

Review article

Green synthesis of nanosilver particles from extract of *Dracocephalum Lindbergii*

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ABSTRACT: We describe the synthesis of silver nanoparticles (Ag-NPs) using aqueous extract of *Dracocephalum lindbergii*. UV-visible spectroscopy, X-ray diffraction (XRD), scanning electron microscopy (SEM), and X-ray energy dispersive spectrophotometer (EDX) were performed to ascertain the formation of Ag-NPs. UV-visible absorption spectra of the reaction medium containing silver nanoparticles showed maximum absorbance at 416 nm. The XRD pattern revealed the crystalline structure of SNPs. The SEM analysis showed the size and shape of the nanoparticles. The environmental friendly method provides simple, easy and cost effective faster synthesis of nanoparticles than chemical methods and can be used in several areas such as food and medicine.

KEYWORDS: Limiting molar conductance, Ion-pair association Constants, Binary Mixed solvents, Walden product, Association constant.

GRAPPHICAL ABSTACT:



Introduction

An important area of research in nanotechnology is the synthesis of nano silver particles. Silver has long been recognized as having an inhibitory effect towards many bacterial strains and

microorganisms [1]. Antibacterial activity of the silver containing materials used in medicine to reduce infections in burn treatment[2] and arthroplasty[3], as well as to prevent bacteria

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colonization on prostheses [4], catheters [5], vascular grafts, dental materials[6], stainless steel materials[7], and human skin[8].

Because of their wide range of applications Synthesis of silver nanoparticles is of much interest to the researcher. Generally, nanoparticles are prepared by a variety of chemical and physical methods which are quite expensive and potentially hazardous to the environment which involve use of toxic chemicals that are responsible for various biological risks. In the search of cheaper and eco-compatible pathways for nanoparticles, scientist used microorganism[9,10] and plant extracts[11,12]. Green synthesis of nanoparticles has proven to be better methods due to slower kinetics, offer better manipulation, control over crystal growth and their stabilization. Greener synthesis provides advancement over traditionally used nanoparticles synthesis methods i.e. chemical[13,14] and physical method as it is cost effective, easily scaled up, environment friendly[15] or large scale synthesis and in this method there is no need to use toxic chemicals. Green synthesis of nanoparticles is a bottom up approach where the main reaction occurring is reduction.

Biogenic synthesis is useful not only because of its reduced environmental impact[16,17] compared with some of the physicochemical production methods, but also because it can be used to produce large quantities of nanoparticles that are free of contamination and have a well-defined size and morphology[18]. Biosynthetic routes can actually provide nanoparticles of a better defined size and morphology than some of the physicochemical methods of production[19]. The methods for obtaining nanoparticles using naturally occurring reagents such as vitamins, sugars, plant extracts, biodegradable polymers, and microorganism as reductants and capping agents could be considered attractive for nanotechnology. But among above mentioned reagents plant extract using leaf, root, stem, latex, resin, seed seems to be the best candidates and they are suitable for large scale "Green synthesis" of nanoparticles[20].

The genus *Dracocephalum* L. (Lamiaceae) consists of around 60 species distributed in the temperate regions of the Northern Hemisphere. In the flora of Iran, the genus is represented by

eight species, which are mainly distributed in the northern and central parts of the country, belonging to the Irano-Turanian phytogeographical region [21].

In the present study, *Dracocephalum lindbergii* was used for source of reducing agent. *Dracocephalum* extract show various biological effects, such as anticancer, antioxidant, antihypoxic, and immunomodulatory activities[22].

In this paper, we report on the biosynthesis of pure metallic nanoparticles of silver by the reduction of aqueous Ag⁺ ions with the aqueous extract of *Dracocephalum lindbergii*

2 – EXPERIMENTAL

2.1 -Preparation of dried biomass

10g of aerial parts of *Dracocephalum lindbergii* were washed thoroughly with double-distilled water and were then cut into small pieces. These finely cut pieces were then mixed with 100 mL doubled distilled water and this mixture was kept for boiling for a period of 15 min. After cooling, it was filtered through Whatman Filter Paper No.1. Filtrate placed at 4°C for further experiment.

2.2 - Biosynthesis of silver nanoparticles

1 mM aqueous solution of silver nitrate (AgNO₃) were prepared and used for the synthesis of silver nanoparticles. 10 mL of extract were taken and 100 mL of AgNO₃ solution was added to it. The colour change from pale green to dark brown due to surface plasmon resonance. This occurs due to the collective oscillation of the conduction electrons confined to metallic nanoparticles. They were incubated at room temperature for 24h. The colour change indicate the synthesis of silver nanoparticles. UV-visible spectra showed strong SPR band at 416 nm and thus indicating the formation of silver nanoparticles The silver nanoparticles (AgNPs) obtained by *Dracocephalum lindbergii* extract were centrifuged at 13,000 rpm for 25 min and subsequently dispersed in sterile distilled water to get rid of any uncoordinated biological materials.

