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Green Synthesis of Selenium Doped Zinc Oxide Nanoparticles using *Mangifera indica* Leaf Extract and Its Photodegradation and Antibacterial Activities

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ABSTRACT

Biomolecule-mediated nanoparticle synthesis has recently gained the attention of researchers due to its eco-friendly and non-toxic nature. Herein, we aimed to describe the rapid green synthesis of selenium doped zinc oxide nanoparticles by using aqueous leaf extract of *Mangifera indica* to overcome the chemical synthesis. The reduction of precursors used for nanoparticles is activated by natural reducing agents present in plant extract which eliminates the neurotoxic reducing agents present in chemicals. The nanoparticles were assessed for their stability by varying parameters like pH, temperature, time and concentration. The highly stable nanoparticles were characterized using UV–visible spectroscopy, X-ray diffraction, Fourier transform infrared spectroscopy and field emission scanning electron microscopy with EDAX. Further the synthesized nanoparticles are investigated for photodegradation and antibacterial activity and found to be more effective.

1. Introduction

The intention of nanotechnology is to provide the living environment with a more qualitative and target specific solutions. Green chemistry emphasizes on the usage of plant extract as reducing agents in lieu of chemicals. The wastage and draining of chemicals can be avoided to a considerable extent in plant mediated synthesis. Biological processes and green chemistry synthesis of nanoparticles arise as alternatives to the conventional methods of synthesis. They use microorganisms, enzymes, algae and plant extracts to obtain the desired nanomaterial [1]. The leaves are generally used to prepare the extracts, since they represent no threat to food security [2-4].

The mango leaves (*Mangifera indica* Linn Anacardiaceae) known for its astringent and antioxidant properties are used to treat stomach related ailments [5]. The mango leaf is rich in phytochemicals, which are vital in health promotion, disease prevention and drug production [6]. Several types of polyphenols (phenolic acid, tannins and flavonoids) show anti-carcinogenic and anti-mutagenic effects [7- 10]. The phytochemicals in leaves like phenolic compounds, flavonoids, phenolic acids, diterpenes, tannin content helps in reduction of metal ions to metal nanoparticles and act as good capping and stabilizing agent [11].

Metal oxide nanoparticles stand out as one of the most versatile materials, due to their diverse properties and functionalities. Among the semiconductor oxide nanomaterials, zinc oxide nanoparticles are highly ionic; wide band gap and it has unique properties such as optoelectronic, ferromagnetic, piezoelectricity, catalysis, conductivity and sensing properties [12,13]. Selenium (Se) is a naturally occurring mineral in soil and being absorbed and accumulated by plants thereby entering the food chain. Selenium, present in trace amount in human systems is known for its toxicity when present in higher concentration.

Zinc oxide nanostructures are nontoxic, biologically compatible [14,15], have faster electron transfer rates [16] and thus find good application in the field of biosensor [17]. In nanoscale dimensions, selenium has shown reduced toxicity which can be applied in many applications in biological and medical fields [18-21] as well as in dye degradation. Based on above mentioned properties and applications, zinc oxide and selenium were chosen for the work.

The optical and electrical properties of ZnO nanoparticles have been changed due to the doping of transition metal ions in ZnO nanoparticles. This is attributed to the exchange interaction between s and p electrons of host ZnO and d electron of transition metal ion. Past few decades, extensive research has been done on the transition metal doped ZnO nanostructures. Cr-doped ZnO, Mn and Co- doped ZnO nanoparticles were synthesized by coprecipitation method [22,23]. Dyes are important organic pollutants and their release as waste water in the eco-system causes pollution. Photocatalytic degradation for the purpose of purifying waste water from industries and households has attracted attention in the recent years [24]. Antibacterial activity and photocatalytic degradation of Se doped zinc oxide nanoparticles synthesized using electrodeposition method has been studied [26]. Mg doped ZnO NPs synthesized using green method [27].

