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Eucalyptus Leaf Extract-Mediated Green Synthesis of Zinc Oxide Nanoparticles: Phytochemical-Driven Band Gap Engineering and Structural Optimization

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Abstract

This study reports a single-step, eco-friendly precipitation route for synthesizing zinc oxide (ZnO) nanoparticles using aqueous Eucalyptus leaf extract as a simultaneous bio-reducing, capping, and stabilizing agent, in place of hazardous chemical reductants and synthetic surfactants. Phytochemical screening confirmed a phenolic- and flavonoid-rich extract matrix, with a total phenolic content of 174.52 ± 4.85 mg gallic acid equivalent (GAE)/g and a total flavonoid content of 58.31 ± 2.14 mg quercetin equivalent (QE)/g, while GC-MS profiling identified 1,8-cineole (46.85%), α-pinene (14.22%), α-terpineol (9.18%), globulol (6.41%), and a gallic acid derivative (5.74%) as the principal constituents. Synthesis parameters were systematically optimized, with an extract-to-precursor ratio of 1:2 (v/v), 0.10 M zinc nitrate, pH 10, and calcination at 400 °C identified as optimal. X-ray diffraction confirmed a phase-pure hexagonal wurtzite structure (JCPDS 36-1451) with an average crystallite size of 18.35 nm, corroborated by FE-SEM/HR-TEM particle sizing (20.42 ± 2.58 nm) and a near-stoichiometric Zn:O ratio of 1:1.01 (EDX). Comparative FTIR analysis confirmed retention of a deprotonated polyphenolic capping layer, conferring a zeta potential of −32.40 mV and a narrow hydrodynamic size distribution (PDI = 0.214). BET analysis revealed a mesoporous structure with a specific surface area of 34.82 m²/g. Critically, UV-Vis diffuse reflectance spectroscopy revealed a narrowed optical band gap of 3.25 eV relative to bulk ZnO (3.37 eV), accompanied by intense defect-related photoluminescence at 535 nm, indicating a high density of singly ionized oxygen vacancies. Tasleem (2017) These findings demonstrate that Eucalyptus-derived phytochemicals function not merely as passive capping agents but as an active, non-chemical route to band gap engineering, establishing a mechanistically substantiated foundation for the visible-light photocatalytic application of this material.

1. Introduction

Zinc oxide (ZnO) is a wide-band-gap (≈ 3.37 eV) II–VI semiconductor with a large exciton binding energy (≈60 meV), high thermal and chemical stability, and multifunctional applicability spanning optoelectronics, sensing, and photocatalysis (Raha & Ahmaruzzaman, 2022; Wang & Song, 2006). Its wide band gap, however, restricts intrinsic photoactivation to the ultraviolet region, motivating extensive research into strategies for extending its optical response toward the visible spectrum (Ong et al., 2018; Djurišić et al., 2014).

Conventional physical and chemical synthesis routes for ZnO nanoparticles, while offering reasonable control over particle morphology, typically require toxic reducing agents, organic solvents, and energy-intensive processing, raising sustainability and scalability concerns (Mittal et al., 2013). Green synthesis using plant extracts has consequently emerged as an environmentally benign alternative, in

which naturally occurring phytochemicals—flavonoids, tannins, and phenolic acids—serve as reducing and capping agents (Kól et al., 2017; Anastas & Warner, 1998).

Eucalyptus has attracted particular interest owing to its phenolic-rich phytochemical profile and prior demonstrated utility in ZnO nanoparticle biosynthesis (Barzinjy & Azeez, 2020; Siripireddy & Mandal, 2017; Ahmad et al., 2020). However, most existing reports remain largely descriptive, confirming nanoparticle formation without establishing a rigorous, multi-technique mechanistic link between phytochemical composition and the resultant electronic structure of the material (Mwangi et al., 2026). In particular, whether the phytochemical capping layer can be deliberately exploited to narrow the ZnO band gap, rather than functioning as a purely passive size-control agent, remains insufficiently substantiated. Bose & Roy (2025)

The present study addresses this gap by (i) comprehensively profiling the phytochemical composition of aqueous Eucalyptus leaf extract, (ii) systematically optimizing the green precipitation synthesis of ZnO nanoparticles, and (iii) applying an integrated structural, morphological, surface-chemical, thermal, and optical characterization strategy to establish direct evidence that Eucalyptus-derived phytoconstituents actively modify the surface defect chemistry and optical band gap of the resulting ZnO nanostructures. Khatun et al. (2024)

2. Materials and Methods

2.1 Materials

Zinc nitrate hexahydrate [$Zn(NO_3)_2 \cdot 6H_2O$] (AR grade, ≥98%) was used as the zinc precursor. Sodium hydroxide (AR, ≥97%) was used for pH adjustment. Standard reagents for phytochemical screening (Mayer's, Wagner's, ferric chloride, Molisch's, Fehling's reagents), Folin-Ciocalteu reagent, aluminium chloride, gallic acid, and quercetin standards were procured at analytical grade. De-ionized water (resistivity ≥ 18.2 MΩ·cm) was used throughout. Pathak (2023)

2.2 Collection and Preparation of Eucalyptus Leaf Extract

Fresh Eucalyptus leaves were collected, authenticated by a qualified taxonomist, shade-dried, and powdered. The powder was extracted in de-ionized water (1:10 w/v) at 60–80 °C for 30–60 min under continuous stirring, and the resulting extract was filtered (Whatman No. 1) and stored at 4 °C prior to use.

