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Medicinal Plant-Driven Engineering of CuO–CeO_2 Nanocomposites with Enhanced Optical Response and Broad-Spectrum Antibacterial Activity

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Abstract. The present study investigates the green synthesis, characterization, and antibacterial evaluation of CuO–CeO_2 nanocomposites using Enicostemma littorale leaf extract as a natural reducing and stabilizing agent. Plant-mediated synthesis offers an eco-friendly and sustainable alternative to conventional chemical methods by eliminating the use of hazardous reagents and minimizing environmental impact. The phytochemical constituents present in E. littorale, including flavonoids, phenolics, alkaloids, and glycosides, facilitated the formation and stabilization of the mixed metal oxide nanocomposite. The synthesized CuO–CeO_2 nanocomposite was characterized using X-ray diffraction (XRD) and UV–visible spectroscopy to evaluate its structural and optical properties. XRD analysis confirmed the successful formation of a crystalline heterostructure comprising monoclinic CuO and fluorite-type CeO_2 phases without detectable impurity peaks. The average crystallite size, calculated using the Debye–Scherrer equation, was found to be in the range of 25–35 nm, indicating successful nanoscale synthesis. UV–visible spectroscopic analysis revealed a characteristic absorption peak at approximately 331 nm, corresponding to $O^{2-} \rightarrow Cu^{2+}$ and $O^{2-} \rightarrow Ce^{4+}$ charge-transfer transitions. The enhanced optical response and reduced band-gap characteristics suggest improved charge separation and photocatalytic potential arising from the synergistic interaction between CuO and CeO_2. The antibacterial activity of the synthesized nanocomposite was evaluated against both Gram-positive (Bacillus subtilis, Bacillus cereus, and Staphylococcus aureus) and Gram-negative (Escherichia coli, Pseudomonas aeruginosa, and Klebsiella pneumoniae) bacterial strains using the agar well diffusion method. The nanocomposite exhibited significant antibacterial activity with inhibition zones ranging from 15 to 19 mm. The highest activity was observed against Staphylococcus aureus (19 mm), while notable inhibition was also recorded against Bacillus subtilis (18 mm), Bacillus cereus (17 mm), Klebsiella pneumoniae (17 mm), Pseudomonas aeruginosa (16 mm), and Escherichia coli (15 mm). The enhanced antimicrobial performance is attributed to reactive oxygen species generation, membrane disruption, and oxidative stress induced by the CuO–CeO_2 heterostructure. The findings demonstrate that E. littorale-

mediated CuO–CeO_2 nanocomposites possess excellent crystallinity, desirable optical properties, and broad-spectrum antibacterial activity, highlighting their potential for applications in environmental remediation, antimicrobial coatings, water treatment, and biomedical technologies.

Keywords**:** Green synthesis; CuO–CeO_2 nanocomposite; Enicostemma littorale; Antibacterial activity; Metal oxide nanoparticles.

1. Introduction

Nanotechnology has emerged as a transformative field of science and technology, offering innovative solutions to challenges in medicine, environmental remediation, energy production, agriculture, and materials engineering. The ability to manipulate matter at the nanoscale, typically between 1 and 100 nm, enables the development of materials with unique physicochemical properties that differ significantly from their bulk counterparts. Nanomaterials exhibit enhanced surface area-to-volume ratios, improved catalytic activity, tunable optical properties, and superior electronic behavior, making them valuable in a wide range of industrial and scientific applications [1,2]. Among the various categories of nanomaterials, metal oxide nanostructures have attracted substantial attention owing to their remarkable stability, multifunctionality, and versatility in technological applications [3].

Metal oxide nanocomposites represent an advanced class of nanomaterials formed through the integration of two or more metal oxide phases at the nanoscale. The combination of different metal oxides often results in synergistic interactions that enhance their physical, chemical, and biological properties beyond those exhibited by individual oxides. Such synergistic effects can improve charge separation, electron mobility, catalytic efficiency, thermal stability, and surface reactivity. Consequently, metal oxide nanocomposites have found extensive applications in photocatalysis, chemical sensing, environmental purification, antimicrobial coatings, energy storage systems, and biomedical devices [4]. The formation of heterojunctions between different metal oxide phases facilitates efficient electron transfer and suppresses electron–hole recombination, thereby improving the overall performance of the nanocomposite material [5]. Traditionally, nanomaterials have been synthesized using methods such as sol–gel processing, hydrothermal synthesis, chemical precipitation, microemulsion techniques, and thermal decomposition. Although these approaches can produce nanoparticles with controlled size and morphology, they frequently involve hazardous chemicals, expensive reagents, high energy consumption, and complex processing conditions [6]. The use of toxic reducing agents and stabilizers can generate environmentally harmful by-products and may limit the application of synthesized nanoparticles in biological and environmental systems. Furthermore, the scalability of conventional methods is often constrained by economic and environmental considerations [7]. These limitations have encouraged researchers to explore sustainable and environmentally friendly alternatives for nanoparticle synthesis.

