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Brown seaweed-mediated biosynthesis of gold nanoparticles

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ORIGINAL RESEARCH ARTICLE

ABSTRACT

In the present study, we investigated biosynthesis of gold nanoparticles using the extracts of brown seaweed, *Turbinaria ornata*. The formation of gold nanoparticles was characterized using the UV-Visible spectroscopy, Fourier transform infrared (FTIR) spectroscopy, X-ray diffraction (XRD), scanning electron microscopy (SEM) and energy dispersive X-ray (EDX) spectroscopy. The synthesized gold nanoparticles showed distinct surface plasmon peaks at 525 nm and also the color of the reaction mixture gradually changed from brown to ruby red. Comparing the FTIR spectra of seaweed extract and synthesized gold nanoparticles, significant changes were observed in phenolic groups, alkynes, amide and esters. The XRD analysis showed intense peaks corresponding to (111), (200), (220), (311) and (222) Bragg's reflection based on the face centered cubic structure of gold nanoparticles. Elemental gold was also confirmed by the EDX analysis. The SEM images illustrate that all the gold nanoparticles were well separated and there were no aggregations. Gold nanoparticles showed spherical morphology with average particle size of 7 – 11 nm.

KEYWORDS
bioreduction; biosorption; biosynthesis; gold; green synthesis; nanoparticles

1. INTRODUCTION

Nanoparticles are of great interest owing to their extremely small size and high surface to volume ratio, which alter their physical and chemical properties (such as sterical properties, catalytic activity, thermal and electrical conductivity, optical absorption and melting point) compared to bulk of the same chemical composition (Iravani et al., 2014). Due to these beneficial properties, nanomaterials have found potential applications in catalysis, photonics, electronics, chemical sensing and imaging, information storage, environmental remediation, drug delivery, biological labelling, cosmetics, biomedical, mechanics, optics, space industries, chemical industries, energy science, light emitters, single electron transistors and nonlinear optical devices (Daniel and Astruc, 2004; Guo and Wang, 2007; Huang et al., 2007; Ahmed et al., 2015). Generally, nanoparticles are synthesized through three different kinds of methods: physical, chemical, and biological (Sastry et al., 2003; Shankar et al., 2004; Chandran et al., 2006; Rai et al., 2006; Birla et al., 2009). Of these, biological methods are often regarded as clean, biocompatible, non-toxic and eco-friendly methods for nanoparticles synthesis.

The biological synthesis of nanoparticles are being carried out using different biomaterials such as bacteria (Hulkoti and Taranath, 2014), fungi (Dhillon et al., 2012), yeast (Moghaddam et al., 2015), virus (Dujardin et al., 2003), microalgae (Schrofel et al., 2011), macroalgae (Singaravelu et al., 2007) and plant biomass/extract (Akhtar et al., 2013). Of various biological materials, seaweed possesses several inert advantages over other microscopic organisms in nanoparticle synthesis. The advantages of seaweed-mediated biosynthesis over other biomaterials include: (i) easy availability; (ii) one-step simple process; (iii) cost-effective; (iv) elimination of elaborate maintenance of cell cultures; (v) safe to handle; (vi) rapid rate of synthesis; (vii) more environmental friendly and

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(viii) composed of various metabolites that may aid in reduction. However, there is only very little literature supporting the use of algae in nanoparticle synthesis. The brown marine algae (*Sargassum wightii*) showed potential to synthesize Au nanoparticles extra-cellularly in the size range of 8 – 12 nm. Other brown seaweeds such as *Turbinaria conoides* (Rajeshkumar et al., 2013) and *Fucus vesiculosus* (Mata et al., 2009) as well as green seaweeds (Sangeetha et al., 2013) and red seaweeds (Priyadharshini et al., 2014) also investigated for nanoparticle synthesis. Thus, the objective of the present manuscript is to explore potential of brown marine alga (*Turbinaria ornata*) to biosynthesize gold nanoparticles. *Turbinaria ornata* is a very common brown alga found throughout the Pacific and Indian Ocean. It is known for its rigidity but is believed to have no commercial importance. Gold nanoparticles was selected for the present study as it found variety of applications in nanoelectronics, biomedical, nonlinear optics, nanodevices and catalysis (Huo and Worden, 2007) owing to its unique properties.

2. MATERIALS AND METHODS

2.1. Chemicals and algae

Stock Au(III) solution was prepared using chloroauric acid ($HAuCl_4$), purchased from Sigma-Aldrich (India).

Turbinaria ornata samples were collected on the beaches of Mandapam (9°16'47"N 79°7'12"E) region of Tamil Nadu, India. They were subsequently sun-dried on the site itself. In the laboratory, around 5 g of seaweed sample was crushed using motor and pestle. The seaweed pastes were then mixed with each 50 mL of distilled water. The extract was filtered using whatman filter paper and the filtrate was used for further experiments.

