Invitro Assessment of Antibacterial and Antioxidant Property of Biofabricated Silver Nanoparticles using Aqueous Extract of *Bauhinia Tomentosa*

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INTRODUCTION

Nanotechnology is the emerging technology in modern research field, which deals with design, manufacture and application of matter/ material by controlling its size and shape at atomic scale in 10⁻⁹ m (nm). Metal nanoparticles have attracted for biomedical application due to its reduced size and large surface area to volume ratio utilized in wound healing, bio imaging, drug delivery and disease treatment [1]. Silver nanoparticles have broad range of importance in various fields like medicine, textile and cosmetics [2]. Green approach to synthesise metal nanoparticles using microbes and plant gained significance such as simple, low cost, non toxic, ecofriendly and one step process compared to other conventional methods [3]. Use of plant extract to synthesise metal nanoparticles is rapid than the microbe mediated synthesis. Because, since the microbes are biohazardous to human kind. The presence of various bioactive components in the plant extract act to reduce the size of the metal particles to nanosize [4]. *Bauhinia tomentosa* is a vellow bell orchid plant widely used in folklore medicine to treat various diseases like diarrohea, leprosy, wound ulcers, skin disorders, dysentery and diabetes [5]. It belongs to Fabacea family, commonly known as Kanjana in tamil. The plant as

ABSTRACT

Biosynthesis of silver nanoparticles is an eye catching area of a modern research field. It is simple, single step, non toxic and eco-friendly method. In the present investigation, the aqueous extract of *Bauhinia tomentosa* is used to synthesise silver nanoparticles and it was confirmed by UV- Visible spectrometer. Due to surface Plasmon resonance, the peak obtained at 440.0 nm. The functional group present at the surface of the silver nanoparticles was confirmed by FT-IR analysis. Further, the synthesized silver nanoparticles were subjected to antibacterial activity by Well diffusion method and also antioxidant activity performed by hydrogen peroxide scavenging assay. As concentration of silver nanoparticles increases the bactericidal activity is raised and also the antioxidant activity is increased. Thus it reveals that due to presence of various bioactive compounds present in the plant extract acting as reducing agent to reduce the size of silver ions into silver nanoparticles. Therefore, the biosynthesized nanoparticles is utilized for various biomedical applications.

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KEYWORDS: Silver Nanoparticles, UV- Visible Spectrometer, FT-IR, Antibacterial, Antioxidant property

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wide range of phytochemical compounds such as carbohydrates, proteins, alkaloids, flavonoids, glycosides, terpenoids and phenols which are for medicinal value of the plant [6]. Thus, in the present investigation the bio fabrication of silver nanoparticles was done using aqueous extract of *Bauhinia tomentosa*. The synthesised silver nanoparticles were characterized by using UV- Vis spectrophotometer and FTIR analysis. Further the silver nanoparticles were subjected to antibacterial activity by using well diffusion method and antioxidant property by hydrogen peroxide assay.

Experimental

2.1 Collection Of Plant:

The diseased free plant leaves was collected in the month of November from the garden of Kanchipuram district, Tamil Nadu and identified as *Bauhinia tomentosa* based on their morphological characters.

2.2 Preparation Of Plant Extract:

The collected plant leaves was cleaned with tap water to remove dust particles and followed by distilled water. 20 g of

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fresh leaves were weighed and with help of mortar and pestel the extract was prepared with using 50 mL of deionised water. And extract was filtered using Whatmann filter paper No:1. The collected filtrate was heat treated for about 80°C for 20 minutes. And the cured extract was used for the further study.

2.3 Bio Fabrication Of Silver Nanoparticles:

1mM concentration of silver nitrate solution was prepared using deionised water. To the 20 mL of 1mM of silver nitrate solution, 20 mL of the aqueous plant extract was added under continuous stirring. The solution mixture was kept under dark condition for 1 hr. The color change from dark green into reddish brown indicates the formation silver nanoparticles.

2.4 Purification Of Silver Nanoparticles:

The synthesized silver nanoparticle solution was centrifuged at 8000 rpm for 10 minutes. The supernatant was collected and stored for further settlement of nanoparticles. The pellet was dried at 50° C in hot plate and powder form of silver nanoparticles was collected.

2.5 Characterization:

UV- Visible Spectrophotometer And FT-IR Analysis:

The synthesized silver nanoparticle solution was analysed using UV- Visible spectrophotometer. The peak obtained at the range of 400 to 500 nm due to surface Plasmon resonance which confirms the formation silver nanoparticles. The powdered silver nanoparticle was mixed with KBr to make the pellet and analysed to know the presence of functional groups using FT-IR.

2.6 Antibacterial Activity:

The antibacterial activity for synthesized silver nanoparticles was studied using agar well diffusion method against *E. coli, S. aureus* and *S. typhi* with four different concentrations 30μ L, 60μ L, 90μ L and 120μ L. Muller Hinton agar medium was prepared and poured into the sterile petriplate and allowed to get solidifies. The wells were punched using sterile cork borer and inoculated with one day over night bacterial cultures and wells were loaded with synthesised silver nanoparticles solution of desired concentrations respectively.

