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## Green synthesis of Zinc oxide nanoparticles from fresh curcuma longa tuber and their anti-bactericidal and photocatalytic activity

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Biological nanoparticle synthesis approach focused on microorganisms, enzymes, and plant extracts have been proposed as eco-friendly alternative to chemical and physical methods. In the current study green approach was used to create zinc oxide nanoparticles (ZnO NPs) from aqueous solution of Zinc sulphate heptahydrate by using fresh *curcuma longa* tuber's methanolic and aqueous extract. The indication of the synthesis of zinc nanoparticles was initially confirmed with the color change from dark orange to brownish yellow. The characterizations of ZnO NPs were determined using UV-Visible spectroscopy, Scanning Electron Microscopy (SEM) and Energy dispersive X-ray analysis (EDX). FTIR analysis confirmed the capping of nanoparticles by phytochemicals present in fresh *curcuma longa* tuber's extract. The presence of zinc and oxygen were confirmed from the EDX of the nanoparticles, and SEM analyses showed uniform and homogeneous morphology with an average size of 40-72 nm for the synthesized ZnO nanoparticles. The bio-synthesized nanoparticles showed significant photo catalytic and antibacterial activity against *staphylococcus aureus* and *Acetobacter*. It was concluded that ZnO NPs show effective activity against *staphylococcus aureus* and *Acetobacter*.

**Keywords:** ZnO nanoparticle, *Curcuma longa*, photo catalysis, antibacterial activity.

### INTRODUCTION

Nanotechnology is one of the most active fields of study in contemporary material science. Nanotechnology is an increasingly growing field with applications in science and technology for the purpose of developing new materials at the nano-scale level. Nano-materials have been dubbed a modern-day medical miracle (Sosa et al.). The word nano comes from the Greek word for "dwarf." A nanometer (nm) is one billionth of a meter, or three atoms located side by side. A DNA molecule has a diameter of 2.5 nm, a protein has a width of about 50 nm, and a flu virus has a width of about 100 nm and a human hair has a thickness of around 10,000 nm. A nanoparticle is a spherical particle of less than 100 nanometers in at least one dimension (Thakkar et al.).

Nanostructures have size and shape-dependent properties useful in a variety of applications, including photodynamic therapy and catalysts, optics, antimicrobial action, computer transistors, electrometers, chemical sensors, and wireless electronic logic and memory schemes. These particles have many uses in a variety of areas, including medical imaging, nano composites, sensors, drug delivery, and tumor hyperthermia (Tan et al.2006), (Lee et al.2008), (Pissuwan et al. 2006), (Panigrahi et al. 2004).

Nano metal oxides can be produced using a variety of methods, including electrochemical processing, decomposition, microwave-assisted techniques, and chemical reduction procedures (Kharissova et al.2013), (Logeswari et al.2013). However, the materials used in such processes

are usually flammable and poisonous (Nadagouda et al.2014). Because of its eco - friendliness, green synthesis of metallic nanoparticles using plant materials such as tissues, plant extracts, and living plants has gained a ton of interest. The method is free of hazardous and toxic chemicals, and it is therefore very cost effective, making it a viable and useful option for large-scale synthesis (Raut2014).

In terms of interaction and environmental impact, the use of plant extracts in the production of nanoparticles has a significant advantage over other approaches. Over the years, the use of engineering nano-materials has expanded in a variety of areas, including medicine, cosmetics, toys, and other consumer goods (Harris et al.2008). They have a wide range of properties, including self-assembly, precision, entrapment, stability, and biocompatibility. Because of their antimicrobial properties, zinc oxide nanoparticles are a popular nanomaterial in the medical and agricultural industries (Linga Rao and Savithramma 2012).

ZnO NPs have been extensively explored because of their photolytic properties and are seeing widespread usage in personal care products due to their UV-blocking performance (Farahani et al. 2012). The current research describes an environmentally safe process in order to synthesize zinc oxide nanoparticles that use aqueous and methanolic extract of *C. longa* tubers for the bio-reduction of ZnO NPs. XRD, FTIR, and SEM analysis were used to classify the shaped ZnO NPs. The nanoparticles that were created were used for photocatalytic degradation of methyl orange and methyl blue dyes, as well as antibacterial action.

