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Biosynthesis, characterization, and application of silver nanoparticles in sustainable plastic

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A R T I C L E I N F O

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A B S T R A C T

The synthesis of silver nanoparticles (AgNP) by microorganisms is an area of growing interest in nanobiotechnology since these nanoparticles are used for biomedical, clinical, and biological imaging, as well as for various products, including cosmetics and biosensors. This study covers the production of colloidal silver nanoparticles through bacterial synthesis. Colloidal silver nanoparticles are characterized by ultraviolet (UV-Vis) spectroscopy, infrared Fourier transform (FTIR), scanning electron microscopy (SEM), and zeta potential. The subsequent particles are sphere-shaped and have an average particle size of 33 to 37 nm. The resolution data show that the emission of silver nanoparticles is inversely proportional to the size of the nanoparticles. The smaller the particles, the greater the emission

K E Y W O R D S

Bacillus spp. | Silver nanoparticles | Biosynthesis | Characterization

C I T A T I O N

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Introduction

Metallic nanoparticles are very important because of their unique electronic, optical, and physic°Chemical properties, which are very different from bulk materials (Mazur M, 2004.). Various methods of physic°Chemical synthesis for the production of metal nanoparticles have been described (Raut *et al.,* 2009). Nevertheless, this pr°Cess has some drawbacks, such as the use of toxic chemicals and the production of hazardous waste. The need to develop environmentally friendly pr°Cesses for the synthesis of nanoparticles is growing (Whitesides, 2003). Microorganisms have the ability to reduce metals in metal ions and play an important role in bioremediation (Fortin 2000). In addition, these small nanostructures can synthesize nanoparticles in an environmentally friendly way. Many metals, gold, and cadmium nanoparticles are synthesized by various microorganisms. Bacteria are environmentally friendly and relatively easy to handle, which makes them the most commonly used microorganism (Parikh *et al.,* 2008).

Traditionally, physical and chemical methods are used for the synthesis of nanoparticles (Huang and Yang, 2005; Mandal *et al.,* 2005). In general, physical methods have a negative impact on the environment, since the yields are low, and chemical methods use toxic solvents and form harmful products (Wang *et al.,* 2007). Currently, scientists are f°Cused on the biosynthesis of nanoparticles using bacteria (He *et al.,* 2005), fungi (Bhainsa and D'Souza, 2016), and plants (Krishnaraj *et al.,* 2010). This biopr°Cess is inexpensive, profitable, safe, and environmentally friendly in comparison with physic°Chemical synthesis pr°Cesses (Emeka *et al.,* 2014).

The properties and behavior of materials to varying degrees compared with the nanoscale or micro level. Therefore, many studies are devoted to precious metal nanoparticles due to their unique properties other than bulk materials. This property depends on differences in the size, shape, and environment of nanoparticles (Piriyawong *et al.,* 2012).

Nanotechnology involves the synthesis and use of nanoparticles (Duran *et al.,* 2005). Silver nanoparticles are used in many applications as antibacterial agents in disinfection devices, cosmetics, household appliances, and sewage treatment plants (Leaper, 2006; Chopra, 2007). Inorganic composite materials are used as preservatives in various products (Duran *et al.,* 2005). Silver nanoparticles were obtained by physical, chemical, and biological methods (R°Co, 2005). Physic°Chemical methods are very expensive (Li *et al.,* 1999). Biological pr°Cesses for the synthesis of nanoparticles eliminate harsh pr°Cessing conditions and provide a low cost of synthesis at physiological pH, temperature, pressure, and at the same time. A comparison of various synthetic methods emphasizes biological synthesis (Vithiya and Sen, 2011). The biosynthesis of nanoparticles is environmentally friendly and has significant advantages compared to other pr°Cesses (Gade *et al.,* 2008; Mukherjee *et al.,* 2008). Microbial synthesis of nanoparticles can be intracellular or extracellular (Ahmad *et al.,* 2003; Mokhtari *et al.,* 2009; Saravanan *et al.,* 2011; Shahverdi *et al.,* 2007; Sharma *et al.,* 2009).

Nanoparticles synthesized in cells undergo additional subsequent pr°Cessing steps, such as sonication and reaction with a suitable detergent to release the nanoparticles from the cells during washing (Kalimuthu *et al.,* 2008). The synthesis of nanoparticles by extracellular pr°Cesses with simpler subsequent pr°Cessing is more economical than intracellular synthesis. Against this background, this study is devoted to the extracellular synthesis of nanoparticles from bacterial sources.

