Volume - 15 | Issue - 09 | September - 2026 | PRINT ISSN No. 2277 - 8179 | DOI : 10.36106/ijsr

INTERNATIONAL JOURNAL OF SCIENTIFIC RESEARCH

GREEN SYNTHESIS OF ZnO NANOPARTICLES USING MUEHLENBECKIA PLATYCLADA LEAF EXTRACT, CHARACTERIZATION, ANTIBACTERIAL AND ANTIOXIDANT ACTIVITY

Chemistry

Dr. N. Gunavathy Assistant Professor and Head, Department of Chemistry, Nirmala College For Women, Coimbatore

Manjula K II.Msc.Chemistry, Nirmala College For Women, Coimbatore

ABSTRACT

Green synthesis of nanoparticles is an environmentally friendly approach that utilizes biological resources such as plant extracts to reduce and stabilize metal ions. This method avoids toxic chemicals, is cost effective and offers a sustainable route for nanoparticle production. In this present study zinc oxide (ZnO) nanoparticles were synthesized using Muehlenbeckia Platyclada leaf extract as a natural reducing and stabilizing agent. The prepared nanoparticles were systematically characterized using standard analytical techniques to evaluate their structural and morphological features. Phytochemical screening confirmed the presence of flavonoids, sterols, anthraquinone, proteins, phenolic compounds, carbohydrates, glycosides and coumarins in Muehlenbeckia Platyclada leaf extract. UV– Visible Spectroscopy confirmed the formation of ZnO nanoparticles by showing a characteristic absorption peak around 260 nm, while FTIR analysis revealed the presence of functional groups such as hydroxyl, carbonyl, amine, aromatic, phenolic, ether and alkene. Low frequency peaks at 671 – 409 cm-1 confirmed Zn – O bond formation, indicating successful synthesis of ZnO nanoparticles. X - ray diffraction (XRD) patterns indicated an average crystallite size of 28 nm. Scanning Electron Microscope (SEM) analysis showed ellipsoidal particles forming irregular agglomerates with rough surfaces. The synthesized ZnO nanoparticles exhibited notable antibacterial activity against selected pathogens and significant antioxidant activity in - vitro assays, confirming their biofunctional potential.

KEYWORDS

Muehlenbeckia Platyclada, Green Synthesis, Zinc Oxide Nanoparticles, Characterization

INTRODUCTION

Nanotechnology has emerged as an important interdisciplinary field that enables the manipulation of materials at the nanoscale, leading to the development of nanomaterials with enhanced physical, chemical and biological properties [1]. Among various metal oxide nanoparticles, zinc oxide (ZnO) nanoparticles have attracted significant research interest due to their wide band gap, high surface area, chemical stability, biocompatibility and cost effectiveness [2]. These properties make ZnO nanoparticles suitable for diverse applications including antibacterial agents, antioxidants, drug delivery systems, sensors, cosmetics and environmental remediation [4]. Although numerous plant species have been explored for the green synthesis of ZnO nanoparticles [5], the use of *Muehlenbeckia Platyclada* remains largely unexplored. This plant is known to possess bioactive constituents that may play a significant role in nanoparticle reduction and stabilization [6]. Exploring such unused plant sources is essential for developing novel nanomaterials with enhanced functionality. Therefore, the present study focuses on the green synthesis of ZnO nanoparticles using *Muehlenbeckia Platyclada* leaf extract, followed by their characterization using standard analytical techniques. The antibacterial and antioxidant activities of the synthesized nanoparticles were also evaluated to assess their potential for biomedical and environmental applications [7].

MATERIALS AND METHODS

Collection of Plant Materials and Chemicals

Fresh and healthy leaves of *Muehlenbeckia Platyclada* plant (Figure 1) were collected from Bisalavadi Village, Chamrajnagar, Karnataka, India. The leaf were collected and washed with tap water for 2 – 3 times and were shade dried and powdered (Figure 2) and stored in air tight container [8]. Zinc sulphate ($ZnSO_4$) solution (Figure 3) used as the precursor for the synthesis of zinc oxide nanoparticles. Distilled water was used for preparing aqueous solutions all over the experiments [9].

