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RESEARCH PAPER

Copper Nanoparticles Against Two Types of Bacteria Staphylococcus Aureus and Escherichia Coli

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ABSTRACT

Nanotechnology is a young scientific field that holds great promise for solving several issues across various domains. The concept of synthesizing nanoparticles from their respective metals has been introduced by the combination of nanotechnology with other scientific disciplines such as chemistry, biology, and physics. Numerous kinds of nanoparticles have been created to date and are employed in numerous industries for a range of purposes. Additionally, scientists are drawn to Copper nanoparticles due to their notable and extensive bioactivity. Copper nanoparticles have been employed as possible antibacterial agents in numerous biomedical applications because of their high surface area to volume ratio. However, overuse of any metal nanoparticle raises the possibility of harm to people other animals and the environment.

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INTRODUCTION

The science of nanotechnology is still in its infancy, yet it has enormous potential to address many problems in a variety of fields. The fusion of nanotechnology with other scientific fields including chemistry, biology, and physics has created the idea of synthesizing nanoparticles from their respective metals. To date, several different types of nanoparticles have been developed and are used for a variety of purposes across many industries. Furthermore, the remarkable and broad bioactivity of copper nanoparticles attracts scientists. Due to their high surface area to volume ratio, copper nanoparticles have been used in many biomedical applications as potential antibacterial agents. Any metal nanoparticle used excessively, though, increases the risk of injury to humans, other animals, and the environment [1].

There are numerous uses for copper oxide nanoparticles, or CuO-NPs. The nanoparticles of copper oxide exhibit better catalytic activity and selectivity when compared to regular powdered copper oxide. It exhibits strong antibacterial properties against a range of bacterial types [2]. CuO-NPs are used in organic catalysis, gas sensors, semiconductors, dye removal, solar energy transformation, and many more applications [3]. There is another use for CuO-NPs in heat transmission. CuO-based Nano fluid has a 12.4% greater thermal conductivity than deionized

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water. A highly effective recyclable catalyst for the environmentally friendly production of 1,8-dioxooctahydroxanthenes in water is the CuO-CeO2 nanocomposite [4]. CuO-NPs catalysts that are supported by graphene oxide are called Nano-hybrid catalysts. Several techniques, including precipitation pyrolysis, hydrothermal microwave, electrochemical, wet chemical, and microwave irradiation, can be used to create CuO-NPs [5]. The purpose of this work was to examine the crystallite size, shape, microstructure, composition, and interactions between the species of CuO-NPs as well as to synthesize Nano-sized copper oxide powder in an efficient and straightforward manner. CuO-NPs were created in this work using the chemical precipitation process and then annealed at 200˚, 400˚, and 600˚ degrees Celsius.

MATERIALS AND METHODS

Chemical Reagents

We obtained copper nitrate, or Cu $(NO_3)_2$, from Merck in India. The supplier of sodium hydroxides was Lobha Chemie in India. In the experiment, analytical reagent-grade compounds were utilized, and they weren't further purified. Throughout the experiment, washing and solution preparation were done with deionized and distilled water.

Synthesis of Nanomaterial (Cu Nanoparticles)

With the use of sodium hydroxide and the copper nitrate Cu $(NO_3)_2$, the creation of copper nanoparticles was completed. The copper nitrate was weighed at room temperature with 3.0 g and 5 ml from a NaOH solution dissolved in 100 ml of deionized water. The NaOH solution was gradually poured down while being constantly stirred, and when the temperature cooled to 250 °C, the maximum amount of blue solution was visible on the magnetic stirrer for 30 minutes. Later, as a catalyst for the Solvthermal process, 5 milliliters of triethylamine (TEA) was added to dilute it. To make sure there were no contaminants in the resultant solution, it was centrifuged and repeatedly cleaned with DI water. The final solution was applied to the glass surface as a thin coating. The glass surface was first washed using a commercial detergent, and then it was cleaned for 30 minutes in an ultrasonic cleaner with ethanol. Ultimately, a DI rinse and symbolic drying were applied to the glass surface. After evaporating the liquid mixture on the glass surface in air below 125 °C until it solidified, the thin film was annealed at 250 °C for 30 minutes in the air.

