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Preparation of cotton fabric using sodium alginate-coated nanoparticles to protect against nosocomial pathogens

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ABSTRACT

There is a vital need for the production of clean, hygienic, and antibacterial textile fabric for use as hospital clothing, surgical scrubs and linens to protect against nosocomial pathogens. In the present study, cotton fabrics were treated with sodium alginate-capped silver nanoparticles and were tested for antibacterial activity against nosocomial pathogens. The sodium alginate-capped silver nanoparticles were synthesized under various experimental conditions by using varying sodium alginate concentrations (0.5–2%), volumes of reducing agent aniline (50, 100, 150 μL), and durations of heat treatment (30–240 s). The synthesized nanoparticles were characterized using ultraviolet-visible spectroscopy, X-ray diffraction, Fourier transform infrared spectroscopy and scanning electron microscopy. Furthermore, the antibacterial efficacy of sodium alginate-capped silver nanoparticles treated cotton fabrics were tested against different nosocomial pathogens (*Escherichia coli, Pseudomonas aeruginosa* and *Staphylococcus aureus*). The results clearly showed that the sodium alginate-capped silver nanoparticles treated cotton fabrics exhibit excellent antibacterial activity for use in different medical textile fields.

1. Introduction

Hospital clothing, surgical scrubs, and linens used currently have been proven to provide insufficient protection from infection for health care workers [1]. Survival and transfer of microorganisms between patients and health care workers have been reported in many studies [1,2]. As a result, modern cotton fabrics require improvements in their antimicrobial properties to protect against nosocomial pathogens. Recently, nanotechnology has become a fast growing, intriguing area of research because of its many potential applications in the medical field. Metal nanoparticles like silver, gold, and iron oxide exhibit interesting optical and electronic properties, and they have a wide scope in biological and medical sciences because of their brilliant colors and inherent antimicrobial activity [3–5]. Specifically, silver nanoparticles (AgNPs) have gained a lot of importance owing to their unique and remarkable properties, including their electronic, electrical, optical, catalytic, mechanical, heat transfer, sensor, conductive, and antimicrobial properties [6–9]. Typical methods for preparing AgNPs are chemical reduction, photochemical reduction, the reverse micelles process, the electrochemical, reflux, and sonochemical methods, UV photolysis, microwave dielectric heating reduction, ultrasonic irradiation radiolysis, solvothermolysis, and green synthesis of metal salts [10–13]. The nanoparticles prepared using these methods must have good dispersibility and thermal stability, which are considered very important for industrial applications [13]. Among these methods, the microwave can be used to synthesize nanoparticles within a few minutes. The microwave synthesis method also ensures uniform heating of the reaction mixture. The advantage of microwave heating over conventional heating is the rapid and uniform internal heating of the solution; microwave irradiation generates very fast nucleation sites in the solution, which significantly enhances the reaction rate [14]. Methods that involve reducing and protecting molecules or organic capping molecules to synthesize nanoparticles are important synthetic approaches to stable nanoparticles synthesis without the challenge of aggregation [15,16]. Nowadays, green synthesis of metal nanoparticles has opened up a new route for the synthesis of different shaped and sized nanoparticles without using any harmful chemical reducing agents. In this method, mainly plant extracts, natural antioxidants, green solvents, amino

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acids, and glucose and its derivatives are used as the reducing agents [17,18]. The nanoparticles synthesized by this method have been used to study antimicrobial activity and dye decoloration [19,20]. Microwave synthesis of metal nanoparticles is considered a green method [21,22].

In the green synthesis of AgNPs, three important factors to be considered are (i) the use of green solvents, (ii) the use of an eco-friendly, benign reducing agent, and (iii) non-toxic material as a stabilizer. The polysaccharide method is a green method for preparing AgNPs. In this method, water is normally used as an eco-friendly benign solvent and polysaccharides as capping agents. Raveendran et al. [6] reported first completely green synthesis of AgNPs using water, starch, and D-glucose as the solvent, capping agent, and reducing agent, respectively. The use of starch makes it possible to avoid the use of relatively toxic organic solvents. Based on modifications of this method, synthesis of AgNPs has been reported using different sugars and biopolymers as reducing agents including carboxymethyl cellulose, glucose, and sodium alginate [23,24]. Sodium alginate (SA) is a naturally occurring polyanionic polysaccharide derived from brown marine algae, and composed of 1, 4-linked β-D-mannuronic and α-L-guluronic residues in varying proportions. The anionic and reducing properties of SA support its utilization as a reducing and stabilizing agent for the synthesis of AgNPs. The negatively charged solubilized SA facilitates the attraction of the positively charged silver cations to the polymeric chains, followed by reduction with the existing reducing groups. This is an inexpensive, biocompatible, and environmentally benign biopolymer with numerous applications in the biotechnology industry as a non-toxic food additive, thickening agent, gelling agent, emulsifier, and colloidal stabilizer [25,26]. Alginate has widespread applications in wound healing, delivery of bioactive agents, and cell transplantation owing to its structural similarities to extracellular matrices of living tissues [26].

