International Journal of Current Advanced Research
ISSN: O: 2319-6475, ISSN: P: 2319-6505, Impact Factor: 6.614
Available Online at www.journalijcar.org
Volume 8; Issue 03 (D); March 2019; Page No.17844-17846
DOI: http://dx.doi.org/10.24327/ijcar.2019.17846.3399

Research Article

ANALYZING ANTIFUNGAL ACTIVITY OF GREEN NANOPARTICLES FROM *SYZYGIUM cumini*

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ARTICLE INFO

Article History:
Received 15th December, 2018
Received in revised form 7th January, 2019
Accepted 13th February, 2019
Published online 28th March, 2019

Key words:

Syzygiumcumini, Silver nanoparticles, SEM, TEM, Antifungal properties

ABSTRACT

The nanoparticles were synthesized under various temperature conditions by organic means from the leaf extracts of Syzygiumcumini (Jamun). The efficacy of these metallo nanoparticles was tested against fungi. Thepresence of Silver nanoparticles wasinitiallyanalyzed by color changes of reaction mixture andby Calorimetric readings. The spherical shape of these nanoparticles was confirmed using Scanning Electron Microscopy (SEM).The diameter of synthesized Silver nanoparticles was found within the range of 20-50 nm as confirmed by Transmission Electron Microscopy (TEM). These nanoparticles were assessed for their antifungal potentials against Aspergillusniger and Penicillium sp. by measuring the zone of inhibition. The maximum zone of inhibition was observed against Penicillium sp. using 4 ml extract concentration at 600 C but the activity of these particles was not observed against A. niger.

INTRODUCTION

Nanotechnology deals with tiny nanoparticles which are extensively used in research [1]. These nanoparticles are synthesized by both Chemical and organic means. But the Organic (Green) synthesis technology has led to economic growth due to its cost effectiveness and hence improved the national security [2]. These organo nanoparticlesare easily available, safe and non-toxic in various cases. Various techniques are used to analyze the shape and size of silver nanoparticles viz. Scanning Electron Microscopy (SEM) is used to see the shape while Transmission Electron Microscopy (TEM) to study the size [3].

Syzygium cumini (Jamun), a tropical plant belonging to Myrtaceae family, has numerous advantages. It has a wide variety of metabolites that can aid in the reduction of silver ions. The plant possesses anti- fungal and anti- inflammatory properties.The leaves have maximum Vitamin A and C content and hence cure a number of skin diseases. The silver nanoparticles of various sizes can be synthesized from this precious plant.These organically synthesized nanoparticles represent antimicrobial properties with possibly novel mechanisms of action.

Thus the present study was envisaged with the objectives of

- To Synthesize and characterize silver nanoparticles from the leaf extracts of *Syzygium cumini*
- To study the antifungal properties of these nanoparticles

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MATERIAL AND METHODS

Preparation of Syzygiumcumini leaf Extracts

Fresh *Syzygium cumini* (Jamun) leaves were collected from garden near Doraha. These leaves were a washed number of times with distilled water and allowed to air dry and grounded.Thirtygm of leaves were weighed and added in 5 times double distilled water. The extract was then percolated at 100^0 C for 2 hours. The leaf extract was filtered through Whatmann filter paper no.1 to obtain the concentrate.The pH of leaf extract was determined and stored it in a refrigerator at 4^0C.

Biosynthesis of Silver Nanoparticles

Reagents and Chemicals

For the biosynthesis of Silver nanoparticles, Silver nitrate solution was prepared by dissolving 1M silver nitrate in 1000ml distilled water. To avoid photo reactivation of silver nanoparticles, the reaction mixture was incubated in the dark at 30^0 C.

Synthesis Process

Both the filtered leaf extract and Silver nitrate solution were mixed for the synthesis of silver nanoparticles in a clean sterilized conical flask. Two sterilized conical flasks of 50 ml capacity were taken and poured 2 ml leaf extract along with50 ml Silver nitrate solution in one and 4 ml leaf extract and 50 ml solution in other flask. Then this mixture was kept on a water bath for 2 hours at 60^0 C and 100^0 C temperatures, respectively.

