Synthesis and Characterization of Silver Nanoparticles from *Ocimum basilicum L. var.thyrsiflorum*

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Abstract: Biosynthesis of nanoparticles is an important area in the field of nanotechnology, which has economic and ecofriendly benefits over chemical and physical methods of synthesis. In the present study leaf extract of *Ocimum basilicum L. var.thyrsiflorum* was assessed for the synthesis of Silver nanoparticles using 1mM Silver Nitrate aqueous solution. Nanoparticles were characterized with the help of UV-Vis absorption spectroscopy analysis, Fourier Transform Infrared (FTIR) analysis, Scanning Electron Microscopy (SEM) and EDX analysis. The Synthesized nanoscale particles were confirmed by analyzing the excitation of Surface Plasmon Resonance (SPR) using UV–visible spectrophotometer at 436nm. Further SEM analysis confirmed the range of particle size between 22 to 42 nm. Fourier transform infrared spectrum confirmed the presence of high amount of phenolic compounds in the plant extract, possibly influenced the reduction process and stabilization of nanoparticles.

Keywords: Silver nanoparticles, Surface Plasmon Resonance, Ocimum basilicum L. var.thyrsiflorum, Silver Nitrate, SEM analysis.

1. Introduction

Nanotechnology is an active area of research with tremendous applications for society, industry and medicine. The non-polluting nanotechnologies have revolutionized the production of nanomaterials as environmentally safe products. Several chemicals used in the chemical and physical synthesis of nanoparticles are toxic which leads to environmental pollution [1]. Therefore biological sources can be an alternative for the synthesis of nanoparticles [2]-[4]. Plants are the richest bioresources of drugs in traditional and modern medicine [5]. Silver nanoparticles have wide range of applications such as catalysis [6], drug delivery [7], biosensing [8], [9] and optics [10]. Biogenic path of nanoparticle synthesis using microorganisms [11]-[13], enzymes [14] and plant extracts [15]–[20] were suggested as possible ecofriendly alternatives to chemical and physical methods. Here we report an in expensive and green method for the synthesis of Silver nanoparticles by reduction process using Ocimum basilicum L. var.thyrsiflorum, the plant is extensively used in traditional medicine. The genus Ocimum

belongs to Lamiaceae family and consists of 160 species with 24 species native to India [21]. The essential oils of Ocimum consists euginole, methyl chavicol, linalool, geraniol, 1,8-cineol, citral and camphor [22]. These essential oils are being used as pharmaceutical agents because of their anticancer, antiasthamatic, antistress, antimicrobial activity [23], antidiabetic [24] and antioxidant activity [25]. Herein we report the Synthesis and characterization of Silver nanoparticles first time from *Ocimum basilicum L. var.thyrsiflorum*.

2. Materials and Methods2.1 Collection of plant material

The genus Ocimum belongs to Lamiaceae family. *Ocimum basilicum L. var.thyrsiflorum* (Fig. 1) is a traditional medicinal plant distributed in most parts of the India. Local name is Adavi sabza or Tella sabza and the trade name is

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Basil. The plant is stout, nearly glabrous and herbaceous with short recemes. Leaves are elliptic and ovate and the flowers are white in terminal thyrsoid panicles (Fig. 2).

Ocimum basilicum L. var.thyrsiflorum leaves were collected from the Medicinal plant garden, SR&BGNR Govt. Arts & Science College of Khammam, Andhra Pradesh and identified by the plant systemic laboratory, Department of Botany, Kakatiya University, Warangal, A.P.India and further the voucher specimen was preserved in the department as a record.



Figure 1: Ocimum basilicum L. var.thyrsiflorum plant



Figure 2: Inflorescence.

2.2 Preparation of leaf extract

Fresh basil leaves were washed several times with tap water and later with deionised water. 10 grams of washed fine cut leaves along with 100 ml double distilled water were taken in 250 ml glass beaker and boiled for 5 minutes at 80°c. The extract was cooled to room temperature and filtered with Whatman No 1 filter paper. The filtrate was centrifuged for 10 minutes at 10000 rpm, the supernatant was collected and stored at 4° C. The filtrates are used as reducing and stabilizing agents.

