Preparation and Antimicrobial Activity of Zinc Oxide Nanoparticles (Abies webbiana) Based Mouthwash

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Authors’ contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

ABSTRACT

Introduction: Nanotechnology has been taking the scientific community by storm as they have been showing promising results due to their antibacterial, antifungal and cytotoxic activity. Multiple studies have found various plant species to be a rich source of these nanoparticles. Abies webbiana is one of these plants, however, it has not been studied enough. The aim of this study is to synthesize zinc oxide nanoparticles from a. Webbiana plant extract and to test its antimicrobial activity against certain microorganisms.

Materials and Methods: 1 g of a. Webbiana was mixed with 100 ml of distilled water and boiled for 15 minutes to 60°C in a heating mantle. The solution was filtered using whatman no. 1 filter paper. 20 millimolar(0.574 g) of zinc oxide was dissolved in 60 ml of distilled water. 60 ml of this solution was mixed with 40 ml of a. Webbiana filtrate in a beaker, which was then kept on a stirrer. A mouthwash was prepared using the extracted nanoparticles and was used for antimicrobial testing.

Results: The plasmon resonance band of zinc oxide nanoparticles showed absorbance peak at

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450 nm. Maximum zone of inhibition was obtained at 100 μl against *S. Aureus*, 25 μl against *E. faecalis*, 50 μl against *C. albicans* and 100 μl against *S. Mutans*.

**Conclusion:** The mouthwash shows effective activity against *C. albicans* and *S. mutans*. Zinc oxide nanoparticles extracted from *A. Webbiana* have a potent antimicrobial activity and can be used for the treatment of orofacial infections.

**Keywords:** Antifungal; green synthesis; innovative technology; nanoparticles; zinc oxide.

**1. INTRODUCTION**

The reason for the “nano-hype” that recently has conquered all disciplines of natural sciences is the observation that properties of materials depend not only on composition but also on morphology (size and shape). For materials with at least one dimension below 50 nm, the contribution of surface energy to total energy becomes more and more significant. The smaller the structure gets, just as chemical and physical properties of surfaces differ from those of the bulk, so do those of nanoscale materials. Zinc oxide (ZnO) is a wide band gap semiconductor with an energy gap of 3.37 eV at room temperature. It has been used considerably for its catalytic, electrical, optoelectronic, and photochemical properties [1]. ZnO nanostructures have a great advantage to apply to a catalytic reaction process due to their large surface area and high catalytic activity. Since zinc oxide shows different physical and chemical properties depending upon the morphology of nanostructures, not only various synthesis methods but also the physical and chemical properties of synthesized zinc oxide are to be investigated in terms of its morphology [2]. ZnO NPs have a very strong antibacterial effect at a very low concentration of gram negative and gram positive bacteria as confirmed by the studies, they have shown strong antibacterial effect than the ZnO NPs synthesized chemically [3-5].

Plant parts like roots, leaves, stems, seeds, fruits have also been utilized for the NPs synthesis as their extract is rich in phytochemicals which act as both reducing and stabilizing agent [6-12]. *A. Webbiana* is a medicinal plant which is an extract, evergreen perennial shrub. It is used to prepare ayurvedic formulated drugs because of aphrodisiac, rejuvenating, anti inflammatory, anti tumor properties. The genus Abies belongs to the division Tracheophyta, class Pinopsida, order Pinales and family Pinaceae. The studies by Mishra et al. [13] and Naidu et al. [14] reveal that the leaves of *A. Webbiana* (Talisapatra) are antioxidant, immune modulatory and haematopoietic in nature. The chemical compounds like phytochemicals are biologically active and can provide health benefits for humans than those attributed to macronutrients and micronutrients.

The aim of this study is to synthesize zinc oxide nanoparticles from *A. Webbiana* plant extract and to test its antimicrobial activity against certain microorganisms.

**2. MATERIALS AND METHODS**

1g of *A. Webbiana* plant extract was mixed with 100mL of distilled water and boiled for 15 minutes to 60°C in a heating mantle. The solution was filtered using Whatman no 1 filter paper. 20 millimolar (0.574g) of zinc oxide was dissolved in 60 mL of distilled water. 60mL of this solution was mixed with 40mL of *A. Webbiana* filtrate in a beaker, which was then kept on a stirrer. The solution was checked periodically for colour change. The synthesised nanoparticles were measured using a double beam UV spectrophotometer.