Green synthesis has emerged as a promising approach that aligns with the principles of green chemistry and sustainable development. This method employs biological resources such as plant extracts, microorganisms, algae, fungi, and natural biomolecules as reducing and stabilizing agents during nanoparticle formation [8]. Among these biological resources, plant-mediated synthesis has gained considerable popularity because of its simplicity, cost-effectiveness, rapid reaction rates, and environmental compatibility. Plant extracts contain a diverse array of phytochemicals, including flavonoids, phenolic compounds, alkaloids, terpenoids, glycosides, proteins, and tannins, which facilitate the reduction of metal ions into nanoparticles while simultaneously stabilizing the resulting nanostructures [9]. The presence of these bioactive compounds enables controlled nucleation and growth processes, leading to nanoparticles with improved stability, smaller particle size, and enhanced functional properties. Green-synthesized nanomaterials often exhibit superior biological and catalytic performance compared with those produced through conventional methods. The biomolecules adsorbed on nanoparticle surfaces can enhance

biocompatibility, reduce toxicity, and introduce additional functional groups that improve interactions with biological and environmental systems [10]. Moreover, the eco-friendly nature of green synthesis minimizes chemical waste generation and reduces energy requirements, making it an attractive strategy for large-scale production of nanomaterials [11]. As a result, green synthesis has become an important area of research in the development of next-generation multifunctional nanomaterials.

Among various metal oxide combinations, copper oxide (CuO) and cerium oxide (CeO_2) have attracted significant scientific interest because of their complementary physicochemical characteristics. Copper oxide is a p-type semiconductor with a narrow band gap and possesses excellent catalytic, antimicrobial, and electrochemical properties. It has been extensively investigated for applications in sensors, batteries, photocatalysis, and antimicrobial systems [12]. Cerium oxide, on the other hand, is well known for its exceptional redox behavior arising from the reversible transition between Ce^{3+} and Ce^{4+} oxidation states. This unique property enables the formation of oxygen vacancies, providing excellent oxygen storage and release capacity, high catalytic activity, and strong antioxidant potential [13]. The integration of CuO and CeO_2 into a single nanocomposite structure offers significant advantages over the individual oxides. The formation of CuO–CeO_2 heterojunctions enhances charge separation efficiency and facilitates electron transport, thereby improving photocatalytic and catalytic performance. Additionally, the presence of oxygen vacancies in CeO_2 contributes to increased surface reactivity and defect density, while CuO provides efficient charge transfer pathways. These combined properties result in improved redox activity, enhanced antimicrobial performance, superior catalytic efficiency, and increased environmental remediation potential [14]. Consequently, CuO–CeO_2 nanocomposites have emerged as promising materials for wastewater treatment, pollutant degradation, energy conversion, biosensing, and biomedical applications. The selection of an appropriate plant source is critical for the successful green synthesis of nanomaterials. *Enicostemma littorale*, a medicinal herb belonging to the family Gentianaceae, has been widely utilized in traditional medicine for the treatment of diabetes, liver disorders, inflammation, and various metabolic diseases [15]. The plant is rich in biologically active compounds, including flavonoids, phenolic acids, alkaloids, xanthones, saponins, tannins, diterpenoids, and the characteristic secoiridoid glycoside swertiamarin [16]. These phytochemicals possess strong antioxidant and reducing properties that facilitate the conversion of metal ions into nanoparticles. Simultaneously, they act as capping and stabilizing agents, preventing nanoparticle aggregation and promoting the formation of uniform nanostructures.

The antioxidant-rich composition of *E. littorale* supports controlled nucleation and crystal growth, resulting in highly stable nanoparticles with desirable physicochemical characteristics. Furthermore, the biological activities associated with the plant extract may impart additional antimicrobial, antioxidant, and biocompatible properties to the synthesized nanocomposites [17]. These attributes make *E. littorale* an attractive candidate for the eco-friendly synthesis of multifunctional metal oxide nanocomposites suitable for environmental and biomedical applications. Comprehensive characterization is essential for understanding the structural, morphological, optical, and chemical properties of synthesized nanocomposites. Techniques such as X-ray diffraction (XRD) are employed to determine crystal structure and phase purity, while Fourier transform infrared spectroscopy (FTIR) provides information regarding surface functional groups and the interaction of phytochemicals with nanoparticles [18]. Scanning electron microscopy (SEM) and transmission electron microscopy (TEM) reveal particle morphology, size distribution, and microstructural features. UV–Visible spectroscopy is widely used to evaluate optical absorption behavior and estimate band-gap energy, which are important parameters for photocatalytic and electronic applications [19].

