

Research Article



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Formulation and evaluation bio-silver nano ointment using leaf extract of *Justicia adhatoda*

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ABSTRACT

Background

Nanotechnology is the most expeditiously emerging technology in the field of therapeutics as drug delivery system. Silver has been known to have effective bactericidal properties for centuries. Silver based topical dressing has been widely used as treatment for infection in burns and open wounds. *Justicia adhatoda* is a full dense evergreen shrub with thick branches, widely distributed in the south Asia and Indo-china. *Justicia adhatoda* is ethanomedically used as treatment of asthma, cough, bronchitis and tuberculosis, joint pain, lumber pain, sprains, eczema, malaria, rheumatism, swelling, venereal diseases.

Methods

The present research work cost effective and environmental friendly silver nanoparticles were synthesized from the ethanolic extract of *Justicia adhatoda* as a reducing agent. The characteristic of the obtained silver nanoparticles were studied using UV-Visible spectrophotometry. The bio-synthesized silver nanoparticles infused ointment formulation were evaluated for colour, odour, solubility, PH, melting point, density, spreadability and skin irritation test.

Results

Reduction of silver ions into silver nanoparticles using ethanolic leaf extract of *Justicia adhatoda* showed colour changed from greyish green to light brown due to excitation of surface vibration in silver nanoparticles. This indicate the formation of silver nanoparticles. The absorption spectrum of the biosynthesized silver nanoparticle showed absorbance peak at 539nm. This indicate that the plasmonic nanoparticles. The physicochemical characteristics of bio-silver nanoparticles infused ointment showed whitecolour, odourless, soluble in petroleum ether, insoluble in water, PH of 4.7, melting point 65°C, density 39gm/cc, spreadability of 6.12-11.50, no irritation and safe.

Conclusion

The present research draw the conclusion that synthesis of silver nanoparticles from the leaf of *Justicia Adhatoda* was confirmed by colour change from greyish green to light brown. This indicate the formation of silver nanoparticles. The characterization of the obtained silver nanoparticles were studied using UV-visible spectrophotometer. The bio-silver nanoparticles infused ointment physicochemical parameters was successfully evaluated. Further microbial and clinical tests are warranted to commercialization this product.

Keywords: Silver nanoparticles, UV-Visible spectrophotometer, *Justicia adhatoda*, *Acanthaceae*

INTRODUCTION

The development of nanoparticles for drug delivery began in 1960s¹. Recent development in multifunctional nanoparticles has offered a great potential for targeted delivery of drugs for treatments of various types of disease. Nanoparticles are basically solid colloidal particles ranging

in size from 1to1000nm. Nanoparticles are made from biocompatible and biodegradable materials such as polymers, either natural or synthetic or solid lipid nanoparticles²

Silver has been known to have effective bactericidal properties for centuries. Now a days, silver based topical dressing has been widely used as treatment for infection in

burns and open wounds³. Nano-sized drug delivery systems of herbal drug have a potential future for enhancing the activity and overcoming problem associated with plant medicines⁴

Justicia adhatoda belong to the family *Acanthaceae*. It is a full dense evergreen shrub with thick branches. The species is distributed in the south Asia and Indo-china. The trunk has many long opposite ascending branches, where the bark is yellowish in colour, flowers are usually white and the inflorescence show large, dense, axillary spikes, fruits are pubescent and are with club-shaped capsules.

Justicia adhatoda used in extensively in the treatment of asthma, cough, bronchitis and tuberculosis, joint pain, lumber pain, sprains, eczema, malaria, rheumatism, swelling, venereal diseases⁵. The literature survey of the plant revealed the presence of quinolone alkaloids vasicine, 7-hydroxyvasicine, 3 deoxyvasicine, vasicol, vasicolone, vasicolinone, adhatodine, betaine, steroids, flavonoids, carbohydrate and alkanes⁶.

The plant exhibited various pharmacological activities such as Anti- asthmatic and bronchodilator activity⁷, antibacterial activity⁸ wound healing activity⁹, Anti-Tursive activity¹⁰ Anti-Tubercular activity¹¹, Hepatoprotective activity¹², Anti-Thrombolytic activity¹³, Anti-diabetic activity¹⁴, Anti-inflammatory activity¹⁵, Abortifacient activity¹⁶.

Green synthesis of nanoparticles has gained attention because of its advantages, including being nontoxic, safe for humans, ecofriendly and economically viable, compared to chemical and physical synthesis method.¹⁷

The purpose of the study is formulation and evaluation of bio-silver nano ointment using leaf extract of *Justicia adhatoda*.

MATERIALS AND METHODS

Plant collection & authentication

Fresh leaves of the young plants of *Justicia adhatoda* were collected from medicinal garden located at B.S.AbdurRahman Crescent Institute of Science and Technology, Vandalur, Chennai, Tamilnadu during the month of March 2021 was authenticated by Dr. P. Jayaraman, Director of Plant Anatomy Research Centre, Chennai. Plant authentication receipt number was PARC/2021/4436 for *Justicia adhatoda*.

Preparation of ethanolic extract of *Justicia adhatoda* [EEJA]

Procedure

The shade dried and coarsely powdered leaf of *Justicia adhatoda*. 10g of the *Justicia adhatoda* powder was added with 100ml of ethanol and soaked overnight and after 17 hours over it is extracted in a distillation apparatus for 7 hours. The liquid extract was concentrated using hot plate to obtain the thick extract.

