International Journal of Zoology and Applied Biosciences **ISSN: 2455-9571**
Volume 7, Issue 1, pp: 17-22, 2022
https://doi.org/10.55126/ijzab.2022.v07.i01.004
http://www.ijzab.com

Research Article

ANTIBACTIRIAL ACTIVITY OF COPPER OXIDE NANO PARTICLES AGAINST GRAM POSITIVE AND NEGATIVE BACTERIAL STRAIN SYNTHESIZED BY PRECIPITATION TECHNIQUE

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Article History: Received 11th January 2022; Accepted 25th January 2022; Published 30th January 2022

ABSTRACT

This work was carried out to investigate the antibacterial activity of CuO nanoparticles (CuONPs), which were produced by synthetic methods of the precipitation technique of the precursor material copper acetate and sodium hydroxide as reducing agents. The purity of the nanoparticles was confirmed by various characterizations. Techniques including X-ray diffraction, Scanning Electron Microscopy (SEM), Ultraviolet (UV), EDAX. The antibacterial activity of nanoparticles was examined with various microorganisms from *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Klebsiella pneumonia.*

Keywords: XRD, SEM, UV, EDAX, Antibacterial studies, CuO-Copper Oxide.

INTRODUCTION

Metal oxide (NP) nanoparticles have gotten a lot of press because of their prospective uses in optoelectronics, nanodevices, nanoelectronics, nanosensors, data storage, and catalysis. CuO has established itself as an excellent catalyst in organic reactions among the various metal oxide NPs (Azam *et al*., 2012; El Trass *et al*., 2012). CuO has recently been used in gas sensors with high TC, superconducting solar cells, emitters, and electronic cathode materials. Solar energy and transfer fluids (Azam *et al*., 2012; Marabelli *et al*., 1995; Ren *et al*., 2009). CuO NPs have antimicrobial properties. CuO NPs were made using a simple precipitation method. X-ray diffraction (XRD), scanning electron microscopy with field emission (SEM), and X-ray energy dispersive spectroscopy were used to investigate the structural features of CuO NPs (EDS). CuO NP disc diffusion was used to test the antibacterial activity of CuO NP against diverse bacterial strains (*Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylo coccusaureus*, *Klebsiella pneumonia*).

MATERIALS AND METHODS

CuO NPs were made utilising an aqueous precipitation approach using copper (II) acetate as the precursor and sodium hydroxide (NaOH) as the reducing agent. In a round-bottomed flask, 0.2M copper (II) acetate solution (300mL) and glacial acetic acid (CH3COOH) (3mL) were combined and heated to boiling with magnetic stirring. The flask was then filled with 15mL of 6M NaOH solution. The solution's hue changed from blue to black very instantly, and a black suspension formed at the same time. The reaction took two hours to complete with constant stirring and boiling. The mixture was centrifuged after cooling to room temperature. The result was a moist CuO precipitate. The precipitates were filtered and washed multiple times with distilled water and 100% ethanol.

Characterization

XRD and SEM analysis methods were used to characterise the structure, morphology, and elemental composition of the ready toughened samples. Victimization EQUNIOX 3000, part meter within the scanning range of 20 - 80 (2),

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employing Cu Karadiations of wavelength 1.5406 was used in the crystallographic research. The morphology and size of NPs were studied using a scanning microscope (model XL30, Philips, Eindhoven). The strength of NPs' absorption peaks was measured using a UV-Vis photometer from 200 to 900nm. The fundamental composition of produced CuO NPs was determined using EDX.

Antibacterial activity

The glass articles are sterilized in an autoclave (121° C for 15 minutes) for antibacterial activity testing after sterilization. After sterilization, the procedure for making plates, as well as using Mullerhiton agar, is followed, followed by the pouring plate technique. This method employs the expanded plate technique (*Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumonia*). Borehole diffusion is used to study antimicrobial action, and the borehole is made with a borehole cutter. According to their concentration, the samples were fed into the wells. (Litres: 100, 80, and 60).