Characterization

Using an X-ray diffractometer (XRD, Bruker D8 Advance, Germany) with a 0.15405 nm Cu-Kα radiation source in the 2θ range of 20^ to 80^ (40 KV, 40 mA, step size 0.020, scan rate 0.50 min^{-1}), X-ray diffraction (XRD) was used to analyze the phase and estimate the crystallite size of the samples. The diffraction intensity versus 2θ XRD patterns was captured. Scanning electron microscopy (JEOL, JSM-7600F, Japan) was used

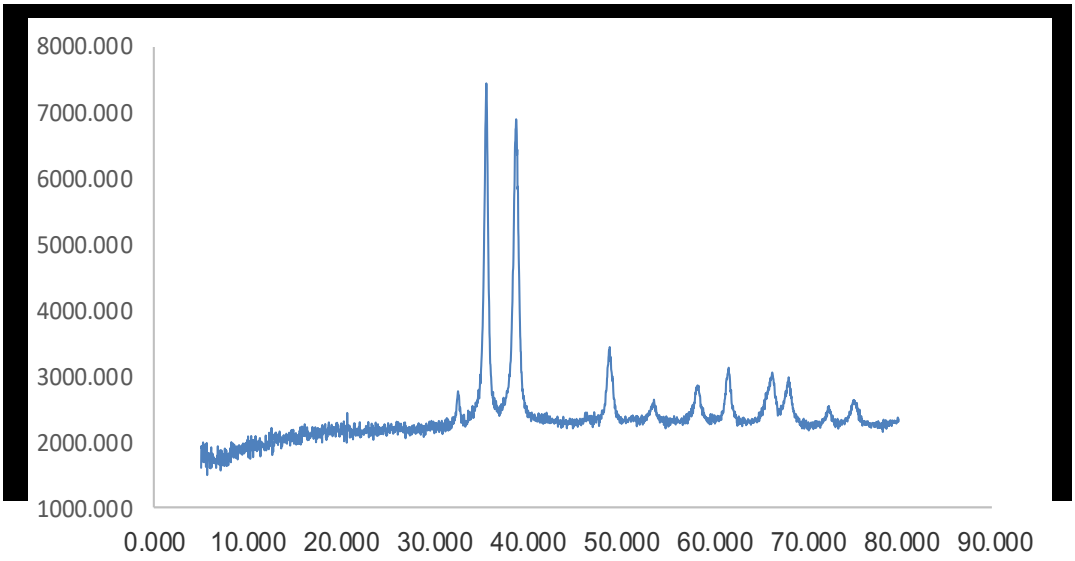

Fig. 1. XRD pattern of Cu nanoparticles

to determine the sample's surface morphology. JEOL JSM-7600F was used to do energy dispersive X-ray (EDX) analysis for composition analysis. The sample's FTIR spectroscopy was obtained in the region between 500 - 4000 cm^{-1} (with Perkin Elmer 1650, USA) on a Thermo-Nicolet Avatar 370 model FT-IR.

RESULTS AND DISCUSSION

X- Ray Diffraction for Copper

The crystalline growth nature of thin films created by solid phase on glass substrate at annealing temperature 300°C can be understood by examining the X-ray diffraction spectrum. created Cu nanoparticles are depicted in Fig. 1. It

