Hindawi Publishing Corporation
Journal of Nanomaterials
Volume 2016, Article ID 9505704, 8 pages
http://dx.doi.org/10.1155/2016/9505704

Research Article

Characterization of Silver Nanoparticle *In Situ* Synthesis on Porous Sericin Gel for Antibacterial Application

Gang Tao,[1] Lina Liu,[1] Yejing Wang,[1,2] Huaipu Chang,[1,2] Ping Zhao,[1] Hua Zuo,[3] and Huawei He[1,2,4]

[1]*State Key Laboratory of Silkworm Genome Biology, Southwest University, Beibei, Chongqing 400715, China*
[2]*Chongqing Engineering and Technology Research Center for Novel Silk Materials, College of Biotechnology, Southwest University, Beibei, Chongqing 400715, China*
[3]*College of Pharmaceutical Sciences, Southwest University, Beibei, Chongqing 400715, China*
[4]*National Laboratory of Biomacromolecules, Institute of Biophysics, Chinese Academy of Sciences, 15 Datun Road, Chaoyang District, Beijing 100101, China*

Correspondence should be addressed to Hua Zuo; zuohua@swu.edu.cn and Huawei He; hehuawei@swu.edu.cn

Received 26 December 2015; Accepted 11 April 2016

Academic Editor: Liangliang Hao

Sericin from *Bombyx mori* cocoon has good hydrophilicity, reaction activity, biocompatibility, and biodegradability, which has shown great potentials for biomedical materials. Here, an ultraviolet light-assisted *in situ* synthesis approach is developed to immobilize silver nanoparticles on the surface of sericin gel. The amount of silver nanoparticles immobilized on the surface of sericin gel could be regulated by the irradiation time. The porous structure and property of sericin gel were not affected by the modification of AgNPs, as evidenced by the observation of scanning electron microscopy, X-ray diffractometry, and Fourier transform infrared spectroscopy. Differential scanning calorimetry analysis showed that the modification of AgNPs increased the thermal stability of sericin gel. The growth curve of bacteria and inhibition zone assays suggested that the sericin gel modified with AgNPs had good antimicrobial activities against both Gram-negative and Gram-positive bacteria. This novel sericin has shown a great potential for biomedical purpose.

1. Introduction

Sericin is a protein which is coated on the surface of fibroin fiber when *Bombyx mori* silkworm spins cocoon for protective and adhesive effects [1]. Sericin could be extracted by boiling cocoon in an alkaline solution or through high temperature and high pressure extraction process [2]. Processing of raw silk cocoon produces about 50,000 tons of sericin worldwide each year. However, sericin is usually discarded in silk processing wastewater [3], resulting in a high chemical oxygen demand level of the degumming wastewater [4], which greatly caused the environmental pollution and the waste of resources. Sericin consists of 18 kinds of amino acids. Most of them have strong polar side groups such as hydroxyl, carboxyl, and amino groups. Sericin is characterized by a high serine content which is about one-third of the total amino acids [5]. Because of its good hydrophilicity, reaction activity, biocompatibility, and biodegradability, sericin has been applied widely in biomedical materials such as cell culture scaffolds, drug carriers, and tissue engineering scaffolds [6–9].

In natural conditions, sericin could spontaneously form hydrogel, but its mechanical property is relatively poor. Though the mechanical property of sericin hydrogel could be improved by adding functional cross-linker, the toxicity of the hydrogel may be introduced by the additives. Thus, it is highly desired to develop a green synthesis method without any cross-linking reagent to produce sericin hydrogel with porous structure from aqueous *Bombyx mori* sericin. According to a previous report, sericin gel could be prepared

with certain mechanical properties from soluble sericin through freeze-drying without adding any chemical reagent [10].

Surface immobilization with antibacterial material is a kind of common method to make antibacterial materials. Silver nanoparticle is a regular and effective antibacterial material, which has shown a broad spectrum of antibacterial activity against fungus, viruses, and Gram-positive and Gram-negative bacteria [11–13]. With the development of nanotechnology, numerous nanomaterials with multifarious particular characteristics have been developed [14–18]. Recently, the immobilization of fibroin fiber with nanomaterials for multifunctional textiles has been developed [19, 20]. Some natural polymers such as gelatin, chitosan, and starch immobilized with silver nanoparticle are also developed for antibacterial purpose [21–23]. Sericin gel has the characteristics of porous and good water absorbability, which shows great potentials as scaffold materials, drug carriers, and mouldable wound dressing [24, 25]. Although sericin gel has a lot of unique properties, the protein nature makes it a matrix for bacterial adhesion and thriving, further resulting in its deformation and degradation [26]. Thus, it is necessary to produce a kind of sericin with antibacterial activity for broad application.