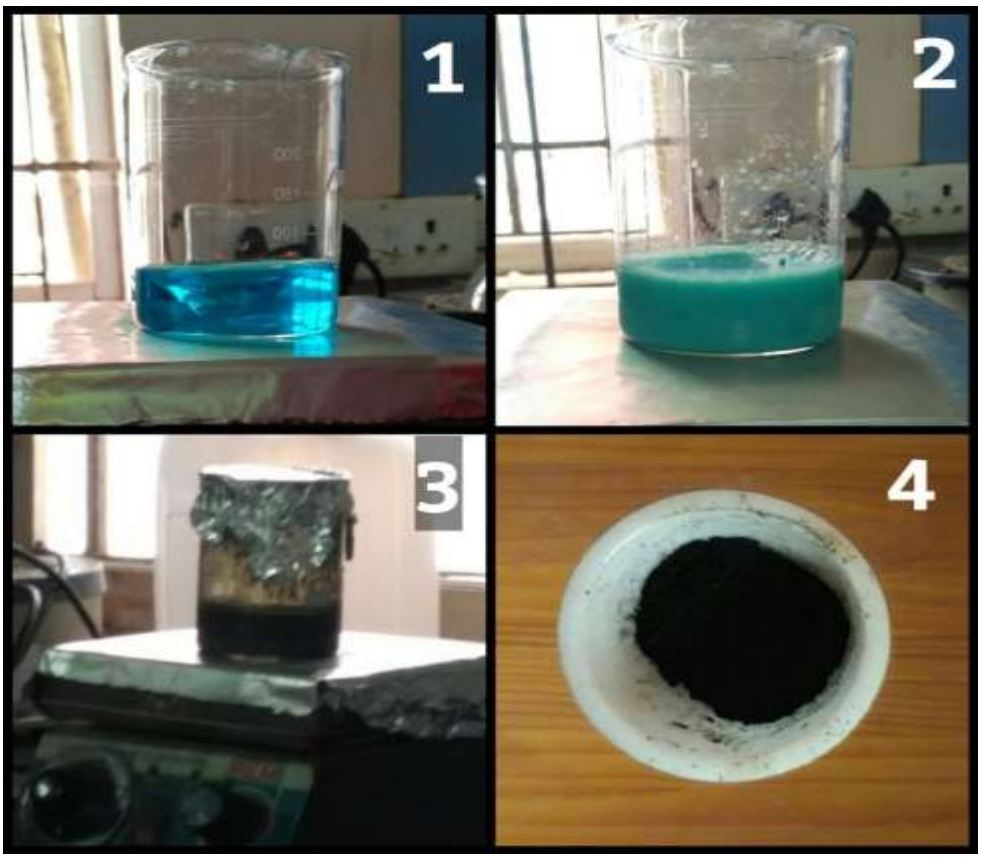

Figure 1. Synthesis methods of CuO NPs.

Antibacterial activity of the synthesized CuO NPs

Figure 1 shows the antibacterial efficacy of CuO-NPs against human bacterial pathogens: *Staphylococcus aureus* (G+), *Pseudomonas aeruginosa* (G-), and *Klebsiella pneumoniae* (G+). The positive control was gentamicin, the negative control was CuSO4sollution (10 mM), and the blank was distilled water. CuSO4 in a specific concentration exhibits no activity against the selected bacterial pathogens, as evidenced by the absence of an inhibition zone in the control. Biosynthesized CuONPs made *Sareus* and *Pesudomonas* extremely sensitive (20 mm). Furthermore, at a concentration of 100 g / mL, biosynthesized CuO NPS suppressed bacterial infections (Figure 2). CuO NPs are extremely reactive due to their enormous surface area. Copper nanoparticles' surface-to-volume ratio allows them to connect with bacteria's cell membrane via their surface, resulting in bacterial death (Usman *et al*., 2013). Copper nanoparticles also have an effect on bacteria's biological mechanisms. Inhibition of cell growth, resulting in bactericidal activity. Copper Nano sol produced from *Ocimum sanctum* plant leaf extract has been shown to have antibacterial action (18 mm) against *S. aureus* (Patel *et al*., 2016). CuNPs and ZnNPs have been found to have substantial anti-fish action. Bacterial pathogens are bacteria that cause disease (Muralisankar *et al*., 2015) Few studies on the antibacterial activity of actinomycetes-derived nanoparticles against bacterial pathogens have been published (Abdelnaby *et al*., 2021; Chauhan *et al*., 2015). Table 1 shows the antibacterial activity of CuO NPs produced utilizing various biological sources in various microorganisms (Figure 2).