Here, the main focus of this study, offers a green technology to synthesize, characterize the selenium doped zinc oxide nanoparticles that can be applied in antimicrobial action, photodegradation, in order to enhance the activity of Selenium, and zinc oxide nanoparticles. Before converging, the basis synthesis is done by employing the leaf extract of *Mangifera indica*, for the reduction of zinc ions to ZnO nanoparticles, selenite ions to selenium nanoparticles (bottom up approach).

2. Experimental Methods

2.1 Chemicals

Zinc acetate [$ZnC_4H_6O_4$], sodium hydroxide, methylene blue and sodium selenite (Na_2SeO_3) were purchased from Merck. Ultrapure water employed for the experiments. To study the antibacterial activity, two bacterial strains *Escherichia coli* ATCC 8739 (Gram-negative), *Staphylococus aureus* ATCC 29736 (Gram-positive) were obtained from Microbiology Laboratory at PSG Hospitals, Coimbatore.

2.2 Preparation of Plant Extract

5 grams of thoroughly washed *Mangifera indica* leaves were finely cut, and soaked in 50 mL of deionized water and heated in the mantle for 20 min. The solution was kept for cooling at room temperature and then filtered using Whatman filter paper no. 1. The filtrate was collected in amber bottle and was stored at 4 °C for further experiment.

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2.3 Synthesis of Se Doped ZnO NPs

The preparation method involves addition of 40 mL aqueous solution of zinc acetate (0.05 M) to 40 mL of sodium selenite (0.01 M) solution and then 20 mL of plant extract was added under constant stirring at ambient temperature for 1 hour. The pH of solution was raised to 12 by the dropwise addition of NaOH solution (1.0 M). In order to monitor the formation of Se doped ZnO NPs, the synthetic procedure was carried out for ZnO NPs and SeNPs separately and characterized using UV-visible spectra (DRS). The formation of Se doped ZnO NPs was further confirmed by X-ray diffraction, FT-IR and FESEM with EDAX.

2.4 Characterization Techniques

The diffuse reflectance UV-Visible spectra (DRS) of the nanosized materials were recorded using a Cary100 UV-visible spectrophotometer. Crystal structure were determined by X-ray diffraction analysis under SEIFERT JSO-DEBYEFLEX 2002 model operated at 40 kV and a current of 30 mA with Cu-Kα radiation of wavelength 1.5406 Å. The XRD pattern was scanned in the 2θ range from 30° to 70° with a step size of 0.04°/s. The different functional groups present in the samples were analyzed by FTIR spectroscopy (Hitachi Ltd., Tokyo, Japan). The FTIR analysis was performed with KBr pellets and recorded in the range of 400–4,000 cm^{-1}. Size distribution and elemental composition of the nanoparticles were determined by using FESEM coupled with EDAX (HITACHI SO-6600, Japan). The photocatalytic degradation of dye was determined by spectrophotometer.

2.5 Determination of Anti-Bacterial Activity

The anti-bacterial activities of selenium doped zinc oxide nanoparticles synthesized by the leaf extract of *Mangifera indica* were carried out by agar well diffusion method. In this study, two different human pathogenic bacteria *E. coli* ATCC 8739 (Gram-negative) and *S. aureus* ATCC 29736 (Gram-positive) grown in Mueller-Hinton agar (MHA) medium. The controls used for *E. coli* are NA-Nalidixic acid (positive control), NV-Novobiocin (moderate control), AK-Amikacin (negative control). The controls used for *S. aureus* are E-Erythromycin (positive control), NV-Novobiocin (moderate control), C-Chloramphenicol (negative control). 150 µL of selenium, zinc oxide and selenium doped ZnO nanoparticles were loaded separately into each well of the petri plates. The plates were incubated at 37 °C for 24 h and the zone of inhibition (ZOI) was measured in terms of millimeter. These assays were carried out in triplicate.