Two types of methods have been developed to produce AgNPs-functionalized natural polymers. One is to coat with presynthesized AgNPs on the surface of natural polymer. This method needs the synthesis of silver nanoparticles and composite beforehand on the surface of the polymer, which has been abandoned gradually by researchers due to its complexity and redundancy [8, 27]. Another one is to grow AgNPs *in situ* directly on the surface of natural polymer without the presynthesis step. Silver ions attach on the surface of polymer materials *via* electrostatic adsorption or ion exchange, followed by a reduction step to form AgNPs *in situ*. Natural polymers with small specific surface area and less active group are frequently required to cross-link polymers such as polyamide network polymer and polyacrylic acid to functionalize the surface [28], which will increase the density of AgNPs immobilized on the surface of natural polymers. However, cross-linking functional materials on the surface of natural polymers not only increase the cost, but also likely introduce potential contaminations to environment. Sericin gel has a porous structure, large surface area, and lots of active groups, which may facilitate sericin gel to adsorb more silver nanoparticles on its surface than other natural polymers without cross-linking. Therefore, it is worth developing a facile and green approach to fabricate porous sericin gel modified with silver nanoparticles for antibacterial application.

In previous reports, ultraviolet (UV) light has been applied to assist in the reduction of silver ions to immobilize AgNPs on the surface of polymer [29]. In this study, we developed a synthesis approach to immobilize AgNPs *in situ* on the surface of sericin gel with the assistance of UV. Scanning electron microscopy (SEM), X-ray diffractometry (XRD), Fourier transform infrared spectroscopy (FT-IR), and differential scanning calorimetry (DSC) were applied to characterize the surface tomography and structure of the AgNPs-sericin material. The antimicrobial tests, including growth curve and inhibition zone, were carried out to investigate the antibacterial activities of AgNPs-sericin gel against *Escherichia coli* (*E. coli*) and *Staphylococcus aureus* (*S. aureus*). Our research suggested the AgNPs-sericin gel with porous structure could effectively inhibit the growth of both Gram-negative and Gram-positive bacteria. This novel antibacterial material may be regarded as potential biomedical materials such as wound dressing material.

2. Experimental

2.1. Materials and Chemicals. Silkworm cocoon was kindly provided by the State Key Laboratory of Silkworm Genome Biology, Southwest University, China. Silver nitrate ($AgNO_3$) (AR, 99.99%) was purchased from Aladdin Corporation (Shanghai, China). MilliQ water was used in the experiments. All other chemicals in this study are of analytical grade and directly used without further purification.

2.2. Preparation of Sericin Gel. Sericin was prepared according to a previous reported procedure with minor modification [30]. The details on the extraction of sericin and preparation of sericin gel were described in Figure 1. *Bombyx mori* cocoons were first cut into small pieces and placed into a beaker containing milliQ water. The beaker was autoclaved with a temperature of 121°C and a pressure of 0.1 Mpa for 30 min to dissolve sericin. The aqueous sericin solution was collected and then used to produce sericin powder by freeze-drying. Sericin powder and milliQ water were mixed with appropriate proportion and heated until the powder was completely dissolved. Sericin solution was freezed under −20°C for 2 hours to form sericin hydrogel. Sericin hydrogel was further dehydrated by freeze-drying to become sericin gel.

2.3. UV-Assisted In Situ Synthesis of AgNPs. Sericin gel was cut into small pieces of sheets and then soaked into 50 mM $AgNO_3$ solution. At the same time, sericin gel sheets were irradiated with a 365 nm UV light lamp (24 W) for 10 min, 30 min, and 60 min to make AgNPs on the surface of sericin gel, respectively. Sericin hydrogel, sericin gel, and AgNPs modified sericin gel were shown in Figure 2. AgNPs modified sericin gel was collected and dried at room temperature, which will be used for the following experiments to characterize its surface tomography, structure, and antimicrobial activity.

2.4. Materials Characterization. The surface morphologies of sericin gel and AgNPs modified sericin gel were imaged using SEM (JCM-5000, JEOL, Tokyo, Japan). Sericin gel and AgNPs modified sericin gel were pressed to form compact pellets for FT-IR measurements (Nicolet iz10, Thermo Electronic Corporation, USA). DSC curves were recorded on a differential scanning calorimetry instrument (DSC 200PC, Nestal, Germany) in the temperature range of 50–400°C. The heating rate for the test was kept at 10°C/min and the flow rate of nitrogen gas was controlled at 50 mL/min. XRD spectra of the

