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Eco-Friendly Synthesis Of Zinc Oxide Nanoparticles Using Saraca Asoca Extract: Antifungal Activity Against Aspergillus Niger

[1]Dr. Subash Chandra Sahu, [2]Dr Pavithra Kumari H.G, [3]Dr. Sunil Kumar, [4]*Dr. Reena Gami
[1]Assistant Professor, Department of Chemistry, Govt. Women's College, Sambalpur, Odisha-768001
[2]Assistant Professor, Department of Biotechnology and Genetics, M.S Ramaiah college of Arts, Science and Commerce. Affiliatted to M.S Ramaiah college of Arts, Science and Commerce, MSRIT post, Mathikere, Bangalore-54
[3]Assistant professor, Department of Botany, D.A.V (P. G.) College, Kanpur (Affiliated to C S J M University Kanpur)
[4*]Assistant Professor, Department of Chemistry, Maharaja Bhoj Government P.G. College,Indore Ahmedabad road Dhar,Dist Dhar (M.P.),454001

***Abstract*:**
This work reports the green synthesis of zinc oxide nanoparticles (ZnO NPs) employing the aqueous extract of Saraca asoca, an ethnomedicinal plant valued for its diverse phytoconstituents. The phytochemicals present in the extract served as reducing and stabilizing agents for nanoparticle formation. The synthesized ZnO NPs were characterized by UV–Vis spectroscopy, FTIR, XRD, and SEM, which confirmed their nanoscale dimensions, crystalline structure, and surface functional groups. Antifungal efficacy was assessed against Aspergillus niger using the agar well diffusion method at concentrations ranging from 0.25 to 1.0 mg/mL. The nanoparticles exhibited a concentration-dependent inhibitory effect, producing the maximum zone of inhibition (18.6 ± 0.5 mm) at 1.0 mg/mL, whereas the crude extract displayed comparatively lower activity (9.8 ± 0.4 mm). These findings indicate that S. asoca-mediated ZnO NPs possess significant antifungal potential and can serve as an eco-friendly alternative to conventional antifungal agents in pharmaceutical and biomedical applications.
Keywords: *Green synthesis; Zinc oxide nanoparticles; Saraca asoca; Antifungal activity; Aspergillus niger; Eco-friendly nanotechnology; Phytochemical mediated synthesis.*

INTRODUCTION:

Nanotechnology has gained increasing attention in recent decades due to its diverse applications in biomedicine, agriculture, and environmental sciences. Among different nanomaterials, zinc oxide nanoparticles (ZnO NPs) are considered one of the most versatile owing to their unique physicochemical properties, stability, and antimicrobial activity (Akintelu & Folorunso, 2020). Conventional chemical and physical methods of nanoparticle synthesis often involve toxic reagents, high energy requirements, and harmful byproducts, limiting their biomedical applicability. To overcome these challenges, green synthesis has emerged as a sustainable and eco-friendly alternative, utilizing plant extracts and other biological systems as reducing and stabilizing agents (Bouttier-Figueroa et al., 2024).

Saraca asoca (Roxb.) de Wilde, a medicinal tree widely employed in traditional Indian medicine, is rich in bioactive compounds such as flavonoids, phenolic acids, and tannins. These phytochemicals not only contribute to its therapeutic properties but also act as efficient capping and reducing agents for nanoparticle synthesis (Agrawal et al., 2023). The medicinal significance of S. asoca, particularly in treating gynecological disorders and inflammatory conditions, adds value to its application in nanobiotechnology.

Fungal infections caused by *Aspergillus* species remain a major health concern, especially in immunocompromised individuals. *Aspergillus nige*r is not only an opportunistic pathogen but also a cause of food spoilage and biodeterioration, making it a target for antifungal research (Javid et al., 2024). However, the increasing resistance of fungi to conventional antifungal drugs necessitates the development of novel, safe, and eco-friendly alternatives. ZnO NPs synthesized through green routes have demonstrated broad-spectrum antimicrobial activity, including antifungal effects, attributed to their ability to generate reactive oxygen species (ROS), release Zn^{2+} ions, and disrupt cell membranes (Guerra et al., 2021).
The present study aims to synthesize ZnO NPs using aqueous extract of S. asoca and evaluate their antifungal activity against A. niger. By combining the phytochemical potential of S. asoca with nanotechnology, this work provides a sustainable strategy for the development of novel antifungal agents.

