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Green synthesis of zinc oxide nanoparticles using orange peel extract and their antibacterial activities

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Abstract

Nanoparticles present a higher surface area to volume ratio with increase in the size, morphology and distribution of the particles. The growing need of environmental friendly nanoparticles, researchers are using green methods for the synthesis of several metal nanoparticles for pharmaceutical applications. Nanoparticles has great number of application in many areas. Most of the antibacterial compounds are of bulk complex materials having increase toxicological side effects. Recent studies on antibacterial activity using nanoparticles have shown to reduce the side effects, although the data are both limited and contentious. The antibacterial activity of nanoparticles is attributed to their nano scale dimensions, changed electronic and optical transitions and high surface to volume ratio which leads to improved surface chemistry and quantum imprisonment effects. Compared to organic antibacterial agents, the main advantages of inorganic antibacterial agents are their good stability at increase the pressure and temperature and their long shelf-life.

Keywords: *Orange peel extract, Green synthesis, ZnO, Antibacterial activity*

1 Introduction

Zinc oxide nanoparticles are of particular concern since they are believed to be non-toxic, biocompatible and safe. Additionally, Zinc oxide nanoparticles have optical and electrical capabilities and catalytic and antimicrobial activities [1,2]. Lately, the use of Zinc oxide nanoparticles in agriculture is appear as a prospective tool to plant science, providing promising aspects for higher quality plant growth and yield to help minimize reliance on chemical fertilizers for sustainable agricultural development and food security and fulfill the nutritional needs of the world's fast-growing population [3,4&5]. In innovative agriculture Zinc oxide nanoparticles are explored to achieve their sustainable development and assess their capability for promoting growth by examining them as nano fertilizers in crops such as corn, wheat, pepper, tomato, onion. Antimicrobial activities of metal oxide, Zinc oxide nanoparticles against pathogenic microorganisms that can cause diseases in plants and animals, including Gram-positive and Gram-negative bacteria were quantitatively estimate in the culture media [6,7]. The detected reactive oxygen species generated by these metal oxide particles could be the considerable mechanism of their antibacterial activity [8]. The antibacterial mechanism of Zinc oxide nanoparticles encloses the direct interaction between Zinc oxide nanoparticles and cell surfaces affecting cell membrane permeability then nanoparticles enter and induce oxidative stress in bacterial cells, which cause the inhibition of cell growth and eventually cell death. Verification of antibacterial activity of Zinc oxide nanoparticles is promoting its application in the seed preservation of crops during the storage period before improvement [9,10].

Nanoparticles have found their usage in medicine, food industry and agriculture. The currently widespread physiochemical method of preparation of Nanoparticles is hazardous, expensive, environmentally unfriendly, requires condition of high temperature, pH, and pressure for synthesis. As such potential adverse effects on human as well as environmental have often been increased. Biological methods of synthesis of nanoparticles can supply a great ecofriendly and safe alternative supplied that it is sustainable and scalable [11,12]. Nanoparticles exhibit new and enhanced biochemical properties as well as clearly improved phenomenon and functionality. Given these size-controlled particles display significantly differing properties as compared to macro particles, these particles should be studied and characterized. Also, these modified chemical, physical, and morphological properties of Nanoparticles allow for a unique interaction with cellular molecules while facilitating the entry into the inner cellular compartments. Also, there is greater surface area and consequently greater reactivity of Nanoparticles when differentiate to the macro sized particles [13,14 &15].

Green synthesis is economically and ecologically viable way for synthesis of Nanoparticles for several industrial application. It is known that extracts may function as reducing as well as stabilizing agent during the synthesis

