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Economical biosynthesis of silver nanoparticles using fruit waste

Anil R Shet*, Shwetha Tantri[2], Arvind Bennal[3]
Department of Biotechnology, B. V. Bhoomaraddi College of engineering and technology, Vidyanagar, Hubli, Karnataka
[3]Department of Physics, B. V. Bhoomaraddi College of engineering and technology, Vidyanagar, Hubli, Karnataka
***Corresponding Author Email: anil_shet@bvb.edu**

ABSTRACT

Silver nanoparticles (Ag-NPs or nanosilver) have proven to be of great interest because of their special properties. Synthesis of these silver nanoparticles by the process of chemical reduction is a known widely. This method involves using of chemicals that are toxic by nature for the synthesis, which may adversely affect the medical applications of these AgNPs and also have perilous effect on the environment. Hence, Green synthesis method which is eco-friendly and economic has been undertaken in this study. Peel extract of Banana and Orange were used as capping agent to reduce 1mM silver nitrate solution to silver nanoparticles. The biosynthesized AgNPs were characterized by UV- Vis spectroscopy, SEM and FT - IR. The anti-bacterial activity of the biosynthesized AgNPs was tested against pathogenic bacterial strains of *Staphylococcus aureus* and *Proteus vulgaris*. In case of Banana peel extract (BPE), peel concentration of 6% and pH 8 and, similarly, for Orange peel extract (OPE), peel concentration of 6% and pH 11 was observed to be effective for the synthesis of AgNPs. Temperature of 60ºC had a positive effect on the biosynthesized AgNPs in case of both the fruit peels. The fruit peel biosynthesized AgNPs are effective against pathogenic strains of bacteria. The antibacterial effect of Banana peel synthesized AgNPs was maximum for both the bacterial strains.

KEYWORDS: Silver nanoparticles; UV- Vis Spectroscopy; FTIR; SEM; Antibacterial activity.

1. INTRODUCTION

Silver nanoparticles are very useful in the present environment because of the distinctive properties which includes its shape, size, electrical, magnetic and optical properties. Thus, these AgNPs are widely exploited for various applications, including; biosensor materials, composite fibers, cosmetic products, electronic components and antimicrobial applications. (Korbekandi, 2012). The process of chemical reduction is a widely known process used to synthesize Ag-NPs because of its willingness to generate Ag-NPs under clement conditions and its ability for large scale synthesis. However, this method of chemical synthesis involves the using of chemicals that are toxic in nature in and have adverse effect on the environment and hinder the application of the biosynthesized AgNPs in the medical fields. Therefore, synthesis of Ag-NPs by green synthesis approach has proven to be more advantageous over the chemical and physical methods as it is environmentally benign, economical and most importantly it does not require adverse environment of high temperature, pressure, energy or the use of toxic chemicals (Shameli, 2012). Hence, this biological method has been employed for the silver nanoparticles. Biological synthetic methods show that the silver nanoparticles synthesized from the plant source are more stable than the AgNPs synthesized from microbial sources. Thus, biodegradable fruit peel waste extracts have been used as capping and stabilizing agents for synthesizing silver nanoparticles from silver ions.

Silver nanoparticles have shown remarkable repressive effects against microorganisms, and are extensively used as antimicrobial agents in a wide range of consumer products. In this present work, silver nanoparticles were synthesized using Banana and Orange peel extracts as capping agents and their antimicrobial effect against pathogenic bacterial strains was measured. The effect of parameters like; pH, temperature and peel extract concentration on the synthesis of AgNPs was also studied.

2. MATERIALS AND METHODS

Preparation of Fruit Peel Extract: The fruits used for biosynthesis of AgNPs (silver nanoparticles) were brought from the household wastes and washed thoroughly with water. The peel of the fruits (Banana and Orange) was then air dried on paper towel. 25gms of the dried fruit peel was added to 50ml of distilled water and boiled at 80ºC for 10mins. The peel extract was then filtered using a filter paper and stored at 4ºC for further use.

Biosynthesis of silver nanoparticles: The peel extract obtained was added to 20ml of 1mM silver nitrate ($AgNO_3$) procured from SRL solutions, and the reaction mixture was left to react for duration of 1 hour. Then, the solution containing synthesized silver nanoparticles was analyzed using a UV 3000 $^+$ UV/ Vis Spectrophotometer made by Lab India, within a wavelength range of 300 - 600nm. The biosynthesis of the AgNPs was undertaken by altering parameters like pH, temperature and peel extracts concentration and their impact on the nanoparticles formed was studied. The samples containing biosynthesized AgNPs were also characterized by FT – IR and SEM methods.

