Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

J P P Journal of Pharmacognosy and Phytochemistry

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2016; 5(1): 264-266
Received: 16-12-2015
Accepted: 18-01-2016

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Flavonoid content of *Anthemis tinctoria* and its application for green synthesis of silver nanoparticles

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Abstract
An aqueous extract of *Anthemis tinctoria* was used for green synthesis of silver nanoparticles. The extract of *Anthemis tinctoria* was prepared with maceration method and the extract was used for preparing silver nanoparticles in $AgNO_3$ aqueous solution. The related tests showed that the extract contains flavonoids. Synthesized silver nanoparticles were characterized by ultraviolet-visible spectroscopy, infra-red spectroscopy, X - ray diffraction spectroscopy and transmission electron microscopy. To confirm the formation of silver nanoparticles, the ultraviolet- visible absorption spectrum was obtained in 420-450 nm and showed that silver nanoparticles were produced. The average size of obtained silver nanoparticles was found to be average 34.77 nm in face center cubic crystalline structure.

Keywords: silver nanoparticles, *Anthemis tinctoria*, echo- friendly

Introduction
In recent years, flavonoids are important because of its nutritional and antioxidant properties. This paper, presents silver nanoparticles preparation by flavonoids and have advantages over methods with chemical agents. Several methods have been used to reduce Ag^+ to Ag^0 nanoparticles. Nanoparticles are prepared by chemical and physical methods which are quite expensive and hazardous to the environment which involve use of toxic chemicals that are responsible for various biological risks. The advantage of green syntheses over chemical and physical methods is: environment friendly, low cost and easily scaled up for large scale syntheses of, furthermore there is no need to use high temperature, pressure, energy and toxic chemicals [1]. Reduction by sodium citrate ($C_6H_7NaO_7$) and sodium borohydrate ($NaBH_4$) are such methods which using toxic and expensive compounds [2].
This green method was carried out in water, an echo-friendly solvent. Bio-organisms in plant extracts contain flavonoids and the reduction of Ag^+ by flavonoids gave Ag NPs. nanoparticles of silver (Ag-NPs) are used widely in optics [3], anti-HIV [4], chemical reaction catalyst [5], medical application [6], absorption of solar energy [7] and anti-bacterial agents [8].
Some Silver nanoparticles had been Synthesysed in spherical shape and 50-100 nm size with the extract of *Alternathera denlate* [9].
Some scientist's synthesized spherical and feck shape silver nanoparticles in 10-30 nm size with the extract of *Vitex negundo* [10]. Rajakumar G, obtained triangles silver nanoparticles from *Eclipta prostrate* extract, with average size 35-60 nm [11].
Successful determination of compounds from plants is depended on the type of solvent used in extraction. Properties of a good solvent includes low toxicity and evaporation at low heat mixture of alcohol and water is suitable for presence of amounts of polyphenols in an aqueous extract.

Experimental
Plant Material and preparation of extract
The aerial parts (leaves, stem and flowers) of *Anthemis tinctoria* were collected from Khalkhal, Ardabil province, Iran in May 2015. The plant material was washed by deionized water and dried in shade under air stream in ambient temperature (25 °C). Dried plant was powdered mechanically by a blender.
In this method (maceration method), 30 g powdered plant is kept in contact with mixture of ethanol and water in 20: 80 ratio, for 48 hours. Ratio of solvent to sample has been 10: 1 (v/w). After that, the extract was placed in rotary evaporator with vaccum source to remove solvent rapidly from extract.

One ml of ethanol extract of the studied plant was mixed with 0.5 ml of hydrochloric acid and magnesium metal and a reddish color was indicating the presence of flavonoids.

Silver nano particles synthesis

10 mL of *Anthemis tinctoria* aqueous extract was added to 90 mL of 1 mM solution of silver nitrate and kept in ambient temperature. After 10 min a brownish solution can be seen due to reduction of Ag^+ cations to Ag by *Anthemis tinctoria* extract.

All reagents and materials were purchased from Merck chemicals.