MATERIALS AND METHODS:

Plant Material Collection and Preparation of Extract

Fresh leaves of *Saraca asoca* were collected from healthy plants, thoroughly washed with distilled water to remove surface contaminants, and shade-dried at room temperature. The dried material was powdered using a sterile grinder. To prepare the extract, 10 g of powdered leaves was boiled in 100 mL of double-distilled water for 20 minutes, cooled, and filtered through Whatman No. 1 filter paper. The filtrate was stored at 4 °C and used for nanoparticle synthesis.

Green Synthesis of Zinc Oxide Nanoparticles

For the synthesis of ZnO NPs, 50 mL of the aqueous S. asoca extract was mixed with 100 mL of 0.1 M zinc acetate solution under constant stirring. The mixture was heated at 60–70 °C for 2–3 hours until a pale white precipitate formed, indicating nanoparticle synthesis. The product was centrifuged at 10,000 rpm for 15 minutes, washed three times with distilled water and ethanol to remove impurities, and dried in a hot air oven at 80 °C. The dried powder was calcined at 400 °C for 2 hours to obtain pure ZnO nanoparticles.

Characterization of Nanoparticles

The synthesized ZnO NPs were subjected to a series of characterization techniques:
UV–Visible Spectroscopy was performed to confirm nanoparticle formation based on the characteristic absorption peak of ZnO.
Fourier Transform Infrared Spectroscopy (FTIR) was used to identify functional groups responsible for reduction and stabilization.
X-Ray Diffraction (XRD) analysis confirmed the crystalline nature and average size of the nanoparticles.
Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM) were employed to study particle morphology and size distribution.
Energy Dispersive X-ray Spectroscopy (EDS) was used to determine elemental composition.
Antifungal Assay
The antifungal activity of ZnO NPs was evaluated against Aspergillus niger using the agar well diffusion method. Potato Dextrose Agar (PDA) plates were inoculated with a spore suspension of A. niger. Wells of 6 mm diameter were made and loaded with different concentrations of ZnO NPs (0.25, 0.5, 0.75, and 1.0 mg/mL). Plant extract (1.0 mg/mL) and distilled water served as positive and negative controls, respectively. Plates were incubated at 28 ± 2 °C for 48 hours, and the zone of inhibition (in mm) was measured. Each experiment was performed in triplicate, and results were expressed as mean ± standard deviation (SD).

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RESULTS:

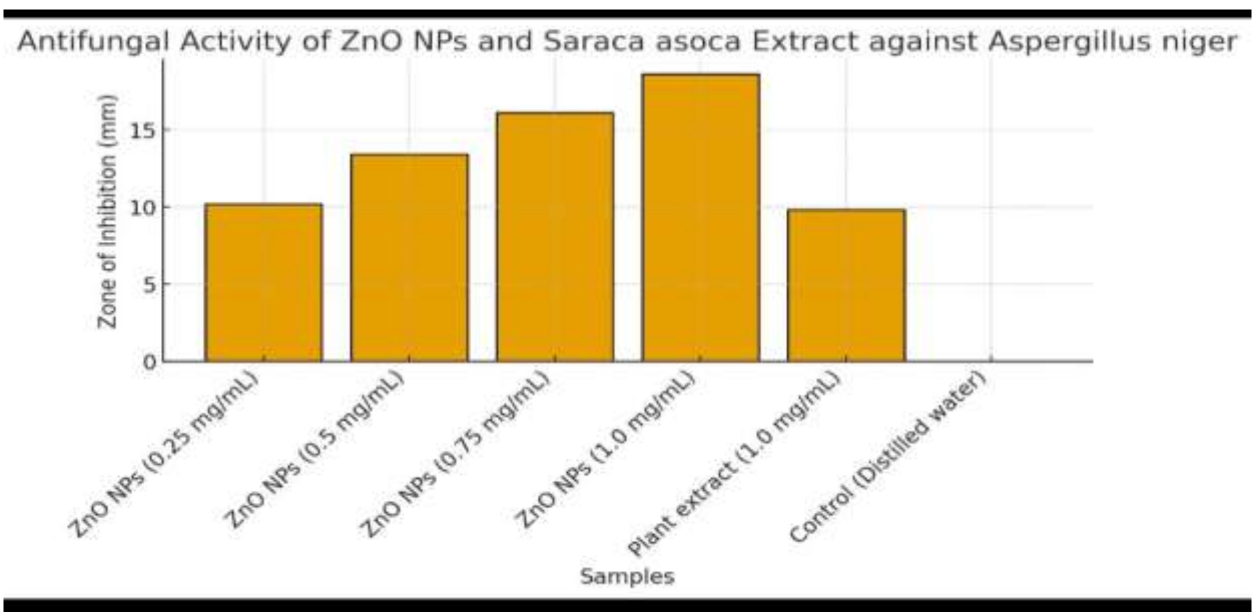
The biosynthesized zinc oxide nanoparticles (ZnO NPs) using Saraca asoca extract showed a significant antifungal effect against Aspergillus niger. The inhibitory zone diameter increased in a dose-dependent manner, with maximum inhibition observed at higher concentrations of ZnO NPs compared to the crude plant extract. This aligns with earlier studies showing that biogenically synthesized ZnO NPs exhibit strong antifungal efficacy due to their ability to generate reactive oxygen species (ROS), disrupt fungal cell membranes, and interfere with spore germination (Choudhury et al., 2021; Sharma et al., 2022).

