Research Article

Phytomediated synthesis of silver nanoparticles using *Cassia auriculata* L: Evaluation of antibacterial and antifungal activity

T. S. Bhuvaneswari, T. Thirugnanam, V. Thirumurugan*

PG & Research Department of Chemistry, A.V.V.M Sri Pushpam College (Autonomous), Poondi, Thanjavur, Tamilnadu-613 503, India

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Abstract

Objective: Now a day's cost effective and environmentally friendly technologies for nano material synthesis have gaining attention in biosynthesis of nanoparticles. *Cassia auriculata* have been used traditionally in Tamilnadu for various ailments. The present study contains by using *Cassia auriculata* silver nanoparticles are biosynthesized from aqueous silver nitrate solution. **Material and Methods:** In 1mM silver nitrate solution plant flower extract is added after 12hrs the color change from dull yellow to blackish brown confirms the formation of nanoparticles. It is further confirmed and characterized through UV-VIS, FTIR, SEM, EDAX and XRD instruments. **Results and conclusion:** A peak at 452 nmconfirms the formation of nanoparticles, FTIR peaks confirm the capping of plant biomoleculs on silver nanoparticles, EDAX result confirmed reduction of silver nitrate to silver ions, SEM exhibits morphology and size of nanoparticles, XRD reveals the formation cubic structure. The nanoparticles proved to be it posses antibacterial and antifungal properties. In this study antibacterial and antifungal potentials are compared with standard chloramphenicol and nystatin.

Keywords: *Cassia auriculata*, bioreduction, nanoparticles, antimicrobial

Introduction

Noble metal nanoparticles gaining important in the past few years due to their applications.In the field of physics, chemistry, medicine, biology and material science (Yokohama and Welchons, 2007). There are many methods are available to synthesis nanoparticles including chemical reduction (Wang et al., 2005), electrochemical, photochemical (Khaydarov et al., 2009; Zaarour et al., 2014) and physical methods, such as physical vapor condensation (Simchi et al., 2007; Erico et al., 2017). Nanobiotechnology and their products derived are significant not only the treatment methodology but also due to their uniqueness particle size, physical, chemical, biochemical properties and broad range of applications (Banerjee et al., 2014).

Now a day's researchers inspired on biological system to develop nanoparticles using yeast, vitamin, sugar, microorganisms, plant or plant extract termed as green chemistry approach (Sinha et al., 2009; Mallikarjuna et al., 2011). If plant extract are used they can act as a reducing agent, but also stabilizing component for the system (Mittal et al., 2013). In the current investigation, the silver nanoparticles are successfully synthesized using *Cassia* (senna) *auriculata* flower extract. In the synthesized nanoparticles characterized by various instrumental techniques such as UV-Vis, FTIR, XRD, EDAX&SEM and to evaluate antimicrobial activity using human pathogens (Dongamanti et al., 2017; Zhang et al., 2010; Sun et al., 2000).

Material and methods

Collection of plant materials

Cassia auriculata (other name: *Senna auriculata*) flowers are collected from Nangikottai of Thanjavur, Tamilnadu, India. The voucher specimen is authenticated by the staff of Rapinat Herberium, St, Joseph's College, Thiruchirapalli, Tamilnadu, India.

Address for Corresponding Author:
Dr. V. Thirumurugan
Assistant professor
PG & Research Department of Chemistry,
A.V.V.M. SriPushpam College (Autonomous), Poondi, Thanjavur (Dt) - 613 503 Tamil Nadu, India.
Email: drv.thirumurugan@gmail.com

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The extraction and synthesis of nanoparticles

The 10g of flower powder with 100mL of deionized water in 250 mL of Erlenmeyer flask and boiled for 10 min.This aqueous extract was separated for the reduction of silver ions to silver. 10 mL of flower extract is mixed to 90mL of 1mM aqueous of $AgNO_3$ and kept in a dark room for 12 Hrs.

UV-Vis spectra analysis

The reduction of pure silver to silver ions is monitored by measuring the UV-Visible spectrum of the reaction mixture after diluting a small aliquot of the sample with distilled water. UV-Vis spectral analysis is done by using PerkinElmer -Model-Lambda 365 in 200 to 1200 nm.

