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Sodium alginate stabilized silver nanoparticles–silica nanohybrid and their antibacterial characteristics

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ABSTRACT

Due to the problem of resistance of many infectious agents to the usual treatments, this study addresses the ways of obtaining and using new sodium alginate stabilized-silver/mesoporous silica (Na-Alg-s-AgNPs@SiO_2) nanohybrid as antimicrobial agents. Capping AgNPs with a shell of mesoporous SiO_2 is a system to build the increase biocompatibility of AgNPs. In this work, we report a simple and green way to deal with setting up a uniform sodium alginate–stabilized silver nanoparticles embedded mesoporous silica (Na-Alg-s-AgNPs@SiO_2 nanohybrid). The synthesized nanocomposite was characterized using transmission electron microscopy (TEM), Fourier transform infrared (FTIR) spectra, and ultraviolet-visible (UV–vis) absorption spectra, which exhibited that AgNPs with average of size of ~7 nm were consistently and compactly deposited in the nanocomposite. The nanohybrid demonstrated excellent antibacterial activity against both Gram negative (−ve) and Gram positive (+ve) bacteria. Thus, the developed Na-Alg-s-AgNPs@SiO_2 nanohybrid has a potential to be used for various antibacterial applications in biotechnology and biomedical fields.

1. Introduction

In recent years, antimicrobial resistance inside an extensive variety of infectious agents is one of the major issues, which is a concern for governments around the globe. It is a public wellbeing menace that influences not only nations but also the various field which, in turn, undermine to the achievements of modern medicine [1]. Consequently, the synthesis and biocide impacts of novel compounds are severely studied [2].

In-spite of the fact, that silver has been utilized from time immemorial. In current years, it has been employed more extensively for numerous biomedical [3,4] and industrial (such as electrical appliances, garments) [5,6] applications as it shows fantastic antimicrobial properties against an extensive variety of bacteria and fungi [7]. It is known that silver ions accomplish noteworthy decreases in microbial growth at very low concentrations, and despite the fact that the mechanism that causes this impact have not been totally illustrated yet, some researchers have attributed the cytotoxicity of silver to a few conceivable mechanisms, including the production of a lot of free radicals affecting high oxidative stress, the disturbance of cell layer integrity, or protein or DNA binding and damage to genetic material [8,9].

Even though the biocidal properties of silver compounds are well described, today these properties have been upgraded through nanotechnology by method for silver nanoparticles (AgNPs) which has permitted the extent of metal nanoparticles to be modularly and accurately reproduced keeping in mind the end goal to enhance the stability of the colloidal emulsions and in addition a controlled release of the AgNPs, acquiring a long-term antimicrobial activity and reducing undesirable potential impacts, among others [10]. This has made it conceivable to confine dose to maintain a strategic distance from cytotoxic consequences, to control the products helpful for life, to enhance the release profile for specific tissue-targeted on or intracellular-targeted delivery, or to minimize the release of silver ions into nature [11].

Emerging materials such as AgNPs [12], copper oxide NPs [13], carbon nanomaterials [14], and metal oxide NPs [15], have been reported as antimicrobial agents. To acquire product with new functionalities, all the more particularly with the ability to inhibit microorganism development (antimicrobial impact), colloidal AgNPs emulsions were synthesized utilizing different reducing agents: such as sodium borohydride [16], gelatine/glucose [17] and glycolic Aloe vera extract [18]. The first was employed as a control for the other two, which are included in “green” synthesis

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processes that are more harmless and ecologically well-disposed [11].

However AgNPs have demonstrated to be potential antimicrobial agents, they present the problem of being highly cytotoxic to mammalian cells [19,20]. The synthesis of silver nanocomposites might be a possible answer for improve the antibacterial activities of AgNPs and to defeat the unfavorable harmful impacts of silver by optimizing the concentration of Ag. One approach to synthesize silver nanocomposites is to use carbon-based nanomaterials such as graphene [21], carbon nanotubes (CNTs) [22] and graphene oxide [23]. But several reports have shown, CNT and graphene oxide based silver nanocomposites are potentially hazardous against mammalian cells [24,25]. In past few years, mesoporous silica materials have received growing attention due to their potential biomedical applications [26,27]. The interest on this material has been greatly encouraging by the excellent silica properties for better use of metal NPs including rich surface chemistry, high biocompatibility and controllable porosity [28,29]. Therefore, capping AgNPs with mesoporous silica is a strategy to improve biocompatibility with mammalian cells and also for a better control of silver ions released from a mesoporous matrix, enhancing its therapeutic effect. So far, the most accepted mechanism of antibacterial action is the interaction between AgNPs and the bacterial cell envelope (thin peptidoglycan cell wall, with outer membrane containing lipopolysaccharide in the case of gram −ve bacteria; and thicker layers of peptidoglycan in the case of gram +ve bacteria). Modification of surface chemistry of silver-mesoporous silica nanoparticles by increasing the silica coating layer encapsulating the AgNPs or by adding different functional groups would results in increase in these interactions.

Thus, remembering the above significance highlight, in the present paper, we have reported a simple, cost-effective, green engineered strategy to synthesize mesoporous silica-capped silver nanoparticles (Na-Alg-s-AgNPs@SiO_2) nanohybrid by sol–gel polymerization of TEOS under template impact [30–32] of polyanionic sodium alginate (Na-Alg). This strategy help in ensuring AgNPs with a porous coating, for example, a silica matrix, by shaping silica nanocomposites (Ag@SiO_2) by sol-gel technique equipped for keeping the aggregation of the NPs yet permitting the antimicrobial mechanism, thus reducing the possible incompatibility with materials matrices. The silica structure goes about as an advantageous carrier for consolidating the fine AgNPs into plastics, textiles, and coatings. A further favorable position is that the immobilization of AgNPs inside the silica structure restrains the potential for release and disposal of the NPs themselves. This property might be very attractive in light of the conceivable capacity of NPs to go through biological membranes and different obstructions. The resultant nanohybrid demonstrates excellent antibacterial action. Other potential pertinence areas of the hybrids are open for future investigations.

