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Biogenic Synthesis of Silver Nanoparticles Using *Tectona grandis* Leaf Extract and evaluation of their Antibacterial Potential

Raju Nalvothula[1], Veera Babu Nagati[1], Rama Koyyati[1], Ramchander Merugu[2], Pratap Rudra Manthur Padigya[1]*

[1]Department of Biochemistry, Osmania University, Hyderabad, India-500007.
[2]Department of Biochemistry, Mahatma Gandhi University, Nalgonda, India

*Corres.author: mpprataprudra@gmail.com,
Tel: 91-40-27097044.

Abstract: Silver nanoparticles were synthesized using *Tectona grandis* leaf extract and were identified by the color change of the silver nitrate solution to dark red. FTIR, SEM and TEM results revealed the functional groups and morphology and size of the silver nanoparticles. Silver nanoparticles synthesized have shown strong peaks within the range of 450-490 nm. The EDS and XRD revealed the binding of leaf extract to silver ions. The size of the formed nanoparticles was found to be within the range 30-40 nm. In the present communication, biogenic synthesis of silver nanoparticles using *Tectona grandis* leaf extract and evaluation of their antibacterial activity are communicated.
Keywods: *Tectona grandis* , Silver nanoparticles, 30-40 nm, Antibacterial activity.

1. Introduction:

Nanotechnology is being applied in various fields like Biology and Medicine[1-3] etc., Further earlier reports indicate that silver nanoparticles are not harmful to humans but act as effective agents against different bacteria[4-6], fungi[7] and yeast. Chemical processes are tedious and are not ecofriendly. Biological methods on the other hand neither requires toxic solvents nor synthesize hazardous by-products, they are fast and cost effective. *Tectona grandis* (Verbenaceae) is a large deciduous tree. The tree is, native to central India, Konkan, South India and Burma[8]. It is commonly known as teak tree[9,10]. Teak is a hardwood species of worldwide reputation[11]. Leaves are 30-40 by 15-30 cm, elliptic or ovate, acute or acuminate. Roots are used in the treatment of anurea and urine retention[12]. The flowers are useful in the treatment of bronchitis and urinary discharges. Bark and wood are useful in the treatment of bronchitis, piles, dysentery, leucoderma and headache. The flowers and the seeds are diuretics. Reports of various researches show that silver nanoparticles synthesized by green synthesis technique[13-17] and exhibited antimicrobial activities against various pathogenic microbes. The aim of this study

includes the synthesis of silver nanoparticles by using *Tectona grandis* leaf extract and investigating the efficacy of the same as an antibacterial agent on the pathogenic bacteria.

2. Materials and Methods:

*2.*1 Collection of plant material:

Tectona grandis leaves were collected from Botanical garden, Osmania University Campus, Hyderabad, Andhra Pradesh, India (Figure 1A).

Fig.1: Tectona grandis Tree

2.2 Preparation of *Tectona grandis* leaf extract:

The 30g of *Tectona grandis* leaves were weighed and washed twice with distilled water and then cut into fine pieces. For these pieces of leaves taken in a 500ml Erlenmeyer flask, 200ml of milli pore water was added and kept on a water bath for 30 minutes at 40^0C. Then the plant material is filtered off through Whatmann No: 1 filter paper and the leaf extract were used for the synthesis of silver nanoparticles. The extract will be stored at 4^0C for one week

2.3 Biosynthesis of Silver nanoparticles Using *Tectona grandis* leaf extract:

1 mM aqueous solution of sliver nitrate ($AgNO_3$) was prepared and for the synthesis of silver nanoparticles. To 1ml of the extract, 9ml of 1mM silver nitrate solution was added and kept at on magnetic stirrer for 2 hours. A color change of yellowish to dark reddish color silver nanoparticles was observed (Figure.2).

The silver nanoparticles solution was then centrifuged for 30minutes at the rate of 20,000rpm. The pellet obtained was washed with MQ water for three times. Then the pellet is dispersed into MQ water, which can be used for the further characterization of the silver nanoparticles.

