INTERNATIONAL RESEARCH JOURNAL OF PHARMACY
www.irjponline.com
ISSN 2230 – 8407

Research Article

GREEN APPROACH FOR THE SYNTHESIS OF ZINC NANOPARTICLES AND ITS ANTIBACTERIAL ACTIVITY

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Article Received on: 01/05/16 Revised on: 02/06/16 Approved for publication: 07/06/16

DOI: 10.7897/2230-8407.07673

ABSTRACT

Green nanotechnology is an eco-friendly approach for the synthesis of metal nanoparticles and a promising alternative for chemical methods. In this study, we have synthesized zinc nanoparticles by utilizing the leaf extract of the plant *Cestrum nocturnum* as a reducing agent. The generated nanoparticles were characterized using Transmission Electron Microscopy (TEM) and Fourier transform infrared spectroscopy (FTIR). TEM analysis revealed spherical nanoparticles with an average size range of 10-30 nm. Bactericidal activity was tested against the bacteria *E. coli*.

KEYWORDS: Nanoparticles, Zinc, Antibacterial, Biosynthesis, TEM, FTIR

INTRODUCTION

In recent times, nanotechnology has been studied extensively due to its potential applications. The advent of nanotechnology has been an advantage to the physical and life sciences[1]. Nanotechnology is the utilization of 1-100 nm size nanomaterial that shows properties different than the bulk samples of the same material[2-4]. Therefore, metal nanomaterials have drawn the attention of researchers due to their potential applications; as compared to the bulk materials[5]. This has lead to commercial exploration of nanotechnology in the biological and medical sciences.

Numerous methods of nanoparticles synthesis have been developed and categorized as physical, chemical and biological[6]. However, most of the physical and chemical techniques of nanoparticle synthesis are expensive and require various hazardous chemicals, stabilizing agents and capping agents[7-8]. Thus, green nanotechnology has been projected as the eco-friendly substitute to these methods[9].

Recently, the use of microorganisms to synthesize nanoparticles has garnered interest, but cell culture maintenance is a tedious task due to continuous sub-culturing and risk of contamination[10]. Therefore, researchers have focused on the green route, due to ease of availability of plants in nature[11].

Nanoparticles are known to be versatile and have a variety of applications in modern biology, medicine and bio-detection of pathogens[12]. Biogenic nanoparticles have been shown to have antibacterial and antifungal properties[13-14]. Due to rising health-care costs and rise in incidents of infections and antibiotic resistance, there is an urgent need to develop new antimicrobial agents.

The current investigation focuses on the development of a simple method for the synthesis of zinc nanoparticles by utilizing plant leaf extract and study its bactericidal activity against *Escherichia coli*.

MATERIALS AND METHODS

Plant Material and Preparation of the Extract

Fresh leaves of the plant *Cestrum nocturnum* were obtained locally from Aurangabad, Maharashtra, India. *Cestrum nocturnum* leaves were used to make the aqueous extract. 25 gm of freshly obtained leaves were cut into small pieces and grounded using a mortar pestle, then homogenized with 100 ml of double-distilled water and filtered through Whatman No.1 filter paper. The filtered aqueous extract use as reducing agent.

Synthesis of Zinc Nanoparticles

For the synthesis of nanoparticles, 10 ml of filtered plant extract was added to 10 ml of 1 mM $ZnSO_4$. The test tube containing the reaction mixture was kept at 80°C-90°C for 20 minutes in a boiling water bath and then incubated at 35°C-37°C in an incubator for 24 hours. The reaction mixture was centrifuged at 3,000 rpm for 5 minutes and used for characterization.

TEM Analysis

The TEM analysis was done using PHILIPS- Model No-CM200 instrument at SAIF-IIT, Bombay. TEM analysis was used to confirm the synthesis of nanoparticles and understand their size and shape. Prior to the sample preparation for TEM analysis, sonication of sample was done for 10 min.

FTIR Analysis

Fourier Transform Infrared Spectroscopy was performed to identify the presence of biomolecules that may have played a role in the synthesis of the nanoparticles. The spectrum was recorded on Bruker Alpha E FTIR spectrophotometer at

Department of Chemical Technology, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad, Maharashtra, India.

Study of Antibacterial Activity

Antibacterial activity of the generated nanoparticles was performed against *E.* coli. The well diffusion method was employed to evaluate the antibacterial activity of the zinc nanoparticles. 25 µl, 50 µl, 75 µl and 100 µl of nanoparticle solution were added to the wells and plant extract was used as control, then incubated at 37°C for 24 hours. The diameters of the zone of inhibition were measured in millimeters.

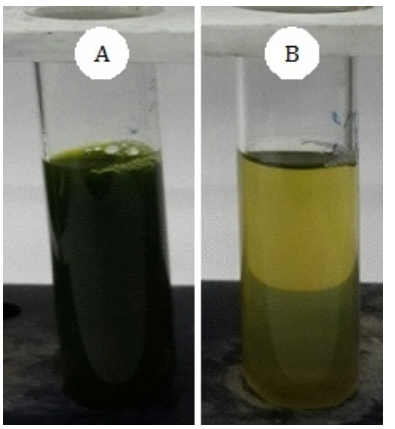

Figure 1: Visual observation after synthesis of nanoparticles: A) Plant extract B) Reaction mixture of zinc nanoparticles showed colour change

