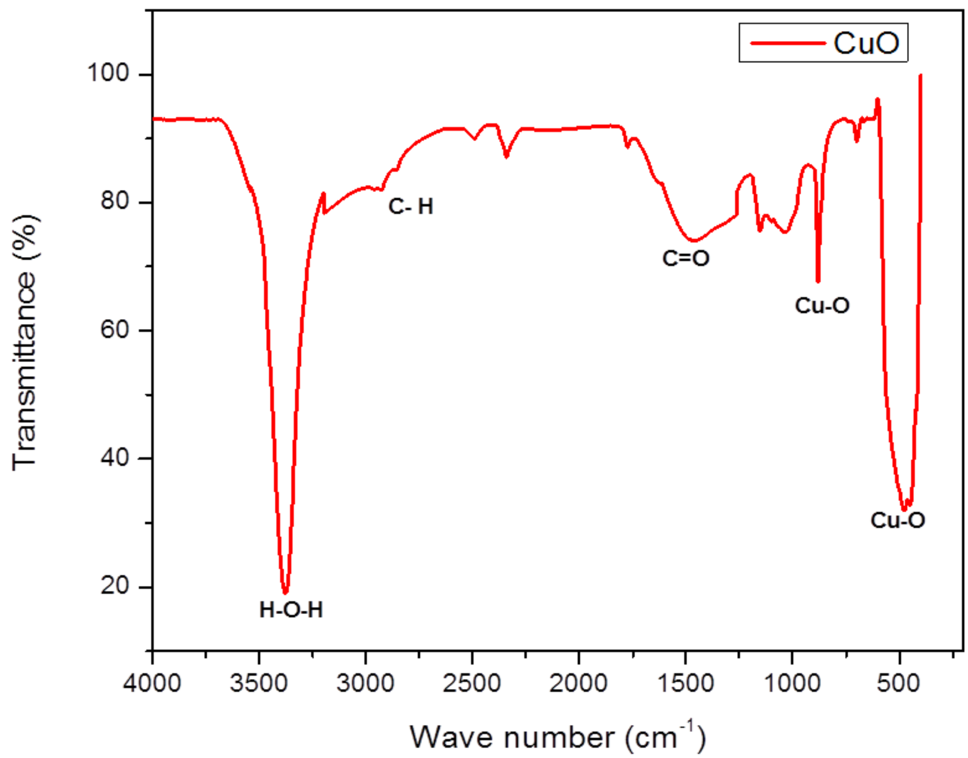

Fig. 4. FTIR spectra of synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu)

The peak in between 2000 cm^{-1} to 3000 cm^{-1} is attributed for C-H stretching due to alkanes and alkyls. The strong C = C noted around 1600 cm^{-1} is for Alkenes. A moderate vibrational frequency (= C- H bend) is designed at 932cm^{-1}. A strong vibrational frequency obtained around 584cm^{-1} is due to CuO vibration[11].

Analysis of Optical Properties

The optical properties of CuO nanoparticles were studied from 190-1100 nm with 2 nm spectral resolution using UV-Vis-NIR spectrometer. Optical properties are very essential for any NLO materials. If they have a wide transparency window, synthesized materials can be used in NLO applications. Optical properties of CuO were studied and it is shown in fig.5 and 6. From the above graph we can understand the cut off wavelength of CuO is 225.2 nm. Direct and indirect energy band gap of CuO were also calculated and values are 3.16 eV and 2.15 eV respectively. Since the green synthezied CuO Nps has wide transparency window, it can be used for nonlinear optical applications [10].