Characterization of Silver Nanoparticles

Preliminary Analysis

Change in Colour

The formation of organically synthesized nanoparicles was first confirmed by colour changes in the extract i.e. light Rosewood to Brown.

Calorimetric Analysis

The Absorbance of these Silver nanoparticles was monitored at 670nm.

Confirmatory Analysis

Scanning Electron Microscopy (SEM)

The shape and surface morphology of Silver nanoparticles was evaluated by using SEM. SEM image was analysed on Model TM3000.These nanoparticles were identified at Sophisticated Analytical Laboratories at Thapar University, Patiala.

Transmission Electron Microscopy (TEM)

The size of the synthesized silver nanoparticles was examined by using TEM. TEM image was recorded on Model H-7650 TEM machine at voltage of 80KV at Nanoscience Technology Department, in Punjab Agricultural University, Ludhiana.

Antifungal Activity

The two fungal cultures, *Aspergillusniger* and *Penicillium* sp. were isolated by pour plating technique on PotatoDextrose Agar (PDA) media.These fungi were characterized by following Standard AOAC methods. A Disc Diffusion method was adopted to evaluate the antifungal activity of AgNPs [4].Then these fungal isolates were cultured separately on PDA by spread plating. The sterilized filter paper discs were dipped in AgNPs solution (2ml and 4ml) and placed on cultured petriplates.

These plates were incubated at 25±2^{0}C for 6 days to see the zone of inhibitionwhich indicates antifungal activity.

RESULTS AND DISCUSSION

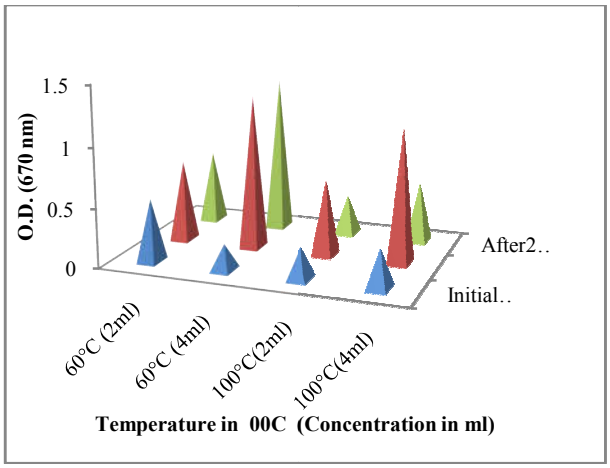
Syzygiumcumini leaf extractfiltrate was used for production of silver nanoparticles. When the leaf extract filtrate was mixed with Silver nitrate solution, Silver ions were reduced into silver nanoparticles. The synthesis of these nanoparticles was represented by thecolour changes in extract filtrate from light Rosewood to brownish at different time and temperatures (Table 1).The similar results were observed by [5, 6]. The pH of the extract filtrate was found to be acidic (5.7). The acidic pH is generally favourable for the growth of Fungi [7].

Table 1 Change in colour of solution during silver nanoparticles synthesis

Time Interval	Color Change
Initial	Light Rosewood
After one hour	Golden
After two hours	Brown

Calorimetry

The Optical Density (O.D.) of the extract filtrate was taken at 670nm. The maximum absorbance of Jamun leaf extract filtrate was observed at 60^0 C after 2 hours. Our results were on consonance with the results of [8].



Graph 1 Absorbance (670nm) of leaf extracts prepared at 60^0 C and 100^0 C (After 1 hr and after 2 hrs)

SEM Analysis

The spherical shape of silver nanoparticles was confirmed by SEM. But the SEM micrograph of the Silver nanoparticles did not represent uniform surface. Similar results were observed by [9].

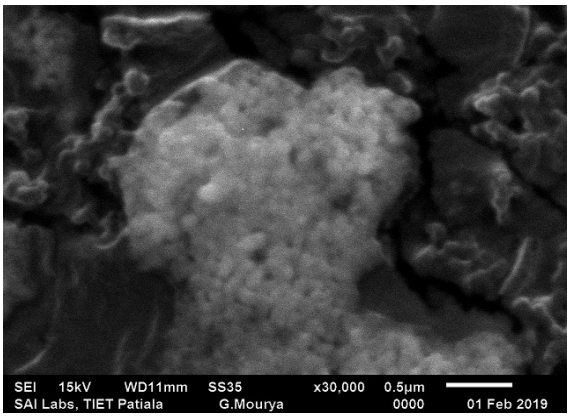


Figure 1 SEM Analysis of AgNPs from *Syzygiumcumini* leaf extracts

TEM Analysis

The synthesized nanoparticles were of 50nm size as determined by TEM analysis. Our results are in consonance with the results of [10,11].

