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## Green synthesis and characterization of *Terminalia chebula* seeds mediated ZnO nanoparticles and their Larvicidal activity

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### Abstract

Green synthesis of nanoparticles can be an important alternative compared to conventional physiochemical synthesis. *Terminalia chebula* seed aqueous extract as a capping and stabilizing agent for the synthesis of pure zinc oxide nanoparticles (ZnO NPs). Further, the synthesized ZnO NPs were subjected to various characterization techniques for their functional group and particle size as 37nm. Larvicidal activity of green synthesized ZnO NPs were tested against Anopheles third instar larvae showed significant LC50 value of 85ppm with regression coefficient R<sup>2</sup> as 0.91. Our study concluded that *T. chebula* seed mediated ZnO NPs showed an effective larvicidal activity against malarial vector Anopheles third instar.

**Keywords:** ZnO NPs, *Terminalia chebula*, FTIR, UV, XRD, Larvicidal activity

## 1. Introduction

Nowadays, there is a lot of interest in the topic of nanotechnology. Three general methodologies have been identified for the process of nanotechnology: computational, wet, and dry. The wet method works with materials found in living beings' cells, tissues, and membranes, whereas the computer approach only works with structures the size of nanometers. Furthermore, the synthesis of inorganic compounds using physical chemistry techniques is handled by the dry process. The primary purpose of nanotechnology is said to be the creation of nanoparticles, which primarily uses three approaches: chemical, biological, and physical. When comparing these techniques to the other two, biological synthesis is a significant factor. Eco-friendly synthesis, which includes plants and plant materials and has the associated benefits of cost reduction and simplification, is a subset of biologically-mediated synthesis. Thus, we choose to concentrate mostly on the environmentally friendly production of nanoparticles (Anzabi et al., 2018; Begum et al., 2018; Fatimah 2018; Elango et al., 2016; Kumar et al., 2014; Madhumitha et al., 2014; Jeyaseelan et al., 2013; Roopan et al., 2013; Velayutham et al., 2013)

Zinc oxide (ZnO) nanoparticles are used in a variety of fields, including material applications, photoprinting, gas sensors, UV lasers, sunscreen lotion cosmetics, and medicinal creams. They are also employed in the biological, pharmacological, cosmetic, environmental, and catalytic fields. ZnO nanopowder is a potential material for many applications, such as transparent electrodes, pH sensors, biosensors, acoustic wave devices, and UV photodiodes. It is currently utilised in products such as plastics, ceramics, glass, cement, rubber, lubricants, and fire retardants, among others (Ma *et al.*, 2013; Ravichandrika *et al.*, 2012; Ahmad and Zhu, 2011).

A member of the Combretaceae family, *Terminalia chebula* is a plant that is used in many countries, including Nepal, India, Sri Lanka, China, Vietnam, and Malaysia, to treat a variety of human maladies, including fever, TB, indigestion, and cardiac and hepatic disorders. Plant material has antimicrobial, antitussive, antispasmodic, antiasthmatic, and antihelminthic properties, among other medical benefits (Singh and Kumar 2013; Gilani et al., 2008a; Ravi Shankara et al., 2019; Gilani et al., 2005a). Toxicologists have come to accept the use of LC50, which is typically the most widely regarded test for determining the possible harm that chemical pollutants may cause to living things (Suganthi 2019). The objective of this study is to green synthesize and characterize the metallic ZnO nanoparticles and also to study their larvicidal property against malaria causing vectors – *Anopheles* larva.

## 2. Materials and methods

### 2.1. Green synthesis of Zinc oxide (ZnO) nanoparticles

The seeds of *Terminalia chebula* are extracted and ground from medicinal plant products. 200ml of distilled water is combined with 40g of plant material, and the mixture is filtered by Whatmann paper. Combine 180ml of aqueous zinc acetate (1mM) with plant filtrate. The combination was maintained for 24 hours at 35°C

during plasmon excitation (incubation) to reduce zinc ions with phytochemicals. According to Naif and Mariadhas (2018), the mixture's colour changed to white powder, which is seen as an indication of the reduction of zinc ions for the creation of zinc nanoparticles. At 10,000 rpm the mixture was centrifuged for 15 minutes after 8 hours. Supernatant was collected and repeatedly rinsed with distilled water. Zinc nanoparticles mediated by *T. chebula* seeds are finally produced.

