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RESEARCH PAPER

Biosynthesis of Gold Nanoparticles Using Aqueous Extract of Stem of Periploca Aphylla Plant

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ABSTRACT

This present work reports an ecofriendly approach for the synthesis of gold nanoparticles (Au NPs) using aqueous stem extract of Periploca aphylla as a reducing and stabilizing agent, has been discussed. This approach is simple, cost-effective and stable for a long time, reproducible at room temperature and in an eco-friendly manner to obtain a self-assembly of Au NPs. Two parameters were optimised for the fabrication of gold nanoparticles including the pH and contact time. The resulting nanoparticles are characterized using UV–vis, TEM, XRD and FT-IR spectroscopic techniques. UV–visible spectra of the aqueous medium containing gold nanoparticles showed a surface plasmon resonance peak at 535 nm. Uniform spherical shapes were observed for biosynthesized Au NPs within range of 25–30 nm by transmission electron microscopy. XRD results confirmed the presence of gold nanoparticles with face centered cubic structure. FT-IR analysis was performed to analyze the biomolecules responsible for the reduction of Au NPs.

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INTRODUCTION

Application of nano-scale materials, ranging from 1–100 nm, is an emerging area of nanotechnology. Nanoparticles exhibit completely new and improved properties based on specific characteristics such as size, distribution and morphology, if compared with larger particles of the bulk material they are made of. Nanomaterials have a long list of applicability in improving the human life and its environment [1, 2]. Nanoparticles present a higher surface to volume ratio with decreasing size of nanoparticles. As specific surface area of nanoparticles is increased, their biological effectiveness can increase due to the increase in surface energy [3].

Different methods are used for the synthesis of gold nanoparticles. A number of methods including physical and chemical processes [4-10], electrochemical reduction [11, 12] photochemical reduction [13, 14] and heat evaporation [15, 16] for synthesis of metal nanoparticles were developed considering the real life application of nanoparticles in the area of medicine [17], catalysis [18] detection [19], drug delivery [20] etc. Chemical synthesis methods lead to some chemically toxic substances being absorbed on the surface that may have negative effects on medical applications, limiting them [21]. Therefore, the need for the development of a clean, reliable, biocompatible, benign, and eco-friendly process to synthesize nanoparticles without using toxic chemicals leads to turning researchers toward “green” chemistry and bioprocesses [22]. Biological methods for nanoparticle synthesis using microorganisms,

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enzymes, and plants or plant extracts have been suggested as possible ecofriendly alternatives to chemical and physical methods [23, 24]. Recently the studies started under green chemistry for the search of benign methods for the development nanoparticles and searching antibacterial, antioxidant, and antitumor activity of natural products. Biosynthetic processes have received much attention as a viable alternative for the development of metal nanoparticles where plant extract is used for the synthesis of nanoparticles without any chemical ingredients [25, 26]. Bioinspired synthesis of nanoparticles has advantages over chemical and physical methods as it is a cost effective and environment friendly and does not need to use high pressure, energy, temperature and toxic chemicals. The main benefit of using the green chemistry route for the fabrication of gold nanoparticles is to fabricate a material that will be compatible for pharmaceutical and other biomedical applications since the toxic chemicals are not used during the fabrication of nanoparticles [27]. Moreover, using a plant extract can decrease the cost of preparation and eliminate the need for any special culture preparation and isolation technique compared with using bacteria and fungi for fabrication of nanoparticles [28].

Using plants for nanoparticle synthesis can be advantageous over other biological processes because it eliminates the elaborate process of maintaining cell cultures and can also be suitably scaled up for large-scale nanoparticle synthesis [29]. Moreover, plants have been used to fabricate nanoparticles since they are readily available, safe to handle, and they have a broad variability of metabolites that can aid in reduction [30].