2.3 Preparation of 1 mM AgNO₃ solution

Accurate concentration of 1 mM AgNO₃ (Merck India Ltd) was prepared by dissolving 0.169 gm AgNO₃ in 1000 ml double distilled water and stored in Amber coloured bottle to avoid auto oxidation of Silver.

2.4 Bio synthesis of Silver nanoparticles

In the single step green synthesis, 5 ml of leaf extract was added to 95 ml of 1 mM aqueous AgNO₃ solution and heated up to 80°C for 5 minutes, the immediate colour change (Fig. 3) indicate the formation of Silver nanoparticles.



Figure 3: *Ocimum basilicum L. var.thyrsiflorum* leaf extract, aqueous AgNo₃, Silver nano particles and color change respectively.

The silver nanoparticles solution thus obtained was purified by repeated centrifugation at 10000 rpm for 15 minutes. The supernatant was transferred to a clean dry beaker for further settlement of particles and repeated centrifugation was carried using cooling microfuge to get dried and purified Silver nanoparticles. The particles obtained were used for further characterization.

3. Characterization

3.1. UV –Visible spectra analysis

Synthesized silver nanoparticles were initially characterized by taking small aliquot of sample in to UV –Visible spectrophotometer absorption spectra at 300-700 nm using Shimadzu UV -1800 Spectrophotometer.

3.2. SEM analysis of silver nanoparticles

Scanning electron microscopic (SEM) analysis was carried by using Zeiss, EV-18 model.

3.3 EDX Analysis

Energy Dispersive X-ray analysis (EDX) was carried out on Zeiss, EV-18 model. The peaks obtained from EDX gives the element composition of the sample.

3.4 FTIR- Spectroscopy

Fourier-transform infra red spectroscopy Bruker Tensor 27 model was used for the analysis of the reduced silver. The spectrum was recorded in mid-IR region of 400-4000 cm⁻¹ with 16 scan speed, using attenuated total reflectance (ATR) technique.

3.5 Preliminary screening of Phytochemicals

About 10 gram of washed fine cut leaves were soaked in 100 ml of deionised water and boiled for 5 minutes to obtain crude extract. Harborne, 1998 method was employed to screen the phytochemicals present in the leaf extract [26]. Natural chemical groups such as amino acids, proteins, carbohydrates, flavonoids, sterols, terpenoids and phenolic compounds were identified and reported in Table No.1. The aqueous extract was further exemplified by FT-IR spectroscopic studies to reveal the presence of characteristic functional groups.

Table No.1 Preliminary Screening of Phytochemicals(+) presence (-) absence

S.No	Phytochemicals	Result
1.	Amino acids	+
2.	Carbohydrates	+
3.	Alkaloids	-
4.	Flavonoids	+
5.	Saponins	-
6.	Sterols	+
7.	Tannins	-
8.	Proteins	+
9.	Phenolic compounds	+

4. Results and Discussion

The present study reports the use of Ocimum *basilicum L. var.thyrsiflorum* for the Synthesis of Silver nanoparticles. The plant extract may act as reducing and capping agents in silver nanoparticles synthesis. Studies have indicated that biomolecules like protein, carbohydrates, flavonoids and phenols not only play a role in the capping of the nanoparticles, but also play an important role in reducing the ions to the nano size [27], [28]. The biomolecules found in these extracts like enzymes, vitamins, proteins, amino acids, and polysaccharides [29] play a vital role in the reduction of Ag+ ions.

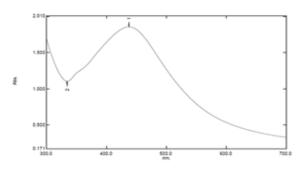


Figure 4: UV-Visible absorption spectrum of *Ocimum* basilicum L. var.thyrsiflorum

The nanoparticles were primarily characterized by UV-Visible Spectroscopy, which is proved to be a very useful technique for the analysis of nanoparticles. As the leaf extract was mixed with the aqueous solution of the silver ion complex it was changed from red to golden yellow colour (Figure 3). This is due to the excitation of the surface plasma vibrations, which indicates the formation of the Silver nanoparticles [30]. UV-Visible Spectrograph of Silver nanoparticles has been recorded as a function of time by using quartz cuvette with distilled water as the reference. The UV spectrum absorption is recorded at 436 nm (Fig. 4).