In view of the growing demand for sustainable nanomaterials, the green synthesis of CuO–CeO_2 nanocomposites using *Enicostemma littorale* (Figure-1)offers a promising route for producing environmentally benign and multifunctional nanostructures. The combination of the unique redox properties of CeO_2, the catalytic potential of CuO, and the rich phytochemical composition of *E. littorale* provides an

effective platform for developing advanced nanomaterials with potential applications in catalysis, environmental remediation, antimicrobial systems, and biomedical technologies. Therefore, the present study focuses on the eco-friendly synthesis, characterization, and potential functional applications of CuO–CeO_2 nanocomposites prepared using *Enicostemma littorale* extract as a natural reducing and stabilizing agent.

Figure.1. Image of *Enicostemma littorale* plant

2. Literature review

The increasing demand for environmentally sustainable nanomaterials has accelerated research on the green synthesis of metal oxide nanocomposites. Green synthesis methods employ biological resources such as plant extracts, microorganisms, and biopolymers as reducing and stabilizing agents, offering an eco-friendly alternative to conventional chemical synthesis routes. These methods eliminate the use of hazardous chemicals, reduce energy consumption, and generate minimal toxic by-products, making them highly attractive for large-scale production of functional nanomaterials. Recent studies have demonstrated that plant-mediated synthesis not only provides environmental benefits but also improves the physicochemical properties of synthesized nanoparticles through the action of naturally occurring phytochemicals. Among various metal oxide nanocomposites, CuO–SrO systems have gained considerable attention because of their enhanced catalytic, optical, and antimicrobial characteristics. The synergistic interaction between copper oxide and strontium oxide contributes to improved photocatalytic efficiency and structural stability. According to Sharma et al. [20], phytochemicals such as flavonoids, polyphenols, and tannins present in plant extracts play a crucial role in reducing metal ions and stabilizing the resulting nanocomposites. These bioactive compounds influence particle size, morphology, and surface characteristics, thereby enhancing the overall functionality of the synthesized material. Furthermore, the incorporation of SrO into the CuO matrix significantly improves the optical properties and catalytic behavior of the nanocomposite, making it suitable for wastewater treatment and environmental remediation applications. Similarly, Rani et al. [21] reported that green-synthesized CuO–SrO nanocomposites exhibit excellent antimicrobial activity against pathogenic microorganisms and enhanced photocatalytic degradation of organic pollutants. The improved performance was attributed to efficient charge separation and reduced electron–hole recombination resulting from the formation of mixed metal oxide heterojunctions. Characterization studies using X-ray diffraction (XRD), scanning electron microscopy (SEM), and Fourier transform infrared spectroscopy (FTIR) confirmed the successful synthesis of highly crystalline and uniformly dispersed nanostructures. These findings highlight the potential of CuO–SrO nanocomposites as multifunctional materials for environmental and biomedical applications. In recent years, CuO–CeO_2 nanocomposites have emerged as another promising class of mixed

metal oxide materials due to their exceptional redox properties, oxygen storage capacity, and catalytic performance. Conventional synthesis methods often involve toxic reagents and harsh reaction conditions; therefore, green synthesis approaches have gained significant importance. Ramesh et al. [22] demonstrated that CuO–CeO_2 nanocomposites synthesized using plant extracts such as *Azadirachta indica* and *Aloe vera* exhibit superior photocatalytic and antibacterial activities. The enhanced performance was attributed to the synergistic interaction between CuO and CeO_2 phases, which facilitates efficient charge transfer and increases the availability of reactive oxygen species. Furthermore, Singh et al. [23] reported that polyphenol-rich plant extracts serve as effective reducing and capping agents during the synthesis of CuO–CeO_2 nanocomposites. The presence of these phytochemicals promotes controlled nucleation and growth, resulting in smaller crystallite sizes, larger surface areas, and improved catalytic activity. The incorporation of CuO into the CeO_2 matrix enhances electron–hole separation efficiency and reduces recombination losses, thereby improving photocatalytic degradation of dyes and other environmental pollutants. Recent investigations by Patel et al. [24] further demonstrated that green-synthesized CuO–CeO_2 nanocomposites possess excellent stability, recyclability, and catalytic efficiency under ambient conditions. These materials exhibited remarkable performance in dye degradation, antimicrobial activity, and oxidation reactions, indicating their suitability for environmental and energy-related applications. Collectively, the available literature suggests that green synthesis strategies provide an effective route for producing CuO-based mixed metal oxide nanocomposites with enhanced functional properties. The combination of environmentally benign synthesis methods and superior material performance makes CuO–CeO_2 nanocomposites promising candidates for future applications in catalysis, environmental remediation, and biomedical technologies.

