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## Coriander Extract Mediated Green Synthesis of Zinc Oxide Nanoparticles and Their Structural, Optical and Antibacterial Properties

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### ABSTRACT

In this work, synthesis, morpho-structural characterization, optical and antibacterial properties of zinc oxide nanoparticles have been reported. Zinc oxide nanoparticles were prepared at room temperature via green route using *Coriandrum sativum* leaf extract. The product was characterized by X-ray diffraction (XRD), Transmission electron microscopy (TEM), Fourier transform infrared spectroscopy (FTIR), Scanning electron microscopy (SEM) with energy dispersive X-ray analysis (EDX) and UV-Visible spectroscopy to ascertain its formation. Morphology study indicates spherical nature of the nanoparticles and the formation of zinc oxide nanoparticles was confirmed by EDX. XRD analysis showed crystalline nature of the ZnO with wurtzite structure having average crystallite size of 60 nm which was also corroborated by SEM and TEM analyses. Furthermore, the estimated band gap was 3.8 eV as determined by UV-Visible spectral analysis. Antibacterial efficacy of the prepared nano particles against the pathogenic bacteria *E. coli* reveals that the ZnO nanoparticles have considerably higher antibacterial potential and is possibly due to a combination of events. Besides, the outcomes of the synthesis certainly contribute to the developing a better understanding of simple, low-cost, green and nontoxic synthesis method and growing the knowledge base needed to design a suitable antibacterial materials and framework for advanced and wise applications.

### 1. Introduction

In past few decades, generation of bacterial infection in industrial sectors including environmental, food, personal care products, synthetic textiles, packaging, healthcare, clinics, and public health threat due to emergence of resistance to antibiotics with their excess uses in bacteria have received worldwide attention of scientific community to develop new age antimicrobial agents, formulations and methods. Metal oxide nanoparticles exhibit remarkable biological applications and are of great interest for both fundamental research and technological development [1-8]. Several investigators are trying to develop the proper growth and processing techniques for the synthesis and to manipulate the features of metal oxide nanoparticles (Nps). Among the metal oxides, zinc oxide being a promising multifunctional material has stimulated intensive research interest due to its unique optical, electrical and chemical properties which is well exemplified by the number of previous articles. Having a large band gap (3.3 eV), large exciton binding energy, good transparency and high electron mobility, it is very useful material for the optoelectronic applications [3, 4]. It is worth noting that zinc oxide nanoparticles can be used in various applications such as biotechnology, drug delivery, medical, optical devices, DNA labelling, bio molecular detection, diagnostics [5] optoelectronics, nanogenerators, biosensors, solar cells, photocatalysts lubricants, rubber, photo detectors, cement ceramics sensors etc., [6,7]. Furthermore, because of their large surface area these nanoparticles are used in waste water treatment [8] and in the removal of impurities like arsenic and sulphur from water or waste waters [9]. There are a number of chemical and physical methods reported in literatures for the preparation of nanoparticles of different sizes, shapes, and compositions which require high temperature, pressure, extremely expensive and toxic chemicals, hazardous reagents and nonpolar solvents, imposes an inordinate challenge before materials scientist [10, 11]. To overcome the limitations of these conventional methods, green synthesis has been emerged as an ecofriendly alternative for low-cost development of nanoparticles, which are highly efficient and biocompatible and can be

used in a variety of applications [12-14]. It has been previously reported that among the different biological synthesis methods of nanoparticles plant-mediated synthesis are rapid, potentially advantageous and have more stability in various shape and size than the other organisms, as the microorganisms based synthesis is not of industrial viability due to necessity of multiple purification steps, highly aseptic conditions and their maintenance [15, 16]. A large number of plants have been reported in recent years for the synthesis of nanoparticles from the extract of various parts of plants such as eucalyptus hybrid, *Coriandrum sativum*, *Cycas*, *sorghum bicolor*, *Azadirachta indica*, *Cassia auriculata*, *Camelliasinensis*, *Aloevera*, *Calotropisgigantea*, *Nelumbo nucifera*, *Ocimum sanctum* and many more [17-25]. In this manuscript, ZnO nanoparticles were synthesized and stabilized using *Coriandrum sativum* (annual herb) leaf extract by the bio reduction method without using any harmful reducing and capping agent, and also different applications have been reported. Image of *Coriandrum sativum* leaves has shown in Fig. 1. *Coriandrum sativum* is a medicinal plant also known as “dhania” belongs to family Apiaceae [26, 27].