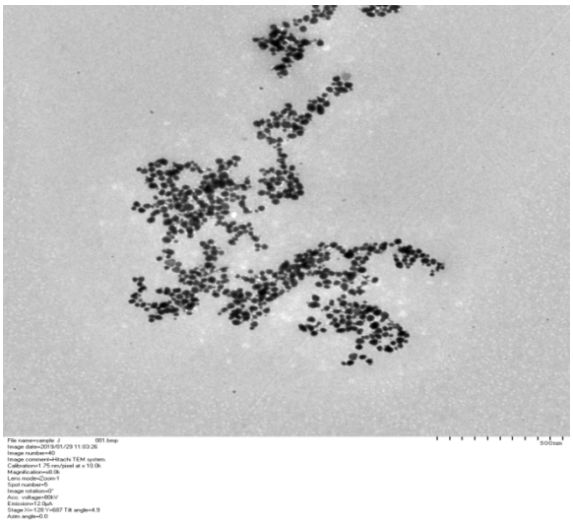


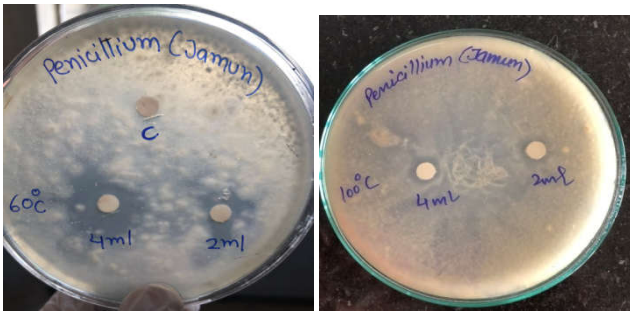
Figure 2 TEM Analysis of Ag NPs synthesized from *Syzygium cumini* leaf extracts

Antifungal Assay

Antifungal tests indicated that the silver nanoparticles synthesized in Jamunleaf extracts showed anti-fungal activity against*Penicillum* sp. only but did not show any activity against *Aspergillusniger*. In *Penicilliumsp.*, the biggestzone of inhibition was recorded at 4ml concentration (12 mm) than at 2 ml concentration (8 mm) at 60^0C. The smallest zones of inhibition (7 mm) were observed at 100^0 C temperature (at 2 ml concentration). Various scientists also reported the increase in halo zones against fungi with the increase in concentration of nanoparticles.

Table 1 Halozones (mm) against *Penicillium sp*. and *A. niger*

Temperatures	**60°C**		**100°C**	
Concentration	**2ml**	**4ml**	**2ml**	**4ml**
Fungal Isolates	Zone of inhibition in (mm)			
Penicilliumsp.	8	12	7	8
Aspergillusniger	Nil	Nil	Nil	Nil



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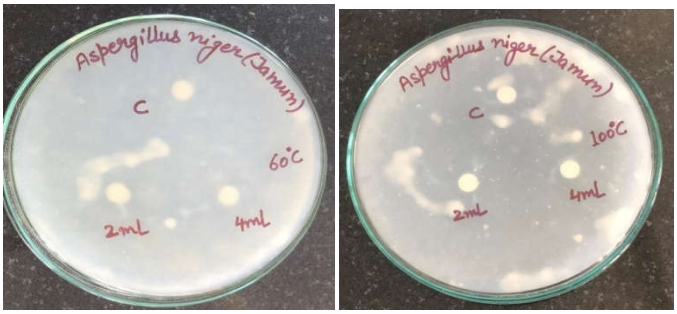
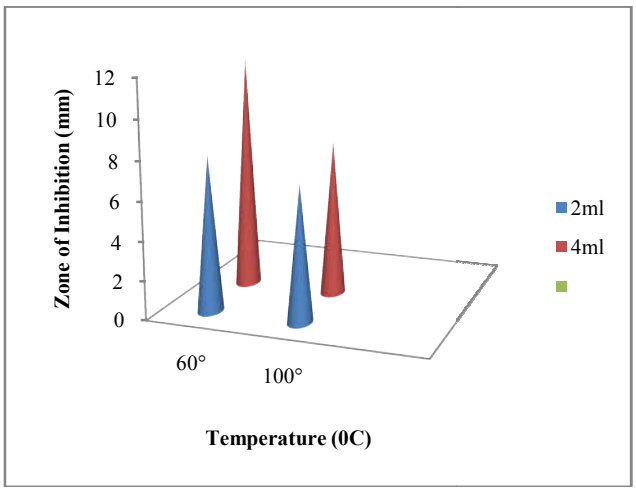