2.3 - UV-vis spectra analysis

The bio reduction of Ag^+ in aqueous solution was monitored by periodic sampling of aliquots (0.2 ml) of the suspension, then diluting the samples with 2 ml deionized water and subsequently measuring UV–vis spectra of the resulting diluents. UV–vis spectroscopy analyses of silver nanoparticles produced were carried out as a function of bio reduction time at room temperature on ELICO UV spectrophotometers at a resolution of 1 nm.

2.4 - XRD analysis

X-ray diffraction (XRD) analysis of drop-coated films of silver nanoparticles in sample was prepared for the determination of the formation of Ag nanoparticle by an X'Pert Pro X-ray diffractometer (X' Pert High Score Plus program) operated at a voltage of 40 kV and a current of 30 mA with $\text{Cu K}\alpha$ radiation.

2.5 - Observation of silver particle size

XRD patterns were analyzed to determine peak intensity, position and width. Full width at half-maximum (FWHM) data was used with the Scherrer's formula to determine mean particle size. Scherrer's equation is given by

$$d = \frac{0.9\lambda}{\beta \cos \theta}$$

where d is the mean diameter of the nanoparticles, λ is wavelength of X-ray radiation source, β is the angular FWHM of the XRD peak at the diffraction angle θ [23].

2.6 - SEM observation of silver nanoparticles

The plant extract biomass after reaction spontaneously precipitated at the bottom of the tubes. After the precipitation, the suspension above the precipitate was sampled for SEM observation. SEM samples of the aqueous suspension of silver nanoparticles were fabricated by dropping the suspension onto clean electric Stubs and allowing water to completely evaporate. SEM observations were carried out on a ZEISS EVO 40 EP Electron microscope.

2.7 - EDX observation of silver nanoparticles

Energy dispersive X-ray spectrometers take advantage of the photon nature of light. In the X-ray range the energy of a single photon is just sufficient to produce a measurable voltage pulse X-ray, the output of an ultra-low noise preamplifier connected to the low noise are a statistical measure of the corresponding quantum energy. By digitally recording and counting a great number of such pulses with in a so called Multi Channel Analyzer, a complete image of the X-ray spectrum is building up almost simultaneously. This digital quantum counting technique makes the energy dispersive spectrometry exceedingly reliable. A semiconductor material is used to detect the x-rays together with processing electronics to analyses the spectrum. EDX observations were carried out by a Bruker LN2 free X-Flash 4010 SDD Detector and analytical Software was QUANTAX 200.

3 - RESULT AND DISCUSSION

3.1- Formation of the silver nanoparticles by reduction of the aqueous Ag^+ during exposure to the aqueous extract of *Dracocephalum lindbergii* were followed by UV–vis spectroscopy. It is well known that silver nanoparticles exhibit dark brown in water[24]. After 3h of the conversion process silver nanoparticle showed dark brown color, suggested the formation of silver nanoparticles in solution. These colors arise due to excitation of surface plasmon vibrations in the silver metal nanoparticles[25].

[Fig.1](#) shows the UV–vis spectra recorded from the aqueous silver nitrate and *Dracocephalum lindbergii* extract. It is observed that the silver surface plasmon resonance band occurs at 416 nm and steadily increases in intensity as a function of time of reaction without any shift in the peak wavelength. The frequency and width

of the surface plasmon absorption depends on the size and shape of the metal nanoparticles as well as on the dielectric constant of the metal itself and the surrounding medium[26,27]. It is generally recognized that UV–vis spectroscopy could be used to examine size- and shape-controlled nanoparticles in aqueous suspensions[28].

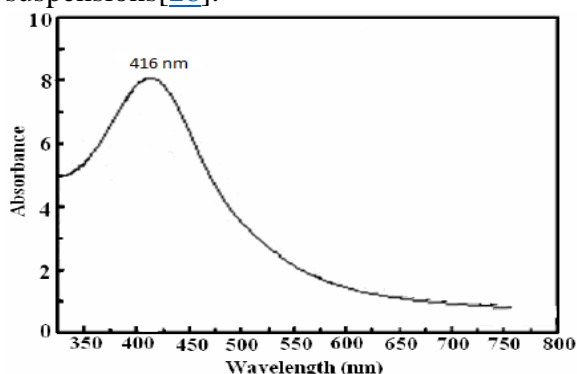


Fig. 1. UV–visible absorption spectra of reduction of silver ions to silver nanoparticles

Fig.2 shows representative SEM images recorded from drop-coated films of the silver nanoparticles synthesized by treating silver nitrate solution with *Dracocephalum lindbergii* extract. The silver nanoparticles formed were predominantly cubical with uniform shape. It is known that the shape of metal nanoparticles considerably change their optical and electronic properties[29].