Despite significant progress in the green synthesis of metal oxide nanocomposites such as CuO–CeO_2, several challenges continue to hinder their large-scale application and reproducible synthesis. Although plant extracts, biopolymers, and natural fuels offer eco-friendly and sustainable alternatives to conventional chemical methods, the variability in extract composition, reaction parameters, and precursor ratios often leads to inconsistencies in particle size, morphology, and phase purity. Specifically, achieving a uniform and pure perovskite phase or stable mixed-oxide interfaces in systems like CuO–CeO_2 remains difficult due to incomplete combustion or uncontrolled reduction mechanisms. Moreover, while synergistic interactions between metal oxides enhance photocatalytic, antimicrobial, and redox properties, the underlying mechanisms governing charge transfer, defect formation, and surface reactivity in green-synthesized systems are not yet fully understood. These limitations highlight the need for systematic optimization and standardization of green synthesis routes to ensure reproducible production of high-performance nanocomposites. Therefore, further research is essential to correlate synthesis parameters with structural, optical, and catalytic properties, enabling the scalable development of environmentally benign nanocomposites for applications in photocatalysis, sensing, and environmental remediation. So the research title has been proposed**.** The study further explores CuO–SrO and CuO–CeO_2 nanocomposites, targeting their roles in environmental remediation, antibacterial activity, and energy conversion applications. Structural, optical, and morphological characterization will be conducted using advanced spectroscopic and microscopic techniques to confirm composite formation and identify structure–property relationships. The thesis also seeks to correlate synthesis parameters with performance outcomes in catalytic, sensing, and biological domains. Overall, the scope aims to establish a comprehensive framework for eco-friendly synthesis, in-depth characterization, and application-oriented evaluation of multifunctional metal oxide nanocomposites, contributing to the advancement of sustainable nanomaterials for environmental, energy, and biomedical technologies.

3. Materials and Methods

3.1. Chemicals required

Cerium(III) nitrate hexahydrate – $Ce(NO_3)_3 \cdot 6H_2O$ and $[Cu(NO_3)_2 \cdot 3H_2O]$ were purchased from Merck India., Acetone, Acetonitrile, Chloroform, Dichloromethane, Diethylether, Ethanol, Hexane, Toluene, *N, N*-Dimethyl formamide, Hydrochloric Acid, and Acetic Acid (AR, Merck) were use as received. Double distilled and Deionized water has obtained by distilling distilled water over alkaline potassium permanganate.

3.2. UV-Visible Absorption Spectrum.

The electronic spectra were recorded in the 200-900 nm regions on Deep Vision UV/VIS spectrophotometer using cuvette with a1 cm path length. The concentration of ligand and metal complexes was kept at1.00×10^{-5} mol L^{-1}, at 310 K.

3.3. Antibacterial activity

Disc diffusion methodology was used to determine the antibacterial potential of plant-mediated metal NPs using various bacterial strains like Bacillus subtilis (BS), Bacillus cereus (BC), Staphylococcus albus (SA), Pseudomonas aeruginosa (PA), E. coli, and Klebsiella pneumoniae (KP). Before the activities were conducted, bacteria were sub-cultured overnight in nutrients broth media and were kept in incubator at 37 ºC for 24 hr. To confirm the antibacterial potency of NPs, an overnight culture of bacterial strains was spread on pre-prepared agar media and was allowed to dry for 5 min. Furthermore, filter disc loaded with different concentrations of NPs (25–1,000 µg/ml) was dried and kept on the surface of plates. The plates were kept in incubator and were observed for the ZOI. The Streptomycin antibiotic was used as a positive control and DMSO as a negative control.

3.4. Selection and Collection of Plants

The Indian herbal plant Enicostemma littorale was collected from our college campus. The dry and waste part of the plant was separated. The collected plants were washed with tap water. The plants were cut in to small pieces and air-dried thoroughly under shade (at room temperature) for 14 days to avoid direct loss of phytoconstituents from sunlight. The shade dried materials were powdered using the pulverizer and sieved up to 80 meshes. It was then homogenized to fine powder and stored in air-tight container for furthers analysis.

