Suspension of Silver Oxide Nanoparticles in Chitosan Solution and its Antibacterial Activity in Cotton Fabrics

Zhigang Hu¹, Jin Zhang¹, Wing Lai Chan², and Yau Shan Szeto¹

¹Institute of textiles and clothing, The Hong Kong Polytechnic University, ITC, PolyU, HungHom, KLN, Hong Kong, Hong Kong
²Department of applied biology and chemical technology, The Hong Kong Polytechnic University, ITC, PolyU, HungHom, KLN, Hong Kong, Hong Kong

ABSTRACT

New stable silver oxide suspension in chitosan solution was prepared from a mixture of silver nitrate and chitosan in dilute acetic acid as precursor, in which the complex interactions between silver ion and chitosan was also investigated through various instrumentation method seriously. The suspension was charactized through laser scan, infrared ray spectroscopy (IR), ultraviolet-visible spectroscopy (UV-vis) and X-ray diffraction (XRD), which indicated that the interactions between silver and chitosan in its precursor were destroyed partially. The measurements of the nanoparticles through scanning electron microscopy (SEM) and transmission electron microscopy (TEM) disclosed the spherical profiles of these silver oxide nanoparticles of 10-20nm in average. Cotton fabrics treated by this emulsion were entitled remarkable antibacterial activity against S aureus and E. coli at PH 5 and 7 with sightless color effect and good washing fastness.

INTRODUCTION

Silver-based antibacterial materials attracts much attention because of their long-term biocidal activity, low volatility and the nontoxicity of the active silver ion to mammalian cells¹,² as well as its perfect antibacterial activity.³-⁵ Many of these materials have been introduced, which are prepared through either doping silver onto the host materials⁶,⁷ or compositing it with other compounds.⁸,⁹ Whereas, to our knowledge, few efforts are done about a aqueous suspension of silver-based materials. A uniform and stable emulsion is believed to be advantageous in many fields, for instance, directly coating of biomaterials, devices or medical textiles in the biologic or therapeutically applications, spraying or dispersing into the circumstance for hygienic purpose. Most of presented works have to use additional surfactant, reductant, emulsifier or other assistant agents, which usually cause side-effects or be toxic. Meanwhile, chitosan is wildly used as a natural polysaccharide deriving from chitin because of its excellent properties, such as antibacterial, nontoxic, biodegradable, biocompatible.¹⁰-¹⁴ Another important property of chitosan is that the amino groups and hydroxyl groups of chitosan can form chelate complex with silver ion.¹⁵,¹⁶ This property provides a possibility to use chitosan as both reductant and surfactant in the preparation of nano scale silver-based materials from silver ion. Works have been done on the preparation of chitosan/silver nanocomposites, which prove the ability of chitosan to reduce silver ion into metallic silver.¹⁷-²¹ But most of these nanocomposites are solid, such as fibers, powders and films, due to their poor stability in aqua.
In this study, we try to use chitosan as not only a reductant but also a surfactant in the preparation of a silver-contained aqueous suspension. The achieved suspension was characterized through kinds of instrumental analysis, such as laser scan, infrared ray spectroscopy (IR), ultraviolet-visible spectroscopy (UV-vis) and X-ray diffraction (XRD), while the growth of silver nanoparticles in the chitosan solution was investigated. Also the positive charge of the suspension was disclosed as well as the spherical modify of silver oxide nanoparticles in the suspension was identified. To obtain a tentative overview about the antibacterial activity of the emulsion, we apply it to treat cotton fabrics whose antimicrobial activity are then enhanced remarkably with sightless colour effect and bearable durability to water washing.

EXPERIMENT

Materials

Chitosan (degree of deacylation: 95%, molecule weight: 500 kDa) was purchased from HDB Ltd. China. Other used chemicals were analytical grade from Aldrich Ltd. without further treatment.

