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RESEARCH ARTICLE

A Marvel on Aqueous Peel Extract (*Musa paradisiaca*): Free Radical Scavenging Activity, Biosynthesis of Silver Nanoparticles and Antibacterial Potential

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ABSTRACT:

Musa paradisiaca is high valued taxa having medical applications. The study aimed to synthesize and characterize the silver nanoparticles using aqueous peel extract of musa paradisiaca and their activity against virulent bacteria. The free radical scavenging activity of the aqueous peel extract was also carried and showed IC₅₀ value of 354μ g/ml. The silver nanoparticles were synthesized by the biological reduction of phytoconstituents present in the peel extract by the addition of silver nitrate. The silver nanoparticles were formed within an hour of incubation. The silver nanoparticles were characterized using UV-visible spectroscopy, Scanning electron microscopy, Fourier transform infra-red spectroscopy, X-ray diffraction. Antibacterial activity were done using well diffusion agar method. The absorption maxima was found in the range between 410 nm and 440 nm in UV-Vis spectroscopy respectively. SEM analysis revealed the size (30-100 nm) and diverse shape of the synthesized nanoparticles. The potential of synthesized silver nanoparticles against the bacterial species *Bacillus subtilis, Escherichia coli, Staphylococcus aureus* and *Pseudomonas aeruginosa* has showed maximum inhibition ranging from 1.2 ± 0.2 to 1.6 ± 0.2 cm. Hence, the study depicts the enormous capability of aqueous peel extract towards the synthesis of silver nanoparticle and was found with enhanced antibacterial potential against test micro organisms.

KEYWORDS: *Musa paradisiaca*, phytoconstituents, free radical scavenging, silver nanoparticles, antibacterial activity

INTRODUCTION:

Bananas are consumed all over the world, after consumption of the pulp; the peels are generally discarded (Bankar et al., $2010)^1$. On the basis of literature banana peels are enriched in polyphenols, hemicelluloses, lignin, pectin and antioxidant so this could be used for medicinal implications. (B. Manu et al., $2017)^2$. Bananas are high in antioxidants, which can provide protection from free radicals.

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They possess potassium, a mineral electrolyte that keeps electricity flowing through the body and pectin, a form of fiber. The low sodium content also helps protect cardiovascular system against high blood pressure according to FDA. Besides, it has high medicinal value of anti-diabetic (Dikshit et al., 2012)³, anti-inflammatory (Phuaklee et al., 2012)⁵, anti-ulcer (Goel et al., 2002)⁴ properties.

The nanotechnology is an emerging and well developing field of science where things at the nanoscale are originated. Nanoparticle research is inevitable because of its application and synthesis (Gopinath et al., $2012)^6$. Nanoparticles have unique electrical, optical as well as biological properties and are thus applied in catalysis,

biosensing, imaging, drug delivery, nanodevice fabrication and in medicine (Jain et al., 2008 and Nair, 2007)^{7.8}. The nanofabricated materials have high intensive properties and have the ability to enhance the features. Silver is one of the most commercialised nanomaterial and is estimated to increase its profound role in field of high sensitivity bimolecular detection, catalysis, biosensors and medicine (Shakeel Ahmed, 2016 et al)⁹. Silver nanoparticles can be synthesized using physical, chemical and biological methods.

In the recent years, green synthesis or plant mediated nanoparticles synthesis have become more advantageous over other biological processes because the elaborated process of culturing and maintaining the cells is eliminated and large-scaled nanoparticle synthesis is scaled up. There is an increasing interest in silver nanoparticles on account of the antimicrobial properties (Choi et al., 2008)¹⁰. Silver is regarded nontoxic, safe inorganic antibacterial agent that is capable of killing about 650 types of diseases causing microorganisms (Jeong et al., 2005)¹¹. Hence in the present study, we have investigated the free radical scavenging activity of the aqueous peel extract of Musa paradisiaca and using the same the biosynthesis of silver nanoparticles and its antibacterial activities were validated. The dimensions and morphology of the synthesized silver nanoparticles were also produced by means of Scanning electron microscopy, FT-IR and XRD analysis.

MATERIALS AND METHODS: Preparation of the extract:

The peels of Musa paradisiaca were collected and thoroughly washed three times using tap water and distilled water to remove the impurities present in it. It was shade dried for three days. About 25 g of peel was taken in a 250 ml beaker containing 50 ml double distilled water and then the peel was boiled at 60°C for 20 min and filtered through Whatman No. 1 filter paper twice remove insoluble fractions to and macromolecules. The filtrate obtained was the aqueous peel extract of Musa paradisiaca, it is stored at 4°C and can be used for further tests.

