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Green Synthesis of Silver Nanoparticles using Fruits of *Coriandrum sativum* Linn and Its Antioxidant Activity

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ABSTRACT

The present work describes a simple and eco-friendly biosynthesis of silver nanoparticles using the fruits of *Coriandrum sativum* Linn [Family: Apiceae] and its determination of in *vitro* antioxidant activity. Methanol and aqueous extracts of dried fruits of *C. sativum* were prepared. The aqueous silver ions (1 mM, 2 mM and 3 mM) with heat and without heat when exposed to fruit extracts were reduced and resulted in silver nanoparticles whose size is in range of 50-200 nm. The silver nanoparticles were characterized by UV-Visible, Fourier transform infra-red spectroscopy (FT-IR) and scanning electron microscopy (SEM) with EDAX techniques. *In vitro* antioxidant activity was carried out using DPPH method with 3.125 to 100 µg/mL concentration of silver nanoparticles. Out of 3 different concentrations, 1 mM silver nitrate reduction was better using methanol and aqueous extract (0.2 mL). The *in vitro* antioxidant activity was found to show biphasic response and inhibitory activity was found between 100 to 25 µg/mL followed by stimulatory activity from 12.5 to 3.25 µg/mL which was not significant. It can be concluded silver nanoparticles synthesized by green synthesis using dried fruits of *C. sativum* possess antioxidant activity.

1. Introduction

In recent years, researchers in the field of nanotechnology are finding that there is an expanding research in the synthesis of metal nanoparticles due to the potential applications for the development of novel technologies. Noble metal nanoparticles are extensively studied because of their wide applications [1-3]. Among the various noble metal nanoparticles, gold and silver have several applications in sensors, detectors, and antibacterial agents [4–6].

Metal and semiconductor nanostructures of different sizes and shapes can now be routinely synthesized by means of various chemical and physical methods. Their performance depends critically on their size, shape, and composition. Though numerous chemical methods are available for metal nanoparticles synthesis, copious reactants and starting materials are used in these reactions that are toxic and potentially hazardous. Increasing environmental concerns over chemical synthesis routes have resulted in attempts to develop bio-mimetic approaches. One of them is the synthesis using plant extracts eliminating the elaborate process of maintaining the microbial culture and often found to be kinetically favorable than other bioprocesses. Bio-molecules as reducing agents are found to have a significant advantage over their counterparts as protecting agents.

Environmentally nanoparticles synthesis procedures do not use any toxic chemicals in the synthesis protocols. In these aspects synthetic methods based on naturally occurring biomaterials provide an alternative means for obtaining these nanoparticles. During recent times several groups have achieved success in the synthesis of Ag, Au, and Pd nanoparticles using extracts obtained from unicellular organisms like bacteria [7-10] and fungi [11-13] as well as extracts from plant parts, e.g., geranium leaves [14], lemon grass [15], neem leaves [16], aloe vera [17] and several others [18-21]. The spectacular success in this field has opened up the prospect of developing bio-inspired methods of synthesis of metal nanoparticles with tailor-made structural properties. Among the various bioreductants, *Coriandrum sativum* leaves extract was chosen for the present study since they have minerals and vitamin contents including calcium, phosphorus, iron, carotene, thiamine, riboflavin, and niacin.

Coriander (*Coriandrum sativum* Linn.) an annual of the Apiaceae family is one of valuable medicinal and seasoning plant. This species comes from the Mediterranean region and it is grown all over the world. The coriander fruit and essential oil isolated from it are used for medicinal purpose. It is used to treat menstrual disorder, secondary infertility, ovaritis and cervicitis. It is used to treat female diseases such as menoxenia, ovulation type dysfunctional uterine bleeding [22]. It is aphrodisiac to enhance sexual function and reproductive capacity. It is used for treating leucorrhea and spermatorrhea. Coriander fruit possess stimulant and carminative properties [23]. Its oil is bactericidal and larvacidal [24]. It is hypoglycemic and anti-inflammatory [25]. The fruits are used as astringent, anthelmintic, emollient, stomachic, antibilious, digestive, appetizer, constipating, diuretic, antipyretic, refrigerant, tonic, expectorant, anodyne, antidiabetic and dyspepsia [26].

