



## BIOMEDICAL APPLICATIONS OF ZINC OXIDE NANOPARTICLES FROM MORINGA OLEIFERA (DRUMMSTICK) LEAVES EXTRACT

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### ABSTRACT

The current study investigated the synthesis and characterization of Zinc oxide Nanoparticles (ZnONPs) from *Moringa oleifera* (*M. oleifera*) leaves extract to investigate the, antimicrobial, potential of *Moringa oleifera* (Drumstick). The antimicrobial activity of green synthesis ZnO nanoparticle (ZnO NPs) was measured against gram-positive *S.aureus* and gram-negative *E.Coli*. The synthesized ZnO nanoparticles were characterized by UV-visible. The outcome of UV-visible reveals broadband absorption for leaf extract of the experimental plants yielding the SPR peaks at *Moringa oleifera* ZnO nanoparticles 340 nm. The antibacterial activities against mentioned pathogens were done by using disc diffusion method; the substantial inhibitory action for leaf extract was observed both in gram-positive and gram-negative bacteria isolates.

**KEYWORDS:** ZnO nanoparticles, *Moringa oleifera* leave, UV-Vis, anti-microbial, disc diffusion method.

### INTRODUCTION

The term “nano” is derived from the Greek word “nanos” for “dwarf”. Nano scale materials are defined as materials having at least one dimension in the 10–100 nm range. Scientists have constantly explored different synthetic methods to synthesize nanoparticles and nanoparticles can be synthesized by physical, chemical and biological routes. Though numerous conventional physical and chemical methods are available for the synthesis of nanoparticles, have some adverse aspects like critical conditions of temperature and pressure, use of expensive and toxic chemicals, long reflux time of reaction, toxic potentially hazardous byproducts etc. Biosynthesis of nanoparticles researchers to develop synthetic strategies using biological entities like enzymes [i], microorganisms [ii] and plant extracts [iii-v] play a major role in the formation of nanoparticles. *Moringa oleifera* (*M. oleifera*) (Family: Moringaceae, English name: drumstick tree) has been reported to be essentially used as an ingredient of the Indian diet since ages. It is cultivated almost all over India and its leaves and fruits are traditionally used as vegetables. Almost all parts of the plant have been

utilized in the traditional system of medicine. The plant leaves have also been reported for its antitumor, cardio protective, hypotensive, wound and eye healing properties [vi].

ZnO nanoparticles, owing to their small size and larger specific surface area exhibit enhanced antimicrobial activities. The ZnO nanoparticles possess antibacterial and antifungal activities even at lower concentrations hence suitable for thin coating applications [vii]. ZnO nanoparticles synthesized using aqueous leaf extract of *Camellia sinensis* was tested for its potential antimicrobial activity against some selected microbes. To analyze the antimicrobial activity of the sample, the samples were subjected to Agar well Diffusion method [viii]. Zinc oxide nanoparticles are known to be one of the multifunctional inorganic nanoparticles with effective antibacterial activity [ix].

## MATERIALS AND METHODS

### BIOSYNTHESIS OF ZnO NPs

29.7 gm of Zinc nitrate [ $\text{Mg}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$ ] obtained from Himedia Pvt.Ltd. chemicals was dissolved in 1000 mL of double distilled water so as to obtain 0.1M of zinc nitrate precursor solution. Synthesis of zinc oxide nanoparticles, 0.1M of zinc nitrate [ $\text{Zn}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$ ] were taken in 50 mL and then 10 mL of *Moringa oleifera* leaves extract was added slowly in a drop wise manner into the solution under magnetic stirring at 70°C for about 2 hours to form zinc hydroxide precipitate. The white precipitate formed after adequate time of stirring was collected by centrifugation at 10000 rpm for 10 minutes. Then the centrifuged particles were washed with water and subjected to another centrifugation at 5000 rpm for 10 minutes the separated white precipitate was dried in an oven at 70°C for 2 hours followed by grinding using mortar. This powdered sample was calcined in a muffle furnace at 350-450°C to get zinc oxide nanoparticles.

### CHARACTERIZATION OF ZnO NPs

#### UV-VISIBLE SPECTRA ANALYSIS

The confirmation of the synthesized ZnO NPs were done by recording UV-Vis spectrum, this was the preliminary step to confirm the result. In the present works, the biosynthesis of ZnO NPs are formed after the addition of *Moringa oleifera* leaves extract. The optical property of nanoparticles was determined by a UV-Visible spectrophotometer in the 200 to 600 nm range. The absorption spectra of the synthesized nanoparticles were measured using Equip-Tronics-826 dual-beam spectrophotometer. The UV –Visible spectrum was recorded for aqueous leaves extracts of *Moringa oleifera* with ZnO NPs. The Surface Plasmon Resonance (SPR) of these ZnO NPs peaks produced during reaction.

