

Antimicrobial Activity of Copper Nanoparticles Synthesised by Ginger (*Zingiber officinale*) Extract

Ipsa Subhankari and P.L.Nayak

P.L. Nayak Research Foundation and Centre for Excellence in Nano Science and Technology,
Synergy Institute of Technology, Bhubaneswar, Odisha, India

Abstracts: Copper nanoparticles are known to be one of the multifunctional inorganic nanoparticles with effective antibacterial activity. This study aims to determine the antimicrobial efficacy of green and chemical synthesized Cu nanoparticle against various bacterial and fungal pathogens. Various microbiological tests were performed using varying concentrations of green synthesised Cu NPs with sizes 40 and 25 nm respectively. Effectiveness of nanoparticles increases with increasing particle dose, treatment time and synthesis method. In addition, the current study has clearly demonstrated that the particle size variation and surface area to volume ratio of green Cu nanoparticle are responsible for significant higher antimicrobial activity. Herein, we are reporting a novel biological approach for the formation of copper nanoparticles using *ginger*. Copper sulphate was made to reduce with aqueous soln of clove extracts. X-Ray diffraction (XRD) pattern reveals the formation of Cu nanoparticles, which shows crystallinity. UV-V in absorption shows characteristic absorption peak of Cu nanoparticles.

Key words: Copper sulphate • XRD • pH • UV-V • Antimicrobial activity • Ginger

INTRODUCTION

The emergence of nanoscience and nanotechnology in the last decade presents opportunities for exploring the bactericidal effect of metal nanoparticles. The bactericidal effect of metal nanoparticles has been attributed to their small size and high surface to volume ratio, which allows them to interact closely with microbial membranes and is not merely due to the release of metal ions in solution.

Plant Description: **Ginger** or **ginger root** is the rhizome of the plant *Zingiber officinale*, consumed as a delicacy, medicine, or spice. It lends its name to its genus and family (Zingiberaceae). Other notable members of this plant family are turmeric, cardamom and galangal.

MATERIALS AND METHODS

Collection of Extracts: Ginger were collected from the local market. They were washed and cleaned and cut into small pieces with triple distilled water and dried with water

absorbent paper. Then it was crushed with the help of mortar and pestle dispensed in 10 ml of sterile distilled water and heated for 2-3 minutes at 70-80°C. The extract was then filtered using Whatman's No.1 filter paper. The filtrate was collected in a clean and dried conical flask by standard sterilized filtration method and was stored.

Synthesis of Copper Nanoparticles: For the synthesis of copper Nanoparticles, both the precursor and the reducing agent were mixed in a clean test tube in 1:1 proportion. For the reduction of Cu ions, 5ml of filtered Copper extract was mixed to 5 ml of freshly prepared 0.001 M aqueous of CuSo₄ solution. It was then kept for incubation for 1hr. Within a particular time, the change in colour was noted from straw yellow to sea green. Thus colour change indicates reduction and reduced copper nanoparticles were obtained.

pH Analysis: The pH was determined by using Digital pH meter. The pH of the reduced solution with Nanoparticle synthesized was found to be 1.91.

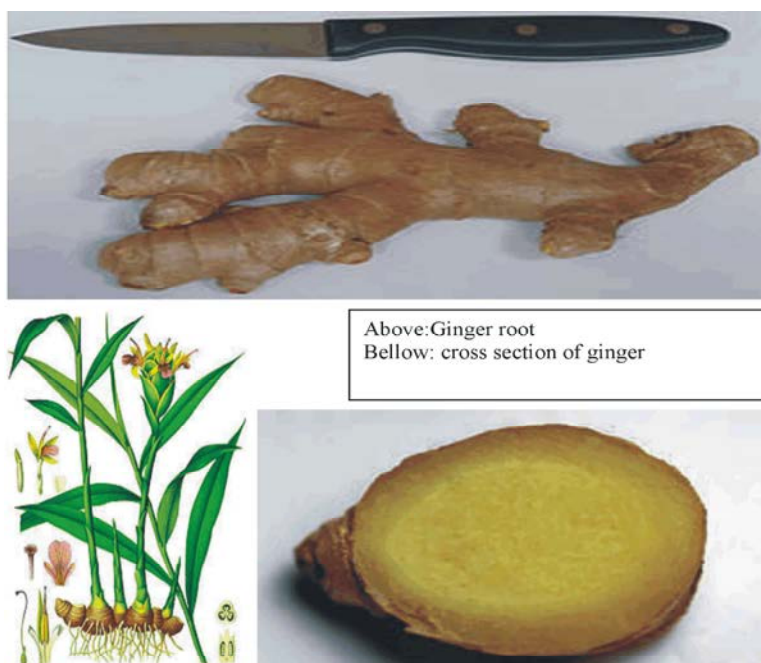


Fig. 1: Images of Ginger



Fig. 2: Tube A- contain copper sulphate solution, Tube B- contain ginger extract, Tube C- contain light green coloured copper nanoparticles solution

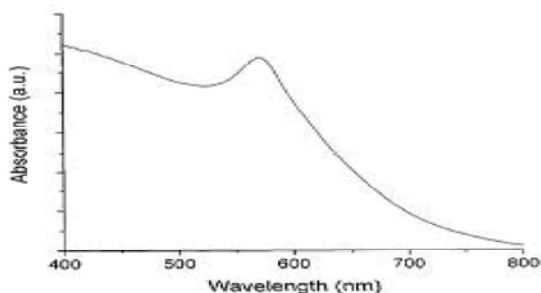


Fig. 3: The absorption maxima of copper nanoparticles at 570 nm

UV-Vis Spectra Analysis: The reduction of pure Cu to nanoparticle was monitored by measuring the UV-Vis spectrum the most confirmatory tool for the detection of surface Plasmon resonance property (SPR) of CuNPs, by diluting a small aliquot of the sample in distilled water. UV-Vis spectral analysis was done by using UV-Vis spectrophotometer Systronics 118 at the range of 300-600 nm.

