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Phytogenic Silver Nanoparticles Using Heteropogon Contortus Leaf Extracts And Its Anti-Bacterial Activity

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ABSTRACT

Nanoparticles are being viewed as fundamental building blocks of nanotechnology. Biosynthesis of nanoparticles by plant extracts is currently under exploitation. In the current study, Heteropogon contortus was used for extra cellular synthesis of metallic silver nanoparticles. Stable silver nanoparticles were formed by treating aqueous solution of $AgNO_3$ with the plant leaf extract as reducing agent at room temperature. UV-Visible spectroscopy was utilized to monitor the formation of silver nanoparticles. Synthesized Ag nanoparticles were effective anti-bacterial agent's against Bacillus subtilis, Staphylococcus aureus, Proteus vulgare and Klebsiella pneumoniae. Our proposed work offers an enviro-friendly method for phytogenic silver nanoparticles production.

Keywords: *Biosynthesis, Heteropogon contortus, Silver nanoparticles, anti-bacterial activity.*

INTRODUCTION

Nanotechnology deals with synthesis of nanoparticles and nanomaterials (generally range from 1-100nm) of variable size, shapes and their application in various fields. Nanoparticles have been studied widely because of their unique physicochemical properties like catalytic activity (1), optical properties (2), electronic properties, antibacterial[1] properties (3) and magnetic properties. The unique property could be attributed to their small sizes and large surface area. Metal nanoparticle such as gold, silver, zinc, and platinum, are extensively used in products that directly come in contact with the human body, such as shampoos, soaps, detergent, shoes, cosmetic products, and toothpaste, also medical and pharmaceutical applications. Metal nanoparticles with unique properties; have been synthesized by chemical (4, 5) and biological methods. In chemical methods ecologically toxic chemicals have been used. This negative aspect can be overcome by biological method. In case of biological method, bio components such as microorganism, plant extract or enzymes are used for the nanoparticle synthesis. Biological synthesis of silver nanoparticle using plant extract of Cissus quadrangularis (6), coriander leaves (7), sundried Cinnamomum camphora leaves (8), phyllanthin extract (9), and purified apiin compound extracted from henna leaves (10) have been reported. Using microorganism like fungi (11, 12), actinomycets and bacteria have also been reported (13).

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In this study, Heteropogon contortus leaf extract was used as a reducing agent to synthesis silver nanoparticle. Heteropogon contortus is a tropical, perennial tussock grass with a native distribution encompassing Southern Africa, southern Asia, NorthernAustralia, Oceania, and southwestern North America. Heteropogon contortus is declared a weed in some regions of America, East Asia and in New-Caledonia (14,15). The reduced silver nanoparticle was characterized by UV-visible spectrometer and their potential antibacterial activity was assessed.

MATERIALS AND METHOD

Plant material and preparation of the aqueous extract

Heteropogon contortus leaves were collected from Srikakulam, Andhra Pradesh, India and used for the preparation of the aqueous extract. 10g of green tender leaves were thoroughly washed, cut into fine pieces, were soaked in 100 ml deionised water, boiled for 10 mins and filtered through Whattman No.1 filter paper (42µm) to get an extract. The aqueous leaf extract was used as a reducing agent for further nanoparticle synthesis.

Synthesis of Silver nanoparticles

After boiling, the solution was decanted, and 5 mL of this broth was added to 100 mL of 10^{-3} M aqueous AgNO3 solution. The bioreduction of the Ag+ ions in solutions was monitored by periodic sampling of aliquots (1 mL) of the aqueous component after 5 times dilution and measuring the UV-vis spectra of the solution. with 0.1 mM $AgNO_3$ solution.

Characterization of synthesized silver nanoparticle

The synthesized nanoparticle was characterized by UV-Vis spectra analysis. The reduction of pure Ag^+ ions was monitored by measuring the UV-Vis spectrum of the reaction medium and the absorption spectra were recorded over the range of 200-800 nm using UV-Vis spectrophotometer (Shimadzu UV-2450).

Antibacterial activity of synthesized nanoparticle

All microorganisms used for this study were purchased from the National Chemical Laboratory (NCL), Pune, India and were maintained at 4^0C on nutrient agar. The antibacterial activity of the nanoparticle was studied by well diffusion method against the following bacteria via, Bacillus subtilis, Staphylococcus aureus, Klebsiella pneumoniae and Proteus vulgare.

RESULTS AND DISCUSSION

Biosynthesis of silver nanoparticle

As the Heteropogon contortus leaf extract was added to the aqueous solution of the silver ion complex, the solution color, started to change from watery to yellowish brown initially and later intensified to brown colour due to reduction of silver ion which indicated formation of silver nanoparticles (16, 17). Reduction of silver ions could be easily followed by color change. Due to excitation of surface Plasmon vibrations in nanoparticle, it exhibits different color than the molecular scale particle.