2.3 Phytochemical Screening and Profiling

Qualitative phytochemical tests were performed following standard protocols. Total phenolic content (TPC) was determined by the Folin-Ciocalteu method (gallic acid standard, λ = 765 nm) and total flavonoid content (TFC) by the aluminium chloride method (quercetin standard, λ = 415 nm). FTIR spectra of the extract were recorded (4000–400 cm^{-1}, KBr pellet). GC-MS analysis was performed using a capillary column with helium carrier gas, and compounds were identified by NIST library matching.

2.4 Green Synthesis of ZnO Nanoparticles

Eucalyptus extract was added dropwise to aqueous $Zn(NO_3)_2 \cdot 6H_2O$ solution under continuous stirring at 60–80 °C. The extract-to-precursor ratio (v/v), precursor molarity, reaction pH, temperature, stirring duration, and calcination temperature were independently varied to establish optimum conditions (Section 3.2). The precipitate was recovered by centrifugation (8000–10,000 rpm), washed with water and ethanol, oven-dried (80–100 °C), and calcined in a muffle furnace to obtain crystalline ZnO.

2.5 Characterization Techniques

Crystal structure was analyzed by powder XRD (Cu-Kα, λ = 1.5406 Å, 2θ = 20–80°); crystallite size was calculated using the Debye-Scherrer equation. Optical absorption and band gap were determined by UV-Vis diffuse reflectance spectroscopy (DRS) with Tauc plot extrapolation. Photoluminescence (PL) spectra were recorded at room temperature (λex = 325/350 nm). Morphology was examined by FE-SEM and HR-TEM/SAED, and elemental composition by EDX. Surface functional groups were analyzed by FTIR; colloidal properties by zeta potential and DLS. Thermal behavior was assessed by TGA/DSC (room temperature–800 °C, 10 °C/min), and textural properties by N_2 physisorption (BET/BJH methods).

3. Results and Discussion

3.1 Phytochemical Composition of Eucalyptus Extract

Qualitative screening confirmed the presence of polyphenolics, flavonoids, tannins, saponins, and terpenoids in the aqueous extract, while alkaloids, steroids, and proteins were not detected, consistent with the polarity-selective recovery expected from aqueous extraction. Quantitative estimation yielded a total phenolic content of 174.52 ± 4.85 mg GAE/g and a total flavonoid content of 58.31 ± 2.14 mg QE/g of dry extract, confirming a substantial pool of hydroxyl-bearing bio-reducing molecules.

GC-MS profiling identified 1,8-cineole (eucalyptol) as the dominant constituent (46.85% relative peak area), together with α-pinene (14.22%), α-terpineol (9.18%), globulol (6.41%), and a gallic acid derivative (5.74%) (Table 1). The presence of the gallic acid derivative corroborates the phenolic character indicated by the TPC assay and supports the proposed role of hydroxyl-rich phenolics in Zn^{2+} bio-reduction.

Table 1. Major bioactive compounds identified in Eucalyptus leaf extract by GC-MS.

Compound	Relative Peak Area (%)	Class
1,8-Cineole (eucalyptol)	46.85	Monoterpenoid
α-Pinene	14.22	Monoterpenoid
α-Terpineol	9.18	Monoterpenoid alcohol
Globulol	6.41	Sesquiterpenoid
Gallic acid derivative	5.74	Phenolic acid

3.2 Optimization of Green Synthesis Parameters

Systematic variation of synthesis parameters established an extract-to-precursor ratio of 1:2 (v/v) with 0.10 M $Zn(NO_3)_2 \cdot 6H_2O$ as optimal, maximizing product recovery while avoiding the organic oversaturation and particle aggregation observed at higher relative extract loadings. Reaction progress, monitored visually and via UV-Vis spectroscopy, reached completion after 2.0 h of stirring at 70 °C under an alkaline reaction medium of pH 10. Calcination at 400 °C was identified as optimal, providing sufficient thermal energy to decompose the zinc hydroxide intermediate and eliminate excess organic matter, while avoiding the grain sintering and coarsening observed at 500–600 °C (Table 2).

Table 2. Optimum green synthesis conditions for ZnO nanoparticles.

Parameter	Optimum Value
Extract-to-precursor ratio (v/v)	1:2
Precursor concentration	0.10 M
Reaction pH	10
Reaction temperature	70 °C
Reaction duration	2.0 h
Calcination temperature	400 °C

3.3 Structural Analysis (XRD)

The XRD pattern of the optimized sample exhibited well-resolved diffraction peaks indexed to the hexagonal wurtzite phase of ZnO (JCPDS Card No. 36-1451), with no detectable secondary or impurity phases, confirming phase-pure crystallization. The average crystallite size, calculated from the (101) reflection using the Debye-Scherrer equation, was 18.35 nm, with lattice parameters a = 3.2497 Å and c = 5.2069 Å, in close agreement with standard bulk ZnO values, and a low micro-strain of 1.89×10^{-3}, indicative of minimal lattice distortion despite the nanoscale crystallite dimensions.