X-ray Diffraction (XRD) Analysis: Diffraction pattern gives information on translational symmetry - size and shape of the unit cell from Peak Positions and information on electron density inside the unit cell, namely where the atoms are located from Peak Intensities. It also gives information on deviations from a perfect particle, if size is less than roughly 100 – 200nm, extended defects and micro strain from Peak Shapes & Widths. XRD measurement of green synthesized CuNPs from copper sulphate, was drop-coated on glass on a Bruker ax- D8 Advance instrument operating at a voltage of 40 KV and current of 20 mA with Cu K α radiation.

Antimicrobial Activity: The emergence of nanoscience and nanotechnology in the last decade presents opportunities for exploring the bactericidal effect of metal nanoparticles. The bactericidal effect of metal nanoparticles has been attributed to their small size and high surface to volume ratio, which allows them to interact

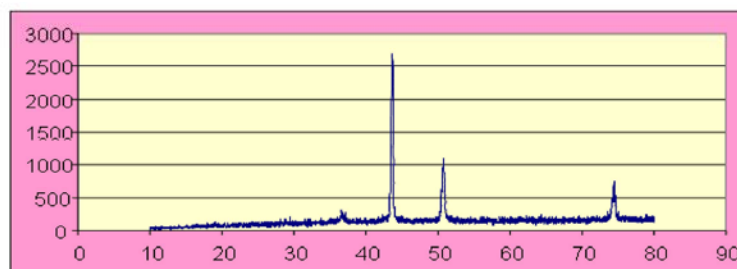


Fig. 4: XRD Pattern of Green Synthesised Cu Nanoparticles



Fig. 5: Inhibition zones against E.Coli by green synthesised copper nanoparticles

Table 1: Antibacterial Activity Table of Copper nanoparticles against *E.Coli*.

Sample in well	Name of bacteria	Variety of bacteria	Inhibition zone diameter (in mm)
Reduced CuSo ₄ + Ginger extract	<i>Escherichia coli</i>	Gram (-)	15
	CuSo ₄	<i>Escherichia coli</i>	Gram (-) 0
Ginger extract	<i>Escherichia coli</i>	Gram (-)	5

closely with microbial membranes and is not merely due to the release of metal ions in solution. A cell wall is present around the outside of the bacterial cell membrane and it is essential to the survival of bacteria. It is made from polysaccharides and peptides named *peptidoglycon*. There are broadly speaking two different types of cell wall in bacteria, called gram-positive and gramnegative. The names originate from the reaction of cells to the gram stain, a test long-employed for the classification of bacterial species. Gram-positive bacteria possess a thick cell wall containing many layers of peptidoglycan. In contrast, gram-negative bacteria have a relatively thin cell wall consisting of a few layers of peptidoglycan. Surfaces of copper nanoparticles affect interact directly with the bacterial outer membrane, causing the membrane to rupture and killing bacteria. Antibacterial activity of

copper nanoparticles synthesized by electrolysis was evaluated by using standard Zone of Inhibition (ZOI) microbiology assay. The sample copper nanoparticles prepared in electrolysis method showed diameter of inhibition zone against *E.Coli* 15 mm.

CONCLUSION

We have come for conclusion that copper nanoparticles synthesized in Gren synthesis method are showing antibacterial activities against gram (+) bacteria. Copper nanoparticles synthesized in green synthesis method are showing more antibacterial activities (For *E.Coli* bacteria) than coppersulphate solution and pure ginger extract. The chemicals involved in the synthesis of nanoparticles are commonly available,

cheap and non-toxic. The experiments suggest the possibility to use this material in water purification, air filtration, air quality management, antibacterial packaging etc.

ACKNOWLEDGEMENT

The authors are sincerely thankful to Physics Department, Pune University to provide XRD and National Chemical Laboratory for Particle size analysis. Also to the Head of the Department, University Department of Chemical Technology, North Maharashtra University, Jalgaon, Maharashtra and to the Directorate of General CIPET, Bhubaneswar, India. The authors are also thankful to Shri Binod Dash, Chairman, Synergy Institute of Technology for providing facilities to carry out this piece of research work.

REFERENCES

1. Tiwari, D.K. and J. Behari, 2009. Advances in Biological Research, 3(3-4): 89-95.
2. Yamamoto, O., M. Komatso, Jun Sawai and Zenbe-E-Nakagawa, 2004. Journal Materials science:Materials in Medicine, 15: 847-851.
3. Omid Akhavan and Elham Ghaderi, 2009. "Enhancement of antibacterial properties of Ag nanorods by electric field", Sci. Technol. Adv. Mater, vol.10 No. 015003, pp: 5. doi:10.1088/1468-6996/10/1/015003.
4. Prakash, N., S. Jayapradeep and P.N. Sudha, April 6-8, 2009. Investigation of Antimicrobial Properties of Silver and Copper Nanoparticles Encapsulated in Chitosan, First International Conference on Nanostructured Materials and Nanocomposites, Kottayam, India, pp: 311-317.
5. Blanc, DS., P. Carrara, G. Zanetti and P. Francioli, 2005. Water disinfection with ozone, copper and silver ions and temperature increase to control Legionella: seven years of experience in a university teaching hospital. J. Hosp. Infect., 60: 69-72.
6. Kolar, M., K. Urbanek and T. Latal Ag, 2001. Antibiotic selective pressure and development of bacterial resistance. Int. J. Antimicrob., 17: 357-63.