International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Green Synthesis and Biological Evaluation of *Couroupita guianensis* Flower Extract

Navnita J Basu
Department of Biotechnology
Sri Krishna Arts and Science College Coimbatore, India

Abstract - The present study investigates the green synthesis of copper nanoparticles (CuNPs) using aqueous flower extract of *Couroupita guianensis* and evaluates their physicochemical characteristics, antioxidant potential, and antibacterial activity. Preliminary phytochemical screening of the flower extract revealed the presence of bioactive compounds including flavonoids, phenolics, alkaloids, terpenoids, saponins, glycosides, steroids, coumarins, carbohydrates, amino acids, and essential oils, which are known to facilitate nanoparticle reduction and stabilization. Successful synthesis of copper nanoparticles was indicated by a visible color change from blue to green and confirmed through UV–Visible spectroscopy, which exhibited a characteristic surface plasmon resonance peak at 564 nm. Fourier Transform Infrared (FTIR) analysis identified functional groups such as O–H, C–H, C=O, and Cu–O, confirming the involvement of phytochemicals in nanoparticle formation and stabilization. X-ray Diffraction (XRD) analysis revealed diffraction peaks corresponding to the crystalline face-centered cubic structure of copper nanoparticles. The synthesized nanoparticles demonstrated significant antioxidant activity in the DPPH radical scavenging assay, showing concentration-dependent free radical inhibition. Antibacterial evaluation against Escherichia coli using the disc diffusion method revealed a maximum zone of inhibition of 18 mm, which was greater than that observed for the crude flower extract and copper sulfate solution. The enhanced biological activity may be attributed to the synergistic effects of copper nanoparticles and phytochemical capping agents. Overall, the findings demonstrate that *C. guianensis*-mediated copper nanoparticles possess promising antioxidant and antibacterial properties and offer a sustainable, eco-friendly alternative for potential biomedical and nanotechnological applications.

Keywords: *Couroupita guianensis*, Green synthesis, Copper nanoparticles, Phytochemicals, Antioxidant activity, Antibacterial activity, Nanobiotechnology.

INTRODUCTION

The rise of microbial resistance has become a critical global health challenge, threatening the effectiveness of conventional antibiotics and increasing the burden of infectious diseases. According to the World Health Organization, antimicrobial resistance is projected to cause millions of deaths by 2050 if effective alternatives are not developed. Consequently, there is an urgent need for new antimicrobial agents that are not only effective but also environmentally sustainable (Iravani, 2011). In parallel, conventional methods for synthesizing metal nanoparticles, which have shown promising antimicrobial properties, often involve toxic chemicals, high energy consumption, and hazardous by-products. These factors have sparked a global interest in exploring eco-friendly and sustainable approaches, particularly in the field of green nanotechnology (Ahmed *et al*., 2016).

The utilization of natural products for medicinal, industrial, and technological purposes dates back to ancient civilizations, where plant-derived materials were widely used for therapeutic, cosmetic, and preservative applications. In recent decades, increasing environmental concerns and the global rise of antimicrobial resistance have renewed scientific interest in sustainable and eco-friendly alternatives to synthetic chemicals. This shift has encouraged the exploration of plant-based resources in advanced fields such as biotechnology and nanotechnology (Iravani, 2011; Ahmed *et al*., 2016).

Green synthesis of nanoparticles utilizes biological materials such as plants, bacteria, fungi, and algae to reduce metal ions into nanoparticles. Among these, plant-mediated synthesis is considered superior due to its simplicity, cost-effectiveness, scalability, and biocompatibility. Plant extracts contain diverse phytochemicals that act as natural reducing and stabilizing agents, eliminating the need for external chemical reagents and minimizing environmental hazards (Sharma *et al*., 2019). The integration of traditional medicinal knowledge with modern nanotechnology thus provides a

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

unique platform for the development of novel therapeutic materials.

Nanotechnology involves the manipulation of materials at the nanoscale, typically ranging from 1 to 100 nm, where materials exhibit unique physicochemical and biological properties distinct from their bulk counterparts. Metal nanoparticles have gained significant attention due to their enhanced surface reactivity, optical behavior, and antimicrobial efficiency. However, conventional physical and chemical synthesis methods often require toxic reagents, extreme reaction conditions, and high energy input, raising concerns about environmental safety and biological compatibility (Rao *et al*., 2017).

To overcome these limitations, green synthesis of nanoparticles has emerged as an environmentally benign alternative. This approach utilizes biological systems such as plants, bacteria, fungi, and algae for nanoparticle production. Among these, plant-mediated synthesis is particularly advantageous due to its simplicity, cost-effectiveness, scalability, and the presence of diverse phytochemicals capable of acting as reducing and stabilizing agents (Sharma *et al*., 2019).

Couroupita guianensis Aubl., commonly known as the Nagalingam or cannonball tree, belongs to the family Lecythidaceae and is widely distributed in tropical regions of India, South America, and Southeast Asia. The tree is valued not only for its aesthetic and religious significance but also for its medicinal properties. Various parts of the plant, including leaves, bark, fruits, and flowers, have been traditionally used in folk medicine to treat skin disorders, wounds, inflammation, fever, and microbial infections. While extensive studies have been conducted on the leaves and bark, the flowers remain relatively underexplored despite their rich biochemical composition and potential medicinal value (Sharma *et al*., 2018).

Plant extracts contain a wide range of secondary metabolites, including flavonoids, alkaloids, phenolic compounds, terpenoids, glycosides, and proteins, which play a crucial role in nanoparticle formation. These biomolecules facilitate the reduction of metal ions while simultaneously stabilizing the nanoparticles, eliminating the need for external chemical agents (Iravani *et al*., 2014). Additionally, phytochemical-capped nanoparticles often exhibit enhanced biological activities, particularly antimicrobial and antioxidant effects (Singh *et al*., 2018).

Couroupita guianensis, commonly known as the Nagalingam tree or Cannonball tree, belongs to the family Lecythidaceae and is widely distributed in tropical regions, especially in India. The plant holds religious, ornamental, and medicinal importance and has been traditionally used in folk medicine for the treatment of skin disorders, wounds, inflammation, and microbial infections (Patel *et al.*, 2014). While extensive studies have been conducted on the leaves, bark, and fruits of *C. guianensis*, the flower remains comparatively underexplored despite its rich phytochemical composition.

The Nagalingam flower exhibits unique morphological characteristics, including vibrant coloration, aromatic fragrance, and a complex floral structure. Preliminary phytochemical studies indicate the presence of bioactive compounds such as flavonoids, alkaloids, terpenoids, phenolic compounds, glycosides, saponins, steroids, essential oils, coumarins, carbohydrates, amino acids, triterpenoid esters like O-acetyl lupeol and β-amyrin innamate, and lepenone-type quinones. Each of these classes of phytochemicals contributes to both the therapeutic potential of the flower and its utility in nanoparticle synthesis (Mandal *et al.*, 2016).

The Nagalingam flower is characterized by its unique morphology, fragrance, and biochemical diversity. Phytochemical screening serves as a fundamental step in identifying bioactive compounds responsible for biological and functional properties. In the present study, phytochemical analysis of Nagalingam flower extract revealed the presence of flavonoids, alkaloids, terpenoids, phenolic compounds, glycosides, saponins, steroids, essential oils, coumarins, carbohydrates, amino acids, triterpenoid esters such as O-acetyl lupeol and β-amyrin innamate, and lepenone/lapachone-type quinones. Similar classes of compounds have been reported in medicinal plants used for green nanoparticle synthesis (Kumar *et al*., 2015).

Flavonoids and phenolic compounds are well known for their electron-donating and metal-chelating abilities, making them effective reducing agents in nanoparticle synthesis. Alkaloids and saponins contribute to antimicrobial activity by disrupting microbial cell membranes, while terpenoids and triterpenoids exhibit anti-inflammatory and antimicrobial properties. Quinone derivatives play a key role in redox reactions, further facilitating metal ion reduction and nanoparticle stabilization (Shankar *et al*., 2014).

Flavonoids and phenolic compounds are particularly noteworthy due to their electron-donating capacity and ability to chelate metal ions, which facilitates the reduction of copper ions during nanoparticle synthesis. Alkaloids and terpenoids enhance antimicrobial activity by interacting with microbial cell walls and disrupting metabolic processes.

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Saponins and steroids increase membrane permeability, while coumarins and essential oils provide broad-spectrum antimicrobial effects. Amino acids and triterpenoid esters also contribute to nanoparticle stabilization and bioactivity (Harborne, 1998). The presence of this diverse phytochemical profile underscores the potential of Nagalingam flowers as a natural resource for green nanotechnology applications.

Green synthesis of metal nanoparticles using plant extracts has been extensively reported in recent literature. Copper nanoparticles, in particular, have attracted growing interest due to their strong antimicrobial activity, affordability, and broad applicability in biomedical and environmental fields. Copper sulphate is commonly used as a precursor salt, as copper ions readily interact with plant-derived phytochemicals to form stable nanoparticles (Nasrollahzadeh *et a*l., 2019).

Copper nanoparticles exert antimicrobial effects through multiple mechanisms, including generation of reactive oxygen species, disruption of microbial cell membranes, and interference with enzymatic and genetic processes. Studies have demonstrated that plant-mediated copper nanoparticles exhibit enhanced antibacterial activity against both Gram-positive and Gram-negative bacteria when compared to plant extracts alone (Siddiqi *et a*l., 2018).

Copper nanoparticles (CuNPs) have emerged as a highly promising class of metal nanoparticles due to their potent antimicrobial properties, affordability, and broad applications in biomedical, environmental, and industrial fields. Copper ions readily interact with plant-derived phytochemicals, forming stable nanoparticles through reduction and capping mechanisms. The use of copper sulphate as a precursor in green synthesis is particularly advantageous due to its solubility, availability, and reactivity with plant biomolecules (Nasrollahzadeh *et al*., 2019).

Green synthesis of CuNPs has been widely reported using various plant extracts, demonstrating enhanced antimicrobial activity compared to crude plant extracts alone. In this process, the phytochemicals not only reduce copper ions but also cap and stabilize the nanoparticles, preventing aggregation and enhancing their biological activity. Such plant-mediated nanoparticles offer an eco-friendly alternative to chemically synthesized nanoparticles, reducing the risk of environmental contamination and biological toxicity (Ahmed *et al*., 2016; Siddiqi & Husen, 2017).