Effect of various parameters on the Biosynthesis of AgNPs:

i) Peel extract concentration: The effect of different concentrations, 2%, 4% and 6% of peel extracts (Banana and Orange) on the biosynthesis of silver nanoparticles was studied.

ii) pH: Different pH (5, 8 and 11) range was used to study its effect on the biosynthesis of silver nanoparticles.

iii) Temperature: Effect of different temperatures on the production of silver nanoparticles was observed at Room

temperature and at 60°C.

Antibacterial Activity of silver nanoparticles: One day old culture of *Staphylococcus aureus* and *Proteus vulgaris* obtained from biotechnology department was spread on pre-incubated nutrient agar plates. Three wells were punched on each of the plates. Streptomycin (100µg/ml) was treated as the positive control; Fruit peel extract as negative control and the biosynthesized nanoparticles solution as test. 20µl of the solutions was added to the previously punched wells. The inoculated plates were incubated for 24 hours and the zone formed around the well was measured.

3. RESULTS

Color change:

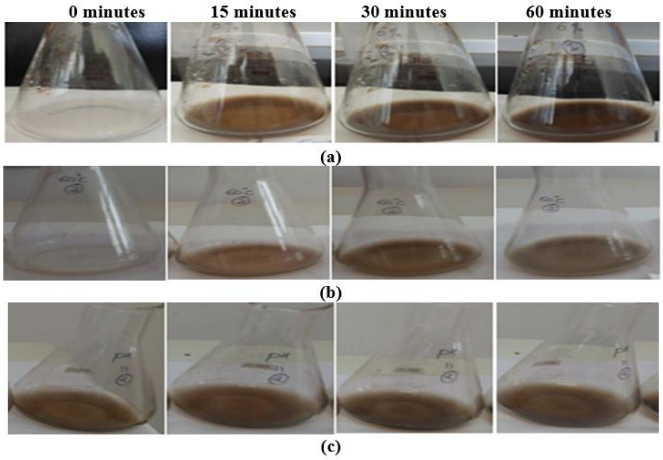


Figure.1.Effect of parameters (a) peel concentration (b) temperature (c) pH on the biosynthesis of AgNPs using Banana peel extract as reducing agent for the duration of one hour

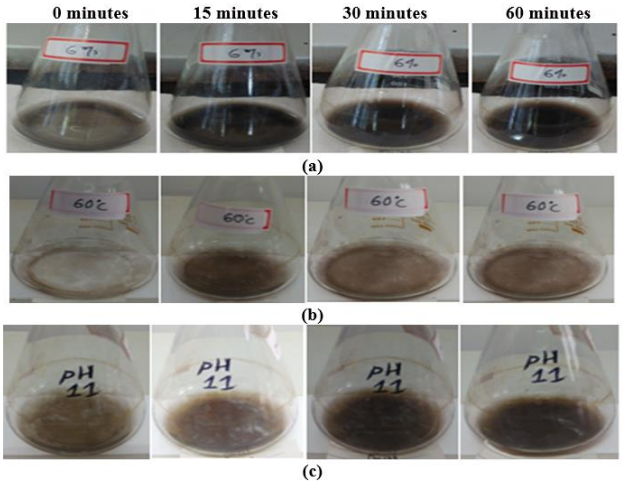


Figure.2.Effect of parameters (a) peel concentration (b) temperature (c) pH on the biosynthesis of AgNPs using Orange peel extract as reducing agent for the duration of one hour

UV- Visible Spectroscopy Analysis:

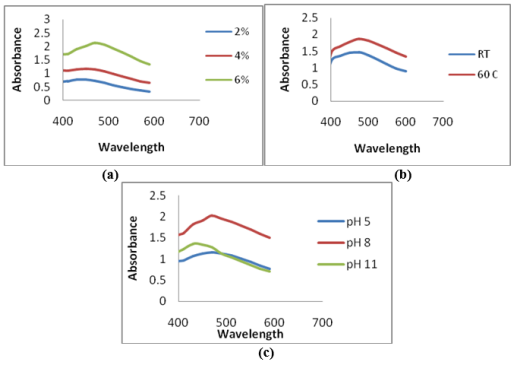


Figure.3.Shows the UV visible Spectra indicating the effect of (a) peel concentration (b) temperature (c) pH on the biosynthesis of AgNPs using Banana peel extract as reducing agent

