Green synthesis of ZnO nanoparticles using Mesua ferrea leaves extract and its antimicrobial activity

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 leaf extract as a stabilizing agent. The synthesized nanoparticles were characterized by using FT-IR (Fourier Transform Infrared Spectroscopy) and XRD (X-Ray Diffraction) and FE-SEM (Field Emission Scanning Electron Microscopy) techniques. The results indicated the successful synthesis of ZnO nanoparticles. These synthesized ZnO nanoparticles were further used for the evaluation of their antibacterial and antifungal activity against *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively.
Results: The successfully synthesized ZnO nanoparticles possess characteristic FTIR peak at 445 cm^{-1} and the average crystallite size was 20 nm. The ZnO nanoparticles has exhibited the antibacterial and antifungal activity with a maximum inhibition zone of 1.5 cm and 2 cm against the growth of *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively.
Conclusion: The *Mesua ferera* plant have some inherent medicinal properties. The ZnO nanoparticles synthesized using plant extract possess antibacterial and antifungal activity due to the synergestic 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 many different techniques like the “bottom up” and “top-down” approaches. Nowadays, bio resource based synthesis of the nanoparticles is gaining interest due to reduced toxic effects as compare to the other chemical and physical synthesis approaches. Generally, metal and metal oxide nanoparticles are synthesized using this 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. The green approach for synthesis of metal and metal oxide nanoparticles is involve 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.
In this study, we developed zinc oxide nanoparticles using *Mesua ferrea* plant extract and used as the antibacterial and antifungal agent against *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively. Since ancient times, *Mesua*

ferrea plant extract is used for medicinal purpose due to its inherent properties such as central nervous system depressant, analgesic, antimoicrobial, antivenom, antioxidant, antispasmodic, anti-inflammatory and immunostimulant. Its use for the synthesis of nanoparticles will enhance the antimicrobial activity of nanoparticles through synergestic effect with the plant extract (3).

2. MATERIAL 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 Leaf Extract - The *Mesua ferrea* leaves were collected, washed, shadow dried and chopped into small pieces. About 20 g of leaf 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 Leaf 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 coloured 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 CuKα radiation (λ= 0.154 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 ferrae* 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

Comment [H1]: Shthesis part of nano partical not apper.

3.1 FTIR Results - FTIR analysis was performed for the identification of synthesized ZnO nanoparticles. Figure 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 (4). A broad peak in the range of 2900-3600 cm^{-1} is found in the plant extract sample (5). 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. But after the formation of ZnO nanoparticles, this broad peak became smaller and shifted towards some lower angle. This broadening and shifting of peaks was observed with the increasing amount of extract from 30 to 120 mL (figure 1).

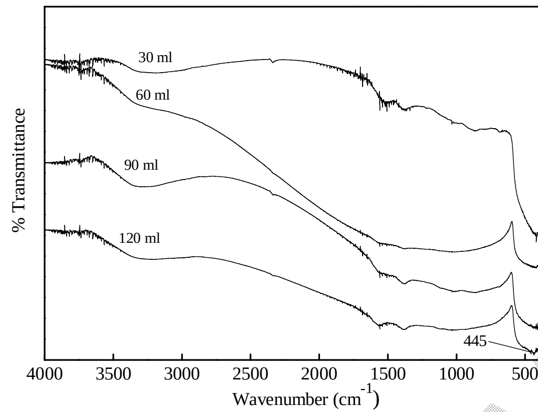


Fig. 1. FT-IR spectra of ZnO synthesized with different amount of extract (a) 30 mL;(b) 60 mL (c) 90 mL (d) 120 mL

3.2 XRD Results - XRD results of ZnO nanoparticles synthesized with different amount of *Mesua ferrea* leaf extract are presented in Figure. All the samples exhibited characteristic ZnO reflections, indicating their successful synthesis. It was observed that there exits 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 (figure 2). These diffraction peaks correspond to the characteristic hexagonal wurtzite structure of zinc oxide nanoparticles (6). Similar XRD patterns were reported by (7,8). The crystallite size for all samples is presented in Table 1.

3.3 FESEM Results - FE-SEM results indicated the presence of near spherical aggregates of ZnO NPs. This agglomeration was probably due to surface polarity and electrostatic attraction of ZnO nanoparticles (figure 2).

