

## Biogenic Synthesis of Copper Nanoparticles Using Leaf Extract of *Hemigraphis colorata* and Characterization

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### ABSTRACT:

**Purpose:** This study aims to synthesize copper nanoparticles (Cu-NPs) and evaluate the antimicrobial activity of the plant *Hemigraphis colorata*.

**Aim & Objective:** The primary objective was to synthesize green Cu-NPs mediated by the leaf extract of the medicinal plant *Hemigraphis colorata*. Integrating the bioactive compounds of this plant with Cu-NPs is expected to offer significant benefits for various ailments. The biogenically synthesized green Cu-NPs were characterized using UV-visible spectroscopy, FT-IR, XRD, SEM, TEM, and EDAX techniques. Additionally, the antimicrobial properties of the green Cu-NPs were assessed against gram-positive and gram-negative bacteria.

**Methodology:** *Hemigraphis colorata* (commonly known as murikooti) is a prostrate herb with rooting branches, deep reddish leaves, and white flowers, belonging to the Acanthaceae family. Leaves were collected from a medicinal garden and used for nanoparticle synthesis. The nanoparticles were characterized using UV-vis, FT-IR, XRD, SEM, TEM, and EDAX techniques. The antimicrobial activity of the synthesized nanoparticles was also evaluated.

**Results and Discussion:** Green synthesis of Cu-NPs was achieved using copper sulfate as the precursor and plant leaf extract as the reducing and capping agent. The color change from blue to light brown visually indicated the formation of Cu-NPs. The synthesized nanoparticles were purified by washing with deionized water and ethanol to remove any impurities.

**KEYWORDS:** *Hemigraphis colorata*, Green Synthesis, Cu NPs Characterization.

### INTRODUCTION:

Nanoparticles are small particles ranging between 1-100 nanometers in size, undetectable by the human eye. These particles can exhibit significantly different physical and chemical properties compared to their larger counterparts. Copper nanoparticles, which are copper-based particles around 100 nm in size, possess excellent catalytic activities and are useful in applications such as biosensors and electrochemical sensors. Copper is one of the nine

essential minerals for humans, playing a crucial role in various physiological processes across all tissues, including the skin[1].

The plant *Hemigraphis colorata* is a versatile tropical low-creeping perennial herb. It grows prostrate with spreading rooting stems. The leaves are opposite, ovate to cordate, featuring well-defined veins, and are slender, lance-shaped with toothed, scalloped margins. The leaves contain various chemical constituents such as natrium, calcium, carboxylic acids, cinnamate, chlorogenate, coumarate, coumarins, flavonoid polyphenols, saponins, tannins, ferulate, gallate, ascorbic acid, E-phenol, carbohydrates, steroids, proteins, and flavonoids[2].

A Drug Delivery System (DDS) is designed for the controlled release of therapeutic agents, monitoring both the release of the drug and its location within the body. Advances in drug delivery systems are focused on areas such as crossing the Blood-Brain Barrier (BBB) in the brain, treating diseases and disorders, enhancing targeted intracellular delivery, and combining diagnosis with treatment. Drug delivery refers to various approaches, formulations, technologies, and systems used to transport a pharmaceutical compound within the body to achieve its desired therapeutic effect, often utilizing nanoparticles to do so[3].

Drug delivery systems often involve precise targeting within the body or enhancing systemic pharmacokinetics, focusing on both the quantity and duration of drug presence. These systems can be based on the chemical formulation of the drug, medical devices, or a combination of both. Drug delivery is closely linked with dosage form and route of administration, sometimes considered part of its definition[4].

Nanoparticles can occur naturally, be by-products of combustion reactions, or be engineered for specific functions. Their use spans various industries, from healthcare and cosmetics to environmental preservation and air purification. In healthcare, one significant application of nanomaterials is in drug delivery. Copper nanoparticles, which are copper-based particles around 100 nm in size, can be produced through material processes or chemical synthesis. These nanoparticles are notable for their historical use as coloring agents and their modern biomedical applications. Due to their high flammability, copper nanoparticles must be stored away from ignition sources[5].

## MATERIALS AND METHODS

**Materials Collection:** The selected plant was collected from the medicinal garden in Palakkad and authenticated at the BSI Southern Regional Centre, with voucher number BSI/SRC/5/23/212. The specimen was submitted for further reference[6].

**Methodology for Green Synthesis of Copper Nanoparticles:** Plant leaves were collected, washed, and the leaf powder was soaked in 1 liter of distilled water. This mixture was held in a shaker at 30°C for 48 hours with continuous stirring at 50 rpm for thorough mixing. The aqueous plant extract was used to reduce CuSO<sub>4</sub>·5H<sub>2</sub>O. 100 ml of plant extract was added to 90 ml of 10 mM copper sulfate solution and stirred continuously at a temperature of 50-60°C.

