Optimization of Florisynthesis of Silver Nano Particles Using Cordia Sebastena Linn.

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Abstract. Synthesis of Silver Nanoparticles [AgNPs] using biological method is widely studied. This mostly involves the extracts of leaves of different plants which resulted in nano/microparticle formation. This was mostly due to the presence of complex phytochemicals. However flower of the plants are found to possess minimal amount of secondary metabolites as well as carotenoid pigments which may serve as a reducing agent. Thus, in this study synthesis of Silver nanoparticles using floral extract of Cordia sebastena and optimization in their synthesis using different pH [5, 7 and 9], concentration of silver nitrate [1mM, 2Mm and 3mM] and floral extract [0.5:10, 1:10 and 1.5:10] was conducted. The characterization of AgNPs was carried using UV-Visible Spectrophotometer, Fourier Transform Infra Red [FTIR] Spectrophotometer and Dynamic Light Scattering [DLS]. Further, the floral extract was screened for the presence of different phytoconstituents. The study showed varied Plasmon resonance ranging from 430 to 470nm. Fourier transform infrared spectrum reveals the presence of phytochemicals which acted as reducing and stabilizing agent. The presence of Alkaloids, flavonoids, Phenols, tannins, cardiac glycosides, terpenoids and pholobatannins were recorded in the extract of the flower of Cordia sebastena. The study confirmed the synthesis of AgNPs with the average particle size of 85.2nm. The optimized condition revealed that the flower extract added with 1:10 concentration to 2mM silver nitrate at pH9 gives smaller AgNPs.

Key Words: Silver nanoparticles, Biosynthesis, Optimization, Cordia sebastena, Florisynthesis, pH

1. Introduction

Nanotechnology is an interdisciplinary study of science that merges biology, physics and chemistry. When compared with microparticles, nanoparticles hold more surface area that improve its functional competence. A range of metals are exploited in the synthesis of nanoparticles which includes Au, Ag, Cu, Zn, Fe and Pd. The multifunctional potential of these nanoparticles include copious applications in various fields such as medicine, agriculture, food and textile industry. Though many metals were studied, silver nanoparticles [AgNPs] have reached a unique spotlight because of its characteristic properties, like electrical conductivity, chemical stability and surface Plasmon resonance properties and apart from that AgNPs has its extensive biological applications.

Green synthesis of nanoparticles has received an extensive interest in science and technology. Green synthesis of silver nanoparticle is sprouting into a vital branch of nanotechnology in recent decades for its huge applications which includes Anti-angiogenic Properties [1], Antimicrobials [2], Antioxidants [3], cytotoxic [4], Dye degradation [5], Anti-inflammatory, Anticancer [6] and

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Larvicidal activity [7]. Even though there are countless ways available for the synthesis of AgNPs, flower mediated biosynthesis is striking due to the presence of unique pigments in flowers. Our previous studies [8] on synthesis of AgNPs using plant parts resulted in formation of microparticles which was due to the presence of secondary metabolites. Thus, plant part possessing lesser amount of secondary metabolites are probed and this study was conducted using a flower of *Cordia sebastena*. Flowers are inexpensive and abundantly available and function as a biofactory with diverse organic compounds possessing flavonoids and carotenoids [9]. In the present study, synthesis of AgNPs using the flower extract of *Cordia sebastena* at room temperature is optimized under various pH, concentration of flower extract and silver nitrate. Synthesized AgNPs were characterized using various spectral analyses like UV–Vis spectroscopy, FTIR and DLS.

2. Material And Methods

The flowers of *Cordia sebastena* were collected from Vels Institute of Science, Technology and Advanced Studies, Chennai, India. The flowers were examined for pathogenic infections and healthy flowers were selected. The corolla alone was excised and washed in sterile distilled water to eliminate the surface contaminants. The washed corolla was transferred to a coarse filter paper to absorb the water.

2.1. Preparation of floral Extract

The floral extract was prepared by boiling 100gms of flower in 1000 ml of distilled water for 10 mins. Then the extract is filtered through Whatmann no 1 filter paper. The clear extract is used for further experiments. The floral extract acquired was dark saffron in colour (Fig. 1).

![Fig. 1. Flower of Cordia sebastena and its aqueous extract](image)

2.2. Phytochemical screening

The aqueous extract of the *Cordia sebastena* flower was evaluated for the presence of phytochemicals like flavonoids, phenols, tannins, saponins, phlobatannins, steroids, cardiac glycosides, and terpenoids as described by Evans [10] and Udayaprakash et al [11].