As we know, micro-organisms are all over the place; for example, in paper and the paper items that are generally utilized in our regular daily existence, from certified receipts to papers, books, and bundling paper. The propagation of this sort

of material can add to the destruction and spreading of irresistible sicknesses. Mulling over this, coordination of antimicrobial measure into cellulose by the consolidation of silver nanoparticles was established.

Finally, the authors demonstrated the successful incorporation of the AgNP-decorated cellulose film. The strategy consisted of the use of Cellulose and AgNPs prepared following a biogenic synthesis mediated by bacteria This is a novel eco-accommodating pr°Cedure to acquire AgNPs in which there is no requirement for high temperatures, high weights, and the creation of poisonous synthetic compounds.

Materials and Methods

Materials: All media components and analytical reagents were developed by Hi-Media Laboratories Pvt Ltd. Related. (India). For the synthesis of silver nanoparticles of Bacillus spp. received from the Department of Biotechnology at Junagadh Agricultural University (JAU) in Junagadh, Gujarat, India.

Methods: - Biosynthesis of Ag nanostructure by $AgNO_3$

The pure culture of Bacillus spp. was freshly in°Culated into on a liquid media (N broth (Hi-Media)) in the flask. The flask containing medium was incubated in an orbitary shaker at 150 rpm at 37 °C for 48 h. After 48 h of growth, the culture-containing medium was centrifuged at 10,000 rpm for 10 min. The supernatant was discarded, and the pellet was re transferred into a sterile flask.

The pellets were re-suspended in 10 ml ethanol/water (75:25, v/v), followed by vigorous mixing.

Then the sample was incubated at 85-90°C for 10 min, and subsequent incubation was done at 4°C for 10 min. Now, the supernatants were collected after centrifugation and were used for the synthesis of silver nanoparticles. The supernatant of strain was added to the flasks containing 1mM silver nitrate solution. The reaction between these supernatants and silver nitrate solution was carried out for 48 h. The bio-reduction of silver nitrate in the solution was monitored by the color change of the solution.

Fresh culture of Bacillus spp. was in°Culated in liquid medium of N-broth (Hi-Media). The flask containing medium was incubated at 37 ° C for 48 hours at 150 rpm on an orbital shaker. After 48 hours of growth, the culture medium was centrifuged at 10,000 rpm for 10 minutes. The supernatant was removed, and the pellet was transferred back to a sterile flask. The precipitate was resuspended in 10 ml of an ethanol / water mixture (75:25 v / v) and vigorously stirred.

Then the samples were incubated at 85-90 ° C for 10 minutes, followed by subsequent incubation at 4 ° C for 10 minutes. After centrifugation, the supernatant is collected and used for the synthesis of silver nanoparticles. The supernatant strain was placed in a flask containing 1 mm solution of silver nitrate. The reaction between the supernatant and the silver nitrate solution was carried out for 48 hours. The bio-reduction of silver nitrate in the solution was monitored by the color change of the solution.

Characterization of AgNPs

After 48 hours of incubation, silver nanoparticles were visually observed for a color change from colorless to brown to confirm the synthesized silver nanoparticles. AgNP samples are stored in crimped vials for further characterization.

Spectroscopic analysis of ultraviolet absorption: The sample used for analysis was diluted with 1 ml of Milli-Q water and then periodically measured in the UV-Vis spectrum. The use of quartz cells. Ultraviolet spectroscopic analysis of silver nanoparticles was carried out using Agilent technology, a Cary 60 ultraviolet spectrophotometer with a scanning speed of 300-700 nm.

Particle size analyser with Zeta potential: Zeta potential is a physical possessions that is given the net surface charge of the nanoparticles when these particles inside the solution are repelling each

other since produced Coulomb explosion between the charges of the nanoparticles giving rise to no tendency for the particles to agglomerate. The criteria of stability of NPs are measured when the values of zeta potential ranged from higher than +30 mV to lower than -30 mV (28). Surface zeta potentials were measured using the laser zeta meter Nanotracwave S3500.

The particle size distribution and zeta potential of biosynthesized silver nanoparticles were performed on the basis of Dynamic Light Scattering (DLS) with Nanotracwave (S3500) at room temperature. This technique gains more attention due to rapid analysis and minimum sample requirement (1 ml).

SEM analysis of silver nanoparticles: Scanning electron microscopy (SEM) was performed using a SEM machine (Zeiss model EVO-18). The thin film sample was made from a layer of aluminum foil, the sample was removed from the scars, the additional solution was removed with baking paper, and the SEM film was dried in hot air. Oven 15 kV for 3 hours at 37 ° C.

