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SYNTHESIS AND CHARACTERIZATION OF SILVER NANOPARTICLES AND THEIR ANTIBACTERIAL ACTIVITY

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ABSTRACT

Silver nanoparticles were prepared by chemical reduction method. Silver nitrate (AgNO3 99.99%) was taken as source of silver. N,N-Dimethylformamide (DMF) was used as solvent as well as reducing agent. Poly vinylpyrrolidone (PVP) was used as stabilizer or capping agent. The formation of the silver nanoparticles was monitored using UV-Vis absorption spectroscopy (Shimadzu UV-1800). The UV-Vis spectroscopy revealed the formation of silver nanoparticles by exhibing the typical surface Plasmon absorption maxima at 415-426 nm from the UV–Vis spectrum. Particle size measurements of colloidal silver were recorded with 90Plus Particle Size Analyser (Brookhaven Instrument Corp.). I have used UV–Vis spectroscopy, particle size measurement and Fourier Transform Raman Spectroscopy (FT-Raman) to characterize the nanoparticles obtained. The synthesized nanoparticles of silver showed high antibacterial activity against gram negative bacteria Escherichia Coli .

Keywords— chemical reduction, Silver nanoparticles, surface Plasmon, UV-Vis absorption Spectrum.

I. INTRODUCTION

Silver Nanoparticles are of great research focus because of their unique functional properties that are significantly different from those of bulk materials [1] which lead to varied applications in the areas of plasmonics [2], Surface enhanced Raman scattering(SERS)[3,4], silver nanoparticles occurs within the visible light frequency range and its precise wavelength is strongly associated with the diameter, spacing and environment of nanoparticles. This has generated a wide interest in their potential for a wide range of optical and photonic applications [5,6]. In recent years nanoparticles of silver have been found to exhibit interesting antibacterial activities [7-10]. Antibacterial activity of the silver-containing materials can be used, for example, in medicine to reduce infections as well as to prevent bacteria colonization on prostheses [11], catheters [12,13], vascular grafts [14], dental materials [15], stainless steel materials [16] and human skin [15,17]. The use of silver nanoparticles as antibacterial agent is relatively new. Because of their high reactivity due to the large surface to volume ratio, nanoparticles play a crucial role in inhibiting bacterial growth in aqueous and solid

media. Silver containing materials can be employed to eliminate microorganisms on textile fabrics [18,19] or they can be used for water treatment [20]. The antibacterial activity of colloid silver particles are influenced by the dimensions of the particles. The smaller the particles, the greater antibacterial effect [11]. Therefore, in developing routes of synthesis, an emphasis was made to control the size of silver nanoparticles. Silver nanoparticles have been produced using different methods: Chemical Vapour deposition , micro emulsion, electrochemical method [12-14], thermal decomposition [15], laser ablation [16], microwave irradiation [17] . The simplest and the most commonly used bulk-solution synthetic method for metal nanoparticles is the chemical reduction of metal salts [18,19]. In fact, production of Nano sized metal silver particles with different morphologies and sizes [20] using chemical reduction of silver salts has been reported [21]. This synthetic method involves reduction of an ionic salt in an appropriate medium in the presence of surfactant using various reducing agents [22].The dispersions of silver nanoparticles display intense colors due to the Plasmon resonance absorption. The surface of a metal is like a plasma, having free electrons in the conduction band and positively charged nuclei. Surface Plasmon resonance is a collective excitation of the electrons in the conduction band; near the surface of the nanoparticles. Electrons are limited to specific vibrations modes by the particle's size and shape. Therefore, metallic nanoparticles have characteristic optical absorption spectrums in the UV-Vis region [23]. The antibacterial characteristics of silver nanoparticles produced have been demonstrating by directly exposing bacteria to colloid silver particles solution [24].

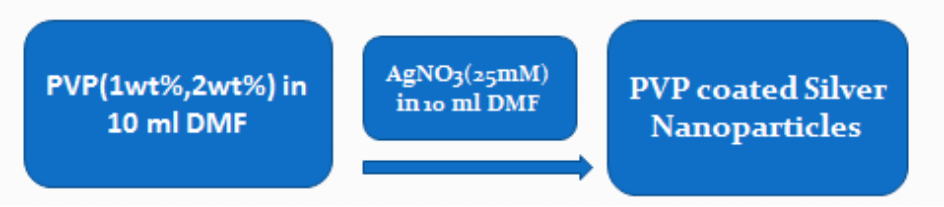
II. EXPERIMENTAL

A. Material

Silver nitrate (AgNO3 99.99%) was taken as source of silver. N,N-Dimethylformamide (DMF) was used as solvent as well as reducing agent. Poly vinylpyrrolidone (PVP) was used as stabilizer or capping agent. All the chemicals used in this experiment were analytical reagent(AR) and without further purification.