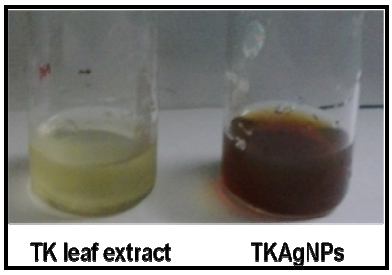

Fig.2: Tectona grandis leaf extract and Silver nanoparticles (dark reddish color).

2.4 Characterization of Synthesized Silver Nanoparticles:

UV-Vis-spectrophotometer is procured from ECLIO SL 159 UV-Vis spectrophotometer for the identification of synthesized silver nanoparticles. Transmission electron microscope (TEM) (Philips CM-10) and Scanning electron microscope (SEM) (Hitachi S - 4500) was used to investigate morphology and size of the particles. FTIR analysis, Energy Dispersive X-ray spectroscopy (EDS) (EDAX XL-30) operating at 15-25 kV) and X-ray Diffraction analysis (XRD)(Phillips, Holland model)was used for the identification on silver nanoparticles binding to leaf extract.

2.5 Antimicrobial Activity by Disc Diffusion Method:

The disc diffusion method was employed for the evaluation of antibacterial activities of the synthesized silver nanoparticles. 0.1 ml from 100cfu/ml of Gram positive (*Staphylococcus aureus*) and Gram negative (*Escherichia coli*) bacteria suspension was spreaded on different plates with LB (Luria-Bertani) media. Filter paper discs (3 mm in diameter) were placed on the plates and then onto the discs samples were impregnated with different concentrations. Ampicillin (10µg/ml concentration) served as the standard for measuring the antibacterial activity. The plates were then incubated at 37^0C for 24h. The Zones of inhibition was measured in mm.

3. Results and Discussion:

Synthesized silver nanoparticles from 1mM silver nitrate solution using leaf extarct were identified by the color change of the solution to dark red. Color of silver colloid is due to Surface Plasmon resonance (SPR). Absorption spectra of silver nanoparticles formed had shown absorbance peaks within the range 450-490 nm (Figure 3)

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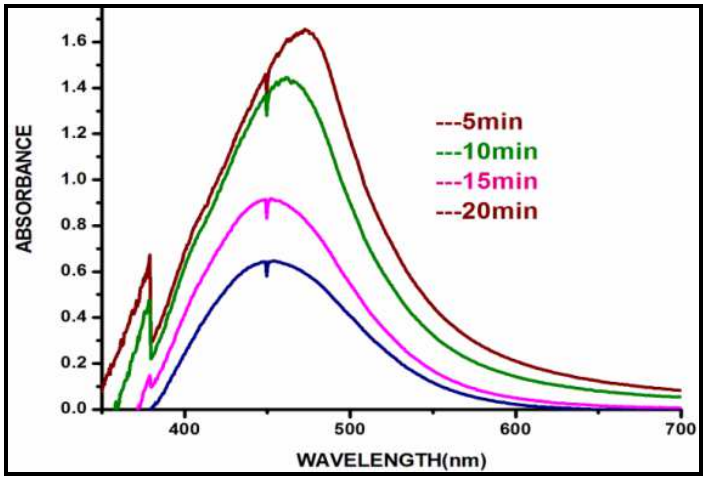

Fig.3: UV-Vis Spectroscopic analysis of Silver Nanoparticles

The FTIR spectra of silver nanoparticles showed broad peaks at 3851 cm-1, 1384cm-1, 858 cm-1 and 433 cm-1(Fig 4).The result indicates that the Carboxyl (-C= hydroxyl (-OH) and Amine (-NH) groups of leaf extracts are mainly involved in synthesis of silver nanoparticles. Other minor peaks indicate that the formed silver nanoparticles were surrounded by proteins, terpenoids and secondary metabolites.