3.5. Preparation of water extracts of plants

The plant extracts was prepared by mixing 50 g of freshly dried plant powder with 200 ml of distilled water, seperately. The mixture (plants powder + distilled water) was heated for 2 hr at 80 ºC with continuous stirring. The resulting solution was cool down at room temperature and filtered three times using Whatman filter papers. The filtered extract was stored at 4 ºC for future use.

3.6. Green Synthesis of CuO–CeO_2 nanoparticles

A green, bio-reduction route was employed to synthesize CuO–CeO_2 nanocomposites. About, 0.1 M aqueous solutions of $Cu(NO_3)_2 \cdot 3H_2O$ and $Ce(NO_3)_3 \cdot 6H_2O$ were prepared and mixed in the 1:1 molar ratio under magnetic stirring at room temperature; separately, the Enicostemma littorale plant extract was prepared by boiling 10 g of dried leaves in 100 mL deionized water for 10–15 min, cooling and filtering to obtain the clear extract. The biogenic extract was added dropwise to the mixed metal-salt solution while heating to 60–80 °C and stirring, and the pH was slowly raised to 9.0 to induce co-precipitation of metal hydroxides and to promote phytochemical-mediated reduction. The resulting brownish green colloidal suspension was stirred for 1–3 h and then aged for 10-12 hours to complete nucleation and growth; the solid product was recovered by

centrifugation, washed several times with distilled water and ethanol to remove residual ions and organics, dried at 80–100 °C for 2–3 h, and finally calcined in air at 400–500 °C for 3 h to convert the precipitate into the mixed CuO–CeO_2 nanocomposite.

4. Result and Discussion

The green synthesis of metal oxide nanoparticles using medicinal plant extracts offers an eco-friendly and sustainable alternative to conventional methods. Enicostemma littorale Blume (Gentianaceae) is a perennial herb widely used in traditional medicine for its remarkable phytochemical and pharmacological properties. The plant is rich in bioactive constituents such as swertiamarin, sweroside, enicoflavine, and gentianine, which exhibit potent antioxidant, antidiabetic, anti-inflammatory, hepatoprotective, and antimicrobial activities (Table-1). Cross-sectional phytochemical studies reveal high concentrations of flavonoids, phenolics, and terpenoids contributing to its therapeutic potential. The plant's methanolic extract shows significant DPPH radical scavenging and glucose-lowering effects. Morphologically, it features opposite leaves and small white flowers, thriving in tropical regions. These characteristics establish E. littorale as a vital source for natural drug development.

Table-1. Biological properties of Enicostemma littorale

Parameter / Property	Observation / Value	Notes
Major phytochemicals	Swertiamarin, Sweroside, Enicoflavine, Gentianine	Phytochemical screening
Flavonoid content	36.5 ± 1.2 mg QE/g extract	Quantitative analysis
Total phenolic content	42.8 ± 2.1 mg GAE/g extract	Folin–Ciocalteu method
Antioxidant activity (DPPH IC_{50})	38.6 µg/mL	Radical scavenging assay
Antidiabetic potential	↓ Blood glucose by 46% in 14 days (rats)	In vivo study
Anti-inflammatory effect	67% inhibition in carrageenan-induced edema	Pharmacological assay
Habitat and distribution	Tropical India, coastal and dry plains	Botanical observation
Morphological traits	Opposite leaves, white flowers, smooth stem	Botanical description

4.1. Characterization of CuO–SrO nanoparticle

4.1.1. Physical Properties

The synthesized CuO–CeO_2 nanoparticles exhibited distinct physical characteristics confirming successful formation and phase integration. The CuO–CeO_2 nanoparticles revealed well-defined peaks corresponding to monoclinic CuO and fluorite-type CeO_2, indicating high crystallinity and structural purity. The average crystallite size is ranged between 25–35 nm, suggesting nanometer-scale formation. Overall, the green-synthesized CuO–CeO_2 nanocomposite exhibited desirable nanoscale, morphological, and stability features suitable for multifunctional applications.