2.6 Determination of Photocatalytic Activity

10 ppm solution of methylene blue (molecular formula: $C_{16}H_{18}ClN_3S$, molecular weight: 319.85 g/mol, λmax = 660 nm) was prepared by dissolving in distilled water. The dye solution was used as a test contaminant for evaluating photocatalytic activities of Se-doped ZnO nanoparticles. The investigation was carried out under sunlight to check the efficiency of Se-doped ZnO nanoparticles.

About 10 mg of selenium doped zinc oxide nanoparticles was added to 50 mL of methylene blue dye solution. The control was maintained without addition of nanoparticles. Before exposing to radiation, the reaction suspension was mixed well using shaker for 30 minutes to make equilibrium solution. To examine the photocatalytic activity, 20 mL of colloidal solution were transferred to centrifuge tube and centrifuged at 800 rpm to remove the dispersed catalyst. For the clear solution, the percentage transmittance was recorded after a regular interval of 30 minutes using a spectrophotometer.

Percentage of dye degradation was estimated by the following formula:

$$\% \text{ Decolorization} = 100 \times (C_0\text{-}C)/C_0 \quad (1)$$

where C_0 is the initial concentration of dye solution and C is the concentration of dye solution after photocatalytic degradation.

3. Results and Discussion

3.1 UV-Visible Spectra

Fig. 1 shows diffuse reflectance spectra of synthesized ZnO, selenium and selenium doped ZnO NPs by green method. ZnO NPs show strong absorption peak at about 360 nm; Se NPs show strong absorption peak near 620nm;Se doped ZnO NPs show peaks in 506nm.Since ZnO is an n-type semiconductor, the decrease in its band gap due to Se doping could be attributed to Burstein–Moss shift, which explains the shift in the absorption edge to higher energies due to merging of Fermi levels with the conduction band [28].

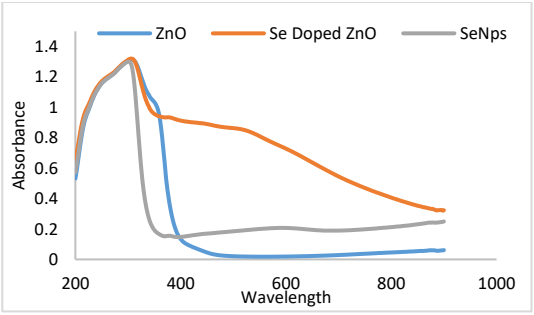

Fig. 1 UV visible spectrum of synthesised nanoparticles

3.2 X-Ray Diffraction

The composition and phase of the resultant samples were examined by XRD. Fig. 2 indicates that particles of synthesized Se doped ZnO NPs were crystalline in nature, which were in good agreement with JCPDS (File No. 06-0362) for SeNPs and JCPDS (File No. 36-1451) for ZnO NPs. From the XRD data it is evident that Se-doped ZnO nanoparticles exhibit dominant diffraction peaks at 21.97 (110), 31.854 (101), 47.61 (112) and 66.9720 (004) which were absent in ZnO [29]. The broadening of XRD peaks ascribed nano-sized formation of Se doped ZnO nanoparticles. The normal size of Se doped ZnO nanoparticles is 27 nm assessed using Debye Scherer's formula.