2.2. Experimental procedure

For synthesis of gold nanoparticles, 25 mL of *T. ornata* extract was contacted with 25 mL fixed volume of 1 mM $HAuCl_4$ solution and then stirred at 50 rpm for 1 h in a magnetic stirrer at room temperature (32±1 °C). The appearance of ruby red color indicates the formation of gold nanoparticles. Control (1 mM $HAuCl_4$) solution amounting 50 mL without seaweed extract was also run along with the experimental flask. Sample of 1 mL was withdrawn at different time intervals and characterized using different instruments.

2.3. Analytical techniques

UV-visible spectra were recorded using Shimadzu UV-1800 instrument in the range 300-800 nm. Synthesized gold nanoparticles were centrifuged at 10,000 rpm for 30 min and the resulting pellet was washed with distilled water and dried for further characterization. Fourier transform infrared (FTIR) spectra of dried gold nanoparticles and extracts were recorded using IR Perkin Elmer Spectrum1 spectrometer between 4000 and 600 cm-1 using KBr pellet procedure. The crystal nature of the newly formed nanoparticles was recorded using X-ray diffractometer (XRD) with CuKα radiation (λ=0.15 nm) in the range of 2θ- 20˚ to 80˚ (BRUKER AXS, D8 Discover). The morphology, size and shape of Au nanoparticles were analyzed by scanning electron microscopy (SEM). The sample was spread on copper gird and then images were taken (Hitachi-S4800).

3. RESULTS AND DISCUSSION

Gold nanoparticle formation was monitored visually (Figure 1) and by UV–Visible spectroscopy (Figure 2). On contacting $HAuCl_4$ solution with *T. ornata* extract, the color of the reaction mixtures gradually changed from brown to ruby red (Figure 1) in 4 h. This color change indicates the generation of gold nanoparticles and it was due to excitation of surface plasmon vibrations in the metal nanoparticles.

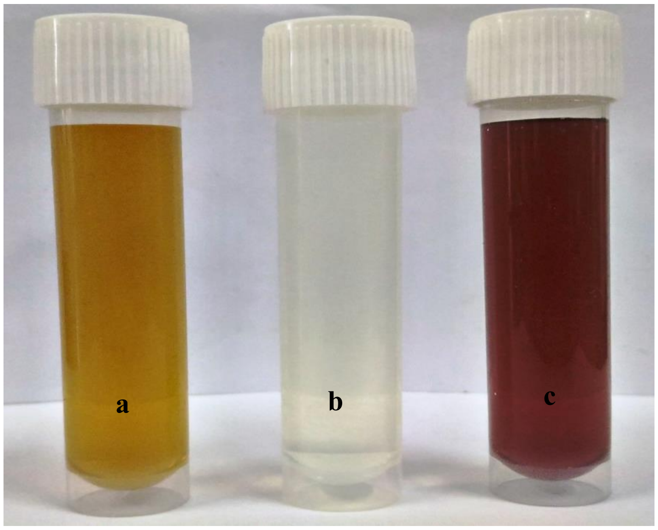

Figure 1. Photograph of vials containing a) seaweed extract of *T. ornata* b) Auric chloride solution c) Gold nanoparticles solution.

UV–visible spectrophotometry analysis is usually the first technique used in characterization of metallic nanoparticles and several authors used this technique to confirm the formation of nanoparticles (Kumar and Yadav, 2009; Noruzi, 2015). UV–visible spectral analysis was performed to confirm the formation and stability of the biosynthesized gold nanoparticles using the extract of *T. ornata* (Figure 2). A band observed in UV–visible spectrum corresponding to the surface plasmon resonance (SPR) occurs at 525 nm, which clearly indicates the formation of gold nanoparticles in solution. This maximum peak is a well-known characteristic of Au nanoparticles, and it is a phenomenon that has been reported in other biological systems (Narayanan and Sakthivel, 2008; Inbakandan et al., 2010). Narayanan and Sakthivel (2008) observed that addition of coriander leaf extract to aqueous HAuCl4 resulted in the color change to pink-ruby red after 12 h of reaction due to the production of Au nanoparticles. Similarly, Ashokkumar et al. (2014) confirmed the formation of gold nanoparticles by observing the color change of reaction mixture from brown to ruby red and surface plasmon resonance centered at 535 nm. The exact position of absorbance depends on a number of factors such as the dielectric constant of the medium and size of the particle. It is observed that the crystallite shape of the biosynthesized gold nanoparticles dependent on the Au^+ ion in solution and the biomolecules or factors from the extract of *T. ornata*.

