



## Evaluate the antibacterial activity by synthesis of silver nanoparticles from *pogostemon Benghalensis* (Burm. F.) Kuntze aerial part

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### Abstract

In recent years green synthesis of silver Nanoparticles has been increased for diverse application. In this study we proposed *Lamiaceae* family plant. *Pogostemon benghalensis* for synthesis of silver nanoparticles and their efficacy in antibacterial activity against five human pathogens. The direct method was chosen for silver nanoparticles synthesis using water extract of *Pogostemon Benghalensis*. Green synthesized silver nanoparticles were characterized by UV-Vis spectroscopy, FTIR, SEM, EDS. The UV-Vis spectro depicts surface Plasmon resonance peak present at 450nm. The three dimensional structure of silver nanoparticles was seen through SEM analysis. The observed FT-IR peaks showed functional groups and the stretch of bonds that responds to AgNPs synthesis. The synthesized nanoparticles showed remarkable antibacterial activity against three gram positive bacteria strains *Staphylococcus aureus*, *Enterococcus faecalis*, *Enterococcus phyogenes* and two gram negative bacterial strains *Salmonella typhi*, *Enterobacter aerogenes*. The highest antimicrobial activity against *Enterococcus pyogenes* (16mm) and the lowest antibacterial against *Enterococcus faecalis* (7mm). The present suggest that AgNPs can be used for potential of antibiotic drugs.

**Keywords:** *pogostemon Benghalensis*, AgNPs, FTIR, SEM

### Introduction

Nanobiotechnology is an emerging field of contemporary modern research that involves the manipulation of materials at the atomic or molecular level for their diverse properties [1-3]. Nanomaterials find their applications in several fields of biomedical sciences, such as gene delivery, biosensing, image analysis, and drug delivery. Nanostructures, such as silver, gold, CuO, and Zn possess the therapeutic potential and hence, used for various biomedical applications including as antimicrobial agents [4-10]. Therefore, studies in the field of nanotechnology and material science are rapidly growing to develop and screen wide ranging materials by using a number of methods [11, 12]. Alternatively, biogenic synthesis of AgNPs that uses plants and microbes has gained wide acceptance because these approaches are simple, rapid, efficient, and cost-effective. [13, 14, 15]. Phyto fabrication of AgNPs depends on the available functional groups in plants or plant extracts that might act as reducing and capping agent [16]. *Pogostemon cablin* Benth. (Patchouli) is a commercial. Medicinal plant belongs to the family *Lamiaceae*. Its raw materials are consumed in the pharmaceutical, fragrance, cosmetic, flavor and perfumery industries [17]. The multidisciplinary field of nanotechnology mainly concern to synthesis and design nano materials and device with in the range of 1-100nm. But the range of size of nanoparticles in some literature greater than 100nm [18]. A large of traditional medicinal plants used in the treatment of cut and injuries, blood pressure, skin diseases, dysentery, blood deficiency, jaundice, urinary trouble, cough, asthma, influenza, malaria etc [19]. These plants are as source of bio-reductant and stabilizers and reported to contain alkaloids, glycosides, tannins, saponins and aromatic compounds [20].

<sup>21</sup>. The most common major materials used for manufacturing nano -products at present is silver, then followed by carbon, Titanium, silicon, Zinc and Gold, Silver nano particles are used as anti-bacterial agents, as catalytic and also as optical sensor. By using plant extract methods, a lot of groups are involved to synthesis silver nanoparticles from plants like *Aloe vera* [22]. To synthesis silver nano particles by plant extract biosynthesis method from leaves of *Pogostemon benghalensis*. (Family-*Lamiaceae*, Assamese vernacular name-*shukloti*) and study their characterization, this traditional medicinal plant is selected for the applications used in cut and injuries by local peoples. Mainly leaves of this plants use by local women's after delivering a child. It is a shrub with a height of 4 to 5ft, leaves are elliptic. The whole plants use as medicinal purpose. Leaves, Young shoots and juice of leaves are used to cure stomach problems, increase digestive power, stop bleeding and mainly for menstrual disorders.

### Materials and Methods

#### Silver Nanoparticle Synthesis

#### Materials and Methods

#### Plant material and preparation of Extracts

Aerial parts of *Pogostemon benghalensis* were dried for 25 days under the shadow at room temperature and ground into fine powder. 5g of powder was used to make aqueous extract using 100ml of distilled water. It was gently heated and filtered through a filter paper

#### Synthesis of Silver Nanoparticles

1mM aqueous solution of silver nitrate ( $\text{AgNO}_3$ ) was prepared and used for the synthesis of silver nanoparticles. 1000 ml of leaf extract was taken in a separate conical flask

and 100µl of 1mM AgNO<sub>3</sub> was added than heated until the colour changed from golden to brown.