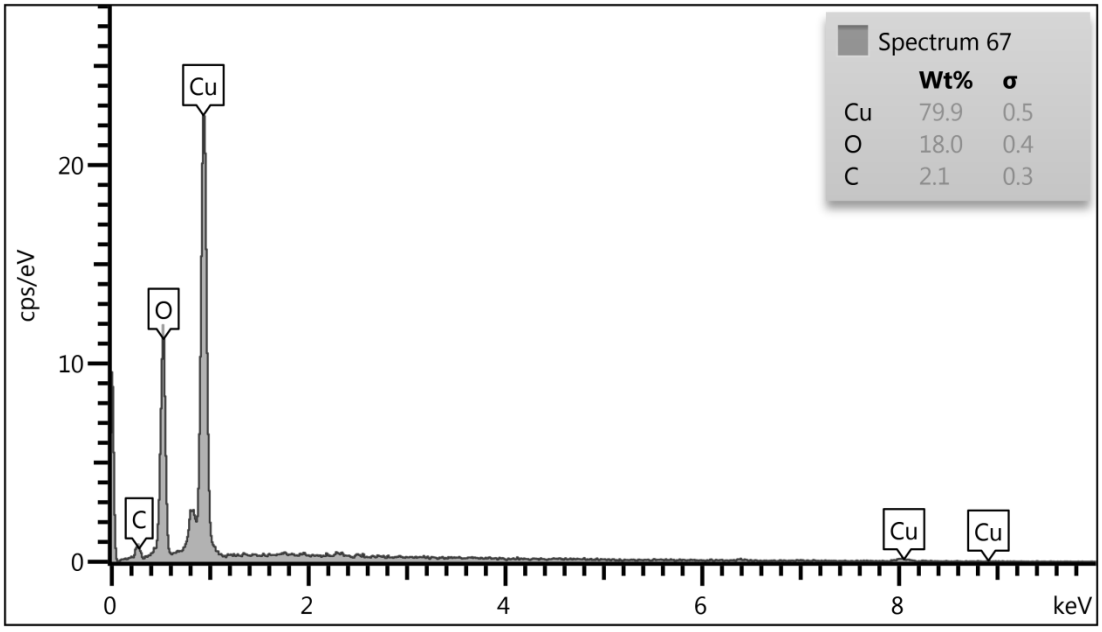

Fig. 2. *The EDS analysis of CuNPs*

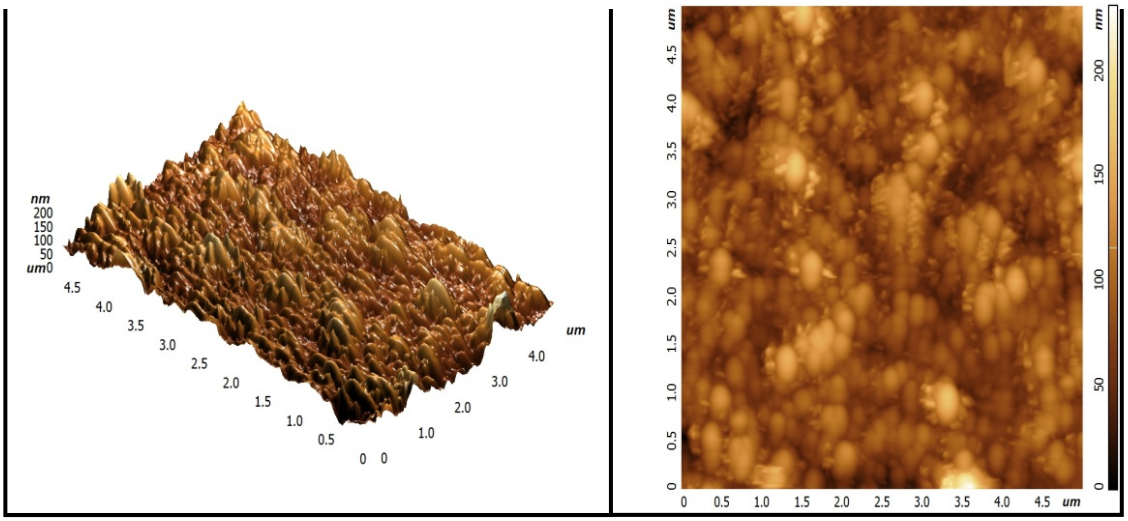

Fig. 3. *AFM images of CuNPs*

yields a monoclinic structure in a single phase. The parameters of the lattice are a = 4.84 Å, b = 3.47 Å, and c = 5.33 Å. The peak positions and intensities match the stated data (JCPDS file No. 05-661) rather well. In accordance with planes (002) and (111), (222), and (113), in that order Using the Debye–Scherrer formula [97], the average crystal size was determined to be d = 45 nm. Additionally, the most intense peak was found at 2θ = 35.45° which corresponds to the spherical nanoparticle diffractions crystallized in the structure with the [002] lattice plane.

EDS Analysis

The EDS analysis is done with: JEOL Model JSM - 6390LV and EDS Machine: JEOL Model JED – 2300 at magnification of 10000 with a voltage of 20KV. The EDS analysis carried out on the sample is depicted in Fig. 2, in which characteristic peaks corresponding to Cu and oxygen are seen and there is no traces of precursors, which shows the purity of the CuO Nano-particles formed and the data indicated the nanoparticles were nearly stoichiometric. The weight percent of copper and oxide calculated from EDX analysis were O: 8.73 weight % (0.525 keV) and Cu: 91.27 weight % (0.804 keV), respectively. There were no other elemental impurities in the EDX spectra. The EDX result showed the presence of uniform distribution of copper to oxygen with atomic ratio of 1:1 in CuO. This result confirmed the formation of pure CuO-NPs. The elemental analysis of the sample shows that the prepared sample was copper oxide, which is in good concord with the results of XRD. Similar results reported elsewhere [6-7].

