CHARACTERIZATION OF SILVER NANOPARTICLES BIOSYNTHESIZED USING FICUS RELIGIOSA PLANT LEAF EXTRACT

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Abstract - Silver nanoparticles were widely used in various fields including pharmaceutical, especially those synthesized from herbal medicinal plants due to their pharmacological importance. Ficus religiosa commonly known as pepal tree belonging to the family Moraceae, is used traditionally as antiulcer, anti-bacterial, anti-diabetic and anti-cancer. In this study the silver nanoparticles were synthesized using Ficus religiosa plant leaf extract and characterized by UV-visible spectroscopy, XRD analysis, SEM analysis and FTIR spectroscopy analysis. The UV-visible spectroscopy showed the peak at 424 nm which confirmed the synthesis of silver nanoparticles. XRD confirmed the crystalline nature of the silver nanoparticles. FTIR confirms the presence of alcohol, alkene, alkyne and alkyl Halide functional groups in the sample and the SEM analysis revealed the morphology of the synthesized silver nanoparticles as spherical.

Key Words: Silver nanoparticles, Ficus Religiosa, UV-Visible spectroscopy, XRD, FTIR, SEM.

1. INTRODUCTION

Synthesis of nanoparticles are evolving into an important branch of nanotechnology [1-2] due to their diverse properties like catalysis, magnetic and optical polarity, electrical conductivity, antimicrobial activity and surface enhanced Raman scattering (SERS) [3-6]. Silver nanoparticles are synthesized by chemical reduction of silver ions in aqueous solutions with or without stabilizing agents, thermal decomposition in organic solvents, chemical reduction and photoreduction have been reported [7-10]. These methods are expensive and involve toxic hazardous chemicals which are potential risk for environment.

Synthesis of silver nanoparticles using plant extract is rapid, low cost, eco-friendly and a single-step method for biosynthesis process and safe for human therapeutic use [11-12]. Many reports are available on the biogenesis of silver nanoparticles using several plant extracts like Azadirachta indica, Pelargonium graveolens, Medicago sativa and Emblica officinalis in India. In the present study, Ficus religiosa, one of the most sacred trees of South Asia, to both Hindus and Buddhists commonly known as Peepul tree, Pipal tree and sacred fig was used to synthesize silver nanoparticles. The bark, leaves and fruits of the trees were traditionally used as antibacterial, antiprotozoal, antiviral, anti diarrhoeal, gonorrhea, cancer, inflammation, asthma, ulcers and various skin diseases [13-17].

2. MATERIALS AND METHODS

2.1. Collection of Plant Materials

Fresh leaves of Ficus religiosa were collected from Chennai, Tamil Nadu and authenticated by Plant anatomy research center, West Tambaram, Chennai, Tamil Nadu (Reg no : PARC/2018/3651).

e-ISSN: 2395-0056

p-ISSN: 2395-0072

2.2. Preparation of Plant Extract

The leaves of Ficus religiosa were thoroughly washed with tap water to remove debris and then rinsed with distilled water several times. The leaves were allowed to shade dry for 3-5 days. The dried samples were blended into fine powder and stored in containers. About 10 gms of sample powder was taken and added into 100 ml of methanol in the ratio of 1:10 (powder/solvent). The extract was placed in a shaker incubator where the temperature is maintained between 35°C to 50°C. After 12 hours of incubation, the extracted sample was collected and stored in refrigerator for further uses.

2.3. Preparation of 1 mM Silver Nitrate Solution

Silver nitrate of 1M stock solution was prepared by adding 1.7 g of AgNO₃ in 10 ml of double distilled water. Solution of 1 mM was prepared by taking 1 ml of 1 M solution and made up to 100 ml with 99 ml of double distilled water. This solution was stored for further use.