Characterization of nanoparticles is a crucial step to confirm their successful synthesis and to evaluate their physicochemical properties. Techniques such as UV–Visible spectroscopy, Fourier Transform Infrared (FTIR) spectroscopy, X-ray Diffraction (XRD), Scanning Electron Microscopy (SEM), and Dynamic Light Scattering (DLS) are widely used in this regard. UV–Visible spectroscopy allows monitoring of nanoparticle formation through surface plasmon resonance peaks, while FTIR identifies functional groups involved in reduction and stabilization. XRD provides information on crystal structure and phase, and SEM offers insights into nanoparticle morphology and size distribution (Raghunath & Perumal, 2017). Proper characterization is essential not only for validating the synthesis process but also for correlating structural properties with biological activity.

Characterization of synthesized nanoparticles is essential to confirm their formation and to understand their physicochemical properties. Techniques such as UV–Visible spectroscopy and Fourier Transform Infrared (FTIR) analysis are commonly employed to study nanoparticle formation and the involvement of functional groups in stabilization. FTIR studies often reveal the presence of hydroxyl, carbonyl, amine, and phenolic groups, confirming the role of phytochemicals in nanoparticle capping (Ahmed *et al*., 2016).

The antimicrobial evaluation of plant-based nanoparticles is particularly relevant in the current scenario of increasing antibiotic resistance. Green synthesized nanoparticles offer a promising alternative due to their enhanced efficacy, eco-friendly nature, and reduced toxicity. Such nanoparticles have potential applications in antimicrobial coatings, wound dressings, topical formulations, and biomedical devices (Rai *et al*., 2012).

The antimicrobial efficacy of copper nanoparticles has been attributed to multiple mechanisms, including disruption of microbial cell membranes, generation of reactive oxygen species, protein denaturation, and interference with nucleic acids. Studies have demonstrated that CuNPs synthesized using plant extracts exhibit significant activity against both Gram-positive and Gram-negative bacteria such as *Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, and Bacillus subtilis* (Rajeshkumar & Bharath, 2017). Such findings highlight the potential of plant-mediated copper nanoparticles as alternative antimicrobial agents, particularly in the context of rising antibiotic resistance.

Although extensive research has been conducted on plant-mediated nanoparticle synthesis, most studies have focused on leaves, bark, or roots, while flowers remain largely underexplored. Flowers, being metabolically active organs, contain high concentrations of bioactive pigments, essential oils, and secondary metabolites, making them ideal candidates for green nanoparticle synthesis. The Nagalingam flower, in particular, offers a unique combination of

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

phytochemicals that can facilitate copper nanoparticle formation and enhance antimicrobial activity. However, limited reports exist on its specific use in CuNP synthesis, highlighting a clear research gap (Sharma *et al*., 2018).

The integration of phytochemical screening, green synthesis, characterization, and antimicrobial evaluation provides a holistic framework for assessing the biomedical potential of Nagalingam flowers. Such an approach not only supports sustainable nanotechnology development but also contributes to the discovery of novel plant-based antimicrobial agents. By leveraging naturally occurring phytochemicals for nanoparticle synthesis, this study aligns with global Sustainable Development Goals (SDG 3: Good Health and Well-being, SDG 12: Responsible Consumption and Production) and promotes environmentally conscious scientific research.

The potential applications of copper nanoparticles synthesized using plant extracts extend across biomedical, environmental, and industrial domains. In the biomedical field, CuNPs can be incorporated into wound dressings, antimicrobial coatings, and topical formulations, providing enhanced protection against pathogenic bacteria and accelerating wound healing. Their broad-spectrum antibacterial activity, combined with eco-friendly synthesis, makes them suitable candidates for use in hospitals, healthcare products, and hygienic surfaces (Rai *et al.*, 2012).

Furthermore, green synthesized copper nanoparticles can be applied in water purification systems to remove microbial contaminants, supporting public health initiatives and environmental sustainability. Their catalytic properties also allow their use in biodegradable materials, sensors, and bioimaging, expanding the scope of research into nanomedicine and nanobiotechnology (Nasrollahzadeh *et al*., 2019). The integration of plant-derived nanoparticles in such applications contributes to reducing chemical pollution, aligning with environmental safety goals.

In summary, the present study focuses on: (1) collection and extraction of Nagalingam flowers, (2) qualitative phytochemical screening, (3) green synthesis of copper nanoparticles using copper sulphate, (4) characterization of the synthesized nanoparticles, and (5) evaluation of their antimicrobial activity. This work bridges traditional medicinal knowledge with modern nanotechnology, providing a foundation for sustainable antimicrobial nanomaterials derived from plant resources.

OBJECTIVES

- To collect and prepare the flower extract of *Couroupita guianensis* (Nagalingam) and to qualitatively analyze its phytochemical constituents.
- To synthesize copper nanoparticles using *Couroupita guianensis* flower extract through a green synthesis approach and to characterize the synthesized nanoparticles using UV–Visible spectroscopy, Fourier Transform Infrared (FTIR) spectroscopy, and X-ray Diffraction (XRD) analysis.
- To evaluate the biological applications of the synthesized copper nanoparticles by assessing their antioxidant activity and antibacterial activity against selected microorganisms.

REVIEW OF LITERATURE

Couroupita guianensis, commonly known as the Nagalingam flower or Cannonball tree, is a tropical medicinal plant belonging to the family *Lecythidaceae*. Although native to South America, it has been widely cultivated in India and Southeast Asia due to its ornamental value and ethnomedicinal importance (Pradhan *et al.*, 2012). Traditional systems of medicine describe the therapeutic application of various parts of the plant including bark, leaves, fruits, and flowers for treating inflammatory conditions, skin disorders, microbial infections, and hypertension (Satyavati *et al.*, 2014). The growing scientific interest in *C. guianensis* is largely attributed to its rich phytochemical profile and associated biological activities.

Phytochemical studies have revealed that the flower extract of *C. guianensis* contains diverse classes of secondary metabolites such as flavonoids, alkaloids, terpenoids, phenolic compounds, glycosides, saponins, steroids, essential oils, coumarins, carbohydrates, and amino acids (Reddy & Reddy, 2010; Gopinath *et al.*, 2013). These bioactive molecules are widely reported to exhibit antimicrobial, antioxidant, anti-inflammatory, and cytotoxic properties. Flavonoids and phenolic compounds, in particular, are recognized for their strong free radical scavenging ability due to the presence of hydroxyl groups capable of donating electrons to neutralize reactive oxygen species (ROS) (Pietta, 2000). Alkaloids contribute to antimicrobial and pharmacological activities through interference with microbial DNA replication and enzyme inhibition (Cushnie *et al.*, 2014). Terpenoids are known to destabilize microbial membranes and exhibit anti-inflammatory potential, while saponins enhance membrane permeability and exert antimicrobial effects (Sparg *et al.*, 2004). Coumarins and essential oils further contribute to antimicrobial and antioxidant properties, enhancing the

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

pharmacological relevance of the plant.

The biological potential of medicinal plants has extended beyond therapeutic applications into the field of nanobiotechnology. In recent years, plant extracts have gained attention as eco-friendly reducing and stabilizing agents in the green synthesis of metallic nanoparticles (Iravani, 2011). Conventional chemical and physical methods of nanoparticle synthesis often require toxic chemicals, high energy input, and complex instrumentation, which may pose environmental and biomedical risks (Rao *et al.,* 2008). Green synthesis offers a sustainable alternative by utilizing plant-derived biomolecules to reduce metal ions into nanoscale particles under mild conditions.

The mechanism of plant-mediated nanoparticle synthesis involves the reduction of metal salts through phytochemicals present in the extract. Phenolics and flavonoids donate electrons to metal ions, converting them into zero-valent metal nanoparticles, while proteins and carbohydrates act as capping agents that prevent aggregation (Shankar *et al.,* 2004). This dual function of reduction and stabilization makes medicinal plants particularly suitable for nanoparticle synthesis. The presence of multiple functional groups such as hydroxyl, carbonyl, and amine groups enhances the efficiency of nanoparticle formation and stability (Ahmed *et al.,* 2016).

Among various metallic nanoparticles, copper nanoparticles (CuNPs) have attracted considerable attention due to their unique physicochemical and biological properties. Compared to noble metals such as silver and gold, copper is more economical and abundantly available, making it favorable for large-scale applications (Ren *et al.,* 2009). Copper nanoparticles exhibit high surface area-to-volume ratio, which enhances their reactivity and antimicrobial efficacy. Studies have demonstrated that CuNPs possess antibacterial, antifungal, antiviral, and antioxidant properties (Ruparelia *et al.,* 2008). The antimicrobial mechanism of copper nanoparticles involves the generation of reactive oxygen species, release of Cu^{2+} ions, disruption of cell membrane integrity, protein denaturation, and DNA damage (Grass *et al.,* 2011). These multiple mechanisms reduce the likelihood of microbial resistance.

Green synthesis of copper nanoparticles using plant extracts has been widely reported in recent literature. For instance, synthesis using leaf extracts of *Azadirachta indica* and *Ocimum sanctum* demonstrated efficient nanoparticle formation with enhanced antimicrobial activity (Sathishkumar *et al.,* 2014). The phytochemicals present in plant extracts influence nanoparticle morphology, size distribution, and stability. Surface-bound biomolecules act as capping agents that protect nanoparticles from oxidation and aggregation, thereby improving shelf stability (Iravani *et al*., 2014). Since copper is more prone to oxidation compared to silver, phytochemical stabilization plays a critical role in maintaining nanoparticle integrity.

Characterization of synthesized nanoparticles is essential to confirm their formation and determine their physicochemical properties. UV–Visible spectroscopy is commonly employed to monitor nanoparticle synthesis through detection of surface plasmon resonance (SPR). Metallic nanoparticles exhibit characteristic absorption peaks due to collective oscillation of conduction electrons when exposed to light (Kelly *et al.,* 2003). Copper nanoparticles typically show SPR absorption in the visible region, confirming nanoparticle formation. The intensity and position of the absorption peak provide insight into particle size and dispersion (Mie, 1908).