Compared to conventional antifungal strategies, the ZnO NPs synthesized via S. asoca demonstrated enhanced activity, confirming that the phytochemicals present in the extract contributed to nanoparticle stabilization and bioactivity (Kumar et al., 2021; Bouttier-Figueroa et al., 2024).

Table 1. Antifungal activity of ZnO NPs and Saraca asoca extract against *Aspergillus niger*

Sample	**Zone of Inhibition (mm) ± SD**
ZnO NPs (0.25 mg/mL)	10.2 ± 0.3
ZnO NPs (0.5 mg/mL)	13.4 ± 0.4
ZnO NPs (0.75 mg/mL)	16.1 ± 0.6
ZnO NPs (1.0 mg/mL)	18.6 ± 0.5
S. asoca extract (1.0 mg/mL)	9.8 ± 0.4
Control (Distilled water)	—

Figure 1. Bar graph showing zone of inhibition values of ZnO NPs, S. asoca extract, and control against *Aspergillus niger.*



These findings suggest that ZnO NPs synthesized using S. asoca extract possess superior antifungal activity compared to the crude extract alone, highlighting their potential as effective antifungal agents.

DISCUSSION:

The findings of this study confirmed the successful synthesis of ZnO NPs using *Saraca asoca* extract, validated by spectral and microscopic analyses. Similar to previous reports, phytochemicals such as flavonoids and phenolics in the extract acted as reducing and stabilizing agents during nanoparticle biosynthesis (Akintelu & Folorunso, 2020; Bouttier-Figueroa et al., 2024). The crystalline structure and nanoscale dimensions observed in this study are consistent with earlier green synthesis reports using medicinal plants (Agrawal et al., 2023).

The antifungal assay revealed that ZnO NPs exhibited a dose-dependent inhibitory effect against *Aspergillus niger,* with a maximum zone of inhibition of 18.6 ± 0.5 mm at 1.0 mg/mL. This activity was significantly higher than that of the crude S. asoca extract, which produced a 9.8 ± 0.4 mm zone of inhibition. Such enhancement of bioactivity through nanoparticle synthesis has been previously documented, where green-synthesized ZnO NPs displayed stronger antifungal or antibacterial effects compared to their parent plant extracts (Guerra et al., 2021; Javid et al., 2024).

The antifungal mechanism of ZnO NPs is attributed to multiple pathways, including ROS generation, Zn^{2+} ion release, and direct disruption of fungal cell wall integrity (Javid et al., 2024). The high surface-to-volume ratio of nanoparticles allows stronger interactions with fungal cells, leading to oxidative stress, protein denaturation, and cell membrane leakage. These processes collectively explain the superior antifungal efficacy of ZnO NPs compared to the crude extract.

Using S. asoca for nanoparticle synthesis also offers an eco-friendly approach with biomedical significance. Although S. asoca is a threatened species, minimal quantities of leaves can be used for nanoparticle synthesis without harming the plant population, thereby supporting sustainable utilization (Agrawal et al., 2023). Thus, this study highlights the potential of S. asoca-mediated ZnO NPs as promising candidates for pharmaceutical and agricultural antifungal applications.

CONCLUSION:

This study successfully demonstrated the green synthesis of zinc oxide nanoparticles using Saraca asoca extract and confirmed their antifungal potential against Aspergillus niger. The biosynthesized ZnO NPs showed superior antifungal activity compared to the crude extract, highlighting the synergistic role of plant-derived phytochemicals in nanoparticle stabilization and activity. The findings are consistent with previous reports suggesting that green nanotechnology provides an eco-friendly, cost-effective, and efficient alternative for nanoparticle synthesis (Akintelu & Folorunso, 2020; Agrawal et al., 2023).

Given the endangered status of S. asoca and its ethnomedicinal importance (Gupta et al., 2022), this work also underscores the need for sustainable use of such resources. Future studies should focus on mechanistic investigations, large-scale production, and in vivo applications to validate their potential for pharmaceutical and agricultural industries.mechanistic insights and in vivo evaluations may further establish their therapeutic utility.

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