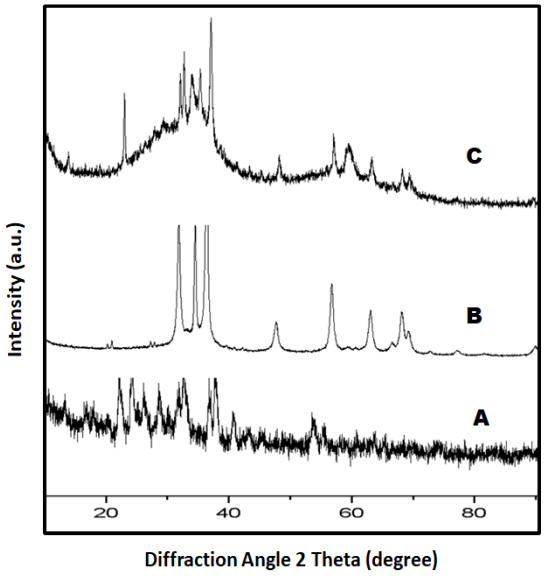

Fig. 2 X- Ray Diffraction of (A) Amorphous Se, (B) ZnO NP, (C) Se-doped ZnO NPs

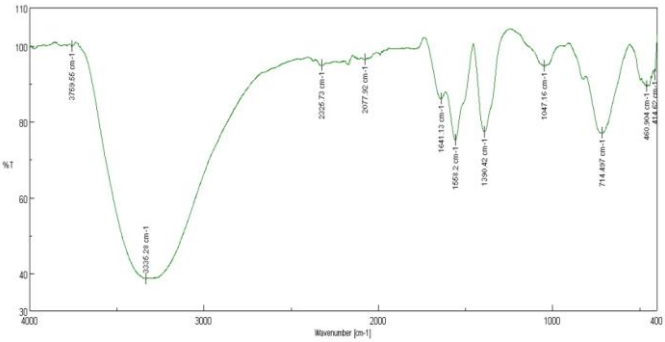

Fig. 3 FTIR spectrum of selenium doped ZnO NPs

3.3 FTIR Spectra

The synthesized Se doped ZnO NPs were characterized using FTIR in order to investigate the biological compounds responsible for the synthesis and stability of the particles. The result (Fig. 3) shows sharp absorption peaks at 3335.28 cm^{-1} and 1568.2 cm^{-1}. Peak at 3335.28 cm^{-1} can be assigned to OH and the one at 1568.2 cm^{-1} corresponds to the C-H vibration of the aromatic ring. Apart from these two prominent peaks, 1641.13 cm^{-1} which correspond to the N-H bend, 1047.16 cm^{-1} which correspond to M-O-M bonding present. Metal oxides generally give absorption bands in fingerprint region i.e. below 1000 cm^{-1} arising from inter-atomic vibration. The peaks at, 460.904 cm^{-1} are correspond to Zn-O

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stretching and deformation vibration, respectively. The presence of Se is confirmed with the appearance of the bands at 414 cm^{-1}, 714.497 cm^{-1} and 1390.42 cm^{-1} which were absent for ZnO [30].

3.4 Field Emission Scanning Electron Microscopy with EDAX

The dimension and morphology of Se doped ZnO NPs obtained from green synthesis were examined by FESEM and shown in Fig. 4. A perusal of figures shows that the synthesized Se doped ZnO NPs were spherical in shape and size ranges from 50 to 100 nm. Further, EDAX were recorded to investigate the purity and compositions of the Se doped ZnO NPs. From Fig. 5, it reveals the presence of Se and ZnO in synthesized NPs.

