

SILVER NANOPARTICLE IMPREGNATED BIOMEDICAL FIBER

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Abstract— Recently, biosynthesis of metal nanoparticles has drawn considerable attention due to environment friendly and sustainable methods. Herein, *Bacillus subtilis* TD 6 was selected as candidate for biosynthesis of silver nanoparticles (AgNPs). These nano-sized silver particles (AgNPs) at concentration of 500 ppm were synthesized using maize and potato starch as both reducing and stabilizing agent. A solution containing AgNPs (500 ppm) was diluted with distilled water to 100 and 50 ppm and applied to cotton fabrics in presence of binder. The finished fabric characterization was carried out using UV-Vis spectroscopy. The antibacterial activity of the treated fabrics loaded with AgNPs was evaluated against *Escherichia coli* (gram negative) and *Staphylococcus aureus* (gram positive) bacteria. Results explored that, binder retains excellent antibacterial properties even after 20 washing cycles reflecting the significance of binder in fixation of AgNPs deposits on the surface of the fabrics.

Keywords—Nanomaterials, nanoparticles, silver nanoparticles, antibacterial activity

I. INTRODUCTION

Nanomaterials are the rapidly developing field of modern science and technology. Nanomaterials utilizes the structures with at least one dimension of nanometer size for the construction of materials, devices or systems with novel or significantly improved properties due to their nano-size. New applications of nanoparticles and nanomaterials in the field of medical technology appear to be emerging rapidly. One of such applications is the introduction of newly devised wound dressing strategy. In order to prevent or reduce infection, a new generation of dressing incorporating antimicrobial agents like silver has been developed. Silver metal and silver dressings, when used in reasonable amounts, has no negative effects on the human health & body and it has a natural antimicrobial activity towards many pathogens such as bacteria, viruses, fungi, yeast, etc. when human beings are infected by these microorganisms (1, 2). Thus silver nanoparticles appears to be an interesting candidate for incorporating in different kind of textile fibers. It is used in textile fibres in two main forms, one is integrating silver ions into the polymer and the other is coating the silver physically onto the yarns. In both these cases the silver prevents the growth of a broad spectrum of microorganisms. Silver nanoparticles have the advantage of high anti-microbial activity even at low concentrations (3, 4). Metal nanoparticles are intensely studied due to their unique optical, electrical and catalytic properties (5). To optimize their chemical or physical properties of nanosized metal particles, a large spectrum of research has been focused to control the size and shape, which is crucial in tuning their physical, chemical and optical properties therefore to utilize them in the best possible way (4, 6). Various techniques exist to prepare metal nanoparticles, which include chemical and physical methods, such as chemical reduction (7, 8, 9), electrochemical reduction

(10, 11, 12), photochemical reduction (13, 14) and heat (15). However the major problems associated with these methods are high energy consumption, use of toxic solvents and generation of harmful by-products (16). Thus in the recent scenario, biosynthesis of metal nanoparticles is considered to be a new growing era to develop. Inspiration from nature comes through yeast, fungi, bacteria and plant extracts for the control synthesis of biocompatible metal and semiconductor nanoparticles (17). Antibacterial agent i.e.; the application of AgNPs which are to be applied to textiles received a great deal of attention (18) for three major reasons: (a) to contain the spread of disease and avoid the danger of injury-induced infection, (b) to contain the development of odor from aspiration, stains and soil on textile materials, and (c) to contain the deterioration of textiles caused by mildew, particularly fabrics made of natural fibres. The present research article involves two major objectives: (1) green synthesis of AgNPs by extracellular enzymes of *Bacillus subtilis* TD-6 strain using economically and commercially viable potato & corn starch as both reducing and stabilizing agent for the formed AgNPs at optimum conditions; (2) incorporation/deposition of AgNPs on to cotton fabrics with binder & evaluating their antibacterial activity.

II. MATERIALS AND METHODS

A. Preparation of AgNPs by *Bacillus subtilis* TD-6

Bacillus Subtilis TD-6 strain was inoculated overnight into 250 ml Erlenmeyer conical flasks containing Luria broth (Difco) and then kept under static conditions. The extracellular enzyme was harvested after 24 h of growth by centrifugation. The enzyme obtained in the form of supernatant was taken in 250 ml Erlenmeyer conical flask containing silver nitrate (AgNO₃) solution (Sigma-aldrich) and kept for 72 h under fluorescent bulb covered in light deficient area. The synthesized nanoparticles were separated from these aqueous solution by centrifugation followed by drying the pellet in the hot air oven overnight. The obtained dried metal ions was routinely monitored by visual inspection of the solution, as well as, by UV-Vis spectroscopy.

