Synthesis Of PEG Coated Ascorbic Acid Functionalized Silver Nanoparticles And Determining its Antioxidant Activity

D. Rajavanciya, D. Venkataraman

Abstract*: *Silver nanoparticles are extensively being studied for various applications in different fields. Functionalized silver nanoparticles are now being used as drug carriers. Ascorbic acid (Vitamin C) is a well-known antioxidant which has been reported to have various beneficial properties to humans. Here we have synthesized silver nanoparticles functionalized with ascorbic acid. The functionalized nanoparticles were further coated with PEG. The nanoparticle synthesis was characterized using UV-Visible spectral analysis, XRD, FTIR and Scanning electron microscope. The average size of the nanoparticle was found to be 24 nm. The synthesized nanoparticle was found to possess antioxidative effects.

Keywords* : *silver nanoparticles, ascorbic acid, PEG, antioxidant

I. INTRODUCTION

The science and technology at the nanoscale levels has huge range of applications of various materials and nanocomposites containing nanoparticles. Essentially the particles with size range < 100 nm are called nanoparticles (Hung *et al*., 2004). The nanoparticles are found to be suitable for widespread applications owing to its properties. For example it is used in drug delivery, cell tagging etc., (Kim *et al*., 2007). The application of the nanoparticles depend on their size, size distribution, structure, chemical and physical environment. Specific control of size, shape and size distribution of nanoparticles are often achieved by various synthesis methods, reducing agents and stabilizers (Zhang *et al*., 2004,). The synthesis of metallic nanoparticles under bottom up approach requires various agents like reducers, various synthesis strategies and most essentially stablizers to prevent the nanoparticles from agglomeration. Among various nanoparticles, the properties of nanoparticles especially their size and shape determines their applications. The applications ranges from acting as catalysts, drug delivery system and even data storage (Choi *et al*., 2007; Nestor *et al*., 2008).

Silver nanoparticle is one among the most studied and commercially explored nanoparticles with a reported production of 500 tonnes per year (Larue et al., 2014). Over the years, silver nanoparticles have found to exhibit biological, chemical and physical properties significantly distinct from the respective bulk materials. The nanoparticles with size less than 100 nm are of great interest as their applications cover wide range of science and industries. The difference in the properties is attributed to the small dimension, high surface area, quantum confinement etc. (Kholoud et al., 2008). Here, we have synthesized silver nanoparticles using ascorbic acid and coated with PEG.

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***** Correspondence Author -

D. Rajavanciya, Department of Biotechnology, Kalasalingam Academy of Research and Education, Krishnankoil, India

D. Venkataraman, Department of Biotechnology, Kalasalingam Academy of Research and Education, Krishnankoil, India.

II. MATERIALS AND METHODS

A. Synthesis ascorbic acid functionalized silver nanoparticles

To 100 ml of borate buffer (50 mM; pH 10) 100mg of ascorbic acid (Sigma; 1 mg/ml) was dissolved and utilized for the synthesis of silver nanoparticles. Then 1 mM of silver nitrate was added to the ascorbic acid solution and it was incubated overnight.

B. PEG-coated silver nanoparticles

To coat PEG, the silver nanoparticles synthesized were collected by adding equal amount of acetone and centrifuging the colloidal silver. To the centrifuged nanoparticels 10 ml of 1% (w/v) PEG was added and mixed using a magnetic stirrer overnight. The PEG coated silver nanoparticles were later collected and characterized.

C. Silver nanoparticles collection and characterization

For characterization, silver nanoparticles were collected by centrifugation and dried under vacuum to obtain a dried powder. The powder was then characterized using UV-visible spectrum, FT-IR, XRD & SEM

D. Fourier Transformed infrared spectroscopy (FT-IR)

The functional groups in the individual components and the silver nanoparticles were analyzed by Fourier transform – infrared spectrophotometer (IR Tracer-100 Shimadzu). The wavenumber for ascorbic acid, silver nitrate, PEG and PEG coated silver nanoparticles were examined from 500 cm^{-1} to 4000 cm^{-1} with a resolution of 0.2 cm^{-1} at room temperature. Obtained FT-IR spectrum was analyzed for characteristic peaks and functional groups.

E. XRD (X-ray diffraction)

owder XRD was performed to determine the nature of the dried powder. Briefly the dried

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powder of silver nanoparticles were analyzed in XRD (Bruker, D8 Advance, US). The crystalline nature of each individual was checked with XRD. The Debye-Scherer equation $\mathbf{D = K\lambda / \beta cos\theta}$ was used to determine the size of the crystal.