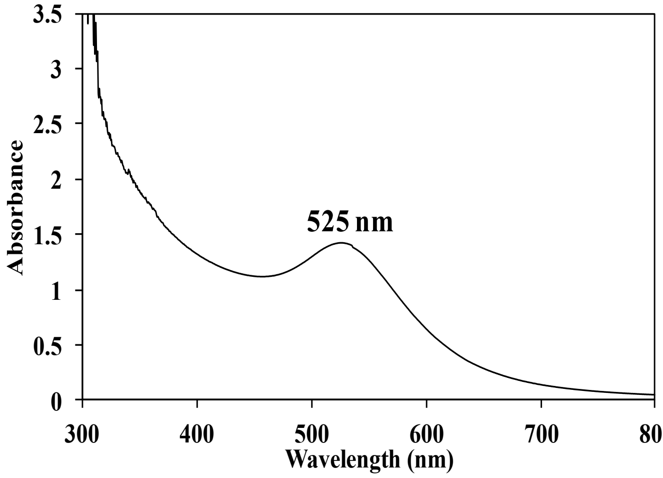

Figure 2. UV–visible spectra of synthesized gold nanoparticles using seaweed extract of *T. ornata*.

FTIR spectra of *T. ornata* seaweed extract and synthesized gold nanoparticles using *T. ornata* are presented in Figure 3. The sample of *T. ornata* seaweed demonstrates peaks at 3428, 2929, 2143, 1603, 1406, 1306, 1257, 1078 and 1037 cm^{-1}. FTIR spectrum of Au nanoparticles synthesized using *T. ornata*, showed distinct peaks in the range of 3407, 2926, 2171, 1634, 1404, 1256 and 1066 cm^{-1} (Figure 3). Comparing both the FTIR spectrum, significant changes were observed in phenolic groups, alkynes, amide and esters. The above results indicate the reduction and stabilization of gold nanoparticles. The FTIR peaks of phenolic and ester groups of *T. ornata* seaweed extract were shifted higher to lower wavelength in synthesized gold nanoparticles. This suggests the loss of the -OH intramolecular hydrogen bond functional groups may responsible for the reduction of Au^{3+} to Au^0 (Dubey et al., 2010; Priyadarshini et al., 2013). The alkynes and amide groups were shifted lower to higher wavelength these functional groups capping and stabilizing molecules of synthesized nanoparticles. Islam et al. (2015) reported the potential of leaf galls extract of *Pistacia integerrima* to reduce ions to Au nanoparticles. Through FTIR spectra, the authors determined that hydroxyl and carboxylic acid groups of polyphenols were responsible for the reduction.

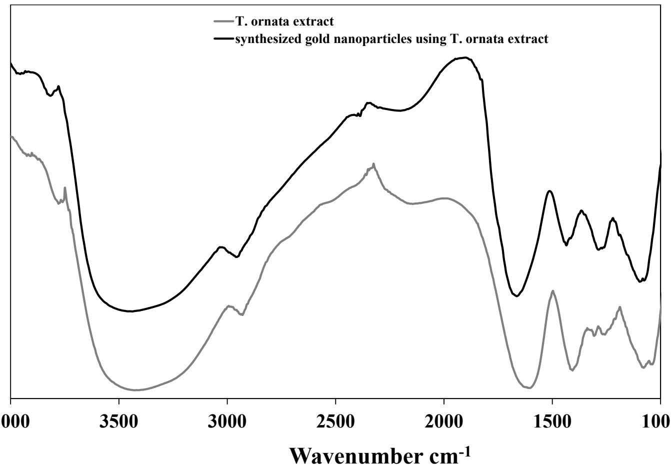

Figure 3. FTIR spectra of *T. ornata* extract and synthesized gold nanoparticles using *T. ornata* extract.

The X-ray diffraction (XRD) technique is used to study structural information about crystalline metallic nanoparticle. The energetic X-rays can penetrate deep into the materials and provide information about the bulk structure (Huang et al., 2007). XRD analysis showed intense peaks corresponding to (111), (200), (220), (311) and (222) Bragg's reflection based on the face centered cubic structure of Au nanoparticles (Figure 4). The broadening of Bragg's peaks indicates the formation of nanoparticle and its crystalline nature. The mean size of gold nanoparticles was calculated using the Debye–Scherrer's equation by determining the width of the (111) Bragg reflection (Borchert et al., 2005). The size of the nanoparticles was thus determined to be about 9 nm. There is no other peak in XRD pattern, which confirmed the stability and high purity of gold nanoparticles. Gao et al. (2014) observed that XRD pattern of newly synthesized Au nanoparticles showed intense peaks of (111), (200), (220), (311) and (222), which confirmed the monophasic nature of pure Au with face centered cubic symmetry.