B

Figure 3 Antifungal activity of AgNPs against a) *Penicillium sp.* and b) *Aspergillusniger by* Disc Diffusion method



Graph 2 Antifungal Effect of AgNPs against *Penicilliumsp.*

CONCLUSIONS

The AgNPs were synthesized by a traditional organic way in the present study. These nanoparticles were inhibitory to *Penicillium* sp.These green nanoparticles can also be used to test their efficacy against a number of other pathogenic microbes too. The average size of synthesized spherical shaped silver nanoparticles is 20-50nm. The reduction of metal ions through leaf extracts and fruit leads to the formation of silver nanoparticles of fairly well defined dimensions.

Bibliography

1. Panigrahi T. Synthesis and characterization of silver nanoparticles using leaf extract of *Azadirachtaindica,* Thesis submitted to Department of Life Science, National Institute of Technology, Rourkela, Orissa, India 2013; 1-70
2. Mallmann EJ, Cunha,FA and Fechine PBA. Antifungal activity of silver nanoparticles obtained by green synthesis. Revista doMedicina Tropical de Sao Paulo2015; 57 (2): 165-167.
3. Bhalodia NR and Shukla VJ. Antibacterial and antifungal activities from leaf extracts of *Cassia fistula*: An ethno medicinal plant. Journal of Advanced Pharmaceutical Technology and Research 2011; 2(2):104-109.
4. Kumar P, Selvi S and GovindarajuM. Seaweed-mediated biosynthesis of silver nanoparticles using *Gracilariacorticata* for its antifungal activity against *Candida spp.*2012; 3: 495-500.
5. Kaur J, Gupta N, Kaur M and Chatli AS. Exploring Antifungal Activity of organically synthesized silver nanoparticles from leaf extracts of *Daturametel*. International Journal of Basic and Applied Research2019; 9(3): 651-658.
6. Gupta N, KomalR and Chatli AS. Development and evaluation of Hemolytic Efficacy of Guava (*PsidiumGuajava*) based green nanoparticles. International Journal of Research in Engineering , Science and Management 2018; 1 (1): 2581-579.
7. Chatli AS, Sood S and Beri V. Effect of pH on inorganic phosphate solubilisation by microorganisms isolated from Trans Himalayan Region of Himachal Pradesh. International Journal of Multidisciplinary Research and Development 2015; 2 (9): 31-35.
8. Kaur J, Gupta N, Kaur M and Chatli AS. Antibacterial Effects of Green Synthesized AgNPs from *Daturametel* Leaf Extracts. International Journal of Pure and Applied Bioscience 2019; 7(1): 247-252.
9. El-Samawaty AM and Yassin MA. Antifungal silver nanoparticles: synthesis, characterization and biological evaluation. Journal Biotechnology and Biotechnological Equipment 2016; 7 (2): 56-62.
10. Butler KS, Peeler DJ, Casey BJ, Dair BJ and Elespuru RK. Silver nanoparticles: correlating nanoparticles size and cellular uptake with genotoxicity. Oxford Academic Journals2015; 30: 577-591.
11. Kaur M, Gupta N, Kaur J and Chatli AS.Biosynthesis and characterization of AgNPs and their antibacterial effects. International Journal of Advanced Research in Biological Sciences 2019; 6 (3): 226-231.