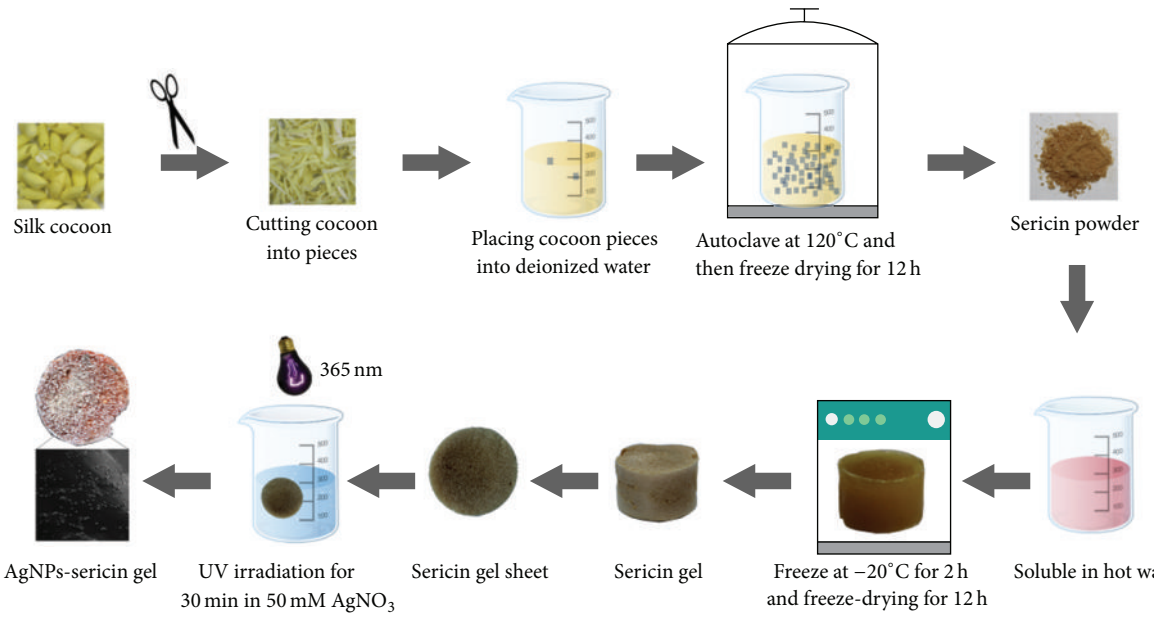

Figure 1: A flowchart illustrates the procedure of the extraction and fabrication of sericin, sericin hydrogel, sericin gel, and AgNPs-sericin gel.

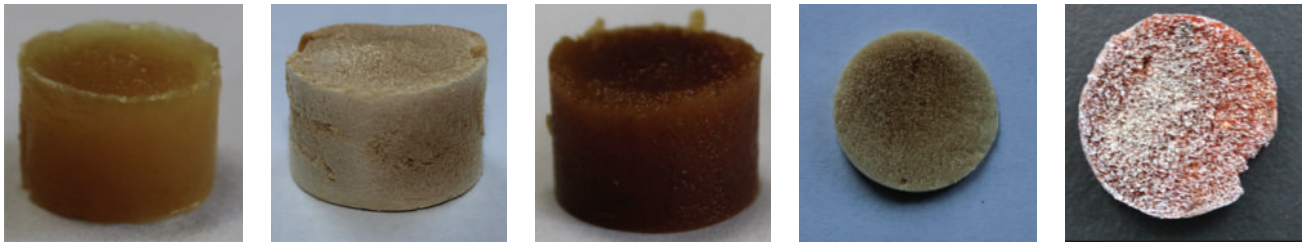

Figure 2: Photographs of the topology of hydrogel prepared from 8% sericin solution (wt): sericin hydrogel (a), sericin gel (b), rehydrated sericin hydrogel (c), sericin gel sheet (d), and AgNPs modified sericin gel (e).

modified and unmodified sericin gel were examined with a 2θ range of 10–70° (X'Pert Powder, PANalytical, Netherlands).

2.5. Analysis of Growth Curve. *E. coli* and *S. aureus* at lag phase were inoculated into 10 mL Luria-Bertani (LB) medium (pH 7.4) in the absence and presence of the modified or unmodified sericin gel and then cultured with constant shaking speed (220 rpm) at 37°C. The optical density at 600 nm (OD_{600}) was measured for 0.5 mL bacterial collected at different intervals. All growth curve tests were made in triplicate to ensure the reproducibility of the tests.

2.6. Inhibition Zone Assay. *E. coli* and *S. aureus* were inoculated in 100 mL LB medium (pH 7.4) and cultured with constant shaking speed (220 rpm) at 37°C for 12 hours, respectively. Bacteria at lag phase were spread uniformly on agar medium plates; then a circular sericin or AgNPs modified sericin gel with diameter of about 1.05 cm was introduced into the plate. After incubation at 37°C overnight, the antibacterial activities of the samples were evaluated based on the diameter of the bacterial inhibition zones. All antibacterial activity tests were done in triplicate to ensure that the assay could be reproduced.

3. Results and Discussion

Sericin gel has lots of hydroxyl and active groups on its surface, which may react with silver ions under the irritation of UV to form AgNPs on the surface of sericin gel [31, 32]. In this work, we developed a facile, green, efficient, and fast approach to fabricate the AgNPs modified sericin gel without addition of any chemicals. The procedures were shown in Figure 1. Sericin concentration may affect the formation of sericin hydrogel from sericin solution. Here, we tested different sericin concentrations and finally determined an appropriate sericin concentration.

Sericin gel has a porous structure and good water absorbency, as shown in Figure 2. Figure 2(a) shows the morphology of sericin hydrogel prepared from 8% sericin solution (wt). After freeze-drying, sericin hydrogel was