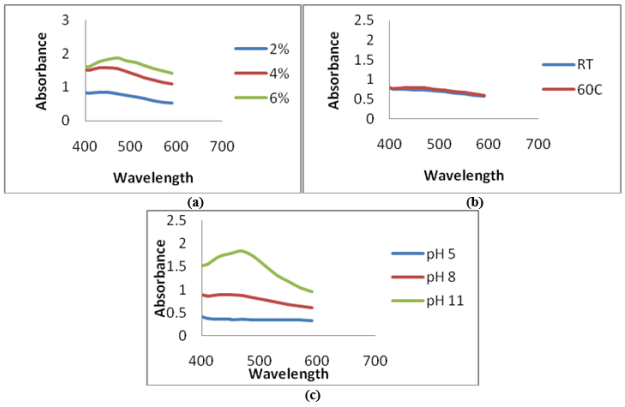


Figure.4.Shows the UV visible Spectra indicating the effect of (a) peel concentration (b) temperature (c) pH on the biosynthesis of AgNPs using Orange peel extract as reducing agent.

FT – IR Analysis:

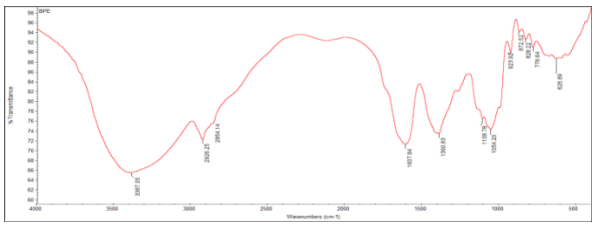


Figure.5.Represents the Infrared spectra of the Banana peel extract used for the biosynthesis of AgNPs.

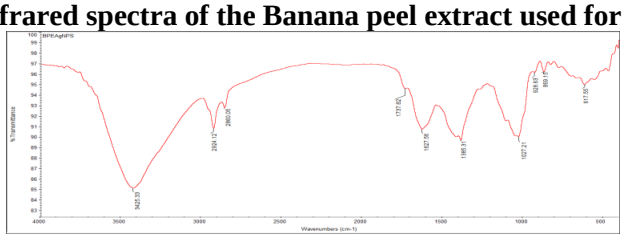


Figure.6.Represents the Infrared spectra of the silver nanoparticles synthesized using Banana peel extract and 1mM silver nitrate solution in the duration of 1hour

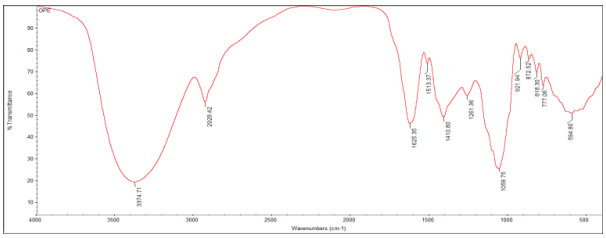


Figure.7.Represents the Infrared spectra of the Orange peel extract used for the biosynthesis of AgNPs

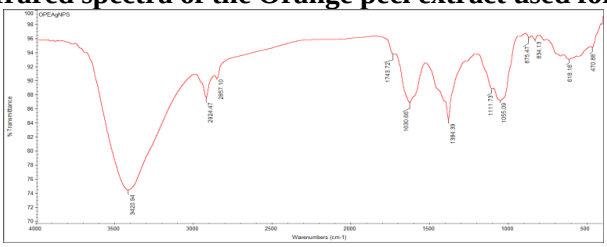
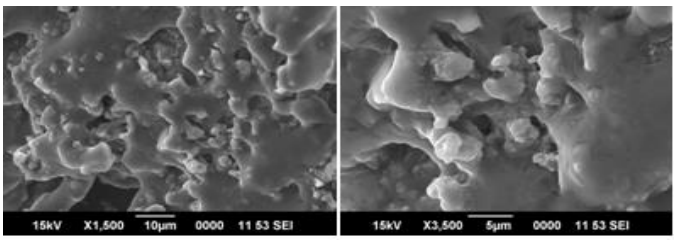
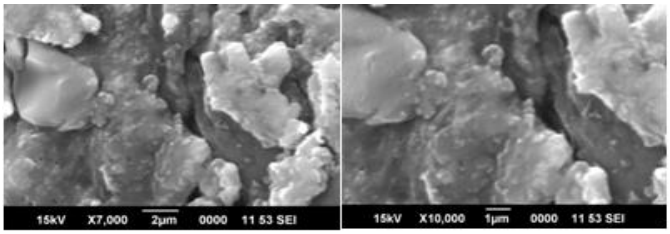


