ORIGINAL RESEARCH PAPER

Engineering

GREEN SYNTHESIS OF SILVER NANOPARTICLES FROM GIVOTIA ROTTLERIFORMIS AND STUDIES ON ITS ANTI BACTERIAL AND ANTI FUNGAL ACTIVITY

KEY WORDS: .

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ABSTRACT

In this paper, silver nanoparticles are synthesized from Givotia Rottleriformis. The silver nanoparticles are well known to possess best antimicrobial, antifungal, antiinflammatory and anticancer properties. Although there are many ways to synthesize the silver nanoparticles, green synthesis is said to be cost effective, eco friendly, good compatibility and exhibit controlled and targeted drug delivery. The silver nanoparticles can absorb ten times as much light which is said to be a major advantage over gold. Hence it can be used in combination with fluorescence emission detection. The reduction process was monitored using a UV-visible spectroscopy with a peak showed at 450nm. It is then tested against the antimicrobial activity using Escherichia coli, Bacillus subtills, Klebsiella pneumonia, Staphylococcus aureus species of bacteria and Aspergillus niger and Aspergillus flavus species of fungi.

Introduction

The development of green processes for the production of nanoparticles is evolving into a significant branch of nanotechnology. Nanoparticle is a core particle which performs as a whole unit in terms of transport and property. The optical, electronic, mechanical, magnetic, and chemical properties are significantly different from those of bulk materials.

Nano size particles are quite unique in nature because nano size increase surface to volume ratio and also its physical, chemical and biological properties are different from bulk materials. So the main aim to study its minute size is to trigger chemical activity with distinct crystallography that increases the surface area. Thus in recent years much research is going on metallic nanoparticle and its properties like catalyst, sensing to optics, antibacterial activity, data storage capacity. Nanotechnology is a fast growing area in the field of science which is a interdisciplinary field of both science and technology that increase the scope of investing and regulating at cell level between synthetic material and biological system.

Preparation of the Extracts

The leaves of Givotia Rottleriformis were collected from parts of Southern India. Primarily they were thoroughly washed with distilled water to remove dirt particles. Cleaned herbal parts (leaves) were dried with water absorbent paper (filter paper). It was then kept to dry completely for 24 hours at room temperature. 10g chopped leaves of the plant leaves was dispensed in 100 ml of deionised water and boiled for 15-20 min. at 80ºC using water bath. The aqueous plant extract was filtered through Whatman filter paper no.1 and is used further for the synthesis of silver nanoparticle.

Synthesis of Silver Nanaoparticles

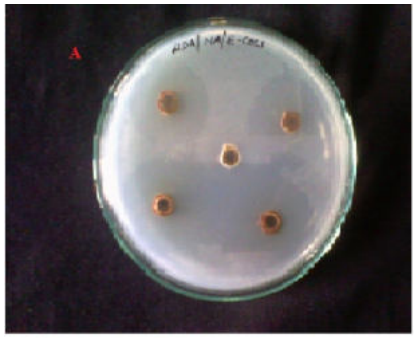
1mM aqueous silver nitrate solution was prepared and stored in brown bottles. 10 ml of herbal extracts was taken in conical flask separately and to this 90 ml of 1mM silver nitrate solution was added. The conical flask was incubated at room temperature. The color change from pale yellow to dark brown was checked periodically. The change in colour visually indicates the formation of Silver Nanoparticles which was used for monitoring the reduction process using UV-Visible Spectroscopy.

Optimization and Production of Silver Nanoparticles by Sunlight Irradiation method

Silver nanoparticles were synthesized by exposing the reaction mixture containing plant extract and silver nitrate (1mM) in the ratio 1:9(w/v) to sunlight for different time intervals (5, 10, 15 minutes). This optimisation leads to the reduction reaction in the mixture. The reduction of pure silver ions were monitored by UV-Visible spectrum of the reduction media. The reaction mixture was kept for incubation throughout the night. It was then centrifuged at 8000 rpm for 20min to recover the silver nanoparticles. Bulk production of the silver nanoparticles was carried out from the optimized time.

3.3.1.1 Antibacterial activity

The comparative antibacterial activities of the plant leaf extracts and of the Ag NPs synthesized from the respective extracts were effectively accessed against one Gram (+)ve (Bacillus) bacteria and one Gram (-)ve (Escherichia coli (E. coli)) bacteria as test microorganisms. Well diffusion method was followed for testing each type of plant leaf extract and their respective Ag NPs containing solution. Nutrient agar was prepared, sterilized and poured in the sterile Petri dishes and allowed to solidify. 24 h growing bacterial cultures E.coli were swabbed on it. Then, 5 wells (8mm diameter) were made by using a sterile cork borer. The different concentrations of sample were loaded in the wells. Standard used was Tetracyclin (10mg/ml). The plates were then incubated at 37ºC for 24h. After incubation the inhibition diameter was measured.