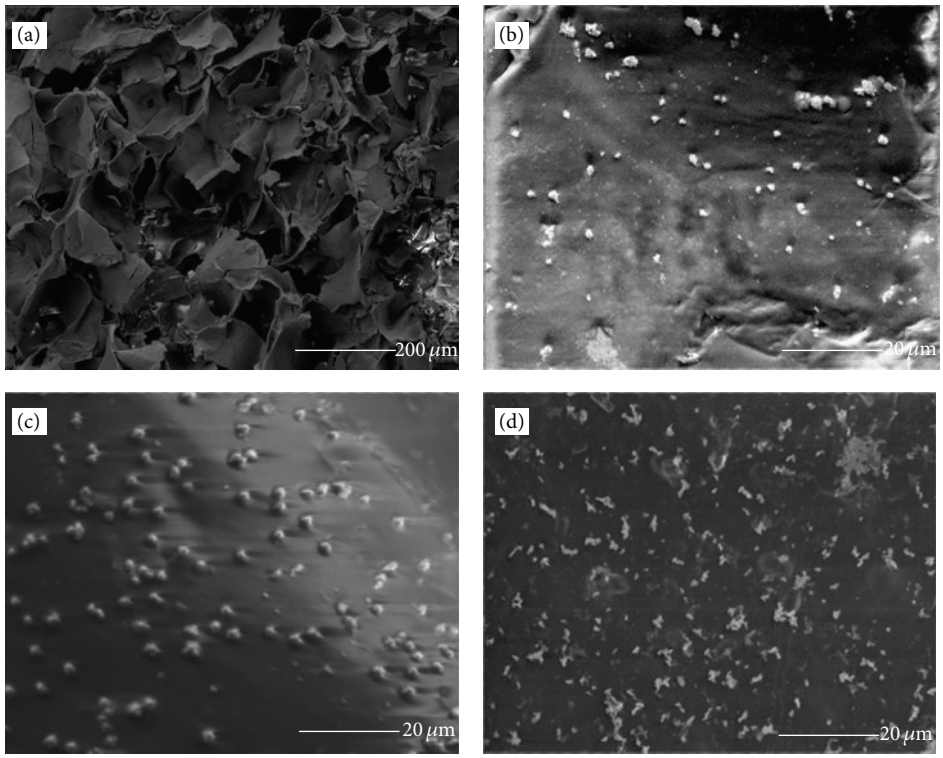

Figure 3: SEM images of sericin gel (a) and sericin gel in silver nitrate solution irradiated with a UV light for 10 min (b), 30 min (c), and 60 min (d).

dehydrated to form dry sericin gel with porous structure (Figure 2(b)). The dry gel reformed hydrogel once absorbing water (Figure 2(c)). Figure 2(d) showed a small piece of sericin gel with porous structure. The AgNPs modified sericin gel was shown in Figure 2(e). This result suggested that sericin could form sericin gel with certain mechanical property without addition of any chemicals, and this kind of sericin gel had porous structure, good permeability, moisture retention, and hygroscopicity. In addition, it was environmentally friendly, biocompatible, and biodegradable. In wound healing process, infection, dehydration, and secondary trauma are key barriers. Therefore, sericin gel is regarded as a potential material for biomedical purpose such as wound dressing material.

SEM was carried out to observe the surface morphologies of the sericin gel before and after the immobilization of AgNPs (Figure 3). As shown in Figure 3(a), sericin gel has a porous structure, which may favor effective nutrient or gas exchange for cells in the process of wound healing [33]. Some small dots on the surface of sericin gel had been observed after an irradiation for 10 min in a silver nitrate solution (Figure 3(b)), indicating a high-efficiency *in situ* synthesis of AgNPs on the surface of sericin gel. As the irradiation time increases to 30 minutes and 60 minutes, the amount of AgNPs on the surface of sericin gel greatly increases (Figures 3(c) and 3(d)). The results showed that *in situ* synthesis of AgNPs on the surface of sericin gel could be regulated by the irradiation time.

Further, XRD was applied to verify *in situ* synthesis of AgNPs on the surface of sericin gel. XRD patterns of unmodified and AgNPs modified sericin gel were shown in Figure 4. In a previous study, two characteristic peaks located at $2\theta = 19.2^\circ$ and $2\theta = 23.2^\circ$ are observed [5]. There was no significant change observed on the peaks of sericin gel with the modification of AgNPs, which suggested that UV irradiation and AgNPs modification did not alter the structure of sericin gel. For the sericin gel modified with AgNPs, one peak was observed at the position of $2\theta = 37.5^\circ$, which may be attributed to the (111) plane of the face-centered cubic structure of AgNPs [34].

FT-IR was widely used to characterize the structure of materials [35]. Here, it was applied to investigate the effect of AgNPs modification on the structure of sericin gel. As shown in Figure 5, there were four characteristic peaks at 3280 cm^{-1}, 1620 cm^{-1}, 1520 cm^{-1}, and 1228 cm^{-1} on both the unmodified sericin gel and the AgNPs modified sericin gel, which may be attributed to the N-H stretching absorption, Amid III, Amid II, and Amid I peaks of sericin [36, 37]. Since the growth of AgNPs on the surface of sericin gel had no influence on the amid peaks of sericin gel, it may suggest that the synthesis procedures did not affect the structure of sericin gel. Amino acid groups on the surface of sericin gel may play an important role in the adsorption and reduction of silver ions. The coordination of nitrogen atom interaction with silver ions may promote the reduction of silver ions [38]. The porous structure of sericin gel will favor the interaction of amino acid residues with silver ions, thus promoting more and more AgNPs to be hybridized on the surface of sericin gel.