Surface Morphology by Atomic Force Macroscopic (AFM)

Fig. 3 shows the AFM image for Cu NPs with many Nano- structure on the surface homogeneously in the film so the particle keep size as deposited on the surface of substrate[8].

Scanning Electron Microscopes (SEM)

structure of growth Cu NPs, was analyzer by the scanning electron microscopic which was utilized and the result is displayed in the Fig. 4 with the Cu NPs was synthesized by the seed growth method, which include chemical reaction with watery solutions of $Cu(NO_3)_2$ as nitrate .5 H_2O 0.1M and NaOH 0.9M solution whose pH value was 13 at room temperature. As can you see from the low – magnification image show as deposited Cu having several aggregated molecular nanostructures. When the nanoparticles were checked at magnifying. The nanoparticles were with a range of 16-55 nm [8]. Fig. 4 shows how the Cu Nanocrystals organized into spherical

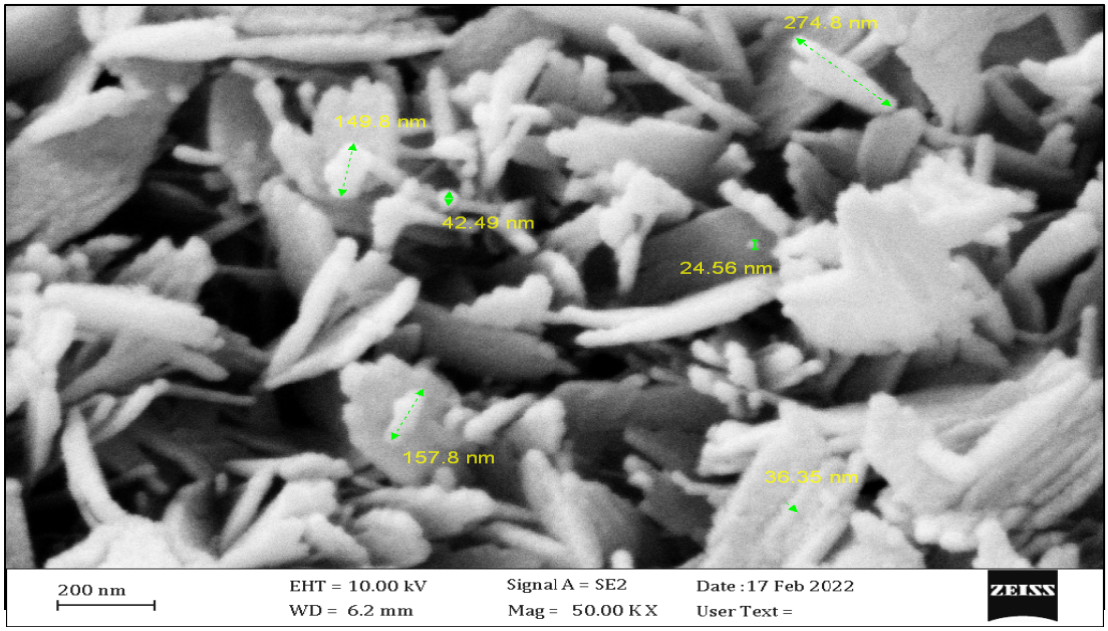

Fig. 4. FE-SEM Cu NPs

assemblies, where with higher magnification on individual particle it appears like dandelion. In the present work, Cu $(NO_{3)}$ $.5H_2O$ was used to prepare spherical shaped Nano Cu at mild reaction temperature. The same morphology was obtained when using Cu $(NO_3)_2$. $3H_2O$ and the surface exhibited smooth particles with a homogenous surface morphology, with a wide size distribution of particles size of aggregated particles.[9].