B. Characterization of AgNPs: UV-Vis Absorption Spectroscopy

Preliminary detection of synthesised nanoparticles was carried out by observing the colour change of the solution. Later the sample was subjected to optical measurements. The AgNPs was taken in the quartz cuvette after vortexing and the absorption spectra were recorded using UV-Visible spectrophotometer (GE, Model: 1-2902). The approximate range of the peak of absorbance for AgNPs is within 300-550 nm.

C. Coating the fabrics with AgNPs

Before being used, cotton fabrics were washed thoroughly and dried. Experiments were performed on samples with maximum dimension of 10 mm x 10 mm. Cotton fabrics were padded with silver nanoparticles solutions at an appropriate concentrations; concentrations were achieved through solubilizing the nanoparticles with maize starch at 7% (w/v) and potato starch 10% (w/v). Gelatinized starch was prepared by boiling aqueous solution at 80°C. For the successive treatment of fabrics with colloidal silver, the solution was agitated continuously. All samples were immersed in such colloid bath for 1 min then squeezed to 100% wet pick up with laboratory padder at constant pressure. Samples were dried in hot air oven for 24h.

D. Antimicrobial activity testing

Anti-bacterial testing was performed for the following fabrics (i) untreated fabrics, (ii) fabrics treated with AgNPs, and (iii) AgNPs treated fabrics subjected to subsequent washings (5, 10 and 15 times washings). Laundering was done with water containing 2% Na₂CO₃ and soap. After each washing (30 mins), clothes were dried in a dryer at 70°C. Anti-bacterial test was evaluated against *E.coli* (gram negative) and *S.aureus* (gram positive). The two bacteria were allowed to grow on luria agar plates (20 ml each plate). Bacterial strains were plated by using luria broth culture as inoculums. One plate was left blank to check for contamination. Two plates with grown culture (both *E.coli* and *S. aureus*) were used as control for checking NP activity. Each plate contains one well, one untreated fabric with only starch suspension, one treated fabric (both maize and potato in different plates), and samples with subsequent washings.

III. RESULTS AND DISCUSSIONS

A. Preparation of AgNPs

AgNPs were synthesized by green synthesis using *Bacillus subtilis* TD-6 strain at optimum conditions using potato and corn starch as binding agent.

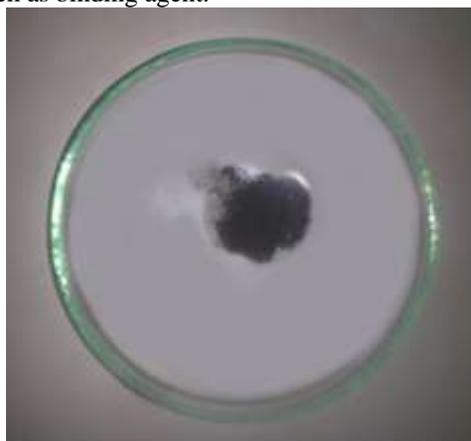


Fig 1: Synthesized starch coated AgNPs on a petriplate.

B. UV-Visible spectroscopy

Preliminary detection of nanoparticles was done by observing the colour change of the solution. The sample was subjected to optical measurements with the optimum peak between 300-550nm. The prepared AgNPs showed its optimum peak at about 420 nm which is in confirmation with previous reports of the literature (19).

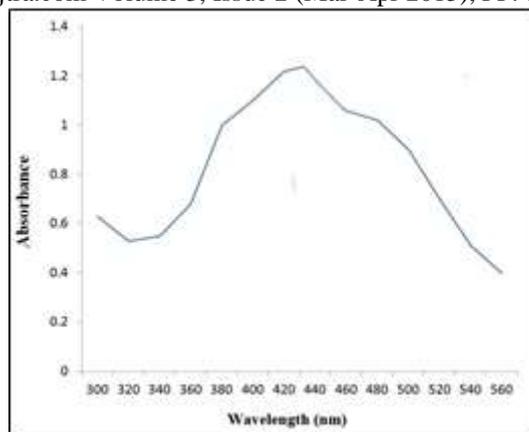


Fig 2: UV- Visible spectroscopy analysis of the prepared AgNPs. Presence of clear peak at about 420 nm confirms the synthesis.

C. Coating of desized cotton fabrics with AgNPs

The nanoparticle coating resulted in a thin film like appearance over the desized fabrics.



(a)



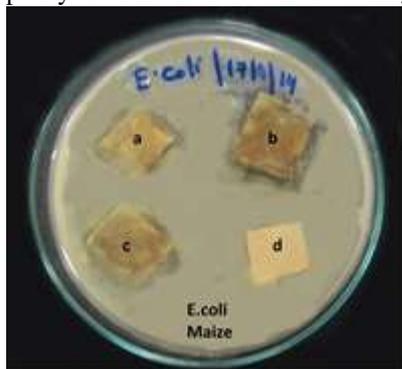
(b)

Fig 3: Desized cloth coated with (a) Maize starch bound and (b) Potato starch bound AgNPs.