FTIR analysis of silver nanoparticles: The interaction between the protein and silver nanoparticles was analyzed using infrared analysis with Fourier transform (FTIR). Samples of silver nanoparticles were dried at 37 ° C and mixed with potassium bromide powder (KBr) and a Fourier transform infrared spectrometer (Shimadzu, 8400S) in the wavelength range of 3500-500 cm; ^{1} analysis.

Incorporation of AgNPs into cellulose based film: Cellulose was extracted from sugarcane baggase by chemical method. Extracted cellulose was dissolved in suitable solvents with a mixture of AgNPs. This silver nanoparticles embedded cellulose based film was characterized by SEM.

Results and Discussion

Primary confirmation: Visual confirmation: The very first characterization of synthesised silver nanostructures was done by observing the colour change from yellow to brown. This can be called a primary confirmation of the formation of silver nanoparticles

Fig. 1: Visual observation of Synthesis of silver nanoparticles (A) AgNO3 solution, (B) Bacterial extract, (C) synthesized silver nanoparticles

UV-Spectrophotometer: The synthesized silver nanoparticles can be traced by a UV-visible spectrophotometer. The UV absorbance v/s wavelength graph indicates that the absorbance peak. The absorption spectra of the silver nanoparticles were taken between 300 and 700 nm using Cary 60 UV–vis spectrophotometer. The MilliQ water was used as the blank. The highest peak of absorbance reported was around 0.86 and wavelength at 440 nm.

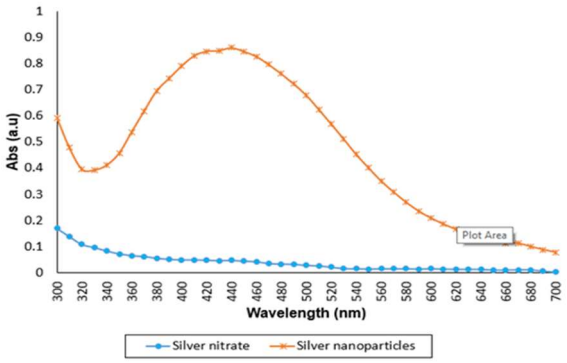


Fig. 2: UV-Visible absorption spectra of silver nanoparticles produced by Bacillus spp. after 48 h

The above data imply that the enzyme nitrate reductase liberated into the cell filtrate solution after the adding of AgNO3. Then the Ag ions are reduced to Ag nanostructures by the act of enzymatic reduction, thus bringing extracellular synthesis of silver nanoparticles.

FTIR analysis: FT-IR estimations were directed to uncover the conceivable potential biomolecules that took part in the bioreduction of silver and adjustment of AgNPs.

Fig. 3 illustrated distinctive broad spectral bands at around 3736–3890 cm−1 are characteristic of the O-H extending vibration sort of hydroxyl useful gathering in polyphenols and N-H extending vibrations in essential and auxiliary amines of amino acids, peptides and proteins. FT-IR information exhibited that the amide linkage of the protein had the higher potential to join silver and subsequently framing protein covering around AgNPs to forestall agglomeration and along these lines balance out the medium.

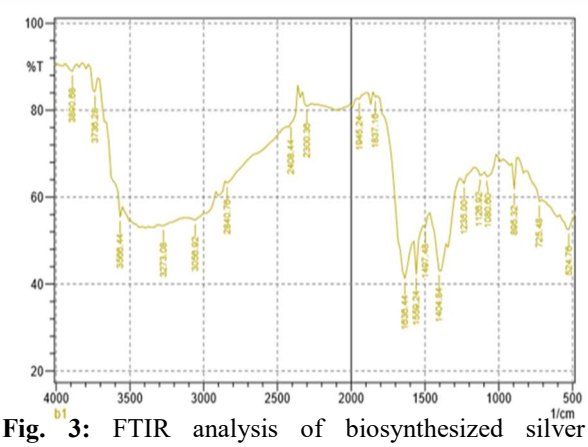


Fig. 3: FTIR analysis of biosynthesized silver nanoparticles from Bacillus spp.after 48 h

SEM Analysis: The biosynthesized Ag NPs are then subjected to centrifuge to obtain a pellet to obtain SEM results image clearly shows silver nanoparticles of size range 33 - 37 nm as showen in Fig. 4.

