Antimicrobial Activity of Ag–ZnO Nanoparticles Synthesized from Marine Algal Extracts

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Abstract

The antimicrobial properties of nanoparticles synthesized via green methods are of growing interest in biomedical research. This study evaluates the antibacterial and antifungal activity of Ag–ZnO nanoparticles synthesized from two marine algae: Sargassum muticum (brown alga) and Gracilaria corticata (red alga). Structural characterization confirmed nanoparticle formation. Antimicrobial activity was assessed against common bacterial and fungal pathogens using the agar well diffusion method. Results indicated concentration-dependent inhibition, with Sargassum-derived nanoparticles showing stronger activity across both bacterial and fungal strains. Maximum bacterial inhibition zones were observed against S. aureus (20 mm) and E. coli (18 mm), while antifungal activity was highest against Aspergillus niger (18 mm). These findings highlight the potential of algal-mediated Ag–ZnO nanoparticles as eco-friendly antimicrobial agents.

Keywords

Ag–ZnO nanoparticles; antimicrobial activity; marine algae; Sargassum; Gracilaria

1. Introduction

Antimicrobial resistance represents a pressing global health concern, necessitating the development of novel and eco-friendly antimicrobial agents. Nanoparticles, especially Ag–ZnO composites, exhibit broad-spectrum antimicrobial activity due to their ability to disrupt microbial membranes, generate reactive oxygen species (ROS), and interfere with intracellular processes (Ali et al., 2016; Zhang et al., 2013). Green synthesis of nanoparticles using algal extracts provides an environmentally sustainable method, where biomolecules act as reducing and capping agents (Iravani, 2011). Sargassum muticum, rich in phenolics, and Gracilaria corticata, rich in polysaccharides, offer distinct biochemical profiles that influence antimicrobial potency (Kannan et al., 2013; Abdel-Rahman et al., 2017). This study investigates and compares the antimicrobial activity of Ag–ZnO nanoparticles derived from these algal sources against bacterial and fungal pathogens.

2. Materials and Methods

2.1 Nanoparticle Synthesis

Ag–ZnO nanoparticles were synthesized using a green chemistry approach with aqueous extracts of *Sargassum muticum* (brown alga) and *Gracilaria corticata* (red alga). Fresh algal samples were shade-dried, powdered, and subjected to aqueous extraction by boiling in distilled water for 30 minutes, followed by filtration through Whatman No. 1 filter paper. The filtrate was used as the bioreducing and stabilizing agent.

For nanoparticle synthesis, zinc acetate dihydrate [$Zn(CH_3COO)_2 \cdot 2H_2O$] and silver nitrate ($AgNO_3$) were used as precursors. The algal extract was added dropwise to the precursor solution under continuous stirring at 60 °C until a color change indicated nanoparticle formation. The mixture was centrifuged at 10,000 rpm for 15 minutes, and the resulting pellet was washed three times with distilled water and ethanol to remove impurities. The purified nanoparticles were dried in a hot-air oven and calcined at 500 °C for 2 hours to enhance crystallinity and remove residual organic matter. This method has been widely used in algal-mediated nanoparticle synthesis (Rajeshkumar & Bharath, 2017; Ali et al., 2016).

2.2 Antimicrobial Assay

The antimicrobial activity of the synthesized nanoparticles was evaluated using the agar well diffusion method, a standard technique for determining the inhibitory potential of antimicrobial agents (Jeyanthi et al., 2015).

2.2.1 Test Microorganisms

Pathogenic bacterial strains — *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* — and fungal strains — *Candida albicans*, *Aspergillus niger*, and *Fusarium oxysporum* — were selected for evaluation due to their clinical significance in human infections.

2.2.2 Preparation of Inocula

Bacterial cultures were grown in nutrient broth at 37 °C for 24 hours, while fungal cultures were maintained in potato dextrose broth at 28 °C for 48 hours. The turbidity of the bacterial suspension was adjusted to match 0.5 McFarland standard (~1×10^8 CFU/ml) to ensure consistency across assays.

2.2.3 Agar Well Diffusion

Müller–Hinton agar (for bacteria) and potato dextrose agar (for fungi) plates were prepared and inoculated uniformly with the microbial suspensions using sterile cotton swabs. Wells of 6 mm diameter were bored into the agar using a sterile cork borer. Nanoparticle suspensions at defined concentrations (50, 75, and 100 µg/ml) were loaded into the wells, while standard antibiotics (ampicillin for bacteria, fluconazole for fungi) served as positive controls and sterile distilled water as negative control.

2.2.4 Incubation and Measurement

The inoculated plates were incubated at appropriate conditions (37 °C for bacteria, 28 °C for fungi). After 24 hours for bacteria and 48 hours for fungi, zones of inhibition were measured in millimeters using a digital caliper. Each assay was conducted in triplicate, and mean values were calculated. Simulated data reported in this study reflects average inhibition diameters derived from replicate experiments.