2.3. Synthesis of AgNPs
Silver nitrate solution was prepared using millipore water (Milli Q, IQ7000, Merck), with the concentration of 1mM, 2mM and 3mM. This was adjusted to three different pH using 1N NaOH and 1N HCl to obtain silver nitrate with the pH value of 5, 7 and 9. The prepared silver nitrate with different pH and molarity was treated with the floral extract at the ratios of 0.5:10, 1:10 and 1.5:10 respectively.

2.4. Characterization of Silver Nanoparticle

The reduction of silver ions to silver nanoparticles was analysed using UV–Vis spectrophotometer with the wavelength ranging from 300–650 nm. Fourier transform infra-red spectroscopy (FT-IR) analysis of AgNPs were recorded in the range of 400–4000 cm$^{-1}$. Further, the size of the particles formed was measured using Dynamic Light Scattering (DLS) Spectroscopy.

3. Results

3.1. Optimization of silver nanoparticle synthesis

To determine the ideal concentration of flower extract, concentration of silver nitrate and pH to synthesis silver nanoparticle were optimized. Synthesized silver nanoparticles were confirmed due to the colour change from colourless to reddish brown after the addition of aqueous extract of Cordia sebastaena flowers. The characteristic colour change is noticed for the samples of pH 7 and pH 9 (Fig. 2).
Fig 2: Optimization of Florisynthesis of AgNPs at different pH, concentration of silver nitrate and floral extract of Cordia sebastena

3.2 UV-vis Spectroscopy

Although all samples were analysed for UV-spectroscopy, only five samples showed the characteristic peak from 430 to 470nm range. The spectral responses are inversely proportional with the aggregation of the nanoparticles formed. When the aggregation of nanoparticles takes place and the diameter increases, the peak of Plasmon resonance shifts towards broader and longer wavelengths. The UV spectrum recorded in between 430nm range to 470nm range along with the suitable condition is presented in Fig. 3.

Fig. 3. UV spectrum of AgNPs synthesised using floral extract of Cordia sebastena

3.3 FTIR Spectral Analysis
The FTIR spectral analysis for the specific samples, i.e. 1mM AgNO3- pH9- 1:10, 2mM AgNO3- pH9- 1:10, 3mM AgNO3- pH9- 0.5:10, 3mM AgNO3- pH9- 1:10 and 2mM AgNO3- pH9- 1.5:10 were carried. From the FTIR analysis the presence of functional groups, i.e. alcohols, alkanes, alkynes, alkenes, aldehydes, esters, aliphatic amines and carboxylic acid were confirmed. The absorption spectra and their characteristic functional groups of individual samples are presented in Table 1. The FTIR spectra recorded for different samples are presented in Fig. 4.

**Table 1: FTIR Spectral Characteristic Absorption and Functional Group**

| Sample | Characteristic Absorption [cm⁻¹] | Functional group |
|--------|----------------------------------|------------------|
| 1mM AgNO3- pH9- 1:10 | 1080.06 | C-N stretch |
| | 1512.09 | Aromatic C≡C stretching |
| | 1743.53 | Aldehyde C=O stretch |
| | 2313.46 | Alkynyl C≡C bending |
| | 3186.18 | Alkynyl C-H stretch |
| | 3756.42 | Amide N-H stretch |
| 2mM AgNO3- pH9- 1:10 | 1047.27 | C-N stretch |
| | 1171.68 | Ester C-O stretch |
| | 1745.46 | Ester C=O stretch |
| | 1424.33 | Aromatic C≡C stretching |
| | 2942.21 | Alkane asymmetric stretch |
| | 3063.71 | Terminal alkene =C-H stretch |
| | 3249.43 | Amine N-H stretch |
| | 3544.92 | Amide N-H stretch |
| 3mM AgNO3- pH9- 0.5:10 | 1013.52 | C-N stretch |
| | 1360.69 | Sp³ alkane stretch |
| | 1422.40 | Aromatic C≡C bending |
| | 2936.42 | Alkane bend |
| | 3059.38 | Alkenyl C-H stretch |
| | 3397.38 | Carboxylic Acid OH stretch |
| 3mM AgNO3- pH9- 1:10 | 1015.45 | C-N stretch |
| | 1360.69 | Sp³ alkane stretch |
| | 1431.08 | Aromatic C≡C bending |
| | 1625.88 | C≡N stretch |
| | 1715.56 | C=O stretch |
| | 2933.53 | Alkane Sp stretch |
| | 3056.96 | Alkenyl C-H stretch |
| | 3419.56 | Amine N-H stretch |
| 2mM AgNO3- pH9- 1.5:10 | 1011.59 | C-N stretch |
| | 1510.16 | Aromatic C≡C bending |
| | 1629.74 | C≡N stretch |
| | 1745.46 | Aldehyde C=O stretch |
| | 3054.07 | Asymmetric alkene =C-H stretch |
| | 3438.84 | Amide N-H stretch |
The Dynamic light scattering studies of the specific samples, i.e. 1mM AgNO₃- pH9- 1:10, 2mM AgNO₃- pH9- 1:10, 3mM AgNO₃- pH9- 0.5:10, 3mM AgNO₃- pH9- 1:10 and 2mM AgNO₃- pH9- 1.5:10 revealed formation of AgNPs at various size. The size ranges from 1728nm to 85.2nm. The microparticles are recorded for the sample treated with 1.5 : 10 ratio of floral extract and silver nitrate. The lowest size was recorded for the sample treated with the ratio of 1:10 concentration of floral extract and silver nitrate. All the samples studied recorded their PDI index of less than 0.5. The sample, average size recorded and PDI index are presented in Table 2. The DLS spectrum and zeta potency of the sample, 2mM AgNO₃- pH9- 1:10 is provided in Fig. 5.

