Biological synthesis of iron oxide nanoparticles using *Streptomyces sp.* and its antibacterial activity

Jositta Sherine, Annie Sujatha, Maheshwaran Rathinam*
Department of Physics and Nanotechnology, SRM University, Kattankulathur – 603 203, India.

*Corresponding author: E-Mail: maheshwaranrathinam@gmail.com

ABSTRACT

This study proposed to synthesis a metal oxide nanoparticle via biological route. The *Streptomyces sp.* is used as a bio-redetection of nanoparticles from iron precursor. Herein we investigate the antibacterial potentials of the biosynthesized nanomaterials. The UV-visible spectra confirms the respective absorbance and particle size. The FTIR analysis confirmed that the biological molecules are responsible for the reduction of nanoparticles. The investigated XRD studies inveterate the crystal antic structure of the particles. The FESEM analysis established that the particles are in spherical shape. The investigated antibacterial studies confirmed that the particles are well suitable for anticancer agent.

KEY WORDS: Iron Oxide, Nanoparticles, Biosynthesis, Antibacterial activity.

1. INTRODUCTION

Nanotechnology refers to an evolving area of science and technology that includes synthesis and development of various nanomaterials. Nanoparticles have vast applications for instance drug delivery, therapeutic products, etc., the development of biologically enthused new processes for the syntheses of nanoparticles is evolving into an important branch of nanotechnology. Living microorganisms, especially *Streptomyces sp.* have a remarkable ability to form exquisite inorganic structures often in nano-dimensions. The development of these eco-friendly methods for the fabrication of nanoparticles is developing into an important division of nanotechnology especially iron oxide nanoparticles. The main impartial of this work is to assess the anti-bacterial effects of iron nanoparticles against various strains. The present study was intended by using *Streptomyces sp.* To synthesis of Fe$_2$O$_3$ nanoparticles and to screen the antibacterial activity.

2. MATERIALS AND METHOD

**Isolation of *Streptomyces* from soil sample:** Isolation of *Streptomyces sp.* can be done by the serial dilution and spread plate technique. One gram of soil sample was suspended in 10ml of sterile double distilled water. The dilution was done out up to 10-5 dilution. 0.1ml of aliquots from 10-2, 10-3, 10-4 and 10-5 were spread on the yeast-malt extract agar medium. Nalidixic acid 100mg/l and cyclohexane 20mg/l were added to minimize the bacterial and fungal contamination. The plates were incubated at 30ºc for 7 days. The suspected colonies of *Streptomyces sp.* were isolated and purified using ISP2 medium.

**Culture Characterization:** The culture characterization of the organism was observed by inoculating the potential isolate in the yeast malt extract medium and incubate at 28ºc for 7 day. After incubation, the culture characteristics of the organism such as aerial and substrate mycelium were observed.

**Nanoparticles Synthesis:** Fresh and clean bacterial cells were taken and it was centrifuged at 12,000 rpm for 10mins, after centrifugation, supernatant were collected. Then 1mM of FeCl$_2$ was taken and dissolved in 50ml of double distilled water. This was kept in a magnetic stirrer to mix thoroughly. After mixing, 1ml of bacterial culture was added in a drop wise manner and it was incubated for 48 hours at 30ºC. After incubation, it was centrifuged at 12,000 rpm for 10 minutes. Then they were washed with distilled water and acetone for several times to remove the impurities. Purified nanoparticles were dried in vacuum oven at 60ºC, followed by annealing at 150ºC for 2 hours.

**Characterization Techniques:** The absorbance and thermal stability were done by (Schimadzu UV–visible spectrophotometer, model UV-1800). The crystalline structure was investigated using XRD-analysis (PAN alytical Xpert PRO diffractometer). The functional groups were identified by Fourier Transform Infrared Spectroscopy (MIDAC 2000M Series). Surface morphology along with chemical composition was determined by Hitachi S3400 SEM (Scanning Electron Microscope) which has accelerating voltage of 15.0 kV.