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of nanoparticles. Nobel metals like copper, Zinc, gold, silver, iron, zinc, platinum, and palladium have been frequently used for the creation of nanoparticles [16]. Nanotechnology is playing a significant role via its unique properties in several fields like engineering, food science, medicine, pest control and animal nutrition. Nanoparticles have large surface area, which leads to increased activity, reactivity, absorption and effectiveness. Thus nano materials could help to produce new pesticides and insect repellants. It is expected that nanotechnology could be effectively used with less dose and application times in insect nuisance control [17]. Recently, nanomaterials are considered as one of the most studied different to traditional neurotoxic grain protectants. For example, many researchers evaluated the toxicity of diatomaceous earths collected mainly of amorphous hydrated silica against a wide range of stored product insect pests, such as moths and beetles. However, the use of nanomaterials in agriculture, especially for production and plant protection is under-explored research area. Zinc oxide has been known as food and feed additive, and it was entitled [18]. Also, Zinc oxide nanoparticles are used in a wide range of applications such as nutrition, antibacterial, drug delivery, antibacterial, fertilizers and batteries. It evaluated the toxicity of zinc oxide and aluminum oxide nanoparticles against adults. Their results appear that both nanomaterials have absolute to strong toxic effect against the tested insects and significantly inhibited the progeny production. Green synthesis of nanomaterials not only reducing the usage and production of hazardous materials, but also it produces highly costly compounds from wastes. The worldwide attention of the Zinc oxide nanoparticle is multipurpose due to its distinctive properties, which differ from its bulkier components, as versatile semiconductor, piezoelectric properties. Zinc oxide nanoparticles have lately been used for various applications such as drugs, photo catalysis, cosmetics and animal dietary supplements [19,20 &21]. Therefore, more sustainable proposition are being developed in preparing Zinc oxide nanoparticles and extensive studies are being carried out to replace traditional methods. Microbial synthesis of metal nanoparticles has recently become irrelevant due to its low cost, environmental friendliness and biocompatibility [22]. Due to their efficiency Zinc oxide nanoparticle synthesis, a variety of microorganisms, including bacteria, yeast and fungi are being studied.

Zinc oxide nanoparticles were synthesized the green process using orange fruit peel extract and investigated the influence of pH and annealing temperature on antibacterial activity and morphology [23]. The morphology and structure of the Zinc oxide nanoparticles were characterized using a X-ray diffraction, and Fourier-transform infrared spectroscopy, Ultraviolent radiation and Energy dispersive X-ray analysis. Furthermore, the antibacterial activity of Zinc oxide nano fluids was tested against and E. coli*,* pseudomonas aeruginosa and Salmonella broth dilution method [24,25]. In order to optimize the bactericidal activity of the Zinc oxide nanoparticles, measurements were carried out with many types of Zinc oxide nanoparticles fabricated at various annealing temperatures and pH levels.

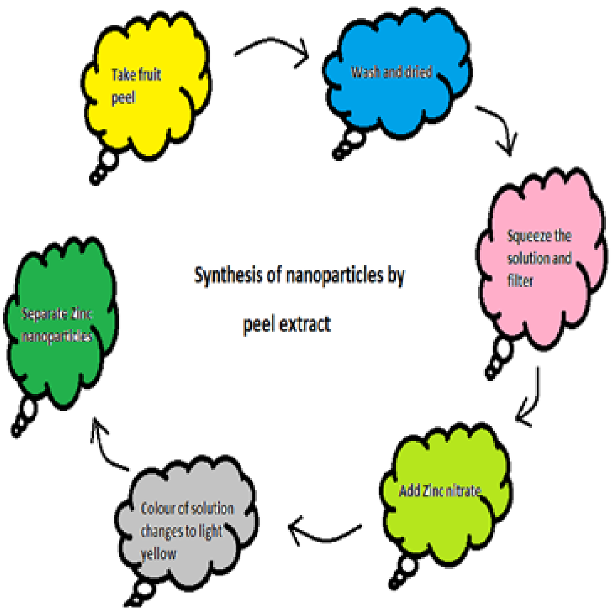

Fig 1. Green Synthesis Of ZnO using various plants and Biological sources

2 Experimental Sections

2.1. Materials and methods:

Oranges were rinsed in deionized water before being dried and peeled as unevenly as possible. The fruit peel was then dried in the sun for a week before being powdered into a fine powder. After that, 3g of fine powder was poured in a glass beaker with 50 mL deionized water and swirled for 3 hours with a magnetic stir. The concoction was then placed in a water bath. Finally these mixtures were filtered and the extract were stirred in normal atmosphere for later use. To create the ZnO nanoparticles, 50 mL of each extract was combined with 6 g of zinc nitrate. These mixtures were then stirred in a magnetic stirrer for 60 minutes before being placed in a water bath for another 60 minutes. The mixes were dried for one hour at 60°C in an air heated furnace at 100° C. Finally, a white colour powder was obtained, which was handled and packed with care for characterization.