Figure 1: ***Muehlenbeckia Platyclada*** **Plant**

Figure 2: ***MPL*** **Powder**

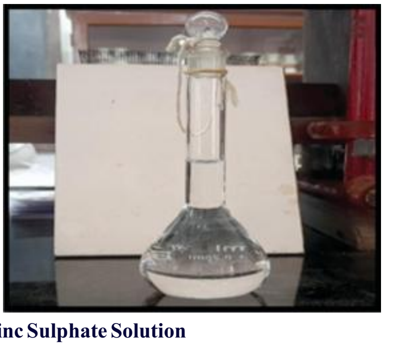


Figure 3: Zinc Sulphate Solution

Preparation of Leaf Extract

5 g of dried and powdered *Muehlenbeckia Platyclada* leaf was dissolved in 250 mL of distilled water taken in 400 mL beaker (Figure 4) and boiled for about 2 h in water bath and filtered using filter paper. The filtered extract was collected and used fresh for further studies (Figure 5) [10].

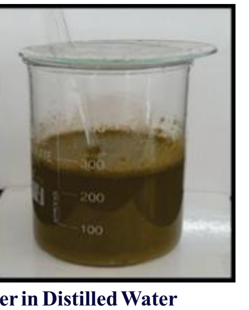


Figure 4: Leaf Powder in Distilled Water

Figure 5: ***MPL*** **Extract**

Synthesis of Zinc Oxide Nanoparticles

1.02 g of zinc sulphate was dissolved in 50 mL of distilled water and stored in a clean, dry flask. 20 mL of zinc sulphate solution was mixed with 10 mL of *Muehlenbeckia Platyclada* leaf extract. Zinc sulphate was dissolved in the *MP* leaf solution under constant stirring using magnetic stirrer. After complete dissolution of the mixture, the solution was kept under vigorous stirring at 760 RPM for 3 h (Figure 6). The appearance of solid pale yellow colour after incubation confirmed the formation of ZnO nanoparticles. The synthesized nanoparticles were used for further characterization (Figure 7) [11].

Figure 6: Preparation of ZnO NPs

Figure 7: Synthesized ZnO NPs

Phytochemical Studies

Preliminary phytochemical screening of the aqueous MP leaf extract was carried out using standard qualitative methods by Kokate (1999) and Harborne (1984) (Table 1) to identify the presence of major secondary metabolites [12].

Table 1 Standard Procedure to Identify Phytochemical Groups Present in MPL Extract

S. NO	GROUP	METHODS OF IDENTIFICATION	INFERENCE
1	Alkaloids	Extract were treated with Mayer's reagent.	Formation of white or yellow coloured precipitate.
2	Flavonoids	The plant extract was dissolved in dilute sodium hydroxide and treated with dilute acid.	Appearance of yellow colour.
3	Terpenoids	The plant extract was dissolved in chloroform, treated with acetic anhydride, and addition of concentrated sulphuric acid.	Formation of blue, green or reddish brown colour.
4	Anthra-quinones	The plant extract was boiled with dilute sulphuric acid, cooled, extracted with chloroform and the chloroform layer was shaken with dilute ammonia.	Appearance of pink, red or violet colour.
5	Phenolic compounds	To 2 mL of the extract, 1 mL of concentrated nitric acid was added, gently heated, cooled and treated with excess sodium hydroxide or ammonia solution.	Formation of a yellow to orange colour
6	Quinones	The plant extract was treated with a few drops of concentrated hydrochloric acid.	Formation of red or violet colour
7	Carbo-hydrates	2 mL of the extract were added to Fehling's solution and boiled for 5 minutes.	Formation of a brick red colour precipitate
8	Tannins	A few drops of ferric chloride solution were added to 2 mL of the plant extract.	Appearance of a greenish black or blue black colour.
9	Saponins	2 mL of the extract were diluted to 10 mL with distilled water and shaken for 15 minutes	Formation of froth
10	Glycosides	The extract in glacial acetic acid was treated with a few drops of ferric chloride, followed by the careful addition of concentrated sulphuric acid.	Appearance of reddish brown colour to bluish green
11	Coumarins	The plant extract was dissolved in chloroform, treated with a few drops of dilute sodium hydroxide, and exposed to UV light.	Appearance of yellow fluorescence

UV – Visible Spectral Analysis

UV – Visible spectroscopic analysis of the synthesized zinc oxide nanoparticles was carried out using a UV–Visible spectrophotometer (Lovibond XD 7000) at Avinashilingam Institute of Home Science for Women, Coimbatore. The formation of ZnO nanoparticles was confirmed by the appearance of a characteristic absorption peak in the UV region [13].