## MATERIALS AND METHODS

### Chemical use

The synthesis of ZnO nanoparticles *Curcuma longa* tubers (from the local market), Zinc sulphate heptahydrate, methanol, and distilled water were used.

### *Curcuma longa* tubers (aqueous) extract preparation

*Curcuma longa* tubers were obtained from a local market, Charsadda KPK. The *C. longa* 20-gram fresh tubers were washed several times with tap water and 3 times with ethanol and then 3 times with distilled water to remove mud particles and possible impurities and chopped with sterile knife then crushed with sterile Mortar and Pestle.

For the preparation of extract 20 grams of crushed fresh *C. longa* tuber was added to 100ml of distilled water in 200 mL beaker and boiled for 30 minutes and allowed to cool down at room temperature for 15 minutes. Then filtered with Whitman No.1 filter paper. Extract was stored at room temperature for further experiments.

### Preparation of Methanolic Extract

20g of fresh *curcuma longa* tuber were washed and then chopped with sterile knife and then crushed with sterile mortar and pestle then 100ml methanol was added to it and boiled for 30 minutes then allowed to cool down at room temperature for 15 minutes. The extract was then filtered with Whitman No.1 filter paper.

### Green synthesis of ZnO nanoparticles

0.01m aqueous solution of zinc sulphate heptahydrate was prepared and used in the synthesis of zinc oxide nanoparticles. 100ml of fresh *curcuma longa* extract was added into 100ml of aqueous solution of 0.01m zinc sulphate heptahydrate under vigorous stirring on hot plate at 70°C for aqueous extract and at 30°C for methanolic extract for 2 hours until the color changed from dark orange to brownish yellow.



ZnSO<sub>4</sub> (7H<sub>2</sub>O)

ZnO NPS

### Study of Antibacterial Activity

Antibacterial activity of *curcuma longa* extract and bio-synthesized zinc oxide nanoparticles against *Acetobacter* and *Staphylococcus aureus* was studied. For antibacterial activity, the agar well diffusion method was used. The test bacterial strains were streaked on nutrient agar in sterile Petri dishes and incubated at 37°C for 24 hours. Using a micropipette, at a concentration of 50µl, 75µl and 100µl in respective agar well plate according to bacterial culture. After 24 hours, zone of inhibition was measured. The antibacterial effect against, *Acetobacter* and *Staphylococcus aureus* was measured based on the inhibition zone.

### Study of Photocatalytic Degradation

Today, environmental safety is becoming highly important for humans, and certain poisonous and stable dye molecules such as Methyl Orange and Methyl Blue, are harmful to the environment. (Wang et al.2008). Metal nanoparticles have large surface area with high reactive activity, are used to reduce dangerous dyes. Methyl orange dye was degraded by the use of *curcuma longa* aqueous extract induced ZnO NPs. Similarly, methyl blue dye was degraded by the use of *curcuma longa* methanolic extract mediated ZnO NPs.

## RESULTS AND DISCUSSION

### UV-visible Spectroscopic Analysis

UV-visible spectrophotometer was used to confirm the method for the biologically synthesized ZnO Nps. The reaction was seen in 50% plant extract, slowly the color of solution was changed then the peak was observed (aqueous medium) at 470 wavelength (nm) and 480 nm (methanolic medium) at maximum absorbance as shown in figure 1,2.

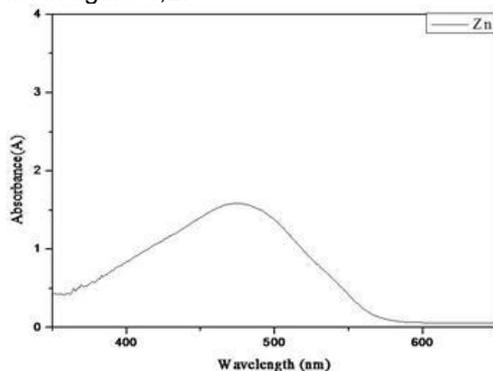


Figure1: UV-Visible spectra of ZnO NPs (aqueous medium)