Fig. 1 Image of *Coriander sativum* leaves

### 2. Experimental Methods

#### 2.1 Materials

Zinc acetate dihydrate and sodium hydroxide from Fisher Scientific (U.K.) were used in the experiments for the synthesis of zinc oxide nanoparticles. All the chemicals were of analytical reagent grade purity.

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## 2.2 Preparation of Coriandrum sativum Leaf Extract

Fresh leaves of *Coriandrum sativum* were washed thoroughly several times with biodegradable and distilled water to remove the dust particles and other unwanted impurities and cut it into fine pieces. 30 g of the leaves were mixed with 120 mL of ethanol and heated at 80 °C for 30 minutes. Resulting extract was cooled at room temperature and filtered by whatman filter paper no.1 [18].

## 2.3 Synthesis of Zinc Oxide Nanoparticles

In typical synthesis, 100 mL of 0.1 M zinc acetate dihydrate was added first to leaf extract in ratio 1:1 with continuous stirring at room temperature for complete dissolution. Subsequently 20 mL of 1.0 M NaOH solution was slowly added under stirring to increase the pH value and in turn for the precipitation [18]. A progressive change in the color of solution was observed during the stirring process and the change from sturdy brown to yellowish white confirms the nanoparticles formation. The obtained precipitate was washed with ethanol to remove the impurity of all the ions produced during the reaction and dried for 4 hours at room temperature. After the calcinations of precipitation at 400 °C in furnace, yellowish white powder was obtained.

## 2.4 Characterization of ZnO Nanoparticles

The crystal structure of the synthesized nanoparticles were examined with powder X-ray diffraction (XRD) and the pattern were recorded using X-ray diffractometer (Phillips X Pert model) equipped with  $\text{CuK}\alpha$  radiation ( $\lambda = 0.1542 \text{ nm}$ ) within the  $2\theta$  range of  $10^\circ$ – $70^\circ$ . Morphological characteristics, Size and composition of nanoparticles were determined by scanning electron microscopy (SEM) equipped with the X-Ray energy-dispersive spectrometry (EDX). SEM images were captured on a LEO 435 VP instrument operated at 25 kV; EDX was carried out by JEOL JXA-8100 EPMA. To obtain further information as well to verify the size and shape of nanoparticles, transmission electron microscope (TEM- 200 KV, TECNAI G2, S-TWIN, FEI, Holland) was employed. Size distribution of particles was examined by the Zitasizer NanoZS-90. Fourier transform infrared spectroscopy analysis was performed by Broker FTIR alpha model to theorize the functional groups involved in the synthesis. The optical properties of synthesized nanoparticles were deliberated with the help of UV- visible spectroscope (EVOLUTION-201).

## 2.5 Antimicrobial Activity of Zinc Oxide Nanoparticles

Herein, Agar well diffusion method was used to assess the antimicrobial activity of synthesized nanoparticles [28, 29]. For this purpose, bacteria *Escherichia coli* were employed as test organism. In typical process, 25 mL of nutrient agar was poured into sterile petri plate. The plates were allowed to solidify and then 100  $\mu\text{L}$  of 24 hrs culture of *E. coli* was spread on the petri plate. Sterile cork borer was used to make the wells in plate. Zinc oxide nanoparticles solution was prepared at a concentration of 100, 200, 300 and 400 mg/mL. About 100  $\mu\text{L}$  of prepared solution were loaded in the wells. Thereafter, antimicrobial activity was screened quantitatively by recording diameter of inhibitor zones plate after incubation for 24 hrs at 30 °C.

## 3. Results and Discussion

In the green synthesis of zinc oxide nanoparticles, biochemical reactions are involved, wherein biological molecules react with the precursor leading to reduction of metallic ions. *Coriandrum sativum* leaf extract contains minerals and vitamin contents including calcium, phosphorus, iron, sodium and also oxalic acid, 2- decenoic acid, capric acid, tridecanoic acid, undecanoic acid and E-11 tetra decanoic acid in larger amount which are responsible for reduction process [30]. Moreover, characterization of green synthesized ZnO nanoparticles by various techniques unfolds their structural and optical properties.