### Characterization of Silernanoparticles by UV- Visible spectroscopy analysis

Bioreduction of aqueous Ag<sup>+</sup> ions can easily be followed by UV - Vis spectrophotometer and one of the most important features in optical absorbance spectra of metal nanoparticles is surface Plasmon band, which is due to collective electron oscillation around the surface mode of the excitation of their surface Plasmon response (SPR), when dissolved in water. The colour of the solution changes from colourless to brownish yellow with in 5 minutes at microwave and than to dark brown.

### Characterization of Silver nanoparticles by Fourier Transform Infrared Spectroscopy (FTIR)

Silver nanoparticles synthesized after 5 hour of reaction of 1Mm AgNO<sub>3</sub> solution with *p.benghalensis* leaves extract were centrifuged at 10,000 rpm for 45 mins at room temperature after which the pellet was re dispersed in sterile distilled water was repeated three times to ensure better separation of free entities from the nanoparticles. The purified pellet was then dried and subjected to Fourier transform infrared (FTIR, IR affinity-1, Shimadzu corporation, Tokyo, japan) spectroscopy measurement using the potassium bromide (KBr) pellet technique in diffuse reflection mode at a resolution of 4cm<sup>-1</sup> The nanoparticle powder was mixed with KBr and exposed to an infrared source of 400 cm<sup>-2</sup> to 4000 cm<sup>-2</sup>

### Scanning Electron Microscope (SEM with EDAX analysis)

In Scanning Electron Microscope analysis, the energy of electron beam current was continuously adjusted from 1 pA to 1µA to suit the type of examination in progress. SEM analysis was performed using Zeiss – SEM machine. Thin film of the sample was prepared on a carbon coated copper grid by just dropping very small amount of the sample on the grid. The film of the SEM grid was allowed to dry by putting it under a mercury lamp for 5 min. The each sample was analysed by the SEM. The presence of silver was confirmed by EDAX

### Antibacterial activity of *Pogostemon benghalensis* leaf aqueous extract from AgNPs synthesis

The antibacterial assays were done by disc diffusion method.

The antibacterial assays of *Pogostemon benghalensis* leaf extract fraction and silver nanoparticles synthesized from aerial part extracts were performed. Fresh overnight culture of inoculums 100µl of each bacterium were spread on to Nutrient agar plates. Sterile discs of 5mm diameter contain 25µl, 50µl, 75µl and 100µl sample (10 mg/1ml conc ) and AgNPs sample (1mg/1ml conc.) with for discs were plate and five antibiotic discs were placed in separate plate for positive control. The inoculated plates were incubated at 37°C for 24 h. The antibacterial activity was evaluated by measuring the diameter of the inhibition zone of the test microorganism

### Result

Silver nanoparticles (AgNPs) were synthesized using *Pogostemon benghalensis* plant extract. The AgNPs showed Yellowish-brown color in aqueous solution due to excitation of Surface Plasmon vibrations. Reduction of silver ions to silver nanoparticles could be followed by color change (Fig.1). AgNPs were investigated to evaluate their Antibacterials activity against Gram Positive bacteria (*Staphylococcus aureus*, *Enterococcus faecalis*, *Enterococcus pyogenes*), Gram Negative bacteria (*Salmonella typhi*, *Enterobacter aerogenes*) using disc diffusion method (Fig2). The Highest zone of inhibition was observed in *Enterococcus pyogenes* (16mm) and Lowest zone inhibition was observed in *Enterococcus faecalis* (7mm). All these results indicates that the AgNPs can be used as control the bacterial growth. So *Pogostemon benghalensis* AgNPs used as therapeutic purpose.

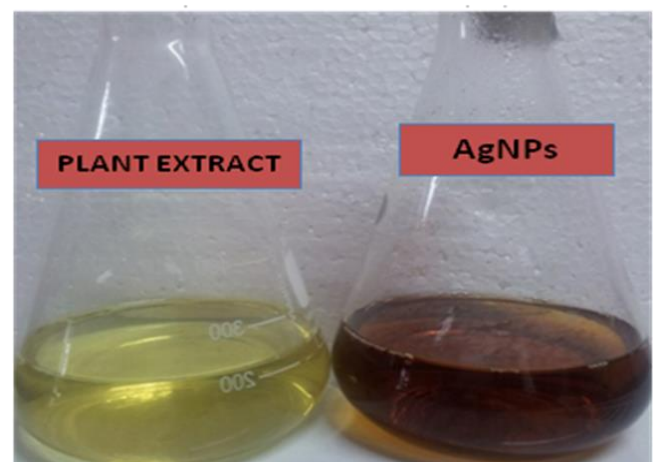
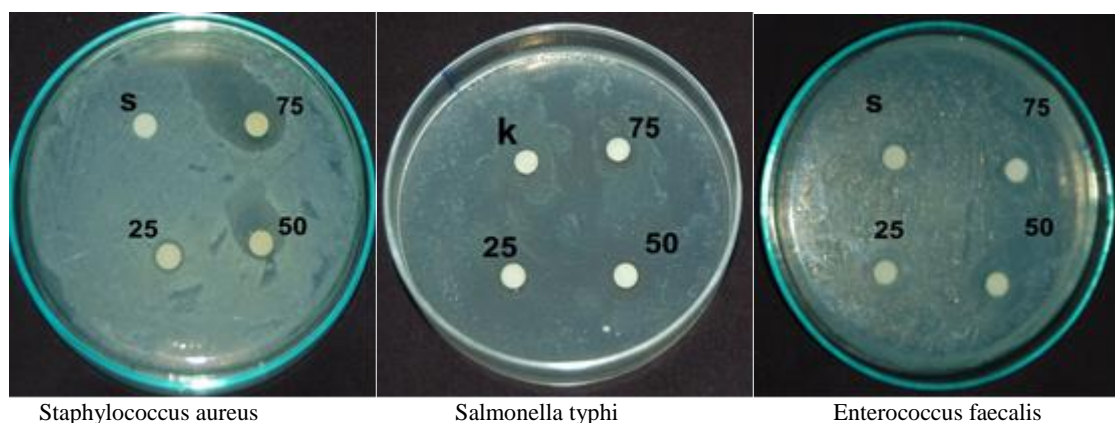