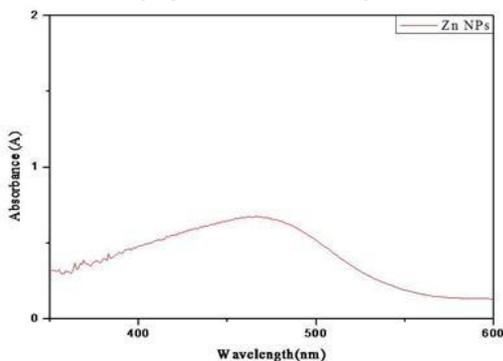


Figure: 2 UV-Visible spectra of ZnO NPs (methanolic medium)

In previous study the UV band was at a

wavelength of 370-400 nm, confirming the synthesis of ZnO NPs (Sharmila et al.2018). Another literature on green synthesized ZnO NPs reported absorbance peak at 380 nm (Bala et al.2015). Padalia and Chanda reported 370nm absorption peak for ZnO NPs (Padalia et al.2017).

### FT-IR Spectroscopic Analysis

In current study the FT-IR spectra of sample of aqueous extracts and synthesized ZnO NPs the Spectra showed broad peak in the range of 3500-3000  $\text{cm}^{-1}$ , which was due to the -OH group. The spectrum shows peak at about 1100  $\text{cm}^{-1}$ , which might be due to the starching vibration of C=O. The band at 1633  $\text{cm}^{-1}$  is due to carbonyl stretching in proteins (Gebru et al.2013). The ZnO peaks are also observed in the range 800-450  $\text{cm}^{-1}$ . The FT-IR spectra sample of methanol extracts and the synthesized Zn NPs showed broad peak in the range of 3500-3000  $\text{cm}^{-1}$ , which might be due to the -OH group. The Spectrum showed peak at about 1100  $\text{cm}^{-1}$ , which might be due to the starching vibration of C=O. The ZnO peaks are also observed at 900  $\text{cm}^{-1}$ . It indicated that the ZnO nanoparticles are successfully synthesized.

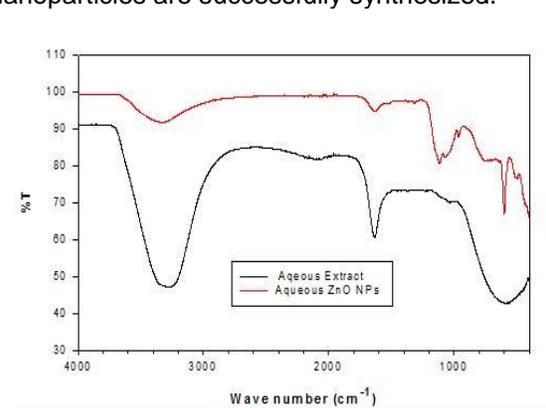


Figure 3: FT-IR spectra

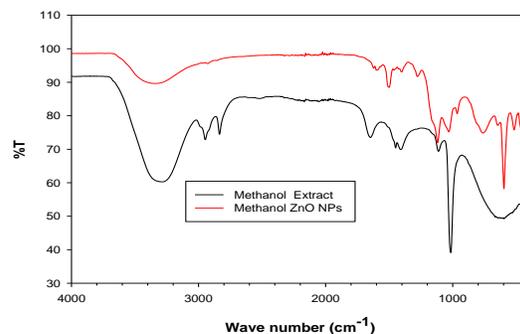
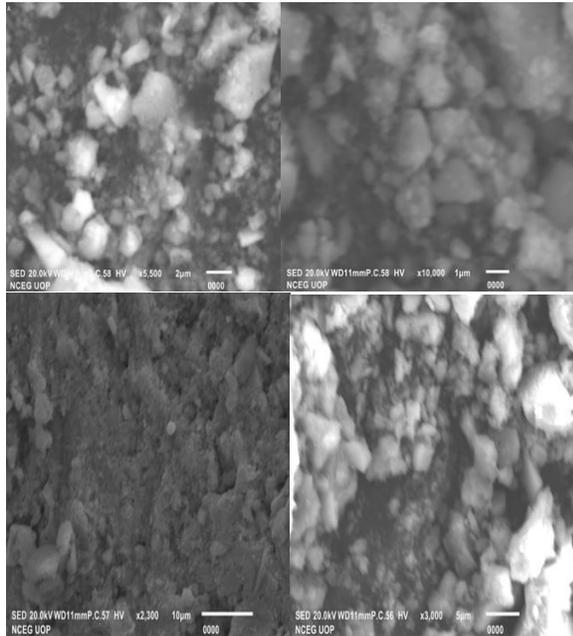