In the present study, microwave-assisted aqueous synthesis of sodium alginate-capped silver nanoparticles (SA@AgNPs) was performed using SA as a stabilizing agent in the presence of a trace amount of aniline as the reducing agent. Synthesized SA@AgNPs were characterized using ultraviolet-visible (UV–vis) spectroscopy, Fourier transform infrared spectroscopy (FTIR), x-ray diffraction (XRD), and scanning electron microscopy (SEM) and were tested for antibacterial activity. Finally, cotton fabrics were treated with SA@AgNPs and tested for antibacterial behavior against nosocomial pathogens. Earlier, Yang and co-researchers tested Ag/SA composites fabricated cotton against bacteria and reported that the cotton fabrics containing the Ag/SA composites showed potent antibacterial properties and maintained washing resistance [27]. The novelty of the present work is SA@AgNPs synthesized by microwave assisted method. The advantage of microwave-assisted synthesis over other physical/chemical treatment is rapid initial heating, effective to increase stability, manipulate homogenous particle size and growth rate. Hence, the method described herein is considered a green chemistry approach for synthesis of SA@AgNPs for large scale production.

2. Experimental

2.1. Materials

Silver nitrate ($AgNO_3 \geq 99.0\%$), Sodium alginate (SA), and aniline were purchased from Sigma-Aldrich, USA. Nosocomial pathogens such as *Escherichia coli*, *Pseudomonas aeruginosa,* and *Staphylococcus aureus* were obtained from the SRM Medical College and Research Centre. Muller-Hinton broth and agar were purchased from Himedia, India. Milli-Q deionized water was used in all experiments (Millipore).

2.2. Microwave synthesis of SA@AgNPs

This experiment was carried out under various conditions by varying the concentration of SA (0.5, 1, 1.5 and 2%), volume of the reducing agents (aniline; 50, 100, 150 μL), and duration of heat treatment (30–240s). In a typical experiment, 32 mL water, 1 mL 0.04 M $AgNO_3$,and 2 mL SA (various concentrations) were mixed with different volumes of aniline at room temperature and heated at 180 °C in a microwave at 80% power for varying time intervals. The samples were cooled down to room temperature and adjusted to 35 mL by adding a small amount of distilled water to compensate for the water lost during microwave irradiation. During the microwave heating process, the color of the reaction mixture changed slowly from colorless to light brown owing to the reduction of Ag^+ to Ag^0. The reaction mixtures were analyzed by UV–vis spectroscopy to confirm the AgNPs formation.

2.3. Characterization of SA@AgNPs

The UV–vis spectra of SA@AgNPs were measured in the wavelength range of 200–1000 nm by using a UV–vis spectrophotometer (Shimadzu, Japan). The XRD analysis was performed using an X'PertProA Analytical X-ray diffractometer using Cu Kα radiation (k = 1.54056 Å) in the range of 30–80 (2θ values) at 40 keV and compared with reference data from the JCPDS database. FTIR (Perkin-Elmer, USA) was performed to analyze the functional groups on the SA@AgNPs in the range of 400–4000 cm^{-1}. The SA@AgNPs and SA@AgNP-treated cotton fabrics were analyzed using SEM (Quanta FEI 200, USA).

2.4. Antibacterial testing of SA@AgNPs

The minimum inhibitory concentration of the SA@AgNPs was assayed against *E. coli*, *P. aeruginosa,* and *S. aureus.* The assay was performed in 96-well plates by using the micro dilution method [28]. Fifty microliters of the bacterial suspension, with a McFarland turbidity of 0.5 (1.5×10^8 CFU/mL), was pipetted into 96-well plates in the presence of various concentrations of the SA@AgNPs in Muller Hinton broth ranging between 0.25 and 128 μg/mL. Double sterilized Millipore water was used as a sterility control. Bacteria were allowed to grow aerobically at 37 °C for 24 h and the absorbance at 600 nm was measured. The lowest concentration inhibiting bacterial growth was taken as the minimum inhibitory concentration (MIC). Furthermore, a microtiter plate method was used to evaluate of the influence of AgNPs on colonizing the substratum. The microplates used for the MIC assay were drained and washed three times using phosphate buffered saline. The biofilm formed on the 96-well plate was fixed with ice-cold methanol for 5 min. One hundred microliters of crystal violet dye (0.1%) was added to each well and incubated for 15–20 min at room temperature to stain the adhered biofilm. Furthermore, 100 μL of 33% acetic acid was added to the wells to solubilize the dye bound to the biofilm. The intensity of the stained suspension was measured by absorbance at 575 nm using 33% glacial acetic acid as the control. Experiments were performed in triplicate.

2.5. Antibacterial testing of SA@AgNPs-treated cotton fabrics

The antibacterial activity of SA@AgNPs-treated cotton fabrics (1 cm^2) was tested against nosocomial pathogens *S. aureus*, *P. aeruginosa* and *E. coli,* which are the most common microbial pathogens encountered in hospitals, by using a modified plate assay [29] and colony counting methods. In the plate assay method, the cotton fabrics (1 cm^2) were sterilized under the UV lamp for 1 h. They were then dipped in the respective MIC value of SA@AgNPs (0.1 mL) and allowed to absorb the solution completely. Further,