FTIR analysis of dried biomass after bioreduction

The solution is centrifuged at 18000 rpm for 20 min and the resulting suspension is redispersed in 10 ml sterile distilled water. The centrifuging and redispersing process is repeated two times. The purified suspension is freezed dried to obtain dried powder. The dried nanoparticles are analyzed by FTIRthe range of 400-4000 cm^{-1}Perkin Elmer -Model - Spectrum TWO.

XRD analysis

X-ray diffraction (XRD) measurement is carried out by Rigaku X-ray diffractometer (Model: ULTIMA IV, Rigaku, Japan) with CuKα X-ray source (λ = 1.54056 Å) at a generator voltage 40 kV, a generator current 40 mA with the scanning rate 2° min−1.

Scanning Electron Microscopy –Energy Dispersive Spectroscopy

The morphology and size of the synthesized silver nanoparticles are identified by using Scanning Electron Microscope (SEM) Hitachi S-4500 SEM Analytical Field Emission Scanning Electron Microscope (FESEM). The EDAX microanalysis system (Oxford instrument) which automatically identifies the elements which are corresponds to the peaks in the energy distribution.

Antimicrobial activity

The antimicrobial activity of silver nanoparticles using *Cassia auriculata* has been studied by well diffusion methods (Patra et al., 2011; Kamaraj et al., 2011). For this sterilized lab wares are used to perform fresh culture of bacterial and fungal study. In the study two bacterial strains namely *Staphylococcus aureus, Pseudomonas aeruginosa* and two fungal strains *Trichophyton megnini, Candida albicans* are studied. For this four media plate are prepared by inoculation of sterilized nutrient agar media and with 50µL of above mentioned bacteria to each plate and similarly fungal strains to each plate individually.The wells are prepared, the control, aqueous flower extract of the sample, nanoparticles sample,control andstandards are added to each plate. The chloramphenicolis used as a standard for bacterial strains and nystatin is used as a fungal strain then the plates are incubated at 37°C for 12hrs.

Statistical analysis

All data were analyzed statistically by following student't' test.

Results and discussion

The *Cassia auriculata* flowers extract is added to 1mM of silver nitrate solution, the reduction of silver ion into silver is seen by color change of solution from dull yellow to blackish brown (Figure 1) The result are consonance with earlier research works (Dongamanti et al., 2017; Thirumurgan et al., 2010; Manimegalai et al., 2015; Anandalakshmi et al., 2016). The biosynthesized nanoparticles are confirmed by the absorption spectra at 452nm by UV-Visible spectroscopy.It is the preliminary confirmation for formation of silver nanoparticles (Figure 2). The result are similar to earlier reports in the literature (Vijayakumar et al., 2018; Leema Rose et al., 2017; Klaus et al., 1999; Muchanyereyi-Mukaratirwa Netai et al., 2017; Ahmed Set al., 2016). The FTIR analysis is used to confirm the extract is the cause for formation of silver nanoparticles and capping of extract molecules by the functional group present in the extract. The peaks at 1111 cm^{-1} denotes –CO-OC-linkage, 1613 cm^{-1} correspond to amide C-O stretching vibration. A broad band 3387 cm^{-1} is due to NH stretching vibration group NH_2 and OHoverlapping of stretching vibration of attributed for water and *Cassia auriculata*. The peak at 2927 cm^{-1} may be due to CH bond of CH_2.The absorbed peaks are may be due to phytoconstituents like terpenoids and flavonoids present in the plant extract (Figure 3) (Anandalakshmi et al., 2016; Chanda 2014; Bharathi et al., 2014).