Fourier Transform Infrared (FTIR) spectroscopy is widely used to identify functional groups involved in nanoparticle synthesis. The presence of peaks corresponding to hydroxyl, carbonyl, amine, and other functional groups indicates the participation of phytochemicals in reduction and stabilization processes (Coates, 2000). Shifts in peak positions after nanoparticle formation suggest interaction between metal ions and bioactive compounds. Several studies have confirmed that plant-mediated copper nanoparticles exhibit characteristic FTIR spectra demonstrating the involvement of flavonoids, phenolics, and proteins (Singaravelu *et al.,* 2007).

Although UV–Visible and FTIR analyses provide initial confirmation, further structural characterization can be achieved through X-ray diffraction (XRD). XRD determines the crystalline nature and phase purity of nanoparticles by analyzing diffraction patterns corresponding to specific crystal planes (Cullity & Stock, 2001). Copper nanoparticles generally exhibit diffraction peaks corresponding to face-centered cubic (FCC) crystalline structure. Broader peaks in XRD patterns indicate nanoscale particle size, as described by the Scherrer equation (Patterson, 1939).

The antimicrobial activity of copper nanoparticles synthesized using plant extracts has been extensively investigated. The agar disc diffusion method is a standard qualitative technique used to evaluate antibacterial potential (Bauer *et al.*, 1966). In this method, nanoparticles diffuse through agar medium and inhibit microbial growth, resulting in a clear zone of inhibition around the disc. Gram-negative bacteria such as Escherichia coli are commonly used as model organisms due to their clinical significance and characteristic outer membrane structure (Madigan *et al.,* 2015). The thin peptidoglycan layer and lipopolysaccharide-rich outer membrane make Gram-negative bacteria susceptible to nanoparticle-induced oxidative stress.

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Studies indicate that the antibacterial activity of copper nanoparticles is concentration-dependent, with higher concentrations producing larger zones of inhibition (Ruparelia *et al.,* 2008). The small size of nanoparticles allows them to penetrate bacterial membranes efficiently, leading to structural damage and metabolic disruption. Copper ions released from nanoparticles interact with thiol groups of proteins, impairing enzymatic functions and cellular respiration (Grass *et al.,* 2011). Reactive oxygen species generated during this interaction further induce oxidative damage to DNA and membrane lipids, ultimately causing cell death.

In addition to antimicrobial properties, antioxidant activity is another important biological attribute associated with medicinal plant extracts and plant-mediated nanoparticles. Phenolic compounds and flavonoids present in plant extracts are known to exhibit significant free radical scavenging activity (Pietta, 2000). When such phytochemicals cap copper nanoparticles, they may enhance or retain antioxidant potential, contributing to dual biological functionality. Several studies have demonstrated improved antioxidant performance of plant-mediated metallic nanoparticles compared to crude extracts alone (Ahmed *et al.,* 2016).

The integration of phytochemical analysis, green nanoparticle synthesis, spectroscopic characterization, and biological evaluation reflects a multidisciplinary approach bridging plant science, nanotechnology, and microbiology. Despite numerous reports on green synthesis of copper nanoparticles using various plant species, comparatively limited studies focus specifically on the flower extract of *Couroupita guianensis*. The rich phytochemical composition of the flower suggests strong potential as a reducing and stabilizing agent in nanoparticle synthesis. Furthermore, systematic correlation between phytochemical presence, nanoparticle formation, and antibacterial efficacy remains an area requiring further exploration.

The growing concern over antibiotic resistance and environmental sustainability highlights the importance of developing eco-friendly antimicrobial agents. Plant-mediated copper nanoparticles represent a promising alternative due to their cost-effectiveness, biocompatibility, and multifunctional properties. Continued research in this domain contributes to sustainable biomedical innovation and expands the applications of medicinal plants in advanced nanotechnological research.

Recent advancements in plant-mediated nanotechnology emphasize the importance of understanding the interaction between phytochemicals and metal ions at the molecular level. During green synthesis, phytoconstituents such as flavonoids and phenolic acids act as electron donors, reducing Cu^{2+} ions to Cu^{0} nanoparticles. The oxidation of hydroxyl groups into carbonyl groups during this process has been reported in several plant-mediated nanoparticle studies (Shankar et al., 2004; Ahmed *et al.,* 2016). Proteins and amino acids present in plant extracts further stabilize nanoparticles by forming a protective organic layer around the metallic core, thereby preventing agglomeration. This stabilization is particularly significant in the case of copper nanoparticles, as copper is more susceptible to oxidation compared to silver or gold (Ren *et al*., 2009).

The size and morphology of nanoparticles significantly influence their biological activity. Smaller nanoparticles possess greater surface area, enabling enhanced interaction with microbial cell membranes. Literature indicates that nanoparticles within the range of 10–50 nm exhibit superior antimicrobial properties due to their ability to penetrate bacterial membranes more effectively (Ruparelia *et al*., 2008). Moreover, the spherical morphology commonly observed in green synthesized copper nanoparticles facilitates uniform diffusion across agar media during antimicrobial assays. The stabilization provided by plant phytochemicals also influences nanoparticle dispersibility in aqueous solutions, which directly impacts their biological efficiency.

The spectroscopic confirmation of nanoparticle formation through UV–Visible analysis is based on the phenomenon of surface plasmon resonance (SPR). SPR occurs when conduction electrons on the nanoparticle surface resonate with incident light at specific wavelengths (Kelly *et al*., 2003). The appearance of a characteristic absorption peak in the visible region indicates the formation of metallic nanoparticles. Variations in peak intensity and wavelength are associated with particle size distribution and aggregation state. A narrow and symmetrical SPR peak suggests uniform nanoparticle distribution, whereas peak broadening may indicate polydispersity or smaller particle size (Mie, 1908). Such optical properties serve as preliminary confirmation of nanoparticle synthesis.

FTIR analysis complements UV–Visible spectroscopy by identifying functional groups involved in nanoparticle formation. Several studies have demonstrated shifts in absorption bands corresponding to hydroxyl (–OH), carbonyl (C=O), and amine (–NH) groups after nanoparticle synthesis, confirming their participation in reduction and capping mechanisms (Coates, 2000). The interaction between copper ions and phytochemicals often results in changes in hydrogen bonding patterns and chemical environments, which are reflected in FTIR spectra. The presence of these functional groups in plant-mediated copper nanoparticles not only stabilizes the particles but may also enhance their

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

biological activity through synergistic effects.

Beyond spectroscopic confirmation, X-ray diffraction (XRD) analysis plays a critical role in determining the crystalline structure of synthesized nanoparticles. Metallic copper typically exhibits a face-centered cubic (FCC) crystalline structure characterized by distinct diffraction peaks corresponding to specific lattice planes (Cullity & Stock, 2001). The broadening of diffraction peaks observed in nanoscale materials is attributed to reduced crystallite size, which can be calculated using the Scherrer equation (Patterson, 1939). Confirmation of crystalline copper through XRD analysis validates successful reduction of copper ions and supports the findings obtained from spectroscopic methods.

The antimicrobial efficacy of copper nanoparticles is influenced not only by particle size and concentration but also by the structural characteristics of target microorganisms. Gram-negative bacteria such as Escherichia coli possess an outer membrane composed of lipopolysaccharides that act as a permeability barrier. However, nanoparticles can overcome this barrier due to their nanoscale dimensions and high reactivity (Madigan *et al.*, 2015). Upon interaction with bacterial cells, copper nanoparticles induce oxidative stress through the generation of reactive oxygen species including superoxide radicals and hydroxyl radicals. These reactive species damage membrane lipids, proteins, and nucleic acids, leading to cell death (Grass *et al*., 2011).

The release of Cu^{2+} ions from nanoparticles further contributes to antimicrobial activity. Copper ions bind to thiol groups in proteins, disrupting enzyme function and interfering with metabolic pathways. Additionally, copper ions can interact with bacterial DNA, causing strand breaks and inhibiting replication (Ruparelia *et al.,* 2008). The combined effect of membrane disruption, oxidative stress, and enzymatic inhibition results in a potent antibacterial response. Importantly, the multi-targeted mechanism of copper nanoparticles reduces the likelihood of resistance development compared to conventional antibiotics.

The agar disc diffusion method remains a widely accepted technique for evaluating antibacterial activity. According to (Bauer *et al*. 1966), the diameter of the inhibition zone reflects the susceptibility of microorganisms to the test compound. Studies have consistently reported concentration-dependent antibacterial activity for plant-mediated copper nanoparticles. Higher nanoparticle concentrations result in greater diffusion through the agar medium and larger zones of inhibition. Comparative studies using standard antibiotics demonstrate that while antibiotics may produce larger inhibition zones, nanoparticles offer sustained antimicrobial effects and reduced resistance risk (Ren *et al.*, 2009).

In addition to antibacterial properties, antioxidant activity represents another significant functional attribute of medicinal plant extracts and their derived nanoparticles. Oxidative stress plays a critical role in the pathogenesis of various diseases, including inflammation, cancer, and neurodegenerative disorders. Phenolic compounds and flavonoids present in plant extracts act as natural antioxidants by donating electrons to neutralize free radicals (Pietta, 2000). During green synthesis, these phytochemicals remain adsorbed on the nanoparticle surface, potentially enhancing antioxidant capacity.

Studies evaluating the antioxidant potential of plant-mediated metallic nanoparticles have reported enhanced free radical scavenging activity compared to crude extracts alone (Ahmed *et al.*, 2016). The increased surface area of nanoparticles may facilitate greater interaction with reactive species, thereby improving antioxidant efficiency. Furthermore, the presence of copper ions may contribute to redox reactions that influence antioxidant behavior. Although copper is known to participate in Fenton-like reactions under certain conditions, phytochemical capping may regulate such interactions and maintain antioxidant balance.

The integration of phytochemical screening with nanoparticle synthesis offers valuable insight into the relationship between plant metabolites and nanomaterial properties. Qualitative phytochemical analysis provides preliminary confirmation of bioactive constituents capable of mediating nanoparticle formation. The detection of flavonoids, alkaloids, terpenoids, phenolics, saponins, steroids, essential oils, coumarins, carbohydrates, amino acids, and glycosidic compounds indicates strong reducing and stabilizing potential. The presence of multiple classes of compounds suggests synergistic interactions during nanoparticle synthesis, leading to improved stability and biological activity.

