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Green synthesis and characterization of silver nanoparticles using leaf and stem aqueous extract of *Pauzolzia Bennettiana* and their antioxidant activity

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Abstract
Silver nanoparticles have received attention due to their physical, chemical, and biological properties and are thus applied in catalysis, nanodevice fabrication, drug delivery and in medicine. Green synthesis of (AgNPs) was achieved by using the leaf and stem aqueous extract *Pauzolzia bennettiana* and AgNO3. Reduction of silver ions into silver nanoparticles was observed as a result of the color change from pale yellow to dark brown. The synthesized nanoparticles were characterized by UV-VIS and FTIR. UV-Visible spectrophotometer showed absorbance peak at 442nm for AgNps leaf extract and at 428nm for AgNps stem extract. Fourier transform infrared spectroscopy (FTIR) analysis revealed that the functional groups responsible for the stabilization of AgNps. The synthesized silver nanoparticles of aqueous leaf extracts have shown good potential source of antioxidants as compared to silver nanoparticles of aqueous stem extracts.

Keywords: Pauzolzia bennettiana, green synthesis, silver nanoparticles, UV, FTIR

Introduction
Plant mediated nanoparticle synthesis is purely a green synthetic route, which has great potential for many applications. Indian medicinal plants have been assured extraordinary importance due to the therapeutic properties of the phytochemicals present in the plant. Medicinal plants are a source of many potent and powerful drugs and used all over the world for the treatment of several chronic diseases. Plant derived nanoparticles appear to play a role in interspecies communication and have been postulated to function against inflammatory diseases and cancers [1].
Nanotechnology can be applied in almost all areas of human life. Silver nanoparticles have attracted and demandable research of interest in the field of nanotechnology. A number of biological approaches are already reported for the synthesis and stabilization of silver nanoparticles [2-6]. The silver nanoparticles exhibits many medical applications such as antifungal, antiplatelet antibacterial, antimicrobial and anticancerous activity [7-11]. Silvernanoparticles are extraordinarily efficient for nanomedicine. Additionally silver nanoparticles are interacting strongly with biomolecules. In this study, the silver nanoparticles were synthesized by using leaf and stem extracts of plant *Pauzolzia bennettiana*and silver nitrate solution. Pouzolzia is a genre of flowering plants in the nettle family. Throughout the tropical world 35 species are distributed.

Materials and Methods
Collection and Preparation of the aqueous leaf and stem extracts
Plant description
Plant Name: Pauzolzia bennettiana var.mysorensis
Family: URTICAEAE
Family (As per the Plant List): Urticaceae
Species: *Pauzolzia bennettiana*
Variety: mysorensis
Habit: Shrub

Plant collection

***Pouzolzia bennettiana* plants were collected from the Jagathala village, Aravankadu in Niligiris District.**

Preparation of extracts

About 20g of leaf and stem of plant *Pauzolzia bennettiana* was weighed separately and transferred into 500ml beaker containing 200 ml of distilled water and stirred with magnetic stirrer at 190–220 rpm for 20 minutes.

Silver Nanoparticle Synthesis

20ml of *Pauzolzia bennettiana* leaf extract was mixed with80ml of 0.01M of aqueous silver nitrate solution ($AgNO_3$). The green coloured solution changed to dark brown within 10 minutes which indicated the formation of AgNPs. Similarly AgNPs were synthesized from the stem extracts also. Here a colour change from pale yellow to brown indicated the formation of AgNPs.

Characterization of AgNPs

UV-absorption spectra of synthesized AgNPs by using leaf and stem extracts of *P. bennettiana*were measured using UV-visible spectrometer (JASCO variant 630 spectrometer). Fourier transform infrared (FTIR) spectral measurements were carried out on the Thermal science-Nicolet Si5, ATR-iD1 spectrometer to identify the potential phytochemical constituents of plant *P. bennettiana*.

Determination of Total antioxidant activity

Phosphomolybdenum method is one of the most commonly employed method for determining the total antioxidant activity [12]. 1.0 ml of the extract was mixed with 1.0 ml of the standard reagent solution (0.6M sulphuric acid, 28mM sodium phosphate and 4mM ammonium molybdate). The tubes were capped and incubated in a thermal block at 95°C for 90 min. After cooling to room temperature, the absorbance was measured at 695 nm against a reagent blank. The total antioxidant capacity was expressed as milligram of Ascorbic Acid Equivalence (AAE) per gram of extract. Phenolic compounds are a major contributor of antioxidant activity [13].