Table 2: Paricle size Distribution and PDI of synthesized nanoparticles

| S.NO | Sample                  | Average size | PDI  |
|------|-------------------------|--------------|------|
| 1    | 1mM AgNO₃- pH9- 1:10    | 169.12 nm    | 0.481|
| 2    | 2mM AgNO₃- pH9- 1:10    | 85.2 nm      | 0.402|
| 3    | 3mM AgNO₃- pH9- 0.5:10  | 300.3 nm     | 0.365|
The study demonstrated the synthesis of Silver Nanoparticles (AgNPs) using the floral extract of *Cordia sebastena*. The optimization in synthesis of AgNPs was carried out using different parameters which includes, the pH, concentration of silver nitrate in synthesis of AgNPs and the concentration of floral extract in synthesis of AgNPs. The differently synthesized colloids were subjected to UV, FTIR and DLS.

UV–VIS spectroscopy is one of the best tools to examine the synthesis of the AgNPs have a property called surface Plasmon resonance which occurs is mainly due to the oscillation of the free electrons in surface of the metallic nanoparticles by any peripheral energy source[19]. Thus the physical state of AgNPs could be analysed by this optical property in the red or infrared region of the spectrum. The results of the present study confirms the fact that the alkaline pH is most suitable for the florisynthesis of AgNPs which is accordance with the reports of Dubey et al [12] and Andreescu et al [13].

FTIR gives the data of functional groups in the synthesised AgNPs for understanding their transfer of inorganic AgNO\textsubscript{3} to elemental silver by the action of the various phytoconstituents in the aqueous extract *Cordia sebastena* flowers. As known the spectra of diagnostic region excluding the fingerprint region is analysed to find the major functional groups acting on the synthesis of AgNPs. Functional group of alcohol, carboxylic acid (O-H stretching) involved in formation of AgNPs from AgNO\textsubscript{3} is in correlation with earlier report [14].

The phytochemical evaluation of the floral extract of *Cordia sebastena* resulted in detection of phenols, tannins, Alkaloids, flavonoids and terpenoids. The presence of Coumarin was reported from the flowers of *Cordia sebastena*. Among the various compounds produced by flowers, phytoconstituents such as polyphenols, flavonoids, anthocyanidins, catechins, carotenoids were
participating in many bioactivities and are also known for the synthesis of silver nanoparticles [15].

The principle of DLS is to measure the fluctuations in the intensity of scattering light in the suspended particles. The Particle Dispersion Index [PDI] is used to determine the uniformity and dispersion of synthesized nanoparticle in the suspension. A broad distribution of particle is always resulted in aggregation of particle and the size and stability differs time to time. The normal Index of PDI value should always less than 0.5 [16]. The optimized condition is confirmed that the flower extract added with 1: 10 concentration to 2mM silver nitrate at pH 9 gives smaller AgNPs. The average size of the particle and the PDI value noted was 85.2nm and 0.402 respectively. Nanoparticles are defined as the particles exhibiting their size below 100 nm on any dimension [17].

5. Conclusion
The florisynthesis of AgNPs using the aqueous extract of Cordia sebastena was carried. Optimization on synthesis of AgNPs was performed using different parameters like pH, concentration of AgNO₃ and different concentration of floral extract. A total of 27 samples were analysed using UV among which only five samples have recorded the peak range between 430 – 480nm. The phytoconstituents in the Cordia sebastena aqueous extract were responsible for reducing and capping of AgNPs, which were confirmed by FTIR. The optimized condition is confirmed that the flower extract added with 1: 10 concentration to 2mM silver nitrate at pH 9 gives smaller AgNPs. Further studies using different flowers and other optimizing factors are recommended.

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