Nanoparticle characterization

UV-VIS spectroscopy: UV-VIS spectroscopy was used to ascertain the formation of Ag-NPs in aqueous solution at the wavelength range from 300-700 nm after different period of times (0- 48hours). The UV-VIS analyses were performed using a Varian Car 300 spectrophotometer. The band for Ag-NPs appears in the 420-450 nm regions. (Figure 1)

FT-IR Spectroscopy: The Ag-NPs synthesized solution was centrifuged at 12000 rpm for 15 min at ambient temperature. The resulting pellet was redisposed in deionized water. The water extract and Ag-NPs pellet was dried by freeze drier (CHRIST alpha1-4 LD plus- Germany) and subjected to FT-IR spectroscopy (Thermo Nicolet Nexus 870- USA) using KBr (Sigma-Aldrich) at 400-4000 cm^{-1} at a resolution of 4 cm^{-1}. (Figure 2A & B)

X-Ray Diffraction (XRD) analysis: The XRD studies were performed using a Sifert XRD 3003 PD (Germany) Equipped with a Cu-Kα_1 radiation with λ of 1.54 A° in the region of 2θ from 20° to 90°, using a dried thin film of synthesized Ag-NPs. XRD pattern of phytochemical synthesized Ag-NPs show four peaks at 27.8375°, 32.227°, 38.1016° and 47.4979°. (Figure 4)

Result and discussion

UV-VIS spectral, FT-IR spectrum and X-Ray Diffraction analysis, were used to confirmation of Ag-NPs formation.

As shown in figure 2, the Ag-NPs were formed 1 h after adding the extract to $AgNO_3$ solution, the mixture color was changed to brown because of excitation of surface plasmon vibrations in the Ag-NPs. The brownish color of Ag-NPs is related to SPR arising due to the grsoup of free conduction electrons induced by an interacting electromagnetic field. The SPR band for Ag-NPs normally appears in the 420-500 nm regions of spectra.

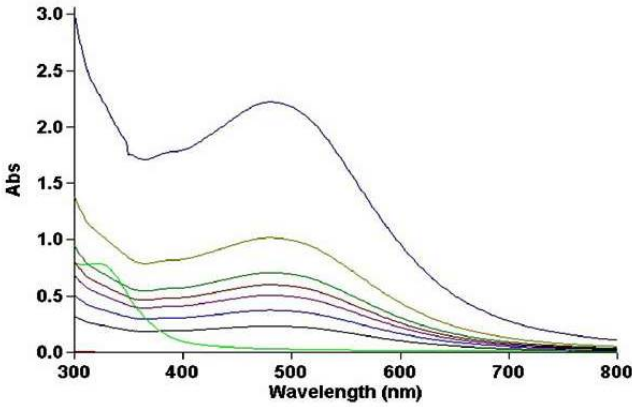

Fig 1: UV- Vis spectrum of Ag-NPs in period of times (0-48 h).

The 3416 cm^{-1} strong peak in FT-IR spectra of aqueous extract of *Anthemis tinctoria,* shows the alcoholic and phenolic O-H groups (fig 2 .A), which disappear after phytochemical reduction of Ag^+ ions due to responsibility of hydroxyl groups in reduction of Ag^+ ions(fig 2 .B) . The 2922cm^{-1}, 1642cm^{-1} and 1389 cm^{-1} absorption are related to alkyl C-C stretch vibration, C=O bond with a benzene ring conjugated with a C=C bond and bending vibration of C-OH, respectively.

