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Research Article

## Green synthesis of silver nanoparticles using water soluble gum of *Sterculia foetida* and evaluation of its antimicrobial activity

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### ABSTRACT

In the present study, green synthesis of Ag<sup>0</sup> nanoparticles using gum extract as reducing and capping agent was investigated. *Sterculia foetida* gum extract prepared at different concentrations (0.1% to 0.01%) was used for the biosynthesis of silver nanoparticles. The gum extract incubated with 1mM AgNO<sub>3</sub> and autoclaved for 15 mins showed gradual change in the colour from yellow to reddish brown indicating the formation of silver nanoparticles. Different spectral analyses were used for confirming nanoparticle formation. Sterile silver nanoparticles devoid of bacteria, viruses, and spores could be produced using this method. Antibacterial activity of the prepared nanoparticles was investigated and the synthesized nanoparticles showed inhibitory activity on Gram-positive and Gram-negative bacteria.

**Keywords:** Silver nanoparticles; *Sterculia foetida*; antibacterial activity

### 1. INTRODUCTION

Green synthesis of nanoparticles is advantageous when compared to chemical methods of synthesis as they involve the use of toxic chemicals. Decreased use of these environmental pollutants protects the environment. In these approaches water is used as a solvent and some biomolecules will be acting as both reducing and stabilizing agents. Many photochemicals are presently being used due to their potential health benefits. Biopolymer used for the production of nanoparticles (NPs) has attracted increasing attention (Ravindran et al., 2003) was the first to introduce the concept of green nanoparticles using 3-d-glucose and starch for synthesis of silver nanoparticles. Studies have also been reported silver nanoparticles formation using polymers such as chitosan (Modrzejewska et al., 2010), heparin (Kemp et al., 2009), Acacia (Mohan et al., 2007), gum kondagogu (Kora et al., 2012) and gum Arabic (Song et al., 2011).

*Sterculia foetida* L. is a tropical plant belonging to the Sterculiaceae family. Polysaccharides (guar gum, locust bean gum, semi-synthetic cellulose derivatives (cellulose ethers and cellulose esters), microbial polysaccharides, algal polysaccharides and animal polysaccharides

have been used earlier for the synthesis of nanoparticles. Synthetic polymers which were used include polyvinylalcohol, polyvinyl pyrrolidone and polyacrylic acid (Sintzel et al., 1996). Green synthesis of silver nanoparticles was also reported using soluble starch (Vigneshwaran et al., 2006). Microwave assisted green synthesis of silver nanoparticles using *Stigmaphyllon littorale* leaves and *Cuminum cyminum* through microwave irradiation, was investigated by Kudle et al. (2012, 2013). *Sterculia foetida* gum was earlier studied for controlled release of ocular dosage forms. In this perspective, a simple and green synthetic way for the production of silver nanoparticles using this gum was investigated as both the reducing and stabilizing agent. The results of the above study are presented in the communication.

### 2. MATERIAL AND METHODS

#### 2.1. Synthesis of silver nanoparticles

Silver nitrate (AgNO<sub>3</sub>) was purchased from Sigma-Aldrich. Bangalore, India. *Sterculia foetida* gum was powdered and then 0.1 -0.05, 0.04, 0.03, 0.02, 0.01 % (w/v) of homogenous gum stock solution was prepared by adding this powder to reagent bottle containing milli Q water and stirred overnight. Insoluble materials were centrifuged and the supernatant was used for other experiments. The AgNPs were synthesized by heating the silver nitrate solutions containing various concentrations of gum extract at 120°C at 15Lbs and for different percentage and different time intervals. The effect of concentration of gum and reaction time on nanoparticles synthesis was studied. Gum AgNPs

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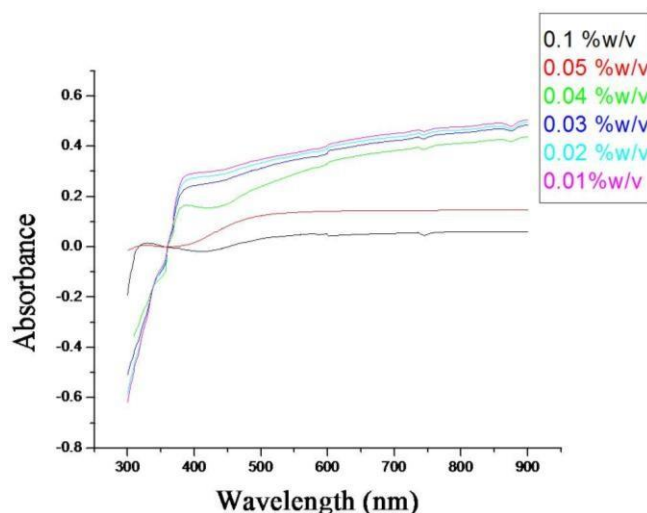


Figure 1: UV-vis absorption spectra of AgNPs

The UV-vis absorption spectra of AgNPs synthesized by autoclaving different concentrations (0.1%w/v to 0.05,0.04,0.03,0.02,0.01%w/v) of gum extract at 1 mM AgNO<sub>3</sub> concentration for 15 min after one 1hr .