XRD patterns (Fig.3) obtained for silver nanoparticles synthesized using *Dracocephalum lindbergii* extract in fig 4 show characteristic peaks (at $2\theta = 38$). The XRD pattern thus clearly shows that the silver nanoparticles are crystalline in nature. The sharpening of the peaks clearly indicates that the particles are in the nanoregime. The size of the silver nano crystallites as estimated from the FWHM of the maximum peak of silver using the Scherrer formula is between 5 and 6nm.

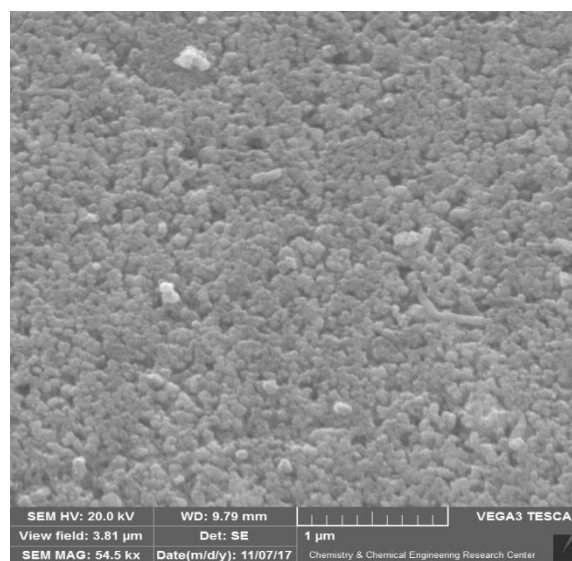


Fig. 2. SEM micrograph of Silver Nanoparticles from *Dracocephalum lindbergii* extract at 34.5 KX magnifications.

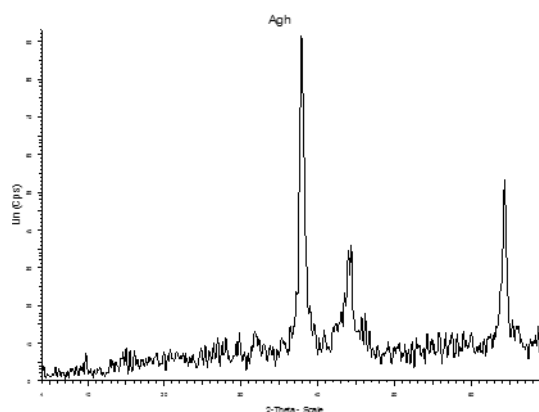


Fig. 1. XRD patterns recorded from drop-coated films on glass substrate of silver nanoparticles synthesized by treating *Dracocephalum lindbergii* extract with AgNO_3 aqueous solutions.

Analysis through Energy Dispersive X-ray (EDX) spectrometers confirmed the presence of elemental silver signal of the silver nanoparticles (Fig. 4). The vertical axis displays the number of x-ray counts whilst the horizontal axis displays energy in KeV. Identification lines for the major emission energies for silver (Ag) are displayed and these correspond with peaks in the spectrum, thus giving confidence that silver has been correctly identified.

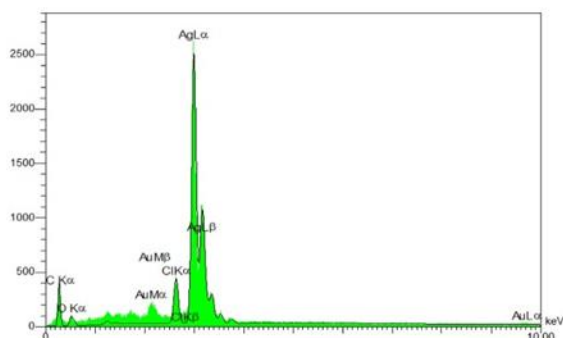


Fig 2. EDX spectrum recorded from a film, after formation of silver nanoparticles. Different X-ray emission peaks are labeled. 4 - Conclusion

In conclusion, it has been demonstrated that the extract of *Dracocephalum lindbergii* are capable of producing silver nanoparticles extracellularly and the silver nanoparticles are quite stable in solution. The phytochemical constituents which present in *Dracocephalum lindbergii* extract are the surface active molecules stabilizing the nanoparticles. Achievement of such rapid time scales for synthesis of metallic nanoparticles contributes to an increase in the efficiency of synthetic procedures using environmentally benign natural resources as an alternative to chemical synthesis protocols and low cost candidate as reductant for synthesizing silver nanoparticles.

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