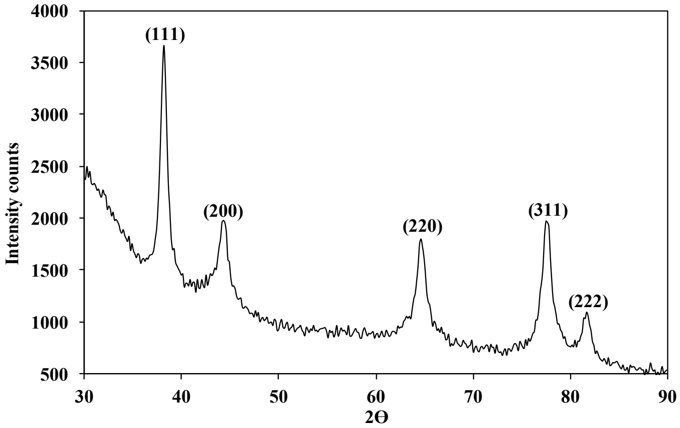

Figure 4. X-ray diffraction pattern of gold nanoparticles synthesized using seaweed *T. ornata* extract.

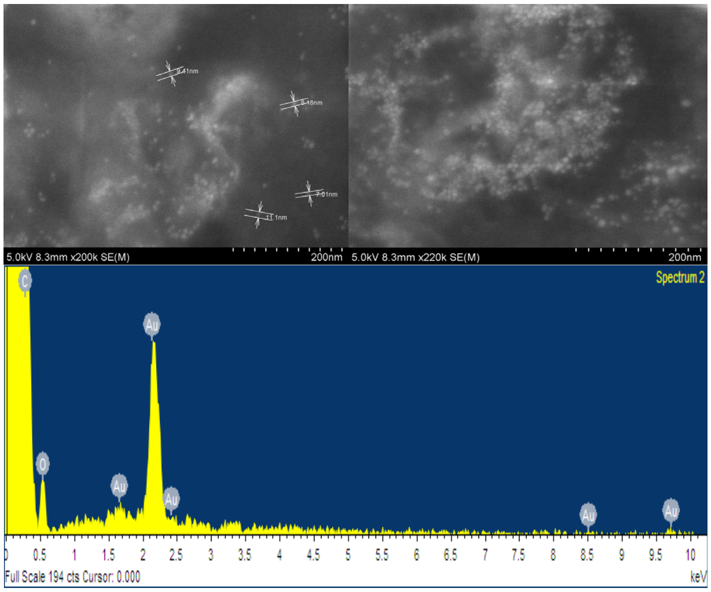

Figure 5. SEM image of synthesized gold nanoparticles (top image); EDX profile of synthesized gold nanoparticles (bottom image).

The surface morphology, crystalline nature and nano size of the gold nanoparticles was confirmed by SEM and the energy dispersive X-ray spectroscopy (EDX) patterns (Figure 5). The SEM images illustrate that all the gold nanoparticles were well separated and there were no aggregations. This indicates the stabilization of gold nanoparticles by the extract of *T. ornata*. Gold nanoparticles showed spherical morphology with particle average size of 7 – 11 nm. Elemental composition of metal nanoparticles can be established using EDX (Strasser et al., 2010). Each element has a unique atomic structure making a unique set of peaks on its X-ray spectrum which, in turn, leads to the characterization of the elements (Noruzi, 2015). The results of EDX analysis are showed in Figure 5. Elemental gold can be seen in the graph presented by the EDX analysis in support of XRD results, which indicated the reduction of gold ions to elemental gold. Sathishkumar et al. (2015) synthesized Au nanoparticles using *Illicium verum* and the EDX spectrum confirmed the presence of Au in the measured sample. The stability of the nanoparticles is an important criterion for the biomedical application under physiological conditions. *T. ornata* seaweed extract was found to be an excellent stabilizing and reducing molecule as other stabilizing agent was not used in the present experiments.

4. CONCLUSIONS

This study examined potential of a brown marine alga, *T. ornata*, to biosynthesize gold nanoparticles from $HAuCl_4$ solution. The main findings can be summarized as follows:

- On contacting $HAuCl_4$ solution with *T. ornata* extract, the color of the reaction mixture gradually changed from brown to ruby red and surface plasmon resonance centered at 525 nm.
- Through FTIR spectra, phenolic groups, alkynes, amide and esters were found to be involved during reduction of Au^{3+} to Au^{0}.
- The XRD results indicated peaks corresponding to (111), (200), (220), (311) and (222) Bragg's reflection based on the face centered cubic structure of Au nanoparticles.
- SEM images confirmed well separated, non-aggregated and stabilized gold nanoparticles with spherical morphology and average particle size of 7 – 11 nm. Thus, the present seaweed-mediated biosynthesis route for nanoparticle synthesis is inexpensive and highly recommended to be used in large-scale production of gold nanoparticles.

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