FTIR Spectral Analysis

FTIR analysis was performed using an FTIR spectrophotometer (Shimadzu IR Affinity-1) at Avinashilingam Institute of Home Science for Women, Coimbatore to identify the functional groups present in the synthesized ZnO nanoparticles [14].

X – ray Diffraction Studies

The crystalline nature and phase purity of the synthesized zinc oxide nanoparticles were analyzed using X – ray diffraction (XRD). The XRD patterns were recorded using an X – ray diffractometer (X'Pert3 Powder Model) at Avinashilingam Institute of Home Science for

Women, Coimbatore. The diffraction data were collected over a 2θ range of 20° – 80° at room temperature. The obtained diffraction peaks were indexed and compared with standard data to confirm the crystalline structure of ZnO nanoparticles. The average crystallite size of the ZnO nanoparticles was calculated using the Debye – Scherrer equation based on the most intense diffraction peak [15].

SEM Analysis
SEM analysis of the synthesized ZnO nanoparticles was performed using a scanning electron microscope (TESCAN – MIRA3 XMU) at Avinashilingam Institute of Home Science for Women, Coimbatore to study their surface morphology [16].

Antibacterial Assay
The antibacterial assessment of ZnO NPs was carried out by using two different test pathogens; gram – negative Escherichia coli and Staphylococcus epidermidis, gram – positive Escherichia coli and Staphylococcus epidermidis. The microbial cultures were maintained by the Microbiology Research Centre, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore, India. Antibacterial activity of the synthesized ZnO NPs was determined using Agar – well diffusion method diffusion method. Petriplates containing 20 mL Mueller Hinton medium were seeded with 0.5 McFarland standard cultures of bacterial strains. Wells were cut using sterile well puncture and 50 µL of samples were added into the wells. The plates were then incubated at 37°C for 24 hours. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well [17].

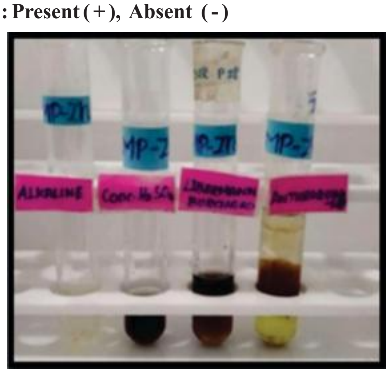
Antioxidant Assay
The antioxidant activity of ZnO nanoparticles (*MP* – Zn) was determined by the DPPH free radical scavenging assay using ascorbic acid (1 mg / mL) as the standard. Varying concentrations of the sample (500 – 1100 µL) were reacted with 0.1 mM DPPH in methanol and incubated in the dark for 30 min [18]. Absorbance was measured at 517 nm (Lovibond XD 7000) at Avinashilingam Institute of Home Science for Women, Coimbatore and % inhibition was calculated using the standard formula.

RESULTS AND DISCUSSION
Phytochemical Analysis
Phytochemical screening of *MPL* extract (Table 2) revealed the presence of flavonoids, sterols, anthraquinone, proteins, phenolic compounds, carbohydrates, glycosides and coumarins (Figure 8).