2.4. Synthesis of Silver Nanoparticles

Preparation of the silver nanoparticles was initiated from 1 mM silver nitrate solution. Plant extract of 10 ml was taken in a conical flask and 100 ml of 1 mM AgNO₃ was added drop wise with constant stirring at 50°C - 60°C. The color change was checked periodically and the beaker was incubated at room temperature. The color of plant extract changes from pale green to faint brown and then faint brown to dark brown indicating the formation of silver nanoparticles. Different concentrations of plant leaf extract and silver nitrate was added into each test tube and kept for incubation for 24 hours.

Volume: 05 Issue: 12 | Dec 2018 www.irjet.net

and Technology (IRJET) e-ISSN: 2395-0056 p-ISSN: 2395-0072

2.5. Characterization of Silver Nanoparticles

Several techniques were used to characterize silver nanoparticles synthesis. The optical absorbance of the sample was taken from 200 nm to 800 nm with Shimadzhu UV-visible spectrophotometer to investigate the reduction of silver nitrate by the leaf extract. The phase identification of crystalline material was analyzed by X-Ray diffraction (XRD) analysis. The size, shape and surface morphology of the nanoparticles were confirmed using scanning electron microscopy (SEM) operated at an accelerated voltage of 120 kV with magnification ranging from 20 X to approximately 30,000 X with spatial resolution of 50 nm to 100 nm. The emission and absorption of infrared spectra by silver nanoparticles were measured using Fourier Transform Infrared spectrometer (FTIR, Bruker spectrometer). The FTIR spectra were recorded from wave number 500 cm⁻¹ to 3500 cm⁻¹.

3. RESULT AND DISCUSSION

3.1. Methanolic Extraction and synthesis of silver nanoparticles

The powdered leaves of *Ficus religiosa* were added to the methanol in the ratio of 1:10 (1g of leaves in 10 ml of methanol). It was kept for 12 hrs in shaker incubation and the extract was stored in refrigerator at 4°C (Fig.1). The extract was added to silver nitrate solution and colour change was observed after 24 hrs. The Fig.2 shows the colour changes during incubation from pale yellow to dark brown due to surface plasma resonance (SPR) vibration indicating the synthesis of nanoparticles.



Fig-1: Methanolic Extraction of Ficus religiosa Leaves.



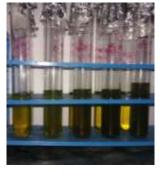


Fig-2: Biosynthesis of silver nanoparticles indicated by the colour change

3.2. Characterization of synthesized nanoparticles

3.2.1. UV- Visible Spectroscopy

Various concentration of silver nitrate 10, 20, 30, 40, 50 and $60 \mu l$ were taken and added to 4ml of the plant extract and made up with 6 ml of double distill water. The synthesized silver nanoparticles were initially detected and characterized using UV-visible spectroscopy. Concentration at 40 µl showed highest production of silver nanoparticles. As the concentration of the plant extract reduces, the synthesis of silver nanoparticles also reduces. The colour of the extract changed from green to brown colour. The metal nanoparticles have free electrons which yields surface plasmon resonance absorption band, due to the mutual vibration of electrons of metal nanoparticles in resonance with light wave. Analyses of the synthesized nanoparticles were carried out from 300 nm to 700 nm (Fig.3). The peak showed the characteristics feature of surface plasmon resonance of silver nanoparticles. The absorbance peak was obtained at 424 nm, indicating the presence of synthesized silver. Gradual decrease was seen in the absorbance of the spectra accompanied by a shift in the wavelength from 430 nm. It was also observed that the peak slowly shifted towards lower wavelength at high concentrations. The shift may be due to the particle size and shape [18]. Similar peak formation from 400 to 450 nm was reported in previous studies [19].

