SYNTHESIS OF PLANT-MEDIATED SILVER NANOPARTICLES USING *FICUS MICROCARPA* LEAF EXTRACT AND EVALUATION OF THEIR ANTIBACTERIAL ACTIVITIES

P. Shanmuga Praba,[a] V. S.Vasantha,[b] J . Jeyasundari[a]* and Y. Brightson Arul Jacob[c]

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In the present work, synthesis of silver nanoparticles has been done using a particular variety of medicinal plant extract. Silver nanoparticles have unique optical, electrical, and thermal properties that play an indispensable role in drug delivery, diagnostics, imaging, sensing, gene delivery, artificial implants and tissue engineering. . We used an environmentally friendly extracellular biosynthetic technique for the production of the AgNPs. The reducing agents used to produce the nanoparticles were from aqueous extracts made from the leaves of *Ficus microcarpa*. We have synthesized silver nano particles by adding the 80 ml of 1 mM silver nitrate solution into the plant extract and characterized by UV-Vis absorption spectroscopy, Infra-red spectroscopy. The UV-Visible spectrum showed a peak at 451 nm corresponding to the Plasmon absorbance of the AgNPs. Moreover, use of plant extracts also reduces the cost of microorganism isolation and culture media enhancing the cost competitive feasibility over nanoparticle synthesis by microorganisms. The antibacterial efficacy also determined by disc diffusion method with *Bacillus cereus*, *Escherchia coli*, *Klebsiella pneumonia, Staphylococcus aureus* and it showed that high level of inhibition. Further, efficient antimicrobial activity of the synthesized silver nanoparticles proves the application potential of green synthesis in the area of nano-medicine

* Corresponding Authors
E-Mail:jjsundari16@gmail.com
[a] PG & Research Department of Chemistry, NMSSVN College, Nagamalai, Madurai-625019, Tamilnadu India.
[b] School of Chemistry, Department of Natural Products Chemistry, Madurai Kamaraj University, Madurai-625021, Tamilnadu, India.
[c] PG & Research Department of chemistry, The American College, Madurai-625002, Tamilnadu, India.

Introduction

In recent years, like other technology developments nanotechnology also expected to grow based on their demand and its wider applications and the number of research being conducted in this filed is rapidly growing throughout the world.[1] Nano technologydeals with the development of nanometer sized materials.[2] In the filled of nanotechnology different concepts of engineering, electronics and material science are applied in molecular or submicron level.[3] Particles with a size up to 100nm are usually referred as nanoparticles and they exhibit completely new particles based on their size,distribution and morphology.[4] A strong surface plasmon resonance absorption is exhibited in the UV-Visible region by the metallic nanoparticle.[5] Metallic nanoparticles are exploited widely because of their excellent antibacterial properties.[6] Silver nanoparticles are used in the development of new technologies in the areas of electronics material science and medicine and because of their extensive applications in various areas more research is being conducted on the silver nanoparticles by the scientists throughout the world.[7] Biological methods for the production of nanoparticles are considered as a safe and environment friendly and it is a cost effective method and toxic chemicals in completely eliminated.[8]

Green nanosynthesis has been proposed for the synthesis of nanoparticles through biological route.[9] The bio synthesis of silver nanoparticles from many plants such as *Euphorbiahitra*,[10] *Coriandrumsativum*,[11] *Solanumtorvum*,[12] have been reported.

In this paper, we report a low-cost convenient green synthesis approach to obtain large quantities of silver nano particles by reduction of silver ions with using *Ficus microcarpa*.

Figure 1. Picture of *Ficus microcarpa* plant

In this communication we report a green method for the synthesis of silver nanoparticles at room temperature by using plant extracts of *Ficus microcarpa* as reducing/stabilizing agents and the probable mechanism for the formation of NPs.

Materials and Methods

Reagents and chemicals: Silver nitrate and all analytical grade chemicals were purchased from TCI Chemicals, Chennai. Freshly prepared triple distilled water was used throughout the experiment.

Preparation of leaf extract by boiling method

Indian medicinal plant, *Ficus microcarpa,* was selected from NMSSVN College campus, Tamilnadu, India, on the basis of cost-effectiveness, ease of availability and medicinal property. Fresh and healthy leaves were collected locally and rinsed thoroughly first with tap water followed by distilled water to remove all the dust and unwanted visible particles, cut into small pieces and dried at room temperature.

