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Green synthesis of selenium nanoparticles using *Allium sativum* extract

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Abstract

The green synthesis of nanoparticles has recently gained the attention of researchers due to its eco-friendly, cost effective and non toxic nature. The present study on the reduction of Selenium ions to Selenium nanoparticles mediated through *Allium sativum* extract were demonstrated with an experimental procedure. The synthesized nanoparticles were characterized using UV-Visible spectrophotometer, Transmission electron microscopy (TEM), Fourier transform spectroscopy (FT-IR) and Energy dispersive X-Ray spectroscopy (EDAX). The selenium nanoparticles synthesized by *Allium sativum*extract observed as hollow and spherical particles in size ranging 7-48nm which found more stable more than two months.

Key words : Green synthesis, Selenium nanoparticles, *Allium sativum.*

INTRODUCTION

Past decades, Nanoparticles biosynthesis is the active area for researcher. Nanotechnology is the attractive area for its vast applications. The field of nanotechnology is one of the upcoming areas of research in the modern field of material science. Nanoparticles show completely new or improved properties, such as size, distribution and morphology of the particles etc. Novel applications of nanoparticles and nanomaterials are emerging rapidly on various fields [1].The most effectively studied nanoparticles silver[2], gold [3]and platinum [4].

Nanotechnology is fundamentally changing the way in which materials are synthesized and devices are fabricated. Incorporation of nanoscale building blocks into functional assemblies and further into multifunctional devices can be achieved through a “bottom-up approach”. Research on the synthesis of nanosized material is of great interest because of their unique properties like optoelectronic, magnetic, and mechanical, which differs from bulk [5].

Nanoparticles can be synthesized using various approaches including chemical, physicaland biological. Although chemical method of synthesis requires short period of time forsynthesis of large quantity of nanoparticles, this method requires capping agents for sizestabilization of the nanoparticles. Chemicals used for nanoparticles synthesis andstabilization are toxic and lead to non-ecofriendly byproducts. The need for environmentalnon-toxic synthetic protocols for nanoparticles synthesis leads to the developing interestin biological approaches which are free from the use of toxic chemicals as byproducts.Thus, there is an increasing demand for green nanotechnology[6]. Many biologicalapproaches for both extracellular and intracellular nanoparticles synthesis have been reported till date using microorganisms including bacteria, fungi and plants [7,8].

Since noble metal nanoparticles are widely used in biological applications. So there is a need to develop eco-friendly processes for nanoparticles synthesis that do not use toxic chemicals. Biological methods of synthesis of Selenium nanoparticles has been synthesized by different approaches like *Bacillus sp. MshI* [9], *Klebsiella pneumonia*[10], *Aspergillusterus*[11], *Saccharomyas-cerevisiae*[12], *Bougainvillea spectabilis*[13], leaves of lemon [14], resin extract of grapes [15].

In the present research work, we report the synthesis of selenium nanoparticles using *Allium sativum*extract as areducing agent. Selenium nanoparticles are formed by reducing the selenium ions present in the solution of sodium selenite by the aqueous extract of *Allium sativum*and characterized using UV-Visible spectrophotometer, Transmission electron microscopy (TEM), Fourier transform spectroscopy (FT-IR) and Energy dispersive X-Ray spectroscopy (EDAX).

MATERIAL AND METHODS

Preparation of *Allium sativum* extract

Buds of *Allium sativum* 10gm were collected in a clean mortar. Buds were crushed using motor pestle and sufficiently diluted with water to make a thick paste. This paste was filtered through whaltman filter paper. The resulting pest was stored in refrigerator and used for further experiments.

Synthesis of metal nanoparticles

Flask containing 25 ml 5 mM Na2SeO3 solutions was kept on magnetic stirrer. Then drop wise addition of *Allium sativum* extract was made in flask containing Na2SeO3 solution until color of sodium selenite solution changed. From this solution 5 ml was taken which was used as a control. Remaining 20 ml solution was kept in shaker in dark for 72 hrs. After few days the color change of the solution was observed.

UV-Vis spectra analysis

The reduction of metallic selenium ions was observed by measuring the UV-Vis spectrum after 10 to 15 min of color change. A small aliquot was drawn from the solution and a wavelength from 250nm to 700nm on UV-Vis spectrophotometer(Optizon Double beam 3220).