Antibacterial activity

The antibacterial potential of the prepared

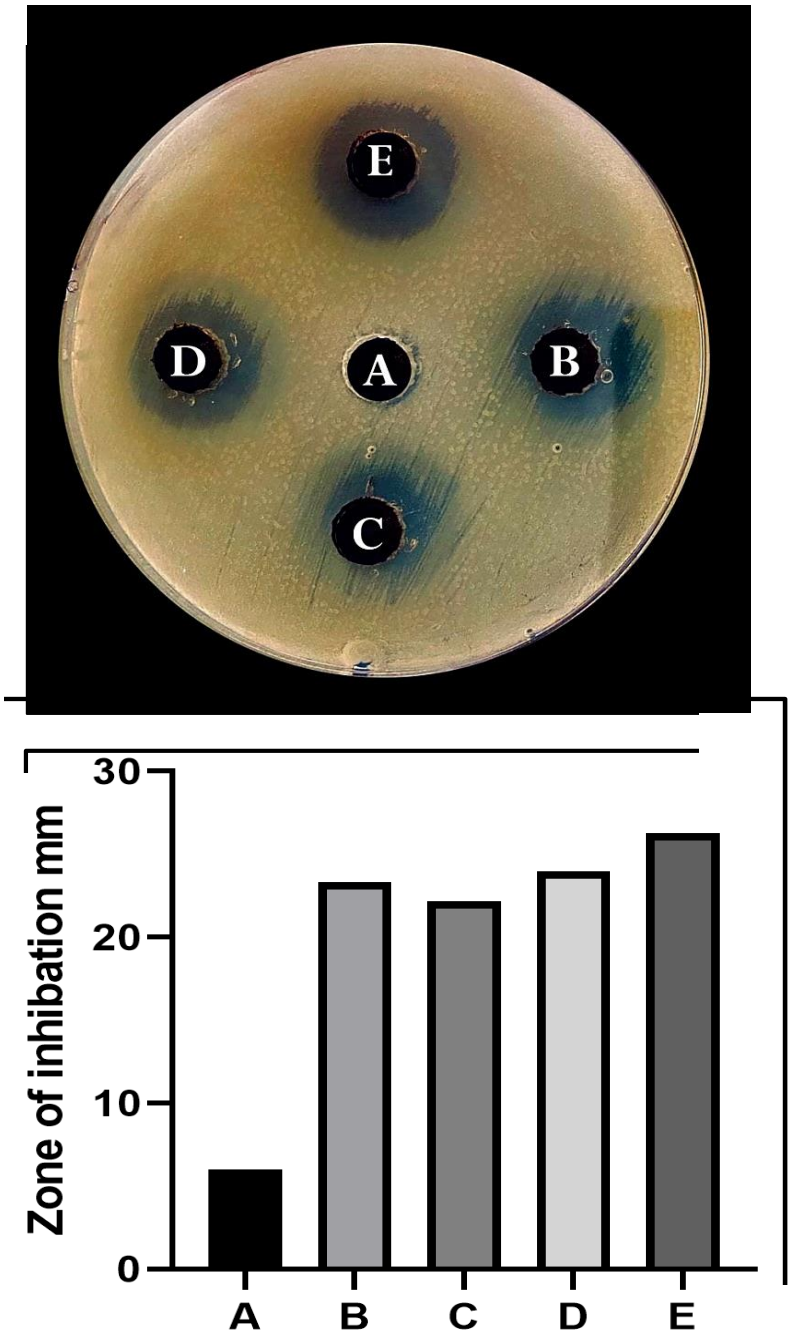

Fig. 5. Antibacterial activity of (N1) against *E.Coli.* A, control. B, 25%. C, 50%. D, 75%. E, 100%

X-SUBSTANCES was investigated against Gram's negative (E.Coli) and Gram's positive (S.aureus) bacterial strains using agar well diffusion assay [1, 2]. About 20mL of on Muller–Hinton (MH) agar was aseptically poured into sterile Petri dishes. The bacterial species were collected from their stock cultures using a sterile wire loop [3]. After culturing the organisms, 6 mm-diameter wells were bored on the agar plates using of a sterile tip. Into the bored wells, different concentrations