About 10 g of these finely incised leaves of each plant type were weighed separately and transferred into 250 mL beakers containing 100 mL distilled water and boiled for about 20 min. The extracts were then filtered thrice through Whatman No. 1 filter paper to remove particulate matter and to get clear solutions which were then refrigerated (4 °C) in 250 mL Erlenmeyer flasks for further experiments. In each and every steps of the experiment, sterility conditions were maintained for the effectiveness and accuracy in results without contamination.

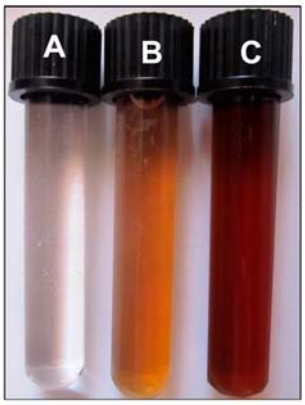

Figure 2. Formation of silver nanoparticles and it is identification by the colour change. (A) Silver nitrate solution, (B) plant extract without silver nitrate, (C) plant extract with silver nitrate solution

UV-Vis spectra Analysis

Samples (1 mL) of the suspension were collected periodically to monitor the completion of bioreduction of Ag^+ in aqueous solution, followed by dilution of the samples with 2 ml of deionized water and subsequent scan in UV-visible (vis) spectra, between wave lengths of 200 to 700 nm in a spectrophotometer (Beckman - Model No. DU - 50, Fullerton), having a resolution of 1 nm.

FTIR analysis

FTIR analysis of the dried Ag NPs was carried out through the potassium bromide (KBr) pellet (FTIR grade) method in 1:100 ratio and spectrum was recorded using Jasco FT/IR-6300 Fourier transform infrared spectrometer equipped with JASCO IRT-7000 Intron Infrared Microscope using transmittance mode operating at a resolution of 4 cm^{-1}.

Estimation of Antibacterial activity

The anti-bacterial activity was done on human pathogenic *Bacillus cereus, Escherichia coli, Klebsiella pneumonia, and Staphylococcus aureus* by the standard disc diffusion method. The discs were soaked with double distilled water, plant leaf extract, silver nitrate solution and solution containing silver nanoparticles of each type separately. Then the discs were air dried in sterile condition. Nutrient agar (NA) plates were seeded with 8h broth culture of different bacteria. In each of these plates, well were cut out using sterile corborer. Using sterilized dropping pipettes, different concentrations (10, 20, 30, 40 µl/well) of sample was carefully added into the wells and allowed to diffuse at room temperature for 2 h. The plates were then incubated at 37 °C for 18-24 h. Gentamicin (10 µg) was used as positive control. Then, the maximum zone of inhibition were observed and measured for analysis against each type of test microorganism

Results and Discussion

Synthesis of silver nanoparticles

Ficus microcarpa extract is used to produce silver nanoparticles in this experiment Ag^+ ions were reduced to Ag nanoparticles when plant extract is mixed with $AgNO_3$ solution in 1:8 ratio reduction is followed by on immediate change in yellowish to brown color in the aqueous solution of the plant extract due to excitation of surface plasmon vibration in silver nanoparticle.[18] Further formation of AgNPs in aqueous extract can be monitored by color change. Fig. 2. Shows the color changes when the aqueous extract of *Ficus microcarpa* plant was mixed with an $AgNO_3$ solution. The mixture was kept at room temperature for 24 hours. The appearance of a yellowish-brown color in the reaction vessel indicated formation of AgNPs. AgNPs exhibit this yellowish-brown color in aqueous solution due to excitation of surface plasmon resonance in the AgNPs.