Table 2 Phytoconstituents Present in MPL Extract

S.No	Phytoconstituents	MPL
1	Alkaloids	-
2	Flavonoids	+
3	Terpenoids	-
4	Sterols	+
5	Anthraquinone	+
6	Proteins	+
7	Phenolic compounds	+
8	Quinones	-
9	Carbohydrates	+
10	Tannins	-
11	Saponins	-
12	Cardiac glycosides	-
13	Glycosides	+
14	Coumarins	+

Key words : Present (+), Absent (-)



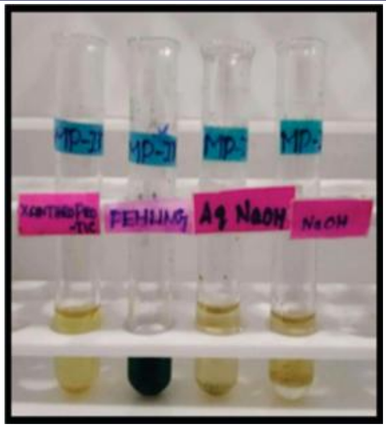


Figure 8: Phytochemical Analysis of *MP* Leaf Extract

UV – Visible Spectroscopy
In Figure 9, zinc oxide nanoparticles prepared from *MPL* extract by green synthesis showed an absorption peak at 260 nm indicating that the synthesized zinc oxide nanoparticles might be a nano sized particle.

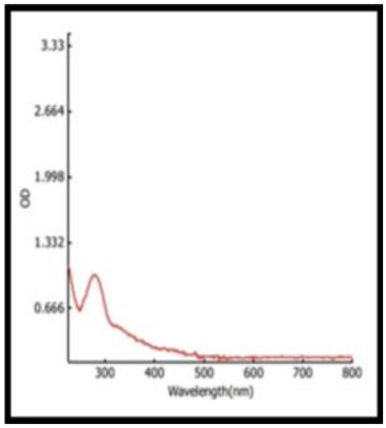


Figure 9: UV – Spectrum of Synthesized Zinc Oxide Nanoparticles

FTIR Analysis
FTIR Analysis of *Muehlenbeckia Platyclada* leaf extract showed the possible presence of the following functional group (Figure 10). The peak at 3271 cm-1 corresponds to O – H stretching, the band at 1643.35 cm-1 corresponds to C = O, the band at 1188.15 cm-1 corresponds to C – O – C, the band at 1087.85 cm-1 corresponds to C – O stretching, the bond at 1041.56 cm-1 corresponds to C – O, the band at 671.73 cm-1 to 408.91 cm-1 corresponds to Zn – O Bending.

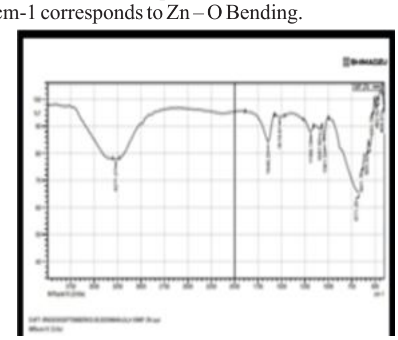


Figure 10 FTIR Spectrum of *MPL*

XRD Analysis
The XRD pattern exhibited three prominent diffraction peaks at 292°, 298° and 309°. The average crystallite size, calculated using the Debye – Scherrer equation from the reflection, was found to be approximately 28 nm.

SEM Analysis
The image obtained by SEM (Figure 11) for the synthesized zinc oxide nanoparticle showed irregular, agglomerated with roughly ellipsoidal shaped nanoparticles.