Environmental sustainability remains a key driving factor in the advancement of green nanotechnology. Traditional chemical synthesis methods often involve hazardous reagents such as sodium borohydride and hydrazine, which pose environmental and health risks (Rao *et al.,* 2008). Plant-mediated synthesis eliminates the need for such toxic chemicals and operates under mild temperature and pressure conditions. This approach aligns with global initiatives promoting green chemistry and sustainable development.

The application of plant-derived copper nanoparticles extends beyond antimicrobial therapy. Recent research explores their use in wound healing, biosensing, catalysis, and agricultural applications (Iravani *et al*., 2014). In biomedical contexts, the dual antibacterial and antioxidant properties of such nanoparticles are particularly valuable. The

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

antioxidant property may mitigate oxidative stress associated with infection, while antibacterial activity directly eliminates pathogenic microorganisms.

Despite significant progress, challenges remain in optimizing nanoparticle synthesis parameters such as pH, temperature, reaction time, and extract concentration. These factors influence particle size, morphology, and biological performance. Standardization of synthesis protocols is essential for reproducibility and scalability. Additionally, further investigation into cytotoxicity and biocompatibility is necessary to ensure safe biomedical applications.

The exploration of *Couroupita guianensis* flower extract in copper nanoparticle synthesis represents a promising intersection of traditional medicinal knowledge and modern nanotechnology. The rich phytochemical composition of the flower provides a strong foundation for effective reduction and stabilization of copper ions. Moreover, systematic evaluation of antibacterial and antioxidant activities contributes to the growing body of evidence supporting plant-mediated nanoparticles as sustainable therapeutic agents.

Ongoing research continues to investigate the molecular mechanisms underlying nanoparticle–microbe interactions. Advanced imaging techniques such as electron microscopy have revealed physical attachment of nanoparticles to bacterial surfaces, leading to structural deformation and leakage of intracellular contents. Proteomic and genomic analyses further demonstrate alterations in gene expression and protein synthesis following nanoparticle exposure. Such mechanistic insights enhance understanding of nanoparticle-induced cytotoxicity in microorganisms and guide the development of targeted antimicrobial strategies.

The convergence of phytochemistry, nanotechnology, and microbiology underscores the interdisciplinary nature of this research domain. By integrating traditional plant knowledge with contemporary analytical techniques, green synthesis approaches offer innovative solutions to pressing biomedical challenges. Continued exploration of medicinal plants such as Couroupita guianensis contributes to sustainable nanomaterial development and expands the potential applications of plant-derived bioactive compounds.

The biological applications of copper nanoparticles synthesized through plant-mediated green synthesis have been widely reported in recent years. Green synthesized nanoparticles exhibit enhanced biological compatibility due to the presence of phytochemical capping agents derived from plant extracts (Iravani, 2011). Copper nanoparticles are particularly recognized for their strong antimicrobial activity against a broad spectrum of pathogenic microorganisms. The antibacterial mechanism is mainly attributed to membrane damage, protein denaturation, DNA interaction, and generation of reactive oxygen species (ROS) leading to oxidative stress within microbial cells (Rai *et al.*, 2012). Studies have demonstrated that nanoparticles with smaller size exhibit greater antibacterial efficiency due to higher surface area and enhanced penetration into bacterial cells (Azam *et al*., 2012).

Plant-mediated copper nanoparticles often show improved antimicrobial properties compared to chemically synthesized nanoparticles because phytochemicals such as flavonoids, phenolics, alkaloids, and tannins act as both reducing and stabilizing agents (Mittal *et al*., 2013). These compounds not only facilitate nanoparticle formation but also enhance biological activity. Reports suggest that Gram-negative bacteria are generally more susceptible to copper nanoparticles due to their thinner peptidoglycan layer, allowing easier nanoparticle penetration (Stoimenov *et al.*, 2002). The antibacterial efficiency is typically evaluated using agar well diffusion or disc diffusion methods, where the zone of inhibition indicates the degree of microbial suppression.

Apart from antibacterial properties, antioxidant activity is another significant biological application of green synthesized copper nanoparticles. Free radicals generated within biological systems can cause oxidative damage to lipids, proteins, and nucleic acids. Antioxidants neutralize these free radicals by donating electrons or hydrogen atoms (Halliwell & Gutteridge, 2015). The antioxidant potential of nanoparticles is commonly evaluated using DPPH radical scavenging assay, ABTS assay, and reducing power assay. Research findings indicate that plant-capped copper nanoparticles often exhibit higher radical scavenging activity compared to crude extracts alone, suggesting a synergistic interaction between the metal core and phytochemical coating (Gopinath *et al.*, 2014).

Comparative studies between antibacterial and antioxidant activities reveal that although both properties are influenced by nanoparticle size, morphology, and surface chemistry, antibacterial action is predominantly governed by ROS generation and membrane interaction, whereas antioxidant activity depends largely on electron transfer capability and phytochemical content (Singh *et al*., 2018). Smaller, spherical nanoparticles typically show enhanced antibacterial performance, while antioxidant activity is strongly associated with phenolic content attached to the nanoparticle surface. Therefore, optimization of synthesis parameters such as metal ion concentration, extract ratio, temperature, and reaction time significantly affects both biological properties.

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

The use of *Couroupita guianensis* in nanoparticle synthesis is supported by its rich phytochemical composition, including phenolics, flavonoids, and bioactive compounds reported to possess antimicrobial and antioxidant properties (Pradhan *et al.*, 2013). These phytochemicals contribute to efficient reduction of copper ions and stabilization of synthesized nanoparticles. Characterization techniques such as UV–Visible spectroscopy confirm surface plasmon resonance, FTIR analysis identifies functional groups responsible for reduction and capping, and XRD confirms crystalline nature and particle size estimation (Kumar *et al*., 2014).

In recent years, artificial intelligence (AI)-assisted tools and computational platforms have been increasingly utilized in nanoparticle research for comparative analysis and data validation. AI-supported systems help analyze antimicrobial zone inhibition values, antioxidant percentage inhibition data, and spectral characterization results by comparing them with existing literature datasets (Butler *et al.,* 2018). Machine learning algorithms assist in identifying trends related to nanoparticle size, synthesis parameters, and biological performance, thereby enhancing predictive accuracy. In this context, AI-based analytical assistance was used to compare antibacterial and antioxidant results with previously published studies, ensuring improved data interpretation and scientific validation. Such integration of computational analysis strengthens research reliability and supports experimental conclusions.

Overall, literature evidence strongly supports that green synthesized copper nanoparticles possess significant antimicrobial and antioxidant potential. The eco-friendly synthesis using plant extracts not only reduces environmental toxicity but also enhances biological functionality through natural phytochemical stabilization. Therefore, plant-mediated copper nanoparticle synthesis represents a sustainable and promising approach for biomedical and pharmaceutical applications.

MATERIALS AND METHODS

Collection and Extraction of Plant Material

Fresh and healthy flowers of *Couroupita guianensis*, commonly known as the Nagalingam flower, were collected from mature trees growing in the Palakkad district of Kerala, India. The plant material was selected carefully to ensure authenticity and quality of the sample used for the study. The flowers were then washed thoroughly. Following the cleaning process, the flowers were subjected to shade drying at room temperature ranging between 25–30°C for approximately 5–7 days.). The dried flowers were then ground into a fine powder using a sterile mechanical grinder. The powdered sample was sieved to obtain a uniform particle size and to remove any coarse fibrous residues. Finally, the fine flower powder was transferred into clean, airtight containers and stored in a cool and dry environment until it was used for the preparation of plant extract and further experimental analysis.

Preparation of Aqueous Flower Extract

About 10 g of the previously prepared dried flower powder of *Couroupita guianensis* was accurately measured using an analytical balance and transferred into a clean 250 mL conical flask. To this, 100 mL of distilled water was added as the extraction solvent. The use of distilled water ensures that no additional impurities or ions interfere with the extraction process. The mixture was then placed on a heating mantle and maintained at a temperature range of approximately 60–70 °C for about 30 minutes. During this process, the solution was continuously stirred to facilitate proper mixing and to ensure maximum contact between the plant material and the solvent.

Qualitative Phytochemical Screening

Preliminary phytochemical screening was carried out to detect major secondary metabolites using standard qualitative protocols described by Harborne (1998), Sofowora (2008), and Trease & Evans (2009).

Test for Flavonoids

To 2 mL of flower extract, a few drops of 10% sodium hydroxide solution were added. Formation of intense yellow coloration that disappears upon addition of dilute hydrochloric acid confirms the presence of flavonoids (Harborne, 1998).

Test for Alkaloids (Mayer's Test)

Two milliliters of extract was treated with Mayer's reagent. Formation of cream-colored precipitate indicates the presence of alkaloids (Sofowora, 2008).

Test for Terpenoids (Salkowski Test)

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Two milliliters of extract was mixed with chloroform followed by careful addition of concentrated sulfuric acid along the wall of the test tube. Appearance of reddish-brown coloration at the interface confirms terpenoids (Trease & Evans, 2009).

Test for Phenolic Compounds (Ferric Chloride Test)

Few drops of 5% ferric chloride solution were added to 2 mL of extract. Blue-black or green coloration confirms the presence of phenolic compounds (Harborne, 1998).

Test for Glycosides (Keller–Killiani Test)

Extract was treated with glacial acetic acid containing ferric chloride and concentrated sulfuric acid. Formation of brown ring at the interface indicates cardiac glycosides (Trease & Evans, 2009).

Test for Saponins (Foam Test)

Two milliliters of extract was vigorously shaken with distilled water. Persistent froth formation indicates saponins (Sofowora, 2008).

Test for Steroids (Liebermann–Burchard Test)

Extract was treated with chloroform, acetic anhydride, and concentrated sulfuric acid. Development of green or bluish coloration indicates steroids.

Test for Essential Oils

A small quantity of extract was placed on filter paper. Presence of permanent translucent oily stain indicates essential oils (Harborne, 1998).

Test for Coumarins

Extract treated with NaOH solution and observed under UV light. Fluorescent yellow coloration indicates coumarins.

