

# Synthesis of colloidal starched silver nanoparticles by sonochemical method and evaluation of its antibacterial activity

D.Jansirani\*, N.Karthick Raja, R.J.Hariprasanth, S.Sweetin Preethi, R.S.A.Sorna Kumar  
Department of Biotechnology, P.S.R.Engineering College, Sivakasi-626 140, Virudhunagar (Dt.)

\*Corresponding author: E-Mail: venkat.janu@gmail.com, 9940782614

## ABSTRACT

The silver ions have been used as an inhibitor to possess a broad spectrum of antimicrobial activity, upon inhibition silver ions were found to have deposited inside the vacuole and cell walls as granules. Silver inhibit cell division resulting in damage to cell envelope and cellular contents of the bacteria. So, the most important application of silver nanoparticles in the pharmaceutical industry such as topical ointments and lotions to prevent infection against burn and open wounds.

Using sonication method, colloidal starch-Ag nanoparticle was synthesized and studied under UV-Visible range. The peak absorption was attained at 435 nm which correspond to yellowish brown color. The molecular size was studied using scanning electron microscope and the colloidal nanoparticle was found to be about  $42\pm 3$ nm in size.

The synthesized colloidal nanoparticle was then studied for five different organisms commonly known for causing pus formations in wounds namely *N.meningitides*, *P.aeruginosa*, *E.coli*, *B.cereus* and *Brucella spp.* Using  $\text{AgNO}_3$  as positive control, antimicrobial activity of the synthesized colloidal starched Ag NPs was studied at three different concentrations 0.1%, 0.5% and 1%. 1% was found to be most optimal in inhibiting growth of organisms to a greater extent, when compared to other concentrations. This concentration gave an inhibition zone of  $7\pm 0.8$ ,  $5\pm 0.7$  and  $6\pm 0.4$  mm for *N.meningitidis*, *E.coli* and *Brucella spp.*, respectively. 0.5% concentration was found to be optimal for *P.aeruginosa* and *B.cereus* showed a zone of  $5\pm 0.4$  and  $3\pm 0.7$ mm more when compared to  $\text{AgNO}_3$ . Based on the study, it was found that the synthesized starched silver NPs was effective in inhibiting the organisms of interest even at low concentration. Therefore, the synthesized colloidal silver NPs can be used in formulation of topical cream and lotions.

**KEY WORDS:** Colloidal starched silver nanoparticles, Silver nitrate, Sonication, Antimicrobial activity, wound pathogens.

## 1. INTRODUCTION

In last decade, nanoparticles appeal to global attentions due to its physical, chemical properties and weighty use in wide applications such as medicine and various fields. In the nano scale level the property of the material are different from that of their bulk material and increased surface area of these nanoparticles is mainly responsible for their different chemical, optical, and mechanical property (Supraja, 2013).

In particular, silver nanoparticles have a significant role in pharmaceutical science. It has antimicrobial property being non-toxic and environmentally safe. Silver nanoparticles have more advantage than conventional antibiotics (Kathiravan, 2014). Silver nanoparticles have attracted more attention because of their antimicrobial activity. Silver nanoparticles play a significant role in biological systems, living organisms and multidrug resistance (Balu, 2012). Silver ions were found to have deposited inside the vacuole and cell walls as granules. Silver inhibit cell division resulting in damage to cell envelope and cellular contents of the bacteria (Karunamoorthy, 2014). So, the most important application of silver nanoparticles is the medical industry such as topical ointments to prevent infection against burn and open wounds (Mahendran, 2013).

Sonochemistry involves using ultrasound techniques in chemical reactions. High surface area nanoparticles have been produced via ultrasonic irradiation. This sonochemical method has been prevent to be useful method for obtaining new materials. The chemical effects of ultrasonic waves arise from acoustic cavitation (Randa, 2014). Cavitation's bubbles collapse can also induce a shock wave in the solutions and drive the rapid impact of the liquid to the surface of the particles (Brajesh, 2014). Due to this, the bulk materials were converted into nanomaterials.

## 2. MATERIALS AND METHODS

**2.1. Chemicals:** Silver nitrate, Starch, Nutrient agar, Muller-Hinton broth, Sodium hydroxide.

**2.2. Synthesis of colloidal starch silver nanoparticles:** 20 ml of 1mM  $\text{AgNO}_3$  was mixed with 50 mg of starch. The pH was adjusted to 11.0 using 0.1N NaOH. Then the mixture was stirred for complete dissolution and agitated under sonication. The operating condition was 20 sec pulse on and 20sec pulse off time with amplitude of 68% at  $25^\circ\text{C}$  for 20 minutes (Brajesh, 2014).