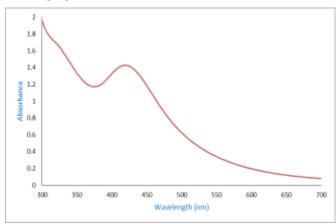


Fig-3: UV-Vis Spectroscopy Reading of silver nanoparticles

3.2.2. XRD

The crystalline nature of nanoparticles was confirmed by X-ray crystallography. The XRD pattern of the synthesized silver nanoparticles is shown in Fig 4. The diffracted intensities were recorded from 10° to 100° . Several distinct Bragg reflections were observed at 27° , 33° , 46° , 55° , 57° and 77° were observed. Three distinct characteristic peaks at 20° values of 33° , 46° and 77° corresponds to 111, 200 and 311 indicating the formation of the face centered cubic structure of silver nanoparticles. The results are similar to several studies that reported the cubic nature of biologically synthesized silver nanoparticles (20)

3.2.3. FTIR Spectroscopy

FTIR analyses were carried out to identify of the presence of various functional groups in biomolecules which are responsible for the bioreduction of Ag+ and stabilization of silver nanoparticles. The observed peaks were compared

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with the standard values to identify the functional groups. The peaks were observed at 3325.45, 2114.88, 1637.47, 584.34 and 523.76 cm⁻¹ as in Fig 5. The peak at 3325 cm⁻¹ corresponds to hydrogen bond (O-H) stretching vibration indicating the presence of alcohol group. The peak at 2114 cm⁻¹ corresponds to (-C=C-) stretching indicating the presence of alkyne group. The peak at 1637 cm⁻¹ corresponds to (C=C) stretching indicating the presence of alkene functional group. The bands at 584 and 523 cm⁻¹ corresponds to the (C-Br) stretching indicating the presence of alkyl halide functional group (21).

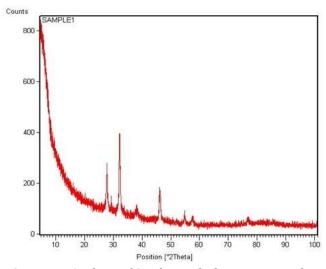


Fig-4: XRD Analysis of Synthesized silver nanoparticles

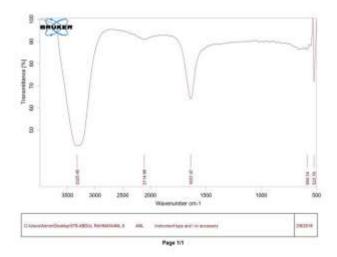


Fig-5: FTIR Analysis of Synthesized silver nanoparticles

3.2.4. SEM Analysis

The morphological studies of synthesized silver nanoparticles were carried out by SEM analysis. The size and shape of the synthesized silver nanoparticles were confirmed as spherical. The analysis showed clear spherical morphology of silver nanoparticles of size from 20 nm to 50 nm as shown in the Fig 6 and Fig 7. The organic compounds or reducing agents present in the extract may be responsible for variation in the shape and size of the nanoparticles, where the compounds interlink with nanoparticles and reduce them as reported in earlier findings (22).

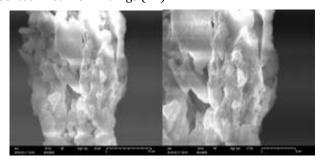


Fig-6: 10μm view of SEM

Fig-7: 5μm view of SEM

e-ISSN: 2395-0056

p-ISSN: 2395-0072

4. CONCLUSION

The biological synthesis of silver nanoparticles using *Ficus religiosa* leaf extract provides environmental friendly, simple and easy way to synthesis nanoparticles. The synthesized silver nanoparticles were characterized using UV-Visible spectroscopy, XRD analysis, FTIR spectroscopy and SEM analysis. The UV-Visible spectroscopy confirms the synthesis of silver nanoparticles by the peak obtained at 424 nm. X-ray diffraction method confirms the crystalline nature of the silver nanoparticles by showing four strong Bragg reflections at 27°, 33°, 46° and 55°. FTIR technique confirms the presence of alcohol, alkene, alkyne and alkyl Halide as the functional groups in the sample. SEM analysis shows the morphology of the synthesized silver nanoparticles as spherical. This work reveals that the Ficus religiosa plant leaf extract can be effectively used for the synthesis of silver nanoparticles.

ACKNOWLEDGEMENT

We thank the department of Biotechnology and the management of SRM Arts and Science College for giving us an opportunity to carry out the research work.

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e-ISSN: 2395-0056

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