

Green synthesis of ferric nano particles using *Justicia adhatoda* leaf extract

And their antibacterial activities

J. Thomas Joseph Prakash*, D. Devi Priya

PG And Research Department of Physics, Government Arts College

(Affiliated to the Bharathidasan University), Trichy-620 022, Tamil Nadu, India.

Abstract

Bio synthesis of iron nanoparticles has gathered an conclusive interest over the last few years to their distinctive properties, applicable in discrete field of science and technology. In the present study, synthesis of iron nanoparticles using *Justicia adhatoda* leaf extract is performed. The successful formation of iron nanoparticles has been confirmed by X-ray diffraction, UV-visible Spectroscopy, Fourier Transform Infrared Spectroscopy, Field Emission Scanning Electron Microscopy and Energy Dispersive X-ray Spectroscopy analysis. UV Visible Absorption Spectroscopy was used to characterize the synthesized ferric nanoparticles. At the range of 262 nm. UV-Visible spectra showed a characteristic absorption peak of ferric nanoparticles. FTIR spectra has presents functional groups. The XRD spectrum displayed three different diffraction peaks corresponding to the crystal plane of crystalline ferric nanoparticles whereas the crystallinity and purity of ferric nanoparticles were indicated by sharp peaks. The FESEM and EDAX study has confirmed. The prepared nanoparticles have

been tested for antibacterial activity for *Pseudomonas aeruginosa*.

Keywords: *Justicia adhatoda*, Iron Nanoparticles, UV, FTIR, Antibacterial activity.

* Corresponding Author

J. Thomas Joseph Prakash

PG And Research Department of Physics,
Government Arts College

(Affiliated to the Bharathidasan University),
Trichy-620 022, Tamil Nadu, India

E-Mail: armyjpr1@yahoo.co.in

Cell: 09842470521.

1. Introduction

Biosynthesis of metal nanoparticles from plant systems is an emerging as a novel and recent development technique. The nanoparticles are of extensive interest suitable to their extremely little size and vast surface to volume ratio, and they exhibited altogether new characteristics compared to the vast particles of bulk material. There is increasing in commercial demand for nanoparticles suitable to their broad applicability in discrete areas such as energy,

electronics, catalysis, chemistry and medicine. Recently, an grand research has been focussed on nano-structured magnetite because it possess differential magnetic and electric properties and its application in medical treatment [1-8].

The main concentration nowadays is to develop economical and environmentally clean synthesis methods to synthesize iron nanoparticles. Numerous eco-friendly materials have been used for synthesis of iron nanoparticles like plant extracts [9] etc for the same. These eco-friendly techniques do not require high temperature, pressure or the use of toxic chemicals. In the present research paper, such an eco-friendly synthesis method for iron nano particles has been reported using the solutions of FeCl₃.

These materials have a substantiate potential to be applied as sensors, in catalysis [10], as high-density magnetic recording media [11], for targeted drug delivery in clinical trials [12], and as substrates in cancer treatment methods [13]. Iron chlorides are relatively passive, non-toxic, and present in living organisms. Iron oxide nanoparticles play an important quota in environmental remediation circles. As it eliminate both of organic and inorganic heavy metal pollutants from polluted water [14]. There are several chemical and physical methods available for synthesis of iron oxide nanoparticles. Those methods are used toxic or

potentially hazardous as starting materials and more energy.

The Iron Nanoparticles are search to be having distinctive magnetic properties and superior biocompatibility. In this study, The Iron Nanoparticles were synthesized using Justicia adhatoda Leaves extract and the antibacterial activity was studied against human pathogen such as *Pseudomonas aeruginosa* which can cause various diseases such as anemia, urinary tract infections, diarrhea etc, although most of the *Pseudomonas aeruginosa* strains are harmless.

2. EXPERIMENTAL METHOD

2.1 Collection of Plant Material

Justicia adhatoda leaves were collected from, Trichy District, Tamil Nadu, India.



Figure.1 Justicia adhatoda Image

2.2 Chemicals

Analytical grade of Ferric chloride were procured from Sigma Aldrich chemicals.