Inspite of its abundant uses, the green synthesis of silver nanoparticle of dried fruit of Coriandrum *sativum* have not been reported but from leaves, nanoparticle was synthesised [27]. The main objective of this study was to propose a simple and eco-friendly biosynthesis of silver nanoparticles using the fruits of *Coriandrum sativum* Linn and its determination of in *vitro* antioxidant activity.

2. Experimental Methods

2.1 Plant Material

The *Coriandrum sativum* fruits were collected from local market in Bangalore, Karnataka, India and it was identified and authenticated by Botanist, Natural Remedies Pvt Ltd., Bangalore. A voucher specimen was deposited in The Oxford College of Pharmacy, Bangalore. The fruits were dried in shade and powdered coarsely, passed through sieve no. 40 and stored in air tight container for further use.

2.2 Preparation of Fruit Extract

Coarsely powdered fruits of *C. sativum* 50 g, each were boiled with methanol and water (200 mL) respectively for 30 minutes and filtered and make up the volume to 100 mL with methanol and water respectively. The solution is preserved for further use.

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2.3 Drugs and Chemicals

Silver nitrate, DPPH were purchased from SD Fine chemicals Ltd, Mumbai. All chemicals and reagents used in this study were at least of analytical grade.

2.4 Synthesis of Nanoparticles

A set of 1 mM, 2 mM and 3 mM aqueous solution of silver nitrate were prepared for synthesis for silver nanoparticles [28]. Exactly 9 mL of each 1 mM, 2 mM and 3 mM silvernitrate solution was added to 0. 1 mL; 0.2 mL; 0.3 mL; 0.4 mL and 0.5 mL aqueous and methanol extract of the fruit of *C. sativum* to obtain silver nanoparticles (i.e., for aqueous extract alone we used 3 concentration of silver nitrate x 5 different dilution of extract x with heat and without heat—total of 30 tubes for 1 extract). The different concentrations of silver nitrate and extracts were used to standardize the optimum concentration of silver nitrate and extract needed for synthesis of silver nanoparticles. The nanoparticles were synthesized at room temperature and with application of heat at 75 °C. Out of 60 tubes for both extracts, 1 mM silver nitrate and 0.2 mL of extract gave higher λ max when compared to other concentrations and formation of nanoparticles with and without heat at 75 °C. Excess quantity of 90 mL 1 mM silver nitrate and 2 mL of aqueous and methanol extract were prepared respectively.

2.5 Lyophilization Procedure for the Reluctant Sample Mixture

After the desired reaction period, the solution containing silver nanoparticles were lyophilized. The reluctant samples were centrifuged 10,000 rpm for 15 minutes. After 15 minutes, discard the supernatant and collected the pellet and freeze dried. The lyophilized samples were kept in the freezer at 4 °C for further analysis.

2.6 Characterization of Silver Nanoparticles

The IR measurements were recorded for silver nanoparticles in the transmittance mode Range: 4000 to 400 cm^{-1} by using Perkin-Elmer-Spectrum RXI FT-IR (RXI FT-IR) instrument individually. Ultra Violet-visible spectroscopy analyses were carried out by UV–visible spectrophotometer Shimadzu in the range of 200 nm – 800 nm, with the scanning speed of 100 nm/min. The morphology examination of dried powder samples were analyzed with Scanning Electron Microscope (SEM) HITACHI-S-3400N model fitted with an energy dispersive X-ray analyzer (EDAX) allows a qualitative detection and localization of elements in the samples. The SEM enables a direct observation of the surface microstructures of the nanoparticles [28].

2.7 In vitro Antioxidant Activity by DPPH Method

In vitro antioxidant activity/Free radicals scavenging potential of silver nanoparticles were tested against a Methanol solution of 1,1–Diphenyl–2 –picryl hydrazyl (DPPH) [29]. Antioxidants reacts with DPPH and convert it to 1,1–Diphenyl–2–Picryl hydrazine. The degree of discoloration indicates the scavenging potentials of the extract. The change in the absorbance produced at 517 nm has been used as measure of Antioxidant activity. Different concentration of silver nanoparticles (3.125 to 100 µg/mL) were mixed with DPPH methanol solution in 3 mL of total reaction mixture and allowed to react at room temperature. After 30 minutes the absorbance values were measured at 517 nm and converted to percent antioxidant activity. For a comparative study, the ascorbic acid was used as the standard. The percentage inhibition activity was calculated by using a formula.