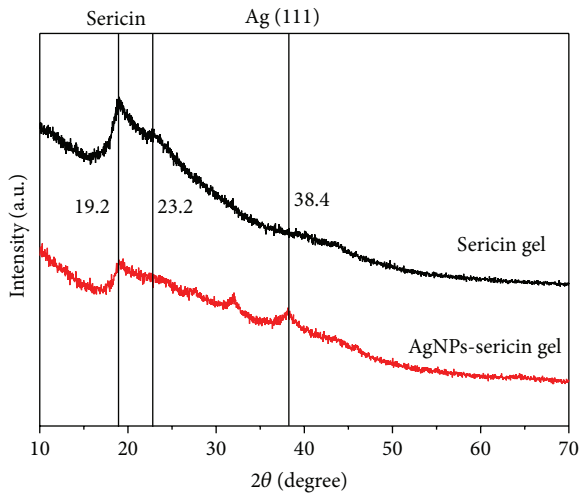

Figure 4: XRD patterns of sericin gel and AgNPs-sericin gel.

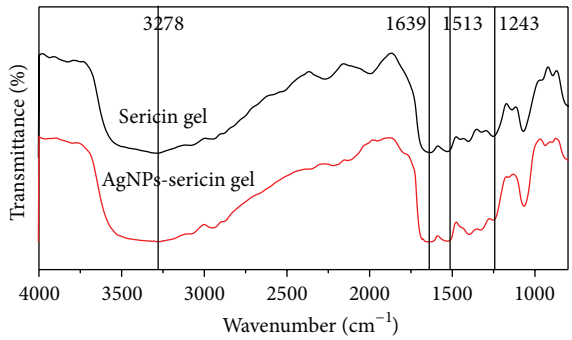

Figure 5: FT-IR spectra of sericin gel and AgNPs-sericin gel.

To further investigate the effects of silver nanoparticles on the structure of sericin gel, the thermal behavior of sericin gel with and without the modification of AgNPs was analyzed by DSC. Figure 6 showed the DSC curves of unmodified and modified sericin gel after freeze-drying. In the case of sericin gel, there were two endothermic peaks at 218°C and 316°C, respectively, which were corresponding to the thermally induced molecular motion of an amorphous region and thermal decomposition of sericin [39]. The DSC curve of sericin gel modified with AgNPs showed that the first endothermic peak slightly shifted to about 223°C, and the second peak shifted to about 381°C. This result suggested that AgNPs modification on the sericin gel may increase the thermostability of sericin gel.

Bacterial growth curves were used to assess the antimicrobial activities of sericin gel and AgNPs modified sericin gel. In this study, *E. coli* and *S. aureus* were chosen as a model of Gram-positive bacteria and Gram-negative bacteria to assess the antibacterial activities of AgNPs modified sericin gel, respectively [40]. As shown in Figure 7, the unmodified sericin gel did not affect the growth of bacteria. However, the AgNPs modified sericin gel showed significantly antibacterial activities against *E. coli* and *S. aureus*. In the presence of AgNPs modified sericin gel, the lag phase of bacteria

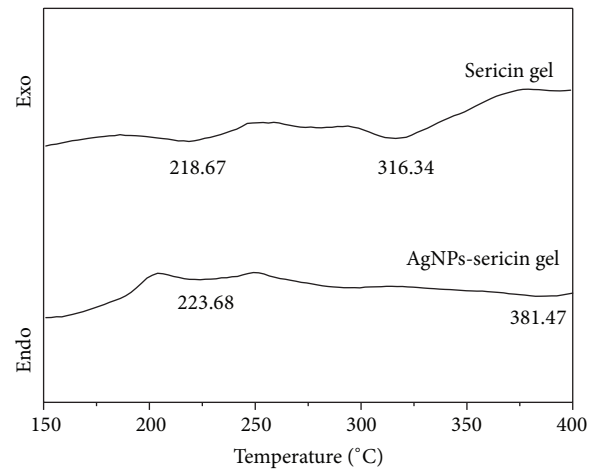

Figure 6: DSC curves of sericin gel and AgNPs-sericin gel.

Table 1: Diameters of inhibition zones of sericin gel and AgNPs-sericin gel against *E. coli* and *S. aureus*. Zone of inhibition is expressed as the diameter in centimeters. Values represent means ± standard deviations of inhibition zones from three individual experiments.

	Control	10 min	30 min	60 min
E. coli	1.05 ± 0.02	1.54 ± 0.01	1.59 ± 0.04	1.79 ± 0.02
S. aureus	1.05 ± 0.02	1.44 ± 0.02	1.48 ± 0.01	1.60 ± 0.01

extended more than 12 h. Our previous result had suggested increasing reaction time facilitated the formation of AgNPs on the surface of sericin gel. This result indicated sericin gel modified with AgNPs for 10 min had already shown significantly antibacterial activities. Further increasing the reaction time had a little effect on the increase of antibacterial activity.

Besides the growth curves, inhibition zone is also regarded as an important indicator for antibacterial activities. The inhibition zone of sericin gel and AgNPs modified sericin gel was shown in Figure 8. No inhibition zone was observed for unmodified sericin gel on both *E. coli* and *S. aureus* agar plates, indicating that sericin gel did not affect the growth of bacteria on the plate. On the contrary, obvious inhibition zone could be observed in the presence of sericin gel modified with AgNPs for 10 min, 30 min, and 60 min. The diameters of the inhibition zones are summarized in Table 1. The result suggested that increasing the reaction time could increase the antibacterial activities of AgNPs modified sericin gel, which was consistent with the assay of the growth curves.