MATERIALS AND METHODS

Preparation of plant extract

Periploca aphylla Stem shown in Fig. 1 was collected from Delfard region in Iran. Fresh Stems were used for the extraction of the active components. The Stems were shade dried for 2–3 weeks at room temperature and then powdered. Briefly, 1 g of Stem powder was weighed and mixed in 100 mL of de-ionized water and boiled at 30°C for 30 min . The obtained extraction was filtered using Whatman No. 1 filter paper and filtered to get the extract. then,the collected extract was centrifuged, in order to eliminate any biomaterials that could interfere with binding of Au ion to the biomass or construction of the nanoparticles. The filtrate is used as reducing agent and stabilizer.

Synthesis of gold nanoparticles

Typical synthesis process of gold nanoparticles, 2 mL of Periploca aphylla extract is added to avigorously stirred 4 mL aqueous solution of $HAuCl_4.3H_2O$ 1mM and the final volume was adjusted to 50 mL with deionised water at room temperature and stirring continued for 30 min. To initialize the reduction of gold ions, the pH of the solution was adjusted to 4. Reduction takes place rapidly less than 1 min as indicated by stable light violet colour of the solution. At the end of reaction, the Au NPs solution thus obtained was purified by repeating the centrifugation thrice at 10000 rpm for 30 min at 4 ◦C followed by redispersion of the pellet in deionised water and studied by adjusting the effects of pH and time parameter on the synthesis rate and amonth of the Au NPs were monitored by UV-vis Spectrophotometer.

Characterization of Au NPs

The synthesized gold nanoparticles were subjected to UV–visible analysis in the wavelength range of 200–800 nm using Shimadzu spectrophotometer (Model UV-1800) operating at a resolution of 1 nm. A transmission electron microscope (EM10C) measurements were carried out to bring out the morphology of the green synthesized nanoparticles in terms of size and shape. Crystalline Au NPs were determined

Fig. 1. Photograph of P.aphylla used in the biosynthesis.

by Brucker D8advance X-ray diffraction (XRD) analysis using Cu K α radiation(ƛ = 1.54060˚A). The purified Au NPs were examined for the presence of biomolecules using FTIR analysis. The spectrum obtained from the dried sample was recorded on FTIR spectrum (Perkin–Elmer). A Metrohm model pH lab 827 meter was used for pH measurements.

RESULT AND DISCUSSION

Adsorption spectra of synthesized nanoparticles

Fig. 2 shows the absorption spectra of the synthesized gold nanoparticles recorded by spectrophotometry in the visible range. As can be seen, the maximum wavelength of it, is in 535 nm. 4 mL aqueous solution of $AuCl_4$ (1 mM) and 2 mL of Periploca aphylla extract was used for this experiment and spectrum was recorded after 30 min of start of reaction.

Effect of pH on Au nanoparticles synthesis

pH play an important role in the nanoparticles synthesis. This factor induces the reactivity of leaf extract with gold ions. The effect of pH on the reduction of gold ion and synthesis of gold nanoparticles by Periploca aphylla extract was investigated in the range 2 to 7. Again, the reaction mixture was consisting of 4 mL aqueous solution of $AuCl_4$ (1 mM) and 2 mL of Periploca aphylla extract. pH of the solutions were adjusted by drop-wise addition of either 0.1 N NaOH or 0.1 N HCl and monitored by a pH meter. After any change in the pH, adsorption of the solution was measured at wavelength 535 nm, after 30 min. Results are summarized in Fig. 3. By increasing pH up to 4, absorption increases and after then it became constant, which means the yield of nanoparticle synthesis reached to its maximum in this pH and beyond. Therefore this pH was selected as the optimum point. Moreover, it was noted that at pH 4, the peak at 535 nm is quite sharp which means that gold nanoparticles are formed mainly in a spherical shape [31, 32]. Several results are reported pH is plays an important role in shape and size control synthesis process of gold nanoparticles[33].

The present investigation indicates acidic pH is more suitable for synthesis of gold nanoparticles using Periploca aphylla of extract of stem. but, Pandey et al. [38] reported that the gold nanoparticles show maximum stability at the pH 10 by aqueous extract of Momordica charantia.