UV-Vis spectroscopy analysis

Silver nanoparticles (AgNPs) appear yellowish brown in colour in aqueous medium as a result of surface plasmon vibrations.[13] As the different leaf extracts were added to aqueous silver nitrate solution, the colour of the solution changed from faint light to yellowish brown to reddish brown and finally to colloidal brown indicating AgNP formation. Similar changes in colour have also been observed in previous studies and hence confirmed the completion of reaction between leaf extract and $AgNO_3$. This was confirmedby the UV-Vis spectrograph of the colloidal solution of silver nanoparticles has been recorded as a function of time is shown in Fig. 3 and 4. Absorption spectra of AgNPs formed in the reaction media has absorption peak at 451 nm due to surface plasmon resonance of AgNPs. The UV-vis spectra recorded, implied that most

rapid bioreduction was achieved using banana many leaf extract as reducing agent. This was denoted by broadening of the peak which indicated the formation of polydispersed large nanoparticles due to slow reduction rates. The UV-vis spectra also revealed that formation of AgNPs occurred rapidly within the first 15 mins only and the AgNPs in solution remained stable even after 24 h of completion of reaction.

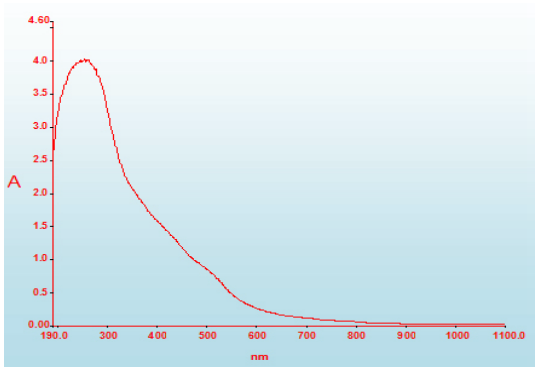

Figure 3. UV-Vis Absorption Spectra of *Ficus microcarpa* leaf extract

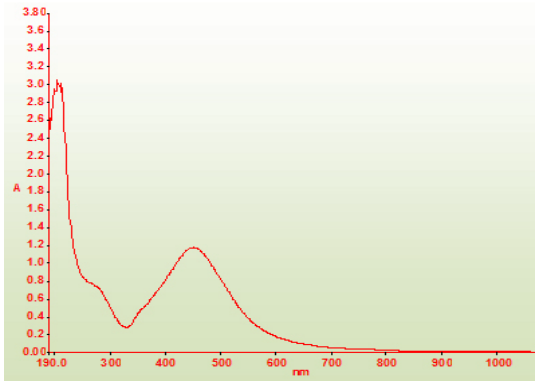

Figure 4. UV-Vis Absorption Spectra of Silver nanoparticles synthesized using *Ficus microcarpa* leaf extract

FTIR Spectroscopy Analysis

The FTIR spectrum of Ag nanoparticle is shown in Fig 5. The IR spectrum of Ag nanoparticles shown band at 3474 m^{-1}, 1638 cm^{-1}, 1455 cm^{-1}, 1079 cm^{-1} corresponds to O-H stretching, H bonded alcohols and phenols, Carbonyl stretching, N-H bond 1°amines correspons to C-N stretching of the aromatic amino group and C-O stretching alcohols, ethers respectively. FTIR spectrum of Ag Nano particles suggested that Ag nanoparticles were surrounded by different organic molecules such as terpenoids, alcohols, ketones, aldehydes and carboxylic acids. Fig 5 shows the plant *Ficus microcarpa* has been effectively involved in the antibacterial activity and also which is used in syntheses the silver nanoparticles

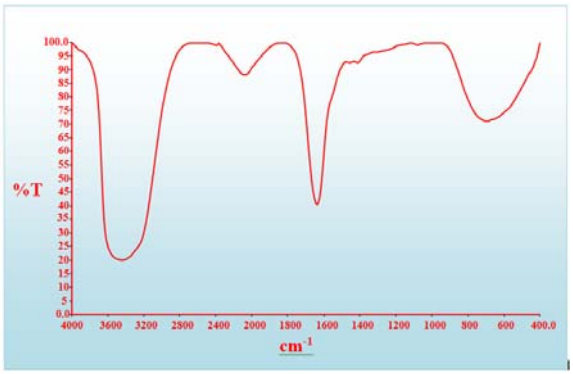

Figure 5. FTIR spectra of silver nanoparticles synthesized using *Ficus microcarpa* leaf extract

Antibacterial activity

The anti bacterial activity was done on human pathogenic *Bacillus cereus, Escherchia coli, Klebsiella pneumonia, Staphylococcus aureus* by the standard disc diffusion method. (Fig. 6.)