4. Conclusions

In this study, we developed a green and efficient approach to synthesize silver nanoparticles *in situ* on the surface of sericin gel. The porous structure of sericin gel will not be affected by the modification of AgNPs, as confirmed by SEM, XRD, and FT-IR. DSC analysis showed that the modification of AgNPs also increased the thermal stability

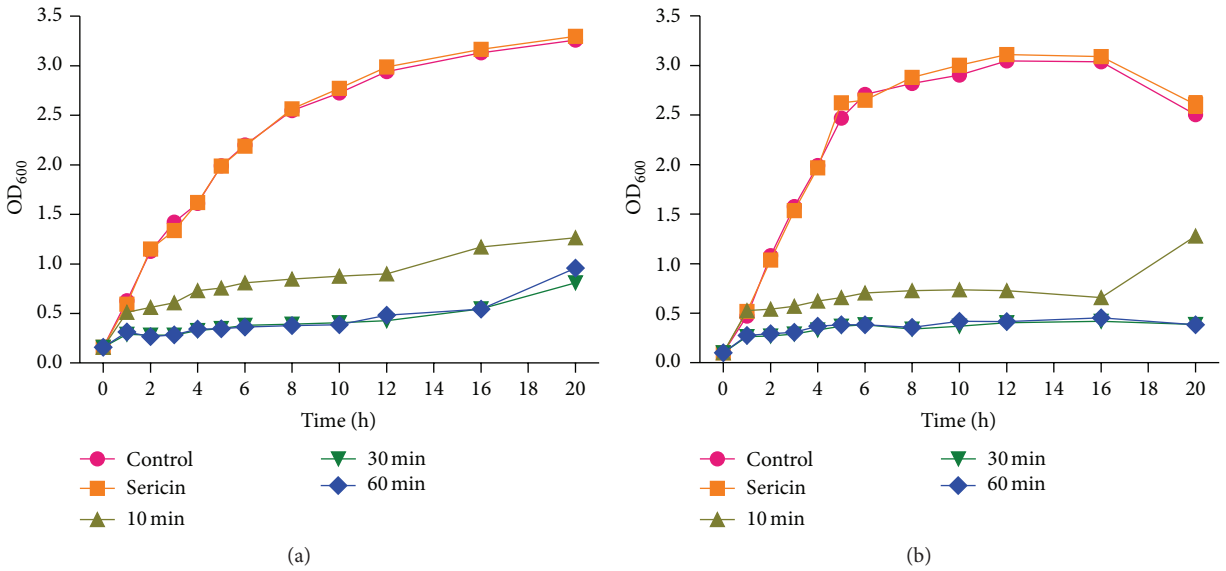

Figure 7: Growth curves of *E. coli* (a) and *S. aureus* (b) in the presence of sericin gel and sericin gel modified with AgNPs for 10 min, 30 min, and 60 min.

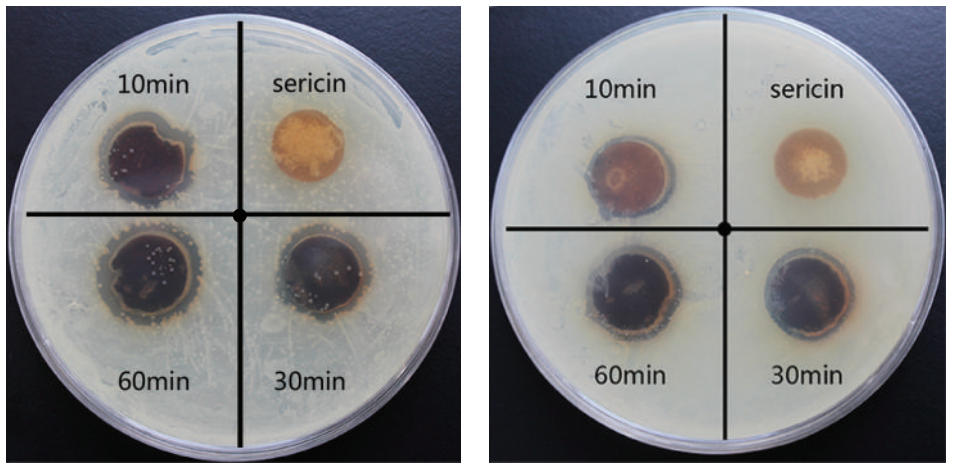

Figure 8: Inhibition zone assay of sericin gel and AgNPs-sericin gel against *E. coli* (a) and *S. aureus* (b).

of sericin gel. Sericin gel immobilized with AgNPs showed good antimicrobial activities against both Gram-negative and Gram-positive bacteria. This novel material shows a great potential for biomedical application.

Competing Interests

The authors declare that they have no competing interests.

Acknowledgments

This work was supported by the National Basic Research Program of China (no. 2012CB114602), State Key Program of National Natural Science of China (no. 31530071), National Natural Science Foundation of China (nos. 31572465 and 31402139), the Open Project Program of National Laboratory of Biomacromolecules (no. 2015kf08), and the Fundamental Research Funds for the Central Universities (SWU112111, SWU112086, XDJK2013A019, and XDJK2013C049), and Chongqing Fundamental and Frontier Research Program (cstc2013jcyjA0500).

References

[1] M. Sasaki, H. Yamada, and N. Kato, "Consumption of silk protein, sericin elevates intestinal absorption of zinc, iron, magnesium and calcium in rats," *Nutrition Research*, vol. 20, no. 10, pp. 1505–1511, 2000.