A) Escheria Coli

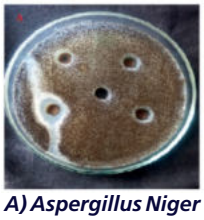
B) Staphylococcus Aureus

C) Klebsiella Pneumonia

Fig 8. Antibacterial Activity of silver nanoparticle

3.3.1.2 Antifungal activity

Potato dextrose agar was prepared, sterilized and poured in the sterile Petri dishes and allowed to solidify. 24 h growing Aspergillus niger and Aspergillus flavus species of fungi cultures were swabbed on it. Then, 5 wells (8mm diameter) were made by using a sterile cork borer. The different concentrations of sample were loaded in the wells. Citric acid solution (10mg/ml) water served as control. Standard used was Fluconazole (10mg/ml). The plates were then incubated at 37°C for 24h. After incubation the inhibition diameter was measured.



A) Aspergillus Niger

B) Aspergillus Flavus

Fig 9. Antifungal Activity of silver nanoparticle

RESULT

Green synthesis of silver nanoparticles by the help of green plants is a very cost effective, safe, non-toxic, eco-friendly route of synthesis which can be manufactured at a large scale.

UV Absorption Peak SEM Analysis of silver nanoparticles

Giovotia Rottleriformis showed great capability to synthesize silver nanoparticles at optimum temperature conditions. The UV absorption peak at 450nm clearly indicates the synthesis of silver nanoparticles.

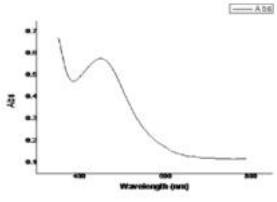


Fig 10. UV-Vis absorption spectrum of obtained silver nanoparticles

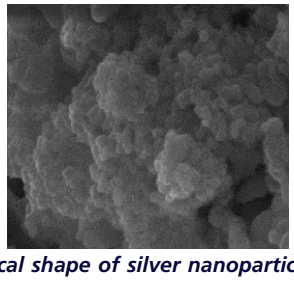


Fig. 11 Spherical shape of silver nanoparticles determined using SEM

The topography of silver nanoparticles was found using SEM. The shape of the silver nanoparticles was determined to be spherical in shape. The figure explains the same.

Antimicrobial Activity of Silver nanoparticles

Minimum Inhibitory Concentration (MIC):

In microbiology, minimum inhibitory concentration (MIC) is the lowest concentration of an antimicrobial that will inhibit the visible growth of a microorganism after overnight incubation. Minimum inhibitory concentrations are important in diagnostic laboratories to confirm resistance of microorganisms to an antimicrobial agent and also to monitor the activity of new antimicrobial agents. A MIC is generally regarded as the most basic laboratory measurement of the activity of an antimicrobial agent against an organism.

Clinically, the minimum inhibitory concentrations are used not only to determine the amount of antibiotic that the patient will receive but also the type of antibiotic used, which in turn lowers the opportunity for microbial resistance to specific antimicrobial agents. Applying MIC testing to a number of bacterial strains in the same species provides an estimate of the concentration that inhibits 50% (MIC_{50}) and 90% (MIC_{90}) of bacterial isolates and can indicate shifts in the susceptibility of bacterial populations to antibiotics. Currently, there are a few web-based, freely accessible MIC databases.

Zone of Inhibition:

The area without bacterial and fungal growth surrounding an antimicrobial-impregnated disk in an antimicrobial sensitivity test is its usual definition. So,the zone of inhibition determines the amount of anti- bacterial and anti- fungal activities of silver nanoparticles.

S. No.	Concentration of samples	Zone Of Inhibition		
		E.Coli	S. Aureus	K. Pneumonia
1.	25	29	25	27
2.	50	27	27	28
3.	75	29	25	27
4.	100	34	33	31

Table 1 : Antibacterial Screening

S. No.	Concentration of the sample	Zone Of Inhibition	
		Aspergillus Niger	Aspergillus Flavus
1.	25	10	11
2.	50	11	11
3.	75	12	12
4.	100	15	14

Table 2 : Antifungal Screening

CONCLUSION

The unique physical and chemical properties of silver nanoparticles only increase the efficacy of silver. Though there are many mechanisms attributed to the antimicrobial activity shown by silver nanoparticles, the actual and most reliable mechanism is not fully understood. Though bacterial, fungal, and plant extract sources can be used for nanosilver synthesis, the easy availability, the nontoxic nature, the various options available, and the advantage of quicker synthesis make plant extracts the best and an excellent choice for nanosilver synthesis. The uses of silver nanoparticles are varied and many, but the most exploited and desired aspect is their antimicrobial capacity and anti-inflammatory capacity. This has been utilized in various processes in the medical field and has hence been exploited well.

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