Figure 4: FT-IR spectra

**SEM Analysis**

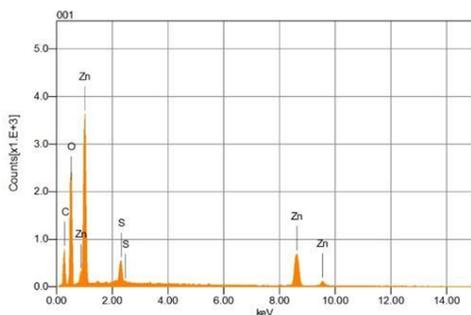
The scanning electron micrographs of ZnO NPs show that synthesized nanoparticles in methanolic medium are irregular however, uniform, and homogeneous in morphology and synthesized aqueous medium ZnO NPs are irregular and dispersed. It also be noted that the particles exist non-agglomerated and well dispersed and the size of particle were 72nm and 40nm.



**Figure 6: SEM images of prepared Zinc oxide Nanoparticles**

**EDX Analysis**

The presence of zinc oxide nanoparticles synthesized in both methanolic and aqueous medium were confirmed by using the Energy dispersive X-ray spectroscopy. The EDX composition showed a strong peak at 3 keV confirmed the efficient synthesis and purity of nano-crystalline ZnO NPs.



**Figure 7: EDX figure of prepared Zinc oxide Nanoparticles**

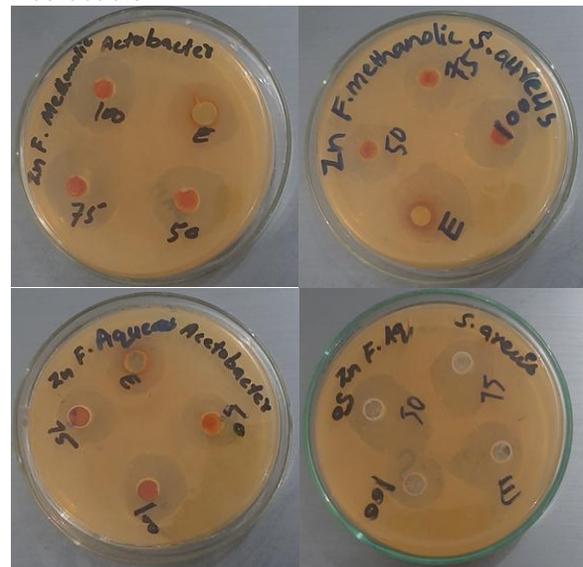
The presence of oxygen clearly confirmed the presence of metal Oxide nanoparticles.

Formula	mass%	Atom%	Sigma	Net	K ratio	Line
C	6.20	38.88	0.04	18901	0.0023840	K
O	39.30	48.95	0.11	60670	0.0349700	K
S	2.58	1.60	0.03	20452	0.0041997	K
Zn	51.92	10.58	0.20	55079	0.0642763	K
Total	100.00	100.00				

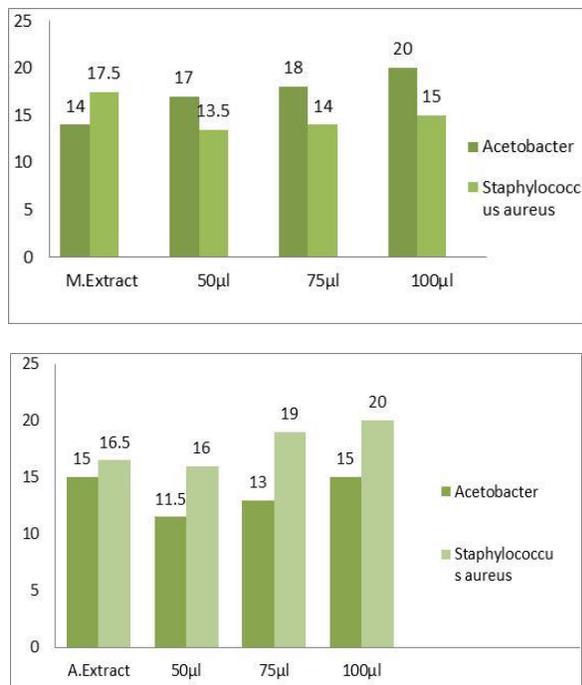
The presence of S and C due to their presence in the extract used for the preparation of ZnO NPs.

