

# Green Synthesis of Silver Nanoparticles Using Sodium Alginate and Lignosulphonic Acid Blends

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**Abstract:** A simple method based on the principles of green chemistry has been developed to synthesize stable silver nanoparticles (AgNP) for possible biomedical applications. Blend of sodium alginate (SA) and lignosulphonic acid (LS) prepared in the ratio of 80/20 mass percent respectively was used as reducing and stabilizing agent. This blend is biocompatible and has shown drug release ability under physiological conditions. Use of blend has an added advantage as LS has the ability to reduce silver while the blend matrix acts as a stabilizing agent. Effect of precursor concentration ( $\text{AgNO}_3$ ) and temperature was investigated. Progress of synthesis was monitored using UV-Vis spectroscopy. Higher temperature and lower silver nitrate concentration showed better synthesis of AgNP.

**Keywords:** Green synthesis, Lignosulphonic acid, Sodium alginate and Silver nanoparticles.

## 1. INTRODUCTION

Silver was known to human beings since ages for antibacterial properties. This image was lost after discovery of antibiotics like penicillin, cephalosporin etc. Indeed Antibiotics saved humanity from many diseases for decades but its overuse is now emerging as bacterial resistance towards these drugs. Failure of antibiotics is going to force humanity towards black age of medicine. Silver is again in news as antimicrobial agent and this time in the nanometer size range.

Particles having at least one dimension less than 100 nm are called nanoparticles [1-2]. Like other nanomaterials, Silver in this size range exhibit unique physicochemical properties and biological activities[3-5]. Having strong anti-microbial activity is a major reason for its application in biomedical field and in consumer applications. A wide category of consumer goods containing nanosilver are already available in market. In medical field, there are wound dressings, contraceptive devices, surgical instruments and bone prostheses coated or embedded with nanosilver [6-8]. Consumer products like room sprays, laundry detergents, water purifiers, wall paints, textiles like underwear socks etc containing nanosilver are being manufactured [10-11].

Successful application requires AgNP with suitable functionality, controlled size, shape and the distribution of size and shape. This makes synthesis process very important. Physical methods of synthesis like evaporation /condensation and laser ablation are commonly used methods[12-13]. These methods can produce monodisperse silver with purity but face challenges like scalability, specialized setup and high energy requirement. Chemical methods based on reduction of silver precursor to atomic silver by some reducing agent and then



stabilizing it by suitable stabilizing agents[14-15] are linked with environmental and health concerns. Contamination of nanoparticles, an undesirable attribute for biomedical application is another issue associated with chemical methods.

Recent studies have reported successful synthesis of crystalline polydispersed AgNP of size below 100 nm in aqueous medium using materials obtained from plants, fungi, algae etc with least of experimental setup. Role of phyto chemicals in the reduction of silver and stabilization of AgNP have been established. These nanoparticles have shown excellent antimicrobial ability[16].

Stabilization of synthesized nanoparticles is critical for its application. Polymer matrix like cellulose acetate hollow fiber membrane [17], porous ceramic composite [18], cotton fiber [19] etc have been reported as effective support materials. Biopolymers not only provide support and stabilization but have added advantage of being renewable, biodegradable and biocompatible. Thus AgNP embedded on biopolymer matrix materials have great applicability in biomedical field. Alginate, polysaccharide obtained from brown sea weed has been used in biomedical field due to hydrogel forming property and biocompatibility. It is a linear polymer of mannuronic acid and guluronic acid monomers. Its applications include wound management, tissue engineering, controlled drug release etc. Applications in wound healing is due to its ability to form gel on absorption of wound exudates, prevention from drying of wound surface [20] and increased healing of skin wounds[21]. It basically provides mechanical integrity in tissue engineering applications [22]. Incorporation of nanosilver into the SA polymer matrix has been reported by researchers. Arockianathan[23] in their experiment used NaBH<sub>4</sub> as reducing agent, Malkar et al[24] used Aminopolycarboxylic acids as reducing agents for in situ synthesis of AgNP.