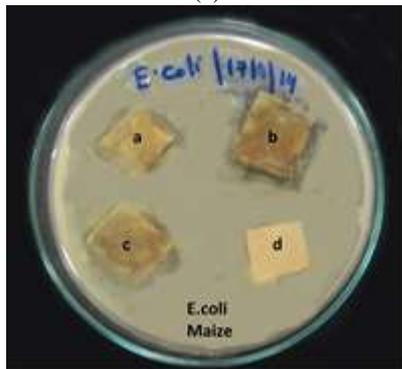
D. Antibacterial assay

Both the strains *Escherichia coli* and *Staphylococcus aureus* showed significant zone of inhibition for the nano-coated fabrics. Maximum zone of inhibition was observed in case of unwashed potato starch bound AgNPs coated fabrics. Among the washed pieces of fabrics significant zone of inhibition was obtained in case of those which were treated with minimum washing of five times and also coated with potato starch bound AgNPs. Antibacterial activity was also noticed for the nanoparticle suspension whereas not even a trace of the same was observed in only starch bound uncoated

fabrics (without nanoparticles). Hence the starch coated AgNPs contain antimicrobial activity and the potato starch is found to be the more efficient binding agent than maize starch having a maximum capacity to withstand five times washing.



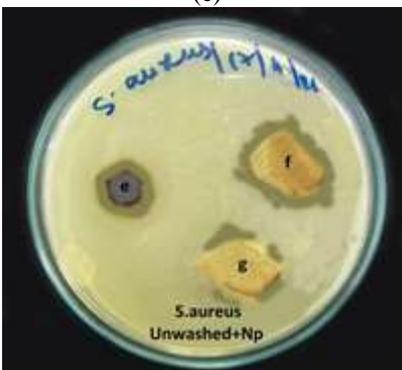
(a)



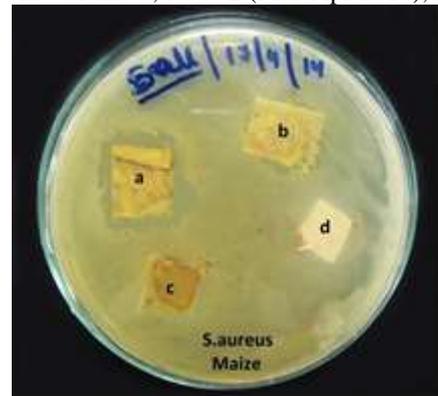
(b)



(c)



(d)



(e)



(f)

Fig 4: Images of the antimicrobial assay of starch bound AgNPs coated fabric pieces. (a) Maize starch bound AgNPs coated fabrics tested against E. coli. (b) Potato starch bound AgNPs coated fabrics tested against E. coli. (c) Both types of starch bound AgNPs coated unwashed fabrics and AgNPs suspension tested against E. coli. (d) Both types of starch bound AgNPs coated unwashed fabrics and AgNPs suspension tested against S. aureus. (e) Maize starch bound AgNPs coated fabrics tested against S. aureus. (f) Potato starch bound AgNPs coated fabrics tested against S. aureus. a- 5 times washed, b- 10 times washed, c- 15 times washed, d- uncoated, e- AgNPs suspension, f- Potato starch bound AgNPs coated unwashed fabrics, g- Maize starch bound AgNPs coated unwashed fabrics.

IV. CONCLUSION

Synthesis of the AgNPs has been carried out by a simple, cost effective, rapid and environmentally benign technique of biological reduction of AgNO_3 by microbial cell extract. Ultraviolet- visible spectroscopy has confirmed the synthesis of the silver nanoparticles. Use of biopolymer based binding agent like potato and maize starch has increased the holding capacity of the fabric for the nanoparticles. Antimicrobial assay revealed the presence of antibacterial activity of these nano-coated fabrics with more efficacy in presence of potato starch coating. This work can be further improvised by the inclusion of various physico-mechanical parameters e.g., tensile strength, rheological stress, flame retardant ability, acid and alkali resistance etc. In terms of binding agents the method can be further optimized to increase the pressure handling ability of the fiber so that it possesses its activity even if subjected to more number of washing cycles. In a significant note, the antibacterial activity of the nanoparticle impregnated clothes even after repeated washing opened up a new horizon for devising a wound healing strategy especially for the defense personnel and others who work in various remote areas or

harsh environments. Covering the wounded area with this nanocoated bandages can decrease the chance of infection as well as this can be used for drug delivery also.

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