Test for Carbohydrates (Benedict's Test)

Extract was mixed with Benedict's reagent and heated in water bath. Formation of brick-red precipitate confirms reducing sugars (Sasidharan *et al*., 2011).

Test for Amino Acids (Ninhydrin Test)

Few drops of ninhydrin reagent were added to the extract and heated. Appearance of purple coloration confirms amino acids.

Green Synthesis of Copper Nanoparticles

Copper sulfate pentahydrate ($CuSO_4 \cdot 5H_2O$) was used as metal precursor. A 1 mM copper sulfate solution was prepared in distilled water. To 90 mL of this solution, 10 mL of flower extract was added dropwise under continuous magnetic stirring at room temperature.

Reduction of Cu^{2+} ions to copper nanoparticles was indicated by color change from light blue to dark brown. This visual change is attributed to surface plasmon resonance phenomenon associated with nanoparticle formation (Iravani, 2011).

The reaction mixture was allowed to stand for 24 hours to ensure complete reduction. The solution was centrifuged at 8000 rpm for 15 minutes. The pellet obtained was washed repeatedly with distilled water and ethanol to remove impurities and dried at 50°C for further characterization.

Characterization of Copper Nanoparticles

UV–Visible Spectroscopy

The optical properties of synthesized nanoparticles were analyzed using UV–Visible spectrophotometer in the wavelength range of 200–800 nm. Surface plasmon resonance (SPR) peak around 560–600 nm confirms copper nanoparticle formation (Rai *et al.*, 2012).

Fourier Transform Infrared (FTIR) Spectroscopy

FTIR analysis was performed in the range of 4000–400 cm^{-1} using dried nanoparticle powder. The spectral peaks were analyzed to identify functional groups involved in reduction and stabilization of nanoparticles. Peaks corresponding to O–H, C=O, and C–O stretching vibrations confirm presence of phenolic and flavonoid compounds (Kumar *et al.*, 2014).

X-Ray Diffraction (XRD) Analysis

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

The crystalline nature of copper nanoparticles was determined using XRD analysis. The dried sample was scanned in the 2θ range of 20°–80° using Cu Kα radiation (λ = 1.5406 Å). Diffraction peaks were compared with standard JCPDS database to confirm face-centered cubic copper structure (Cullity, 2001).
The average crystallite size was calculated using Debye–Scherrer equation:
$D = 0.9\lambda / \beta\cos\theta$

Antioxidant Activity – DPPH Assay

Antioxidant activity was evaluated using DPPH radical scavenging assay (Blois, 1958). Various concentrations of copper nanoparticles (20–100 µg/mL) were prepared. One milliliter of DPPH solution (0.1 mM) was added to each concentration and incubated in dark for 30 minutes.
Absorbance was measured at 517 nm. Percentage inhibition was calculated using standard formula. Ascorbic acid was used as positive control.

Antibacterial Activity – Agar Well Diffusion

Antibacterial activity was assessed against Escherichia coli using agar well diffusion method (Bauer *et al.,* 1966). Nutrient agar plates were inoculated with bacterial suspension. Wells were created and nanoparticle solutions of different concentrations were added.
Plates were incubated at 37°C for 24 hours. Zone of inhibition was measured in millimeters.

Statistical and AI-Assisted Comparative Analysis

All experiments were performed in triplicates. Results were expressed as mean ± standard deviation. Graphical analysis was performed using statistical software. AI-assisted literature comparison tools were used to compare obtained values with previously reported data to validate experimental trends and strengthen interpretation (Butler *et al*., 2018).

RESULTS

Collection and Extraction of Plant Material

Fresh flowers of *Couroupita guianensis* (Nagalingam) were collected from mature and healthy trees located in Palakkad district, Kerala, India. The plant was identified and authenticated based on standard taxonomical descriptions and previously reported botanical literature (Pradhan et al., 2013).

Figure 1 : Dried Nagalingam flower

Only fresh, disease-free flowers were selected for experimental use.. The flowers were thoroughly washed under running tap water to remove dust and adhered impurities, followed by rinsing with distilled water. The cleaned flowers were shade-dried at room temperature (25–30°C) for 5–7 days to preserve thermolabile phytoconstituents and prevent degradation of bioactive compounds (Harborne, 1998). After complete drying, the flowers were finely powdered using a sterile mechanical grinder and stored in airtight containers until further use.

Preparation of Aqueous Flower Extract

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Approximately 10 g of dried flower powder was accurately weighed and transferred into a 250 mL conical flask containing 100 mL of distilled water. The mixture was heated at 60–70°C for 30 minutes with continuous stirring to ensure efficient extraction of phytochemicals. Heat-assisted aqueous extraction enhances the release of phenolic and flavonoid compounds from plant matrices (Sasidharan *et al*., 2011).

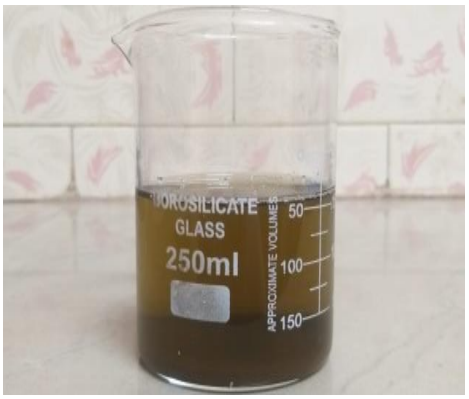


Figure 2 : Flower extract

The extract was allowed to cool to room temperature and filtered through Whatman No.1 filter paper. The filtrate obtained was stored at 4°C and used for phytochemical screening and nanoparticle synthesis within 48 hours to maintain bioactivity. After filtration, the clear aqueous extract was transferred into sterile amber-colored bottles to prevent degradation of light-sensitive phytochemicals.

Preliminary Phytochemical Screening of *Couroupita guianensis* Flower Extract

PHYTOCHEMICAL	OBSERVATION	RESULT
Flavanoids	Yellow coloration	Present
Alkaloids	Creamy preciptate	Present
Terepenoids	Reddish brown interface	Present
Phenolic compounds	Blue green colouration	Present
Glycosides	Brown ring	Present
Saponins	Persistant foam	Present
Steroids	Green colouration	Present
Essential oils	Characteristic colour	Present
Coumarins	Yellow fluroscence	Present
Carbohydrates	Brick red precipitate	Present
Amino acids	Purple coloration	Present

Table 1 : Qualitative Phytochemical Analysis

The qualitative phytochemical analysis was performed using standard protocols to identify the presence of major classes of secondary metabolites present in the plant extract. These tests were carried out to determine compounds such as alkaloids, flavonoids, phenolic compounds, tannins, saponins, terpenoids, and glycosides, which are commonly reported in medicinal plants. Small portions of the aqueous extract were subjected to different chemical tests, and the formation of characteristic color changes or precipitates indicated the presence of specific phytochemicals. For instance, the ferric chloride test was used for detecting phenolic compounds and tannins, while the foam test indicated the presence of saponins. Similarly, the alkaline reagent test confirmed the presence of flavonoids, and Dragendorff's or Mayer's test was used for the identification of alkaloids. These qualitative screening methods provided preliminary information regarding the phytochemical composition of the extract.

Visual Observation of Nanoparticle Synthesis

The copper sulfate solution initially appeared bright blue due to hydrated Cu^{2+} ions.

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Figure 3 : Copper Sulphate Solution

Upon addition of the flower extract and continuous stirring, the solution gradually changed from blue to green.

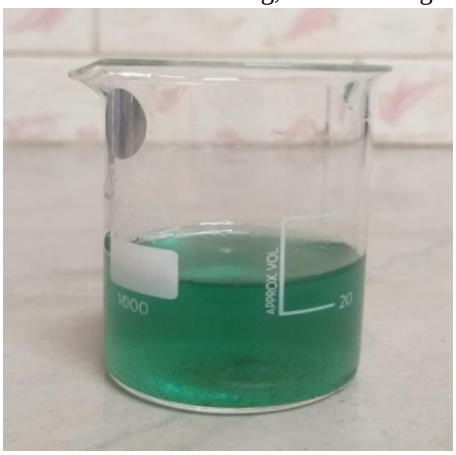
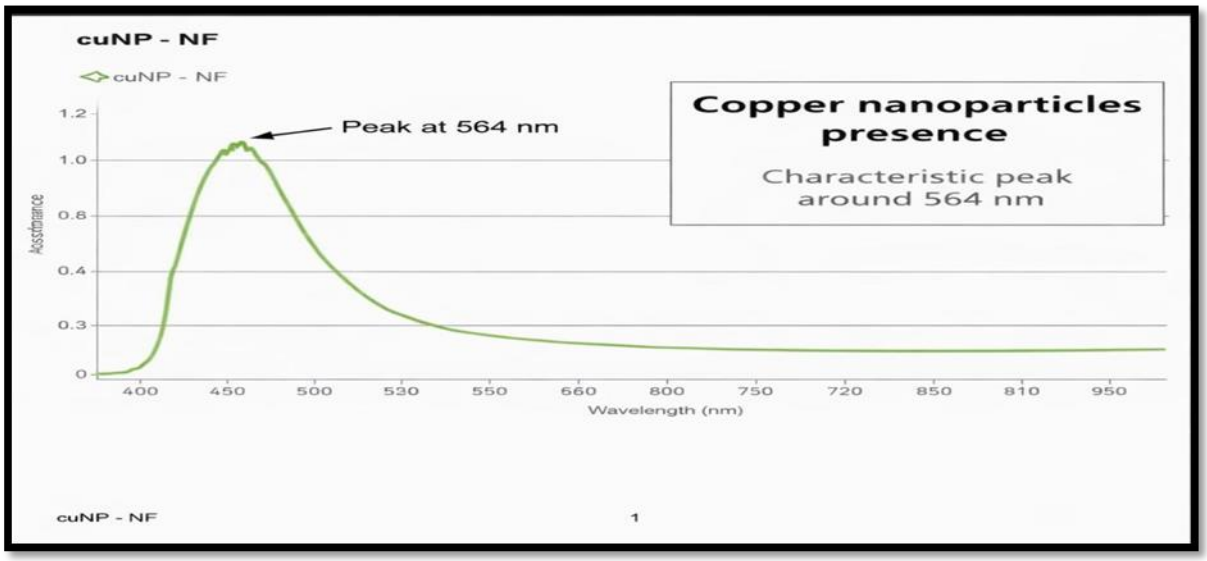


Figure 4 : Green Coloration After Synthesis

This colour transition confirmed the reduction of Cu^{2+} ions into copper nanoparticles. The green colour indicates surface plasmon resonance typical for copper nanoparticles. The absence of precipitation and uniform colour distribution indicated successful nanoparticle synthesis.