Percent Inhibition = 100 (Absorbance of Blank- Absorbance of Sample) / Absorbance of Blank

3. Results and Discussion

Different concentration of silver nitrate and extract were tried with and without heat. 0.2 mL of extract and 1 mM concentration of silver nitrate was found to be optimum concentration for production of nanoparticles. Both methanol and aqueous extract were mixed with 0.2 mL extract each with 1 mM silver nitrate solution at room temperature and at 75 °C and 4 silver nanoparticles were synthesized (CS-01: 1 mM aqueous extract; CS-02: 1 mM methanol; CS-03: 1 mM aqueous extract with heat 75 °C; CS-04: 1 mM methanol extract with heat 75 °C respectively). The UV visible spectroscopy showed λmax at 460 nm confirming the formation of silver nanoparticles (Fig. 1).

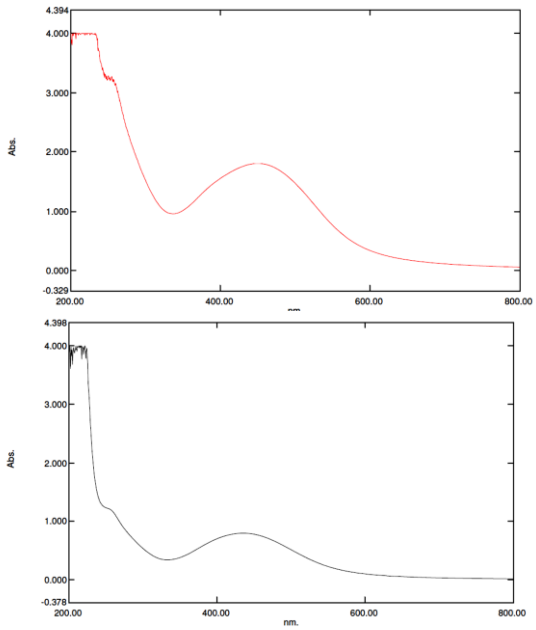


Fig. 1 UV spectrum of a) CS-02 and b) CS-02

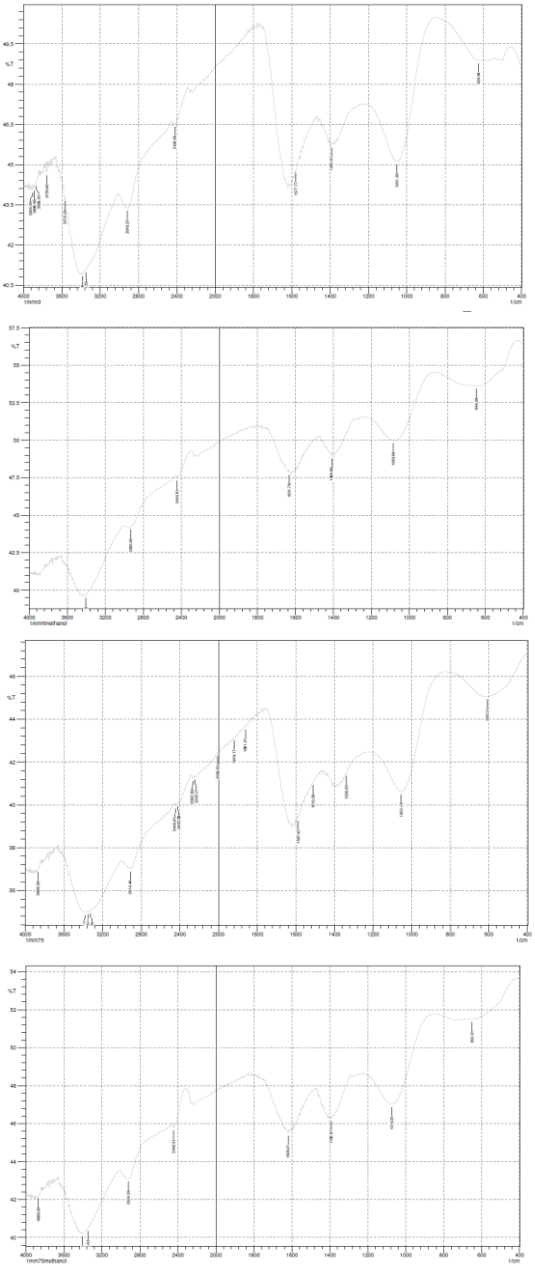
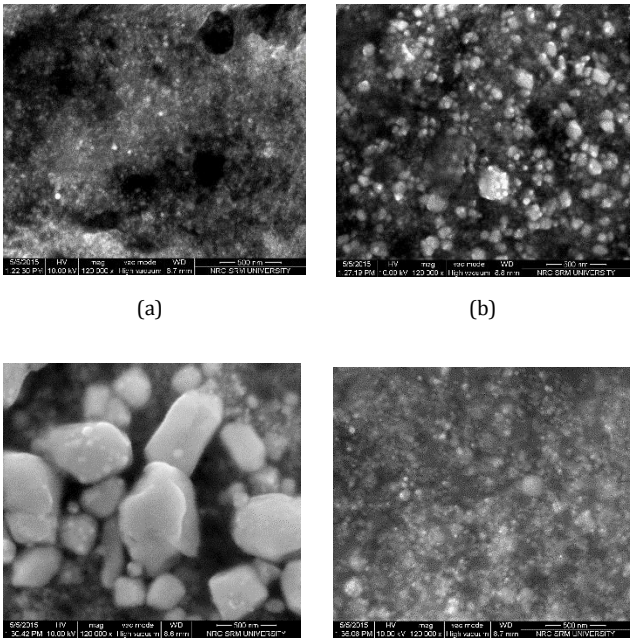


Fig. 2 FT-IR spectrum of a) CS-01, b) CS-02, c) CS-03, and d) CS-04

Fig. 2 shows the FTIR spectra of aqueous silver nanoparticles prepared from the *Coriandrum sativum* fruit extract (aqueous and methanol without and with heat) respectively. The peaks near 3430 cm^{-1} and near 2920 cm^{-1} were assigned to O–H stretching and aldehydic C–H stretching, respectively. The weaker band at 1635 cm^{-1} corresponds to amide I, arising due to carbonyl stretch in proteins. The peak at 1038 cm^{-1} corresponds to C–N stretching vibrations of the amine. IR spectroscopic study confirmed that the carbonyl group form amino acid residues and proteins has the stronger ability