Table 1: Absorbance of nanoparticles at different concentrations of the gum

S.No	Different con.c (w/v)	Wavelength (nm)	Absorbance
1	0.1%	414.17	0.0248
2	0.05%	459.66	0.1175
3	0.04%	444.63	0.1644
4	0.03%	439.97	0.2527
5	0.02%	432.64	0.3008
6	0.01%	431.97	0.3064

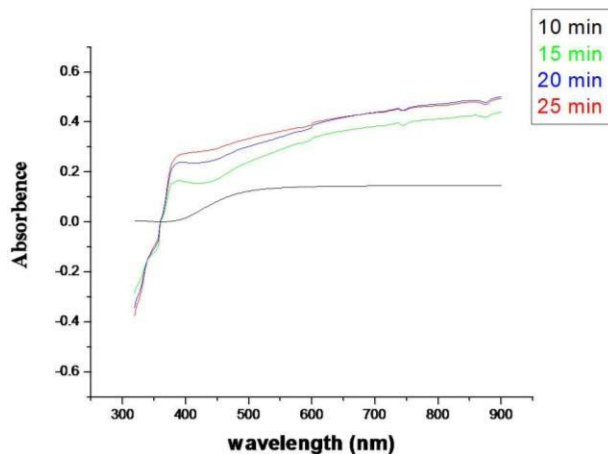


Figure 2: UV-VIS absorption spectra of AgNPs

characterization was done by UV-vis Spectroscopy, FTIR, TEM.

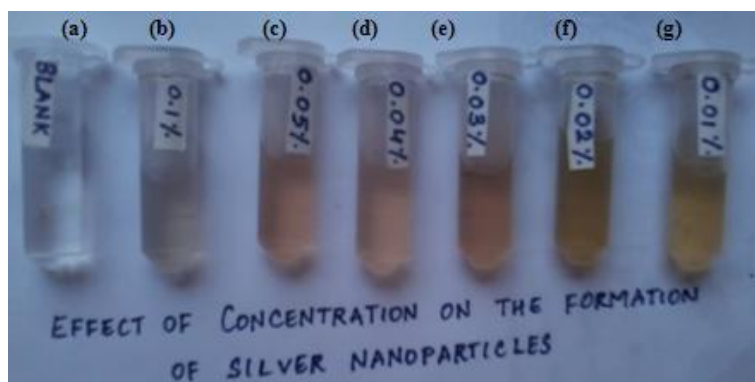
## 2.2. Antibacterial assay

Pure cultures of *Bacillus*, *E.coli*, *Pseudomonas*, *Klebsiella* and *Staphylococcus* were procured from the Department of Microbiology, Osmania University. The sensitivity testing of the plant Gum extract was determined by using disc diffusion method. Bacterial suspension was prepared by growing a single colony overnight in LB broth and by adjusting the turbidity to 0.5. Mueller Hinton agar plates were inoculated with this bacterial suspension and 2, 5  $\mu$ l ( $\mu$ g) of silver nanoparticles was added to the disc center with a diameter of 5

mm. The culture plates loaded with Ampicillin at a concentration of 5  $\mu$ g were included as positive controls. These plates were incubated at 37°C for overnight in a bacteriological incubator and the zone of inhibition was measured by subtracting the well diameter from the total inhibition zone diameter. Triplicate experiments were carried out with each bacterial strain.