TEM analysis

Transmission Electron Microscopic (TEM) analysis was performed with Techni 20 (Philips, Holland). A thin film of the sample was prepared on a carbon coated copper grid by dropping a very small amount of the sample on the grid. The *Allium sativum*extract containing Se nanoparticles were subjected to centrifugation at 13000 rpm for 10 min. The pellet thus recovered was subjected to washing by its re-suspension in de-ionized water followed by centrifugation at 13000 rpm for 10 min, to remove possible organic contamination present in nanoparticles. Finally, pellet was freeze dried using a lyophilizer (Labconco, Kanas, USA).

EDAX analysis

EDAX analysis was carried out on EDAX XL-30 operating at 15-25KeV.Incorporation of selenium nanoparticles in gauze cloth. Nanoparticles suspension was poured on the gauze cloth discs (diameter 1cm) and there discs were dried at for 7 days.

Sample preparation for Fourier Transform Spectroscopy (FTIR)

Metal containing *Allium sativum* extract for Fourier Transform Infrared (FT-IR) analysis was prepared by mixing 5 mg metal salt in 10 ml *Allium sativum*extract. This metal containing *Allium sativum*extract was incubated at room temperature for 1 hour. After 1 hour incubation, this metal containing leaf extract was dried in Petri plate. After drying, particles were scraped using blade. So, powder of synthesized nanoparticles was obtained. Then spectral scan analysis was carried out at wave number ranging from 400-4000 cm^{-1} by using a FT-IR spectrometer (Perkin Elmer, Spectrum GX) with resolution of 0.15 cm^{-1} to evaluate functional groups that might be involved in sorption process.

RESULTS

Visual observation

Reduction of metal salts into metal nanoparticles by the bio-molecules is always accompanied by the color change of reaction medium. In the present study the colorless solution of sodium selenite is changed in light pink color after dropwise addition of *Allium sativum* extract at zero hour. As the reduction proceed, the color of reaction medium is gradually changed to dark pink color after 24 hours.

UV Visible Spectroscopy

In order to determine the formation of Selenium nanoparticles in theextract of *Allium sativum*, a spectral scanning procedure was carried out from 250 nm to 700 nm. Colloidal solution exhibited absorption maxima at 400 nm (Fig. 2).

Initially the colloidal solution appeared white in color but after incubation of a period of 24 hours, it turned to reddish brown