## 2.2. Characterization of green synthesized ZnONPs

The active groups of the green synthesised zinc nanoparticles are examined using potassium bromide pellets and FTIR spectroscopy (Nicolet 660 Avatar, US). The associated particles are detected in the 400–4000/cm range (Nandiyanto et al., 2019). Using Shimadzu UV-2550 UV visible spectroscopy, the functional and active groups of the phytochemical-mediated silver nanoparticles are examined (Yulizar et al., 2019). XRD (Shimadzu 7000) is utilised to analyse the Zn NPs' 2 $\theta$  angle (Klekotko et al., 2019).

## 2.3. Larvicidal activity

Malarial vector *Anopheles* larvae were cultivated in the KIRND laboratory (Tiruchirappalli, Tamil Nadu), Larvicidal investigations were conducted using collected third-instar larvae (Benelli 2018). ZnO NP concentrations, ranging from 1, 5, 10, 20, 40, 80, and 160 ppm, were generated using sonication and then added to 2L experimental tanks containing ten individuals each. A commercial insecticide called "Neemazal" was employed as a control (Volker et al., 2013). Regression analysis was performed on the mortality data, and an LC50 was determined (Suganthi 2019).

## 3. Results and Discussion

### 3.1. Green synthesis and Characterization of ZnO NPs

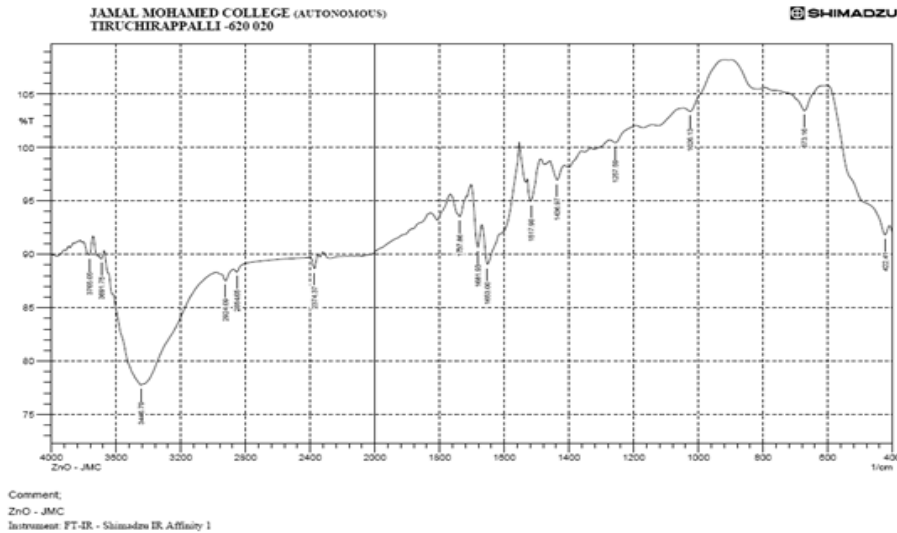
**FTIR:** The green synthesised ZnO NPs (Figure 1) FTIR spectra showed the wide absorption in the 3750–3000cm<sup>-1</sup> frequency range was attributed to O–H stretching from Zn–OH, water, and leftover alcohols. For ZnO NPs, the absorption peaks are seen at 3446cm<sup>-1</sup>. For ZnO NPs, the CO<sub>2</sub> peaks were measured at 2374cm<sup>-1</sup>. These CO<sub>2</sub> bands could be the result of some CO<sub>2</sub> being trapped in the ambient air. The ZnO samples' H-OH bending vibration bands were centred at 1517cm<sup>-1</sup>, indicating a trace of H<sub>2</sub>O in both samples. The stretching of the vibration in ZnO is responsible for the most intense broad absorption band at about 438cm<sup>-1</sup>. For ZnO NPs, the Zn–O stretching bands are 422cm<sup>-1</sup>.

**UV- Vis spectroscopy:** Sharp peaks were seen in the absorption spectrum of the ZnO sample at 382nm (Figure 1), which are thought to be caused by free excitons at the band edge. It was anticipated that the ZnO NPs would exhibit a slight redshift in relation to bulk ZnO. ZnO NPs were discovered to have band gap energies (E<sub>g</sub>) of 3.2 eV. revealed a little "red shift" of 0.17 eV from the ambient temperature conventional bulk band gap (E<sub>g</sub> = 3.37 eV).

