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"GREEN SYNTHESIS OF SILVER NANOPARTICLES(AgNPs) FROM THE AQUEOUS LEAF EXTRACT OF *MUTINGIA CALABURA* AND ITS CHARACTERIZATION STUDIES"

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ABSTRACT

Nanotechnology is a field of science which deals with production, manipulation and use of materials ranging in nanometers. Nanoparticles have expressed significant advances owing to wide range of applications in the field of bio-medical, sensors, antimicrobials, catalysts, electronics, agricultural, bio-labeling and in other areas. Green synthesis of nanoparticles is a promising area, have prominent applications in the field of physics, chemistry, biology and medicine. There are several methods used for the synthesis of silver nanoparticles. It includes both physiochemical and biological methods. The use of environmentally benign materials like plant leaf extract for the synthesis of silver nanoparticles offers number of benefits in both pharmaceuticals and biomedical fields.

KEYWORDS: Nano technology, green synthesis, AgNPs, characterization studies.

INTRODUCTION

Nanotechnology is a field of science which deals with production, manipulation and use of materials ranging in nanometres. In nanotechnology nanoparticles research is an important aspect due to its innumerable applications. Nanoparticles have expressed significant advances owing to wide range of applications in the field of bio-medical, sensors, antimicrobials, catalysts, electronics, agricultural, bio-labelling and in other areas. As opposition with the conventional methods to produce nanoparticles, the term herbal nanotechnology has become of more interest as it makes use of nanoparticles which are made from herbal extracts and are less hazardous when interacted with human as it uses less toxic chemicals.

Nanotechnology is a fast expanding area of science.this area of research is anticipated to lead to the development of novel, multifunctional applications which can recognize cancer cells, deliver drug to target tissue, aid in reporting outcome of therapy. In nanotechnology a particle is defined as a small object that behave as a whole unit with respect to its properties and transport. Particles are further classified according to diameter. Green synthesis of nanoparticles is a promising area, because of its applications in the field of physics, chemistry, biology and medicine. The synthesized nanomaterials are used in medicinal and technological aspects. There are several methods used for the synthesis of silver nanoparticles. It includes both physiochemical and biological methods.

Biological synthesis also called green synthesis of nanoparticles where biological enzymes from plant extracts, fungi, bacteria are used. This method is ecofriendly and reduces toxicity and waste production. Silver nano particles synthesized by green chemistry methods offer a novel and potential alternative to chemically synthesized nano particles. Evaporationcondensation and laser ablation are the two important physical methods used for the production of silver nanoparticle. The absence of solvent contamination in the prepared thin films and the uniformity of nano particle are the main advantages of physical synthesis methods.

The present study focus on the synthesis of silver nanoparticle by an aqueous leaf extract of *Muntingia calabura*. These silver nanopartilces were found to be extremely effective against different bacterial and fungal pathogens. The common name for *Muntingia calabura* is calabur tree, panama berry, ornamental cherry, jam fruit tree, etc. Various phytochemical compounds have been isolated from different parts of *M. calabura*. the bioactive compounds from the roots of *M. calabura* were first isolated by Kaneda *et al.*, (1991). They isolate 12 flavanoids from methanol extract of *M.calabura* roots namely such as (2S)-5'-hydroxy-7,3',4'-tri methoxy flavan, (2S)-2'-hydroxy-7,8,3',4',5'-penta methoxy flavan, (2S)-7,8,3',4',5'- penta methoxy flavan, (2S)-8,8''-5'-tri hydroxy-7,7'-3',3'''-4',4'''-5'''-hepta methoxy-5,5''-bi flavan ... etc.

MATERIALS AND METHODS

Collection of leaf samples from M. calabura

Leaves that appeared healthy were collected from different branches of *Mutingia calabura* from Ottapalam. The plant leaves were collected and brought to the laboratory in a sterile container.

Preparation of plant extract

The leaves was taken and subjected to aqueous extract preparation, 25g of green tender leaves were thoroughly washed with tap water followed by double distilled water twice. The leaves are cut in to small pieces and were boiled in 100ml of distilled water. After 15 minutes the aqueous extract was filtered through What man No.1 filter paper. The aqueous extract of the plant used as reducing agent for the synthesis of silver nano particles.

Preparation of silver nitrate solution

A concentration of 1M AgNO₃ solution was prepared by dissolving 0.169 AgNO₃ in 1000ml of double distilled water and used for the green synthesis of silver nano particles (AgNPs).

Green synthesis of AgNPs

The filtered aqueous extract of *M.calabura* leaves was added individually to 90 ml of 1M AgNO₃ in a 250 ml Erlenmeyer flask. Then kept in room temperature for 48hrs at dark. The process was continued till the change of colour occurred from yellow to dark brown indicating the completion of silver nanoparticle synthesis.

UV- visible spectra analysis

The reduction of pure silver ions was observed by UV - visible spectroscopy. The absorption maxima were measured by using UV spectrophotometer between 300-800nm wavelength.

Characterization of silver nanoparticle using electron microscope (SEM)

SEM analysis was carried out to determine the particle morphology. The biologically synthesized silver nanoparticle sample was centrifuged. The pellet was collected and dried. The fine sample was used for SEM analysis. The characterization of silver nanoparticle by SEM was done at the Department of Nano sciences & Technology, Bharathiar University, Coimbatore.

FTIR analysis

The Fourier Transform Infrared Spectroscopy (FTIR) analysis was carried out to know the different functional groups that act as bioreductors into reduce Ag^+ ions to Ag_0 . The leaf extract and nanoparticle sample were subjected to FTIR in which the samples were irradiated by a broad spectrum of infra-red light and the level of absorbance at a particular frequency was plotted after fourier transformation of the data. Compounds contained

in the extracts were identified according to standard infrared chart.