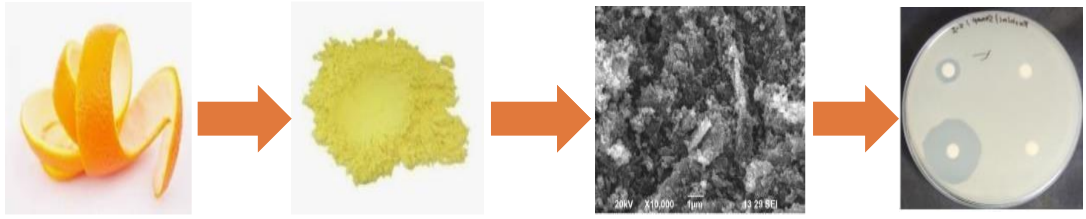

Fig 2. Work flow of green Synthesis Of NPs using orange peel

2.2 MATERIALS CHARACTERIZATION:

2.2.1. X-Ray Diffraction (XRD) Analysis

X – ray diffraction works by irradiating a material with incident x – rays and then measuring the intensities and scattering angles of the x – rays that leave the material.

2.2.2. FTIR SPECTROSCOPY:

Fourier Transform Infrared Spectroscopy analysis method uses infrared light to scan test samples and observe chemical properties. The resulting signal at the detector presents as a spectrum, typically from 4000cm-1 to 400cm-1 representing a molecular fingerprint of the sample.

2.2.3. UV - VISIBLE SPECTROSCOPY:

UV - visible spectrometer used for confirming the synthesized zinc oxide nanoparticles. The UV - visible spectra result revealing a strong absorbance between 360 – 380 nanometers suggesting the formation of zinc oxide nanoparticles.

2.2.4. SEM - EDAX SPECTROSCOPY:

Scanning electron microscopy with energy dispersive X - ray spectroscopy is used to surface analytical techniques. The range of these analysis is x5,500 , x10,000, x20,000 is resulting the analysis.

3. RESULT AND DISCUSSION

3.1. X-Ray Diffraction (XRD) Analysis

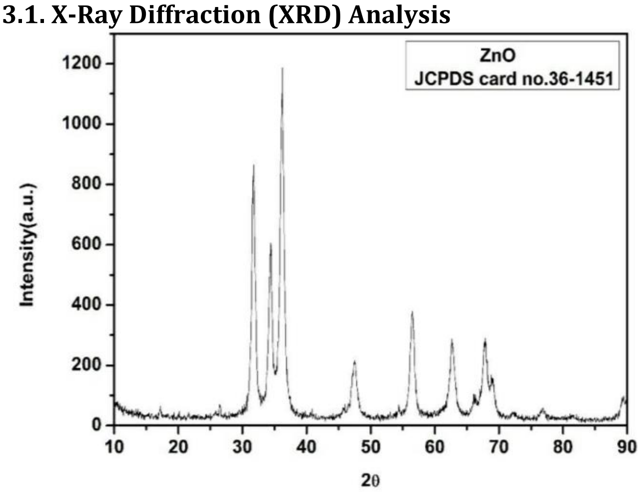

Fig 1. XRD graph of ZnO nanoparticle

The XRD sample exhibited preferred diffraction peaks at 2θ = 36.21^0 with corresponding to the lattice plane. There are agreed well with the standard card of Zinc oxide powder sample (JCPDS file No: 36 - 1451). The Zinc oxide nanoparticles exhibited a hexagonal structure. The temperature of the ZnO nanoparticles has increased, as has the strength of the diffraction peaks and the crystalline size. For this sample, the smallest average crystalline size was found to be around 12nm. The peaks appeared at 2 value ranging in fig 3. from 33.74, 35.38, 37.22, 46.50, 55.56 and 57.81 degree values are corresponding to pure Zinc oxide.

3.2 SEM- EDAX Analysis

SEM analysis is done to visible the shape and size of nanoparticles. Scanning electron microscope was used to determine the shape of Orange fruit peel of ZnO nanoparticles. Fig 4. shows SEM image were seen in different magnification ranges like 2μm – 20kV which clearly demonstrated the presence of spherical shaped nanoparticle with mean the average diameter. The presence of metallic Zinc oxide in the synthesis of ZnO nanoparticles was confirmed by EDAX analysis.

3.3 Analysis of UV Spectroscopy:

The band at 290 and 570 cm_{-}^{1} are due to stretching vibrations of Zn-O bond in ZnO NPs. Two bands observed at 379 and 409 cm_{-}^{1} are attributed to the metal oxygen, stretching vibrations of ZnO NPs. The green approach for the formation of Zinc oxide nanoparticles using Orange fruit peel extract studied. The entire reaction mixture is turned to light yellow color and exhibit an absorbance peak around 379 nm characteristic of Zinc Oxide nanoparticles due to its surface plasma resonance absorption.