Free radical scavenging assay (DPPH):

The aqueous extract of the banana peel was taken in different concentrations of 100, 200, 300, 400 and 500μ g/ml respectively which was made upto 1ml using methanol. 1ml of 0.01 Mm DPPH dissolved in methanol was added to all the test concentrations and maintained in the dark for 30minutes, at room temperature. The absorbance was measured at 517nm against blank. Methanol was used as a blank, DPPH solution as control and butylated hydrox-anisole (BHA) as the reference (udayaprakash et al., 2014)¹². Antioxidant activity of each extract by DPPH method was determined by

calculating percentage of antioxidant activity using reduction of DPPH absorbance. IC50 of DPPH scavenging activity of each extract was calculated using the calibration curve.

Synthesis of silver nanoparticles:

The typical reaction mixture was setup by adding 1ml of aqueous peel extract to the test tube containing 9ml of distilled water. 20μ l of silver nitrate was dropped into the respective test tube and it was incubated in the dark room for 24 hours. The reaction between the silver nitrate and aqueous peel extract occurred within 5 minutes after incubation. It was indicated by the mixture turned into reddish brown colour.

Characterization of silver nanoparticles:

The complete reduction of silver nitrate to silver nanoparticles in the test tube was evidently proved by absorbance peaks produced by the double beam UV-Vis spectrophotometer. The characteristic external view was analysed using the Scanning electron microscope whereas the amino groups of peptides and functional groups present in the nanoparticles was observed in Fourier transform infrared spectroscopy. The crystalline structure of the nanosized particle was also confirmed by XRD.

Evaluation of antibacterial activities:

The minimum inhibitory concentration of the silver nanoparticles was tested at different concentrations (10, 20, 30, 40 and 50µg/ml) against four common pathogenic test organisms Staphylococcus aureus, Bacillus subtilis, Pseudomonas aeruginosa and Escherichia coli. The test was conducted by well diffusion agar method (Shanmuga et al.,2002)¹³ using gentamicin as control. The petri dishes containing nutrient agar medium were divided into eight parts and the well was diffused using t-rod. Each well was added with 30 µg/ml of aqueous peel extract, control and silver nitrate along with different concentrations of silver nanoparticles. The plates were incubated for 24 hours. The zones of inhibition in all the wells were noted after incubation.

RESULTS AND DISCUSSIONS:

Free radical scavenging assay (DPPH) for aqueous peel extract:

The percentage inhibition and IC_{50} values were calculated with DPPH as the control and butylated hydrox-anisole (BHA) as the reference (Table 1). The different concentration of aqueous peel extract inhibits the formation of DPPH radicals by 50% which is defined as the IC ₅₀ value. The IC₅₀ value of the solvent was found to be $354.59\mu g/ml$ (Fig 1).

 Table 1: The DPPH free radical scavenging assay of the aqueous extract of banana peel.

Concentration(µg/ml)	% Inhibition
100	12.22
200	24.44
300	43.33
400	52.22
500	75.56

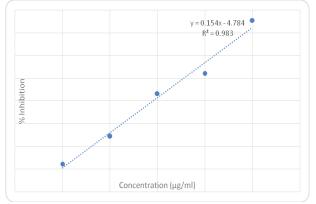


Fig 1: The graphical representation of IC50 value of aqueous extract of banana peel.

Synthesis of silver nanoparticles using banana peel extract:

The biosynthesis of Silver nanoparticles occurred due to the reduction of silver ions in silver nitrate by the aqueous peel extract that are enriched in active compounds like polyphenols (Ahmed M. Aboul-Enein et al., 2016)¹⁴. After adding silver nitrate the mixture immediately turned from colourless mixture to reddish dark brown colour indicating the formation of silver nanoparticles.

Characterization of Silver nanoparticles: UV-Vis spectroscopy:

The presence of silver nanoparticles were indicated using the maximum absorbance peaks in the surface plasmon resonance of double beam UV-Vis spectrophotometer. The formation and stability of the reduced silver nanoparticles in the colloidal solution were monitored. The absorption maxima of silver nanoparticle lies exactly in the range of 410-440nm (Fig 2a).