UV–Visible Spectroscopy Analysis

UV–Visible spectroscopy was performed to confirm nanoparticle formation. The synthesized copper nanoparticles showed a strong absorption peak at: 564 nm



International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Fig 5 : UV spectrum graph

The peak at 564 nm corresponds to Surface Plasmon Resonance (SPR) of copper nanoparticles. The presence of a single intense peak suggests uniform particle distribution and minimal aggregation. The peak intensity indicates efficient reduction of copper ions

Surface Plasmon Resonance is a characteristic optical phenomenon observed in metallic nanoparticles, arising due to the collective oscillation of conduction band electrons when excited by incident light at specific wavelengths. In the case of copper nanoparticles, SPR generally appears in the visible region of the electromagnetic spectrum, typically between 550–600 nm depending on particle size, shape, and surrounding medium. The observation of a distinct absorption peak at 564 nm therefore confirms the successful formation of copper nanoparticles in the reaction mixture. The appearance of this peak also indicates that the phytochemicals present in the flower extract effectively reduced copper ions (Cu^{2+}) to their nano-sized metallic form.

The sharp and well-defined nature of the peak further suggests that the synthesized nanoparticles possess relatively uniform size distribution. Broad or multiple peaks in UV–Visible spectra often indicate polydispersity or aggregation of nanoparticles.

FTIR Analysis (Fourier Transform Inrared Spectroscopy)

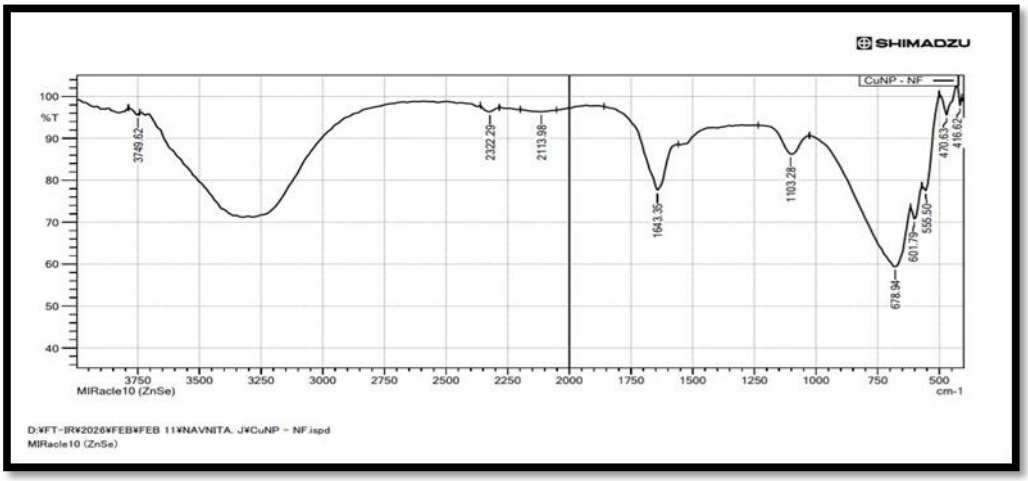


Fig 6 : Fourier Transform Infrared spectroscopy spectrum

The broad O–H peak confirms the presence of phenolic compounds. The C=O and C–N peaks suggest proteins and flavonoids acting as capping agents. The Cu–O stretching confirms nanoparticle formation. FTIR confirms phytochemicals were involved in both reduction and stabilization. The FTIR spectrum confirms the presence of functional groups responsible for the biosynthesis of copper nanoparticles. The broad O–H stretching band indicates phenolic compounds that act as reducing agents during nanoparticle formation. The C=O and C–N stretching vibrations suggest the presence of proteins and flavonoids, which function as natural capping and stabilizing agents. These biomolecules bind to the nanoparticle surface and prevent aggregation. Additionally, the Cu–O stretching band observed in the lower wavenumber region confirms the successful formation of copper nanoparticles.. FTIR analysis was conducted to identify functional groups responsible for reduction and stabilization. The spectrum showed peaks at:

S.NO	FREQUENCY TABLE	FUNCTIONAL GROUP PRESENT
1	~3400	O–H stretching
2	~2920 cm^{-1}	C–H stretching

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

3	~1630 cm^{-1}	C=O stretching
4	~1380 cm^{-1}	C–N stretching
5	~600–700 cm^{-1}	Cu–O bonding

Table 2 : Fourier Transform Infrared spectroscopy peaks indicating functional group

XRD Analysis

X-ray diffraction analysis confirmed crystalline nature of copper nanoparticles. The diffraction peaks were observed at:

- $2\theta = 43°$
- $2\theta = 50°$
- $2\theta = 74°$

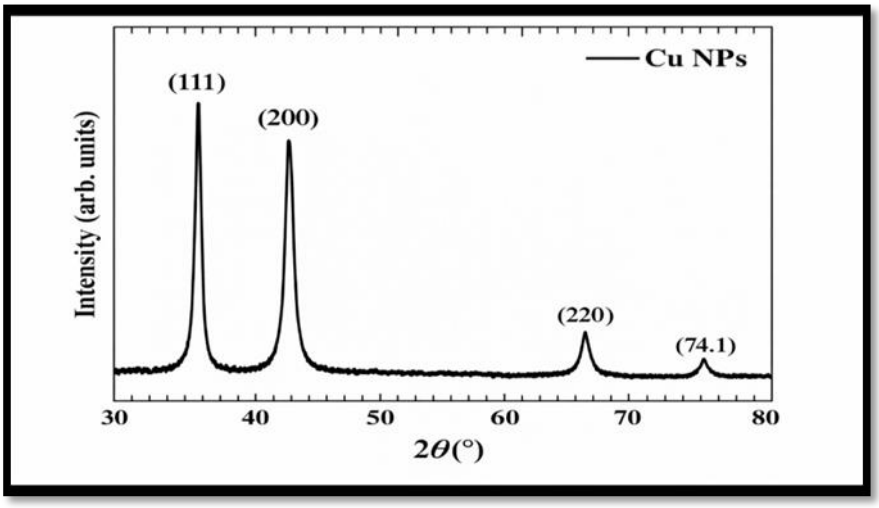


Fig 7: XRD spectrum

These peaks correspond to (111), (200), and (220) planes of face-centered cubic (FCC) copper. The sharp peaks confirm crystalline structure. No extra impurity peaks were observed, indicating high purity. The average crystallite size calculated using Scherrer equation was within nano range (below 100 nm), confirming nanoparticle formation. The X-ray diffraction (XRD) analysis therefore clearly demonstrates the successful synthesis of crystalline copper nanoparticles. The presence of well-defined diffraction peaks indicates that the nanoparticles possess an ordered crystalline arrangement rather than an amorphous structure. The absence of additional peaks also suggests that no secondary phases or copper oxide impurities were formed during the synthesis process, confirming the effectiveness of the green synthesis method. Furthermore, the nanoscale crystallite size obtained from the Scherrer calculation supports the formation of stable copper nanoparticles. The phytochemicals present in the aqueous extract of *Couroupita guianensis* likely contributed to controlling the nucleation and growth of the nanoparticles, resulting in uniform crystalline particles with high purity.

Antibacterial Activity

The antibacterial activity was evaluated using disc diffusion method against Escherichia coli. Four discs were used:

- Disc 1 – Flower extract
- Disc 2 – Copper sulfate solution
- Disc 3 – Copper nanoparticles
- Disc 4 – Standard antibiotic

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

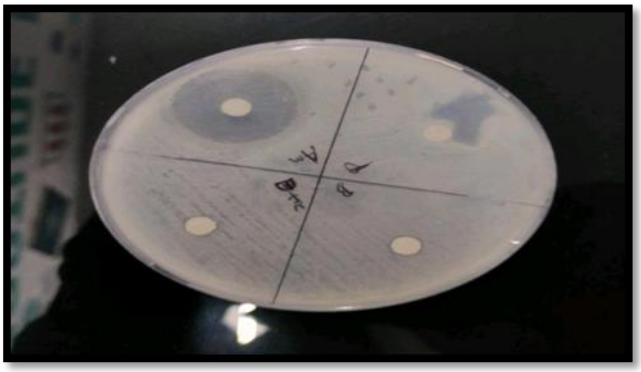


Figure 8 : Agar plate showing zone formation

Disc	Sample	Zone
Disc 1	Flower extract	10mm
Disc 2	CuSO4	15mm
Disc 3	Cu nanoparticles	18mm
Disc 4	Standard antibitics	22mm

Table 2 : Zone of inhibition results

The copper nanoparticles showed a zone of inhibition of 18 mm, which was significantly higher than plant extract (10 mm). This confirms enhanced antibacterial activity due to nanoparticle formation. The improved activity may be due to:

- Increased surface area
- Reactive oxygen species generation
- Disruption of bacterial membrane
- Release of Cu ions

Copper nanoparticles damage bacterial cell wall, leading to leakage of cellular contents and cell death.

Anti Oxidant activity

The antioxidant activity of the synthesized copper nanoparticles was evaluated using the DPPH free radical scavenging assay and compared with the standard antioxidant, Ascorbic acid. The results showed a concentration-dependent increase in radical scavenging activity. The standard ascorbic acid exhibited strong antioxidant activity, showing % inhibition values of 40.27%, 51.38%, 59.72%, 65.27%, and 73.61% at concentrations of 10, 11, 12, 13, and 14 µg/mL, respectively, as shown in Table 3

CONCENTRATION	INHIBITION
10	40.27%
11	51.38%
12	59.72%
13	65.27%
14	73.61%

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

Table 3: Percentage inhibition of DPPH radicals by standard Ascorbic Acid at different concentrations.