B. Preparation of silver nanoparticles

For the preparation of silver nanoparticles Silver nitrate (AgNO3 99.99%) was taken as source of silver. N,N-Dimethylformamide (DMF) was used as solvent as well as reducing agent. Poly vinylpyrrolidone (PVP) was used as stabilizer or capping agent.



The preparation of silver nanoparticles takes place by simple addition of solution of silver nitrate to a PVP solution in DMF. The concentration of Ag NO3 varied from 10 mM to 25 mM for two different concentration of PVP as 1wt% and 2wt%. Herein the effect of concentration of both AgNO3 as well as PVP on optical properties was studied. The in-situ formation of Silver nanoparticals was studied from optical absorbance spectra with

respect to time. PVP of two different molecular weights were tested for their ability to stabilize silver colloidal formed in chemical reduction by DMF. DMF acts as reducing agents. It reduces silver ions even at room temperature. The oxidation of DMF produces hydrogen from water –DMF mixtures as shown in the following reaction.[2]

$$HCONMe2 + 2Ag+ H2O \rightarrow 2Ag^{0} + Me2NCOOH + 2H^{+}$$

The most widely used substances for the stabilization of metal nanoparticles are ligand and polymers. Such substances can also control the reduction rate of the metal ions and aggregation process of zero valent metal atoms. PVP has been demonstrated to be one of the most versatile polymer.

C. Characterization techniques

Characterization is an important step in the development of superior material. UV-Vis absorption measurement is one of the most important method to revel the optical properties of the metal nanoparticles. The absorption spectra of samples were recorded with Shimadzu UV-1800. The studies of size, morphology and composition of the nanoparticles were performed by means of partical size Analyzer (Brookhaven Instrument Corp.), FT-Raman spectroscopy(Broker FT-Raman Spectrometer).

D. Antibacterial assay:

All the glassware, media and reagents used were sterilized in autoclave at 121 ^{0}C for 20 min. For studying the antibacterial assays, Escherichia coli(E.coli) was used as a model test strain for gram negative bacteria. Bacterial suspension was prepared by growing a colony overnight in nutrient broth. Well diffusion method was used to compare the antibacterial activity of silver nanoparticles capped with PVP on E coli. The agar plates were inoculated with bacterial suspension and 25 µl of silver nanoparticles were added to a centre well with a diameter of 5mm. Plates were incubated at 37 0C for 24 hrs and zone of inhibition (ZOI) was measured by subtracting well diameter from the total inhibition zone diameter.

III. RESULTS AND DISCUSSIONS

Silver nanoparticles were synthesized according to the method described in the previous section. Figure 3.1 shows the photographs of samples obtained at different conditions by varying the concentration of silver nitrate and PVP (MW 40000).

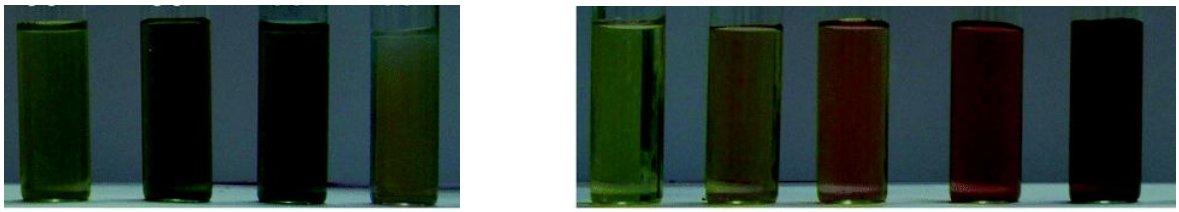


Fig 3.1: Photographs of colloidal silver nanoparticles with different concentration of $AgNO_3$ and PVP

UV-visible spectroscopy is one of the most widely used techniques for structural characterization of silver nanoparticles. The absorption spectra of samples were recorded with Shimadzu UV-1800. All the spectrums

were recorded at 300 nm to 900 nm scanning range. The fig.2 shows the absorption spectrum of AgNO3 onto PVP (MW 40000) 1wt% and 2wt% in DMF respectively. The peak positions were determined to be at 420 nm and 415 nm corresponding to PVP (MW40000) concentration of 1wt% and 2wt % respectively. The peak is shifted towards red on decreasing PVP concentration. Silver colloids showed a surface Plasmon absorption band with a maximum of 420 nm indicating the presence of spherical or roughly spherical Ag nanoparticles. The bands are sharper and more symmetrical which reflects more uniform size distribution.