Results and Discussion

UV-Visible spectroscopy of synthesized AgNPs

One of the most important features in the optical absorbances spectra of metal nanoparticles is surface plasmon band, its resonance frequency is the plasma frequency adjusted by the size and the shape of the particle [14]. The spectrum displayed the characteristic surface plasmon resonance band of silver nanoparticles synthesized from stem extract at 428nm (Fig.1). Similarly spectrum showed a peak at 442nm (Fig 2) for silver nanoparticles of leaf extract.

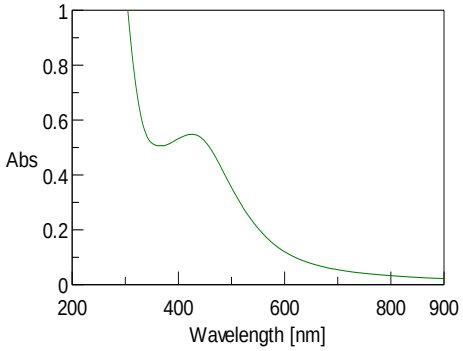


Fig 1: UV-Visible spectra of AgNPS stem extract

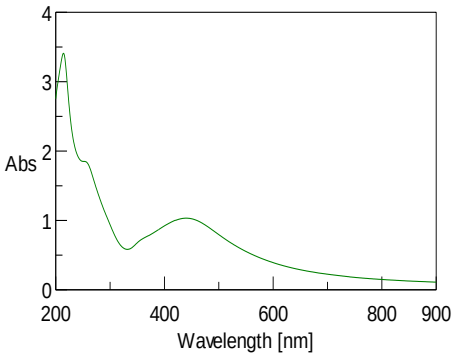


Fig 2: UV-Visible spectra of AgNPS leaf extract

FTIR spectra of AgNPs from *Pouzolzia bennettiana* stem and leaf

The functional groups of *Pouzolzia bennettiana* leaves and stem responsible for the bio-reduction of $AgNO_3$ into Ag nanoparticles can be explained from FT–IR analysis. In figure 3, 4 prominent bands of absorption observed at around 3980, 3307, 2925, 1636, and 1281cm-1. The peak at 3751cm-1 corresponding mainly to OH stretching vibration of free alcoholic group. The peak at 3307 cm-1 corresponding to N-H stretching frequency of amine group. The peak at 2925-2852 cm^{-1}corresponding to C-H aldehydic stretching frequency. The peak at 1736 cm^{-1} is corresponding to carbonyl stretching frequency. The peak at 1636 cm^{-1} corresponding to enol stretching frequency. The peak at 1282-1164 cm^{-1}C-OH stretching frequency. These stretching vibrations represent compounds like alkaloids, tannins and terpenoids.

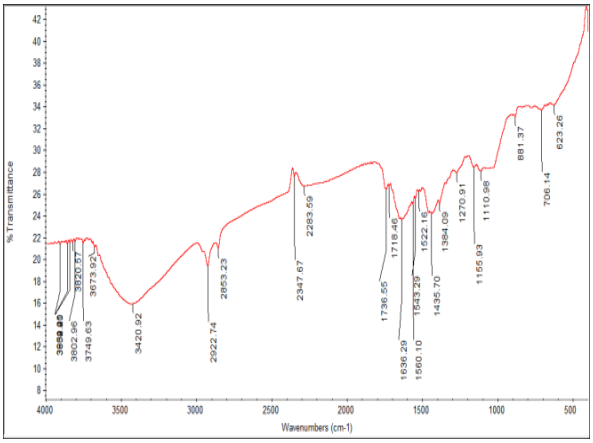


Fig 3: FTIR spectra of AgNPs (*P. bennettiana*stem)