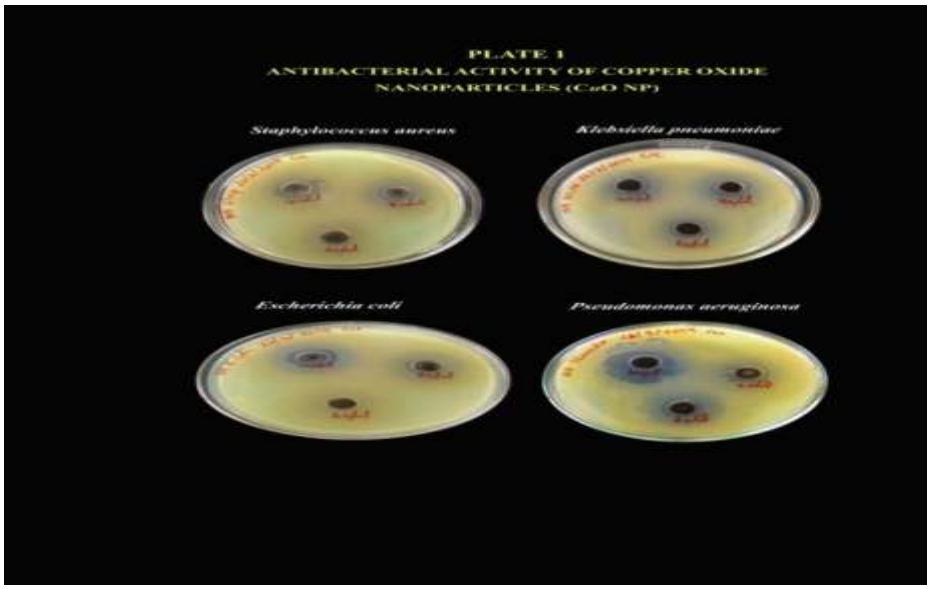

Figure 2. Antimicrobial activity of CuO NPs.

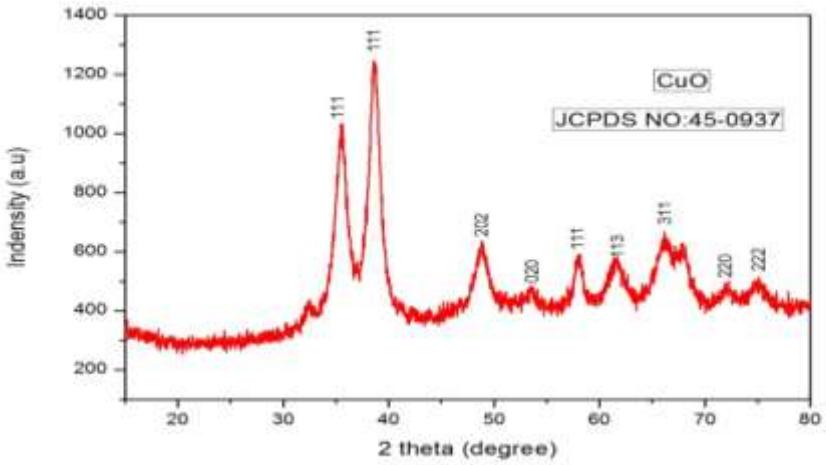

Figure 3. XRD pattern of CuO NPs.

