

Green Synthesis of ZnO Nanoparticles using *Mesua ferrea* Leaves Extract and Its Antimicrobial Activity

Anushree Jatrana ^{a*}, Sonu Chauhan ^a, Sheetal Maan ^a and Monika Kayasth ^b

^a Department of Chemistry, Chaudhary Charan Singh Haryana Agricultural University,
Hisar, Haryana-125001, India.

^b Department of Microbiology, Chaudhary Charan Singh Haryana Agricultural University,
Hisar, Haryana-125001, India.

Authors' contributions

This work was carried out in collaboration among all authors. Author AJ designed and conducted the synthesis and characterization, managed the literature searches, wrote the protocol and wrote the first draft of the manuscript. Authors SC and SM managed the analysis of the study. Author MK conducted the biological activity. All authors read and approved the final manuscript.

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ABSTRACT

Aims: Green synthesis of nanoparticles using plant extracts has become an emerging field of nanotechnology because some of the essential phytochemicals present in plant extract, helps in formulating and enhancing the bioactivity, functionality and compatibility of the nanoparticles. In this study, the green synthesis of ZnO nanoparticles using plant extract and its antimicrobial activity was explored.

Place of Study: The study was conducted at Functionalized Materials Research Lab, Department of Chemistry, CCSHAU Hisar, Haryana, India.

Methodology: In this study, ZnO nanoparticles were synthesized using *Mesua ferrea* leaves extract as a stabilizing agent. The synthesized nanoparticles were characterized by FT-IR (Fourier Transform Infrared Spectroscopy), XRD (X-Ray Diffraction) and FE-SEM (Field Emission Scanning Electron Microscopy) techniques. The results indicated the successful synthesis of ZnO nanoparticles. The synthesized ZnO nanoparticles were further used for the evaluation of their antibacterial and antifungal activity against *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively.

Results: The synthesized ZnO nanoparticles possess characteristic FTIR peak at 445 cm^{-1} corresponding to Zn-O stretching vibration. XRD revealed that the average crystallite size of ZnO was 20 nm. The ZnO nanoparticles exhibited good antibacterial and antifungal activity with a maximum inhibition zone of 1.5 cm and 2.0 cm against the growth of *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively.

Conclusion: The *Mesua ferrea* plant have some inherent medicinal properties. The ZnO nanoparticles synthesized using plant extract possess antibacterial and antifungal activity due to the synergistic effect of the ZnO nanoparticles and the plant extract.

Keywords: Nanoparticles; zinc oxide; plant extract; antibacterial; antifungal.

1. INTRODUCTION

Nanotechnology is an emerging technology which can bring a revolution in almost all scientific fields because of surface and morphological properties of nanomaterials. Nanoparticles can be developed using different techniques like the “bottom up” and “top-down” approaches. Nowadays, bio resource based synthesis of nanoparticles is gaining interest due to reduced toxic effects as compared to the other chemical and physical synthesis approaches. Generally, metal and metal oxide nanoparticles are synthesized using plant extract assisted method [1]. These metal oxide nanoparticles possess promising applications in almost all scientific domains such as environment, energy, medicine, electronics, optics, catalysis, sensors, agriculture etc. This green approach for synthesis of metal and metal oxide nanoparticles involves the reduction of metal salts in the presence of plant extract. The biomolecules present in the plant extract can reduce the metal ions from positive oxidation state to less positive or zero oxidation state and can also stabilize the formed nanoparticles by acting as capping agents [2]. Generally, the size of formed nanoparticles depends on the strength of reductant, stronger the reductant, higher will be the reaction rate and will result in smaller size [3].

Since ancient times, *Mesua ferrea* plant extract is used for medicinal purpose due to its inherent properties such as central nervous system depressant, analgesic, antimicrobial, antivenom, antioxidant, antispasmodic, anti-inflammatory and immunostimulant [4]. Its use for the synthesis of nanoparticles will enhance the antimicrobial activity of nanoparticles through synergistic effect. In this study, we developed ZnO nanoparticles using *Mesua ferrea* plant extract and studied its antibacterial and antifungal properties against *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively.