Fig 3.2: Time evolution of UV- Visible spectra after addition of Ag NO_3 onto PVP (MW 400000) 1 wt % and 2wt % solution in DMF respectively

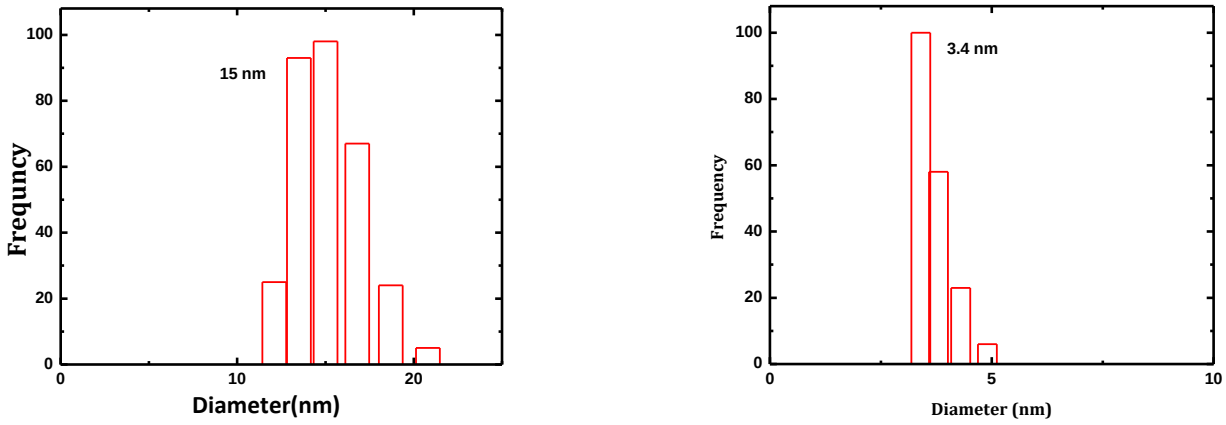


Fig 3.3: Particle size distribution of silver colloid (25 mM) in 1wt% and 2wt % PVP

Figure 3.3 shows that the particles range in size from 3.4 nm to 15 nm. Particle size measurements of colloidal silver were recorded with 90 plus particle size analyzer. All the measurements were carried out at 90 degree scattering angle and at room temperature. With decreasing PVP concentration from 2wt% to 1wt%, the average particle size changes from 3.4 nm to 15 nm for MW

40000.