Scanning electron microscopy:

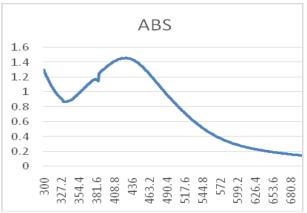
The size and morphology of silver nanoparticles was studied using SEM. The size of the nanoparticles ranged between 30-100nm (Fig 2b). The silver nanoparticles were spherical in shape and crystalline in structure.

FTIR analysis:

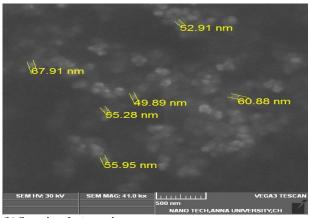
The FTIR analysis produced a set of peaks that confirms the presence of silver nanoparticles in the sample (Fig 2d). The analysis reveals the presence of proteins, phytoconstituents and carbohydrates which are biostabilizers that caps the silver nanoparticles.

XRD analysis:

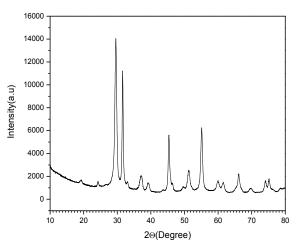
The XRD analysis was carried to determine the crystalline nature of the silver nanoparticles. The peaks in the XRD pattern represents some biochemical compounds like proteins present in aqueous peel extract (Fig 2c).



(a) UV-VIS spectrophotometer



(b) Scanning electron microscopy



^{IIS,} (c) X-ray diffraction

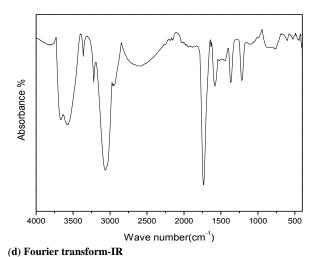


Fig 2: Silver nanoparticles characterization were done using a) UV-VIS spectrophotometer b) Scanning electron microscopy c) X-

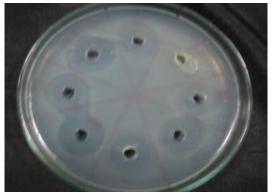
ray diffraction d) Fourier transform-IR

Antibacterial activities:

The antibacterial activity was studied by performing minimum inhibitory concentration against the pathogenic microbes *Staphylococcus aureus*, *Bacillus subtilis*, *Pseudomonas aeruginosa* and *Escherichia coli* (Fig 3). Among the four organisms, gram positive *Staphylococcus aureus* and gram negative *Pseudomonas aeruginosa* showed good results of inhibitions ranging from 1.2 ± 0.2 to 1.6 ± 0.2 cm when compared to aqueous peel extract, control and silver nitrate in their respective wells.(Table 2).

Table 2: The minimum inhibi	itory concentrations of the silver nanoparticles of banana peel against the above mentioned microorganisms.	
Organism	Zone of inhibition (in cm)	

Organism	Zone of inhibition (in cm)								
	Control	10µ1	20 µl	30 µl	40 µ1	50 µl	AgNO3	Plant extract	
Staphylococcus aureus	1.8	1.6	1.8	1.7	1.8	1.7	1.9	1.3	
Bacillus subtilis	1.9	-	-	-	-	-	0.7	0.6	
Pseudomonas aeruginosa	2	1	1.2	1.4	1.6	1.7	0.6	-	
Escherichia coli	1.9	-	-	-	-	-	0.8	-	



(a) Pseudomonas aeruginosa



(b) Staphylococcus aureus







(d)Bacillus subtilis Fig 3: The antibacterial activity of silver nanoparticles against a) Pseudomonas aeruginosa b) Staphylococcus aureus c) Escherichia coli d) Bacillus subtilis

CONCLUSION:

Among the different tests for antioxidants the free radical scavenging assay plays a vital role in detecting the antioxidant activity of aqueous peel extract of musa paradisiaca as it aids in investigating the drug action role by finding the inhibitory concentration. The silver nanoparticles were synthesized biologically using aqueous peel extract of Musa paradisiaca in a very ecofriendly and cost effective method. These nanoparticles were characterized by the UV-Vis spectrophotometer and its morphology were discovered from SEM, FTIR and XRD results. The silver nanoparticles had great efficiency towards the gram positive and gram negative pathogenic bacteria at various concentrations that helps to suggest the medical implication as an antibacterial agent. Thus, a eco-friendly, green method was adopted for the development of potential silver nanoparticles using the natural plant species which is accessible in all moist areas.

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CONFLICT OF INTEREST:

We declare no conflict of interest.

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