3.4 FT-IR Spectroscopy Analysis

Fourier transform infrared spectroscopy is used to identify the possible biomolecules in the Orange fruit peel extract. The result of the FTIR spectrum of hot and gold methods of Orange fruit peel extracts of Zinc oxide nanoparticles are possess in the FTIR image. The FTIR peaks are obtained at 3434.18, 1430.34, 1111.60, 884.28, 842.28, 521.07 and finally 422.24 corresponded to C=C, C=O, C-N, C-H Stretching and bending respectively.

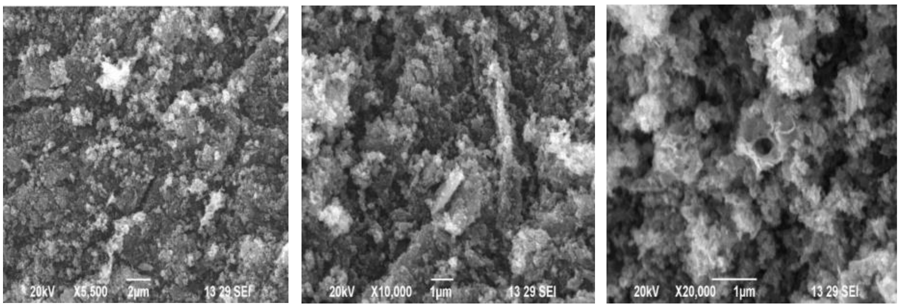

Fig 4. SEM-EDAX images of ZnO nanoparticles.

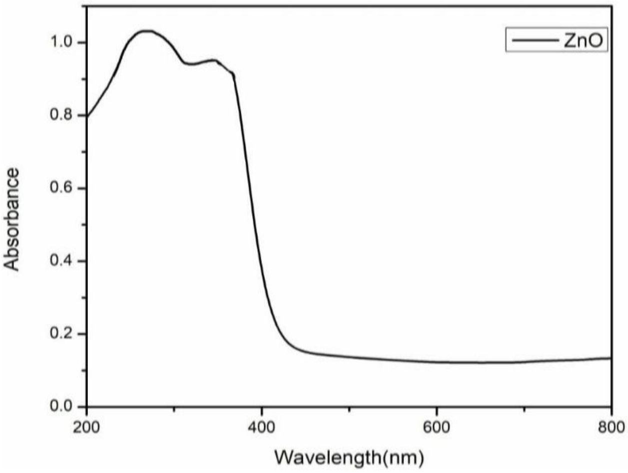

Fig 5. UV-Vis image of ZnO nanoparticles

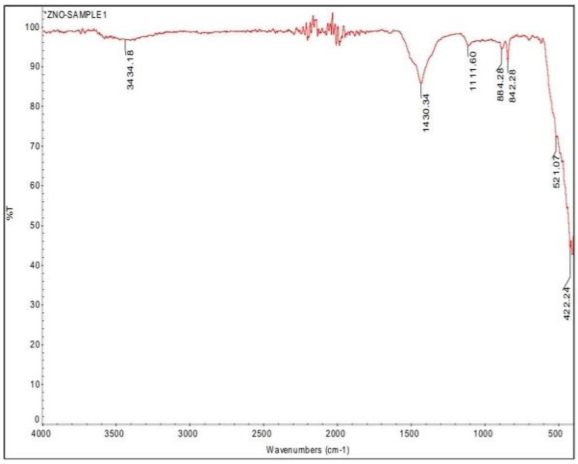

Fig 6. IR Spectrum of ZnO nanoparticle

4.Antibacterial activity:

4.1. Antibacterial Activity of Orange peel of zinc oxide

The antibacterial activity of the Zinc oxide nanoparticles was tested against gram positive bacteria Streptococcus sp. and E. coli and the Gram negative bacteria Salmonella typhi. The results for the antibacterial activity of Zinc oxide nanoparticles was given in the table below. The results of the bacterial activity of the Zinc oxide nanoparticles towards Streptococcus species, Salmonella typhi and E.coli. All samples are performed excellently in sterilizing E.coli with bacterial rates 1000, 750 and 500 in 1mg/ml. The ZnO NPs sample synthesized with 1000 ml orange peel exhibit zone of inhibition against *Escherichia coli,* Streptococcus species, *and Salmonella typhii* were 12, 8, 7 and 29 mm.

The ZnO NPs sample synthesized with 750 ml orange peel exhibit zone of inhibition against *Escherichia coli,* Streptococcus sp, *and Salmonella typhii* were 8, 8, 7 and 13 mm.