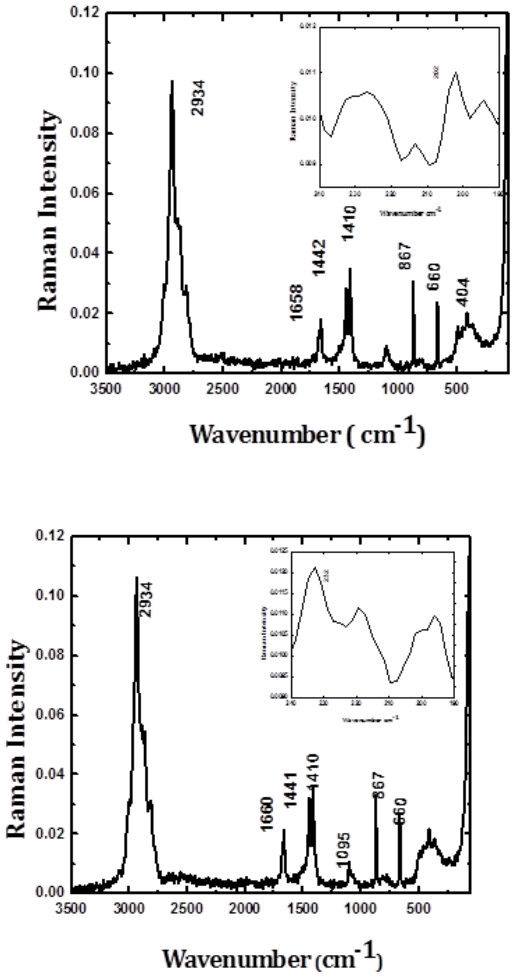
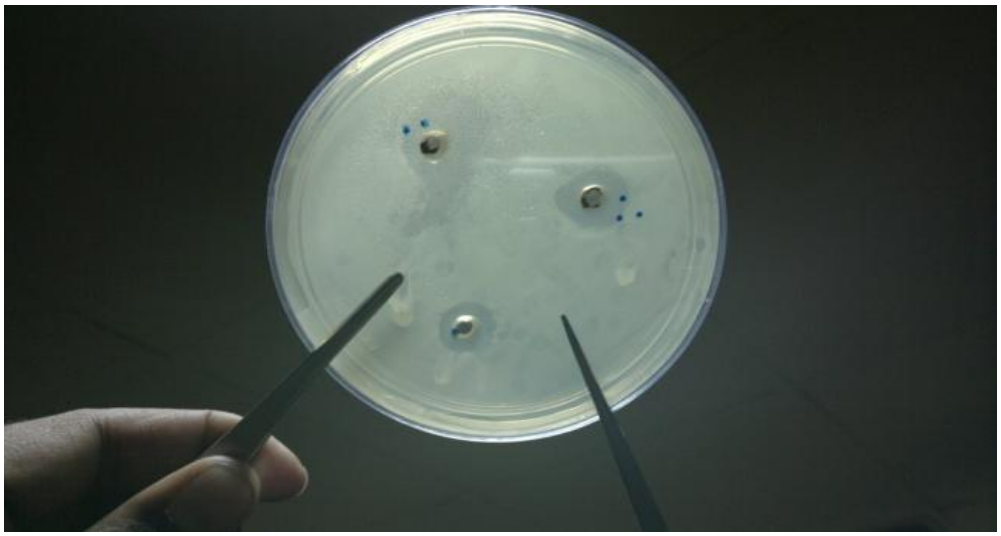


Fig 3.4: FT Raman spectrum

FT Raman spectra were recorded with a Broker FT Raman spectrometer. An excited wavelength at 9395.2 nm was provide by a Ln- Ge I diode laser with a power of 500 mW . All data were collected at a spectral resolution of 4 cm^{-1} from 50 cm^{-1} to 3500 cm^{-1}. FT Raman spectrum of samples is shown in fig 3.4. The entire spectrum of all samples shows strong band at 2934 cm^{-1} corresponds to C-H stretching. Raman bands at 1442 and 1410 cm^{-1} are attributed to C-N and N2C7 stretching along with contributions from ring CH bend. Also 1658, 1660 and 1661 cm^{-1} band are implying to inplane bend of H2O. The 867 cm^{-1} band is attributed to C-H out of plane bend whereas 660 cm^{-1} is ring CH(NH) out of plane bend. The band at 202 cm^{-1} and 232 cm^{-1} are attributed to Ag-N stretch shown in inset figures. The peak of C-N and Ag- N are red shift. This change of spectrum indicates the coordination between N and Ag. The main reason of PVP protecting silver nanoparticles is N in PVP coordinated with silver and formed the protection layer.

Antibacterial activity:

For studying the antibacterial assays, Escherichia coli(E.coli) was used as a model test strain for gram negative bacteria. Bacterial suspension was prepared by growing a colony overnight in nutrient broth. Well diffusion method was used to compare the antibacterial activity of silver nanoparticles capped with PVP on E coli.



Zone of inhibition (ZOI) was measured by subtracting well diameter from the total inhibition zone diameter.Nanosilver disc exhibit zone of inhibition(ZOI) of around 8mm diameter. It is reasonable to state that the binding of the particles to the bacteria depends on the surface area available for interaction. Smaller particles having the larger surface area available for interaction will give more bactericidal effect than the larger particles.

IV. CONCLUSION

In summary, silver nanoparticles with mean diameters of 3.4, 8.5, 15 and 19.8 nm were synthesized using DMF as solvent as well as a reducing agent and PVP as the stabilizer or capping agent. The nanoparticles were characterized by UV/Vis, particle size analyser, FT-Raman spectroscopy. UV/Vis spectra show the characteristic Plasmon absorption peak for the silver nanoparticles ranging from 415 to 420 nm. Additionally, the antibacterial activity of the nanoparticles dispersion was measured by *Well diffusion* method. The results of this study clearly demonstrated that the colloidal silver nanoparticles inhibited the growth and multiplication of the tested bacteria, including highly multiresistant bacteria such as *Escherichia coli*. Such high antibacterial activity was observed at very low total concentrations of silver.

V. ACKNOWLEDGMENT

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