F. Scanning Electron Microscope (SEM)

Dried PEG coated silver nanoparticles was checked for the size under a scanning electron microscope (Carl Zeiss *EVO 18*). The EDX (Bruker and Quantax, USA) was used to determine the elemental compositions of the silver nanoparticles.

G. DPPH assay

The antioxidative activity of the silver nanoparticles was determined by DPPH assay. Briefly the ascorbic acid synthesized silver nanoparticles were mixed with DPPH and the decrease in OD was determined by a continuous assay. Here ascorbic acid was used as control.

III. RESULTS

A. *Ascorbic acid synthesized silver nanoparticles*

Silver nanoparticles were synthesized when silver nitrate and ascorbic acid was mixed together. The primary indication for the synthesis of silver nanoparticles the change of colour of the solution from colourless transparent solution to brown courted opaque solution, the colour kept became darker with time. The synthesis was faster in alkaline pH for silver nanoparticles rather than neural or acid pH.

B. *Coating of ascorbic acid functionalized silver nanoparticles with PEG*

To prevent the silver nanoparticles from agglomeration, we coated the synthesized silver nanoparticles with PEG. The synthesized silver nanoparticles were collected by centrifugation and coated with PEG. The synthesized nanoparticles were then characterized using the following methods.

C. *Characterization of the synthesized silver nanoparticles*

1. Visual confirmation

As previously mentioned in section 3.1 the primary indication of silver nanoparticle synthesis i.e. the change in colour of the solution from transparent colourless form to opaque brown colour was observed. A spectrum between 300 – 700 nm was obtained to determine the surface plasmon resonance, which showed a peak at 430 nm, showing it to be the silver nanoparticles.

2. FT-IR analysis

Fig.1 shows the FT-IR peaks for ascorbic acid, PEG and synthesized silver nanoparticles. The –OH stretching is observed at 3410 cm^{-1} and CH stretching is observed at 2914 cm^{-1}. Similarly distinct peaks were observed for pure PEG including peaks at 1282 cm^{-1}, 960 cm^{-1} and 840 cm^{-1}. But the peaks observed in silver nanoparticles are unlike the pure counterparts.

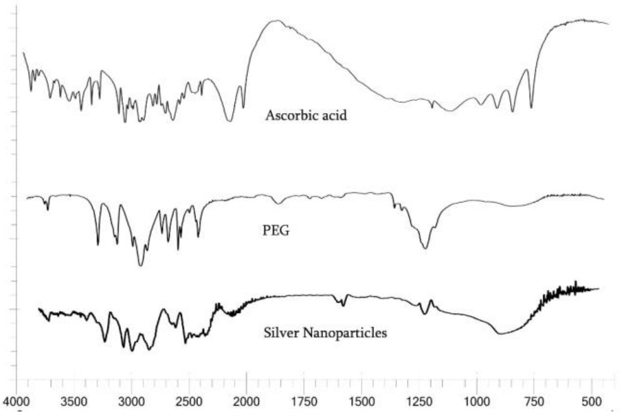

Fig.1 Ft-IR spectra of ascorbic acid, PEG and Silver nanoparticles coated with PEG.

3. XRD analysis

Silver nanoparticle alone is a crystal. The distinct XRD patterns for various kinds of crystals have been studied extensively previously. But when the silver nanoparticles were coated with PEG, they turn into amorphous form. The XRD pattern of PEG coated ascorbic acid functionalized silver nanoparticles is shown in Fig. 2 which shows that the nanoparticle is in amorphous form.

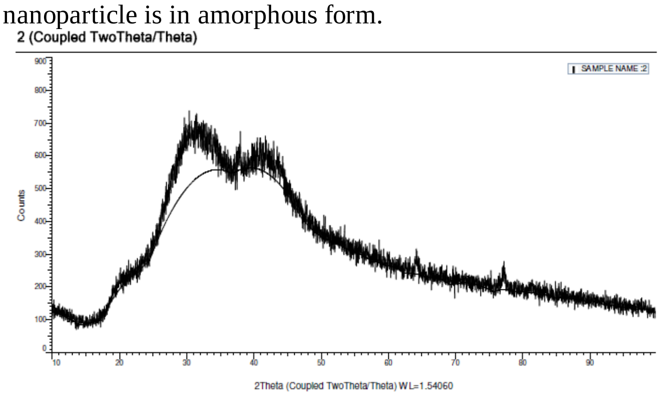

Fig.2 XRD spectrum of ascorbic acid functionalized silver nanoparticles shows the nanoparticles are in amorphous form

4. SEM analysis of PEG-coated silver nanoparticles

Fig.3 shows the synthesized silver nanoparticles under SEM. The morphology of the PEG-coated nanoparticle was found to be spherical. This may be due to the coating of PEG over silver nanoparticles. Individual particles could be visualized and the average diameter of the nanoparticles was about 24 nm. The size of nanoparticles was found to be nearly uniform. The elemental analysis was performed later.