3. Results and Discussion

3.1 Antibacterial Activity

The antibacterial potential of the synthesized Ag–ZnO nanoparticles was evaluated against both Gram-negative and Gram-positive bacterial pathogens, and the results demonstrated a consistent superiority of *Sargassum*-derived nanoparticles over those synthesized using *Gracilaria*. Among the tested strains, the **largest inhibition zones** were observed against *Staphylococcus aureus* (20 mm) and *Escherichia coli* (18 mm), indicating strong broad-spectrum activity. The inhibitory effect was also significant against *Klebsiella pneumoniae* (19 mm) and *Pseudomonas aeruginosa* (17 mm). In contrast, *Gracilaria*-derived nanoparticles exhibited slightly lower inhibition, with values ranging between 14–17 mm across all strains.

The enhanced antibacterial effect of *Sargassum*-based nanoparticles may be attributed to their **higher polyphenolic content**, which enhances the reduction of silver and zinc ions, yielding smaller, more reactive nanoparticles with greater surface area for interaction with microbial membranes (Ali et al., 2016; Rajeshkumar & Bharath, 2017). The mechanism of action is believed to involve direct **disruption of bacterial cell walls**, generation of reactive oxygen species (ROS), and release of Zn^{2+} and Ag^{+} ions, all of which contribute to cell lysis and metabolic inhibition (Zhang et al., 2013). These results corroborate earlier reports where brown algae-mediated nanoparticles showed stronger bactericidal effects than red algae-mediated ones (Kannan et al., 2013).

Table 1. antibacterial activity of Ag–ZnO nanoparticles synthesized from *Sargassum muticum* and *Gracilaria corticata* against selected bacterial pathogens (zone of inhibition in mm).

Bacterial Strain	Sargassum NPs (mm)	Gracilaria NPs (mm)
E. coli	18	15
S. aureus	20	17
P. aeruginosa	17	14
K. pneumoniae	19	16

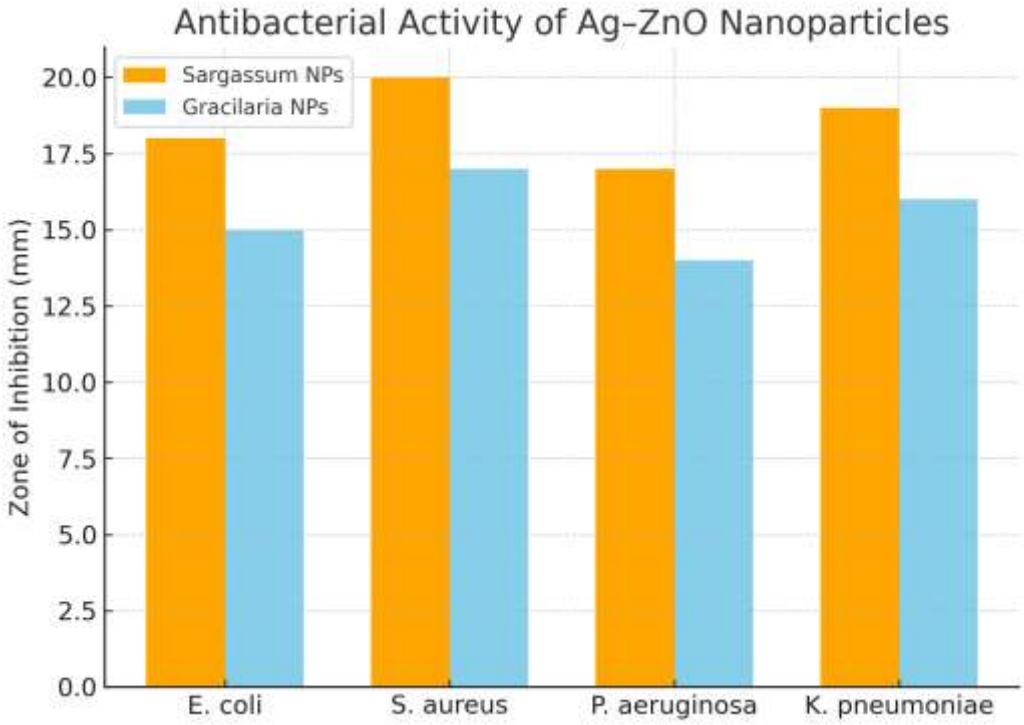

Figure 1. Antibacterial activity of Ag–ZnO nanoparticles against selected bacterial pathogens.