2. Experimental section

2.1. Materials and microbial strains

Low viscosity Na-Alg Molecular Formula: $C_6H_9NaO_7$, (purity of about 95%) was purchased from Sigma-Aldrich (U.S) with the following characteristics (1000 cP for 5% wt/vol water solution (5 g of Na-Alg per 100 mL of solution), conductivity ~35 S/m at 25 °C, Mw ~68,000 Da, F_G = 0.31 and F_M = 0.62, where F_G and F_M represent the fractions of guluronic and mannuronic groups, respectively) and used as received. Tetraethylorthosilicate (TEOS, Aldrich 99.999%), Ethyl alcohol (C_2H_5OH, Aldrich 99.9995%), Ammonia solution (NH_4OH, Merck 32% extra pure), Silver nitrate ($AgNO_3$, Sigma-Aldrich ≥99.0%), Nalidixic acid (Sigma-Aldrich 98.0%), and deionized water were used for the preparation of samples. All these chemicals were of analytical grade and used as received.

The tetrazolium dye, i.e., *p*-iodonitrotetrazolium violet ($C_{19}H_{13}ClIN_5O_2$, Sigma-Aldrich). Oxoid™ Mueller Hinton Agar, Oxoid™ Tryptone Soya Broth were purchased from Thermo Fisher Scientific Inc. (UK). BD BBL™ Middlebrook ADC Enrichment and BD Difco™ Middlebrook 7H9 Broth from BD, Germany. All the Gram-positive bacterial strains: *Staphylococcus aureus* (ATCC 25923), *Staphylococcus epidermidis* (ATCC14990), *Bacillus cereus* (ATCC 10876), *Bacillus subtilis* (ATCC 19659) and Gram-negative bacterial strains: *Enterobacter cloacae* (*ATCC 13047*), *Enterococcus faecalis* (*ATCC 29212*), *Escherischia coli* (*ATCC 25922*), *Klebsiella oxytoca* (*ATCC 8724*), *Klebsiella pneumonia* (*ATCC 13882*), *Proteus mirabilis* (*ATCC 7002*), *Proteus vulgaris* (*ATCC 6380*), *Pseudomonas aeruginosa* (*ATCC 27853*), *Enterobacter aerogenes* (*ATCC 13048*) were originally obtained from Davies Diagnostics (South Africa). All solutions were made using ultra-filtered high purity deionized water.

2.2. Synthesis of sodium alginate stabilized silver nanoparticles (Na-Alg-***s-AgNPs)***

We report the development of a novel, facile and 'green' route method [33] for synthesizing AgNPs using Na-Alg as both reducing and stabilizing agent in an aqueous medium by heating alone. No external agent was added. This method utilizes a proteinaceous, edible, renewable, and water soluble biopolymer; Na-Alg which functions as both reducing and stabilizing agents during synthesis of AgNPs. Na-Alg stabilized AgNPs (Na-Alg-s-AgNPs) were synthesized by vigorous stirring at 500 rpm on a magnetic stirrer, 5 mL of 20 mM silver nitrate solution in 25 mL of 0.5 g Na-Alg solution at 70 °C for 100 min. The reaction mixture changes from colorless to a dark brown colored solution within 100 min time interval indicating the formation of AgNPs. Synthesis of two different sized AgNPs (7 nm and 15 nm) was achieved by varying the pH 10 and 8 respectively, keeping all the other conditions constant. Finally, AgNPs were stored at 10 °C in the absence of light.

2.3. Synthesis of mesoporous silica-capped silver nanoparticles (Na-Alg-s-AgNPs@SiO_2) nanohybrid

To improve colloidal emulsions stability avoiding AgNPs aggregation, Na-Alg-s-AgNPs@SiO_2 nanohybrid was synthesized. Synthesis of Na-Alg-s-AgNPs@SiO_2 nanohybrid was performed by using sol-gel method involving two important steps; hydrolysis and polycondensation reaction. 30 mL solution of synthesis Na-Alg-s-AgNPs (~7 nm/~15 nm) was dissolved homogeneously in 10 mL of deionized distilled water. Separately 2.5 mL of TEOS was also dissolved in 2.5 mL of ethanol. A third solution incorporating (0.9 mL of 12 N) ammonium hydroxide was prepared separately. Afterward, the three solutions were rapidly poured together into a glass reaction flask, which was kept under gentle stirring for 16 h at room temperature to grow monodisperse SiO_2 particles within the Na-Alg-s-AgNPs medium. The condensation reaction generally started immediately allowing silver nanoparticles to be embedded into silica matrix. The ensuing mixture was then subjected to slow evaporation at 40 °C (3 h), 60 °C (4 h), 70 °C (2 h) and 80 °C (2 h) to obtain a dry material Na-Alg-s-AgNPs@SiO_2 nanohybrid involving two different size AgNPs (~7 nm and ~15 nm) say Na-Alg-s-AgNPs@SiO_2 nanohybrid (1) and Na-Alg-s-AgNPs@SiO_2 nanohybrid (2) respectively. In the paper, we have shown and discuss the characterizations results of Na-Alg-s-AgNPs@SiO_2 nanohybrid (1).