LS is a natural biodegradable polymer available in plenty as a plant byproduct and is large tonnage wastes obtained from paper industry. SA/LS form stable blend in aqueous medium [25] with reduced crystallinity as compared to original polymers [26]. This blend has also shown good compatibility with Ciprofloxacin drug [27] and control release ability in a pH medium of 7 at a rate suitable for the biomedical applications [26, 28].

Owing to better property than individual polymers this blend has been used in this study to synthesize AgNP. This *in situ* synthesis with SA/LS (80/20) blend is performed without assistance of external reducing agent. Entire process has been done in aqueous medium. The blend matrix acts as a template for the synthesis of nanoparticle. Since studies have indicated that AgNP combined with various antibiotics show improved results than AgNP or antibiotic alone [29] this study can pave way for development of materials which can be used for loading of both drug and AgNP.

## 2. Materials and Methods

**Chemicals:** A seaweed product sodium alginate (SA) and plant byproduct Lignosulphonic acid (LS) are purchased from Sigma Aldrich Bangalore, India and silver nitrate from Ranbaxy Chemicals, India. Aqueous solutions of chemicals are prepared using distilled water. All chemicals are of analytical grade and used without any further purification.

**Preparation of SA/LS Blends:** 1% w/v aqueous solutions of SA and LS were prepared in 80/20 ratio and stirred using magnetic stirrer for 30 min.

**Preparation of AgNP:** The SA/LS blends solution is mixed with equal amount of silver nitrate (0.25%, 0.5%, 0.75% and 1%) solution. In this study, the AgNP are synthesised in the SA/LS matrix. In a typical synthesis 25 mL of SA/LS solution is added to a 25 mL aqueous solution of AgNO<sub>3</sub> in a 100mL beaker to obtain the AgNP at a temperature of 25°C and 40°C.

**Characterization Methods and Instruments:** The AgNPs synthesised are examined using ultraviolet-visible (UV-Vis) spectroscopy, The UV-Vis spectra were recorded over the range of 300-500 nm with a Shimadzu 2600 UV-Vis spectrophotometer.

### 3. Discussion

Since the blend prepared by mixing 80/20 mass percent of SA and LS had shown better properties for biomedical applications it was selected for *in situ* AgNP synthesis in order to explore it for possible application. AgNP Synthesis was done by addition of the mentioned SA/LS blend solution (1%) to different concentration (0.25, 0.50, 0.75 and 1 %) of AgNO<sub>3</sub> precursor. The effect of temperature and AgNO<sub>3</sub> concentration on the synthesis of AgNP was investigated using UV-Vis spectra. Earlier reports indicate that SA being a soft reducing agent cannot effectively reduce silver which requires higher potential for reduction [30]. Successful synthesis of AgNP using SA/LS blend, without any external reducing agent indicates that LS is reducing silver while SA/LS blend matrix is stabilizing AgNP.

As blend solution was added to 0.25, 0.50, 0.75 and 1.0 w/v percent solution of AgNO<sub>3</sub>, first visual observation was emergence of gel for the higher two concentrations especially more for 1% solution [Fig.1]. This observation can be explained by the fact that alginate can form silver alginate and this process can be more with higher Ag<sup>+</sup> ion concentration. Silver alginate is known to form gels in water. Ag<sup>+</sup> ions being large can create more interaction than Na<sup>+</sup> with the alginate molecules which may increase insolubility of SA [31] as shown in Fig.1.

Formation of AgNP can be observed by colour change as they exhibit Yellowish brown colour in aqueous solution due to excitation of surface plasmon vibrations in AgNP [32]. Colour change was not observed for synthesis at RT for very long period of time indicating poor synthesis of NP at lower temperature. At 40°C, characteristic colour immediately appeared after mixing of blend and silver nitrate solution which indicated that higher temperature is favouring synthesis of AgNP. Visual observation of colour change is shown in Fig.2.