Preparation of the suspension

Typically, 5% (w/v) silver nitrate solution was dropped into a chitosan solution of 0.5% (w/v) dilute acetic acid under magnetic stirring. Then sodium hydroxy solution was dropped into the chitosan/AgNO₃ solution. Black precipitates produced and aged at pH of 10. After adjusting the pH of the solution with acetic acid, the suspension was obtained with brown color and pH of 4.

Morphology characterization

Zetasizer 3000HSA was used for laser scanning investigation; Scanning electron microscopy (SEM) analyses were performed through JOEM JSM 6335F; Transmission electron microscopy (TEM) analyses were performed through Philips CM-20 TEM.

Treatment of cotton samples and relative tests

Cotton fabrics were soaked with the suspension of certain concentration for 3 minutes, subsequently padded with Pick Up Weight around 80% through a laboratory pad machine (Rapid Vertical Padder, Taiwan). All padded samples were dried at 100 °C for 3 minutes and cured at 150 °C for 3 minutes, then rinsed with tap water and dried again. Flask Shake Method was used for the antibacterial test against E. coli and S. aureus. Durability to home laundry test was performed according to AACTT 63 option 1.

DISCUSSION

Growth of silver nanoparticles in the chitosan solution
Adding of silver nitrate changed the chitosan solution phenomenally from colorless to brown slowly. **Fig. 1** shows the UV–vis spectrum illustrating the slow change, in which two gradually increasing peaks are observed from 350nm to 700nm wavelength. Silver nanoparticles absorb radiation in the visible region of the electromagnetic spectrum due to the excitation of surface plasmon vibrations responding to the striking violet colour of silver nanoparticles in various media. 19-21 The peak at 390nm wavelength in **Fig. 1** should respond to the plasmon vibration in this case with increasing intensity due to the size change of produced silver particles. 17 **Fig. 2 a** shows the SEM of a film from the silver-contained chitosan solution, in which silver nanoparticles with size of around 20nm can be observed. The other peak shifting from 500nm to 600nm wavelength is supposed to relative with the non-spherical morphology of the obtained particles. After longer stirring duration, hexagonal silver crystals with bigger size are observed, shown in **Fig. 2 b**. These nanoparticles kept growing until black precipitates appeared finally after stirring of several days.

![UV-vis spectrum of the chitosan/AgNO₃ solution at different stirring duration](image1.png)

**Fig. 1.** UV-vis spectrum of the chitosan/AgNO₃ solution at different stirring duration

![SEM and TEM images](image2.png)

**Fig. 2.** (a) SEM microscopy of the films from the silver-contained chitosan solution (b) TEM of the silver crystal generated in the reactive solution after 30 hours

We also investigated the possible chemical interaction between chitosan and silver. The differences between the two spectrums focus on 2800-3500cm⁻¹, 1500-1700cm⁻¹, 900-1100cm⁻¹ and 700-400 cm⁻¹, which mainly correspond to the stretching vibration and bending vibration of amino group and hydroxyl group. The peaks of NH₂ and OH groups between 3000 cm⁻¹ and
3500 cm\(^{-1}\) have significant changes, indicating that the \(\text{NH}_2\) and \(\text{OH}\) groups bear certain interaction affecting their vibration characteristics. The 1680 cm\(^{-1}\) peak of \(\text{CONH}_2\) and 1600 cm\(^{-1}\) peak of \(\text{NH}_2\) shifted separately to 1640 cm\(^{-1}\) and 1560 cm\(^{-1}\), which implies that the amino groups build new relationship with the silver. New bands observed in 700-400 cm\(^{-1}\) are assigned to stretching vibration of N-Ag and O-Ag. Banding with Ag(I) causes the substantial redistribution of different types of vibrations associating with the amino groups and hydroxyl groups.