The calibration curve obtained for the standard showed a good linear relationship between concentration and percentage inhibition with the regression equation y = 8.0556x – 38.611 and a correlation coefficient R^2 = 0.9871, indicating high reliability of the assay, as illustrated in Table 4.

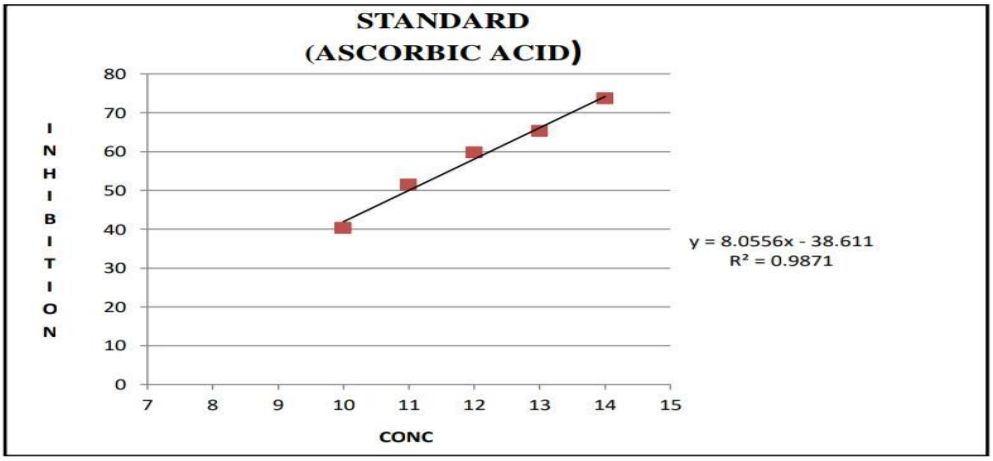


Table 4: Standard calibration curve of Ascorbic Acid.

Similarly, the synthesized copper nanoparticles (Sample code: CuNP-NF) also exhibited significant antioxidant activity. The percentage inhibition increased with increasing concentration of nanoparticles, indicating effective free radical scavenging ability. The CuNP-NF sample showed % inhibition values of 12.5%, 25%, 44.44%, 59.72%, and 76.38% at concentrations of 100 µL, 200 µL, 300 µL, 400 µL, and 500 µL, respectively

CONCENTRATION (MICROLITER)	INHIBITION
100	12.5%
200	25%
300	44.4%
400	59.72%
500	76.38%

Table 5: Antioxidant activity (% inhibition

The highest antioxidant activity was observed at 500 µL, showing 76.38% inhibition, which indicates strong radical scavenging potential

DISCUSSION

The present study demonstrates the successful green synthesis of copper nanoparticles using *Couroupita guianensis* flower extract and evaluates their structural characteristics and biological activities. The findings of phytochemical screening, spectroscopic analysis, and biological assays collectively confirm the effectiveness of plant-mediated nanoparticle synthesis.

Preliminary phytochemical analysis revealed the presence of flavonoids, phenolic compounds, alkaloids,

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

terpenoids, saponins, glycosides, steroids, coumarins, carbohydrates, amino acids, and essential oils. The abundance of phenolic and flavonoid compounds plays a crucial role in green nanoparticle synthesis due to their electron-donating hydroxyl groups.

Plant-derived phenolics and flavonoids are well documented as strong reducing agents capable of converting metal ions into their nanoform (Iravani, 2011). Similar phytochemical-mediated synthesis of copper nanoparticles has been reported using various plant extracts where phenolic compounds were primarily responsible for reduction and stabilization (Shah *et al*., 2014). Flavonoids act through oxidation of hydroxyl groups to carbonyl groups, facilitating electron transfer to Cu^{2+} ions (Mittal *et al.*, 2013). In addition, saponins and glycosides may serve as natural surfactants, preventing nanoparticle aggregation by providing steric stabilization (Ahmed *et al.*, 2016). The presence of amino acids also supports nanoparticle stabilization, as amino and carboxyl groups bind effectively to nanoparticle surfaces (Singh *et al*., 2018). Therefore, the phytochemical profile of C. guianensis extract strongly supports its suitability for green synthesis.

The observed color change from blue to green during synthesis is a primary indication of nanoparticle formation. The blue coloration corresponds to hydrated Cu^{2+} ions in solution. Upon reduction, the solution gradually turned green, suggesting formation of copper nanoparticles or copper oxide nanoparticles. Color variation during copper nanoparticle synthesis has been widely reported in literature, where transition from blue to green or brown indicates reduction and surface plasmon resonance effects (Nasrollahzadeh *et al.*, 2015). The formation of green coloration may indicate intermediate CuO or Cu_2O nanostructures along with metallic copper nanoparticles. This visual confirmation aligns with previous reports where plant extracts successfully reduced copper salts, resulting in observable color transitions (Sathiyavimal *et al.*, 2017).

UV–Visible analysis of the synthesized nanoparticles showed a characteristic absorption peak at 564 nm. This peak corresponds to surface plasmon resonance (SPR) of copper nanoparticles. Surface plasmon resonance arises due to collective oscillation of conduction band electrons when excited by incident light (Kelly *et al.*, 2003). For copper nanoparticles, SPR peaks are typically observed between 560–580 nm, depending on particle size and surrounding medium (Philip, 2010).

The presence of a single distinct peak at 564 nm suggests relatively uniform particle distribution with minimal aggregation. Broadening of the peak may indicate polydispersity, which is common in green synthesis methods (Song & Kim, 2009).

Comparable SPR peaks for plant-mediated copper nanoparticles have been reported at 560–570 nm in previous studies (Rajesh *et al.*, 2015), confirming that the present findings are consistent with established research.

FTIR analysis revealed major absorption peaks around 3400 cm^{-1} (O–H stretching), 2920 cm^{-1} (C–H stretching), 1630 cm^{-1} (C=O stretching), and 600–700 cm^{-1} (Cu–O bond vibration).

The broad O–H stretching band confirms the involvement of phenolic compounds and alcohol groups in reduction and stabilization. Similar FTIR patterns have been reported in green synthesized copper nanoparticles, where hydroxyl groups acted as reducing agents (Sutradhar *et al*., 2014).

The presence of amide or carbonyl groups suggests protein interaction with nanoparticles, providing a capping layer that enhances stability (Elumalai *et al*., 2015). The Cu–O stretching band further confirms nanoparticle formation. The shift in peak positions compared to pure extract indicates binding of phytochemicals to nanoparticle surfaces. Such shifts are commonly observed due to coordination interactions between metal ions and functional groups (Kumar *et al.*, 2017).

XRD analysis showed diffraction peaks at 2θ values of 43°, 50°, and 74°, corresponding to (111), (200), and (220) planes of face-centered cubic copper. These peaks confirm crystalline structure of the synthesized nanoparticles. The absence of additional impurity peaks suggests high purity synthesis. Similar diffraction patterns have been reported for green synthesized copper nanoparticles in earlier studies (Dhand *et al.*, 2016). The sharpness of peaks indicates good crystallinity, which is essential for enhanced biological activity. The nanoscale crystallite size, typically calculated using the Scherrer equation, supports the formation of particles in the nanometer range (Cullity & Stock, 2001). Crystallinity plays an important role in antimicrobial efficiency, as smaller crystalline particles exhibit higher surface area and increased reactivity (Raghavendra *et al.*, 2019).

The synthesized copper nanoparticles demonstrated concentration-dependent DPPH radical scavenging activity. The observed color change from deep violet to pale yellow confirms reduction of DPPH radicals. Nanoparticles synthesized via plant extracts often retain phytochemical capping agents on their surface, enhancing antioxidant activity

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

(Bar *et al*., 2009). The presence of phenolics and flavonoids on nanoparticle surfaces contributes to hydrogen donation and free radical neutralization. Similar antioxidant potential of plant-mediated copper nanoparticles has been reported in several studies (Gopinath *et al.,* 2014). Increased surface area and presence of active functional groups enhance radical scavenging efficiency. The synergistic interaction between copper ions and phytochemicals may amplify antioxidant properties compared to crude extract alone (Sathishkumar *et al.,* 2012).

The antibacterial activity results showed that copper nanoparticles exhibited a zone of inhibition of 18 mm, which was significantly higher than plant extract (10 mm) and copper sulfate solution (15 mm). Copper nanoparticles are known to exhibit strong antibacterial activity due to multiple mechanisms:

1. Generation of reactive oxygen species (ROS)
2. Disruption of bacterial cell membrane
3. Release of Cu^{2+} ions
4. Interaction with bacterial DNA and proteins

These mechanisms have been widely reported in literature (Raffi *et al.,* 2010). Smaller particle size enhances penetration through bacterial membranes, leading to increased cytotoxic effects (Azam *et al*., 2012). The enhanced activity observed in this study may be attributed to:

- Increased surface-to-volume ratio
- Synergistic effect of phytochemical capping agents
- Sustained release of copper ions

Similar findings were reported by Ren et al. (2009), where copper nanoparticles showed higher antibacterial activity compared to bulk copper. The greater inhibition zone compared to plant extract confirms that nanoparticle formation enhances biological efficacy.

The present study aligns strongly with global sustainability initiatives, particularly the United Nations Sustainable Development Goals (SDGs). The green synthesis approach adopted in this research directly supports SDG 3 (Good Health and Well-being) by developing biologically active copper nanoparticles with significant antibacterial and antioxidant potential. The enhanced antibacterial activity observed against *Escherichia coli* suggests possible applications in infection control, antimicrobial coatings, and biomedical formulations, which contribute to improved public health outcomes.

Furthermore, this work supports SDG 12 (Responsible Consumption and Production) by employing an eco-friendly, plant-mediated synthesis method instead of conventional chemical reduction techniques. Traditional nanoparticle synthesis often involves hazardous chemicals, high energy consumption, and toxic by-products (Iravani, 2011). In contrast, the use of Couroupita guianensis flower extract eliminates the need for harmful reducing agents, thereby minimizing environmental impact.

The process also aligns with SDG 13 (Climate Action) by promoting sustainable laboratory practices and reducing chemical waste. Green nanotechnology has emerged as a promising field that integrates environmental responsibility with technological advancement (Anastas & Warner, 1998). By utilizing renewable plant resources, this study demonstrates an approach that is both economically feasible and environmentally sustainable.