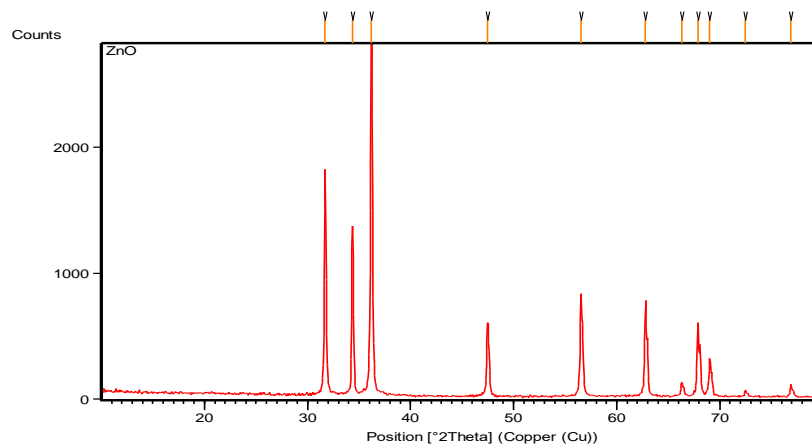
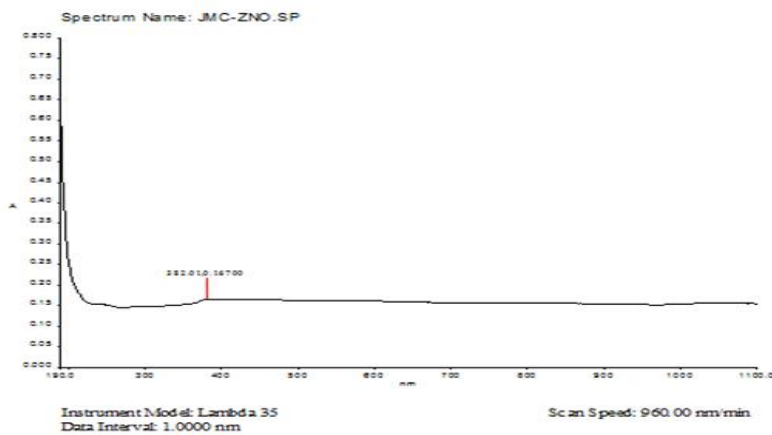
**XRD:** The (100), (002), and (36.183) angles (2 $\theta$ ) at which the XRD peaks were found correspond to the (100), (002), and (36.183) planes of the ZnO NPs, respectively (Figure 3a). Similar to the above, ZnO NP planes (102, (110), (103), (200) (112), (201), (004), and (202) were corresponding to other peaks observed at

angles ( $2\theta$ ). ZnO NPs' Zn-O bond length was measured to be 1.9795Å. The average size of ZnO NPs is 37nm.

**Figure 1. FTIR, UV-Vis and XRD Characterization of green synthesised ZnO NPs**



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ZnO NPs made from pure leaf powder from *Scadoxus multiflorus* were capping and stabilising agents made from an aqueous extract. An average particle size of  $31 \pm 2$  nm was reported after green synthesised ZnO NPs were exposed to

different characterization techniques, including UV (with the greatest absorbance of 274 nm), FTIR (with peaks at 3003 and 1730 $\text{cm}^{-1}$ , ~417  $\text{cm}^{-1}$ ), and XRD (Naif and Mariadhas 2018).