[2] P. Aramwit, S. Damrongsakkul, S. Kanokpanont, and T. Srichana, "Properties and antityrosinase activity of sericin from various extraction methods," *Biotechnology and Applied Biochemistry*, vol. 55, no. 2, pp. 91–98, 2010.

[3] Y.-Q. Zhang, "Applications of natural silk protein sericin in biomaterials," *Biotechnology Advances*, vol. 20, no. 2, pp. 91–100, 2002.

[4] C. Fabiani, M. Pizzichini, M. Spadoni, and G. Zeddita, "Treatment of waste water from silk degumming processes for protein recovery and water reuse," *Desalination*, vol. 105, no. 1-2, pp. 1–9, 1996.

[5] W. Tao, M. Li, and R. Xie, "Preparation and structure of porous silk sericin materials," *Macromolecular Materials and Engineering*, vol. 290, no. 3, pp. 188–194, 2005.

[6] B. C. Dash, B. B. Mandal, and S. C. Kundu, "Silk gland sericin protein membranes: fabrication and characterization for potential biotechnological applications," *Journal of Biotechnology*, vol. 144, no. 4, pp. 321–329, 2009.

[7] B. B. Mandal, A. S. Priya, and S. C. Kundu, "Novel silk sericin/gelatin 3-D scaffolds and 2-D films: fabrication and characterization for potential tissue engineering applications," *Acta Biomaterialia*, vol. 5, no. 8, pp. 3007–3020, 2009.

[8] A. Nishida, M. Yamada, T. Kanazawa, Y. Takashima, K. Ouchi, and H. Okada, "Use of silk protein, sericin, as a sustained-release material in the form of a gel, sponge and film," *Chemical and Pharmaceutical Bulletin*, vol. 58, no. 11, pp. 1480–1486, 2010.

[9] T. Siritientong, T. Srichana, and P. Aramwit, "The effect of sterilization methods on the physical properties of silk sericin scaffolds," *AAPS PharmSciTech*, vol. 12, no. 2, pp. 771–781, 2011.

[10] H. Zhang, M. Yang, S. Min, Q. Feng, X. Gao, and L. Zhu, "Preparation and characterization of a novel spongy hydrogel from aqueous *Bombyx mori* sericin," *e-Polymers*, vol. 8, no. 1, pp. 761–770, 2008.

[11] S. Eckhardt, P. S. Brunetto, J. Gagnon, M. Priebe, B. Giese, and K. M. Fromm, "Nanobio silver: its interactions with peptides and bacteria, and its uses in medicine," *Chemical Reviews*, vol. 113, no. 7, pp. 4708–4754, 2013.

[12] J. A. Lemire, J. J. Harrison, and R. J. Turner, "Antimicrobial activity of metals: mechanisms, molecular targets and applications," *Nature Reviews Microbiology*, vol. 11, no. 6, pp. 371–384, 2013.

[13] I. Sondi and B. Salopek-Sondi, "Silver nanoparticles as antimicrobial agent: a case study on *E. coli* as a model for Gram-negative bacteria," *Journal of Colloid and Interface Science*, vol. 275, no. 1, pp. 177–182, 2004.

[14] J. B. Addison, T. M. O. Popp, W. S. Weber, J. S. Edgerly, G. P. Holland, and J. L. Yarger, "Structural characterization of nanofiber silk produced by embiopterans (webspinners)," *RSC Advances*, vol. 4, no. 78, pp. 41301–41313, 2014.

[15] Z. S. Lu and C. M. Li, "Quantum dot-based nanocomposites for biomedical applications," *Current Medicinal Chemistry*, vol. 18, no. 23, pp. 3516–3528, 2011.

[16] Y. Kuang, D. Yuan, Y. Zhang, A. Kao, X. Du, and B. Xu, "Interactions between cellular proteins and morphologically different nanoscale aggregates of small molecules," *RSC Advances*, vol. 3, no. 21, pp. 7704–7707, 2013.

[17] Y. D. Lei, T. X. Wang, J. W. Mitchell, L. Zaidel, J. Qiu, and L. Kilpatrick-Liverman, "Bioinspired amphiphilic phosphate block copolymers as non-fluoride materials to prevent dental erosion," *RSC Advances*, vol. 4, no. 90, pp. 49053–49060, 2014.

[18] S. Das, P. K. S. Magut, S. L. De Rooy, F. Hasan, and I. M. Warner, "Ionic liquid-based fluorescein colorimetric pH nanosensors," *RSC Advances*, vol. 3, no. 43, pp. 21054–21061, 2013.

[19] M. Meng, H. He, J. Xiao, P. Zhao, J. Xie, and Z. Lu, "Controllable in situ synthesis of silver nanoparticles on multilayered film-coated silk fibers for antibacterial application," *Journal of Colloid and Interface Science*, vol. 461, pp. 369–375, 2016.

[20] Z. Lu, J. Xiao, Y. Wang, and M. Meng, "*In situ* synthesis of silver nanoparticles uniformly distributed on polydopamine-coated silk fibers for antibacterial application," *Journal of Colloid and Interface Science*, vol. 452, pp. 8–14, 2015.

[21] P. Kanmani and J.-W. Rhim, "Physicochemical properties of gelatin/silver nanoparticle antimicrobial composite films," *Food Chemistry*, vol. 148, pp. 162–169, 2014.