Additionally, the valorization of plant materials for high-value nanomaterial production contributes indirectly to SDG 9 (Industry, Innovation and Infrastructure) by encouraging innovation in bio-nanotechnology and supporting sustainable industrial development.

Significance of Green Nanotechnology Approach

Green nanotechnology emphasizes the design of nanomaterials using environmentally benign methods. The successful synthesis of copper nanoparticles using *C. guianensis* flower extract confirms the effectiveness of plant-mediated reduction pathways. Compared to physical and chemical methods, green synthesis offers advantages such as:

- Lower toxicity
- Cost-effectiveness
- Simplicity
- Biocompatibility

Previous studies have demonstrated that plant-mediated nanoparticles exhibit improved biological compatibility due to the presence of natural capping agents (Ahmed *et al.,* 2016). The phytochemical coating observed in FTIR analysis in the present study likely enhances stability and bioactivity.

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

In the present study, artificial intelligence (AI)-assisted tools were utilized for literature comparison, data organization, and interpretation support. AI-based academic tools can enhance research efficiency by facilitating rapid comparison of experimental values such as UV–Visible peak ranges, XRD diffraction positions, and reported antibacterial zones from previous studies.

AI-assisted literature screening enables identification of similar nanoparticle synthesis studies and helps in validating experimental findings against published data. For instance, the UV–Visible absorption peak observed at 564 nm was cross-referenced with reported SPR ranges for copper nanoparticles (Philip, 2010; Rajesh *et al.*, 2015). Such computational assistance improves accuracy in scientific comparison. Additionally, AI-supported data structuring can help in organizing results systematically and minimizing interpretation bias.

SUMMARY

The present study investigates the green synthesis of copper nanoparticles using *Couroupita guianensis* (Nagalingam) flower extract and evaluates their physicochemical and biological properties. Preliminary phytochemical screening confirmed the presence of flavonoids, phenolic compounds, alkaloids, terpenoids, saponins, glycosides, steroids, coumarins, carbohydrates, amino acids, and essential oils, indicating strong reducing and stabilizing potential. Upon addition of the aqueous flower extract to copper sulfate solution, a visible color change from blue to green was observed, confirming nanoparticle formation.

UV–Visible spectroscopy showed a characteristic surface plasmon resonance peak at 564 nm, indicating successful synthesis of copper nanoparticles. FTIR analysis revealed functional groups such as O–H, C=O, C–H, and Cu–O, confirming the involvement of phytochemicals in reduction and stabilization. XRD analysis displayed diffraction peaks at 2θ values of 43°, 50°, and 74°, corresponding to the (111), (200), and (220) planes of crystalline copper, confirming nanoscale crystalline structure.The synthesized nanoparticles demonstrated significant antioxidant activity in DPPH assay, showing concentration-dependent radical scavenging potential. Antibacterial evaluation using disc diffusion method revealed enhanced activity against Escherichia coli, with a maximum zone of inhibition of 18 mm, which was greater than the plant extract and copper sulfate solution. Overall, the study confirms that *C. guianensis*-mediated copper nanoparticles possess promising antioxidant and antibacterial properties and can serve as eco-friendly nanomaterials for potential biomedical applications.

ACKNOWLEDGEMENTS

I would like to express my deepest and most heartfelt gratitude to my mother, **Mrs. Kalpana M.N.,** whose unwavering love, sacrifices, encouragement, and belief in me formed the foundation of this work. Her constant support during every challenge and her confidence in my abilities gave me the strength and determination to complete this research. This accomplishment would not have been possible without her presence, guidance, and endless encouragement. I dedicate my sincere appreciation to her for being my greatest source of inspiration throughout this journey. I am profoundly grateful to my grandfather, **Mr. M.K. Narayanan,** and my grandmother, **Mrs. M.T. Shyamala**, for their unconditional love, blessings, wisdom, and constant encouragement. Their faith in me and their support throughout my academic journey have been invaluable and deeply cherished. I would also like to acknowledge my father, **Mr. K. Jyothi Basu**. The experiences and challenges I encountered throughout my journey motivated me to work harder, persevere, and strive toward completing this research successfully. These experiences played a significant role in shaping my determination and resilience. My sincere thanks go to my best friends, **Ms. Nooha Saleem and Ms. Fidha K.A.,** whose constant motivation, encouragement, and belief in my potential inspired me to push beyond my limits. Their support and positivity greatly contributed to my confidence and determination during the course of this work. I would also like to thank my dear college friend, **Ms. Harini**, for her friendship, encouragement, and support throughout my academic journey. Finally, I extend my sincere gratitude to my guide, **Ms. Vishnu Priya**, for her guidance and support during the completion of this research work. Her assistance and encouragement are gratefully acknowledged. To everyone who contributed directly or indirectly to the successful completion of this research, I offer my sincere thanks and appreciation.

REFERENCE

[1]. Ahmed, S., Ahmad, M., Swami, B. L., & Ikram, S. (2016*). A review on plants extract mediated synthesis of silver nanoparticles for antimicrobial applications. Journal of Advanced Research*, 7(1), 17–28. https://doi.org/10.1016/j.jare.2015.02.007

International Journal of Engineering Research & Technology (IJERT)
ISSN: 2278-0181
Vol. 15 Issue 06 , June - 2026

[2]. Ali, K., Dwivedi, S., Azam, A., Saquib, Q., Al-Said, M. S., Alkhedhairy, A. A., & Musarrat, J. (2016*). Aloe vera extract functionalized zinc oxide nanoparticles as nanoantibiotics against multi-drug resistant clinical bacterial isolates. Journal of Colloid and Interface Science*, 472, 145–156.
[3]. Bauer, A. W., Kirby, W. M. M., Sherris, J. C., & Turck, M. (1966). *Antibiotic susceptibility testing by a standardized single disk method. American Journal of Clinical Pathology*, 45(4), 493–496.
[4]. Blois, M. S. (1958). *Antioxidant determinations by the use of a stable free radical. Nature*, 181, 1199–1200.
[5]. Chandra, H., Patel, D., Kumari, P., Jangwan, J. S., & Yadav, S. (2020). *Phytochemicals as green reductants for nanoparticle synthesis. Materials Science for Energy Technologies*, 3, 335–345.
[6]. Dauthal, P., & Mukhopadhyay, M. (2016*). Noble metal nanoparticles: Plant-mediated synthesis and their antimicrobial activity. Industrial Crops and Products*, 82, 10–21.
[7]. Elumalai, K., & Velmurugan, S. (2015). *Green synthesis of silver nanoparticles using plant extracts and their antimicrobial activities. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy,* 143, 158–164.
[8]. Iravani, S. (2011*). Green synthesis of metal nanoparticles using plants. Green Chemistry*, 13(10), 2638–2650.
[9]. Jeevanandam, J., Barhoum, A., Chan, Y. S., Dufresne, A., & Danquah, M. K. (2018). *Review on nanoparticles and nanostructured materials. Beilstein Journal of Nanotechnology,* 9, 1050–1074.
[10]. Khandel, P., & Shahi, S. K. (2018). *Mycogenic nanoparticles and their bio-applications: A review. Journal of Nanostructure in Chemistry, 8*, 1–18.
[11]. Kumar, V., Yadav, S. K., & Yadav, S. C. (2010). *Syzygium cumini leaf and seed extract mediated biosynthesis of silver nanoparticles. Colloids and Surfaces B: Biointerfaces*, 75(1), 335–340.
[12]. Li, X., Schuler, M. A., & Berenbaum, M. R. (2007). *Molecular mechanisms of metabolic resistance to synthetic and natural xenobiotics. Annual Review of Entomology*, 52, 231–253.
[13]. Makarov, V. V., et al. (2014). *"Green" nanotechnologies: Synthesis of metal nanoparticles using plants. Acta Naturae*, 6(1), 35–44.
[14]. Mittal, A. K., Chisti, Y., & Banerjee, U. C. (2013). *Synthesis of metallic nanoparticles using plant extracts. Biotechnology Advances*, 31(2), 346–356.
[15]. Nasrollahzadeh, M., Sajadi, S. M., & Atarod, M. (2016). *Plant-mediated green synthesis of copper nanoparticles. Journal of Colloid and Interface Science*, 469, 191–205.
[16]. Oyaizu, M. (1986*). Studies on products of browning reaction: Antioxidative activities of browning reaction products. Japanese Journal of Nutrition*, 44, 307–315.
[17]. Prabhu, S., & Poulose, E. K. (2012). *Silver nanoparticles: Mechanism of antimicrobial action. International Nano Letters*, 2, 32.
[18]. Rai, M., Yadav, A., & Gade, A. (2009). *Silver nanoparticles as a new generation of antimicrobials. Biotechnology Advances*, 27(1), 76–83.
[19]. Sathishkumar, M., Sneha, K., & Yun, Y. S. (2009). *Immobilization of silver nanoparticles synthesized using plant extract. Bioresource Technology*, 100(14), 3466–3470.
[20]. Sharma, V. K., Yngard, R. A., & Lin, Y. (2009). *Silver nanoparticles: Green synthesis and antimicrobial activities. Advances in Colloid and Interface Science*, 145, 83–96.
[21]. Singh, J., Dutta, T., Kim, K. H., Rawat, M., Samddar, P., & Kumar, P. (2018*). Green synthesis of metals and their oxide nanoparticles. Journal of Nanobiotechnology*, 16, 84.
[22]. Sowndarya, P., & Ramkumar, G. (2012). *Green synthesis of copper nanoparticles. Journal of Nanotechnology,* 2012, 1–6.
[23]. Subramanian, K. S., et al. (2015*). FTIR and XRD characterization of metal nanoparticles. Applied Nanoscience*, 5, 1–8.
[24]. Thatoi, H., Patra, J. K., & Das, S. K. (2014). *Free radical scavenging and antioxidant potential of medicinal plants. Journal of Medicinal Plants Research*, 8(21), 776–790.
[25]. Yallappa, S., et al. (2013). *Antibacterial activity of plant mediated copper nanoparticles. Journal of Nanoscience and Nanotechnology*, 13, 1–6.