Effect of time on Au nanoparticles synthesis

The effect of the incubation time was studied by monitoring the absorption spectra of gold nanoparticles formed in the reaction media at different durations every 5 min at wavelenght 535 nm after adding 2ml of Periploca aphylla extract to 4 ml aqueous solution of $HAuCl_4.3H2O$ 1mM which was adjusted to pH 4 and the reaction was monitored using UV/Vis spectrophotometer from 0 to 30 min in order to optimise the time

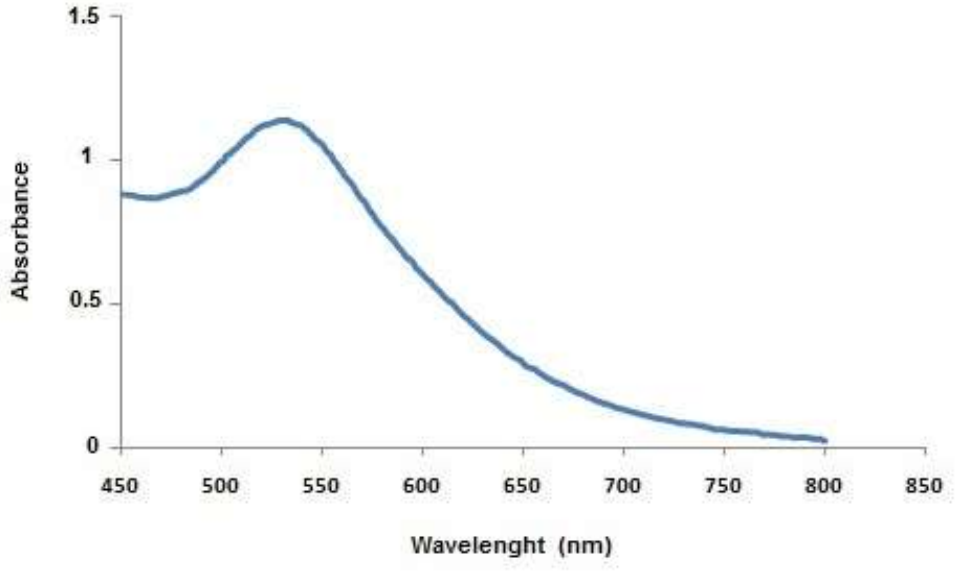

Fig. 2. UV visible spectra of Au nanoparticles synthesized by stem extract of P.aphylla

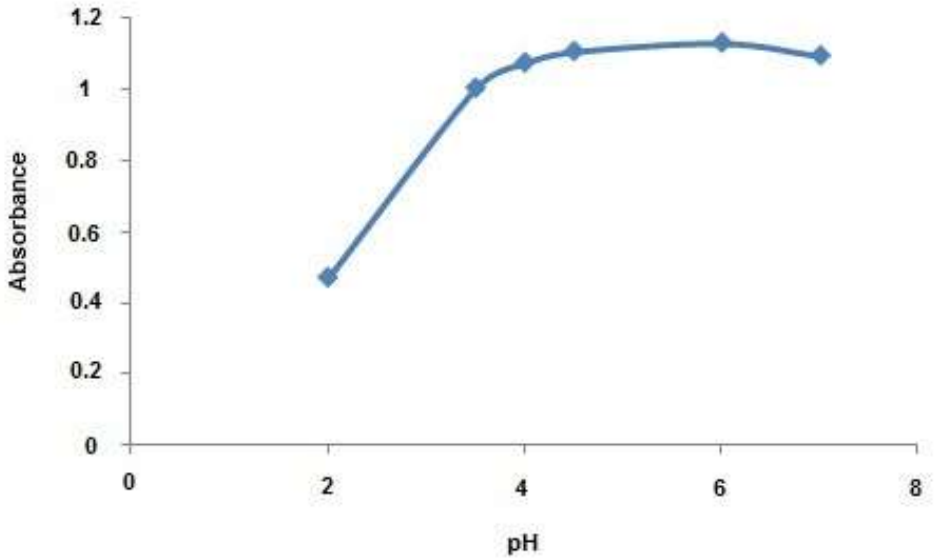

Fig. 3. The effect of pH on synthesis of Au nanoparticles by tem extract of P.aphylla at wavelenght 535 nm