2.3 Extraction Preparation

The *Justicia adhatoda* (Tamil name Aadathoda) leaves were washed and stored. For the production of extract, ground, air-dried *Justicia adhatoda* samples (leaf) about 5 gram were boiled with double distilled water (100 ml) in an Erlenmeyer flask (Fig. 1) while being continuously stirred for 15 min. The extract was cooled to room temperature after that filtered, and stored at $-4\text{ }^{\circ}\text{C}$ for further use.

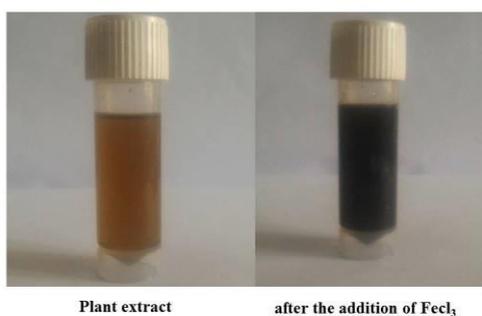


Figure.2

3. Characterization

The prepared sample has to go under a number of characterization techniques to investigate its various properties. The results of the sample give the information about the different optical and structural properties. The synthesized nanoparticles were further analyzed with the help of ultra violet-visible spectroscopy in 200 nm to 1000 nm wave length range.

Obviously Iron nanoparticles has wide range of application in the field of optoelectronics devices. On putting the sample in the following characterization techniques, the information related to optical properties can be obtained [15-19].

- UV-Visible spectroscopy (UV) UV-VISIBLE SPECTROPHOTOMETER LAMBDA 35 PERKIN ELME which ranges between 190 nm-1100 nm.
- Fourier transform infrared spectroscopy (FTIR SPECTRUM 1000 PERKIN ELMER SPECTROMETER) of the supernatant and synthesized iron nanoparticles was recorded using KBr pellets and the spectrum was collected at a resolution of 4cm^{-1} in wavenumber region of $400\text{-}4000\text{ cm}^{-1}$.
- Powder X-ray diffraction (XRD) was recorded by XRD “X” PERT PRO Diffractometer.

In order to get relevant information about the surface morphology, particle size etc. the below mentioned characterization techniques are applicable

- Field emission Scanning Electron Microscope and Energy dispersive X-ray spectroscopy using the model were performed on FEI QUANTA-250 FEG equipped with an EDAX instrument.

3.1 Antibacterial Activity test

The purpose of this study was to examine the antibacterial activity of the sample toward selected pathogens using disc diffusion method.

Collection of test organisms:

To examine the antibacterial activity of samples, One gram negative bacterial strains *Pseudomonas aeruginosa* (MTCC 27853), were prepared as test organisms. All the strains were procured from the Microbial Type Culture and Collection (MTCC) at Chandigarh, India. Bacterial strains were cultivated at 37°C and maintained on nutrient agar (Difco, USA) slant at for 4°C.

Antibacterial activity of samples was determined using the disc diffusion method. The petridishes (diameter 60 mm) was prepared with Muller Hinton Agar and inoculated with test organisms. Sterile disc of six millimeter width were impregnated with 10 µl of various samples respectively. Prepared discs were placed onto the top layer of the agar plates and left for 30 minute at room temperature for compound diffusion. Positive control was prepared using the 10 µl of Amoxicillin as standard antibiotic disc. The dishes were incubated for 24 h at 37°C and the zone of inhibition was recorded in millimeters and the experiment was repeated twice.

4. Results and Discussion

4.1 UV-Visible Spectroscopy

Synthesized FeNPs using UV-visible spectroscopy were studied and the recorded

spectra are shown in Figure 3. The absorption behavior arises due to surface Plasmon resonance (SPR) [20]. As the *Justicia adhatoda* leaf extract was added to the aqueous FeCl₃ solution, the color of the solution changed from pale yellow to black color indicating FeNPs formation. The UV-Vis spectra of the synthesized FeNPs which gives a broad absorption band at around 262 nm. [21]