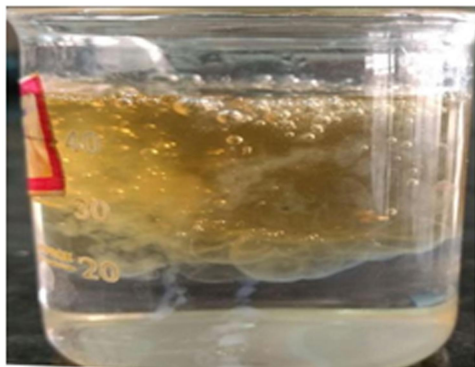


Fig.1: Gel formation immediately after addition of SA/LS blend in 1% AgNO<sub>3</sub> solution due to formation of silver alginate

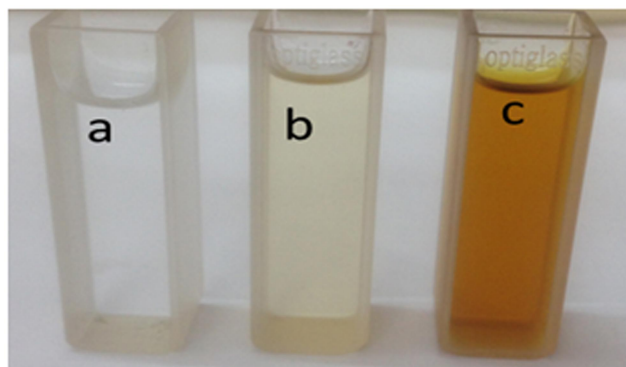


Fig.2: a) AgNO<sub>3</sub> solution b) SA/LS (80/20) blend c) AgNP in blend matrix

### Study of synthesis of AgNP using UV-Visible spectra

Formation and stability of AgNP in aqueous solution can be monitored by study of UV-Vis absorption spectra. Dependence of absorption spectrum of metal nanoparticles on particle size, shape and particle–particle interaction (agglomeration) with the medium [33] has made it possible to monitor synthesis of AgNP by study of UV-Vis spectra. Fig.3 shows the UV-Vis spectra of progress of synthesis of AgNP with AgNO<sub>3</sub> concentrations 0.25, 0.50, 0.75 and 1% and at two temperatures, room temperature (RT) and 40°C recorded at regular interval of time. The lower sets of curves in each graph are for RT while upper sets are for 40°C in Fig.3. The results reveal that the synthesis of AgNP using SA/LS blend is significantly faster at elevated temperature (40°C) than at RT. This could be due to faster reduction of silver ions at elevated temperatures. Here reduction of silver is carried out by LS and the synthesized NPs are stabilized in the SA/LS blend matrix.

There is no indication of AgNP synthesis for all samples at RT for first 3h as we can see no distinct peak for AgNP. A low intensity peak at 415nm was observed for 0.25% AgNO<sub>3</sub> concentration in the fourth hour which is an indication of beginning of synthesis of AgNP. Higher concentration of AgNO<sub>3</sub> did not show any sign of AgNP synthesis. The probable reason could be formation of alginate gel. Formation of gel not only prevents availability of stabilizing sites in the gel matrix but also entraps the LS thereby making it less available for reduction. Another factor that can operate here is increased viscosity. It is reported that high viscosity adversely affects silver synthesis in presence of SA [30]. Since synthesis of AgNP with 0.25% AgNO<sub>3</sub> concentration at RT started after 4h it was investigated for longer time. A distinct surface Plasmon resonance (SPR) peak at 432nm indicating synthesis of AgNP inside the blend matrix was observed at 24 h [Fig.4]. As the nucleation process was very slow from the beginning and the reaction took almost 24 h to complete it is expected that the size of synthesized AgNP would be large and the observance of shift in SPR peak from 415nm to 432nm is also supporting this fact.