2. MATERIALS AND METHODS

2.1 Chemicals

Mesua ferrea (Nagkesar) leaves were collected from Botanical garden of Department of Botany, C.C.S. University Meerut, and zinc nitrate was procured from Central Drug House (P) Ltd.

2.2 Preparation of Leaves Extract

The *Mesua ferrea* leaves were collected, washed, shadow dried and chopped into small pieces. About 20 g of leaves were taken in a beaker containing 100 mL distilled water and then heated at 70 to 80°C for 30 min. The yellow coloured extract was filtered twice and stored in the refrigerator.

2.3 Synthesis of ZnO using Leaves Extract

Green synthesis of ZnO was carried out using the different amounts of *Mesua ferrea* extract. 30 mL, 60 mL, 90 mL, 120 mL of extract was taken in different beakers, and 1 g of zinc nitrate was added to each beaker. The mixture was boiled until the formation of brown colored paste. The paste was transferred to a ceramic crucible followed by heating in muffle furnace at 400°C for 2 h.

2.4 Characterization Techniques

The Infrared induced vibrations were recorded on a Thermo Scientific spectrometer (Nicolet-is 50), at a scan speed of 4 cm^{-1} from 4000 to 400 cm^{-1} . The XRD analysis was performed on Bruker AXS D8 diffractometer equipped with $\text{CuK}\alpha$ radiation ($\lambda = 0.154\text{ nm}$). The diffractogram was collected in the 2θ range of 20-80°, at a scanning rate of 2° min^{-1} . The surface morphology of MOF was studied by Quanta 200F microscope FE-SEM with an accelerating voltage of 20 kV.

2.5 Antimicrobial Activity of ZnO Nanoparticles

The antimicrobial activity of the ZnO nanoparticles synthesized using *Measua ferrea* extract (30 ml), was analyzed against *Pseudomonas aeruginosa* and *Aspergillus awamorii* by using agar well diffusion method. Control experiment was carried out by using standard drugs i.e., chloramphenicol for antibacterial activity against *Pseudomonas aeruginosa* and nystatin for antifungal activity against *Aspergillus awamorii* as standard drugs.

3. RESULTS AND DISCUSSION

3.1 FTIR Results

FTIR analysis was performed for the identification of synthesized ZnO nanoparticles. Fig. 1 shows the FTIR spectra of zinc oxide nanoparticles, which are synthesized using different amounts of the plant extract. Generally, metal oxides have characteristic peaks below 1000 cm^{-1} (fingerprint region). It was observed that the characteristic peak for Zn-O stretching vibration band is present at 445 cm^{-1} indicating the successful synthesis of ZnO nanoparticles [5]. A broad peak in the range of $2900\text{-}3600\text{ cm}^{-1}$ is also found in the sample [6]. This region can be assigned to NH_2 , OH, COOH, OCH_3 and C-H aromatic stretching vibrations in the bioactive compound. The absorption region between 1000 to 1700 cm^{-1} belongs to the C=N, C=C and C=O stretching. The broadening and shifting of peaks

was observed with the increasing amount of extract from 30 to 120 mL (Fig. 1).

3.2 XRD Results

XRD results of ZnO nanoparticles synthesized with different amount of *Mesua ferrea* leaves extract are presented in Fig. 2. All the samples exhibited characteristic ZnO reflections, indicating its successful synthesis. It was observed that there exists strong diffraction peaks (2θ) at 31.8° , 34.49° , 36.2° , 47.6° , 56.6° , 62.7° , 66.4° , 67.8° and 69.3° corresponding to the (100), (002), (101), (102), (110), (103), (200), (112) and (201) crystal planes, respectively (Fig. 2). These diffraction peaks correspond to the characteristic hexagonal wurtzite structure of ZnO nanoparticles [7,8]. Similar XRD patterns were reported by [9,10]. The crystallite size for all samples is presented in Table 1. Crystallite size of synthesized nanoparticles indicated that 30 ml of *Mesua ferrea* leaves extract was sufficient for synthesis of ZnO nanoparticles with crystallite size of around 20 nm.