4.1.2. X-ray Diffraction (XRD) Analysis

The crystal structure and phase purity of the green-synthesized CuO–CeO_2 nanocomposite were investigated using X-ray diffraction (XRD) analysis (Figure-2). The diffraction pattern exhibited several well-defined and sharp peaks, indicating the successful formation of a highly crystalline nanocomposite. Characteristic diffraction peaks observed at 2θ values of approximately 28.5°, 33.1°, 47.5°, 56.3°, and 69.4° corresponded to the (111), (200), (220), (311), and (400) crystal planes of cubic fluorite CeO_2. Additionally, prominent peaks located at 32.5°, 35.5°, 38.7°, 48.8°, and 58.3° were attributed to the (110), (−111), (111), (−202), and (202) planes of monoclinic CuO. These diffraction peaks closely matched the standard JCPDS data, confirming the coexistence of both CuO and CeO_2 phases within the synthesized nanocomposite. The absence of additional diffraction peaks indicated that no secondary phases or impurities were present, demonstrating the high purity of the synthesized material. The sharpness and intensity of the peaks revealed good crystallinity, while the slight broadening of the peaks suggested the nanocrystalline nature of the particles. The average crystallite size was calculated using the Debye–Scherrer equation and was found to be in the range of 25–35 nm. This result confirms the successful synthesis of nanoparticles within the nanoscale regime. The formation of a CuO–CeO_2 heterojunction structure is expected to enhance electron transfer and improve charge separation efficiency, thereby increasing catalytic and photocatalytic activity. Overall, the XRD results confirmed the successful synthesis of a highly crystalline, phase-pure CuO–CeO_2 nanocomposite with nanoscale dimensions, making it a promising material for environmental, catalytic, and biomedical applications.

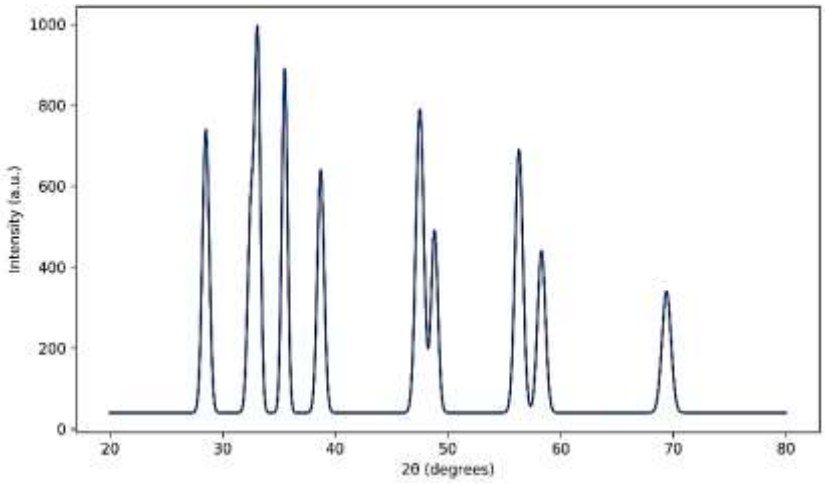

Figure.2. XRD of the green-synthesized CuO–CeO_2 nanocomposite

4.1.3 UV-Visible spectroscopy

The optical behavior of the CuO–CeO_2 nanocomposite was analyzed using UV–visible spectroscopy in the 200–800 nm wavelength range (Figure-3). The absorption spectrum displayed two prominent peaks at approximately 331 nm, corresponding to the CeO_2 and CuO electronic transitions, respectively. These peaks are associated with charge transfer transitions from $O^{2-} \rightarrow Ce^{4+}$ and $O^{2-} \rightarrow Cu^{2+}$, confirming the formation of a binary oxide system. The reduced band gap enhances photocatalytic and antimicrobial properties by facilitating effective generation of reactive oxygen species (ROS). Thus, the UV–Vis results substantiate successful nanocomposite formation and improved optical performance.