**Antimicrobial activity:** With the intention of testing the antimicrobial activity of FeO$_2$ nanoparticles, nutrient agar media was prepared and it was autoclaved. Thereafter, it was poured into petriplates. After solidification of the media, the wells of 5mm diameter were prepared with the help of sterilized steel cork borer then 50 µl of fresh overnight culture of gram negative organisms such as *E.coli, Pseudomonas auroginous* and gram positive organisms like *mycobacterium tuberculosis, streptococcus aurous* were swabbed separately over the media in the petriplates. Incubate the petriplates at 37ºC for 24 hrs. After incubation, different concentrations such as 50µl, 100µl and 150µl of the synthesised nanoparticles were added separately into the wells of the petriplates and they were incubated at 37ºC for 24 hrs. The zone of inhibition was noted after 24 hrs of incubation.
3. RESULT AND DISCUSSION

The synthesized FeO$_2$ nanoparticles were preliminary confirmed by Ultra violet visible spectroscopy (Fig.1). The UV absorbance was ranged from 200-800nm. The absorbance peak at 559 nm represents the formation Fe$_2$O$_3$ nanoparticles.

**Figure.1. Absorbance spectrum of biosynthesised iron oxide nanoparticles**

FTIR is one of the widely utilized tool for the detection of functional groups in pure compounds and mixtures and for compound comparison. Infrared analysis is connected to the vibrational motion of atoms and presented molecules. The peak (Fig.2) at 1632cm$^{-1}$ and 3443cm$^{-1}$ corresponds to the vibrational mode of -C= C- and O-H groups respectively. The –C=C- stretch was allotted alkene function group and N-H stretch was dispensed the primary amine group. These may perform as a reducing agent for the Fe$_2$O$_3$ nanoparticles.

**Figure.2. FTIR analysis for biosynthesised iron oxide nanoparticles**

The phase and the crystallographic characterises (Fig.3) of biosynthesized nanoparticles were characterized by XRD patterns. The XRD peaks at 23°, 31°, 37°, 43°, 63°, 76° with lattice plan (111), (220), (311), (400), (422) and (440) were well matched with the JCPDS No. 653107 and confirms that the particles were in Face centred cubic system. The crystalline size was calculated using Debye-Scherrer’s formula and found that at 39nm.

**Figure.3. Crystalline analysis for biosynthesised iron oxide nanoparticles**

FESEM analysis (Fig.4) is employed to visualize the size and morphology of the synthesized Fe$_2$O$_3$ Nanoparticles. It is observed that the biosynthesised Fe$_2$O$_3$ nanoparticles were spherical in shape with smooth surface. Agglomeration is caused due to the non-addition of capping agent, so that the particles are homogeneously distributed without much of agglomeration and ensured the average size ranging between 200-500 nm in diameter.

**Figure.4. FESEM image of FeO$_2$ nanoparticles**

X-ray fluorescence is a non-destructive analytical method used to fine the elemental composition of materials. Fig.5, showed that the Fe particles have higher intensity peak and oxygen was presented in low intensity level. The particles has higher purity which was confirmed by the calculation of mass of the particles. In the study the Fe have 89.95% of mass and O have 10.05% of mass.
The antibacterial activity (Fig.6) of the biosynthesised Fe₂O₃ nanoparticles were tested against four strains i.e. Mycobacterium tuberculosis, Streptococcus aureus were gram positive and E.coli, Pseudomonas aeruginosa were gram negative strains. Observations showed that the nanoparticles are more active against Pseudomonas aeruginosa. Various concentration such as 50µl, 100µl, and 150µl of synthesised nanoparticles were performed against all the selected isolates to find the greater zone of inhibition. The synthesised Fe₂O₃ nanoparticle was also tested against the selected strains. The zone of shyness pragmatic for Fe₂O₃ against Mycobacterium tuberculosis was 2mm at 50µl, 2.6mm at 100µl and 3.5mm for 150µl concentration. For E.coli 1.5mm at 50µl, 2mm at 100µl, 2.6mm at 150µl concentration, then for Pseudomonas aeruginosa 2mm, 3.5mm, 4.2mm inhibition zone was empirical at 50µl, 100µl, 150µl concentration and for Streptococcus aureus 1.2mm, 2.5mm and 3mm inhibition zone was observed at 50µl, 100µl, 150µl concentration.

4. CONCLUSION

Streptomyces sp. was used for the production of Fe₂O₃ nanoparticles in aerobic culture in limited specification is an advantage and the strain was able to tolerate higher level of dosage. This is an eco-friendly approach and simple step for the biosynthesis of nanoparticles. The biosynthesised nanoparticles were characterized by UV spec., XRD, FTIR, FESEM and XRF techniques. The antibacterial activity of the magnetically responsive Fe₂O₃ nanoparticles was also investigated. The use of such nanoparticles for antibacterial activity against virulent pathogens may implicate higher advantage to reduce the contamination in environment.

REFERENCES