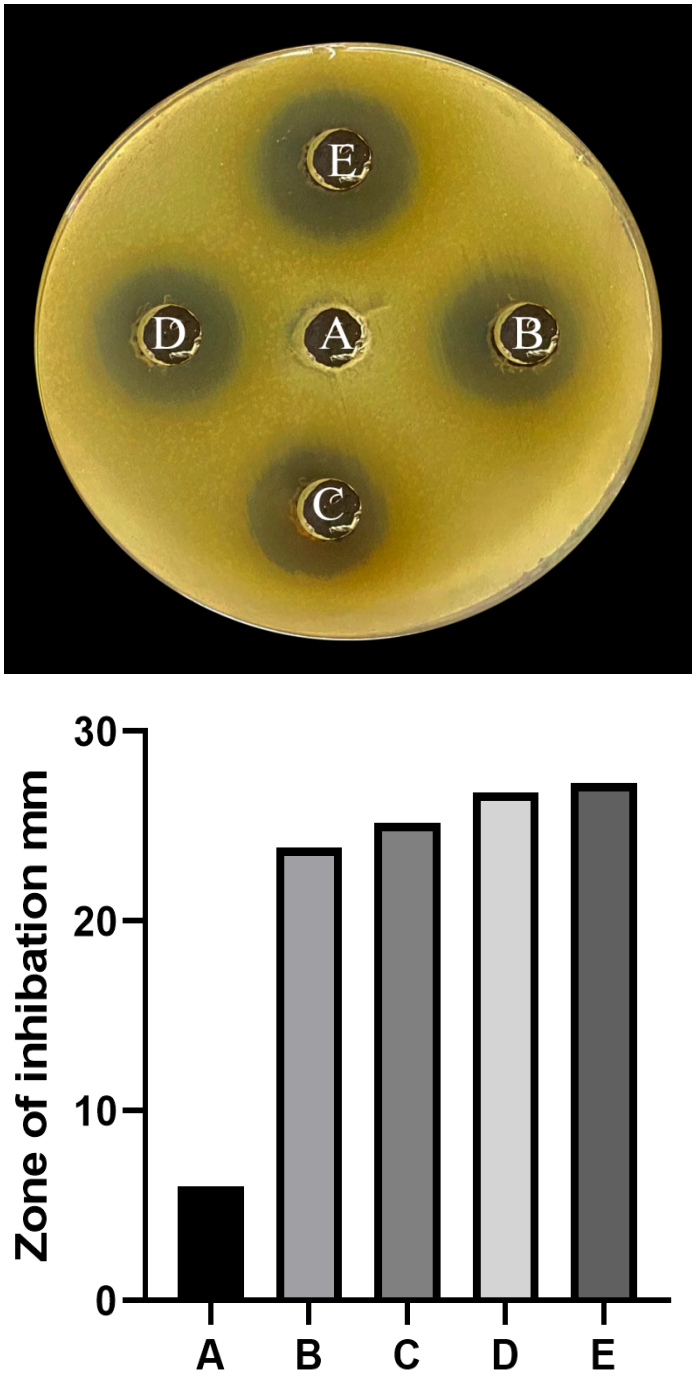

Fig. 6. Antibacterial activity of (N2) against *E.Coli.* A, control. B, 25%. C, 50%. D, 75%. E, 100%.

of the X-SUBSTANCES were used. The cultured plates containing the X-SUBSTANCES and the test organisms were incubated overnight at 37°C before measuring and recording the average the zones of inhibition diameter (Figs. 5-8) [11-13].

Statistical analysis

Data were statically analysis using Graphpad prism program [14]. Data are represented as mean