A color change in the solution was observed visually, confirming the synthesis of copper nanoparticles [7]. The nanoparticles were characterized using techniques such as UV-Vis, FT-IR, XRD, SEM, TEM, and EDAX. The antimicrobial properties of the copper nanoparticles were also determined[8].

### CHARACTERIZATION TECHNIQUES:

**UV-VISIBLE:** An absorbance spectrum was recorded by scanning wavelengths between 200 and 800 nm. The absorbance of the nanoparticle solution was measured against a blank extract solution.

**FT-IR:** Fourier transform-infrared spectroscopy (FT-IR) spectrum (FT-IR PerkinElmer) was recorded using KBr pellets in the range of 400-4000  $\text{cm}^{-1}$ .

**XRD:** X-ray diffraction (XRD-Shimadzu X-ray diffractometer) (PXRD-7000) analytical technique was used to reveal the crystalline nature of the green synthesized Cu NPs.

**SEM:** Scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM-EDX, EVO18 model with low vacuum facility and ALTO 1000 Cryo attachment) was used to analyze the surface morphology and elemental composition of the nanoparticles.

**TEM:** Transmission electron microscopy (JEM-2100plus HRTEM) was used for understanding the morphological and structural features of the green synthesized Cu NPs.

### RESULTS AND DISCUSSION:

The green synthesis of copper nanoparticles (Cu NPs) was achieved using copper sulfate as a precursor and plant leaf extract as a reducing and capping agent. The visual change in color from blue to light brown indicated the formation of Cu NPs. The synthesized Cu NPs were washed with deionized water and ethanol to remove any impurities.

#### Characterization of Green Synthesized Copper Nanoparticles:

**UV-Vis Spectroscopic Analysis of Copper Nanoparticles:** UV-Vis spectroscopy within the 200-800 nm range was used to verify the development of Cu NPs. A characteristic peak at 575 nm indicated the absorption spectrum of the green synthesized Cu NPs.

**FT-IR Spectral Analysis:** Fourier transform infrared spectroscopy (FT-IR) was utilized to confirm the formation of nanoparticles. This technique provides a fingerprint of the vibrational and rotational modes of present molecules, aiding in the identification of functional plant molecules involved in the reduction of copper nanoparticles. (Fig. 1)

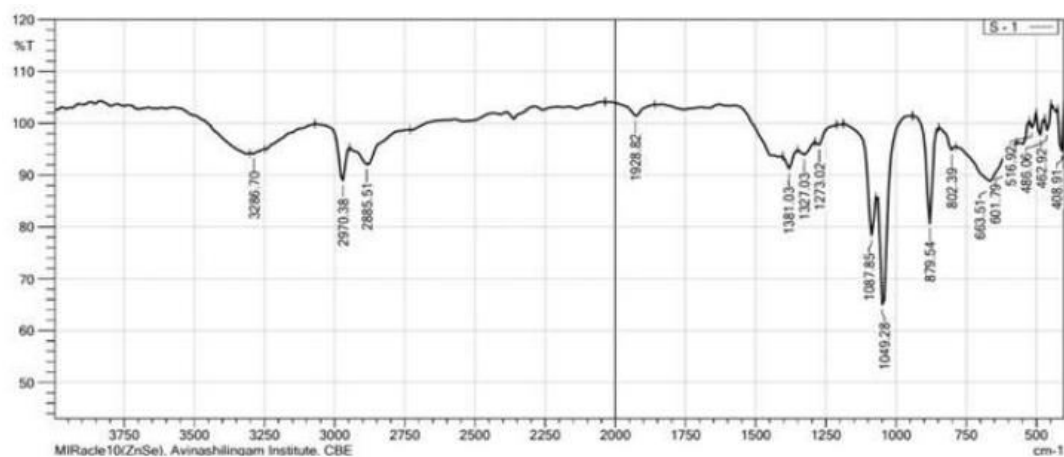


Fig 1: FT-IR Analysis of Copper Nano Particles

**XRD Analysis:**

The structure of the synthesized copper nanoparticles was determined using X-ray diffraction (XRD) analysis. Diffraction peaks were observed at  $2\theta$  values of  $44.32^\circ$  and  $65.25^\circ$ , confirming the crystalline nature of the copper nanoparticles. (Fig. 2)