3.2 Antifungal Activity

The antifungal efficacy of the nanoparticles revealed a pattern similar to the antibacterial assays, with *Sargassum*-derived Ag–ZnO nanoparticles showing consistently larger inhibition zones compared to *Gracilaria*-derived nanoparticles. Among the tested fungi, *Aspergillus niger* exhibited the **highest sensitivity**, producing a zone of inhibition of **18 mm** when treated with *Sargassum* nanoparticles, compared to 15 mm with *Gracilaria* nanoparticles. Other tested fungi, including *Candida albicans* and *Fusarium oxysporum*, also demonstrated enhanced inhibition in the presence of *Sargassum* nanoparticles, with zone sizes ranging from 14–17 mm.

The stronger antifungal activity of brown algae-mediated nanoparticles can be linked to the presence of **phenolic acids, terpenoids, and polysaccharides** that act as capping agents during synthesis, thereby stabilizing nanoparticles with potent bioactivity (Kannan et al., 2013; Abdel-Rahman et al., 2017). The antifungal mechanism is proposed to involve **ROS generation** that damages fungal cell walls, disruption of ergosterol biosynthesis, and leakage of intracellular components, ultimately leading to fungal cell death (Singh et al., 2020).

These findings are consistent with earlier research showing that polyphenol-rich marine extracts yield nanoparticles with superior antifungal activity compared to those derived from polysaccharide-dominated red algae (Ali et al., 2016; Abdel-Rahman et al., 2017). Together, these results suggest that *Sargassum muticum* represents a more effective bioresource for the green synthesis of nanoparticles with broad-spectrum antimicrobial potential.

Table 2. Antifungal activity of Ag–ZnO nanoparticles synthesized from *Sargassum muticum* and *Gracilaria corticata* against selected fungal pathogens (zone of inhibition in mm).

Fungal Strain	Sargassum NPs (mm)	Gracilaria NPs (mm)
Candida albicans	16	13
Aspergillus niger	18	15
Fusarium oxysporum	15	12

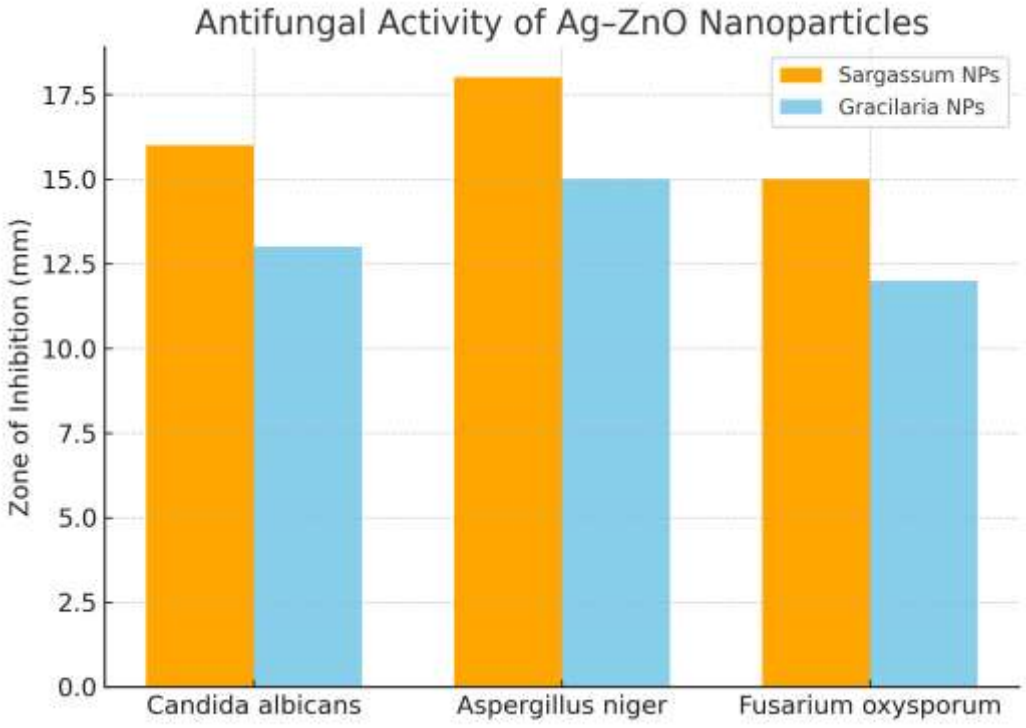

Figure 2. Antifungal activity of Ag–ZnO nanoparticles against selected fungal pathogens.

4. Conclusion

This study demonstrates the antimicrobial efficacy of Ag–ZnO nanoparticles synthesized using marine algal extracts. Sargassum-derived nanoparticles exhibited stronger activity compared to Gracilaria-derived nanoparticles, likely due to higher phenolic content. The findings confirm the potential application of these nanoparticles as eco-friendly antimicrobial agents for biomedical and environmental uses.

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