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# Rapid biogenic synthesis of silver nanoparticles using black pepper (*Piper nigrum*) corn extract

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**Abstract**

Rapid biogenic synthesis of silver nanoparticles using extract of black pepper corn was attempted. Aqueous solution of $AgNO_3$ was treated with black pepper corn extract and can reduce most of the silver ions into silver nanoparticles within 120 s of reaction time with the aid of microwave. The formation of nanoparticles is reported by UV-vis spectrophotometer and complemented with characterization using Transmission Electron Microscopy, X-ray Diffraction and Fourier Transform Infrared Spectroscopy. Transmission Electron Microscopy revealed the presence of spherical silver nanoparticles with size ranging of 5 to 50 nm. X-ray diffraction studies corroborated that the biosynthesized nanoparticles are crystalline silver. Furthermore, this green biogenic approach was rapid and simple alternative to chemical synthesis.

**Key words:** *Piper nigrum*, Silver nanoparticles, Microwave, Transmission Electron Microscopy, Transform Infrared Spectroscopy

## Introduction

In 21st century, nanotechnology has emerged as a rapidly growing field with numerous applications in science and technology. Metal nanoparticles have attracted considerable attention for their unusual chemical and physical properties such that they show great potential applications in biotechnology, bioremediation of radioactive wastes, gene delivery for treatment or prevention of genetic disorder, catalysis, medical imaging, particularly in medicine such as delivery of antigen for vaccination, novel electronics, optics, better drug delivery methods, chemical deposition for environmental pollution cleanup [1–12] inspired the scientists to develop environment friendly procedures for the synthesis of nanoparticles and to avoid use of hazardous chemicals, which are traditionally used. Their novel optical, thermal, chemical and physical properties are because of higher surface energy of nanoparticles compared to the bulk solid and short mean free path of an electron in a metal (10–100 nm) for many metals at room temperature. Physical methods such as vapor deposition, molecular beam epitaxy [13], etc. require expensive high technology and are neither suitable for mass production nor energy-efficient. Although they are relatively more affordable and suitable for mass production as well as allowing tight control over the particle size distribution in many ways, chemical methods [14] are not environment friendly. Biosynthesis of nanoparticles offers many advantages over the corresponding physical and chemical methods. Contrary to alternative physical and chemical methods employing toxic chemicals which are unacceptable for medical applications, biological synthesis methods use bacteria [15–17], yeast [18], fungi [19-21] or biomass of oat (*Avena sativa*) and wheat (*Triticum aestivum*) have been reported for irregular and rod shaped gold nanoparticle (GNP) with size range spaning 10-30 nm [22], bengal gram bean(*Cicer arietinum*) was also reported for gold nanotriangles [23]. Similarly for the synthesis of silver nanoparticle, plants such as *Capsicum annuum* [24], Quercetin [25] were used. In addition to the independent synthesis of silver and gold nanoparticles various plants such as sun dried biomass of *Cinnamommum camphora* was used for synthesis of triangular and spherical gold and silver nanoparticles synthesis with size range between 55-80 nm [26], fruit extract of Amla (*Emblica officinalis*) for silver and gold nanoparticles synthesis with dimensions spanning 10-20 nm and 15-25 nm respectively [27], leaf extract of *Aloe vera* produced triangular and spherical silver and gold nanoparticles [28], and neem (*Azadirachta indica*) leaf broth was used for

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silver, gold and Ag–Au bimetallic Au core-Ag shell nanoparticles[29]. Overall material and energy consumptions in biological methods are extremely lower, offering a low-cost green alternative. A number of plants such as alfalfa (*Medicago sativa*) produced gold nanoparticles of 4-10 nm size range [30]. These methods are very efficient in the production of silver and gold nanoparticles because of their simplicity, accuracy and cost effectiveness, also from the green chemistry perspective and their biomedical application.

We herein report the synthesis of silver nanoparticles using extract of black pepper corn with the aid of microwave. Dried black pepper corn is used medicinally for gastrointestinal disorders [31]. In addition to these medicinal uses, black pepper continues to be valued around the world as important cooking spice. The approach followed by us is cost efficient alternative to conventional methods and completely biogenic method of silver nanoparticles synthesis.

## MATERIALS AND METHODS

### S*ynthesis and characterization of silver nanoparticles*

$AgNO_3$ was purchased from Qualigens Fine Chemicals, Mumbai, India. Black pepper corns were purchased from a local market, washed to remove any impurities, dried under sunlight to completely remove the moisture and grinded in a mixer grinder. 10 gm powder of black pepper corn was taken and mixed with 100 ml of water and was exposed to microwave for 180 s to suppress the enzymes present in the solution. The raw extract obtained was cooled and centrifuged at 4000 rpm for 10 min then supernatant was filtered with 11-micron mesh to remove impurities. The resultant clear extract was used for the synthesis of silver nanoparticles. For reduction of $Ag^+$ ions, 10 ml corn extract was added to 50 ml of $10^{-3}$ M aqueous $AgNO_3$ solution and the solution mixture was exposed to microwave radiation at a fixed frequency of 2.45 GHz and power of 450 watt. Periodically, aliquots of the reaction solution were removed and subjected to UV–vis spectroscopy measurements. The formation of the silver nanoparticle is monitored using UV–vis spectra by employing a UV–visible Systronics double beam spectrophotometer 2203 operated at a resolution of 0.1 nm with optical path length of 10 mm. UV–vis analysis of the reaction mixture was observed for a period of 210 s.