to bind metal indicating that the proteins could possibly form a layer covering the metal nanoparticles (i.e., capping of silver nanoparticles) to prevent agglomeration and thereby stabilize the medium. This suggests that the biological molecules possibly perform dual functions of formation and stabilization of silver nanoparticles in the aqueous medium.



(a) (b)

(c) (d)

Fig. 3 SEM images of a) CS-01, b) CS-02, c) CS-03, and d) CS-04

The SEM-EDAX spectra obtained for the study samples are given in Fig. 3. From SEM images, by morphologically all the 4 samples look different while the particle size showing variations which ranges between 50 to 200 nm. As it can be seen from Fig. 3, the 1 mM room temperature with both aqueous and methanol and 75 °C methanol, the particle were looking like spherical and 1 mM aqueous extract at 75 °C showed slightly bigger particles with rhomboidal crystals. The Fig. 4 of EDAX analysis revealed that the percentage of silver was found to be 100% with weight being 57.21; 47.29; 64.33 and 37.16 % wt of Silver for 1 mM water, 1 mM methanol, 1 mM water with 75 °C and 1 mM methanol with 75 °C respectively.

Table 1 shows the *in vitro* antioxidant activity of silver nanoparticle. The *in vitro* antioxidant activity was found to show biphasic response and inhibitory activity was found between 100 to 25 µg/mL followed by stimulatory activity from 12.5 to 3.25 µg/mL which was not significant with both 1 mM silver nanoparticles synthesized using methanol and water at room temperature. The *in vitro* activity of nanoparticle synthesized at 75 °C did not show good activity.