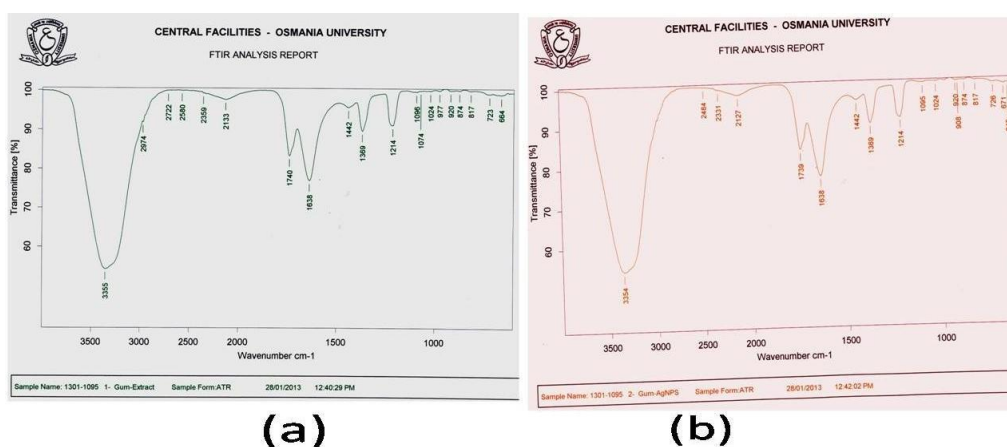
## 3. RESULTS AND DISCUSSION

UV-Visible spectra was used for the formation of nanoparticles in the range of 300 to 700 nm (Figure 1). The influence of parameters such as concentration of gum and reaction time were studied for optimization. The role of gum concentration on the synthesis was

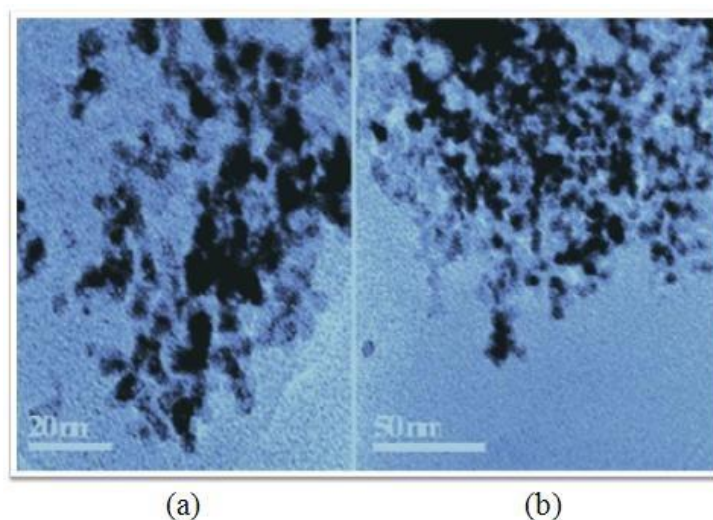


**Figure 3: Representative images of silver nanoparticle dispersions**

Representative images of silver nanoparticle dispersions synthesized with different concentration of gum extract solution at 120°C after a 15mins reaction time and purification by centrifugation (a) Blank, (b) 0.1% w/v, (c) 0.05 w/v, (d) 0.04%w/v, (e) 0.03%w/v, (f) 0.02w/v and (g) 0.01w/v.



**Figure 4: FTIR Spectra of Liquid (a) Aqueous extract of Gum Sterculia Foetida (b) Silver Nano Particles**



**Figure 5: TEM analysis for the size determination of (a) 0.05% gum silver nanoparticles (b) 0.01% gum silver nanoparticles**

studied by autoclaving these gum solutions (0.1–0.05 to 0.01%). UV–Vis spectra of nanoparticles with various concentrations of the gum from 0.1-0.05% was studied (Figure. 2). After autoclaving, yellow to reddish brown color was seen indicating the formation of nanoparticles by the gum extract. The efficiency of nanoparticle synthesis increased with increasing concentration of

gum extract. The synthesis was also evaluated by varying the reaction time of 10 to 25 min (Figure 3) and reduction was studied with 0.1 and 0.05% gum at 1 mM AgNO<sub>3</sub> (Figure 4.) The reduction capacity of the gum increased with increase in reaction time. In the UV-Vis spectra a single strong peak with a maximum around 431.97 nm was observed at 0.01% (Table 1).