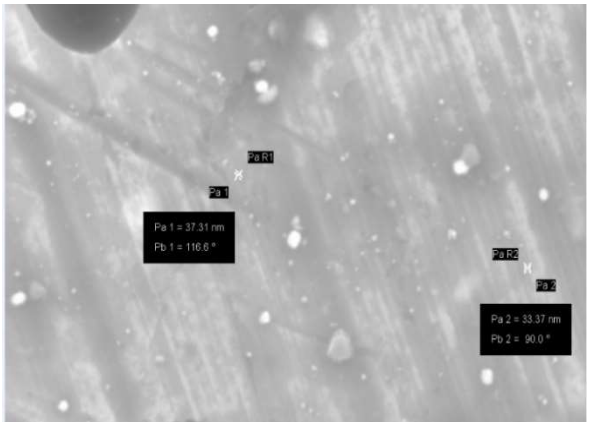
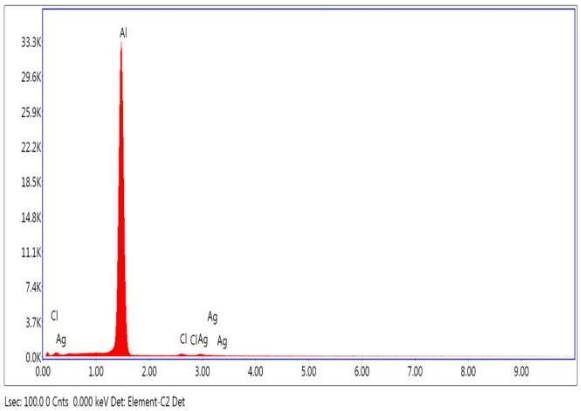


Fig. 4: SEM-EDAX image of biosynthesized silver nanoparticles from Bacillus spp. after 48 h



Particle size analysis and Zeta potential: The particle size analysis showed that the AgNPs synthesized in the bioreduction pr°Cess using bacterial spp. were extensively distributed in the solution. The particle size of the AgNPs was approximately 21 nm, and the zeta potential of the nanoparticles was 0.2 mV, and it showed good stability (Fig. 5).

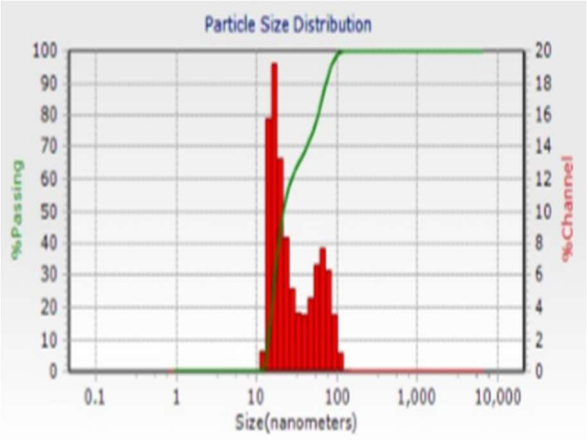


Fig. 5: PSD image of biosynthesized silver nanoparticles from Bacillus spp.after 48 h

SEM analysis of AgNPs embedded film:

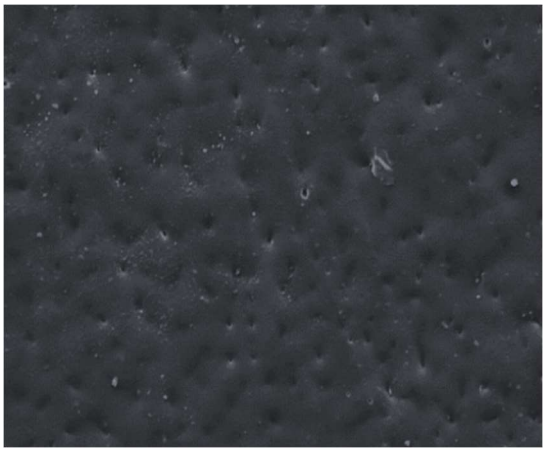


Fig. 6: SEM image of silver nanoparticles embedded cellulose based film

Discussion

Current study states a unified approach to the biosynthesis of AgNP from AgNO3 using an aqueous extract of Bacillus spp. Since the bacterial extract acts as a bifunctional molecule as a reducing agent and stabilizer of AgNP synthesis, the method used follows the principles of green chemistry. Since biological methods are environmentally friendly and economical, they always find a better approach than chemical and physical methods. The biosynthesis method showed the maximum production of nanoparticles within 48 hours. An interesting conclusion can be made from the study is that the shape and size of the synthesized nanoparticles have a direct and strong influence and depend on the pr°Cess parameters used in the experiment. The ultraviolet spectrum confirms a decrease in silver ions at 440 nm after a 48-hour incubation period. The average size of AgNP is 33-37 nm from SEM. Briefly, silver nanoparticles embedded cellulose based plastic films were prepared.

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