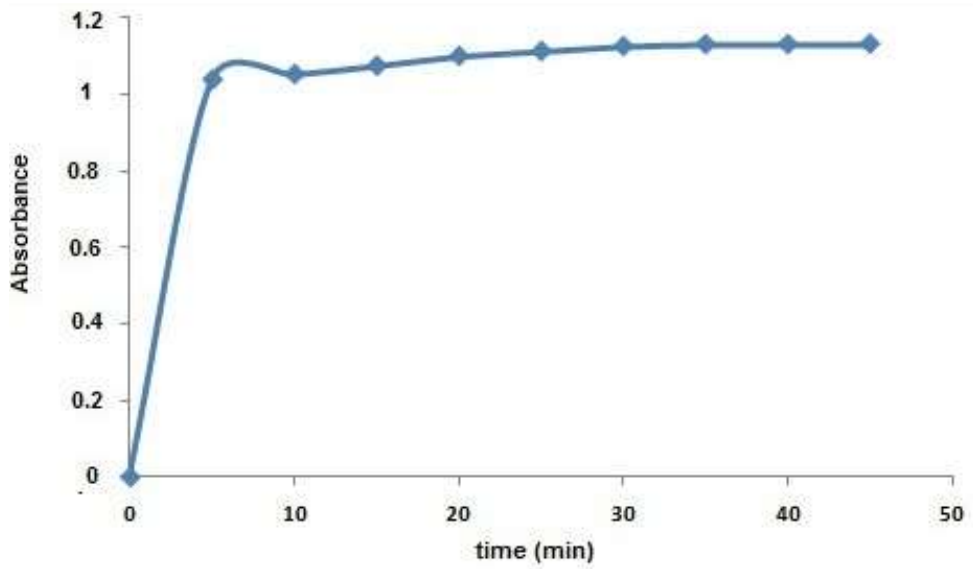

Fig. 4. The effect of time on synthesis of Au nanoparticles by P.aphylla at wavelenght 535 nm

required for the reaction completion. Fig. 4 shows the UV-Vis spectra of reduction of gold ions to gold nanoparticles using Periploca aphylla extract at different reaction times. It was found that when the incubation time was less than 1 min, gold nanoparticles appeared. Moreover, it was observed that by increasing the time of reaction, the absorbance intensity and amount of formed GNPs at wavelenght 535 nm increased within 30min ,but after that only a slight variation can be observed.. From the spectrum, it was found that the optimum incubation time for the completion of reaction in this study was 30 min. In addition, it was observed that increasing the incubation time to more than 30 min did not increase the absorption significantly, which indicates the stability of biosynthesized nanoparticles, whereas synthesis from other plant extracts takes 2 to 4 h. When employing microorganisms, the formation of gold nanoparticles is a slow process

(reaction time between 24 and 120 h).[35]. This stability is resulting from the peresence of organic compounds that are along with nanoparticles that those existence ascertained by helping FT-IR spectra.

transmition Electron Microscope (TEM)

In addition to UV spectroscopy which confirms reduction and shape of gold ions to gold nanoparticles. The size, shape and distribution of synthesized gold nanoparticles were characterized by Transmition Electron Microscope. TEM image of the GNPs synthesised at pH 4 shows the presence of spherical monodispersed nanoparticles ranging from 25 to 30 nm and has an average size of about 27 nm (Fig. 5).