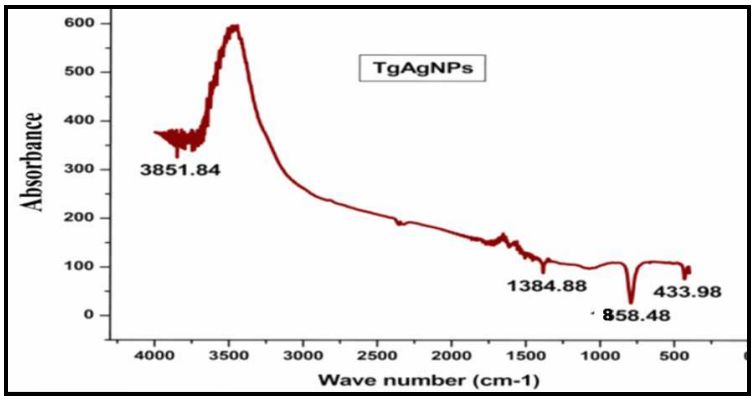

Fig.4: FTIR analysis of synthesized silver nanoparticles

X-ray diffraction (XRD) confirmed the existence of silver colloids. Braggs reflections were observed at 2θ = 38,, 44.16, 64.17 and 77.12 (Fig.5). A strong diffraction peak located was ascribed to the 32 facets of silver. The results thus illustrate that the synthesized *Tectona grandis* silver nanoparticles were crystalline in nature.

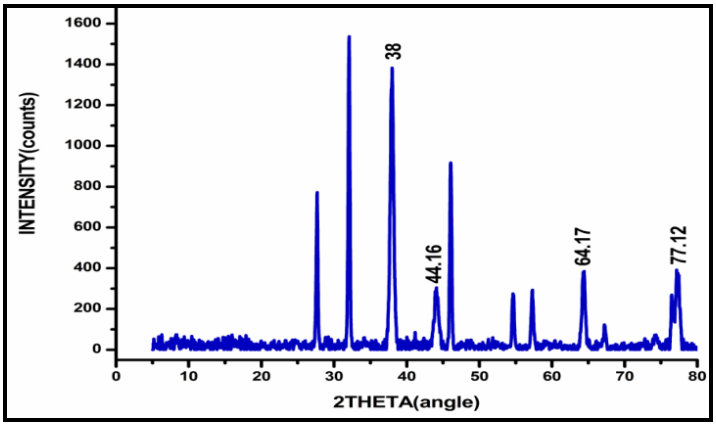

Fig.5: XRD analysis of Tectona grandis silver nanoparticles

Scanning Electron Microscopic studies were carried out to study the morphology of the silver nanoparticles. The SEM pictures indicated that the formed particles were from spherical to oval in shape with a smooth morphology. EDS analysis confirmed the presence of elemental silver as the major constituent. The SEM figures are shown in Figure 6. The EDS results are shown in (Fig 6 and Table 1).

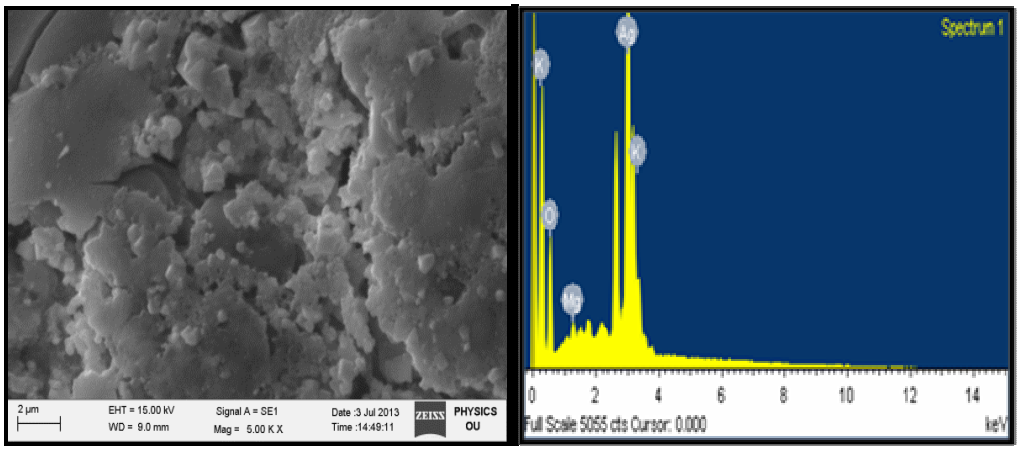

Fig.6: SEM analysis and EDS spectrum of synthesized silver nanoparticles

Table.1: The composition of silver nanoparticles synthesized from Tectona grandis leaf extract

Element	Weight%	Atomic%
O K	24.84	67.76
Mg K	0.77	1.39
K K	1.08	1.20
Ag L	73.30	29.65
Totals	100.00	

From the figure 7, the size of the formed nanoparticles was found to be within the range 30-40 nm..