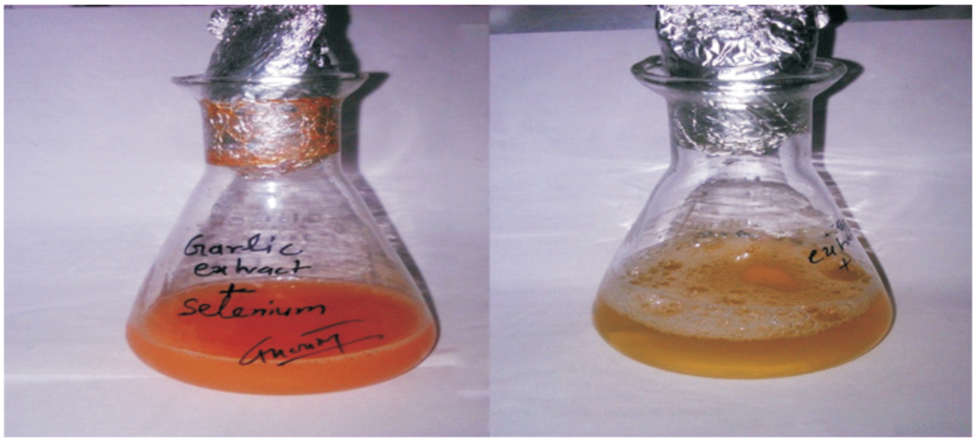

Figure 1: Color change of reaction medium

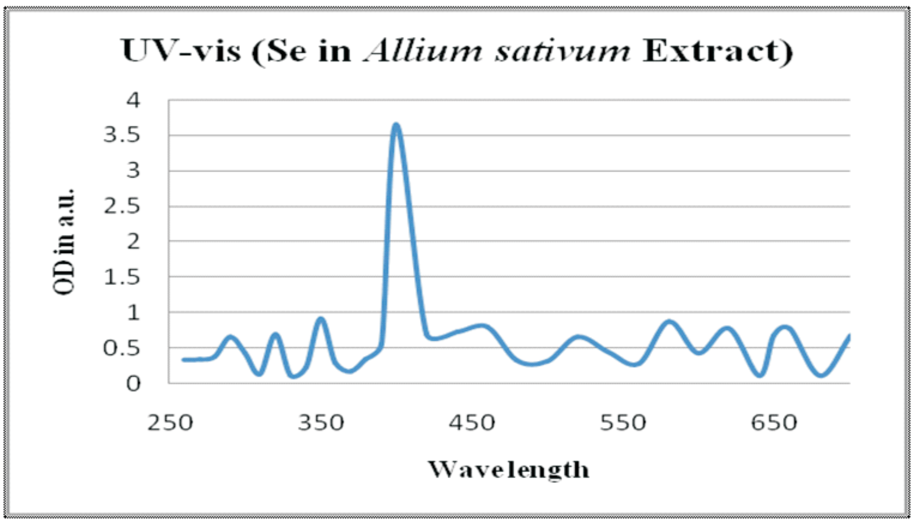

Figure 2: UV- VIS spectra selenium nanoparticles

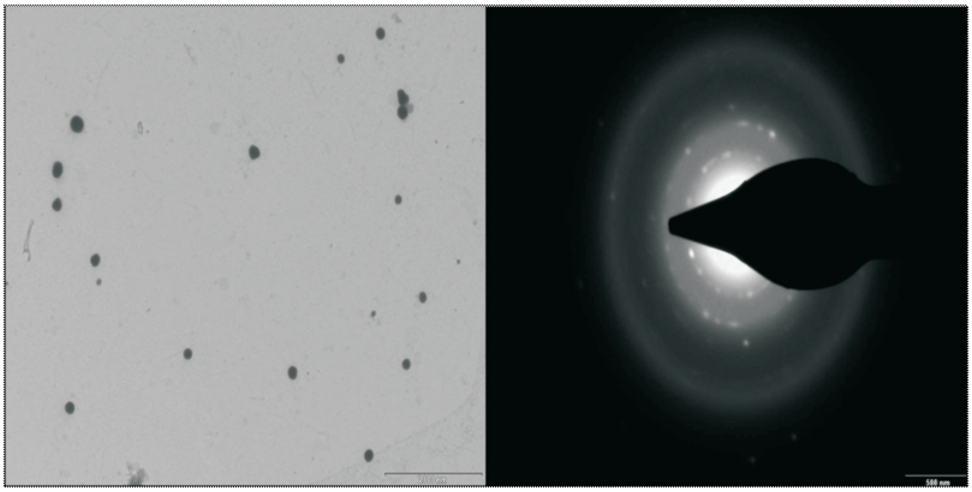

Figure 3: ETEM analysis of Selenium nanoparticles reveled size of particles 7-45 nm.

in color.Building of absorbing maximum at 400 nm clearly indicates the gradual formation of particles during the incubation period.

Transmission Electron Microscopy (TEM):

TEM analysis of colloidal solution indicated the formation of selenium nanoparticles. (Fig.3) shows that size of particles, generated using *Allium sativum* extract ranges from 7 45 nm. Formation of variable size of particles indicates that particles suggest that *Allium sativum* extract could form polydisperse nanoparticles. Fig.3 shows Selected Area Electron Diffraction (SAED) of selenium nanoparticles.

Results shows that particles are crystalline in nature as diffraction ring appeared which correspond to diffraction angle of (111, 121 and 311).

Fourier Transform Infrared Spectroscopy (FT-IR)

FT-IR analysis was carried out to identify the possible bio molecules and plant extract-metal ions interaction responsible for formation and stabilization of selenium nanoparticles. The result of FT-IR analysis of *Allium sativum* extract is presented in figure 4. The figure 4 shows the spectrum of both the sample control (A) and test (B). The fig. 4 (B) shows the spectrum of the sample that contains selenium metal in *Allium sativum* extract or fig. 4 (A) shows the spectrum of the *Allium sativum* extract that did not contain metal selenium. Spectra B show the peaks of both control and test, similarly the Fig. 4 (A) is showing transmission peaks of the control sample. Around 600 and 500 may be due to the partial duitriation of amine or carboxyl group.