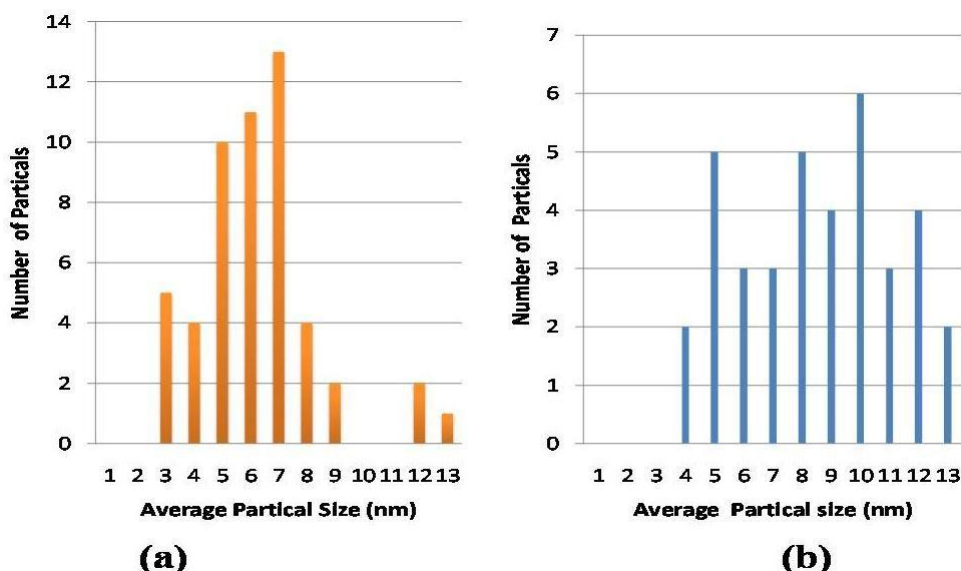


Figure 6: Particle Size Distribution different conc. (a) 0.01%w/v (b) 0.05%w/v of Silver Nanoparticles Synthesized by gum Extract

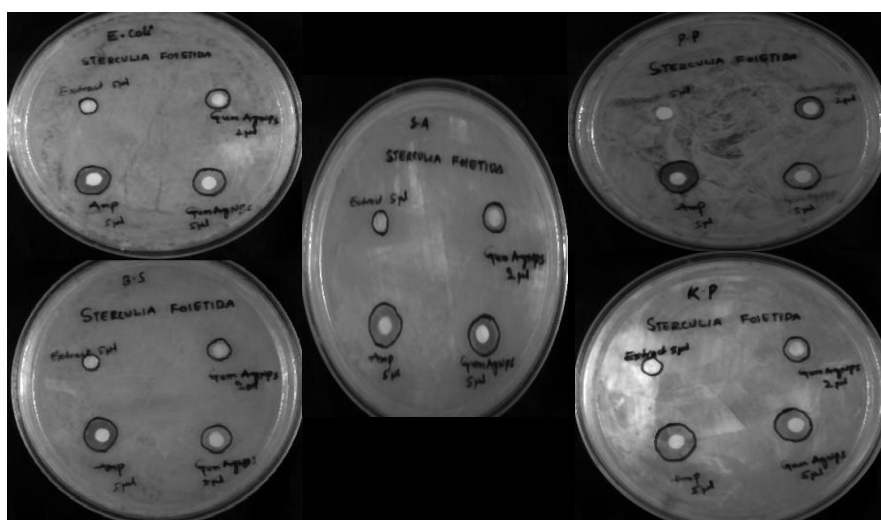


Figure 7: Bacterial culture plates showing the inhibition zones around the loaded wells.

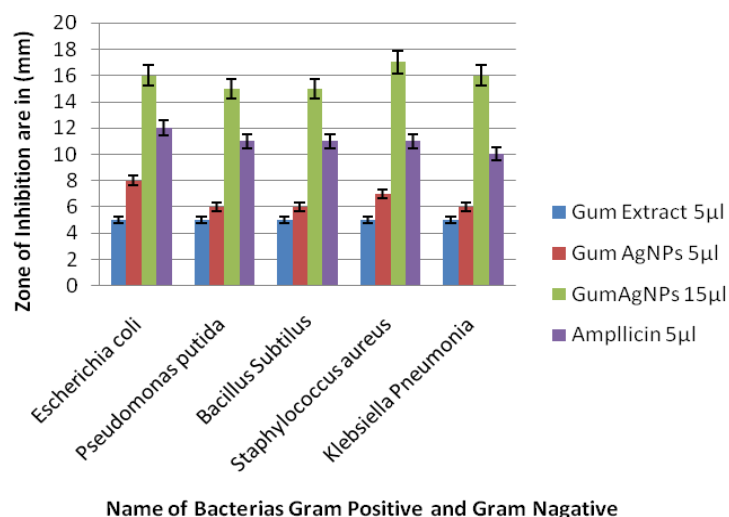
Left well: autoclaved gum extracts 5µl (0.01%w/v) and right two well: 2 µl (µg) and 5 µl (µg) of silver nanoparticles and stranded drug (Ampicillin, 5µl).