The transmission electron microscopy (TEM) image of the sample was obtained using FEI Philips Morgagni 268D instrument operated at 100 kV. TEM samples of the aqueous suspension of silver nanoparticles were prepared by placing a drop of the suspension on carbon-coated copper grids and the films on the TEM grids were allowed to stand for 2 min, after which the extra solution was removed using a blotting paper and the grid was allowed to dry prior to measurement.

The FTIR spectrum was recorded using Bruker – FTIR Alpha instrument at a resolution of 4 $cm^{-1}$.

For XRD measurements silver nanoparticles powdered sample was prepared by centrifuging the synthesized silver nanoparticle solution at 10,000 rpm for 10 min. The solid residue layer containing silver nanoparticle was redispersed in sterile deionised water for three times to remove unattached biological components to the surface of nanoparticles that are not responsible for bio-functionalization or capping. The pure residue was dried perfectly in an oven overnight at $65^0$C.

For the crystallinity studies, powder is used for X-ray diffraction (XRD) study. The data was obtained using Bruker X-ray diffractometer, operated at 30 kV and 10 mA current with Cu K(α) radiation of 1.54 nm wavelength.

## RESULTS AND DISCUSSIONS

### UV–*visible absorbance studies*

*Th*e addition of black pepper corn extract to silver nitrate solution resulted in color change of the solution from transparent to yellowish brown after exposure to microwave radiation. The color intensity increases with increased exposure of reaction mixture to microwave radiation. These color changes arise because of the excitation of surface plasmon vibrations with the silver nanoparticles (32). The metal ions reduction occurs very rapidly and reduction of most of the $Ag^+$ was completed in 120 s. Above 120 s and up to 150 s, the reaction process is drastically reduced as the reaction rate changes to linear phase. The surface plasmon resonance (SPR) of silver nanoparticles produced a peak at 360 nm. UV–vis absorbance of reaction mixture was taken at 30 s interval (Fig. 1). Blank analysis was carried out by exposing 50 ml of $10^{-3}$ M silver nanoparticle solution to microwave for an extended period of 210 s and validation is done

by UV-vis study. It is observed that there is no color change of aqueous $AgNO_3$ solution indicating the particles are not formed.
The microwave-assisted method is much faster as compared to conventional studies with other biological routes. The time required for the conventional synthesis of silver nanoparticles from other plants was 2-4hrs [33-34] and from bacteria was 24-120 hrs [35-39] and thus are rather slow.

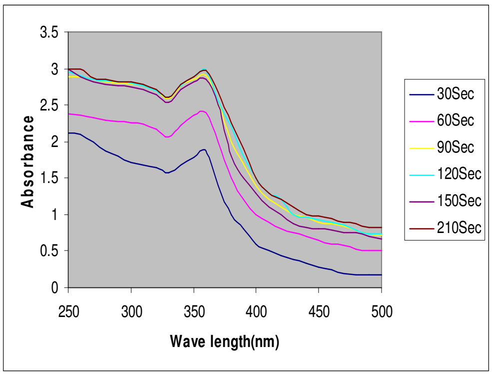


**Fig. 1: UV-Visible spectra of rapid biogenic synthesis of silver nanoparticles at various time interval**

### *Transmission electron microscopy*

Transmission electron microscopy (TEM) was performed for characterizing size and shape of biosynthesized silver nanoparticles. The TEM images of the prepared silver nanoparticles at 50 nm scale are shown in Fig. 2. It was observed that Ag nanoparticles were circular in shape with maximum particles in size range within 5 to 50 nm. It was also observed that silver nanoparticles were evenly distributed in the sample.

### *Fourier transform infrared spectroscopy*

Fig. 3 shows the FTIR spectra of biosynthesized silver nanoparticles and carried out to identify the possible interaction between protein and silver nanoparticles. Results of FTIR study showed sharp absorption peaks located at about 1631cm$^{-1}$ and 3433 cm$^{-1}$. Absorptiion band at 1631cm$^{-1}$ suggested the presence of amide group, raised by the carbonyl stretch of proteins. These results indicated that the carbonyl group of proteins adsorbed strongly to metals, indicating that proteins could have also formed a layer along with the bio-organics, securing nanoparticles. Absorption peak at 3433cm$^{-1}$ are assigned to OH stretching in alcohols and phenolic compounds (40). The absorption peak at 1631 cm$^{-1}$ is close to that reported for native proteins (41) which suggest that proteins are interacting with biosynthesized nanoparticles and also their secondary structure were not affected during reaction with $Ag^+$ ions or after binding with Ag nanoparticles (42). These IR spectroscopic studies confirmed that carbonyl group of amino acid residues have strong binding ability with metal suggesting the formation of layer covering metal nanoparticles and acting as capping agent to prevent agglomeration and providing stability to the medium (43). These results confirm the presence of possible proteins acting as reducing and stabilizing agents.