Table 1 *In vitro* antioxidant activity by DPPH assay

Concentration	% inhibition			
[µg/mL]	CS-01	CS-02	CS-03	CS-04
100	40.4 ± 0.033	30.3 ± 0.015	9.09 ± 0.060	17.17 ± 0.018
50	15.15 ± 0.025	14.14 ± 0.003	9.09 ± 0.028	13.13 ± 0.035
25	10.1 ± 0.045	0	5.05 ± 0.040	2.02 ± 0.028
12.5	0	-3.03 ± 0.063	2.02 ± 0.060	5.05 ± 0.029
6.25	1.01 ± 0.028	-7.07 ± 0.018	-4.04 ± 0.015	-4.04 ± 0.045
3.125	-3.03 ± 0.075	-6.06 ± 0.020	-4.04 ± 0.020	-2.02 ± 0.055

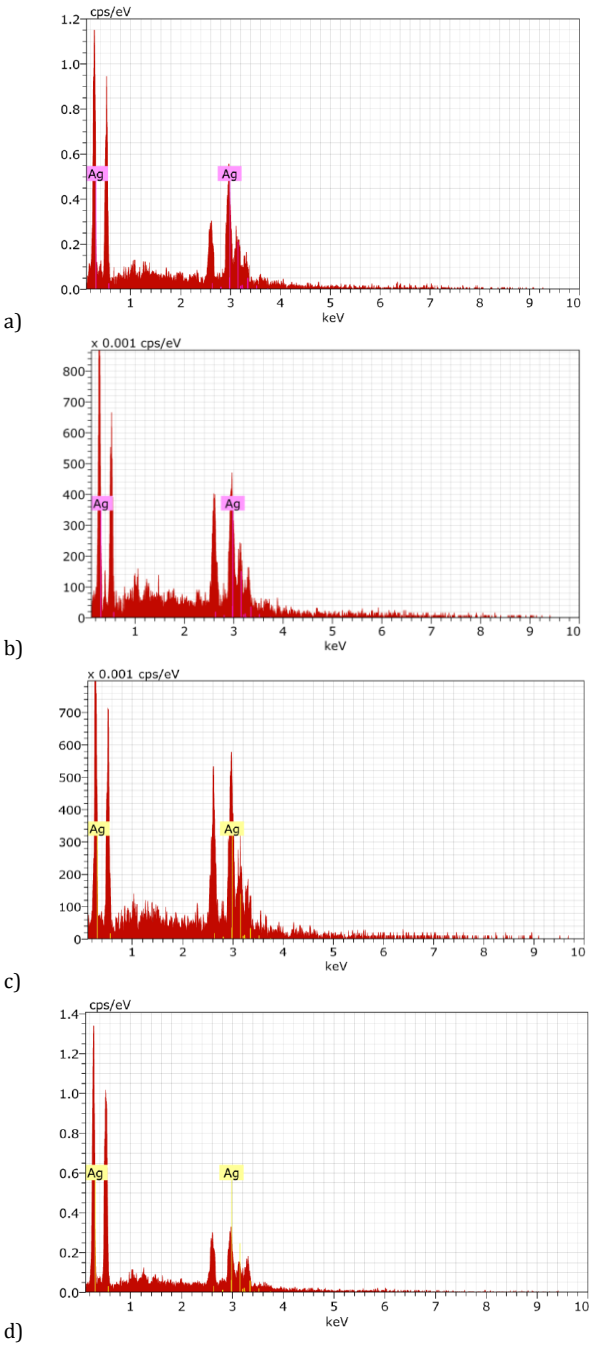


Fig. 4 EDAX of a) CS-01, b) CS-02, c) CS-03, and d) CS-04

4. Conclusion

As conclusion this present preparation is a simple, fast, and economical biological procedure to synthesize Ag nanoparticles using *Coriandrum sativum* fruit extract. The characterization studies SEM, UV-visible and FTIR spectroscopic techniques confirm the formation of silver nanoparticles and also the synthesized nanoparticles showed its *in vitro* antioxidant activity.

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