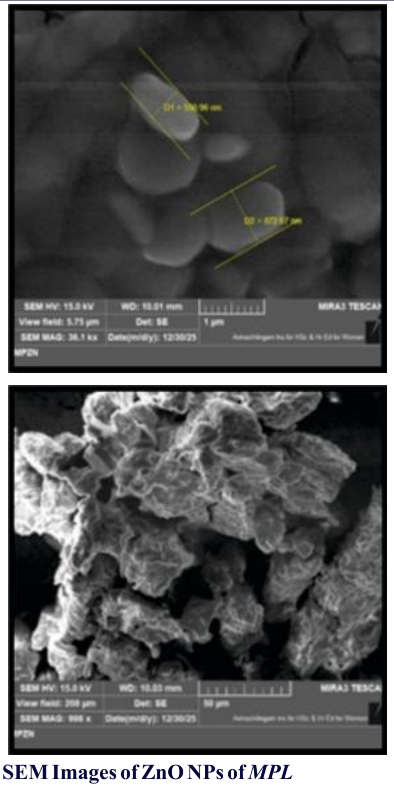


Figure 11: SEM Images of ZnO NPs of *MPL*

Antibacterial Assay

The synthesized nanoparticles showed significant antibacterial activity against the tested bacterial strains, (Figure 12) as indicated by clear zones of inhibition, which may be attributed to their small particle size and enhanced interaction with bacterial cell membranes. The zone of inhibition values for the bacteria are given in the (Table 3) as compared with the standard antibiotic, Chloramphenicol. The activity was comparable, indicating good antibacterial potential.

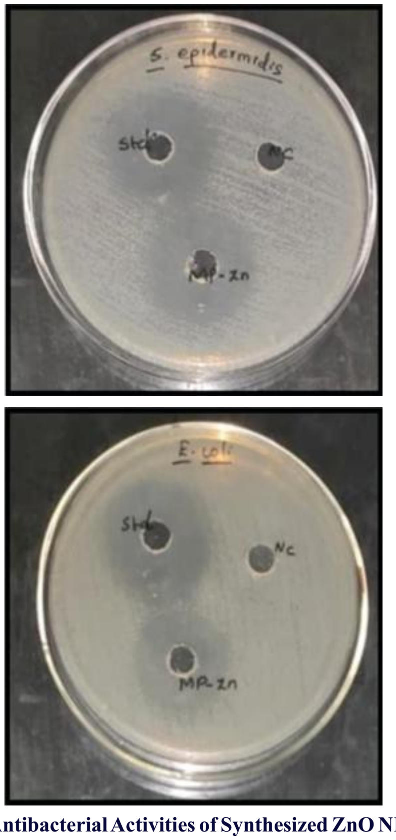


Figure 12 Antibacterial Activities of Synthesized ZnO NPs Against Bacteria Staphylococcus Epidermidis and Escherichia Coli

Table 3 Zone of Inhibition for the Synthesized ZnO using *MPL* Extract for Bacteria

Samples	Zone of Inhibition in diameter (mm)	
	Staphylococcus epidermidis	Escherichia coli
Standard antibiotic (Positive Control)	32.5 ± 0.51	30.4 ± 0.69
MPZN – 50 µL	32.6 ± 0.52	29.5 ± 0.51

Antioxidant Activity

The synthesized nanoparticles demonstrated significant antioxidant activity (Table 4), as indicated by their effective free radical scavenging ability, which can be attributed to the presence of bioactive phytochemicals acting as reducing and stabilizing agents. The antioxidant activity of ZnO nanoparticles increased with concentration, showing maximum 76.38 % inhibition at 1100 µg / mL. This indicates that the synthesized zinc oxide nanoparticles have good free radical scavenging potential.

Table 4 Antioxidant Activity of Synthesized NPs Evaluated by DPPH Radical Scavenging Assay

Concentration (µL / mL)	% Inhibition
500	55.55
600	58.33
700	59.72
800	63.88
900	65.27
1000	70.83
1100	76.38

CONCLUSION

The present investigation demonstrates an eco-friendly synthesis of nanoparticles using *Muehlenbeckia Platyclada* leaf extract. Phytochemical screening confirmed the presence of flavonoids, sterols, anthraquinone, proteins, phenolic compounds, carbohydrates, glycosides and coumarins in *Muehlenbeckia Platyclada* leaf extract. UV–Visible analysis showed a λ_{ax} at 260 nm, confirming ZnO nanoparticle formation. Low frequency peaks at 671 – 409 cm-1 confirm Zn – O bond formation, indicating successful synthesis of ZnO nanoparticles. XRD analysis revealed ZnO nanoparticles with size of 28.93 nm. SEM images showed ellipsoidal shaped ZnO nanoparticles. The synthesized zinc oxide nanoparticles had good antibacterial activity against the selected bacteria. The synthesized zinc oxide nanoparticles had good antioxidant activity. The eco - friendly synthesis approach, combined with promising biological performance, highlights the potential of these nanoparticles for biomedical and environmental applications.

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