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**Antibacterial activity of Silver Nanoparticles synthesized by using
Strychnos potatorum leaves**

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ABSTRACT

Biologically synthesized silver nanoparticles are being widely using in the field of medicine. Extracellular biosynthesis of silver nanoparticles was carried out by using Strychnos potatorum leaf extract for the reduction of silver ions in short period. The silver nanoparticles formation was confirmed by the colour change of plant extracts. These silver nanoparticles were tested for antibacterial activity by using disc diffusion method. The test cultures are Escherichia coli, proteus vulgaris, Staphylococcus aureus, Staphylococcus epidermidis, Pseudomonas aeruginosa, and Klebsiella sp., were used. The antibacterial property of silver nanoparticles was analysed by measuring the inhibition zone. The silver nanoparticles synthesized from leaf extracts of Strychnos potatorum. Whereas the growth of Pseudomonas sp., and Proteus sp., were inhibited maximum by the silver nanoparticles synthesized from leaf extract of Strychnos potatorum. The results indicates that the silver nanoparticles may have an important advantage over conventional antibiotics.

Key words: *Strychnos potatorum, Silver nanoparticles synthesis, Pathogens, Antibacterial activity.*

INTRODUCTION

Strychnos potatorum (Family - Loganiaceae) is one such medicinally important forest tree species which is popularly known as clearing nut tree or sirungamaram or theththamkottai¹. The plant is a medium sized deciduous tree having height upto 12 meters, leaves are simple, opposite, elliptic, acute, 15 × 6.25 cm, glabrous, shining. *Strychnos potatorum* is a moderate sized tree found in central and Southern India's also in Sri Lanka and Myanmar; it has been used extensively as a folklore medicine and in ayurvedic practice, notably the fruit as an antidiabetic, antidystentric, emtic, while the pulp is useful as an expectorant². According to World Health Organization (WHO), it was estimated that 80% of the population in developing countries rely mostly on traditional medicine like plant drugs, for their primary health care needs. Medicinal plants being natural, cost effective, having no side effects, preventive and curative therapies which could be useful in achieving the goal of Health for all in a cost of effective manner³.

Nanotechnology is now creating a growing sense of excitement in the life sciences especially biomedical devices and Biotechnology⁴. Nanoparticles exhibit completely new or improved properties based on specific characteristics such as size, distribution and morphology. The silver nanoparticles have various and important applications. Historically, silver has been known to have a

disinfecting effect and has been found in applications ranging from traditional medicines to culinary items. It has been reported that silver nanoparticles are non-toxic to humans and most effective against bacteria, virus and other eukaryotic micro-organism at low concentrations and without any side effects⁵. Moreover several salts of silver and their derivatives are commercially manufactured as antimicrobial agents. In small concentrations, silver is safe for human cells, but lethal micro-organisms. Antimicrobial capability of silver nanoparticles allows them to be suitably employed in numerous household products such as textiles, food storage containers, home appliances and in medical devices. Silver nanoparticles is in medical industry such as tropical ointments to prevent infection against burn and wounds. Biological synthesis of nanoparticles by plant extracts is at present under exploitation as some researchers worked on it and testing for antimicrobial activities⁶.

MATERIALS AND METHODS

Collection of Plant Material

Strychnos potatorum leaves were collected from Velur near Viralimalai, Pudukottai district, Tamil Nadu India. The leaves were separated from the plant and dried under shade. After drying, it was powdered and used for our studies.

Silver nanoparticles synthesis

1 mm silver nitrate was added to the plant extracts separately and make up a final solution of 100 ml, 50 ml of solution was fresh and another 50 ml of solution was boiled and it was allowed to stand for 12-24 hours after that the solution were observed for colour changes and it was centrifuged at 12000 rpm for 15 minutes. The colour changes indicate the formation of silver nanoparticles. Supernatant and pellets were separated gently.

Phase Contrast Microscope

After synthesizing silver nanoparticles observed the Phase Contrast Microscope, different views of photos were identified.

Microorganisms

Pure culture of *Escherichia coli*, *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Klebsiella pneumonia*, *Staphylococcus aureus*, and *Staphylococcus epidermidis* species of bacteria were obtained from PG and Research Department of Microbiology, of Jamal Mohamed College, Trichy. The antibacterial activity carried out in the Department of Microbiology.

Antibacterial activity

The antibacterial activities of silver nanoparticles were carried out by disc diffusion method. Muller Hinton Agar medium plates were prepared, sterilized and solidified. After solidification bacterial cultures were swabbed on these plates. The sterile discs were dipped in silver nanoparticles solution and placed in the Muller Hinton Agar plate and kept for incubation at 37° C for 24 hours. After incubation the zones of inhibition were measured.