RESULT AND DISCUSSION

The powder X-ray diffraction of pure CuO nanoparticles is performed using automated powder X-ray diffract meter. Operating at wavelength 0.15405 nm. From the XRD pattern the particle size D be calculated using scherrer formula

$$D = K\lambda / \beta \cos \theta,$$

Where K is a shape factor, λ is the wavelength of the incident X-ray and ɵ is diffraction angle and β is the Full width half maximum. From the value of the particle size (D). From the figure 6.1 it is shown that sharp peaks are observed, which shows the crystalline perfection of CuO nanoparticles. The peaks of the diffraction patterns can be compared with standard available data for the confirmation of the structure, with the use of JCPDS Card no; 45-0937. The XRD patterns agree well with the monoclinic phase of the CuO and it is confirmed that both pure CuO nanoparticles are of monoclinic crystalline structure. The particle size 10.92.

Tauc's plots are drawn between the worths of gauge boson energy hν and (αhν)2 are shown in figure 6.2 From the Tauc's plot, the calculated band gap energies of pure CuOs are 3eV respectively. The band gap value of bulk CuO is 3eV. However the band gap values of CuO nanoparticles are more than that of Bulk CuO. It's thanks to the quantum confinement result of those nanostructures. It may beCuO nanoparticles can be activated by the visible light (Rejith & Krishnan, 2012; Rejitha & Krishnan, 2013). The surface morphology of the synthesized nanoparticles is analyzed by scanning electron microscopy analysis. Figure 6.3 shows the scanning electron microscope image of synthesized pure CuO nanoparticles, which was scanned with a 20KV electron beam at 10,000 times magnification with the SEM HITACHI S3400 instrument. The morphology of the CuO nanoparticles is changed to ice rock. The elements present in the synthesized nanoparticles are analyzed by energy-dispersive X-ray analysis EDAX. It is usually taken in conjunction with an SEM analysis. The EDAX spectrum not only identifies the elements that correspond to each of its peaks, but also the type of X-ray radiation that it corresponds well to. The higher a peak is in a spectrum, the more concentrated the element is in the spectrum. Figure 6.4 shows the EDAX spectrum of the nanoparticle-like structure of pure CuO(Azam *et al*., 2012; Rejith & Krishnan, 2012).

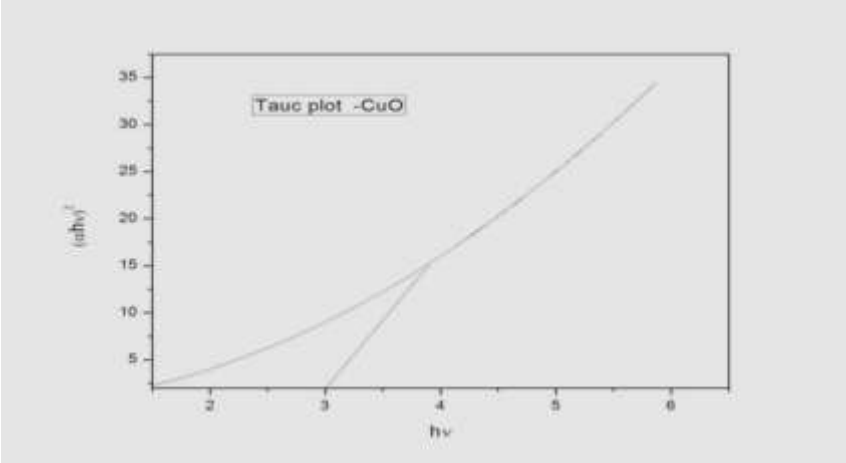

Figure 4. UV- patterns of CuO NPs.