**Anti-bactericidal activity**

ZnO Nps synthesized using *curcuma longa* methanolic and aqueous extract are subjected to antibacterial testing against *Staphylococcus aureus* and *Acetobacter*. Synthesized ZnO nanoparticles showed remarkable antibacterial activity. ZnO NPs (methanolic extract) showed significant activity against *Acetobacter* while showed moderate activity against *Staphylococcus aureus*. However, ZnO NPs from aqueous extract were most effective against *Staphylococcus aureus* while showed moderate activity against *Acetobacter* and the aqueous and methanolic extract were most effective antibacterial agent against *Staphylococcus aureus* as compared to *Acetobacter*.



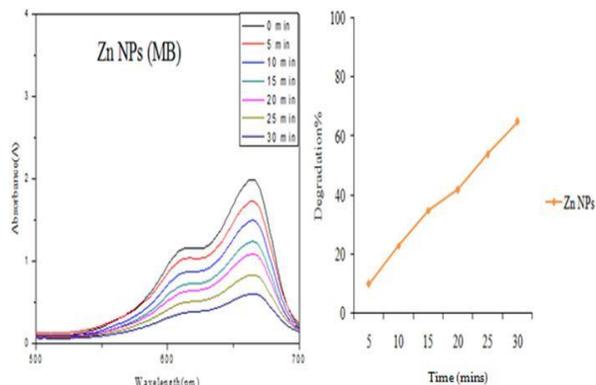
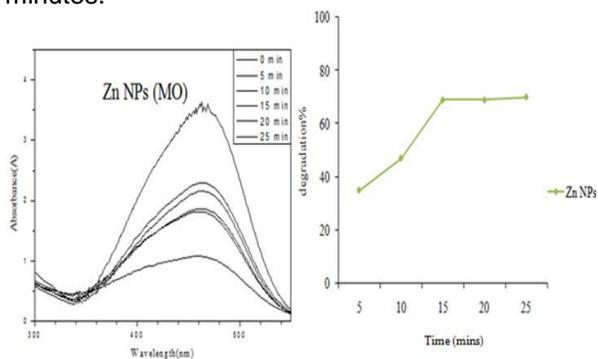
**Figure: 8 images showing an anti-bacterial activity against selected bacterial strains**



**Figure 9: Inhibition Zones of ZnO NPs and C.longa extracts against selected bacterial strains**

**Photocatalytic Degradation Of MB And MO Dyes By Using ZnO NPs**

The methyl orange and methyl blue dye was degraded by the use of *curcuma longa* aqueous and methanolic extract induced Zn-NPs. 0.02g of the prepared ZnO NPs were added to 10ml (50ppm) solution of MO dye and run under the UV light scan in UV-Visible spectrophotometer. Different peaks were observed at various time intervals from 5 to 25 min the MO original peaks were observed one at 480 nm and the % degradation were observed 30% at 5 minutes and 70% at 15 minutes. Similarly, MB original peaks were observed at 680nm and the % degradation were observed 20% at 10 minutes and 70% at 30 minutes.



**Figure 10: Degradation spectra of MO and MB Dyes by ZnO NPs**

**CONCLUSION**

Our current research concludes that bio-assisted method of nanoparticle synthesis is a good alternative compared with hazardous physical and chemical methods. Our results indicated the effectiveness of nanoparticles against different bacterial strains. The synthesized zinc oxide nanoparticles indicated excellent antibacterial activity toward *Acetobacter* and *staphylococcus aureus*. ZnO nanoparticles illustrated the catalytic degradation of methyl blue and methyl orange dyes and showed very positive degradation result. Our research work showed that NPs play an important role in biological assays and also in photocatalytic degradation.

**CONFLICT OF INTEREST**

The authors declared that present study was performed in absence of any conflict of interest.

**AUTHOR CONTRIBUTIONS**

A performed the experiments and wrote the manuscript, FA designed the experiments, critically reviewed and edited the manuscript, TS analyzed the data, performed few experiments and reviewed the manuscript and SS reviewed the manuscript

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with these terms.

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