### ANTIMICROBIAL ASSAYS

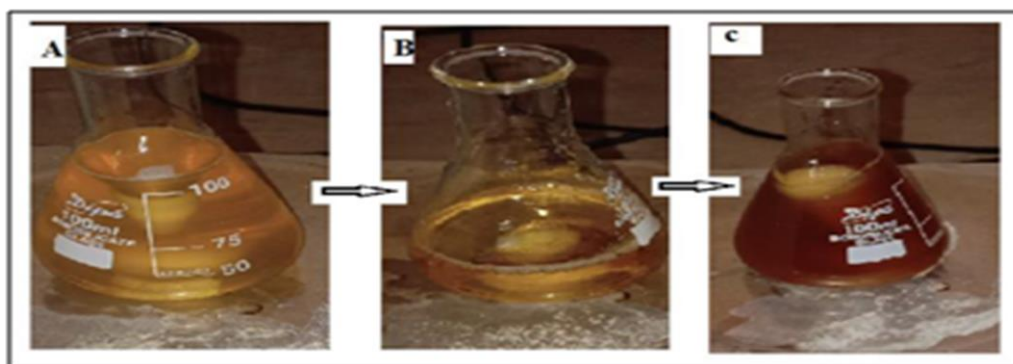
Antibacterial study of the Zinc oxide nanoparticles synthesized using plant extracts were tested by the modified Kirby – Bauer disc diffusion method against various pathogenic bacteria of gram positive (*S. aureus*) and gram negative strains (*E. coli*). Two major pathogens (*S. aureus*, *E. coli*) were taken for analysis. Two plates for each organism in such away 2 plates containing solidified agar media were prepared and swabbed with respective microbial inoculum. Four disks in each plate were fixed at equal distance. The first disk of each plate underlined were loaded with a concentration of 25 µg/ml (A), ZnO nanoparticles. The second disk of plate red underlined were impregnated with 50 µg/ml (B) concentration of ZnO nanoparticles. The third disk of each plate was impregnated with 50 µg/ml (C) concentration of plant extract and the fourth disk (D) was maintained as blank without nanoparticle and plant extract. All these plates were kept at 37 °C for incubation overnight. After 24 hrs of incubation the zone of inhibition were measured and compared. The antibacterial potential of test compounds was evaluated on the basis of mean diameter of zone of inhibition around the disc in millimeters. Zone of inhibition is the area in which the

bacterial growth is stopped due to bacteriostatic effect of the compound. Finally the diameters (mm) of ZOI were measured [x].

## RESULT AND DISSCUSSION

### SYNTHESIS OF ZnO NPs OF *M.OLEIFERA*

Mostly identification of Zinc oxide nanoparticles synthesis was done by visual observation, an appearance of light yellow to dark brown color in the reaction mixture denote the formation of nanoparticles. The leaf extract shows light yellow color while Zinc nitrate solution shows yellow color before the addition of Zinc nitrate and leaf extract these changed after 24 hours of incubation into dark brown color indicating the completion of reaction as shown in fig 1. The all different parts of the plants have already been exploited for the synthesis of various metal nanoparticles with precious bioactive mechanisms. This report proposed on bio-synthesis of ZnO nanoparticles leaf extracts of *M. oleifera*. The AgNPs from *M. oleifera* leaves have been investigated to possess significant antifungal activity against *Candida albicans* [xi]. The silver nanoparticles reduced by the gum of *M. oleifera* were reported to showed antibacterial activity against *Staphylococcus aureus*, *E. coli* and *Pseudomonas aeruginosa* [xii].



**Fig.1. Zinc nitrate solution B) *M.oleifera* leaves extract C) Zinc nitrate and *M.oleifera* leaves extract solution.**

### UV-VIS.SPECTROSCOPY

The ZnO nanoparticles were synthesized by using leaf extract of *M. oleifera* as detailed above. The reduction of zinc metal ions to zinc oxide nanoparticles in the reaction medium was initially analyzed using UV-Vis Spectrophotometer between 200 to 600 nm. The UV-Vis spectrum of ZnO Nps is shown in Fig. 2. Confirmation of the synthesized ZnO product in nano-scale was exhibited by the highly blue shifted absorption maximum occurring around 344 nm.