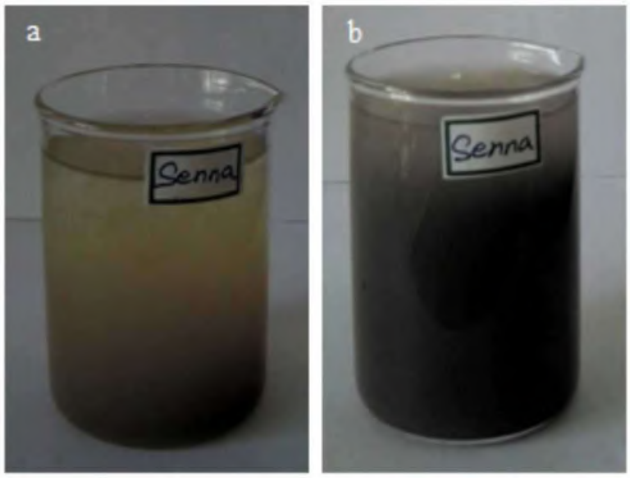

Figure 1. The conversion of silver nitrate to nanosilver by *Cassia auriculata* flower extract (a) at starting stage (b) after 12hrs.

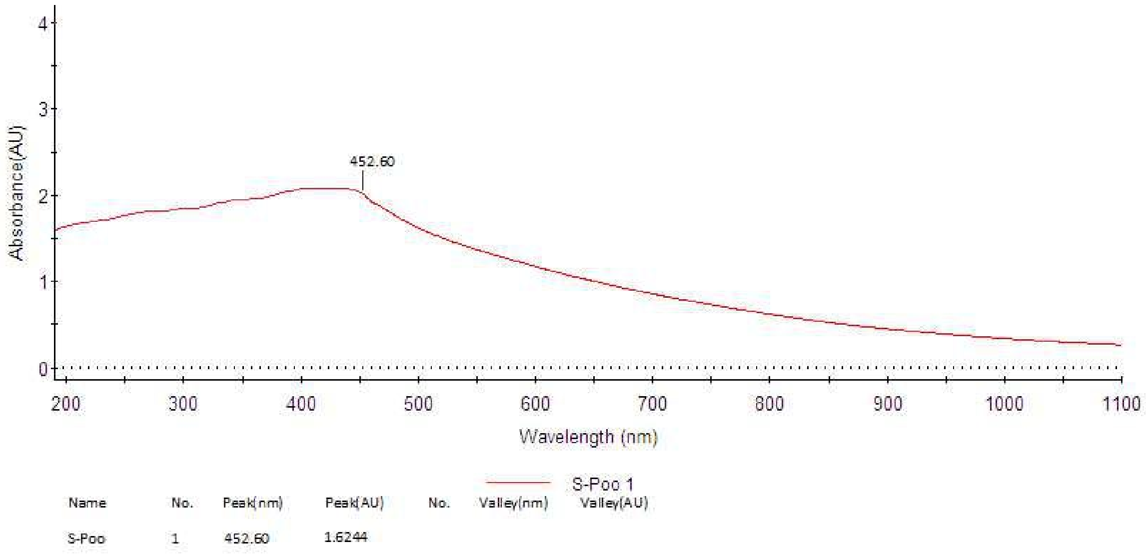

Figure 2. UV Spectra of synthesized nanoparticles using *Cassia auriculata* flower extract