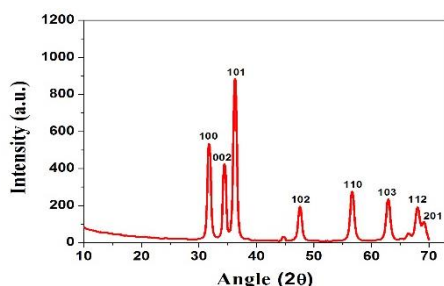


Fig. 2 XRD pattern of zinc oxide nanoparticles

XRD pattern of synthesized sample is shown in Fig. 2. The sharp and narrow diffraction peaks suggest that synthesized product has the good crystallinity with relatively larger crystallites, and signifies the effects of experimental conditions on the nucleation and growth of the crystal. The XRD patterns shows the noticeable peaks corresponding to  $2\theta$  values of  $31.64^\circ$ ,  $34.43^\circ$ ,  $36.29^\circ$ ,  $47.75^\circ$ ,  $56.51^\circ$ ,  $62.90^\circ$ ,  $67.98^\circ$  and  $69.23^\circ$ . The prominent peaks correspond to (hkl) values of (100), (002), (101), (102), (110), (103), (112), (112) and (201). These plane values are closely matched with wurtzite structure of ZnO [31]. Additionally, no characteristic peaks other than the ZnO appears which in turn specify the high purity of our sample. The average crystalline size of nanoparticles was estimated using the Debye-Scherrer formula [32],  $D = 0.89 \lambda / \beta \cos\theta$ , where,  $\lambda$  (1.54 Å) is the wavelength of X-ray,  $\theta$  being Bragg's diffraction angle and  $\beta$  is the full width at half maximum. The calculated value of crystalline size was found to be 60 nm, which obviously supports the SEM and TEM results.

Typical SEM images of the ZnO nanoparticles are shown in Fig. 3. SEM images revealed that the particles are well shaped and spherical in the size range 30-150 nm, and there is fairly large number of individual small nanoparticles along with very few agglomerated relatively bigger particles. As depicted in Fig. 4, the EDX micrograph of the ZnO reveals the purity of the sample. Both zinc (Zn) and oxygen (O) are present in the sample with weight percentage 90.86% and 9.14% successively; and atomic percentage 70.88% and 29.12% proves that the prepared ZnO is essentially free from impurities.

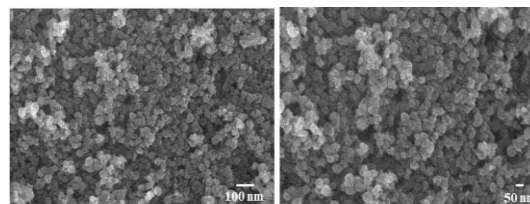


Fig. 3 SEM images of ZnO nanoparticles synthesized using Coriandrum leaf extract

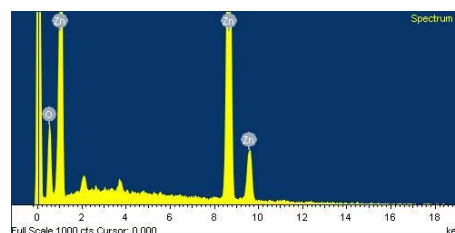


Fig. 4 EDX spectra of ZnO nanoparticles synthesized using Coriandrum leaf extract

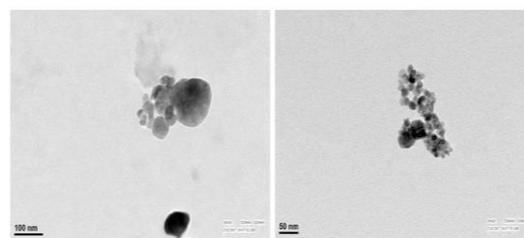


Fig. 5 TEM images of ZnO nanoparticles synthesized using Coriandrum leaf extract

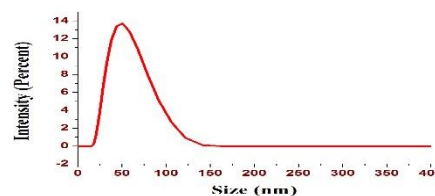


Fig. 6 Particle size distribution of ZnO nanoparticles synthesized using Coriandrum leaf extract

TEM images shown in Fig. 5 too confirm the nanosize and spherical morphology of zinc oxide prepared, as indicated in SEM images. Average particle size estimated to 40 nm, which nearly matched with crystalline size of zinc oxide nanoparticles determined from the XRD analysis.