Similarly, The ZnO NPs sample synthesized with 500 ml orange peel exhibit zone of inhibition against *Escherichia coli,* Streptococcus species, *and Salmonella typhii* were 8, 8, 7 and 12 mm.

The antibacterial activities of Zinc oxide nanoparticles are due to their small size and large surface area and it also involves in electrostatic interactions between the nanoparticles. Therefore by increasing the concentrations the antibacterial activity is highest in Gram positive compared to Gram negative bacteria. The darker zone in the plates represents the antibacterial activity against the bacteria. The Zinc oxide nanoparticles inhibit the growth of microorganism by permeating into the cell membrane. The highest antibiotic value is 29nm recorded against Streptococcus species. The average antibiotic value is 13nm recorded against Salmonella typhi. The lowest antibiotic value is 12nm recorded against E. coli.

Table 2. antibacterial activity of ZnO nanoparticles

Organisms	Zone of Inhibition (mm) Sample (1mg/ml)			Antibiotic (1mg/ml)
	1000	750	500	
Streptococcus sp.	12	8	7	29
Salmonella typhi	8	7	7	13
Escherichia coli	8	7	7	12

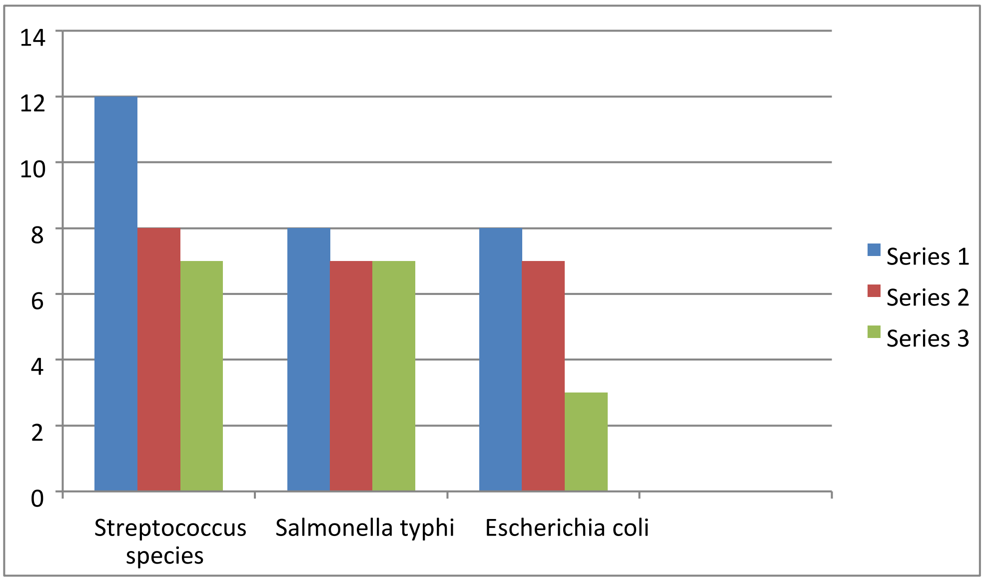

Fig 7. Graph of antibacterial activity of ZnO nanoparticles

Fig 5. Antibacterial activity of ZnO nanoparticles

5. Conclusion:

In this study, we successfully synthesized Zinc oxide nanoparticles by using a green synthesis method using orange fruit peel extract as the reducing agent. The metal nanoparticles have been produced chemically and physically for a long time. The biological synthesis of Zinc oxide nanoparticles using peel extract provides and environment friendly , simple and efficient route for synthesis of nanoparticles. The structure and size of the prepared Zinc oxide nanoparticles were characterization by XRD, SEM-EDAX, FTIR and UV-visible analysis. The XRD is present in 2θ value shows the synthesis of Zinc oxide nanoparticles. Then the SEM-EDAX analysis is shows the size of the Zinc oxide nanoparticles from 2-0.5μm. The FTIR spectra is shows that functional groups of stretching bands for Zinc oxide nanoparticles found in 3434-884cm^{-1}. And then finally the UV- visible spectra is analysis the presence of Zinc oxide by absorption peaks at 250-390nm. The synthesis of Zinc oxide nanoparticles was studied for antibacterial activities against bacteria like Streptococcus species, Salmonella typhi and E.coli. The green synthesis of Zinc oxide nanoparticles using fruit peel extract which has the potential to reduce the use of toxic chemicals and the production of nanoparticles.

Conflict of Interest

The authors have no affiliation with any organization with a direct or indirect financial interest in the subject matter discussed in the manuscript

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