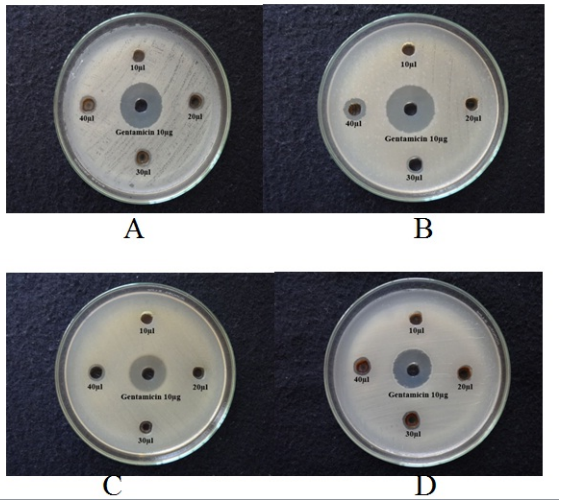

Figure 6. Antibacterial activity of synthesized silver nanoparticles (*Ficus microcarpa* leaf extract) on a) human pathogenic *Bacillus cereus*, b) *Escherchia coli*, c) *Klebsiella pneumonia*, d) *Staphylococcus aureus* by the disk diffusion method.

Table 1. Effect of silver nanoparticles on human pathogens

Pathogens	Zone of Inhibition	
	Plant silver	Gentamicin
Bacillus cereus	10 mm	24 mm
Escherchia coli	12 mm	26 mm
Klebsiella pneumonia	12 mm	26 mm
Staphylococcus aureus	14 mm	28 mm

Conclusion

The present study concluded that the plant *Ficus microcarpa* can be used as an excellent source for synthesizing the silver nanoparticles. The primary

conformatory for the silver nanoparticles was color change and UV-Vis absorption spectra of silver nanoparticles formed (451 nm) and FTIR spectroscopy confirms the presence of silver nanoparticles as well.

The green synthesized nanoparticles have more effective antibacterial activity to the pathogens. So green synthesis of nanoparticles can be ecofriendly involved in the many applications of clinical, electronics and etc.

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References

[1]Roco, M. C., *Curr. Opin. Biotechnol*., **2003**, *14,* 337–346.

[2]Zhang, L., Gu, F. X., Chan, J. M., Wang, A. Z., Langer, R. S., Farokhzad, O. C., *Clin. Pharmacol. Ther*., **2008**, *83*, 761–780.

[3]Daniel, M. C., Astruc, D., *Chem. Rev*., **2004,** *104*, 293–346.

[4]Wong, T. S., Schwaneberg, U., *Curr. Opin. Biotechnol,*. **2003,** *14,* 590–596.

[5]Callegari, A., Tonti, D., Chergui, M., *Nano Lett*., **2003**, *3*, 1565–1568

[6]Tsuji, M., Hashimoto, M., Nishizawa, Y., Tsuji T., *Chem. Lett*., **2003,** *32*, 1114–1115.

[7]Kundu, S., Maheshwari, V., Saraf, R., *Nanotechnology*., **2008**, *19(6)*, 065604.

[8]Okitsu, K., Mizukoshi, Y., Yamamoto, T. A., Maeda, Y., Nagata, Y., *Lett. Materials*., **2007**, *61*, 3429–3431.

[9]Narayanan, K. B., Sakthivel, N., *Adv. Colloid. Interface. Sci*., **2010**, *22(156)*, 1–13.

[10]Gan, P. P., Ng, S. H., Huang Y., Li, S. F., *Bioresour. Technol*., **2012**, *113*, 132–135.

[11]Raveendran, P., Fu, J., Wallen, S. L., *Am. Chem. Soc.,* **2003**, *125(46)*, 13940–13941.

[12]Sharma, H. S., Ali, S. F., Hussain, S. M., Schlager, J. J., Sharma, A., *J. Nanosci. Nanotechnol*., **2009,** *9(8)*, 5055–5072.

[13]Narayanan, S., Sathy, B. N., Mony, U., Koyakutty, M., Nair, S. V., Menon, D., *ACS Appl. Mater. Interfaces*., **2012**, *4(1)*, 251–260.

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