To determine the distribution of particle size in the synthesized sample, we have used dynamic light scattering technique. To ensure this, synthesized ZnO nanoparticles in powder form were fully dissolved in hydrochloric acid for the preparation of representative solution followed by ultrasonication for uniform distribution of nanoparticles. Fig. 6 shows the size distribution of the dispersed nanoparticles. The image reveals a

maximum intensity at the average particle size of 55 nm, and thereby results suggest that the synthesized ZnO is in nanosize form. Here, difference in the average particle size obtained by DLS and TEM images may clearly be seen. The slightly larger diameter given by DLS may be attributed to the presence of a few aggregates of nanoparticles.

Fig. 7 shows the FTIR spectra of green synthesized ZnO nanoparticles. The spectra were recorded in the series of absorption band 4000  $\text{cm}^{-1}$  to 500  $\text{cm}^{-1}$ . The band at 3500  $\text{cm}^{-1}$  is possibly attributed to the O-H stretching of hydroxyl group, whereas the peak observed at 1630  $\text{cm}^{-1}$  identifies the presence of asymmetrical stretching of COO<sup>-</sup> (carboxylate) of Zn [33]. Broad peak at 1029  $\text{cm}^{-1}$  occurs due to the stretching vibration between C-N bonds of amine [34]. Moreover, absorption band at 548  $\text{cm}^{-1}$  identifies the occurrence of ZnO nanoparticles [35]. Thus, it can be observed from the FTIR spectrum that *Coriandrum sativum* leaf extract is significantly rich in different chemical groups such as -OH, -COOH and amine. Further, hydroxyl group probably due to the presence of phenols [36]. The presence of phenols probably indicates the existence of polyphenolic tannins which also prevent aggregation, play an important role in the reduction and stabilization of ZnO nanoparticles. Thus, physicochemical properties of *Coriandrum sativum* act as a bio template that prevents agglomeration.

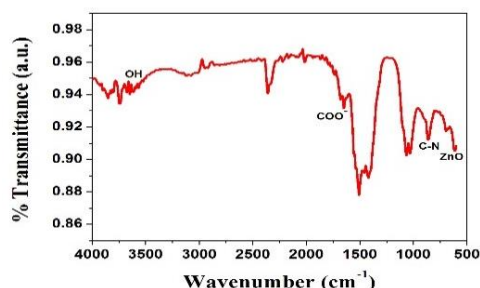


Fig. 7 FTIR of ZnO nanoparticles synthesized using *Coriandrum* leaf extract

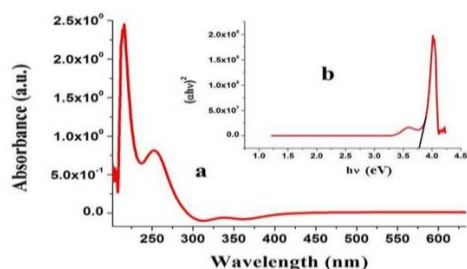


Fig. 8 (a) Absorption spectrum of ZnO nanoparticles (b) Tauc plot of ZnO

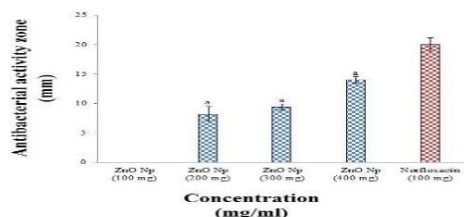


Fig. 9 Antibacterial activity (zone of inhibition, mm) of zinc oxide nanoparticle solution against pathogenic *Escherichia coli*. (ZnO Np = Zinc oxide nanoparticle; Average value given is mean of three replicates with standard deviation and significant value were predicted by using ANOVA (<sup>a</sup>  $p < 0.01$ , <sup>b</sup>  $p < 0.05$ ) by comparing data of each concentration of ZnO Np with 100mg Norfloxacin).