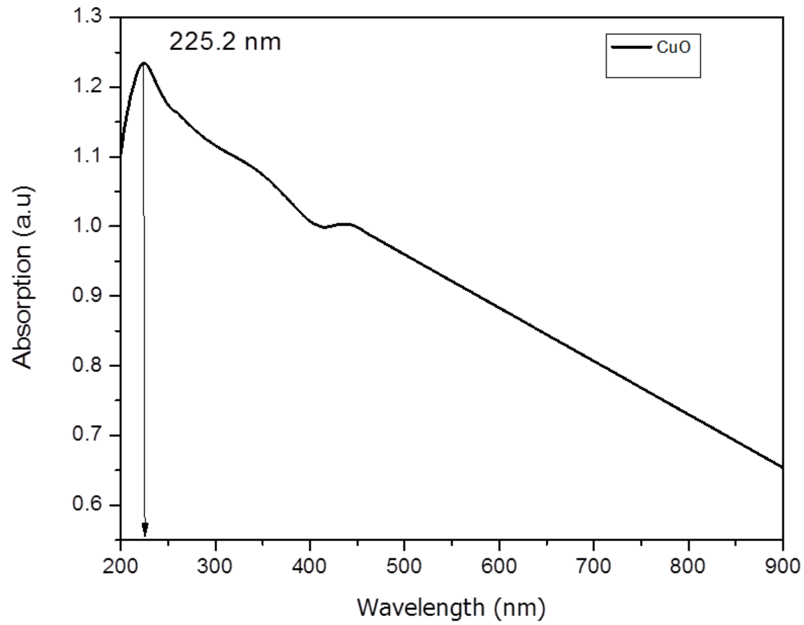

Fig. 5. Absorption spectra of synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu)

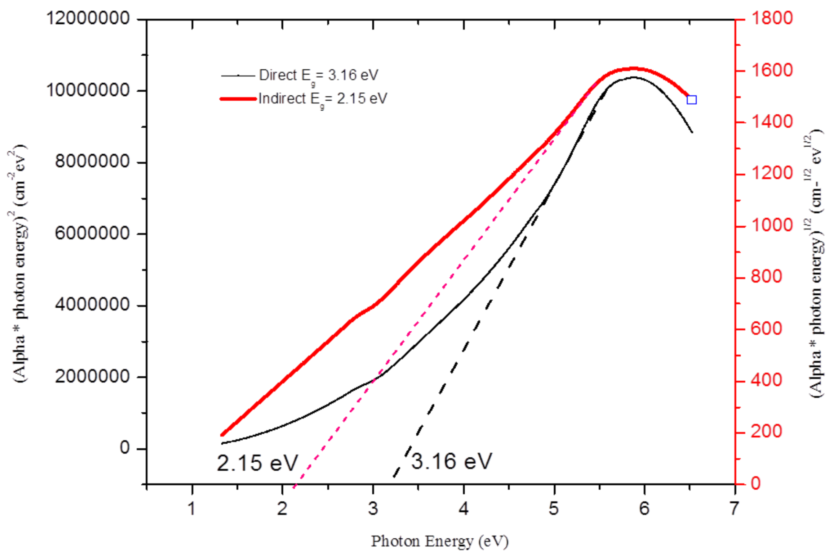

Fig. 6. Direct and indirect band gap of green synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu)

SCANNING ELECTRON MICROSCOPE (SEM)

The surface morphology of green synthesized Copper Oxide nano particles using $CuSO_4$ and Plant extracts (Areca Catechu) was analyzed using SEM. Fig.7. shows the surface morphology of the CuO NPs. SEM image shows the Copper nano particles are gravel like structure.

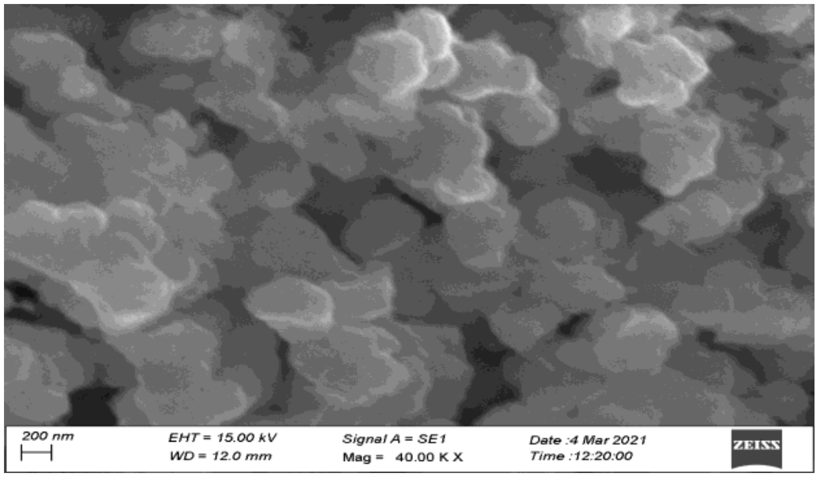

Fig. 7. SEM image of green synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu)

Antimicrobial Analyzes

Antibacterial analysis hinders the growth of bacteria by preventing the formation of microbial colonies. Bacteria are of two types, gram positive and gram negative. We chose gram *Streptococcus oralis* and *Pseudomonas aeruginosa (*positive and negative bacteria) for the study.