Figure.8.Represents the Infrared spectra of the silver nanoparticles synthesized using Orange peel extract and 1mM silver nitrate solution in the duration of 1 hour

SEM Analysis:

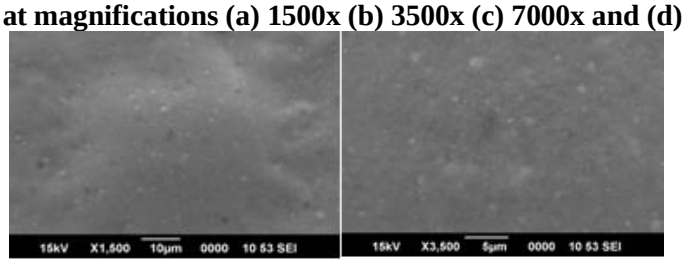


(a) (b)

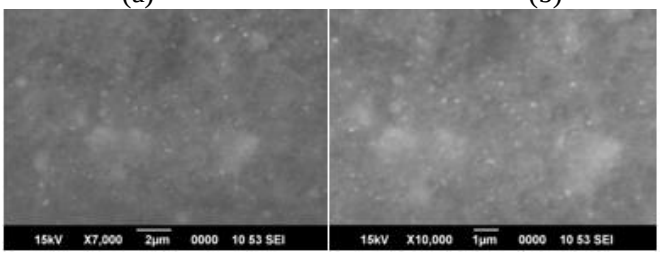


(c) (d)

Figure.9.SEM images of banana peel extract synthesized silver nanoparticles using 1mM silver nitrate solution at magnifications (a) 1500x (b) 3500x (c) 7000x and (d) 10000x



(a) (b)



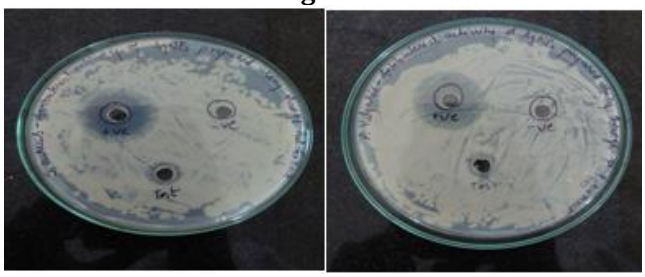
(c) (d)

Figure.10.SEM images of Orange peel extract synthesized silver nanoparticles using 1mM silver nitrate solution at magnifications (a) 1500x (b) 3500x (c) 7000x and (d) 10000x

Antibacterial Activity:

(a) (b)

Figure.11.(a) and (b) represent the antibacterial activity of Banana peel synthesized silver nanoparticles on *Staphylococcus aureus and Proteus vulgaris* respectively. In the figure +ve indicates positive control (0.1µg/ml Steptomycin), -ve indicates Banana peel extract and Test indicates Banana peel synthesized AgNPs



(c) (d)

Figure.12.(c) and (d) represent the antibacterial activity of Orange peel synthesized silver nanoparticles on *Staphylococcus aureus* and *Proteus vulgaris* respectively. In the figure +ve indicates positive control (0.1µg/ml Steptomycin), -ve indicates Orange peel extract and Test indicates Orange peel synthesized AgNPs

Table.1.Indicates the zone of inhibition of the biosynthesized silver nanoparticles using fruit peel extract

Organism	Diameter of the inhibited zone obtained in mm		
	Streptomycin (0.1mg/ml)	Banana peel extract synthesized AgNPs	Orange peel extract synthesized AgNPs
Staphylococcus aureus	17	8	7
Proteus vulgaris	17	10	5

DISSCUSSION

The fruit peel extracts of banana and orange were chosen to react with 1mM of $AgNO_3$ at different peel concentration, temperature and pH. In all the cases there was a color change, which demonstrated the generation of metal nanoparticles. The above figures 1 and 2 showed that there was a progressive change in the color from yellow to reddish yellow and from reddish yellow to dark brown for mixtures containing the solution of 1mM silver nitrate and banana and orange peel extracts respectively indicating the formation of AgNPs. These characteristic color variations are because of the metal nanoparticles (Shet, 2015). The intensity of the color was observed to increase with the incubation duration which denoted the formation of more amounts of nanoparticles. Orange peel extract transformed the colorless $AgNO_3$ solution to black in minimal time. It is the fastest reaction, which may be owing to its high Vitamin C content. The Vitamin C content of orange peel has antioxidant property, which acts as reducing agent and facilitates the reduction of Ag+ of $AgNO_3$ to Ag^o. The intensity of color changed with the change in peel extract concentration, temperature and pH.