[22] V. Thomas, M. M. Yallapu, B. Sreedhar, and S. K. Bajpai, "Fabrication, characterization of chitosan/nanosilver film and its potential antibacterial application," *Journal of Biomaterials Science, Polymer Edition*, vol. 20, no. 14, pp. 2129–2144, 2009.

[23] S. M. Yakout and A. A. Mostafa, "A novel green synthesis of silver nanoparticles using soluble starch and its antibacterial activity," *International Journal of Clinical and Experimental Medicine*, vol. 8, no. 3, pp. 3538–3544, 2015.

[24] M. Yang, Y. Shuai, G. Zhou, N. Mandal, L. Zhu, and C. Mao, "Tuning molecular weights of *Bombyx mori* (*B. mori*) silk sericin to modify its assembly structures and materials formation," *ACS Applied Materials and Interfaces*, vol. 6, no. 16, pp. 13782–13789, 2014.

[25] G. H. Altman, F. Diaz, C. Jakuba et al., "Silk-based biomaterials," *Biomaterials*, vol. 24, no. 3, pp. 401–416, 2003.

[26] H.-J. Jin, J. Park, R. Valluzzi, P. Cebe, and D. L. Kaplan, "Biomaterial films of *Bombyx mori* silk fibroin with poly(ethylene oxide)," *Biomacromolecules*, vol. 5, no. 3, pp. 711–717, 2004.

[27] J.-M. Lee, J.-E. Yu, and Y.-S. Koh, "Experimental study on the effect of wavelength in the laser cleaning of silver threads," *Journal of Cultural Heritage*, vol. 4, no. 1, pp. 157–161, 2003.

[28] D. S. Zhang, G. W. Toh, H. Lin, and Y. Y. Chen, "*In situ* synthesis of silver nanoparticles on silk fabric with PNP for antibacterial finishing," *Journal of Materials Science*, vol. 47, no. 15, pp. 5721–5728, 2012.

[29] X. M. Wang, W. R. Gao, S. P. Xu, and W. Q. Xu, "Luminescent fibers: *in situ* synthesis of silver nanoclusters on silk via ultraviolet light-induced reduction and their antibacterial activity," *Chemical Engineering Journal*, vol. 210, pp. 585–589, 2012.

[30] J.-H. Wu, Z. Wang, and S.-Y. Xu, "Preparation and characterization of sericin powder extracted from silk industry wastewater," *Food Chemistry*, vol. 103, no. 4, pp. 1255–1262, 2007.

[31] Y. Shin, I.-T. Bae, and G. J. Exarhos, "'Green' approach for self-assembly of platinum nanoparticles into nanowires in aqueous glucose solutions," *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, vol. 348, no. 1–3, pp. 191–195, 2009.

[32] N. Vigneshwaran, R. P. Nachane, R. H. Balasubramanya, and P. V. Varadarajan, "A novel one-pot 'green' synthesis of stable silver nanoparticles using soluble starch," *Carbohydrate Research*, vol. 341, no. 12, pp. 2012–2018, 2006.

[33] S. Y. Park, C. S. Ki, Y. H. Park, H. M. Jung, K. M. Woo, and H. J. Kim, "Electrospun silk fibroin scaffolds with macropores for bone regeneration: an *in vitro* and *in vivo* study," *Tissue Engineering Part A*, vol. 16, no. 4, pp. 1271–1279, 2010.

[34] X. Zou, E. Ying, and S. Dong, "Preparation of novel silver-gold bimetallic nanostructures by seeding with silver nanoplates and application in surface-enhanced Raman scattering," *Journal of Colloid and Interface Science*, vol. 306, no. 2, pp. 307–315, 2007.

[35] C. S. Ki, J. W. Kim, H. J. Oh, K. H. Lee, and Y. H. Park, "The effect of residual silk sericin on the structure and mechanical property of regenerated silk filament," *International Journal of Biological Macromolecules*, vol. 41, no. 3, pp. 346–353, 2007.

[36] X. M. Zhang and P. Wyeth, "Using FTIR spectroscopy to detect sericin on historic silk," *Science China Chemistry*, vol. 53, no. 3, pp. 626–631, 2010.

[37] M. Nagura, R. Ohnishi, Y. Gitoh, and Y. Ohkoshi, "Structures and physical properties of cross-linked sericin membranes," *Journal of Sericultural Science of Japan*, vol. 70, no. 2, pp. 149–153, 2001.

[38] A. Frattini, N. Pellegri, D. Nicastro, and O. De Sanctis, "Effect of amine groups in the synthesis of Ag nanoparticles using aminosilanes," *Materials Chemistry and Physics*, vol. 94, no. 1, pp. 148–152, 2005.

[39] K. Lee, H. Kweon, J. H. Yeo et al., "Effect of methyl alcohol on the morphology and conformational characteristics of silk sericin," *International Journal of Biological Macromolecules*, vol. 33, no. 1–3, pp. 75–80, 2003.

[40] S. A. Khan, A. Ahmad, M. I. Khan et al., "Antimicrobial activity of wool yarn dyed with *Rheum emodi* L. (Indian Rhubarb)," *Dyes and Pigments*, vol. 95, no. 2, pp. 206–214, 2012.