Synthesis of silver nanoparticles using ethanolic extract of *Justicia adhatoda* [EEJA]

Silver nitrate solution was prepared by adding 0.169g of silver nitrate in 1000ml of distilled water, using water

distillation apparatus for 5 hours by single distillation method. About 6-7 drops of the *ethanolic* leaf extract of *Justicia adhatoda* was added to the silver nitrate solution, the solution colour was gradually changed from greyish green to light brown colour is shown in Fig 1. The 1000ml of silver nitrate solution was evaporated by using Rotary Evaporator with 60 rpm at 95°C. Finally concentrated 20 ml of the silver nitrate solution were obtained and further the conc. solution refluxed additional for 15min by using hot plate. The heat was then turned off and the solution had been stirred using magnetic stirrer at 200 rpm and it will cool in a room temperature. Finally silver nano particles of 20g were obtained. It was stored at 4°C for further use.¹⁸

Characterisation of synthesized bio-silver nano particles by UV Spectrophotometer

The synthesized silver nanoparticles (solution of 1 mg/mL in distilled water as a dispersive medium) were monitored using UV-Vis spectrophotometer (Systronics double beam spectrophotometer 2202, India) between the range of [300nm and 550nm]. Distilled water was used as blank for UV-Spectrophotometer analysis. The UV spectrum of silver nanoparticle is shown in Fig 2. Characterization of synthesized silver nano particles by UV Spectrophotometer.

Formulation of bio silver nanoparticles infused ointment.

Required quantity of cetosteryl alcohol is melted in a china dish by using hot plate and then liquid paraffin (17.4g) were added and heated. At the same time in another china dish white soft paraffin (6g) was added and melted, and followed by cetosteryl alcohol and liquid paraffin mixture were added. Finally accurately weighed 20 gm of the extract infusing nanoparticles dissolved in 30ml of water add drop wise while stirring and allowed to cool, to obtain ointment state as the final product.

Evaluation of physicochemical parameters for bio-silver nanoparticles infused ointment

Physicochemical parameters with regards to silver nanoparticles using *Justicia adhatoda* such as colour, odour, solubility, melting point, density and skin irritation test were checked. Aqueous solution (1%) of the formulation was measured by using a calibrated digital PH meter at constant temperature. The results are tabulated in Table 1.

Spreadability

Glass slides with standard dimension (length of 6.0 cm) were taken. Topical ointment formulation was placed on the one side of the glass slide and sandwiched with the help of another slide. Remove the adhering ointment on the outer surface of the glass slides by wiping. Slides are fixed in a stand that only upper slide to slip off freely without any disturbance by force of weight (5 g) tied to it. Time taken for the movement of upper slide to the distance of 6.0 cm was measured. Measurement of spreadability was done in triplicate and calculated by using the following formula:

$$\text{Spreadability} = (\text{Weight} \times \text{Length}) / \text{Time}$$

Where,

S = Spreadability,

m=Weight tied to the upper slide (20 g),
l=Length of the glass (6.0 cm),
t=Time taken in seconds.

RESULTS AND DISCUSSION

Characterization of Silver Nanoparticles

Colour changes indicating the synthesis of silver nanoparticles.

Reduction of silver ions into silver nanoparticles using ethanolic extract of *Justicia adhatoda* was evidenced by the visual change of colour from greyish green to light brown due to excitation of surface vibration in silver nanoparticles as shown in Fig 1.



Fig 1: Silver nanoparticles synthesized by using silver nitrate using ethanolic extract of *Justicia adhatoda* [EEJA]

UV visible spectroscopy

UV visible spectroscopy is utilized for analyze size and shape of nanoparticles in aqueous suspensions and the UV visible spectrum was recorded after few hour incubation of

the synthesized. Silver nanoparticles in dark place. The absorption spectrum of the silver nanoparticles have an absorbance peak at 539nm is shown in Fig 2 This indicate that the plasmonic nanoparticles.



Fig: 2 UV- Visible spectron of Ag Nanoparticles, synthesized by reducing AgNO_3 using ethanolic extract of *justicia Adhatoda* [EEJA]

Evaluation of Physicochemical parameters of bio-silver nanoparticle infused ointment

Physicochemical parameters of bio-silver nanoparticle infused ointment showed in the following result and displayed in Table 1.

Table 1: Physicochemical parameters of bio-silver nanoparticles infused ointment		
Sl.No	Parameters	Observation
1.	Colour	White
2.	Odour	Odorless
3.	Solubility	Soluble in Petroleum ether Insoluble in water
4.	pH	4.7
5.	Melting Point	65°C
6.	Density	39g/cc
7.	Spreadability	6.12mm-11.50mm
8.	Skin Irritation Test	No irritation

CONCLUSION

The present research draw the conclusion that synthesis of silver nanoparticles from the leaf of *Justicia Adhatoda* was confirmed by colour change from greyish green to light brown. This indicate the formation of silver nanoparticles. The characterization of the obtained silver nanoparticles were studied using UV-visible spectrophotometry. The bio-silver nanoparticles infused ointment physicochemical parameters was successfully evaluated. Further microbial

and clinical tests are warranted to commercialization this product.

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