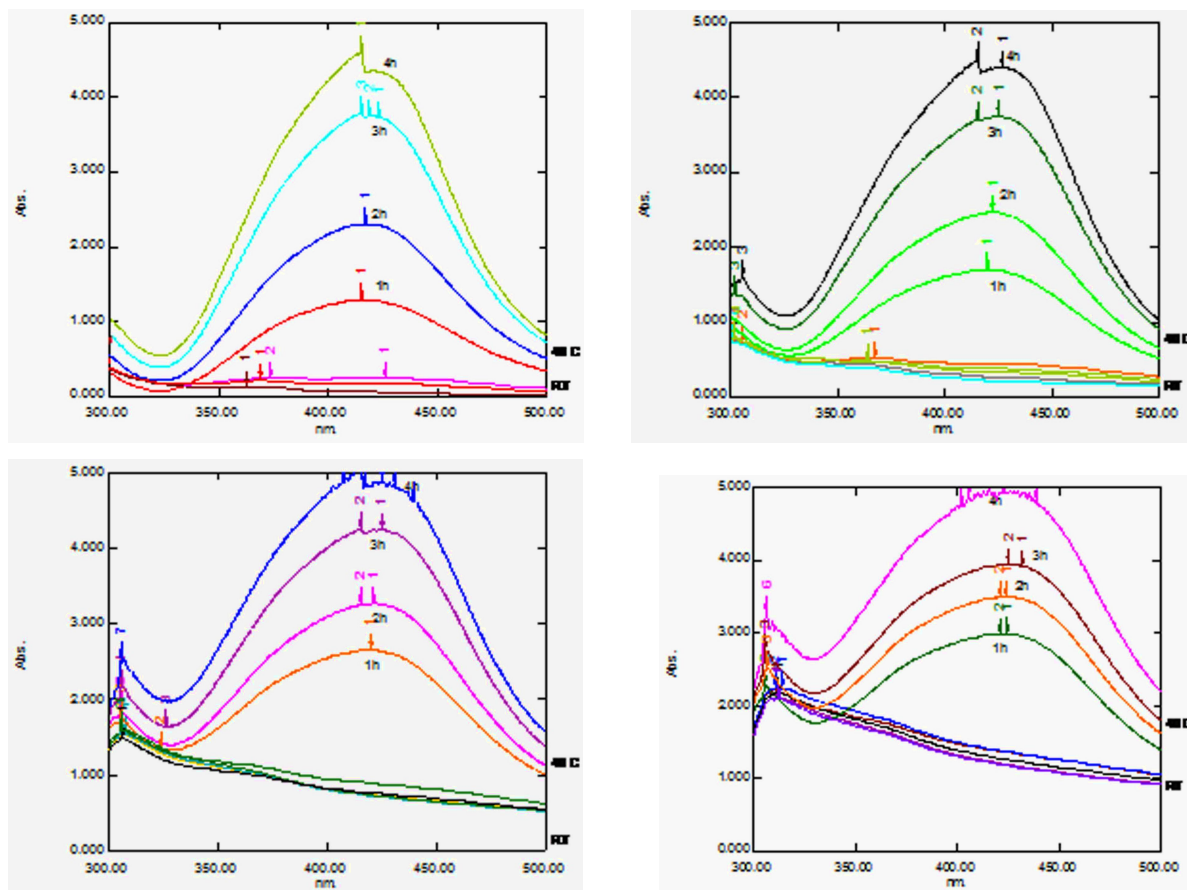


Fig.3: UV-Vis spectra of synthesis of AgNP in SA/LS (80/20) solution at RT and 40 C with different  $\text{AgNO}_3$  concentrations of a) 0.25% b) 0.5% c) 0.75% d) 1%