3.4 Morphological and Elemental Analysis (FE-SEM, HR-TEM, EDX)

FE-SEM and HR-TEM imaging revealed uniform, isotropic spherical to quasi-spherical nanoparticles with a narrow size distribution centred at 20.42 ± 2.58 nm, in close agreement with the XRD-derived crystallite size, indicating predominantly single-crystalline particles. EDX confirmed a high-purity composition with a near-stoichiometric Zn:O atomic ratio of 1:1.01 and a trace carbon signal of 4.24 wt%, attributable to the residual phytochemical capping layer.

3.5 Surface Chemistry and Colloidal Stability (FTIR, Zeta Potential, DLS)

Comparative FTIR analysis of the synthesized nanoparticles, referenced against the raw extract spectrum, confirmed the retention of deprotonated polyphenolic hydroxyl and conjugated carbonyl functionalities on the nanoparticle surface, alongside the characteristic Zn–O stretching vibration, providing direct spectroscopic evidence of phytochemical capping. This organic passivation layer conferred a strongly negative zeta potential of −32.40 mV, indicating good electrostatic colloidal

stability, and a narrow hydrodynamic size distribution (PDI = 0.214) with an average hydrodynamic diameter of 68.74 nm, the larger DLS value relative to the dry TEM size being consistent with the inclusion of the solvation and capping shell in the hydrodynamic measurement.

3.6 Thermal and Textural Analysis (TGA/DSC, BET)

TGA/DSC analysis revealed a distinct organic burnout stage between 150 and 400 °C, corresponding to the thermal decomposition of residual phytochemical capping constituents, with the material remaining thermally stable up to 800 °C thereafter. N_2 physisorption analysis produced a Type IV isotherm with an H3-type hysteresis loop, characteristic of a mesoporous architecture, with a BET specific surface area of 34.82 m^2/g and an average BJH pore diameter of 14.25 nm.

3.7 Optical Properties and Band Gap Narrowing (UV-Vis DRS, PL)

UV-Vis diffuse reflectance spectroscopy combined with Tauc plot extrapolation revealed an optical band gap of 3.25 eV for the green-synthesized ZnO nanoparticles, narrowed relative to the bulk ZnO value of 3.37 eV, corresponding to a measurable shift of the absorption edge toward the visible region. Room-temperature photoluminescence spectroscopy revealed a broad, high-intensity deep-level emission band centred at 535 nm (green emission), attributed to radiative recombination involving singly ionized oxygen vacancies (V_o^+). This defect-mediated emission, considered alongside the band gap narrowing observed by UV-Vis DRS, provides convergent evidence that the phytochemical capping layer introduces surface/sub-surface defect states into the ZnO electronic structure.

3.8 Proposed Mechanism of Bio-Reduction, Capping, and Band Gap Modification

Based on the integrated evidence presented in Sections 3.1–3.7, ZnO nanoparticle formation is proposed to proceed via initial complexation of Zn^{2+} ions with hydroxyl-bearing phenolic and flavonoid constituents of the extract, followed by bio-reduction to form a zinc hydroxide intermediate, phytochemical-mediated capping that restricts crystal growth, and thermally induced dehydration to yield crystalline ZnO. A portion of the capping layer remains bound to the nanoparticle surface following calcination, as confirmed by FTIR (Section 3.5) and EDX (Section 3.4), distorting the local surface coordination environment and generating oxygen vacancy defect states that narrow the effective optical band gap (Section 3.7). This mechanism establishes that the Eucalyptus phytochemical matrix functions as an active band-gap-engineering agent rather than a passive capping template, providing a mechanistic foundation for the visible-light photocatalytic application of this material, to be reported separately.

4. Conclusion

Aqueous Eucalyptus leaf extract, rich in phenolic and flavonoid constituents (TPC = 174.52 mg GAE/g; TFC = 58.31 mg QE/g), was successfully employed as a simultaneous reducing, capping, and

stabilizing agent for the green synthesis of phase-pure, hexagonal wurtzite ZnO nanoparticles (crystallite size 18.35 nm) under optimized conditions (1:2 extract-to-precursor ratio, 0.10 M precursor, pH 10, 400 °C calcination). The synthesized nanoparticles exhibited a mesoporous structure (34.82 m^2/g), good colloidal stability (−32.40 mV), and, most significantly, a narrowed optical band gap of 3.25 eV accompanied by defect-related visible emission, confirming that the residual phytochemical capping layer actively modifies the electronic structure of ZnO. These findings establish Eucalyptus-mediated green synthesis as a viable, dopant-free route to visible-light-responsive ZnO nanomaterials, with direct relevance to subsequent photocatalytic and environmental remediation applications.

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