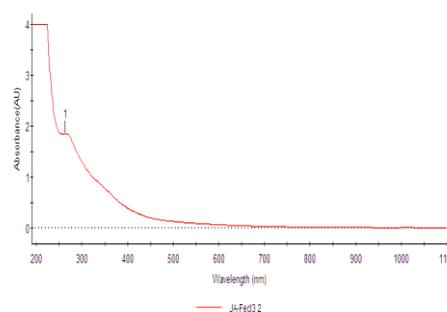


Figure.3 UV-Visible Spectroscopy

4.2 Fourier Transform Infrared Spectroscopy

FTIR analysis was carried out to identify the possible interaction between the biomolecules and Fe³⁺ during the biogenic reduction reactions. The FTIR data for FeNPs containing *Justicia adhatoda* leaf extract is shown in Figure 4. The band at 3422 cm⁻¹ is assigned for O-H stretching vibration of alcohol and phenol compounds and bands observed at 2922 cm⁻¹, 2852 cm⁻¹, 2560 cm⁻¹, 1629 cm⁻¹, 1421 cm⁻¹, 1118 cm⁻¹, 777 cm⁻¹, 625 cm⁻¹ are due to the C-O stretching, C=O stretching mode of the carbonyl functional groups in alcohol, ethers, acids, and esters. The carbonyl band at 1421 cm⁻¹ was shifted to 1690 cm⁻¹ during the formation of

FeNPs. The shift in bands at 1629cm^{-1} was clearly indicating the coordination of carboxylic acid with FeNPs and 777 cm^{-1} is attributed to zero valent iron, Fe⁰ as reported in the literature. From the analysis of FTIR study, we revealed that the carbonyl group from the amino acid residue, carbohydrates, and phytochemical constituents has the stronger ability to bind metal NPs (capping of FeNPs) to prevent the agglomeration and thereby stabilize the medium. This suggests that biological molecules could possibly perform the dual function of formation and stabilization of FeNPs in the aqueous medium. Water soluble heterocyclic compounds such as flavonoids, alkaloids were mainly responsible for the reduction and stabilization of NPs. This results implied that tannins, spanin, flavonoids, steroids, carbohydrates, polyphenol, glycosides present in *Justicia adhatoda* leaf extract play a major role in the reduction of Fe³⁺ [22]

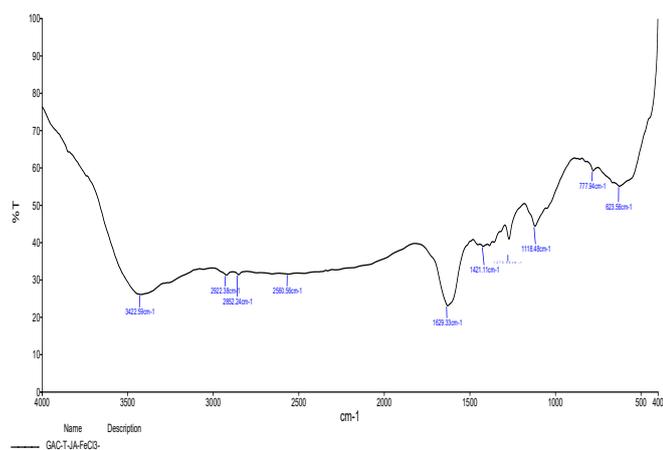


Figure.4 Fourier Transform Infrared Spectroscopy

4.3 X-Ray Diffraction

Phase purity and crystallinity of the synthesized Ferric chloride NPs can be identified XRD analysis. The XRD patterns synthesized Ferric chloride -NPs are shown in Fig. 5. A broad diffraction peak was observed. All peaks in this pattern were found to be in good agreement with JCPDS NO: 65-4899. The diffraction peaks of synthesized Ferric Chloride-NPs were detected at $2\theta=30.4^\circ, 35.8^\circ, 43.5^\circ$, which are assigned to the crystal planes of (200), (311), (511) respectively. The analyzed diffraction peaks were matched well with the standard magnetite XRD patterns declared the crystallographic system of cubic structure [23].