UV-Vis spectra at 40°C are in line with visual observations. The 0.25%  $\text{AgNO}_3$  sample clearly shows a peak at 415nm in 1h and at 417nm in 2h beyond this time peak is broadening which is an indication of agglomeration. For 0.50%  $\text{AgNO}_3$  concentration SPR peaks is observed at 420nm and 422nm in 1h and 2h respectively and later hours it shows broadening. 0.75%  $\text{AgNO}_3$  sample showed absorbance peak at 419nm in the 1h and then broadening of peak observed while 1%  $\text{AgNO}_3$  solution show broad peak in the very first h itself. Broad peak indicates presence of AgNP with different sizes probably due to agglomeration of silver particles which could be due to absence of adequate stabilization [34]. If we compare the absorbance which is an indication of degree of synthesis, it is clear that only 0.25% sample is able to give better synthesis in 2h as intensity of SPR bands is maximum. Observed poor synthesis of AgNP at higher  $\text{AgNO}_3$  concentration is again due to gelation related factors like unavailability of reducing group of LS and stabilizing SA/LS blend matrix and also increase in viscosity.

A comparison of UV-Vis spectra obtained for 0.25% sample was made for RT (24 h) and 40°C (2 h) [Fig.5]. Absorbance peak for 40°C is at 417nm while at RT is at 432nm. The shift toward the shorter wavelength with increase in temperature is characteristic of a reduction in nanoparticle's size, probably due to an increased nucleation because of faster reduction process. In contrast, increase in synthesis time results in further growth (Ostwald ripening) of



the NPs, which gives rise to a red shift of the SPR band (toward a longer wavelength)[38]. The absorbance peak, due to the surface SPR effect of metallic nanoparticles, is known to undergo a red shift following the particle size increase [39]. There is an increase in intensity at higher temperature which is indicative of more synthesis of AgNP at higher temperature. [37].

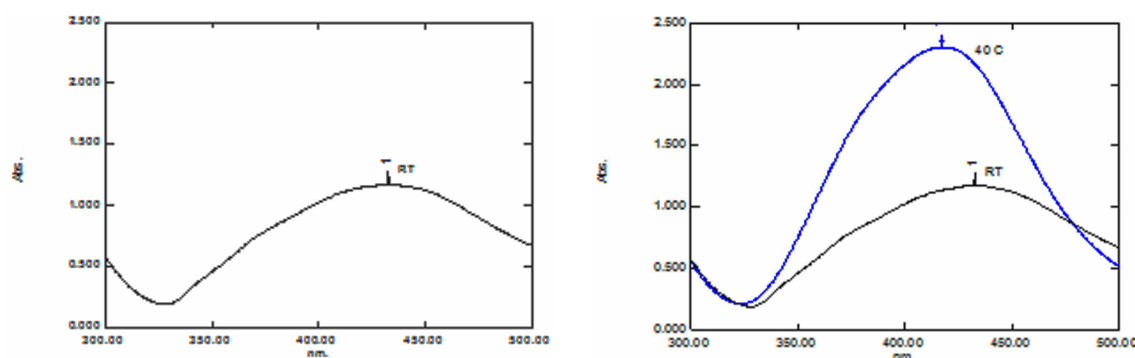


Fig.4: UV-Vis Spectra of a) Synthesis of AgNP at RT in 24h with 0.25%  $\text{AgNO}_3$  and RT in 0.25%  $\text{AgNO}_3$  .

b) Comparison of synthesis of AgNP at 24h and 40°C in 2h with 0.25%  $\text{AgNO}_3$  .

#### 4. Conclusion

Synthesis of *in situ* AgNP could be achieved without use of any reducing agent at 40°C. Effect of temperature and  $\text{AgNO}_3$  concentration was investigated. LS in the blend (SA/LS) can successfully reduce and blend matrix are able to stabilize the AgNP. It was observed that the concentration of the  $\text{AgNO}_3$  exerted a significant effect on the NP synthesis. Lower concentration of  $\text{AgNO}_3$  precursor is giving better synthesis results. Higher precursor concentration is showing increased gelling which acts as a retarding factor for synthesis. Finally the method is green as the reaction medium is water, no extra chemical is used for synthesis and both the reducing agent and stabilizing agents are product of nature, renewable, cost effective and biocompatible.

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