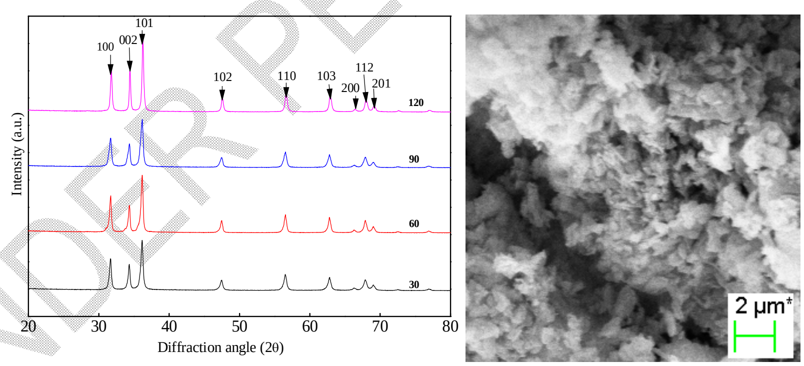


Fig. 2. XRD of ZnO synthesized with *Mesua ferrea* leaf extract and FESEM of ZnO (30 mL)

Table 1. Crystallite size of synthesized ZnO NPs

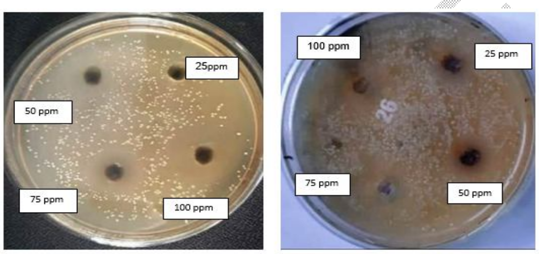
Volume of extract	Peak position (2θ)	FWHM	Crystallite size D (nm)
30 ml	36.1	0.427	19.5
60 ml	36.1	0.375	22.3

90 ml	36.1	0.472	17.7
120 ml	36.2	0.309	26.9

Antimicrobial

Activity of ZnO Nanoparticles - The results obtained during the analysis of antimicrobial activity (figure 3), indicated that the synthesized ZnO nanoparticles possess very good antibacterial and antifungal activity against *Pseudomonas aeruginosa, Aspergillus awamorii* and their zones of inhibition in cm are presented in Table 2. There is increase in the zone of inhibition with the increase in the concentration (ppm) of the ZnO nanoparticles. The antibacterial activity may be due to the larger surface area of the 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 minearls, proteins and other cell materials and hence inhibiting the cell growth (9).

Comment [H2]: antibacterial or antimicrobial not clear, please rewrite this part or re work it.



(a) (b)

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	***Pseudomonas aeruginosa***	***Aspergillus awamorii***
	Zone of inhibition in cm	
25	-	0.25
50	0.5	0.45
75	1	1
100	1.5	2

4. CONCLUSION

In this study, a green approach was used for the synthesis of ZnO nanoparticles using *Mesua ferrea* leaves extract. The successful synthesis of these ZnO nanoparticles was confirmed by peaks obtained in FT-IR spectrum and crystallite size was confirmed through XRD technique. FESEM results indicated the presence of near spherical aggregates of ZnO NPs. These synthesized ZnO nanoparticles have shown good antibacterial and antifungal effect against *Pseudomonas aeruginosa* and *Aspergillus awamorii*, respectively at 100 ppm concentration of ZnO nanoparticles.

Comment [H3]: Rewrite the conclusion part, what is application of this nano particals.

REFERENCES

1. Naseer M, Aslam U, Khalid B, Chen B. Green route to synthesize Zinc Oxide Nanoparticles using leaf extracts of Cassia fistula and Melia azadarach and their antibacterial potential. Sci Rep. 2020;10: 9055 (2020).

2. Demissie MG, Sabir FK, Edossa GD, Gonfa BA. Synthesis of Zinc Oxide Nanoparticles Using Leaf Extract of Lippia adoensis (Koseret) and Evaluation of Its Antibacterial Activity. J Chem. 2020;2020: 7459042.

3. Rajendran NK, George BP, Houreld NN, Abrahamse H. Synthesis of Zinc Oxide Nanoparticles Using Rubus fairholmianus Root Extract and Their Activity against Pathogenic Bacteria. Molecules. 2021;26(10): 3029.

4. Nethravathi PC, Shruthi GS, Suresh D, Nagabhushana H, Sharma SC. Garcinia xanthochymus mediated green synthesis of ZnO nanoparticles: photoluminescence, photocatalytic and antioxidant activity studies. Ceram Int. 2015;41(7): 8680-8687.

5. Matinise N, Fuku XG, Kaviyarasu K, Mayedwa N, Maaza MJASS. ZnO nanoparticles via Moringa oleifera green synthesis: Physical properties & mechanism of formation. Appl Surf Sci. 2017;406: 339-347.

6. Chen C. Joint committee on powder diffraction standards. Diffraction data file. 2000: 36-45.

7. Chen C, Yu B, Liu P, Liu J, Wang L. Investigation of nano-sized ZnO particles fabricated by various synthesis routes. J Ceram Process Res. 2011;12(4): 420-425.

8. Janaki AC, Sailatha E, Gunasekaran S. Synthesis, characteristics and antimicrobial activity of ZnO nanoparticles. Spectrochim Acta – A: Mol Biomol. 2015; 144: 17-22.

Comment [H4]: Added some other recent ref