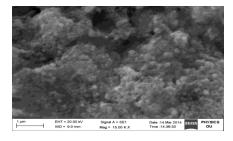


Figure 5: SEM image of silver nanoparticles formed by Ocimum basilicum L. var.thyrsiflorum

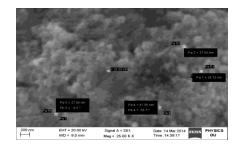


Figure 6: SEM image showing the size of silver nanoparticles.

The SEM image (Fig. 5) is showing the high density silver nanoparticles synthesized from the leaf extract, further confirmed the development of silver nano structure. The SEM image shows the formation of porous surface with spherical nano particles. They are clearly distinguishable in 22.92-41.50 nm in size (Fig. 6).

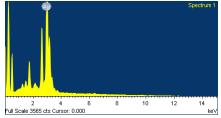


Figure 7: EDX image of silver nanoparticles produced from Ocimum *basilicum L. var.thyrsiflorum*.

The EDX spectra exhibit the purity of the material and the complete chemical composition of synthesized silver nanoparticles. In the present synthesis EDX analysis (Figure.7) shows high percentage of silver indicating the purity of the synthesized sample.

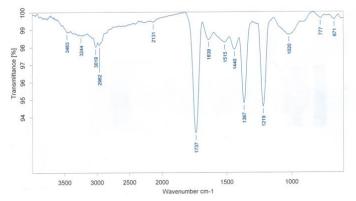


Figure 8: FTIR-spectrum of bio synthesized silver nanoparticles formed by Ocimum *basilicum L. var.thyrsiflorum.*

The FTIR spectrum of silver nanoparticles is shown in Figure.8. The band at 3463 cm^{-1} is assigned to the O-H stretching of H-bonded alcohols and phenols. The peak formed at 2962 cm⁻¹ is because of C-H stretching and symmetric stretching of methoxy groups. The strong peak at 1737 cm⁻¹ shows the stretching vibrations of C=O. The band at 1639 cm⁻¹ corresponds to the N-H bending of primary amines. The value 1440 cm⁻¹ is related to the C-C stretching of aromatic ring structure. The bands at 1367cm⁻¹ are related to the C-N streching of aromatic amine group. Whereas in the region 1020 cm⁻¹ are corresponding to the C-C stretching of alcohols, carboxylic acids, ethers and esters which are binding to metal to form a silver nanoparticle.

The preliminary phytochemical analysis of leaf extract revealed the presence of amino acids, carbohydrates, flavonoids, sterols, terpenoids, proteins, and phenolic compounds as shown in Table.1. FT-IR predicts the molecular configuration of different functional groups present in the extract. Considerable absorption peaks are found at 3463cm⁻¹, 2962 cm⁻¹, 1737 cm⁻¹, 1639 cm⁻¹, 1440 cm⁻¹, 1367cm⁻¹, 1020 cm⁻¹ respectively (Fig. 8). In the present study ,the peaks are more charecteristic of eugenol [32],linalool, methylchavicol and flavanoids.

5. Conclusion

The present study reveals that the plant species of *Ocimum basilicum L. var.thyrsiflorum* is good source for the synthesis of silver nanoparticles at a faster rate. The formation of silver nanoparticles was confirmed by the colour change within 30 minutes. The bioreduced silver nanoparticles were characterized using UV-Vis, SEM, FTIR techniques.

The Silver nanoparticles formed were quite stable in the solution. SEM analysis confirmed the range of particle size between 22 to 42 nm. The carbohydrates, flavanoids and poly phenols constituents present in leaf extract act as the surface active stabilizing molecules for the synthesis of Silver nanoparticles. The method was unique and cost effective. Still more clinical trials are needed to support its therapeutic uses.

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