RESULTS AND DISCUSSION

The synthesis of silver nanoparticles through *Strychnos potatorum* leaf extracts were carried out. It is well known that silver nanoparticles exhibit brown colour in aqueous solution due to the excitation of surface plasmon vibrations in silver nanoparticles. The appearances of brown colour in the reaction vessels suggest the formation of silver nanoparticles (Fig.1). After silver nanoparticles synthesizing we observed the different views of photos in Phase Contrast Microscope (Fig.2).

Fig.1: The colour change of plant extracts after addition of silver nitrate



(a)



(b)



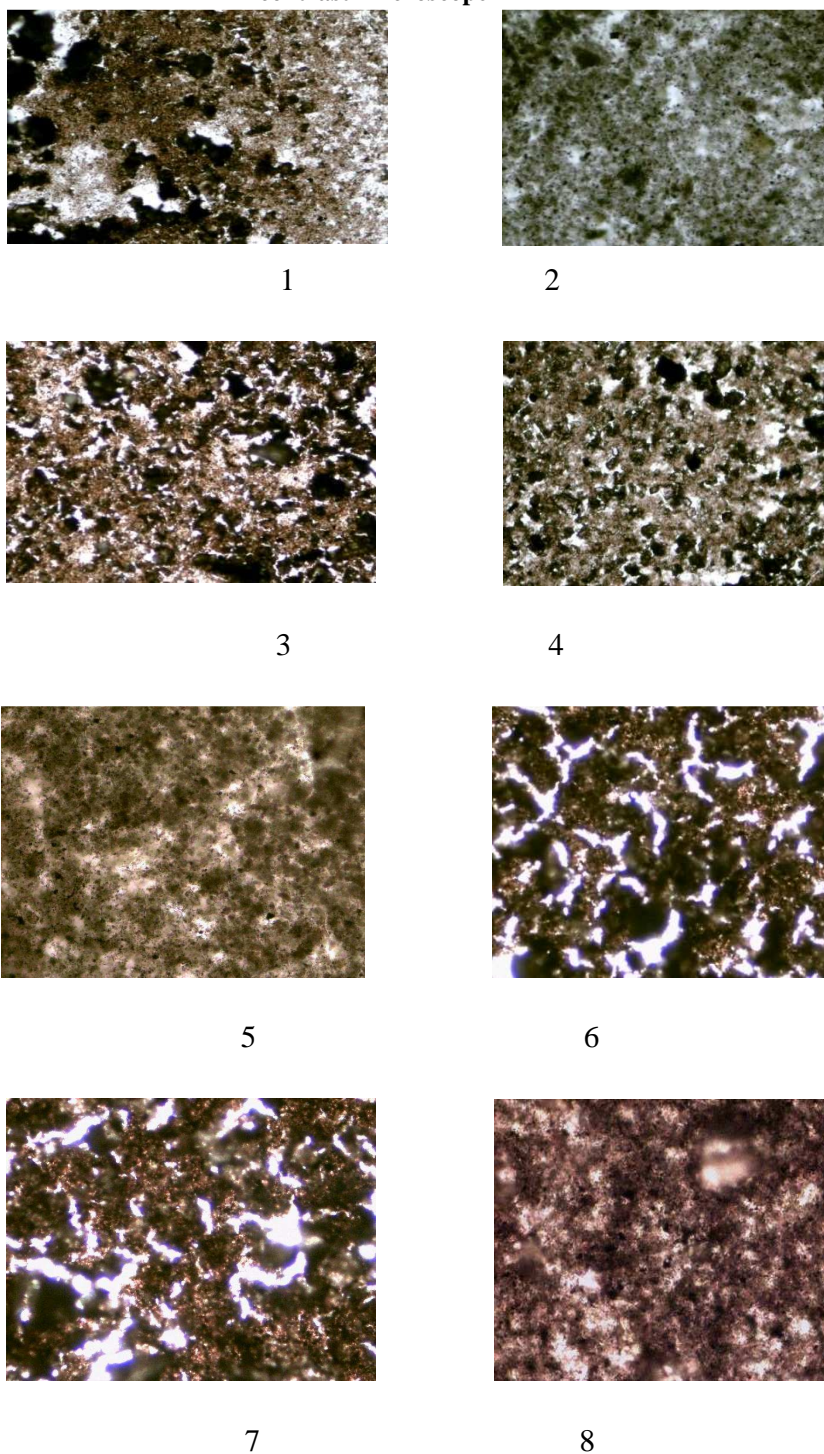
(c)



(d)

(a) plant extract of *S. potatorum* leaf (b) silver nanoparticles synthesized by fresh leaves (c) boiled leaf extract (d) silver nanoparticles synthesized by boiled leaf extract

Fig. 2: After silver nanoparticles synthesizing different views of photos observed in Phase contrast microscope



(1-4 fresh leaves added with AgNO_3), (5-8 boiled leaves added with AgNO_3).