Fig 1: Reduction of Silver Nanoparticles indicating by colour change



Staphylococcus aureus

Salmonella typhi

Enterococcus faecalis

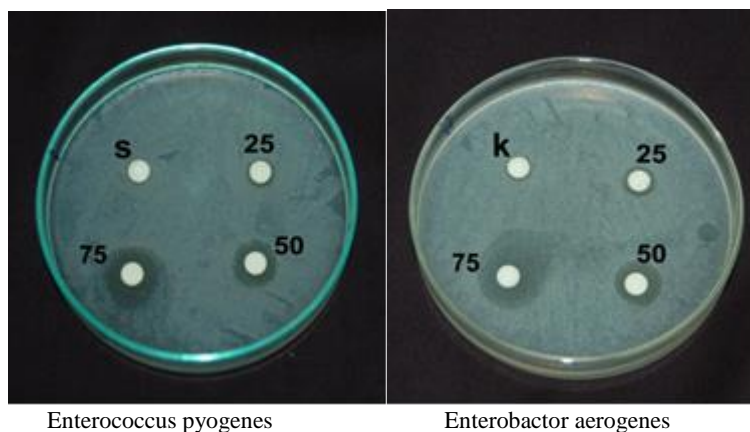


Fig 2: Antibacterial activity of AgNPs synthesis from *pogostemon benghalensis* Aerials part aqueous extract

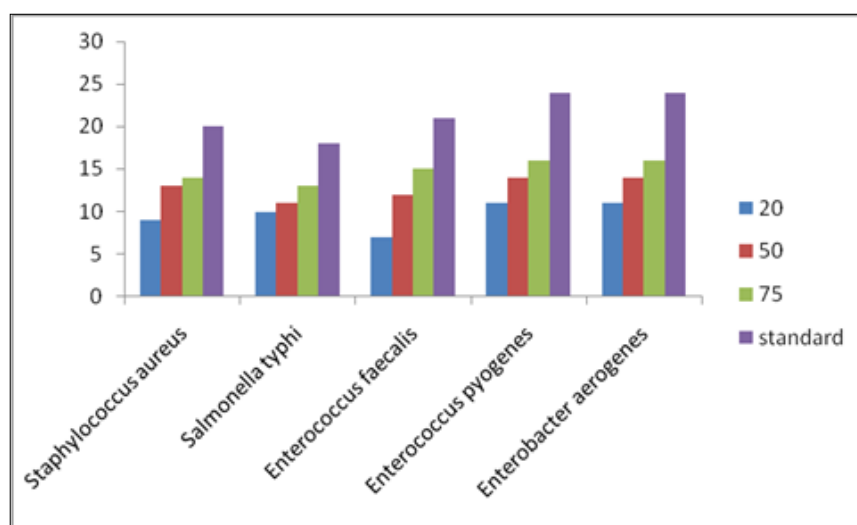


Fig 3

Table 1: Antibacterial activity of AgNPs synthesis from *pogostemon benghalensis* Aerials part aqueous extract

Bacteria name	Zone of inhibition (mm)			Antibiotic disc
	25µl	50µl	75µl	Standard disc
<i>Staphylococcus aureus</i>	9 mm	13 mm	14 mm	20
<i>Salmonella typhi</i>	10 mm	11 mm	13 mm	18
<i>Enterococcus faecalis</i>	7 mm	12 mm	15 mm	21
<i>Enterococcus pyogenes</i>	11 mm	14 mm	16 mm	24
<i>Enterobacter aerogenes</i>	8 mm	10 mm	13 mm	23

### Conclusion

In conclusion, the present study reveals that the AgNPs inhibits the growth of all the tested pathogenic gram-positive and gram-negative bacteria, which indicates that the Ag<sup>+</sup> with plant extract may contain a broad range of antibacterial activity. The AgNPs can be used as biocompatible, effective therapeutic drug against bacteria.

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### Conflict of interests

The authors have nothing to declare

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