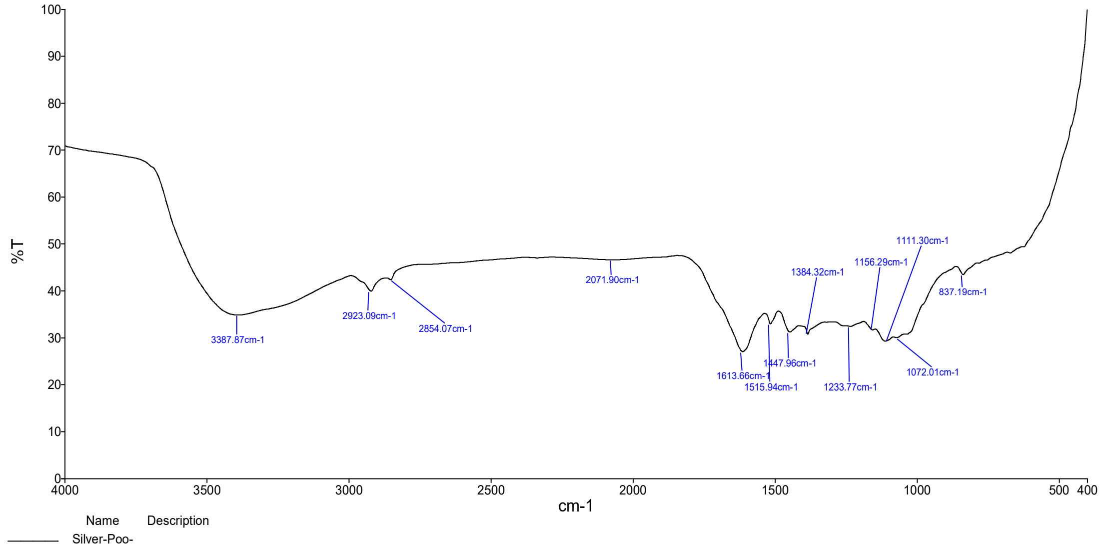

Figure 3. FTIR spectra of synthesized silver nanoparticles with capping of *Cassia auriculata* flower extract

SEM analysis and EDAX analysisof silvernanoparticles

The synthesized nanoparticle using *Cassia auriculata* flower extract SEM image is shown in (Figure 4). The nano images are clear having randomized biscuit like crystal structure showed the particle size of about 50 to 100 nm. EDAX confirms the reduction of silver from silver nitrate which is shown in (Figure 5).

XRD analysis

XRD pattern confirms the structure by JCPDS card number 04-0783. The peaks are observed at $2\theta \approx$ 38.31,46,14 and 77.45 corresponding to the plates 111, 200, 311 which confirm the cubic structure (Figure 6).

Antimicrobial activity

The antimicrobial activity is evaluated for control, plant flower extract, synthesized nanoparticles and standard. For antibacterial study *Staphylococcus aureus*, *Pseudomonas aeruginosa*

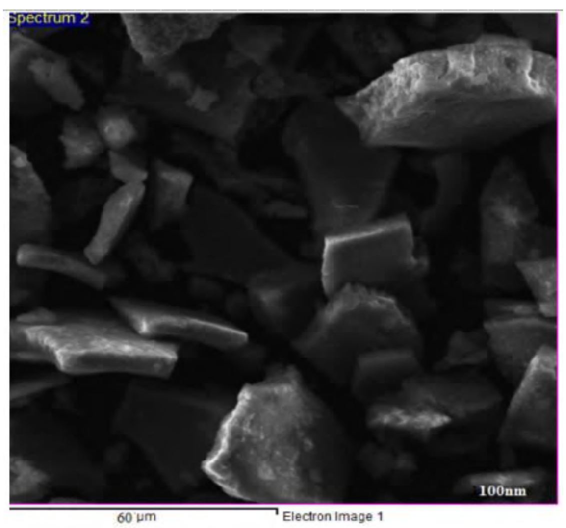

Figure 4. FE-SEM of synthesized silver nanoparticles using *Cassia auriculata* flower extract

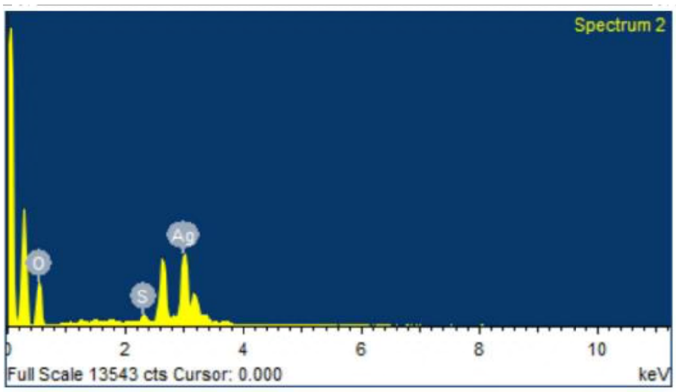

Figure 5. EDAX of synthesized silver nanoparticles using *Cassia auriculata* flower extract