UV-Vis Spectra analysis

The formation of silver nanoparticle by reduction of the aqueous silver ions during exposure of Heteropogon contortus leaf extract may be easily followed by UV-Vis spectroscopy. Silver nanoparticle exhibit brown color in aqueous solution due to the surface Plasmon resonance phenomenon (18, 19). In this study, the surface Plasmon resonance band of the silver nanoparticle was observed at 442 nm (fig1).

Antibacterial activity

Antibacterial activity of silver nanoparticle was evaluated by well diffusion method against the following microorganism: Bacillus subtilis, Staphylococcus aureus, Klebsiella pneumoniae and Proteus vulgare and the results were tabulated below. The silver nanoparticle has shown antibacterial against all tested microorganism and maximum activity was found against Staphylococcus aureus. The second maximum activity was observed against Klebsiella pneumoniae and least activity was found against Bacillus subtilis and Proteus vulgare. The silver nanoparticle's activity was compared with the plant extract and silver nitrate solution. Nanoparticle has shown maximum activity than silver nitrate solution and there was no anti-bacterial activity with plant extract. From table it was clear that silver nanoparticle has shown more activity than the silver nitrate solution. The mechanism involved in antibacterial activity of silver nanoparticle is not well known. It may be due to the attachment of silver nanoparticle to the surface of the cell membrane and disquieting the power function of bacteria such as permeability and respiration (5). Since nanoparticle has large surface area and small size than, the nanoparticle binds and interacts to the cell more than the large particle. It may be a reason why the silver nanoparticle has shown more activity than the silver ions (20).

CONCLUSION

There is an increasing commercial demand for nanoparticles due to their wide applicability in various areas. Metallic nanoparticles are traditionally synthesized by wet chemical techniques, where the chemicals used for quite often are toxic and flammable. In this work an environmental friendly technique for the synthesis of nanoparticles from 1 mM $AgNO_3$ solution using leaf extract of Heterpogon contortus was carried out. Synthesised nanoparticles were characterized using UV-Visible spectroscopy. Later, the synthesized nanoparticles were subjected to toxicological studies. Silver nanoparticle had showed antibacterial activity against the tested microorganisms. Thus, this particle can be further analysed for the bacterial growth inhibition and can be used to treat certain microbial diseases.

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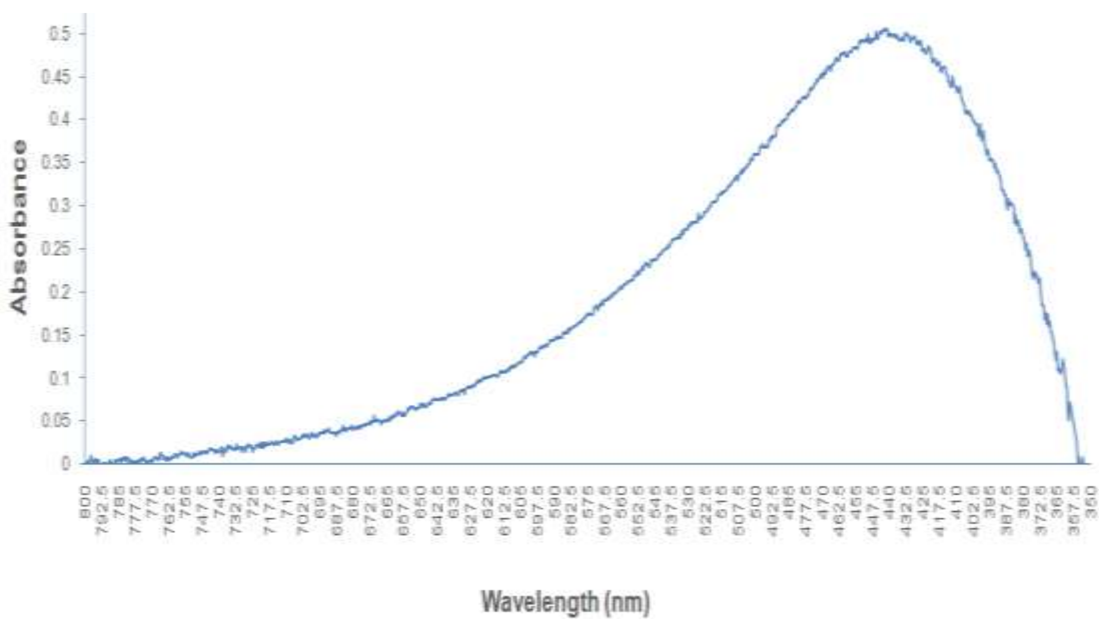

Figure 1: UV-Vis spectra analysis silver nanoparticle synthesized using Heteropogon contortus leaf extract

Table 1: Antibacterial Activity of Silver Nanoparticles

Microorganism	**Zone of Inhibition (mm)**		
	20µl of 1mM $AgNO_3$	20µl of plant extract	20µl of AgNPs
Bacillus subtilis	Nil	Nil	12.23
Staphylococcus aureus	Nil	Nil	13.35
Proteus vulgaris	6	Nil	12.27
Klebsiella pneumoniae	7	Nil	12.34