In order to check the antibacterial analysis, well diffusion technique was implemented for the samples on the microorganisms. In order to prepare nutrient medium in 150 ml double distilled water, 5.3 g of agar (Muller Hinton) was dissolved and retained at 121°C for 20-25 minutes. The nutrient medium was allowed to cool up to 70^{o}C and then was made to settle in gel form in the petri dishes. Wells of approximately 2.5 mm diameter were made in solidified medium and *Streptococcus oralis* and *Pseudomonas aeruginosa* were sponged in their respective petri dishes. The concentration of CuO NPs (500, 250, 100 and 50 µg/ml) were infused into the wells and it was incubated at 310K for about one day. The size of inhibition zone was determined using Gentamicin as reference. The antibacterial activity of *Streptococcus oralis and Pseudomonas aeruginosa* is shown in Fig.8 and 9. The areas of inhibition of CuO NPs tested against respective bacteria *Streptococcus oralis* and *Pseudomonas aeruginosa* is listed in Table.1. The CuO nanoparticles of concentration 500 µg/ml inhibit the growth of Pseudomonas auroginosa compared to other concentrations. Increased concentration eventually increases the production of Reactive Oxygen Species , which is what causes the antibacterial behaviour of the expanding zone of inhibition [11, 12].

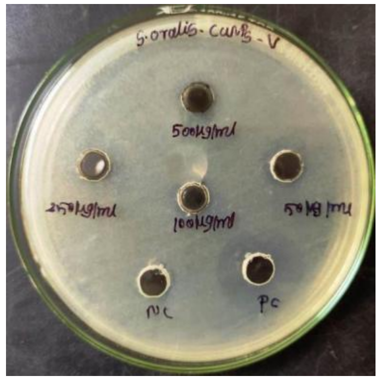

Fig.8. Effect of green synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu) against *Streptococcus oralis*.

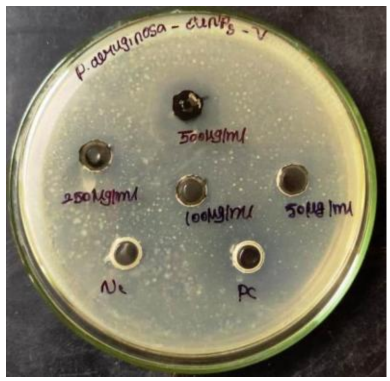

Fig.9. Effect of green synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu) against *Pseudomonas aeruginosa*.

Table.1 The zone of inhibition of green synthesized CuO nanoparticle using $CuSO_4$ and Plant extracts (Areca Catechu) tested against respective bacteria *Streptococcus oralis and Pseudomonas aeruginosa*

S.NO	**Bacteria**	**Sample**	**Inhibition Region (mm)** SD ± Mean				
			500 µg/ml	**250 µg/ml**	**100 µg/ml**	**50 µg/ml**	**PC**
1	*Streptococcus oralis*	CuNPs-V	0	0	0	0	9.5± 0. 7
2	*Pseudomonas aeruginosa*		10.5± 0.7	0	0	0	10.25± 0.35

CONCLUSION

Green synthesized method was used to prepare Copper Oxide nanoparticles from Copper sulphate and Areca Catechu leaves. X-ray diffraction spectra were taken and the structural characterizations study validated the crystalline structure of the synthesized CuO nanoparticles. Functional group of CuO nanoparticles were confirmed using FTIR analysis. Optical property of of bioinspired copper nanoparticles was studied using UV absorption and energy band gap was also calculated. The surface morphology of the green synthesis of copper nanoparticles was determined and they are pebble like structure. Antimicrobial study of green synthesized CuO NPs is effective against gram negative bacteria *Pseudomonas aeruginosa*. The bioinspired copper nanoparticles can be used in nonlinear optical and antimicrobial applications.

CONFLICT OF INTERESTS

The authors declare that there is no conflict of interest.

All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

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