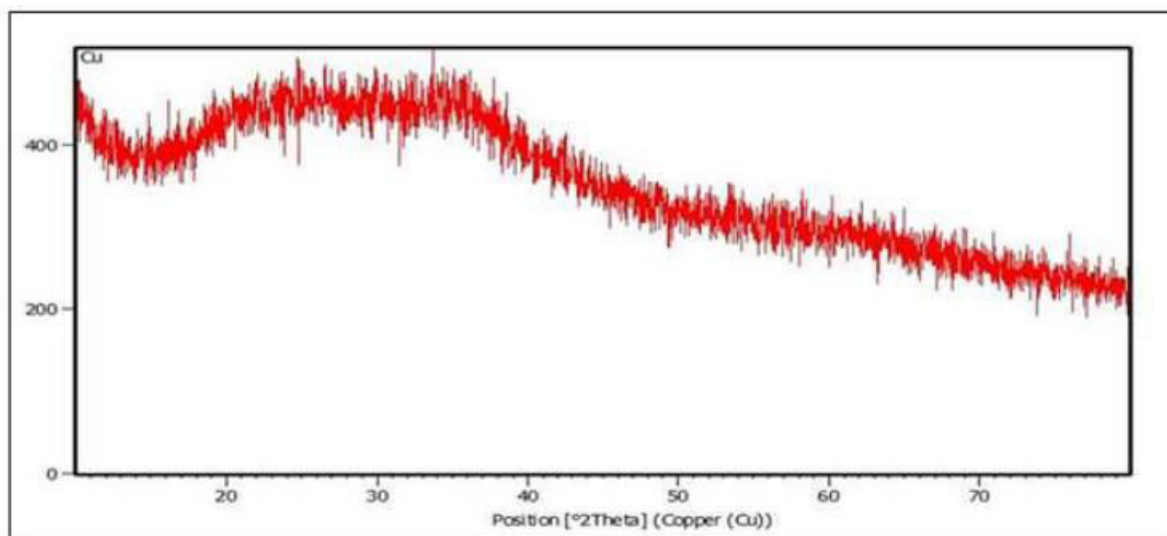


Fig.2: XRD Analysis of Copper Nano Particles

**Morphological Analysis by SEM:**

Field emission scanning electron microscopy (FE-SEM) was employed to study the shape and size of the copper nanoparticles. The analysis was conducted using the Schottky FE-SEM (Japan) TESCAN MIRA3 LMH model. The microscopy results revealed that the copper nanoparticles have a nano-range particle size of approximately 500 nm, with a spherical shape and homogeneous distribution. (Fig. 3)

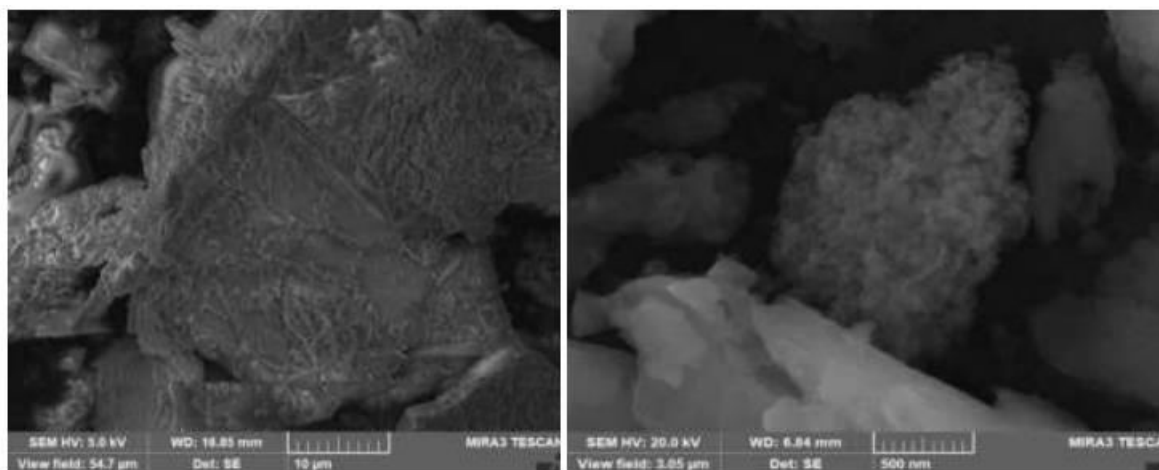


Fig.3: FE-SEM observation of green synthesised copper nanoparticles

**Morphological and Compositional Analysis by TEM and EDAX:**

Transmission electron microscopy (TEM) was used to gain deeper insight into the morphology, size, and crystalline nature of the copper nanoparticles. The TEM images of green synthesized copper nanoparticles (Figure 4) revealed that the nanoparticles exhibited various shapes. The plant extract served as a capping and stabilizing agent. (Fig. 4)