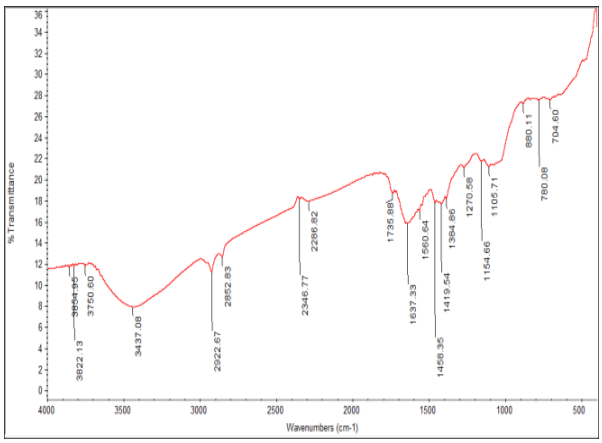


Fig 4: FTIR spectra of AgNPs (*P.bennettiana*leaf)

Phytochemical analysis of leaf and stem extracts of *P.bennettiana*

Phytochemicals are non-nutritive plant chemicals that have protective or disease preventive properties. Medicinal plants are a source for a wide variety of natural products, such as the phenolic acids and flavonoids which are very interesting for their antioxidant properties [15]. The phytochemical analysis was done by the method of standard procedure sand the reports were tabulated (Table 1)

Determination of Total antioxidant activity

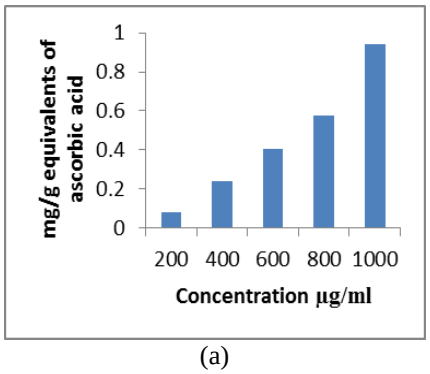
Now a days there is the demand of more effective antioxidants in natural origin. Reducing compound in the herbal plant leaves could be involved in the oxidative defense mechanism [16]. Antioxidants are nutrients, provide some protection against toxic by-products and fights off infection in the body. The radical scavenging properties of medicinal plant leaves is related to the total phenolic content[17].The phosphomolybdenum method of total antioxidant capacity test based on the reduction of Mo (VI) to Mo (V) by the antioxidant compound and formation of a green phosphate/Mo(V) complex. From the absorbance value, the potential of antioxidant activity was determined. The results presented in Fig.5, 6 showed the antioxidant activities of stem and leaf extracts of AgNps (Fig. 1&2).

Conclusion

The silver nanoparticles were synthesized by using leaf and stem extracts of plant *Pauzolzia bennettiana*and silver nitrate solution. UV-Visible spectrophotometer showed absorbance peak at 442nm for AgNps leaf extract and at 428nm for AgNps stem extract. (FTIR) analysis confirmed the functional groups phenols, tannins, terpenoids and other bioactive compounds. Antioxidant studies revealed that the synthesized silver nanoparticles of aqueous leaf extract was found moderate activity and stem extract was found lesser activity. In future, these green synthesised silver nanoparticles can potentially be used for different medical applications.

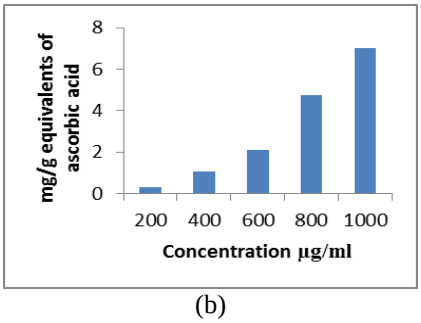
Table 1: Phytochemical analysis of leaf and stem extracts of *P.bennettiana*

Phytochemicals/Tests	P. bennettianaleafextract	P.bennettianastemextract
Alkaloids-Wagner's Test	Present	Present
Flavonoids-Sodium hydroxide Test	absent	absent
Tannins-Braymers Test	Present	Present
Phenols-Ferric Chloride	Present	Present
Protein-Ninhydrin Test	Absent	Absent
Anthocyanin-sodium hydroxide test	Absent	Absent
Triterpenoids/steroids-Salkowaski Test	Present	Present
Glycosides-Legal Test	Present	Present



(a)

Fig 1: Antioxidant activity of AgNPs stem extract



(b)

Fig 2: Antioxidant activity of AgNPs leaf extract

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