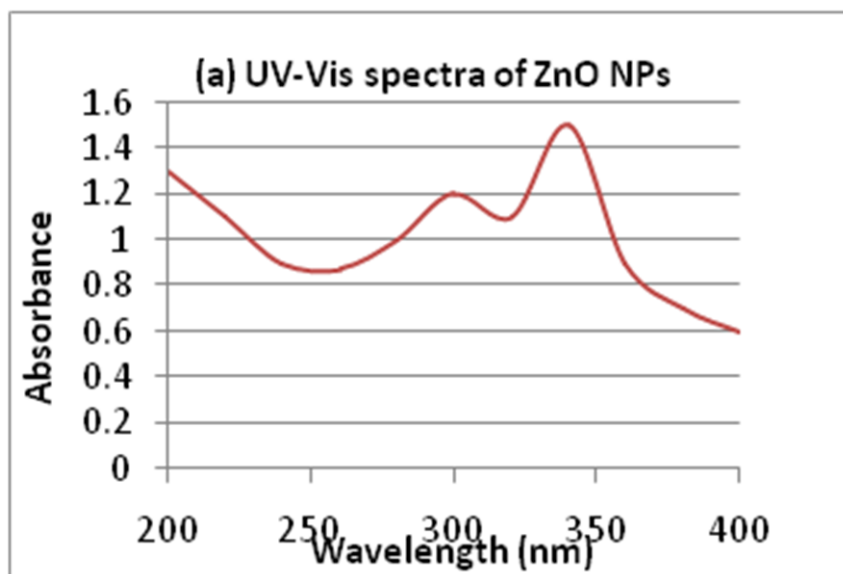


Fig. 2: UV-Vis. spectra of ZnONPs obtained using leaves of *M. oleifera* extract.

#### ANTIMICROBIAL ACTIVITY OF ZnO NPs

The antimicrobial activities of ZnO NPs against gram-positive (*Staphylococcus aureus*) strain reference NCIM 2079 and gram negative bacteria (*Escherichia coli*) strain reference NCIM 2109 were performed using the disc diffusion method and their activity was compared to a well-known commercial antibiotic Gentamicin are shown in fig.3. The results are evaluated and listed in Table 1. From the antibacterial inhibition results, ZnO NPs observed bacterial activity against all bacterial strains. The *S.aureus* gram-positive found more zone of inhibition ( $9.74 \pm 0.2$ ) as compared to *E.coli* ( $8.70 \pm 0.2$ ) gram-negative. Especially, *S.aureus* gram-positive showed higher activity than standard drug Gentamicin ( $9.50 \pm 0.2$ ). Therefore, we can conclude that the biosynthesized ZnO nanoparticles were shown considerable antibacterial activity.

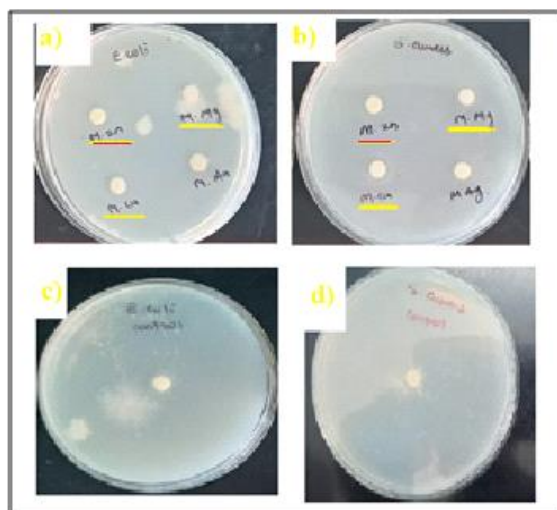


Fig.3. Plate having red underline disc showing antibacterial activity of ZnO NPs against (a) *E.coli* (b) *S.aureus* (c) positive control of *E.coli* and (d) positive control *S.aureus*

Table .1.Zone of Inhibition (ZOI) of ZnO NPs against *S.aureus* *E.coli* and positive (P) control

Pathogenic Bacteria	Zone of Inhibition (mm)	
	ZnO NPs	Positive control

	(Gentamicin)	
<b>S.aureus</b>	<b>9.74± 0.2</b>	<b>9.50± 0.2</b>
<b>E.coli</b>	<b>8.70± 0.2</b>	<b>9.10± 0.2</b>

\*Zone of inhibition values are expressed as the mean of triplicate measurements  $\pm$  standard deviation Figure.3 Antimicrobial activity of ZnO NPs against bacterial pathogens.

## CONCLUSION

The green synthesis of ZnO NPs using aqueous extracts of *Moringa oleifera* (Drumstick) leaves is an eco-friendly and cost-effective method for the bio synthesis preparation of ZnO NPs. The outcome of this study show that the tested pathogenic strains (*S. aureus*, *E. coli*) are susceptible to ZnO NPs, which confirm the potential efficacy of these NPs, aligned with certain bacterial strains. This makes it possible to enlarge the use of these NPs in the biomedical field.

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