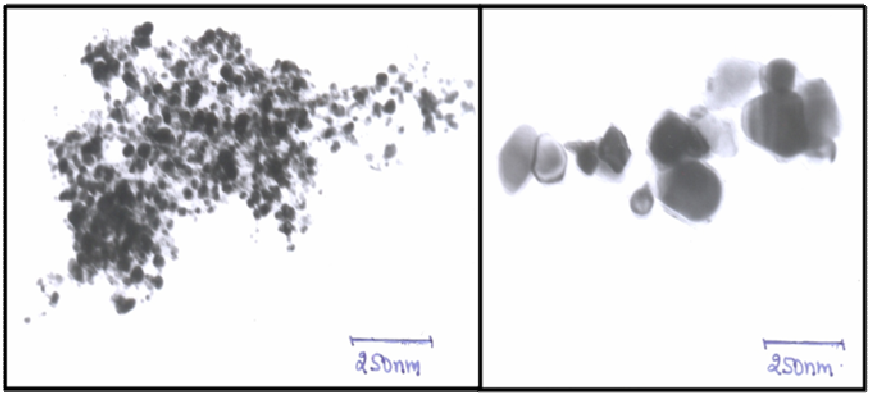

Fig.7: TEM images of biosynthesized silver nanoparticles using Tectona grandis leaf extracts at 250 nm scales

The formed silver nanoparticles have shown antibacterial activity against Gram positive (*Staphylococcus aureus*) and Gram negative (*Escherichia coli*) bacteria (Fig. 8). Ampicillin was taken as the positive control for the measurement of Zone of Inhibition (in mm) as shown in Table 2. The results obtained are shown.

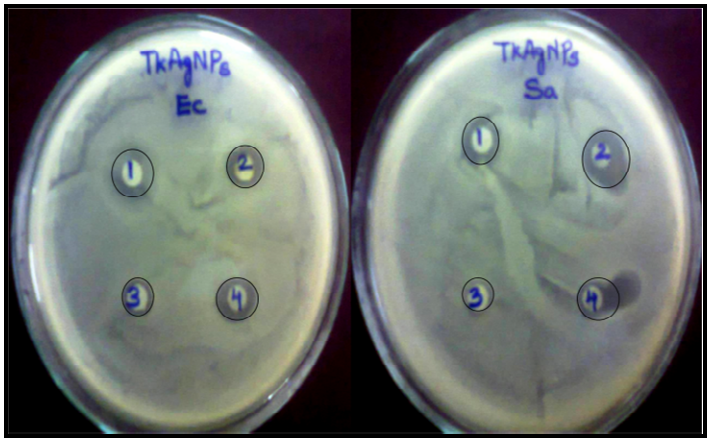

Fig.8: Antibacterial activity of synthesized silver nanoparticles, 1.Ampicillin (5µl); 2.AgNO3(10µl); 3. Tg Leaf extract (10µl); 4. TgAgNPs(10µl).

Table 2 Measurement of Zone of inhibition by Disc diffusion method

Name of the Organism	Zone of inhibition in mm			
	Ampicillin 5µl	1Mm $AgNO_3$ 5µl	Tg Leaf Extract 10µl	TgAgNPs 10µl
Escherichia coli	10	8	4	12
Staphylococcus aureus	8	12	5	8

Conclusion:

In this present study silver nanoparticles were synthesized using *Tectona grandis* leaf extract. These particles have shown considerable anti-bacterial activity against human pathogens and could have potential applications in industry.

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