3.3 FESEM Results

Fig. 2 Shows the FESEM micrograph of ZnO prepared by using 30 ml of *Mesua ferrea* leaves extract. The results indicated the presence of near spherical aggregates of ZnO NPs, indicating that particles are held together because of weak forces. This agglomeration was probably due to surface polarity and electrostatic attraction of ZnO nanoparticles [11].

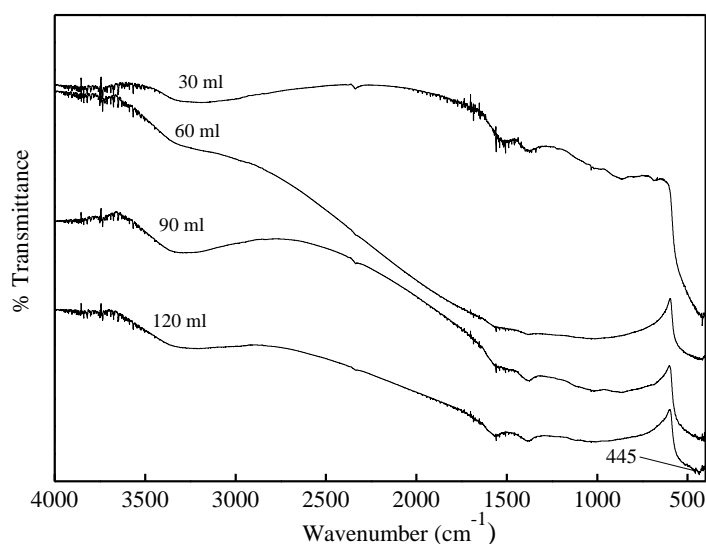


Fig. 1. FT-IR spectra of ZnO synthesized with different amount of extract

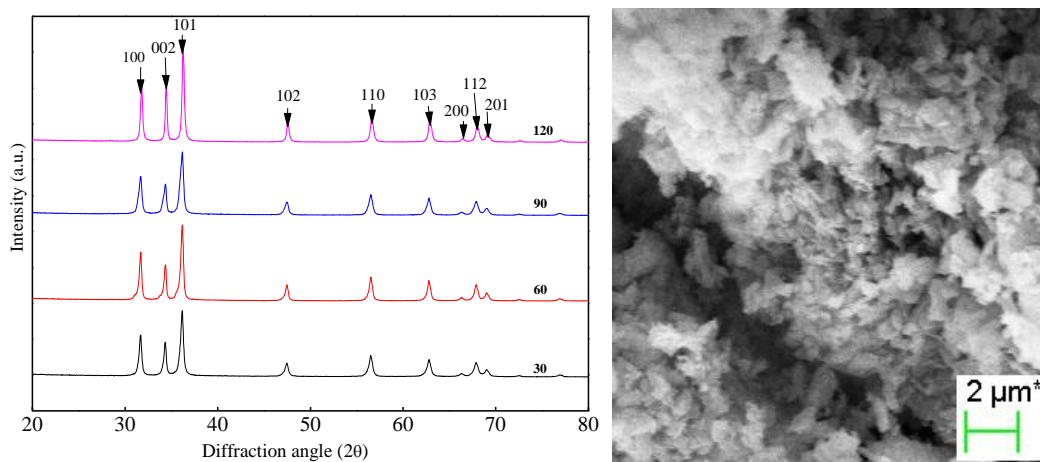


Fig. 2. XRD of ZnO synthesized with *Mesua ferrea* leaves extract and FESEM of ZnO (30 mL)

Table 1. Crystallite size of synthesized ZnO NPs

Volume of extract	Peak position (2θ)	FWHM	Crystallite size D (nm)
30 ml	36.1	0.427	19.5
60 ml	36.1	0.375	22.3
90 ml	36.1	0.472	17.7
120 ml	36.2	0.309	26.9