Two absorption peaks located around 3400 and 4000 can be assigned as the absorption peak of N-H. The peaks located around 3000 and 3200 may be due to the presence of C-H group. The absorption peaks around 2300 and 2000 can be assigned as the peaks of CO_2. The absorption peaks around 1500 and 1800 can be assigned as the absorption peaks of C=O / C=N / C=C. The peaks around 1200 and 1100 were attributed to the stretching vibration of carboxyl group (C=O). The peaks around 1100 and 1000 may be due to the presence of C-O group. Two absorption peaksaround 600 and 500 may be due to thepartial deuteration of amine or carboxyl group.

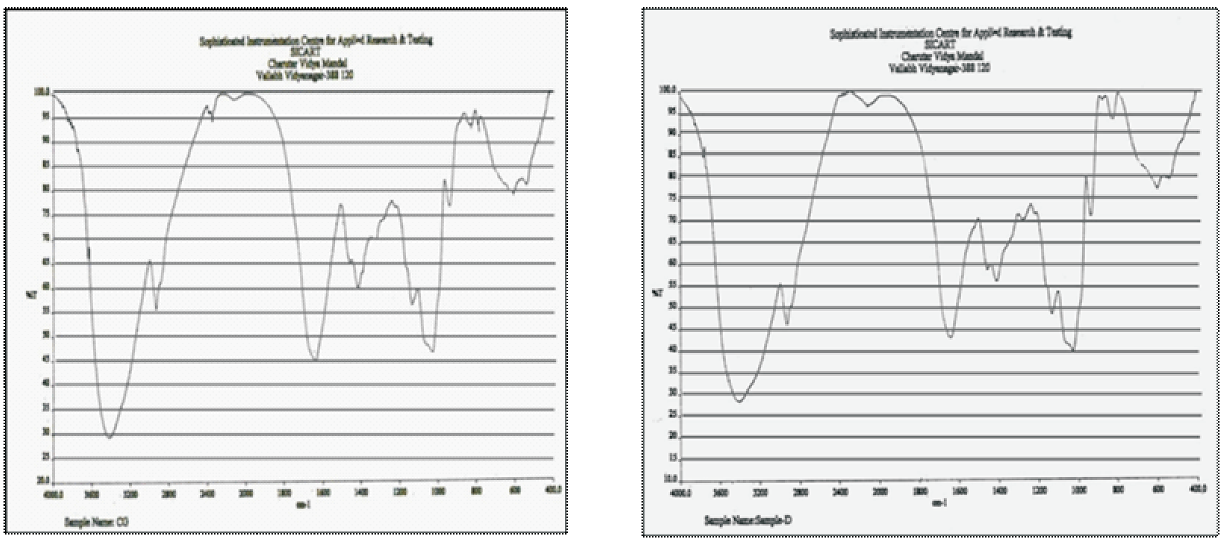

Figure 4: FTIR spectrum of (A) *Allium sativum* extract and (B) Selenium nanoparticles synthesized by *Allium sativum* extract

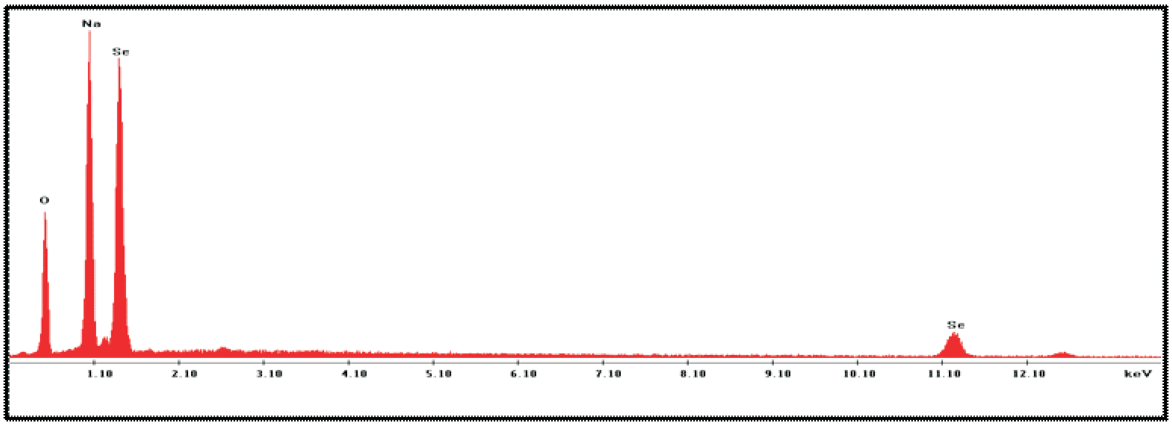

Figure 5: EDAX spectrum of selenium nanoparticles

Energy Dispersive X-Ray Spectroscopy (EDAX)