UV-VIS spectroscopy results established the formation of silver nanoparticles by biological reduction (green synthesis) method using fruit peel extracts (orange, Banana). UV-VIS measurements showed that the surface plasmon band of silver nanoparticles solution was observed to be specifically between 400-480nm throughout the observation period which showed the generation of nanoparticles. The wavelength did not change much with the parameter change, but there was a variation in the absorption. The increase in absorption indicates formation of numerous nanoparticles. As the diameter of the particle enlarges, the plasmon resonance peak varies to longer wavelengths and broadens (Phuphansri, 2012). From the figures 3 (a), (b) and (c) it can be inferred that for the Banana peel extract, 6% peel extract has maximum effect whereas 2% has minimum effect on the biosynthesis of AgNPs respectively. Similarly, pH of 8 and temperature of 60°C has maximum effect on the synthesis of AgNPs. From figures 4 (a), (b) and (c) it can be inferred that 6% and 2% of Orange peel favored maximum and minimum biosynthesis of silver nanoparticles. Similarly, pH of 11 and temperature of 60°C is observed to be favorable for the generation of AgNPs.

To identify the major functional groups in fruit peels that have possible involvement in the formation of silver nanoparticles and its stabilization, the FT-IR measurements were done. The intricate nature of biological material will be indicated by the several peaks, representing various functional groups. The shift in the following peaks (wave number) at 3387, 2926, 1607, 1390, 1054, 828, 629 in BPE and peaks at 3374, 1625, 1410, 1058, 872 and 470 in OPE indicates the participation in the process of nanoparticles synthesis (Bankar, 2012). Banana and orange peel are composed of mainly complex carbohydrates like and hemicelluloses, cellulose and pectin (Emaga, 2007) and their functional groups and also the associated polymers and proteinaceous matter which may be involved in reducing the Ag^+ to Ag^o. These biological compounds interact with the metal salts through their functional groups and facilitate their reduction to particles of nanoscale. (Bar, 2009).

SEM analysis shows AgNPs synthesized by peel extracts of banana and orange (Figure 9 and 10). The SEM results confirmed the presence of nanoparticles. In figure 9, silver particles of large diameters are observed which may be the result of clustering of smaller silver particles. It is observed that the nanoparticles are not directly in contact with the aggregates, indicating that the nanoparticles are stabilized by a reducing agent. From the above figure 10, it is observed that all the particles show almost spherical morphology.

Antibacterial activity of the biosynthesized AgNPs was determined qualitatively by testing the zone of inhibition and the level of inhibition was estimated by using differently prepared colloidal silver nanoparticles. The anti-bacterial activity may because of the decrease in the size of the particle formed, which can cause the increase in the distinct outer surface of a the specimen, enhancing their ability to infiltrate the cell membrane and induce its antibacterial action (Padma, 2012).The bacterial activity is assumed to be due to the changes caused in the membranal structure of microbial cell because of its interaction with the rooted AgNPs which leads to the increase the permeability of the cell membrane and consequently, leading to their death (Awad, 2014).From the above images, the zone of clearance obtained is observed to be maximum for AgNPs synthesized from peel extract of banana for both the bacteria, *S.aureus* and *P.vulgaris*

4. CONCLUSION

A crucial requirement in the field of nanotechnology is for the development of dependable and biodegradable processes for the generation of metallic nanoparticles. The parameters considered for the studies; peel extract concentration, temperature and pH showed a positive effect on the synthesis of metal nanoparticles. FTIR and SEM

results confirmed the presence of nanoparticles. Banana peel synthesized silver nanoparticles were observed to be more effective in inhibiting both the bacterial strains when compared to the orange peel extract synthesized AgNPs. The process of synthesizing the metal nanoparticles by using a biological process has proved to be of great significance because of its myriad advantages over the flaws and drawbacks of the physiochemical processes.

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