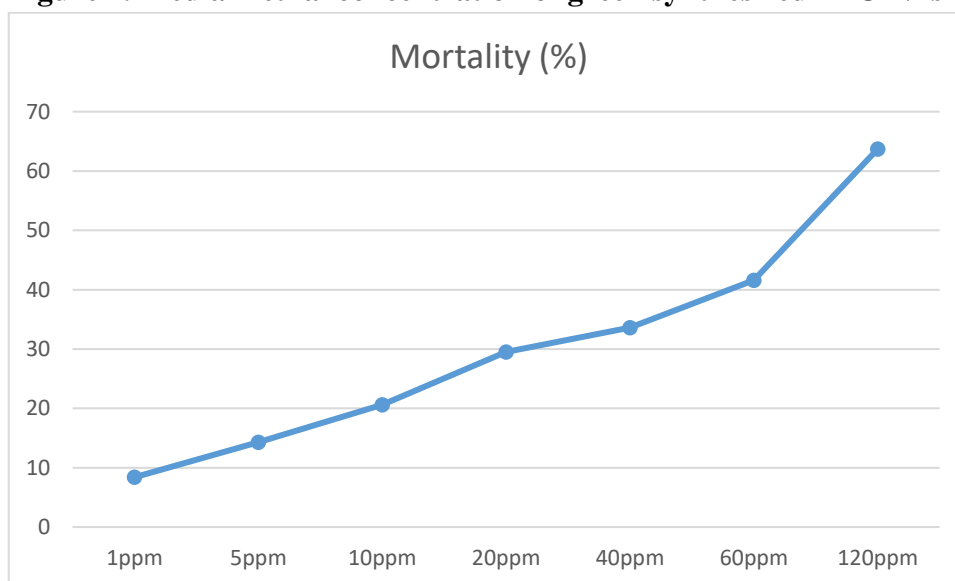
### 3.2.Larvicidal property

Larvicidal Activity of ZnO NPs on malaria-causing vectors (*Anopheles* larva) were studied by treating the third instar larva with ZnO NPs at various concentrations: 1, 5, 10, 20, 40, 60 and 120 ppm. The percentage mortality (Table 1) were 8.4 $\pm$ 1.2, 14.3 $\pm$ 1.6, 20.6 $\pm$ 1.9, 29.5 $\pm$ 2.5, 33.6 $\pm$ 3.0, 41.6 $\pm$ 3.1 and 63.7 $\pm$ 3.5% respectively whereas the control (NeemAzal) group mortality was observed as 30.5 $\pm$ 2.3% (Table 1). This mortality percentage indicates a dose-dependent reaction at higher concentrations, as well as an increasing death rate. The regression coefficient of treated groups were statistically observed as  $R^2=0.91$  whereas the LC50 was observed at 85ppm (Figure 2).

**Table 1. Green synthesized ZnO NPs mortality activities on *Anopheles* (3<sup>rd</sup> instar) larva**

Group	Mortality (%)	Regression Coefficient	LC50 value
Control (NeemAzal)	30.5 $\pm$ 2.3	$R^2=0.91$	85ppm
1ppm	8.4 $\pm$ 1.2		
5ppm	14.3 $\pm$ 1.6		
10ppm	20.6 $\pm$ 1.9		
20ppm	29.5 $\pm$ 2.5		
40ppm	33.6 $\pm$ 3.0		
60ppm	41.6 $\pm$ 3.1		
120ppm	63.7 $\pm$ 3.5		

**Figure 2. Median lethal concentration of green synthesized ZnO NPs**



ZnO nanoparticles made from *T. chebula* fruits shown effective photodegradation of methylene blue dye in addition to increased scavenging and lowering capabilities against reactive oxygen radicals (Rathika et al., 2023). Elango et al. (2016) and Ashokan et al. (2017) reported similar findings, indicating that ZnO nanoparticles had decreased larvicidal action. This could be because the particles' surface did not contain a bio-organic phase. ZnO NPs produced by *Sargassumwightii* have a greater LC50 value (49.22 ppm) (Ishwarya et al., 2018a). ZnO NPs made from *Ulvalactuca* were tested for their larvicidal potential against *A. aegypti*, and the results indicated an IC50 value of 22.38 ppm (Ishwarya et al., 2018b).

According to Naif and Mariadhas (2018), ZnO NPs mediated by *Scadoxusmultiflorus* have remarkable LC50 value of 34.04 ppm against dengue-causing vector *Aedes aegypti* larvae. According to Velayutham et al. (2013), extracts from *Terminalia chebula* have larvicidal efficacy against *A. aegypti* and *Culex quinquefasciatus*, two disease-causing vectors.

### Conclusion

According to the study's findings, ZnO NPs mediated by *Terminalia chebula* seeds have potential uses in the pharmaceutical and biological fields and can be employed as useful instruments for controlling mosquito larvae populations.

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**Conflict of interest:**None

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