2.7 Antioxidant Property:

The antioxidant property of synthesized silver nanoparticles was studied using hydrogen peroxide scavenging assay. The modified Dehpour method was used to determine the scavenging action of the synthesized silver nanoparticles towards Hydrogen peroxide using standard ascorbic acid. The solution of hydrogen peroxide (40mM) was prepared in phosphate buffer (pH=7.4). The silver nanoparticles of different concentration were taken in test tubes. The hydrogen peroxide prepared with phosphate buffer solution was added to it and incubate for 10 minutes. The absorbance was measured at 560 nm using UV spectrophotometer against blank solution containing phosphate buffer without hydrogen peroxide. The percentage of hydrogen peroxide scavenging activity of the silver nano materials was calculated by using the formula:

% Scavenging

= Absorbance of Control – Absorbance of Sample X 100 Absorbance of Control

Result and Discussion:

3.1 Bio fabrication of Silver Nanoparticles:

The aqueous extract of *Bauhinia tomentosa* is mediated to reduce the size silver ions into silver nanoparticles. It was confirmed by the visual observation of solution mixture color change into reddish brown (FIGURE 3.1). The silver nanoparticles were purified by centrifugation at 8000 rpm for 10 minutes (FIGURE 3.2).



FIGURE 3.1- Synthesizing of Silver Nanoparticles Using Aqueous Extract of Bauhinia tomentosa

FIGURE 3.2- Centrifuged Silver Nanoparticles Solution

3.2 Characterization:

3.2.1 UV- Visible Spectrophotometer Analysis:

The synthesized silver nanoparticles were characterized using UV- Vis spectrophotometer for primary conformation to know the formation of silver nanoparticles. The UV- Vis spectra showed the absorbance peak obtained at 440.0 nm (FIGURE 3.3). It is due to the Surface Plasmon Resonance of the surface electrons of silver nanoparticles. The appearance of single indicates that the sphere shape of silver nanoparticles.



FIGURE 3.3- UV- Visible spectra of synthesized silver nanoparticles using aqueous extract of Bauhinia tomentosa

3.2.2 FT-IR Analysis:

To analyze the presence of functional groups in the synthesized silver nanoparticles were performed. The FT-IR spectra registered the presence of phenol group at 3263.56 cm⁻¹, C-H stretch at 2916.37 cm⁻¹, C=C bending at 1602.85 cm⁻¹, C-O stretch at 1020.34 cm⁻¹, C-H bonding at 1346.31 cm⁻¹ and Aromatic C-H bending at 821.68 cm⁻¹. (FIGURE 3.4)



3.3 Antibacterial Activity:

The synthesized silver nanoparticles were subjected to antibacterial activity by well diffusion method. The bacterial cultures such as *Escherichia coli*, *Salmonella typhi* and *Staphylococcus aureus* were used for the bactericidal activity study. The minimum zones of inhibition of the plates (FIGURE 3.5) were measured in milimeter measurement (Table 3.1). The test was performed for duplicate plates.



FIGURE 3.5- Antibacterial Activity of Synthesised Silver Nanoparticles using Bauhinia tomentosa Against E. Coli

| S. <u>No</u> | Microorganism | Silver Nanoparticles Concentration | | | |
|-----------------|--------------------------|---------------------------------------|-----|-----|-----|
| | | Zone of inhibition in mm diameter | | | |
| | | 30 | 60 | 90 | 120 |
| | | μL | μL | μL | μL |
| 1. | Escherichia coli | 1.8 | 1.9 | 2.0 | 2.5 |
| 2. | Staphylococuus aureus | 1.5 | 1.8 | 2.1 | 2.3 |
| 3. | Salmonella typhi | 1.3 | 1.7 | 1.8 | 2.0 |

TABLE 3.1- Antibacterial Activity Of Synthesized Silver Nanoparticles – Zone Of Inhibition

3.4 Antioxidant Property:

The synthesized silver nanoparticles were subjected to hydrogen peroxide scavenging assay to analyse the antioxidant property. The silver nanoparticles showed gradual increase in antioxidant property according to their concentration utilized for the assay, ascorbic acid were used as standard. (FIGURE 3. 6)





4. Conclusion:

The green synthesis of metal nanoparticles has wide application in medicinal field due its biocompatibility property and low toxicity. The investigation was deals with Bauhinia tomentosa mediated synthesis of silver nanoparticles was confirmed by color change into reddish brown and further confirmed with UV- Visible spectra the peak obtained at 440.0nm and functional groups presence was confirmed with FT-IR spectra. The antibacterial activity was performed against Escherichia coli, Salmonella typhi and Staphylococcus aureus. The antioxidant property was done using ascorbic acid as standard, which showed the gradual increase of antioxidant activity with concentration increases. Thus, the investigation eye opens young scientists to shine their research into the green synthesis of metal nanoparticles which can be utilized for pharmaceutical purpose in future.

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