The result of the antibacterial activity of *Strychnos potatorum* fresh leaf extract with silver nanoparticles is given in Table 1. Best zone of inhibition was produced against *Pseudomonas aeruginosa* (30 mm), and better zones of inhibition against *Proteus vulgaris* (24 mm), *Escherichia coli* (20 mm), *Staphylococcus aureus* (20 mm), *Staphylococcus epidermidis* (18 mm), and least was produced against *Klebsiella* sp., (16 mm). Maximum zone of inhibition were observed in *Pseudomonas* sp., and *proteus* sp., and least zone of inhibition observed in *Klebsiella* species (Fig.1).

Table 1. Antibacterial activity of *Strychnos potatorum* fresh leaf extract with AgNO₃ (zone of inhibition in mm)

S. No	Sample	Bacterial strains used	Fresh leaf extract with silver nitrate
1	<i>Strychnos potatorum</i> leaf	<i>E. coli</i>	20
2		<i>Klebsiella sp.</i> ,	16
3		<i>P. vulgaris</i>	24
4		<i>P. aeruginosa</i>	30
5		<i>S. aureus</i>	20
6		<i>S. epidermidis</i>	18

The result of the antibacterial activity of boiled leaf extract with silver nanoparticles is given in Table 2. best zone of inhibition was produced against *Pseudomonas aeruginosa* (20 mm), and better zones of inhibition against *Proteus vulgaris* (16 mm), *Klesiella sp.*, and *Staphylococcus aureus* (14 mm), least zone of inhibition produced against *Escherichia coli* (13mm) and *Staphylococcus epidermidis* (12 mm). Maximum zone of inhibition were observed in *Pseudomonas aeruginosa* and *proteus vulgaris*, least zone of inhibition produced against *Klebsiella sp.*, and *Escherichia coli* fig 2.

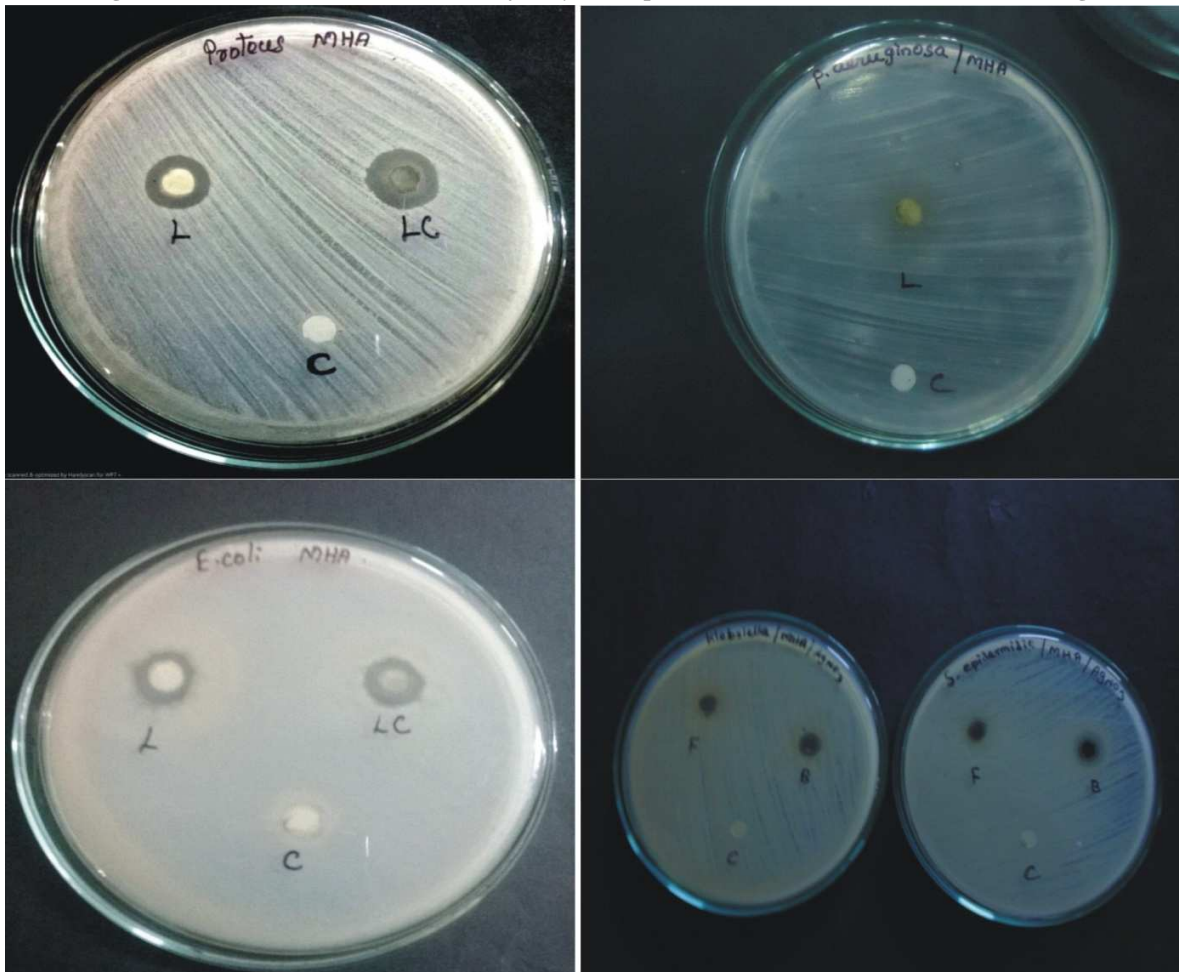
Table 2. Antibacterial activity of *Strychnos potatorum* boiled leaf extract with AgNO₃ (zone of inhibition in mm)