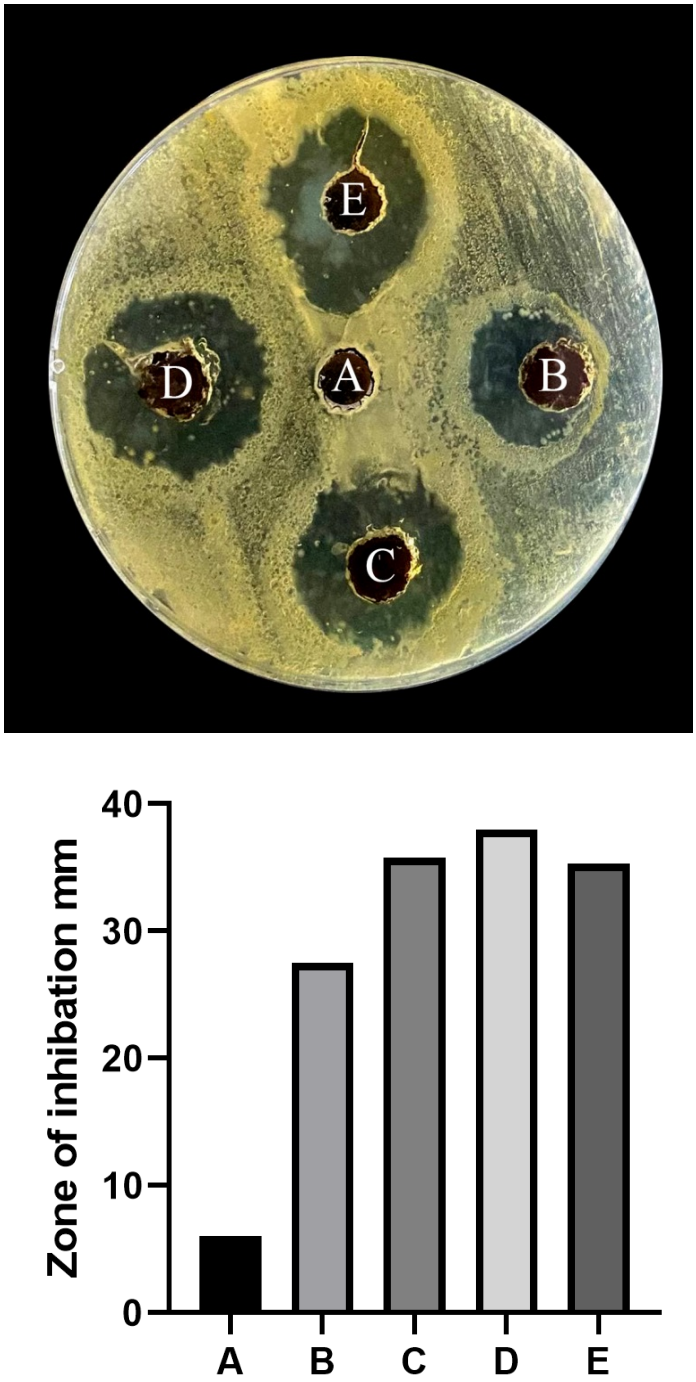

Fig. 7. Antibacterial activity of (N1) against *S.aureus.* A, control. B, 25%. C, 50%. D, 75%. E, 100%.

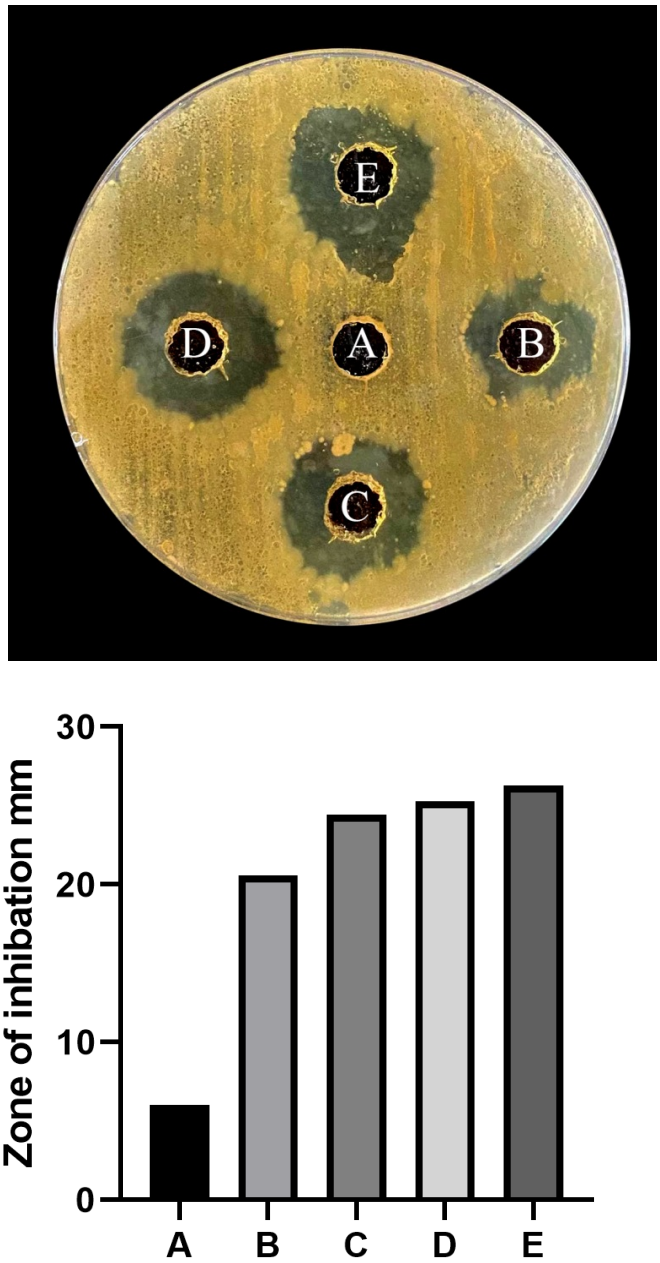

Fig. 8. Antibacterial activity of (N2) against *S.aureus.* A, control. B, 25%. C, 50%. D, 75%. E, 100%.