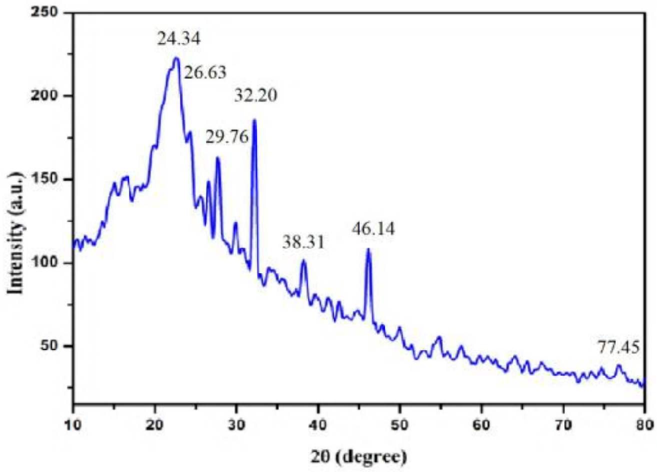

Figure 6. XRD of synthesized Silver nanoparticles using *Cassia auriculata* flower extract

bacterial strains and standard chloramphenicol are used. For fungal study *Trichophyton megnini, Candida albicans*and standard nystatin are used. The zone of diffusion is measured after 12hrs of incubation at 37°C. For gram positive bacterial strains *Staphylococcus aureus* and *Pseudomonas aeruginosa* strains have more or less same zone of inhibition for aqueous extract of *Cassia auriculata,* silver nanoparticles and standard chloramphenicol. This information gives that nanoparticles have

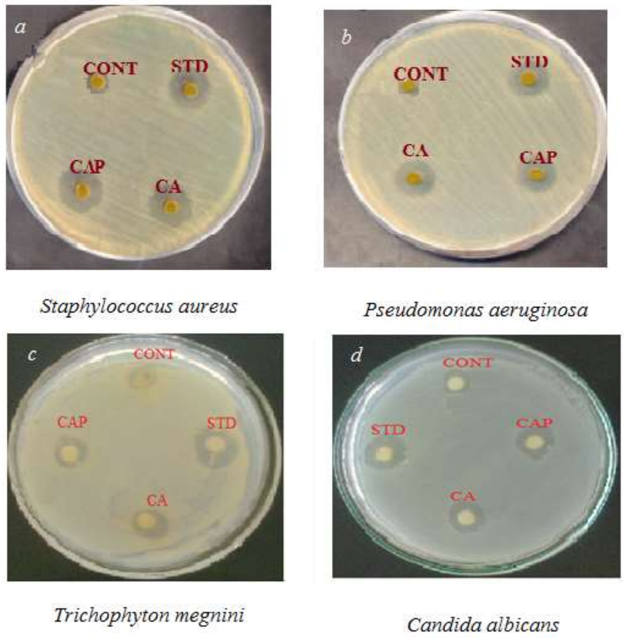

Figure 7. Antimicrobial activity ofsynthesized silver nanoparticles using *Cassia auriculata* (CAP), *Cassia auriculata* aqueous extract (CA), and standard Chloramphenicol for a & b and nystatin for c & d (STD)

same effect in both gram positive and gram negative strains.For antifungal activity the result are very fruitful in controlling the fungal pathogens whereas when comparing to standard nystatin, the standard has little bit more pronounce result (Figure 7). The results are found to be statistically significant.

Conclusion

The above biogreen methodology for synthesizing nanoparticles is easy technique, simple, low cost technology and eco-friendly. The phytoconstituents like alkaloid, flavonoid, terpenoid present in the plant extracthave been considered to be responsible for the bioreduction process. The results proven that the synthesized nanoparticles from *Cassia auriculata* plant flower extract and aqueous extract posses both antibacterial

Table 1. Antimicrobial activity of synthesized silver nanoparticles using *Cassia auriculata*

Organisms	Control	Water extract CA (mm)	Nanoparticles CAP(mm)	Chloromphenical (Standard)
Antibacterial				
Staphylococcus aureus	0	30±0.49	30 ± 0.70	32 ± 0.77
Pseudomonas aeruginosa	0	31 ± 0.42	31 ± 0.63	33 ± 0.65
Antifungal				**Nystatin (Standard)**
Trichophyton megnini	0	25±0.13	26 ± 0.18	30 ± 0.29
Candida albicans	0	30 ± 0.45	32±0.37	34 ± 0.55

and antifungal activity. Among the two activity, nanoparticls are more potent in antibacterial effect than antifungal effect. This technology infuture will have more potential application in biomedical fields.

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