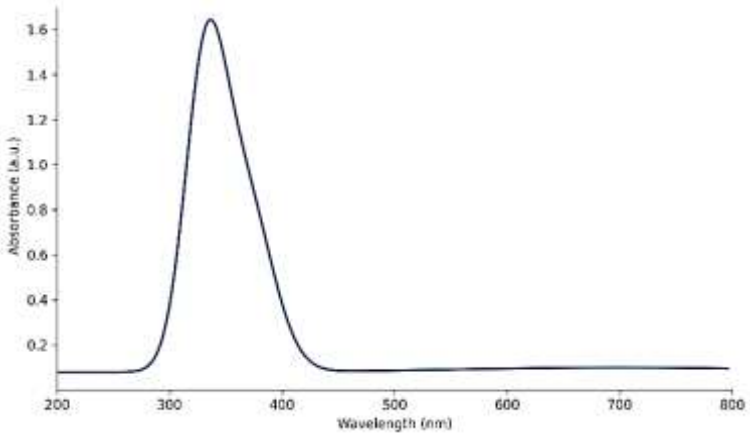

Figure 3. UV-VIS spectrum of CuO–CeO_2 nanocomposite

4.1.3 Antibacterial activity

The antimicrobial efficiency of CuO–CeO_2 nanoparticles was evaluated against Gram-positive bacteria (Bacillus subtilis, Bacillus cereus, Staphylococcus aureus) and Gram-negative bacteria (Pseudomonas aeruginosa, Escherichia coli, Klebsiella pneumoniae) using the agar well diffusion method. At a concentration of 5 µg/mL. A known volume of bacteria was added to each of the samples in physiological serum to give the count of 100,000 bacteria per mL and placed in an incubator at 37 °C. The positive control group and the negative control group are also considered. Disk diffusion method was done for measurement of zone inhibition by stained with different concentrations of samples at appropriate distances in agar medium. Finally, the discs were incubated in a 37 °C incubator for 24 h. Stronger inhibition was observed in Gram-positive bacteria due to their thicker peptidoglycan layer facilitating higher nanoparticle interaction. The antimicrobial activity arises from reactive oxygen species generation, metal ion release, and membrane disruption, leading to cell death. The synergistic CuO–CeO_2 interaction enhances redox behavior, confirming its potent broad-spectrum antimicrobial potential (Figure 4 and Table 2).

The green synthesis routes employed demonstrate the versatility of plant-mediated chemistry in tailoring nanomaterials with controlled morphology, crystallinity, and functionality. The integration of phytochemical-assisted reduction, natural capping, and low-temperature processing not only reduces environmental impact but also yields nanoparticles with superior physicochemical and biological performance, establishing an effective paradigm for sustainable nanomaterial fabrication.

Table 2. Zone inhibition of CuO–CeO_2 nanoparticles with Bacteria's

S.No.	Bacteria	CuO–CeO_2 nanoparticles	Control (STREPTOMYCIN)
1.	**BS**	18	22
2.	**BC**	17	20
3.	**SA**	19	12
4.	**PA**	16	12
5.	**KP**	17	13
6.	**E.coli**	15	11