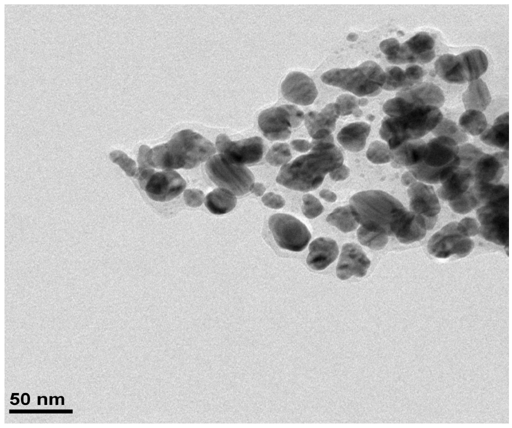


**Fig. 2: TEM image of silver nanoparticles**

### *X-ray diffraction measurements*

*Fig.* 4 shows the XRD pattern of silver nanoparticle synthesized from corn extract of black pepper. A number of Bragg reflections with 2θ values of 38.030, 46.18$^0$, 63.43$^0$, and 77.18$^0$ correspond to the 111, 200, 220, and 311 sets of lattice planes are observed which may be indexed as the band for face centered cubic structures of silver. The XRD pattern thus clearly illustrates that the silver nanoparticles synthesized by the present green method are crystalline in nature.

## CONCLUSION

Synthesis of Silver nanoparticles using black pepper corn extract provides an environmental friendly, simple and efficient route. The nanoparticles produced were of spherical in shape with size range from 5 to 50 nm. The bio-synthesized silver nanoparticles were characterized using UV-Vis, XRD, TEM and FTIR spectroscopic techniques. The synthesized silver applications in biomedical field, medical and pharmaceutical applications as well as large scale commercial production.

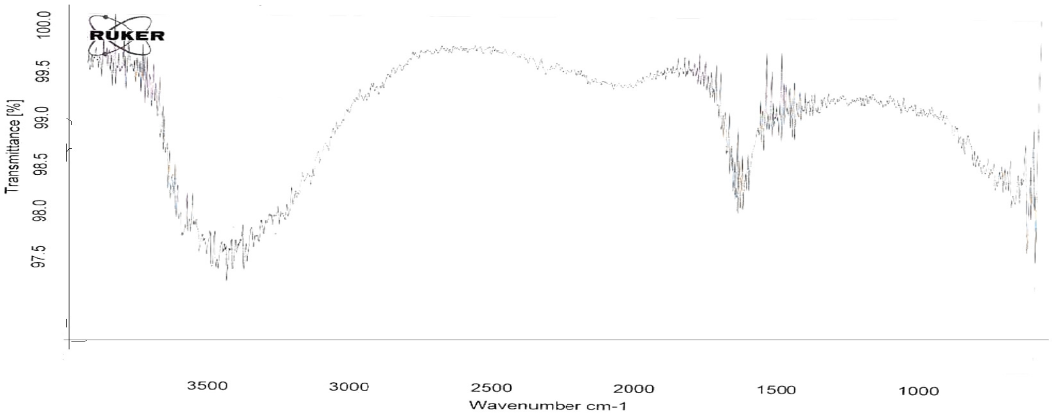


**Fig. 3: FTIR spectra of silver nanoparticles**

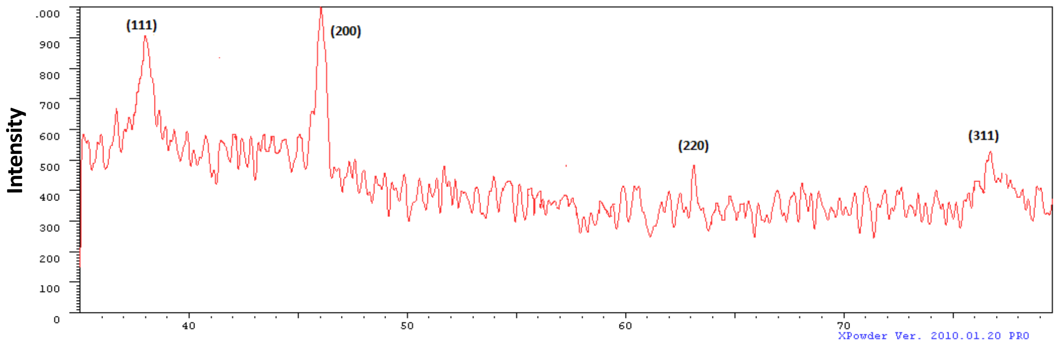


**Fig. 4 : XRD spectra of silver nanoparticles**

nanoparticles were seen to be stable due to the presence of proteins which act as capping agent and prevent the particle from aggregation. These nanoparticles were crystalline in nature. From a technological point of view, these obtained silver nanoparticles have several advantages such as more surface availability and can function effectively. In this synthesis, material is environmentally safer (plant material) compared to chemically synthesized nanoparticles, which often involve harmful chemicals or are environmentally less acceptable. These synthesized silver nanoparticles have various

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