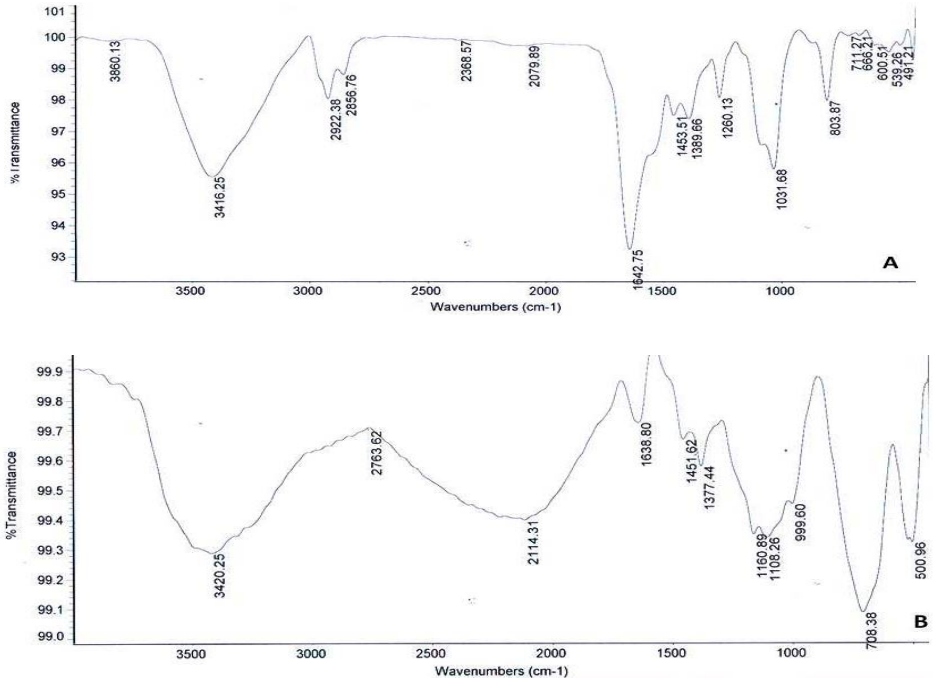

Fig 2: FT-IR spectra of *Anthemis tinctoria* before (A) and after (B) reduction of Ag^+ ions. The FESEM images of nanosilvers are shown in Fig 3. It was found that the particles are nanometer-sized spheres.

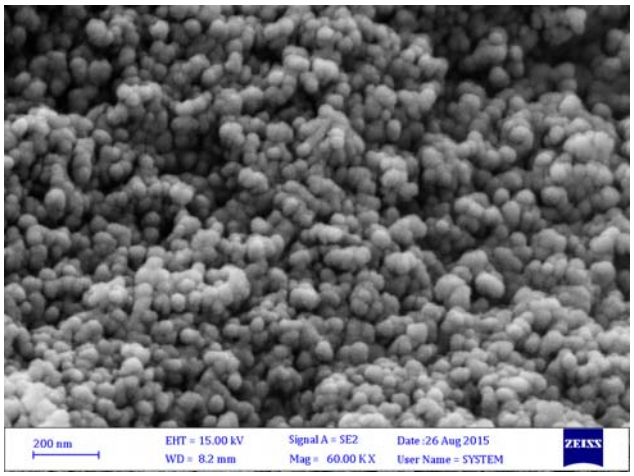

Fig 3: F.E.S.E.M, image of phytochemical synthesized Ag-NPs using the extract

The crystallographic structure of the sample was confirmed by a powder X-ray diffraction spectroscopy.

XRD spectrum of Ag-NPs synthesized shows four intensive peaks at 2θ of 27.8375°, 32.227°, 38.1016° and 47.4979 ° related to (111), (200), (311) and (222) HKL values, respectively, which shown the Ag-NPs were appeared in a face center cubic (fcc) lattice system. The average size of Ag-NPs was calculated to 34.77 nm by Debye-Scherer's equation.

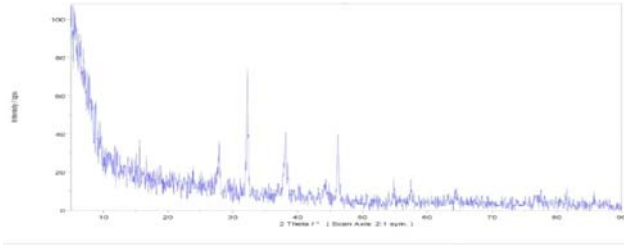

Fig 4: XRD pattern of phytochemical synthesized Ag-NPs

The obtained data were confirmed these nanoparticles are in face center cubic lattice system with the average size 34.77 nm.

Conclusion

Nature has elegant and ingenious ways of creating the most efficient miniaturized functional materials. An increasing awareness towards green chemistry and use of green route for synthesis of metal nanoparticles. Many reports have been published about the syntheses of silver nanoparticles using plant extracts like those as already discussed. This study shows that Silver nanoparticles were synthesis using water extract of *Anthemis tinctoria* in to replace hazardous chemical reducing agent with an eco- friendly reducing agent. The synthesized Ag nanoparticles were characterized by UV-VIS, FT-IR, XRD and TEM. The data were confirmed that these nanoparticles are in face center cubic (fcc) system with the average size 34.77 nm. . There is a significant variation in chemical compositions of plant extract of same species when it collected from different parts of world and may lead to different results in different laboratories

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