Absorption spectra analysis of synthesized sample was performed to study the optical property and to confirm the presence of ZnO nanoparticles. This spectrum is displayed in Fig. 8(a). A perusal of Fig. 8(a) reveals strong absorption peak at 272 nm within the absorption range 200–650 nm, which further confer ZnO nanoparticles formation [37]. When compared to bulk ZnO (369 nm), the observed blue shift clearly indicate a widening of band gap. Intense and sharp absorption further reveals the individual distribution of the nanoparticles. The band gap energy was determined using Tauc equation  $\alpha h\nu = B (h\nu - E_g)^m$  for direct band gap material [38], where,  $\alpha$  is the absorption coefficient for direct band gap material,  $\beta$  is the constant and has different values for the different transitions, B is constant and has different values for different transitions.  $E_g$  is the energy gap and  $h\nu$  is the photon energy while m denotes an exponent which have values  $\frac{1}{2}$ ,  $\frac{3}{2}$ , 2 and 3 depending on the electronic transition [39]. Tauc plot as shown in Fig. 8(b) has the photon energy ( $h\nu$ ) on the X axis and a quantity  $(\alpha h\nu)^2$  on the Y axis; and extrapolating the linear portion of the curve to the X axis yields the energy band gap of the

material. The gap was found to be 3.8 eV which is comparable to the values obtained by others [40–43]. Moreover, our calculated value is higher than that of bulk ZnO (3.3 eV), which indicates that the as prepared ZnO nanoparticles exhibit strong quantum confinement by modifying valence and conduction bands of ZnO semiconductor.

The observed sizes of the inhibition zones are illustrated in Fig. 9, which specifies that synthesized zinc oxide nanoparticles exhibit antibacterial activity against *E. coli*. *E. coli* is a gram negative bacterium which produces predominantly extended-spectrum beta-lactamases enzymes which contribute to tolerance or resistance to many antibiotic drugs such as penicillin, cephalosporin and carbapenems etc. [44]. The results of antibacterial activities of our samples were compared with standard antibiotic norfloxacin (100 mg). From the column graph, it is fascinating to note that ZnO nanoparticles exhibited strong toxicity to bacteria except at the concentration 100 mg/mL. With increasing concentration of Nps, zone of inhibition increases and is maximum at the concentration of 400 mg/mL; which shows efficient and comparable antibacterial activity as the norfloxacin. These results are consistent with the literature. Though there are several mechanisms are attributed to the bactericidal activity of ZnO nanoparticles, the exact mechanism is not known and is still under debate. Nevertheless, the possible mechanism for the bactericidal activity, explained as follows, involves the factors including mainly reactive oxygen species (ROS) and the release of Zn ion [45–49]. It has been found in the literature that the activity depends up on crystallite size, morphology, composition, specific surface and phase of crystalline material [48]. Because of larger surface area than the bulk and distinguishable porosity as the particle size reduces, greater number of reactive oxygen species (ROS), namely  $O_2^-$ , hydrogen peroxide ( $H_2O_2$ ),  $OH^-$ , and organic hydroperoxides are generated due to small crystallites, which in turn are considered responsible for the damage of cellular composition such as lipids, phosphorus-containing elements (DNA) and disabling of proteins by oxidative stress [50, 51]. It should be noted that oxidative stress results when reactive oxygen species production surpasses the capability of antioxidant defense of the cell [51].

Moreover, it is written somewhere [52] that the ZnO nanoparticles have positive zeta potential. Owing to this, nanoparticles provide enhanced particle surface reactivity and adherence with the microbial pathogens; and so surface properties also affect the interactions with the cell walls of the bacteria by allowing easy penetration into the bacteria. The exposure of nanoparticles to the surface of bacteria or accumulation of nanoparticles in the cell or even in the cytoplasm inhibit several functions in the cell thereby inducing structural abnormalities such as distraction of cellular function or deformation or rupture or blebs in membranes, leading eventually to death of bacterial cells [53, 54]. It is therefore evident from the above analysis that the interplay between the membrane biology of the pathogen and chemical and physical properties of medium appears to be most important aspect of bactericidal properties

#### 4. Conclusion

In summary, synthesized zinc oxide nanoparticles by *Coriandrum sativum* have significant shape and size, which were confirmed by SEM, XRD and TEM techniques, also with greatly reduced toxicity than the chemical and physical synthesis methods. FTIR and EDX showed the chemical contents and purity of the sample. TEM results demonstrate the presence of spherical nanoparticles of average particle size 40 nm, whereas dynamic light scattering examination shows average particle size of 55 nm. Green synthesized ZnO NPs exhibit useful and potent antibacterial activity against the test bacteria *E. coli*. The interplay between the membrane biology of the pathogen and chemical and physical properties of medium appears to be most important aspect of bactericidal properties. Conclusively, green method provides progression over chemical and physical method and synthesized material by this approach will not be used only in engineering field but also in drug delivery advantageously.

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