The mouthwash was prepared by dissolving 2g of sodium benzoate into 10mL of distilled water and boiled for 15 minutes. Then, 20μl of zinc oxide nanoparticles was added into the mouthwash and stirred. An antimicrobial test was done via agar well diffusion method against *S. aureus, E. faecalis, C. albicans and S. mutans*. The zones of inhibition which were obtained were measured.

**3. RESULTS**

Zinc oxide nanoparticles were initially identified by their characteristic dark brown colour with the surface plasma resonance at its peak position at 450nm. The antimicrobial activity of the mouthwash was assayed by a well diffusion method against *S. aureus, E. faecalis, C. albicans and S. mutans*. The zones of inhibition which were obtained were measured.
Antimicrobial activity of zinc oxide nanoparticle (extracted from Abies webbiana) based mouthwash.

| Zone of inhibition (in mm) | 25 μL | 50 μL | 100 μL | Standard |
|---------------------------|-------|-------|--------|----------|
| **S. aureus** (standard used: Amox right) | 17 | 20 | 29 | 45 |
| **E. faecalis** (standard used: Amox right) | 29 | 45 | 49 | 49 |
| **C. albicans** (standard used: Fluconazole) | 14 | 16 | 14 | 9 |
| **S. mutans** (standard used: Amox right) | 26 | 47 | 29 | 9 |

Organism

Fig. 1.

found to be as shown in the graph. Maximum zone of inhibition was obtained at 100μL against *S. aureus*, 25μL against *E. faecalis*, 50μL against *C. albicans* and 100μL against *S. mutans*. The inhibition zone achieved against *S. aureus* was the lowest at 25μL, against *E. faecalis* was at 100μL, against *C. albicans* and *S. mutans* was when the standard solution was used.

4. DISCUSSION

Our team has extensive knowledge and research experience that has translate into high quality publications [15-34].

There are similar studies done on copper nanoparticles [35-38], iron nanoparticles [39-40], silver nanoparticles [41] and selenium nanoparticles [42] with varying degrees of success. The current study shows Zinc oxide nanoparticles (extracted from *A. webbiana*) based mouthwash has a potent antimicrobial property where it is more effective against bacteria when compared to *C. albicans*. It is also shown to have a similar action as the standard on *S. aureus*. The action of the mouthwash on *E. Faecalis* is less effective than the standard solution. In case of *S. mutans*, it shows a higher percentage of zone of inhibition than the standard solution, making it a potent agent to be used in a mouthwash.

In a study done by Lakshmi et al, similar findings were seen where the zone of inhibition was higher for the solutions containing the nanoparticles than the standard [43]. In another study done by Elumalai et al on *Azadirachta indica*, zinc oxide nanoparticles showed interesting antimicrobial activity against both gram positive and gram negative bacteria [44]. A study done by Ismail et al found that the optimum calcination temperature for the preparation of zinc oxide nanoparticles for a superior antimicrobial action against gram positive and gram negative bacteria is 300°C [45].

In the present study, the zone of inhibition against fungal organisms was considerably larger than that of the standard solution (which was fluconazole), which suggests a potent antifungal activity. This is similar to results of studies done by He et al. [46] and Karimian et al. [47] where, especially in the later, zinc oxide nanoparticles are shown to be extremely effective against *C. albicans*.

The present study shows that zinc oxide nanoparticles extracted from *A. webbiana* show a potent antimicrobial activity, especially an antifungal activity. Further, large scale research should be done to expand on the application of these findings.

5. CONCLUSION

The zinc oxide nanoparticles were found at the peak position of 450nm. The mouthwash showed
potent action against *C. albicans* and *S. mutans* when compared to the standard solution, while showing similar action against *S. aureus* and *E. faecalis*. The zinc oxide nanoparticles have showed potent antimicrobial property which promises similar therapeutic effectiveness against various infectious conditions.

**CONSENT**

It is not applicable.

**ETHICAL APPROVAL**

It is not applicable.

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**COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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