X-ray diffraction

X-ray Diffraction pattern was recorded for the

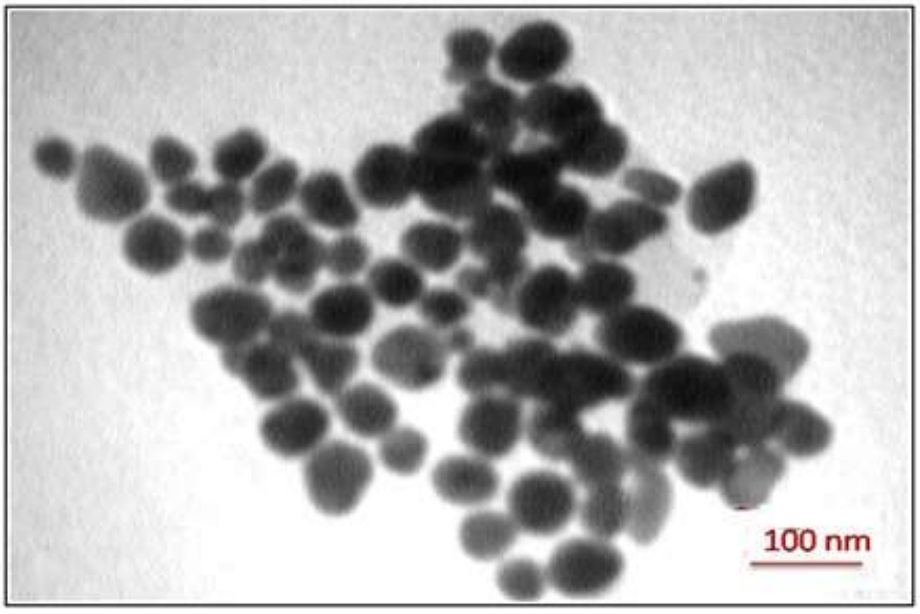

Fig. 5. TEM image of gold nanoparticles synthesized by Stem extract of P.aphylla

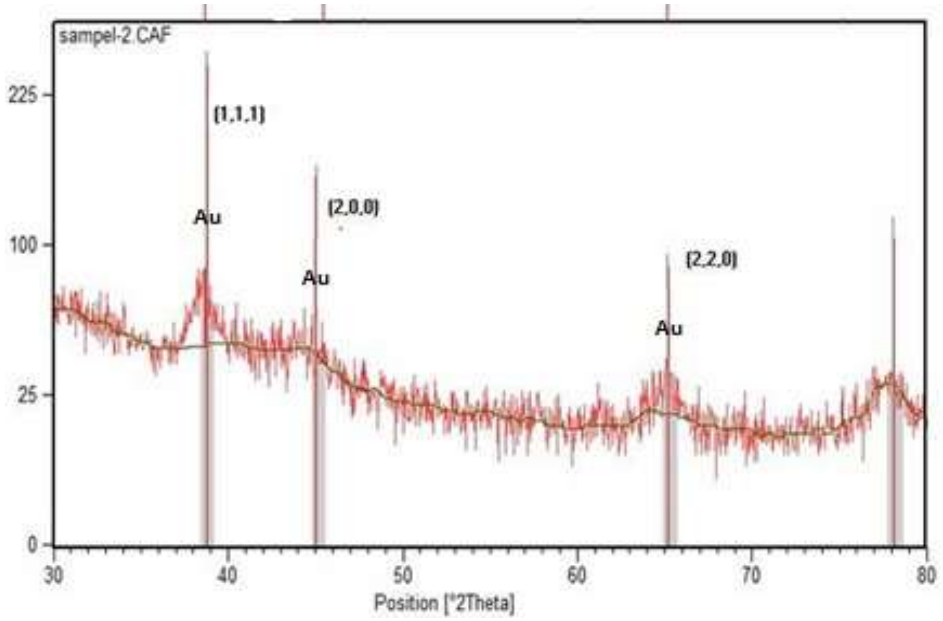

Fig. 6. XRD pattern of Aunanoparticles synthesized by Stem extract of P. aphylla

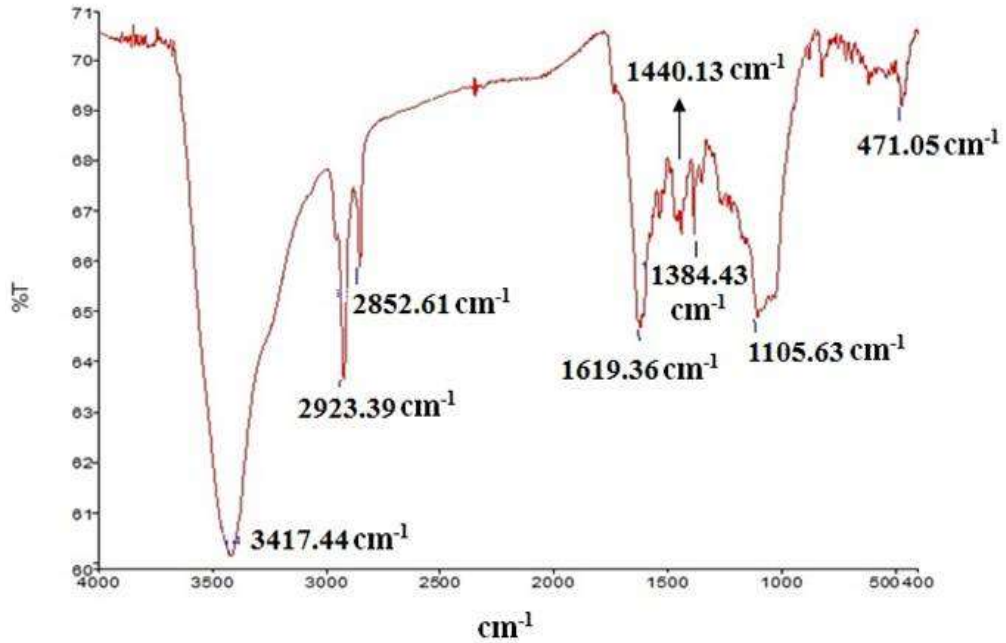

Fig. 7. FTIR spectra of Au nanoparticles synthesized by stem extract of P.aphylla

synthesized Au NPs (Fig. 6). The diffraction peak at 2θ values 38.68°, 45.48°, 65.15° assigned to the (1 1 1), (2 0 0), (2 2 0) of lattice plane of face centered cubic (fcc) for Au nanoparticle, respectively. The average size of nanoparticles was calculated using Debye-Scherrer's equation [39] by determining the width of the peaks and found to be 28, 22 and 32 nm respectively which is fairly in agreement with the TEM measurement.

Fourier transform infrared spectroscopy

FTIR measurement was carried out to identify the possible biomolecules in extract of stem of Periploca aphylla responsible for capping leading to efficient stabilization of the gold nanoparticles. Prominent IR bands (Fig. 7) are observed at 3417.44, 2923.39, 2852.61, 1619.36, 1440.13 and 1105.63 cm^{-1}. Most of the FT-IR bands are characteristic of flavonoids and tepenoids present in the plant extract. The sharp bands at 3417.44, and 2914, 2847 cm^{-1} arise from –N-H and C–H stretching modes respectively. The absorption bands located at 1440.13, 1384 and 1105.63 cm^{-1} may be attributed to –C-N, -C-O-C and –C–O stretching modes. The vibrational bands corresponding to bonds such as-N-H, –C=C, –C–O, –C–O–C and –C–N are derived from the water soluble compounds such as flavonoids, terpenoids and porotein present in extract of Stem of periploca aphylla plant. The presence of these biomolecules in the extract could be responsible for the reduction of gold ions, formation and stabilization of the biosynthesized nanoparticles.

CONCLUSION

The spanking new and simple method for biosynthesis of Au NPs by P. aphylla extract offers a valuable contribution in the area of green synthesis and nanotechnology without adding using any toxic reagents and thus has potential for use in biomedical and agricultural applications. The nanoparticles were characterized by UV–Vis Spectrophotometer, TEM, XRD and FTIR measurements. Rapid synthesis and amount of gold nanoparticles was attained maximum when altering such pH effective parameters. The optimum conditions for biosynthesis of Au NPs are observed to be at pH 4 and incubation time 30 min. The NPs synthesized were predominantly spherical with size of 25-30 nm P.aphylla extract was acts as a capping agent and reducing agent in the nanoparticles synthesis. It is a better source for the rapid synthesis of gold nanoparticles . The synthesis of Au NPs by P.aphylla extracts indicated that flavonoids, terpenoids and porotein compounds could be responsible for the reduction of gold ions, formation and stabilization of the biosynthesized nanoparticles. This green method is a single step process, economic viability, non-toxic, ecofriendly and rapid production of

nanoparticles.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this manuscript.

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