**Fig. 3** shows the XRD spectrums of pure chitosan and the film. There are mainly two strong peaks at about 10 and 20 degree for the pure chitosan due to the high degree of crystallinity, which disappear in the spectrum of the later. The disappearance of the peaks reflects great disarray in chain alignment of chitosan with production of new peaks identifying silver and silver oxide. When a matrix substance crystallizes with a solute additive, this substance can observe either a matrix lattice parameter alteration, or the formation of a new crystalline phase which is characterized by new bands in the XRD pattern, and the later case occurs when the solute atoms occupy well-defined positions in the matrix lattice. The above crystalline behaviors and the analyses of far-IR spectrums reveal that amino groups and hydroxy groups of chitosan are involved in the interactions between chitosan and silver. However, these complex interactions between silver and chitosan can not take the role as stabilizers for the generated nanoparticles because these particles keep growing, aggregating, and deposit from the solution finally.

![Fig. 3. XRD spectrums of (a) pure chitosan and (b) the film](image1)

![Fig. 4. XRD spectrums of the dried suspension](image2)

**The suspension of the silver oxide nanoparticles**

The black suspension of the silver nanoparticles in the chitosan displays good stability during the storage of over three months. The result of laser scan indicates that the suspension contains positively charged silver oxide nanoparticles with average size of around 20 nm and zeta potential of +70 mV. **Fig. 5** illustrates the SEM and TEM of the silver nanoparticles in the suspension, in which the spherical profile of the silver oxide nanoparticles can be identified with around size of 20 nm close agreeing with the result of laser scan.
The far-IR spectrum investigation indicates the disappearance of the complex interactions. The spectrum of the precipitates is similar with that of pure chitosan. Also, these peaks in Fig. 3b are not observed in the XRD spectrum of the dried suspension while the crystalline peaks appeared again companying peaks of silver oxide, as Fig. 4 shows. Alkali treatment obviously destroyed the complex interactions between chitosan and silver, resultanty freed the chemical groups of chitosan to form inter and intra hydrogen bonds which recovered the crystallinity of chitosan. Silver was oxidized simultaneously. But the weak of peaks in Fig. 4 implies that some kind of interaction between chitosan and silver still exist. A single peak at 440nm wavelength was observed in the UV-vis spectrum of the suspension according to the plasma vibration of silver instead of the 390 nm one in Fig. 1.

Fig. 5. (a) SEM and (b) TEM microscopy of the suspension

*Treated cotton fabrics and antibacterial activities*

The nanoparticles built up a rough layer on the surface of cotton fibres, in which small silver oxide nanoparticles were dispersed while some of them were still visible. The colour of silver is a wildly-met problem in the application of textile because treated cotton will become yellow when higher concentration of silver is used. In this study, the cotton fabrics treated with the emulsion containing 6ppm chitosan and 2ppm silver oxide still obtain 100 % bacterial reduction to S. aureus and E. coli at pHs 5 and 7 when no obvious colour change occurs; after washed equal to twenty times of standard home laundry, the bacterial reductions still keep in 100%, as Fig 6 shows.

Fig. 11 Comparison of bacterial reduction before and after treating cotton fabrics with the composite chitosan/silver oxide nanoparticles (Sample A), or the treated cotton fabrics after standard home laundry of 20 times (Sample B)
CONCLUSIONS

The growth of silver nanoparticles from silver nitrate in chitosan solution was studied. Basing on the obtained results, silver ion formed complicated chelate complex with both amino groups and hydroxy groups of chitosan. The conducted silver nanoparticles are not stable in the air and become black precipitates finally. After further treatments, the chemical interactions between silver and chitosan were destroyed partially and the suspension of silver oxide nanoparticles is obtained, which is composed of silver oxide composite nanoparticles and has perfect stability in the air. Cotton fabrics have excellent antibacterial activity against S. aureus and E. coli at pH 5 and 7 after treated with the emulsion at very low concentration with good washing fastness. Applying of such low concentration avoids the boring color effect of silver successfully.

ACKNOWLEDGMENTS (OPTIONAL)

We thank the Nanotechnology Centre of the Hong Kong Polytechnic University. Z.G. H thanks the Hong Kong Polytechnic University for the studentship.

REFERENCES

17. Mormi NM, Mohamed NS, Arof AK, Mat Sci Eng B, 1997; 45:140-146.