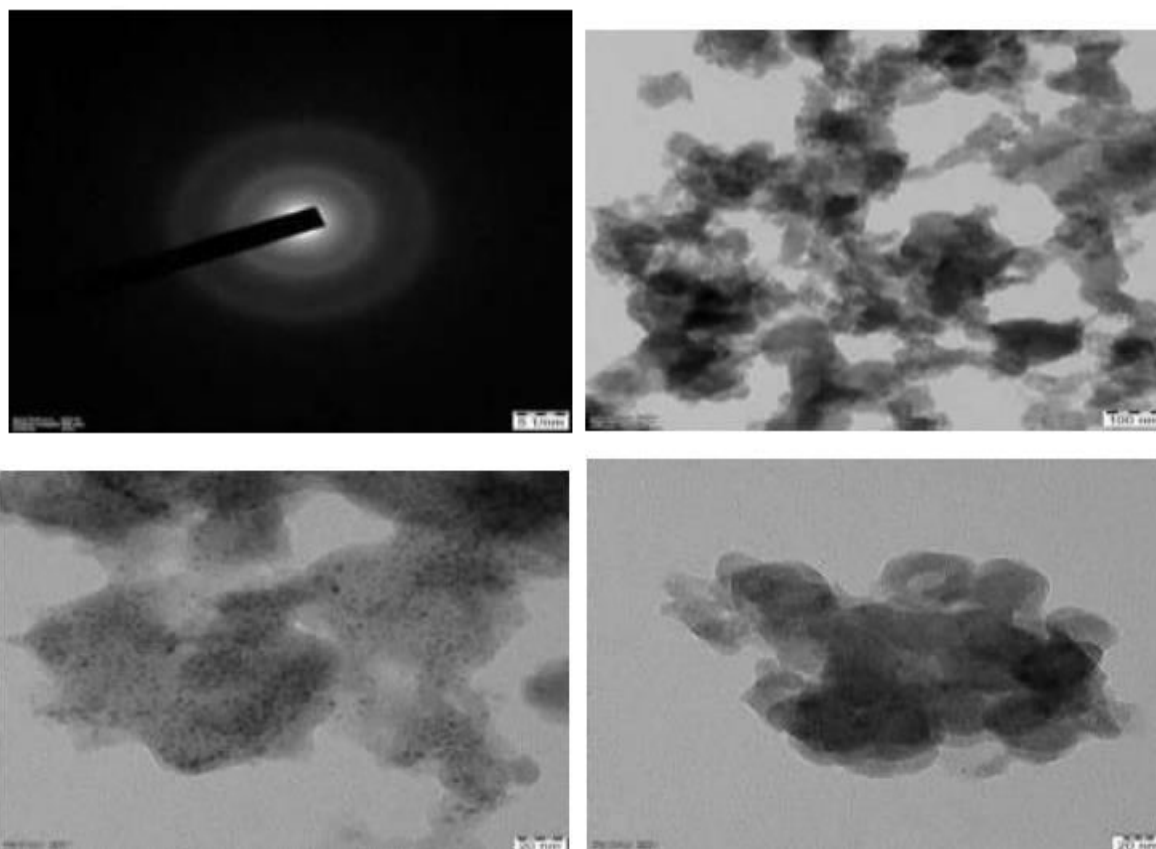


Fig 4: TEM images of green synthesised copper nanoparticles at lower magnification (100nm) and higher magnification (20 nm).

## CONCLUSION:

Green copper nanoparticles (g-Cu NPs) were successfully synthesized using the medicinal plant *Hemigraphis colorata*. The biogenically synthesized g-Cu NPs were characterized using UV-visible spectroscopy, FT-IR, XRD, SEM, TEM, and EDAX techniques. This research provides a comprehensive report on the synthesis and antimicrobial activities of g-Cu NPs, which are expected to have significant biomedical applications.

The UV-visible absorbance spectrum showed a maximum absorption ( $\lambda_{max}$ ) at 575 nm, while FT-IR spectra confirmed the presence of capping agents on the surface of the g-Cu NPs. The crystalline nature and composition of the g-Cu NPs were verified by XRD patterns and EDAX spectra, respectively. SEM and TEM micrographs provided evidence of the spherical shape and homogenous distribution of the nanoparticles, with an average size of 500 nm.

The antimicrobial properties of the g-Cu NPs were evaluated against gram-positive and gram-negative bacteria. The g-Cu NPs synthesized using the leaf extract of *Hemigraphis colorata* showed a broad range of antibacterial activity, with zones of inhibition of 11 mm and 15 mm against *Staphylococcus aureus* and *Escherichia coli*, respectively. These findings confirm the potential of g-Cu NPs as an effective remedy for infectious diseases caused by the tested bacterial pathogens.

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