Phytochemical analysis (Qualitative approach)

The phytochemical analysis was performed to identify the presence or absence of different secondary metabolites like alkaloids, tannins, flavonoids, sterols.. etc.

A: Test for alkaloids

Mayer's test: To 2 ml of plant extract 3 ml of ammonia was added and allowed to stand for few minutes. 3ml of chloroform was added to it and allowed to stand in a water bath till chloroform evaporates. Add Mayer's reagent, cream colour indicate the presence of alkaloids.

B: Test for steroids and triterpenoids

To 2 ml Plant extract few drops of acetic anhydride added. Boiled and then cooled. Concentrated H_2SO_4 was added from the sides of the test tube and then observed for the formation of brown ring at the junction of two layers.

C: Test for phlobatannin

To 1 ml of plant extract few drops of concentrated HCl was added. It allowed to stand in a water bath for few minutes. Red colour precipitate indicate the presence of phlobatannins.

D: Test for reducing sugar

To 2 ml of plant extract was taken in a test tube and 1 ml ethanol and Fehlings solution A and B was added and boiled for two minutes. Red colouration indicate the positive result.

E: Test for flavonoids

To 5 ml of ammonia and few drops of concentrated sulphuric acid was added to 2 ml of plant sample. Yellow colour indicate the presence of flavonoids.

F: Test for saponins

To 1 ml extract was treated with 1% lead acetate solution. Formation of white precipitates indicates the presence of saponins.

RESULT AND DISCUSSION *Muntingia calabura* **PLANT**



Phytochemicals	Aqueous extract
Alkaloids	++
Reducing sugar	++
Terpenoids	+
Steroids	+
Flavanoids	++
Saponins	-
	Alkaloids Reducing sugar Terpenoids Steroids Flavanoids

Table 1: Phytochemical evaluation of Muntingiacalabura leaf extract.

(+) Presence; (++) High level; (-) Absence

Phytochemicals are naturally found in plants, they are biologically active and function to protect plants against invasion, disease and infection. Phytochemicals



Aqueous leaf extract with silver

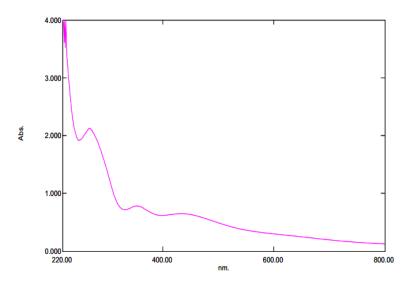
Nitrate solution Characterisation

Characterisation of Green Synthesised Silver Nanoparticle

1. UV spectrum analysis of biologically synthesised nanoparticle

The absorption spectrum showed maximum peak at 430nm for the nanoparticle synthesized using aqueous extract of *Muntingia calabura* leaves.

UV-Visible spectrum of silver nanoparticles using the aqueous leaf extract of Muntingia calabura.



produced by plants through primary and secondary metabolism, and play an important role in plant growth. In the present study is based on the synthesis of silver nanoparticles using aqueous extract of *Muntingia calabura* leaves.

Biological Synthesis Of Silver Nanoparticles

Silver nano particles were syntheised by using aqueous extract of *Muntingia calabura* leaves. The plant extract were pale yellow in colour before the addition of silver nitrate solution .The synthesis of silver nano particles exhibited as the colour change from yellow to brown. The samples were observed periodically for the change of colour from pale yellow to different shades of brown.



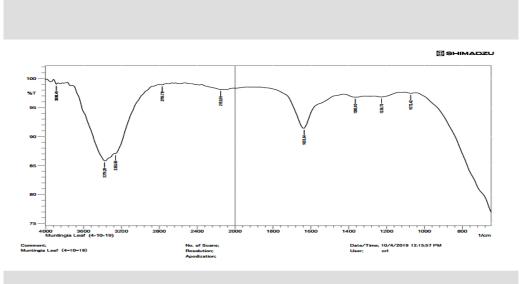
After the formation of AgNPs

2. Fourier Transform Infrared Spectroscopy

The FTIR analysis of M. calabura leaf extract shows peaks at 2862.36 indicates the presence of C-H stretch. The peak at 2098.55 shows the presence of C=C stretch (preferably belonging to a benzene ring). The peak at 1990.54 shows the presence of C-N groups while the peak at 1080 indicate the presence of C-F stretch.

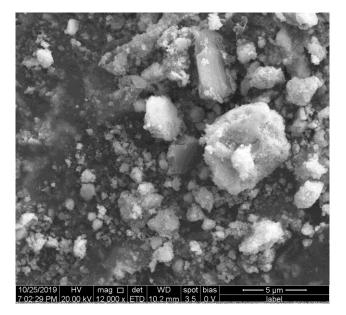
The FTIR analysis of nanoparticle sample (aqueous leaf extract of *M.calabura*) shows peak at 2769.78 indicate the presence of OH group (indicating the presence of alcohol). The peak at 2152 shows the presence of C-N group. The peak at 1072.42 indicate the presence of C-F group (alkyl halides)

FTIR spectra of AgNPs synthesized aqueous extract of *M. calabura*



SEM analysis

SEM images shows similar appearance for the presence of silver nanoparticles synthesized from *M.calabura*. The SEM images shows cluster of nanoparticles in different size. The SEM analysis confirmed the presence of nanoparticles.



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