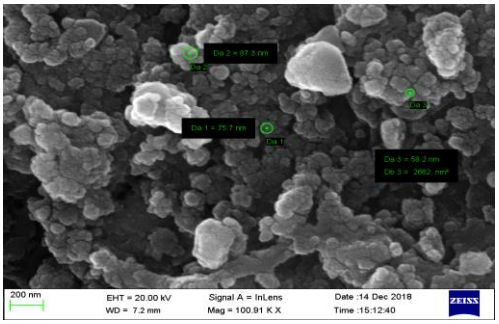

Fig. 4 FESEM of selenium doped ZnO NPs

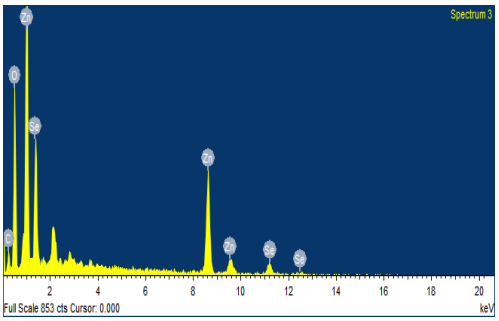

Fig. 5 EDAX of selenium doped ZnO NPs

3.5 Antibacterial Activity

This study shows that, the positive control (NA-Nalidixic acid), has highest antibacterial activity for *E. coli* bacteria is compared with synthesised Se doped ZnO NPs and the zone of inhibition is calculated as 18 mm for Se doped ZnO NPs. The positive control (E-Erythromycin) used for *S. aureus* is compared with synthesised Se doped ZnO NPs and the zone of inhibition is calculated as 13 mm for Se doped ZnO NPs. Using Se doped ZnO NPs, zone of inhibition is high in *E. coli* compared with *S. aureus*.

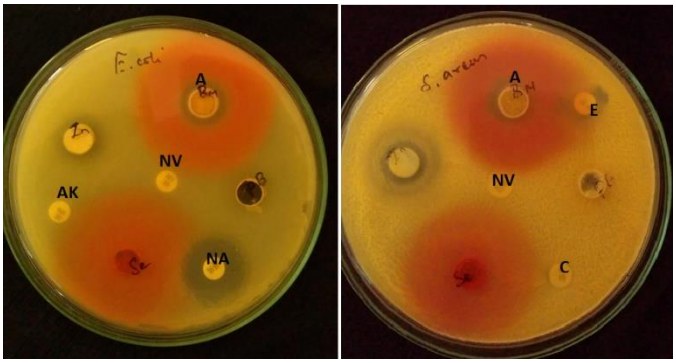

Fig. 6 Antibacterial activity of selenium doped ZnO NPs [NA-Nalidixic acid, NV-Novobiocin, AK-Amikacin E-Erythromycin, NV-Novobiocin, C-Chloramphenicol, A-Se Doped ZnO NPs]

3.6 Effect of Light Intensity

The kinetic study of photodegradation of methylene blue dye was studied under sunlight. The photodegradation rate was increased in sunlight for Se-doped ZnO NPs. The degradation of dye noted at different time intervals (Fig. 7), the absorbance shows as decreased. The completion of photocatalytic degradation of dye shows gradual decrease of absorbance value. The percentage of maximum degradation of methylene blue using selenium doped zinc oxide nanoparticles was calculated as 94.46%. Also, it depicts that Se doped ZnO acts as a good photocatalyst and is very active under sunlight.

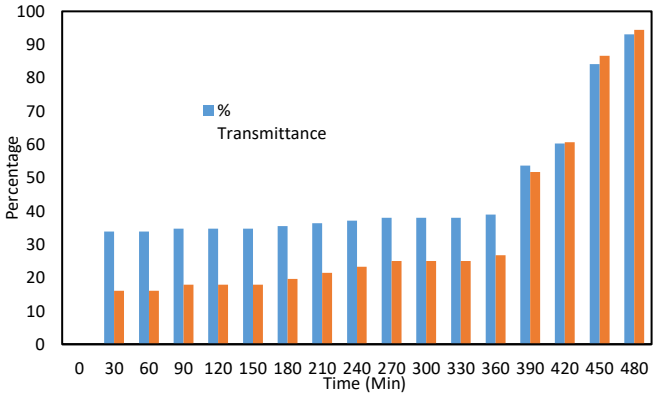

Fig. 7 Transmittance and decolourisation of methylene blue dye

4. Conclusion

Through greener routes, Se doped ZnO NPs are synthesized in highly alkaline pH. The size and structures of the obtained NPs were characterized by FESEM and XRD. Moreover, this plant mediated synthesis method represents a considerable improvement for the preparation of Se doped ZnO NPs because it allows better control over their nanostructures. The synthesized Se doped ZnO NPs had significant antibacterial activity and photocatalytic degradation.

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About the Conference…

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