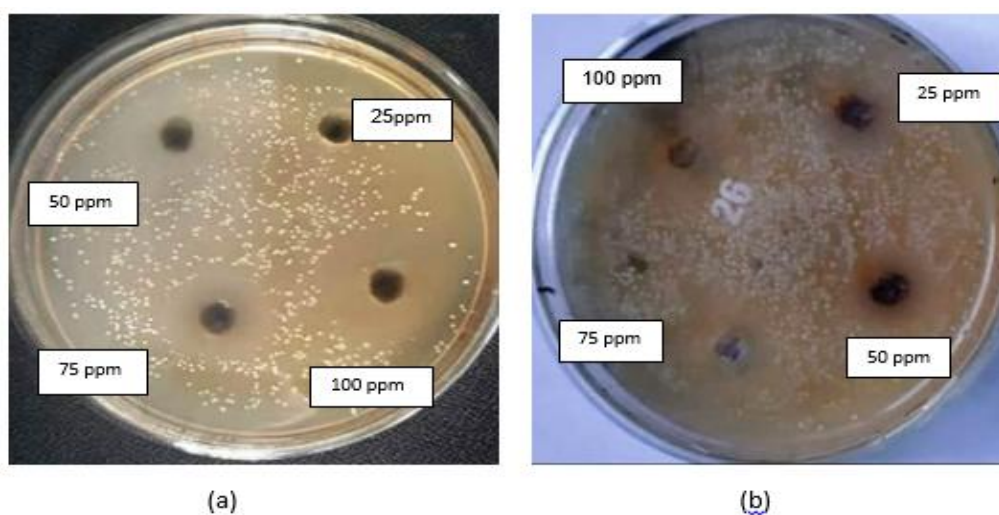


Fig. 3. (a) Antibacterial activity against *P.aeruginosa* (b) antifungal activity against *Aspergillus awamorii* at different concentrations

Table 2. Antimicrobial activity of ZnO NPs

Concentration in ppm	<i>Pseudomonas aeruginosa</i>	<i>Aspergillus awamorii</i>
	Zone of inhibition in cm	
25	-	0.25
50	0.5	0.45
75	1	1
100	1.5	2

3.4 Antimicrobial Activity of ZnO Nanoparticles

The antimicrobial activity of ZnO nanoparticles synthesized with 30 ml of *Mesua ferrea* leaves extract was evaluated at different concentrations, i.e., 25, 50, 75 and 100 ppm. The zones of inhibition against *Pseudomonas aeruginosa* and *Aspergillus awamorii* are presented in Table 2. The results obtained during the analysis of antimicrobial activity (Fig. 3), indicated that the synthesized ZnO nanoparticles possess very good antibacterial and antifungal activity against *Pseudomonas aeruginosa*, *Aspergillus awamorii*. It was observed that there was an increase in the zone of inhibition with the increasing concentration (ppm) of the ZnO nanoparticles. Results indicated that the increased concentration of ZnO nanoparticles upto 100 ppm presented positive effect on its antimicrobial activity.

This antibacterial activity may be due to the larger surface area of ZnO nanoparticles, giving more contact area with the microorganism and due to the insertion of the reactive oxygen species (generated from ZnO nanoparticles) into the small pores of microbial cell wall, leading to the leakage of minerals, proteins and other cell materials and hence inhibiting the cell growth [12].

4. CONCLUSION

In this study, a green approach was utilized for the synthesis of ZnO nanoparticles, where *Mesua ferrea* leaves extract was taken as stabilizing and capping agent. The successful synthesis of ZnO nanoparticles was confirmed by the bands obtained in FT-IR spectrum, while the crystallite size was obtained by XRD analysis. FESEM results indicated the presence of near spherical aggregates of ZnO NPs. These synthesized ZnO nanoparticles have shown good antibacterial and antifungal activity against *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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