Table 2: Results of the anti bacterial activity of the synthesized silver nanoparticles Zone inhibition (mm)

Name of Bacteria's	Inhibition zones of AgNPs synthesized from the Gum Extract of sterculia foetida against bacterial species			
	GumExtract ( 5µl)	Gum AgNPs (5µl)	GumAgNPs (15µl)	Amplicine (5µl)
<i>Escherichia coli</i>	5	8	16	12
<i>Pseudomonas putida</i>	5	6	15	11
<i>Bacillus Subtilus</i>	5	6	15	11
<i>Staphylococcus aureus</i>	5	7	17	11
<i>Klebsiella Pneumonia</i>	5	6	16	10

The FTIR spectra of the gum extract and Gum -AgNPs were recorded in order to identify the functional groups of gum involved in the reduction and capping/stabilization of the synthesized nanoparticles. Figure 5a, b shows the FTIR spectra of the gum extract and silver nanoparticles. The spectrum of gum extract showed absorbance bands at 3355, 2722, 2133, 1740,

1638, 1442, 1369, 1214, 1074, 723 and 664  $\text{cm}^{-1}$ . The broad band observed at 3355  $\text{cm}^{-1}$  could be assigned to stretching vibrations of O-H groups in gum extract. Asymmetric stretching (2722), scissoring (1442), twisting and rocking vibrations (1214  $\text{cm}^{-1}$ ) of  $\text{CH}_3$  groups are seen. The broad band at 2133  $\text{cm}^{-1}$  only peak in the spectrum of gum could be assigned to various carbonyl



**Figure 8: Zone of inhibition are in (mm) of Autoclaved based synthesized silver nanoparticles against Gram positive and Negative bacterial pathogens**

species. The stronger band found at  $1638\text{ cm}^{-1}$  could be assigned to characteristic asymmetrical stretch of COO-group. The symmetrical stretch of carboxylate group can be attributed to the band present at  $1369\text{ cm}^{-1}$ . The peaks at  $1214$  and  $1074\text{ cm}^{-1}$  were due to the C–O stretching vibration of ether and alcoholic groups, respectively. The spectrum of Ag nanoparticles showed characteristic absorbance bands at  $3354, 2484, 2127, 1739, 1638, 1442, 1369, 1214, 1094, 729$  and  $649\text{ cm}^{-1}$ . Nanoparticles showed a new band at  $1739\text{ cm}^{-1}$  corresponding to carbonyl stretching vibrations in aldehydes, ketones, and carboxylic acids. Further, the occurrence of the peak at  $1739\text{ cm}^{-1}$  and disappearance of the peak at  $2127\text{ cm}^{-1}$  confirm that the reduction of the silver ions is coupled to the oxidation of the hydroxyl and carbonyl groups. The intense band at  $1094\text{ cm}^{-1}$  can be assigned to the C–N stretching vibrations of aliphatic amines. Study indicates that the carboxyl (–C=O), hydroxyl (–OH) and amine (N–H) groups of mulberry gum extract are mainly involved in reduction of  $\text{Ag}^+$  to  $\text{Ag}^0$  nanoparticles.

The TEM images of the silver nanoparticles synthesized with 0.01% gum and 1 mM  $\text{AgNO}_3$  autoclaved for 15 min are presented in the Figure 6a, b. The average size obtained was about 5–7nm and 5–10nm. Morphology of the average silver nanoparticles was investigated with 0.01% gum and 0.05% (w/v) and 1 mM  $\text{AgNO}_3$ . (Figure 7 a, b). The synthesized nanoparticles were stable for three months. In the present study, the AgNPs synthesized using gum extract exerted a fairly significant antibacterial action on the tested bacteria. Ampicillin was used as a reference drug. Table and Figure shows the zones of inhibition of *Escherichia coli*, *Pseudomonas putida*, *Bacillus subtilis*, *Klebsiella pneumonia* and *Staphylococcus aureus* against AgNPs, gum extract as control. A very small but noticeable zone of inhibition was observed for gum extract. Zones of 16 mm and 17mm were observed for *E. coli* and *Staphylococcus aureus*.

#### 4. CONCLUSION

The natural phytoexudate used in this study acts as a reducing and stabilizing agent. The wide occurrences of this plant and production of the gum in large amounts can be scaled up for large-scale production of silver nanoparticles. The inhibitory activity of the gum against Gram negative and Gram positive reveals the potential of stabilized AgNPs for application in tackling the problem of antibiotic resistance in microorganism.

#### 5. ACKNOWLEDGMENTS

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