S. No	Sample	Bacterial strains used	Boiled leaf extract with silver nitrate
1	<i>Strychnos potatorum</i> boiled leaf	<i>E. coli</i>	13
2		<i>Klebsiella sp.</i> ,	14
3		<i>P. vulgaris</i>	16
4		<i>P. aeruginosa</i>	20
5		<i>S. aureus</i>	14
6		<i>S. epidermidis</i>	12

Figure 1. Zone Inhibition formed by *Strychnos potatorum* fresh leaf extract with AgNo₃



Figure 2. Zone Inhibition formed by *Strychnos potatorum* boiled leaf extract with AgNo₃



In earlier studies¹⁰, the synthesis of silver nanoparticles through plant extracts were carried out. It is well known that silver nanoparticles exhibit brown colour in aqueous solution due to the excitation of surface plasmon vibrations in silver nanoparticles. The appearances of brown colour in the reaction vessels suggest the formation of silver nanoparticles.

In previous studies¹¹, shows the biological and green synthesis of silver nanoparticles. In this work, the synthesis of stable silver nanoparticles by the bio-reduction method was investigated. Aqueous extracts of the manna of hedysarum plant and the soap-root (*Acanthe phylum bracteatum*) plant were used as reducing and stabilizing agents, respectively. UV-Vis absorption spectroscopy was used to monitor the quantitative formation of silver nanoparticles. Biologically synthesized silver nanoparticles could be of immense use in medical textiles for their efficient antibacterial and antimicrobial properties (Shahverdi et al., 2007).

The present study shows the synthesis of silver nanoparticles through *Strychnos potatorum* leaf extract were carried out. It is well known that silver nanoparticles exhibit brown colour in aqueous solution. The appearances of brown colour in the reaction vessels suggest the formation of silver nanoparticles. Fresh and boiled leaf extract with silver nitrate solution were tested the antibacterial activity.

Silver nitrate is used as a reducing agent as silver has distinctive properties such as good conductivity, catalytic and chemical stability. The aqueous silver ions when exposed to herbal extracts were reduced in solution, there by leading to the formation of hydrosol. The time duration of change in colour varies from plant to plant. *Strychnos potatorum* is door for nanotechnology applications in medicine. Biological synthesis of metal nanoparticles is a traditional method and the use of plant extracts has a new awareness for the control of disease, besides being safe and no phytotoxic effects. The biologically synthesized silver nanoparticles using medicinal plants were found to be highly toxic against different pathogenic bacteria of selected species.

The silver nanoparticles of *Strychnos potatorum* shows highest antibacterial activity was observed against *Pseudomonas aeruginosa* followed by *Proteus vulgaris* and *E.coli* species. The silver nanoparticles synthesized from leaf extract showed higher toxicity, leaf extract synthesized higher concentration of silver nanoparticles. Moreover green leaves are the site of photosynthesis and availability of more H⁺ ions to reduce the silver nitrate to silver nanoparticles. The molecular basis of biosynthesis of these silver crystals is speculated that organic matrix contain silver binding protein that provide amino acid moieties that serve as the nucleation sites. The efficiency of various silver based antimicrobial fillers in polyamide toward their silver ion release characteristics in an aqueous medium was also investigated and discussed in their number of plants including fungi, yeast and algae¹³.

CONCLUSION

The present study included the bio-reduction of silver ions through *Strychnos potatorum* plant leaf extracts and testing for their antibacterial activity. The aqueous silver ions exposed to the extracts, the synthesis of silver nanoparticles were confirmed by the change of colour of plant extracts. The results indicated that silver nanoparticles have good antibacterial activity against different microorganisms. It is confirmed that silver nanoparticles are capable of rendering high antibacterial efficacy and hence has a great potential in the preparation of drugs used against bacterial diseases.

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