± SD of three experiments. Indicate statistically significant difference at $p<0.05$ (Figs. 5-8) [15].

CONCLUSION

Various studies have revealed that copper nanoparticles can be synthesized by chemical, physical, and biological routes. The physical and chemical methods are time-consuming and tedious. Moreover, some chemical methods include use of hazardous chemicals, which may exert adverse effects to the user. Therefore, ecofriendly, easy, and rapid methods are needed. Biological synthesis is an effort in this direction

Studies on bioactivities of copper nanoparticles

proved their effectiveness against the wide range of pathogenic bacteria, fungi, algae.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

REFERENCES

1. Anyaogu KC, Fedorov AV, Neckers DC. Synthesis, Characterization, and Antifouling Potential of Functionalized Copper Nanoparticles. Langmuir. 2008;24(8):4340-4346.
2. Beveridge TJ, Murray RG. Sites of metal deposition in the cell wall of Bacillus subtilis. Journal of bacteriology. 1980;141(2):876-887.
3. Blosi M, Albonetti S, Dondi M, Martelli C, Baldi G. Microwave-assisted polyol synthesis of Cu nanoparticles. J Nanopart Res. 2010;13(1):127-138.
4. Borkow G, Gabbay J. Copper, An Ancient Remedy Returning to Fight Microbial, Fungal and Viral Infections. Curr Chem Biol. 2009;3(3):272-278.
5. Bennett Chandran C, Subramanian TV, Arthur Felse P. Chemometric optimisation of parameters for biocatalytic reduction of copper ion by a crude enzyme lysate of Saccharomyces cerevisiae grown under catabolic repression conditions. Biochem Eng J. 2001;8(1):31-37.
6. Chang Y-N, Zhang M, Xia L, Zhang J, Xing G. The Toxic Effects and Mechanisms of CuO and ZnO Nanoparticles. Materials. 2012;5(12):2850-2871.
7. Chatterjee AK, Sarkar RK, Chattopadhyay AP, Aich P, Chakraborty R, Basu T. A simple robust method for synthesis of metallic copper nanoparticles of high antibacterial potency against E. coli. Nanotechnology. 2012;23(8):085103.
8. Chattopadhyay DP, Patel BH. Effect of Nanosized Colloidal Copper on Cotton Fabric. Journal of Engineered Fibers and Fabrics. 2010;5(3):155892501000500.
9. Bahjat HH, Ismail RA, Sulaiman GM, Jabir MS. Magnetic Field-Assisted Laser Ablation of Titanium Dioxide Nanoparticles in Water for Anti-Bacterial Applications. Journal of Inorganic and Organometallic Polymers and Materials. 2021;31(9):3649-3656.
10. Khashan KS, Abdulameer FA, Jabir MS, Hadi AA, Sulaiman GM. Anticancer activity and toxicity of carbon nanoparticles produced by pulsed laser ablation of graphite in water. Advances in Natural Sciences: Nanoscience and Nanotechnology. 2020;11(3):035010.
11. Khashan KS, Badr BA, Sulaiman GM, Jabir MS, Hussain SA. Antibacterial activity of Zinc Oxide nanostructured materials synthesis by laser ablation method. Journal of Physics: Conference Series. 2021;1795(1):012040.
12. Jihad MA, Noori FTM, Jabir MS, Albukhaty S, AlMalki FA, Alyamani AA. Polyethylene Glycol Functionalized Graphene Oxide Nanoparticles Loaded with Nigella sativa Extract: A Smart Antibacterial Therapeutic Drug Delivery System. Molecules (Basel, Switzerland). 2021;26(11):3067.
13. Mohammed MKA, Mohammad MR, Jabir MS, Ahmed DS. Functionalization, characterization, and antibacterial activity of single wall and multi wall carbon nanotubes. IOP Conference Series: Materials Science and Engineering. 2020;757(1):012028.
14. Ali IH, Jabir MS, Al-Shmgani HSA, Sulaiman GM, Sadoon AH. Pathological And Immunological Study On Infection With Escherichia Coli In ale BALB/c mice. Journal of Physics: Conference Series. 2018;1003:012009.
15. Luksic I. Global and regional estimates of the incidence and case-fatality rates of childhood bacterial meningitis caused by streptococcus pneumoniae. Morressier; 2016.