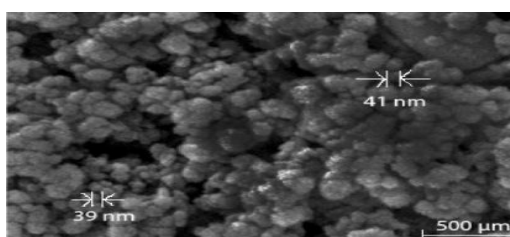
**2.3. Characterization of Starch-Ag nanoparticle:** The prepared solution was subjected to absorbance under UV-Visible spectrophotometer (Systronics 117) at wavelength varying between 200 to 750 nm. This was followed by analysis using SEM microscopy (SEM-Carl Zeiss EVO 18).

**2.4. Study of antimicrobial activity:** Five different organisms were chosen to study the antimicrobial activity namely *P.aeruginosa*, *N.meningitidis*, *B.cereus*, *E.coli* and *Brucella spp.*, which were isolated from the wounds of human subjects. The antimicrobial activity of the prepared colloidal starch-Ag nanoparticle was studied by Disc diffusion method using various concentrations of  $\text{AgNO}_3$  as positive control.

**2.5. Disc diffusion method:** The pure cultures of organisms were subcultured on Muller–Hinton broth at  $37^\circ\text{C}$  on rotary shaker at 210 rpm. Each strain was swabbed uniformly on the individual nutrient agar plates. 50  $\mu\text{l}$  of the crude solution of 0.1%, 0.5% and 1%  $\text{AgNO}_3$  was added into sterile circular disc of 6 mm diameter which were then placed into the nutrient agar plate. After incubation at  $37^\circ\text{C}$  for 24 hours, the different levels of zone of inhibition were measured. This same procedure was repeated for synthesized Starch-Ag NPs.

### 3. RESULTS AND DISCUSSION

The aqueous silver ions were reduced to silver nanoparticles by sonication when starch was added. The initial colour was white which turns into brownish yellow at the end of the process. This indicated the formation of Starch–Ag NPs. The formation and stability was studied under UV-Vis spectrophotometer and maximum absorbance was obtained at 435 nm. It is already reported that absorbance at 430 nm for silver is the characteristics of their noble particles (Nestor, 2008). The sample was then subjected to SEM analysis which showed the formation of starch–Ag NPs.



**Fig.1.SEM micrograph of Synthesized Ag-NP**

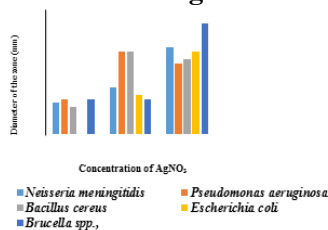
The antimicrobial study was done for starch–Ag NPs on five different organisms namely *N.meningitides*, *P.aeruginosa*, *E.coli*, *B.cereus* and *Brucella spp.*, showed zones ranging from  $5\pm 0.7$  to  $7\pm 0.8$  mm more than the zones obtained for  $\text{AgNO}_3$ , which was used as control. Ag has been used since a century in treatment of burns (Ghosh, 2012), yet the colloidal starch- NP as comparatively more effect in suppressing the growth of the pathogens under study.



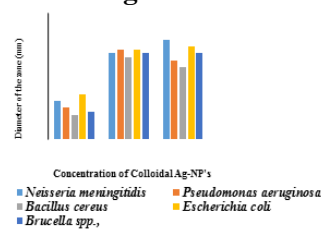
**Fig.2.Antimicrobial activity of  $\text{AgNO}_3$  solution**



**Fig.3.Antimicrobial activity of synthesized Starch-Ag NP**



**Fig.4.Antimicrobial activity of  $\text{AgNO}_3$  against selected pathogens**



**Fig.5.Antimicrobial activity of Colloidal Ag-NP's against selected pathogens**

This may be due to the fact that these particles are very small ( $41\pm 3$  nm), which is about 227 times smaller than the size of these bacteria. Therefore metal nanoparticles are harmful for bacteria (Chwalibog, 2010). This result corresponded to the dimension of the colloidal NPs (Kaviya, 2011) and their smaller size (Prashanth, 2011). These particles have high surface area so are very effective against microorganisms (Kvitek, 2008). As reported, these

colloidal nanoparticles release silver ions in the bacterial cells thereby attacking the respiratory chain, cell division and ultimately leading to death (Morones, 2005; Sondi, 2004).

#### 4. CONCLUSION

The colloidal starch-Ag NPs synthesized using starch and AgNO<sub>3</sub> by sonochemical method is economical, effective and eco-friendly. Analysis using UV-Vis spectrophotometer and SEM microscopy confirmed the reduction of AgNO<sub>3</sub> and formation of colloidal starch-silver nanoparticles. The zones of inhibition formed during antimicrobial studies corresponded to the antibacterial activity of the synthesized nanoparticles against the studied pathogens. The synthesized colloidal nanoparticle could be of immense use in formulation of topical creams and lotions.

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