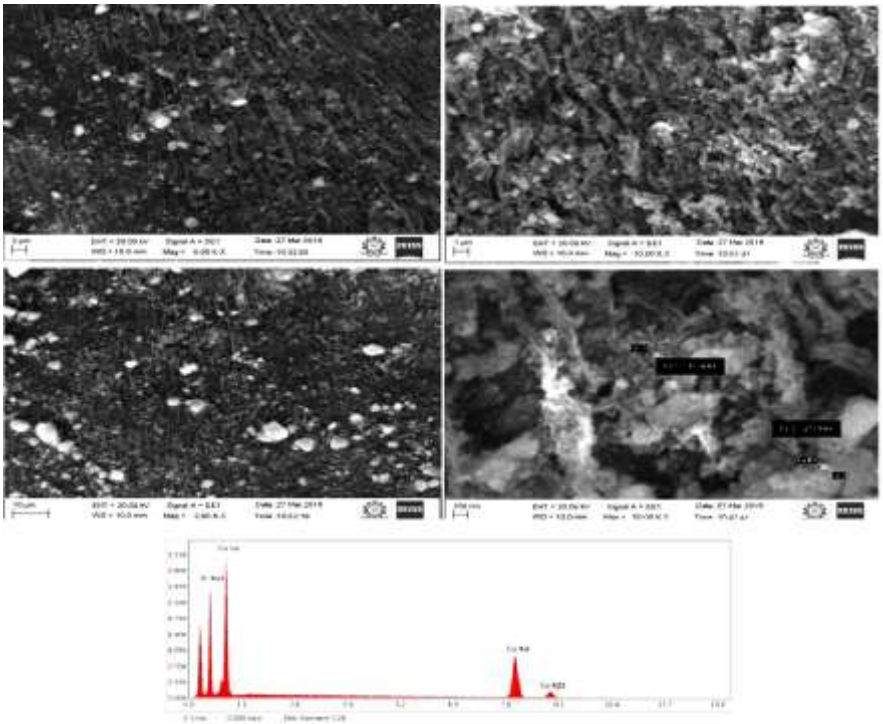

Figure 4. SEM & EDAX Analysis of CuONPs**.**

Table 1. Antibacterial effects of various extract of *Girardinia diversifolia*(link) friis and Vitamin C.

S. No	Tested organisms	Positive control (mm)	Negative control (mm)	Concentration of Nanosolution (µl)	Inhibition zones (mm)
1	*Escherichia coli*	15	-	100	18 ± 0.97
			-	80	15 ± 0.53
			-	60	12 ± 0.26
2	*Pseudomonas aeruginosa*	12	-	100	20 ± 0.33
			-	80	18 ± 0.12
			-	60	17 ± 0.51
3	*Staphylococcus aureus*	12	-	100	20 ± 0.11
			-	80	18 ± 0.24
			-	60	15 ± 0.31
4	*Klebsiella pneumonia*	18	-	100	19 ± 0.13
			-	80	18 ± 0.21
			-	60	15 ± 0.67

Excellent antimicrobial interesting opposition to various bacteria. The diameter of inhibition area displays value of susceptibility of microbes, the traces susceptibility of microbes. The diameter of inhibition area displays value of susceptibility of microbes. The traces prone toCuO NPs exhibited large area of inhibition, while resistant traces showcase smaller area of inhibition. According to area of inhibition *Pseudomonas aeruginosa*, *Staphylococcus aureus* exhibited the very best sensitivity towards CuO NPs even as *E. coli*, *K. pneumonia* confirmed the least sensitivity some of the examined microbes.

CONCLUSION

CuO NPs was prepared by a simple precipitation method**.** The XRD patterns agree well with the monoclinic phase of the CuO and it is confirmed that both pure CuO nanoparticles are of monoclinic crystalline structure. The surface morphology of the synthesized nanoparticles is analyzed by scanning electron microscopy analysis. Antibacterial activity area of inhibition *Pseudomonas aeruginosa*, *Staphylococcus aureus* exhibited the very best sensitivity towards CuO NPs even as *E. coli*, *K. pneumonia* confirmed the least sensitivity some of the examined microbes.

ACKNOWLEDGMENT

The authors are very grateful for the constant support provided by Rev. Dr. S. Mariadoss S.J, the principal of St. Xavier's College, Palayamkottai, and Rev. Dr. Alphonse Manickam S.J, the secretary of St. Xavier's College, Palayamkottai.

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