EDAX analysis gives qualitative as well as quantitative status of elements that may be involved in formation of nanoparticles. Figure shows the elemental profile of synthesized nanoparticles using *Allium sativum* extract. The analysis revealed the highest proportion of Selenium (55%) in nanoparticles followed by oxygen (15%), sodium (28%) etc.

DISCUSSION

In present work for the green synthesis of selenium nanoparticles Fig.1 represents change in color during nanoparticles synthesis. Initially the colloidal solution appeared colorless but after an incubation period of 24h, it turned to light pink. Building of absorbing maximum at 400nm (Fig.2) clearly indicates the gradual formation of particles during the incubation period. Selenite ions were reduced to selenium nanoparticles after addition of the aqoues *Allium sativum* extract followed by incubation of the mixture for studied period of time. Reduction of colloidal selenite more over leading to change in color can be attributed to surface plasmone resonance of selenium nanoparticles.

TEM analysis of colloidal solution indicated the formation of selenium nanoparticles. Fig.3 shows that size of particles generated using *Allium sativum* extracts ranges 7-45nm. Formation of variable size of particles suggested that *Allium satvum* extract could form polydispersed nanoparticles and Fig.3 also shows selected area electron diffraction (SEAD) pattern of selenium nanoparticles. Results indicated that particles were crystalline in nature as diffraction angle of 111, 121 and 311 respectively.

FT-IR analysis was carried out to identify the possible formation and stabilization of selenium nanoparticles.The Fig. 4(a) shows the spectrum of the sample that did not contain metal selenium while, Fig. 4(b) shows the spectrum of the sample that contained selenium metal. Spectra 4(a) show peaks at 3415.80, 2934.42, 1634.86, 1415.53, 1049.81 and 607.61 cm−1. Similarly Fig. 4(b) that contained selenium was obtained at 3418.28, 2930.93, 2356.55, 1616.66, 1517.75, 1404.49, 1319.54, 1074.93, 914.94, 816.85, 773.41, 673.81 and 612.46 cm−1. Two absorption peaks located around 3400 and 4000 can be assigned as the absorption peak of N H. The peaks located around 3000 and 3200 may be due to the presence of C H group. The absorption peaks around 2300 and 2000 can be assigned as the peaks of CO2. The absorption peaks around 1500 and 1800 can be assigned as the absorption peaks of C O/C N/C C. The peaks around 1200 and 1100 were attributed to the stretching vibration of carboxyl group (C O). The peaks around 1100 and 1000 may be due to the presence of C O group. Two absorption peaks around 673.81 and 612.46 may be due to the partial deutriation of amine or carboxyl group. The analysis indicated that the selenium nanoparticles formed by *Allium sativum* extract.

The energy dispersive X-Ray spectroscopy analysis confirmed the presence of elemental selenium nanoparticles in given sample as depiciated by Fig.5. The selenium nanoballs showed characteristics absorption peaks at 1.37 KeV and 11.22KeV. The peak located on the left of the spectrum at around 0.5 KeV clearly indicates the presence of oxygen. The lack of other elemental peaks and high amount of selenium in the spectra confirms the purity of the selenium metal in the transformed product.

CONCLUSION

Present green synthesis shows that the environment friendly *Allium sativum*can be used as an effective capping as well as reducing agent for the synthesis of selenium nanoparticles. Selenium nanoparticles synthesized by the above method are quite stable and no visible changes are observed even after a month or so, if the nanoparticles solutions are kept in light proof condition. UV-Vis Spectra at 400 nm with *Allium sativum* extract and observed as hollow and spherical particles in size ranging 7-48nm which is found more stable more than two months. EDAX analysis was carried out to check the presoak of Selenium in nanoparticles. Results of EDAX, confirmed its present. TEM and SEAD represented addition evidence of formation of nanoparticles whereas SEAD indicates the particles were crystalline in nature.

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