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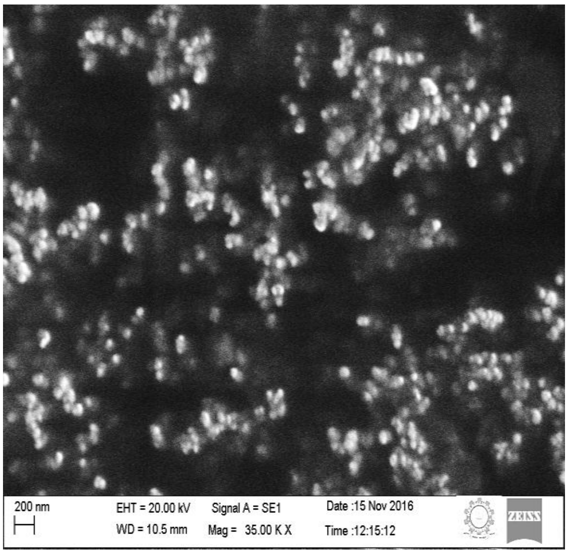

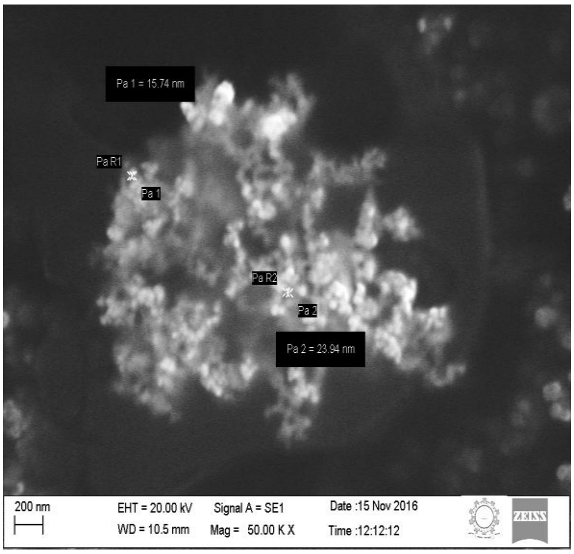

Fig. 3 SEM images of PEG coated AgNps where average size of the nanoparticles were found to be 24nm

5 Anti-oxidative activity of PEG-coated silver nanoparticles

DPPH assay was employed to determine the antioxidative activity of PEG-coated ascorbic acid functionalized silver nanoparticles. The antioxidative activity showed a concentration dependent response where increase in the concentration of PEG-coated silver nanoparticles contributed to the increase in antioxidative activity. The data are presented in Table. 1.

Table. 1 Antioxidative activity of PEG coated silver nanoparticle by DPPH assay

Concentration of Silver Nanoparticle (µg)	Antioxidant activity
200	9.7 ± 1.2
400	22.7 ± 2.1
600	37.3 ± 1.5
800	56.3 ± 2.9
1000	69.0 ± 2.6

IV. DISCUSSION

PEG coated silver nanoparticles were synthesized using ascorbic acid as reducer. Ascorbic acid or vitamin C is a well-known antioxidant. Here the silver nitrate is reduced to silver with the help of ascorbic acid. The nanoparticle synthesis was primarily characterized by visual change of solution colour to brown and surface plasmon resonance. The size of the nanoparticle was 24 nm after coating with PEG. The nanoparticle synthesized are coated with PEG in order to prevent agglomeration. PEGylation or coating of PEG on various nanoparticle surface has been reported to improve the efficiency of gene and drug delivery to the target tissues and cells. They also have been studied for increasing the bioavailability of the nanoparticle and immobilized drug, immunogenicity etc., (Suk et al., 2016). Ascorbic acid has been reported to improve the efficiency of cancer chemotherapeutic drugs (Liu et al., 2011 & Volbracht et al., 2011). The synthesized nanoparticle exhibited antioxidant activity which shows that the ascorbic acid still elicits its antioxidant activity even after reducing the silver to silver nanoparticles.

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AUTHORS PROFILE

Rajavanciya has completed her B. Tech. at Kalasalingam Academy of Research and Education and her Master's in Business Administration at Karpagam University. The work is a part of her final year project thesis.

D. Venkataraman has completed his Bachelor's and Master's of Technology at Kalasalingam University. He then later joined as a doctoral student and completed his doctoral studies at the same institute. His field of interest is nanotechnology and cancer biology.

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