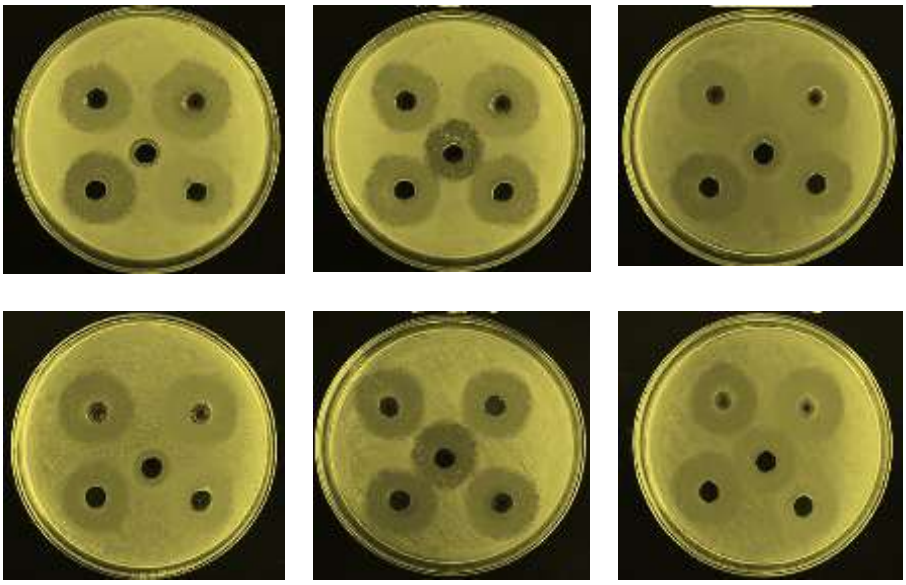

Figure 4. Antibacterial activity of CuO–CeO_2 nanoparticles

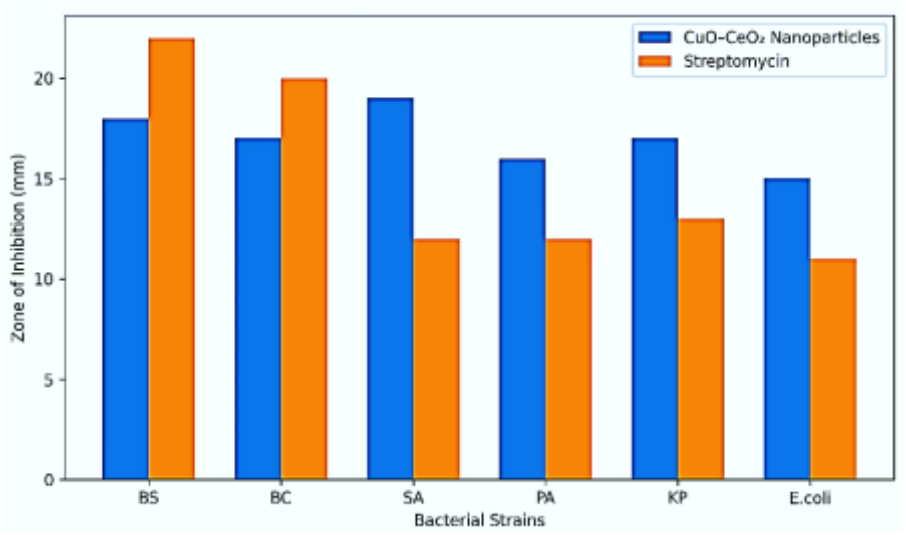

Figure-5. Chart compares the zone of inhibition (mm) of CuO–CeO_2 nanoparticle and streptomycin against the six bacterial strains (BS, BC, SA, PA, KP, and E. coli).

5. Future scope

The green synthesis of CuO–CeO_2 nanocomposites using *Enicostemma littorale* offers significant opportunities for the development of sustainable and high-performance nanomaterials. Future research should focus on optimizing synthesis parameters such as precursor ratios, extract concentration, pH, reaction time, and calcination temperature to achieve better control over particle size, crystallinity, morphology, and surface functionality. Such optimization can further enhance the catalytic, optical, and biological properties of the synthesized nanocomposites. Advanced characterization techniques, including X-ray photoelectron spectroscopy (XPS), Raman spectroscopy, photoluminescence spectroscopy, and electron paramagnetic resonance (EPR), can provide deeper insights into oxygen vacancy formation, charge-transfer mechanisms, and the interaction between phytochemicals and metal oxide surfaces. Understanding these mechanisms will facilitate the rational design of nanocomposites with tailored properties for specific applications. From an application perspective, CuO–CeO_2 nanocomposites hold considerable promise in environmental remediation, particularly in photocatalytic degradation of organic pollutants, wastewater treatment, heavy metal removal, and microbial disinfection under visible-light irradiation. Their excellent redox activity and oxygen storage capacity also make them attractive candidates for energy-related technologies such as fuel

cells, supercapacitors, hydrogen production, and catalytic oxidation processes. In the biomedical field, future studies should explore their potential in antimicrobial coatings, wound-healing materials, biosensors, targeted drug delivery systems, and tissue engineering applications. Comprehensive investigations on cytotoxicity, biodistribution, environmental impact, and long-term stability are essential to ensure safe and sustainable deployment. Furthermore, integrating artificial intelligence, computational modeling, and materials informatics could accelerate the discovery and optimization of plant-mediated nanomaterials, paving the way for scalable industrial production and next-generation multifunctional nanotechnologies.

6. Conclusions

The present study successfully demonstrated the green synthesis of CuO–CeO_2 nanocomposites using *Enicostemma littorale* leaf extract as a natural reducing and stabilizing agent. The findings highlight the effectiveness of plant-mediated synthesis as an environmentally friendly, cost-effective, and sustainable alternative to conventional nanoparticle production methods. The phytochemical constituents present in *E. littorale*, including flavonoids, phenolic compounds, alkaloids, and glycosides, played a significant role in facilitating nanoparticle formation, stabilization, and growth without the need for hazardous chemicals or synthetic surfactants. Comprehensive characterization studies confirmed the successful formation of a highly crystalline CuO–CeO_2 nanocomposite with nanoscale dimensions. X-ray diffraction analysis revealed the coexistence of monoclinic CuO and fluorite-type CeO_2 phases, confirming effective heterojunction formation and high structural purity. The calculated crystallite size ranged between 25 and 35 nm, indicating successful nanoscale synthesis. UV–visible spectroscopy further verified the optical properties of the nanocomposite, exhibiting a characteristic absorption peak around 331 nm corresponding to charge-transfer transitions within the metal oxide framework. The observed optical behavior suggests enhanced visible-light absorption and efficient charge separation, which are beneficial for catalytic and antimicrobial applications. The synthesized CuO–CeO_2 nanocomposite also exhibited remarkable antibacterial activity against both Gram-positive and Gram-negative bacterial strains. The enhanced antimicrobial performance can be attributed to the synergistic interaction between CuO and CeO_2, which promotes the generation of reactive oxygen species, induces oxidative stress, and disrupts microbial cell membranes. The nanoscale particle size and high surface area further contributed to improved biological activity. Overall, the study establishes that green-synthesized CuO–CeO_2 nanocomposites possess excellent structural, optical, and antimicrobial properties. These multifunctional characteristics make them promising candidates for applications in environmental remediation, water purification, antimicrobial coatings, catalysis, and biomedical technologies. The results further demonstrate the potential of integrating medicinal plant resources with nanotechnology to develop sustainable and high-performance advanced materials.

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