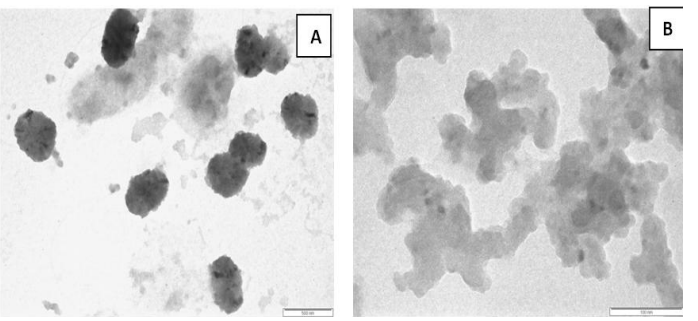

Figure 2: TEM analysis of generated zinc nanoparticles A) At 500 nm and B) At 100 nm

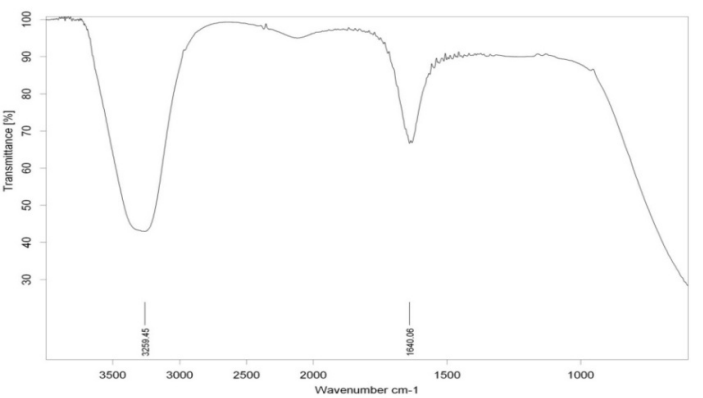

Figure 3: FTIR spectra of zinc nanoparticles

Table 1: Antibacterial activity of zinc nanoparticles

Microorganism	Zone of inhibition (mm)				
	Control	25 µl	50 µl	75 µl	100 µl
E. coli	-	25	28	30	33

RESULTS AND DISCUSSION

Various biological methods have been employed for the synthesis of nanoparticles. Gunalan synthesized zinc oxide nanoparticles of size 25-40 nm using zinc nitrate as a precursor and *Aloe vera* leaf broth for the reduction[15]. Banumathi *et al.* synthesized zinc oxide nanoparticles using the leaf extract of *Lobelia leschenaultiana* and zinc acetate as precursor[16]. Similarly, in the present investigation, zinc nanoparticles were produced using the leaf extract of *Cestrum nocturnum* as a reducing agent and zinc sulphate as precursor.

Visual Observation

10 ml precursor solution of 1 mM $ZnSO_4$ was added to 10 ml of leaf extract, colour change was observed in the reaction mixture after a period of incubation with respect to the control (Figure 1). The qualitative analysis of zinc nanoparticles was carried out based on the visual observation of color formation.

TEM Analysis

Transmission Electron Microscopy was utilized for understanding actual size and shape of the nanoparticles. Figure 2 shows TEM micrograph of the generated nanoparticles at different magnifications.

It was observed that there is variation in particle sizes, ranging from 10 nm to 30 nm. The synthesized Zn nanoparticles were spherical in shape (Figure 2). This provided conclusive proof that leaf extract of *Cestrum nocturnum* plant has the ability to produce zinc nanoparticles.

FTIR Analysis

Figure 3 showing FTIR Spectra of aqueous zinc nanoparticles synthesized from *Cestrum nocturnum* leaf extract was carried out to identify the possible biomolecules responsible for capping and stabilization of nanoparticles. Absorbance bands were observed at 3259.45 cm^{-1} and 1640.06 cm^{-1} which suggests that these biomolecules may have played a role in the nanoparticle synthesis. The peak at 3259.45 cm^{-1} might be alcohol O-H stretch; peak is broad due to H-bonding.

Study of Antibacterial Activity

In this study, antimicrobial assay was performed to determine the biological activity of the synthesized zinc nanoparticles. The generated zinc nanoparticles were tested against Gram-negative bacteria *E. coli*. The zones of inhibition around each well are showed in Table 1. From the results it was observed that the generated zinc nanoparticles have efficient antibacterial activity against *E. coli*.

The largest zone of inhibition was measured at 33 mm for 100 µl of zinc nanoparticle solution. It was found that the diameter of the clear zones were proportional to the volume of the nanoparticle solution.

CONCLUSION

We have successfully developed a simple and eco-friendly method for the synthesis of zinc nanoparticles using *Cestrum nocturnum* leaf extract. The biosynthesized nanoparticles were characterized using Transmission Electron Microscopy (TEM), which revealed the presence of spherical nanoparticles. FTIR analysis revealed the presence of biomolecules that may have played a part in the formation of zinc nanoparticles. The nanoparticles showed antibacterial activity against *E. coli*. The synthesized zinc nanoparticles might be applicable as a potential antibacterial agent.

ACKNOWLEDGEMENT

Authors are thankful to SAIF-IIT, Bombay, for providing facilities to analyze the nanoparticles by Transmission Electron Microscopy, and Department of Chemical Technology, Dr. Babasaheb Ambedkar Marathwada University, for providing the facility of Fourier Transform Infrared Spectroscopy (FTIR).

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Cite this article as:

Nilesh Paul, Asifuddin Syed, Parth Vyawahare, Ravindra Dakle, Balaji Ghuge. Green approach for the synthesis of zinc nanoparticles and its antibacterial activity. Int. Res. J. Pharm. 2016;7(6):99-102 http://dx.doi.org/10.7897/2230-8407.07673

Source of support: Nil, Conflict of interest: None Declared