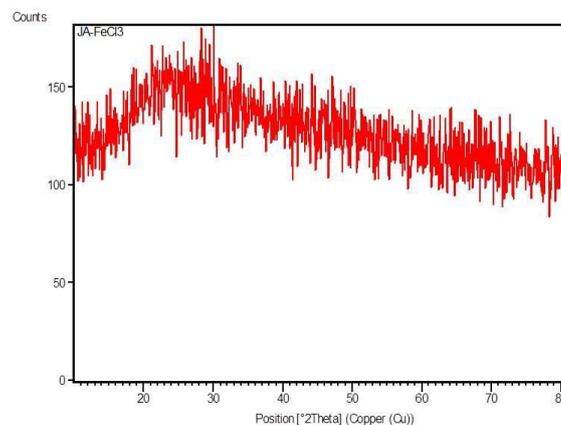


Figure.5 X-Ray Diffraction

4.4 Field Emission Scanning Electron Microscopy and Energy dispersive X-ray spectroscopy

The Field Emission Scanning Electron Microscopy can be employed to characterization the size (40 to 70 nm), shape and morphologies of formed iron nanoparticles. The FESEM images for samples are shown in Fig.6 (a)

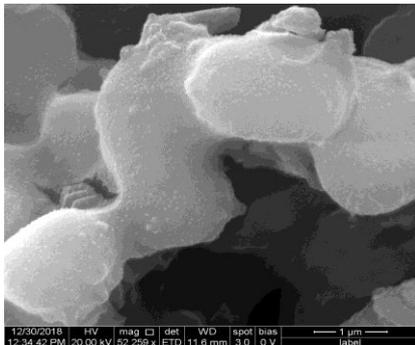


Figure.6 (a) Field Emission Scanning Electron Microscopy

EDAX analysis has confirmed zero-valent which is present in the sample [24]. The spectrum represent a distinctive peak at 6.4 KeV due to ferric nanoparticles as shown in Fig.6 (b).

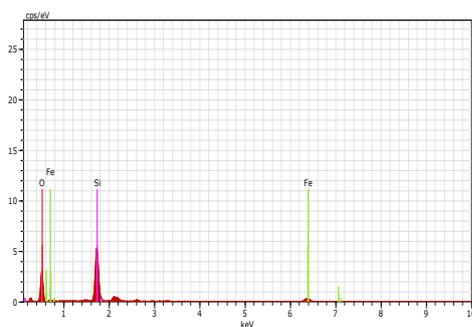


Figure.6 (b) EDAX

4.6 Screening of Antibacterial Activities

Antibacterial activity of samples(disc diffusion method)

The results of the antibacterial activity of various samples were tested against pathogens

by disk diffusion method. The Sample D showed growth inhibitory activity against *Pseudomonas aeruginosa* (5 mm). At sample C exhibited the antibacterial activity all the four bacteria, but was more susceptible against *Pseudomonas aeruginosa* (4 mm). However, the crude extract and synthesized nanoparticles showed better inhibitory actions against pathogens as shown in Fig.7

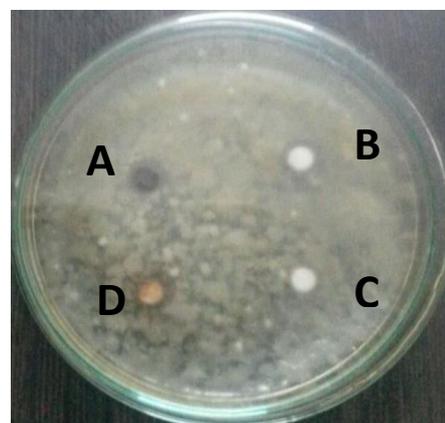


Figure.7 *Pseudomonas aeruginosa*

CONCLUSION

Biosynthesis of iron nanoparticles using *Justicia adhatoda* leaf was carried out and the results are interpreted using different analytical instruments like UV-spectrophotometer which indicate the formation of iron nanoparticles at 262 nm, followed by FTIR that was used in identifying the functional groups present in synthesized nanoparticles and FESEM images showed the relative size of the synthesized nanoparticles in the range of 40nm-70 nm and

EDAX was confirmed the presence of absorption peak at 6.4 KeV. XRD analysis was confirmed. Iron nanoparticles showed good antibacterial activity against *Pseudomonas aeruginosa*.

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