Liuyang Zuo,
Feng Zhang,
Bing Gao,
Baoqi Zuo*

Fabrication of Electrical Conductivity and Reinforced Electrospun Silk Nanofibers with MWNTs

DOI: 10.5604/01.3001.0010.1687

Department of Soochow University,
National Engineering Laboratory for Modern Silk,
Ren ai road 199, Suzhou, China
* E-mail: bqzuo@suda.edu.cn

Abstract
Electrospinning is an effective technique for fabricating submicron to nanoscale fibers from synthetic polymer as well as natural proteins. In this study, multiwalled carbon nanotubes (MWNTs) were embedded via electrospinning by adding MWNTs into the spinning dope, and found to be well aligned along the fiber axis in the silk fibroin nanofibers. The morphology and microstructure of the electrospun nanofibers were characterised using a field emission scanning electron microscope (FESEM) and Transmission electron microscopy (TEM). X-ray diffraction (XRD) and TG-DTA were used to study the crystal structure of the silk/MWNTs composite nanofibres, carried out to alter the strength, toughness and electrical conductivity of silk nanofibers by adding a small amount of MWNTs. The electrospun random silk mats with 1% MWNTs had a Young's modulus, ultimate tensile strength and strain of 107.46 ± 9.15MPa, 9.94 ± 1.2MPa and 9.25 ± 1.5%, respectively, and electrical conductivity increased to 1.2×10^{-4}S/cm. The silk/MWNTs composite nanofibres could potentially be applied in nerve repair materials owing to their excellent mechanical properties and electrical conductivity.

Key words: *silk, MWNTs, electrospinning, reinforce electrical conductivity.*

Introduction

Bombyx mori silk, a protein-based polymer with unique structural and mechanical properties, has been used as textile materials for a long time [1-6]. Degummed bombyx mori silk, which only consists of silk fibroin, can be reprocessed in various forms, such as porous film, electrospinning silk nanofibers and gel. Electrospinning is a simple method for producing nanofibers from both natural [7] and synthetic polymers, with diameters ranging from 2 nm to several micrometers. Electrospun random silk mats, with their good biocompatibility, high specific surface area and high porosity, are ideal candidates for biomedical applications, including tissue-engineered scaffolds, wound dressings and drug delivery [2-6]. However, the mechanical properties of electrospun silk fibers are lacking in comparison with natural silk fiber because regenerated silk fibroin changes some of the structure during reprocessing. Introducing a reinforced agent to electrospun silk fibers is a good way to alter the mechanical performance.

Carbon nanotubes (CNTs) as typical reinforcing agents have attracted considerable attention on account of their unique mechanical and electrical properties since Dr. Iijima invented them in 1991 [8]. Introducing a little amount of MWNTs in the form of small bundles or individual nanotubes can dramatically improve the mechanical strength, such as polypropylene [9], polyamide [10], polyacrylonitrile [11] and so on. Wei K [12] introduced a small amount of MWNTs in electrospun silk fibers, and clearly increased the strength. Viviana Lovat [13] demonstrated the possibility of using carbon nanotubes (CNTs) as potential devices able to improve neural signal transfer while supporting dendrite elongation and cell adhesion. The electrical conductivity of polymers, however, ranges from 10^{-14} to 10^{-17} S/cm. Polymeric materials are composed of carbon fillers to improve their electrical conductivity, such as carbon fibers, carbon black and synthetic graphite [14]. Several scholars have focused on fabricating polymer composite nanofibres using MWNTs as a filler. J.S. Jeong [15] produced conductive MWNTs/nylon composites and improved their electrical conductivity.

In this study, we fabricated silk/MWNTs composite nanofibres using the electrospinning technique. The effect of MWNTs on the morphology, crystal structure, mechanical properties and electrical conductivity of electrospun silk nanofibres were investigated. The silk/MWNTs composite nanofibres can be potentially applied in nerve repair materials owing to their excellent mechanical properties and electrical conductivity. As a developing country Serbia is in a constant race with the world economy in all spheres of industries, not to mention the textile industry. The poor financial situatment and evaluation of social, psychological, philosophical, economic and other aspects, as key links between science and economy.

Materials and experiment

Bombyx mori silk were kindly supplied by Gains cocoon silk co., LTD, China. All reagents were purchased from Sigma Aldrich, unless otherwise mentioned, and all concentrations were weight to weight (w/w).

MWNTs (JEIO Co., Korea) used in this paper were 1 μm in length and 15~20 nm in diameter, synthesised by the thermal chemical vapour deposition (CVD) method, and their purity was 97%. They were treated with 3 M HNO_3 and then flushed in 5 M HCl to obtain pure MWNTs. A required amount of purified MWNTs was dispersed in formic acid containing sodium dodecysulphate (SDS), whose ratio of weight between the SDS and MWNTs was 1:1. An ultrasonic generator (Kyungill Ultrasonic, Korea) with a nominal frequency of 28 kHz and power of 600 W was used for treatment for 2 h.

Bombyx mori silk was degummed twice with 0.05% Na_2CO_3 (w%) solution at 100 °C for 1 h, and rinsed thoroughly in warm distilled water to remove the glue-like sericin. A new method of dissolving silk was adopted in this paper. Degummed silk was first dissolved in 98% formic acid with 4% $CaCl_2$ for 1 h to obtain an 8% silk fibroin solution. Then

Zuo L, Zhang F, Gao B, Zuo B. Fabrication of Electrical Conductivity and Reinforced Electrospun Silk Nanofibers with MWNTs.
FIBRES & TEXTILES in Eastern Europe 2017; 25, 3(123): 40-44. **DOI:** 10.5604/01.3001.0010.1687

the solution obtained was cast onto a polystyrene plate and the solvent evaporated at room temperature [14].

Here, MWNTs were dispersed in a required amount of formic acid by ultrasound for 2 h to obtain a stable dispersed MWNT solution. Then a spinning dope was prepared by dissolving the regenerated silk film in the MWNT solution and mechanically stirring for 2 h to obtain an 8% spinning dope with several amounts of MWNTs. The blended solution was transferred to a 10 ml syringe with an 18-G needle. The flow rate was 0.6 ml/h, and the voltage was gradually increased to 18 kV. Random nanofibres got deposited on the collection plate. The distance between the transmitting and receiving electrodes was 15 cm and the experiment was carried out in the conditions of 30% relative humidity and 20 °C temperature.

Result and discussion

Images of electrospun fibers were obtained using a field emission scanning electron microscope (FESEM S-4200, Hitachi, Japan). SEM images of the silk/MWNT composites with different concentrations of MWNTs are shown in ***Figure 1.a-1.d***, demonstrating that nanoscale fibers can be fabricated by adding a small amount of MWNTs, and that the nanofibers spun are oriented randomly, whose average diameter is about 85 nm. The diameter was measured with Image-Pro Plus 6.0 software. Pictures of the MWNTs are shown in ***Figure 1.e*** and ***1.f***. The diameter of the MWNTs is about 20 nm, with some tangled up. The diameters of nanofibres show no obvious change and there is little entanglement between the nanofibres with an increase in MWNT content. With an increase in the MWNT content, it is easy for MWNTs to get tangled in the spinning dope, and in consequence the appearance of nanofibres is slightly coarse in ***Figure 1.d***, compared with others. For the spinning dope, an increase in the apparent viscosity increases the surface tension of the solution, which is also a conceivable reason for the result (***Figure 2,*** see page 42).

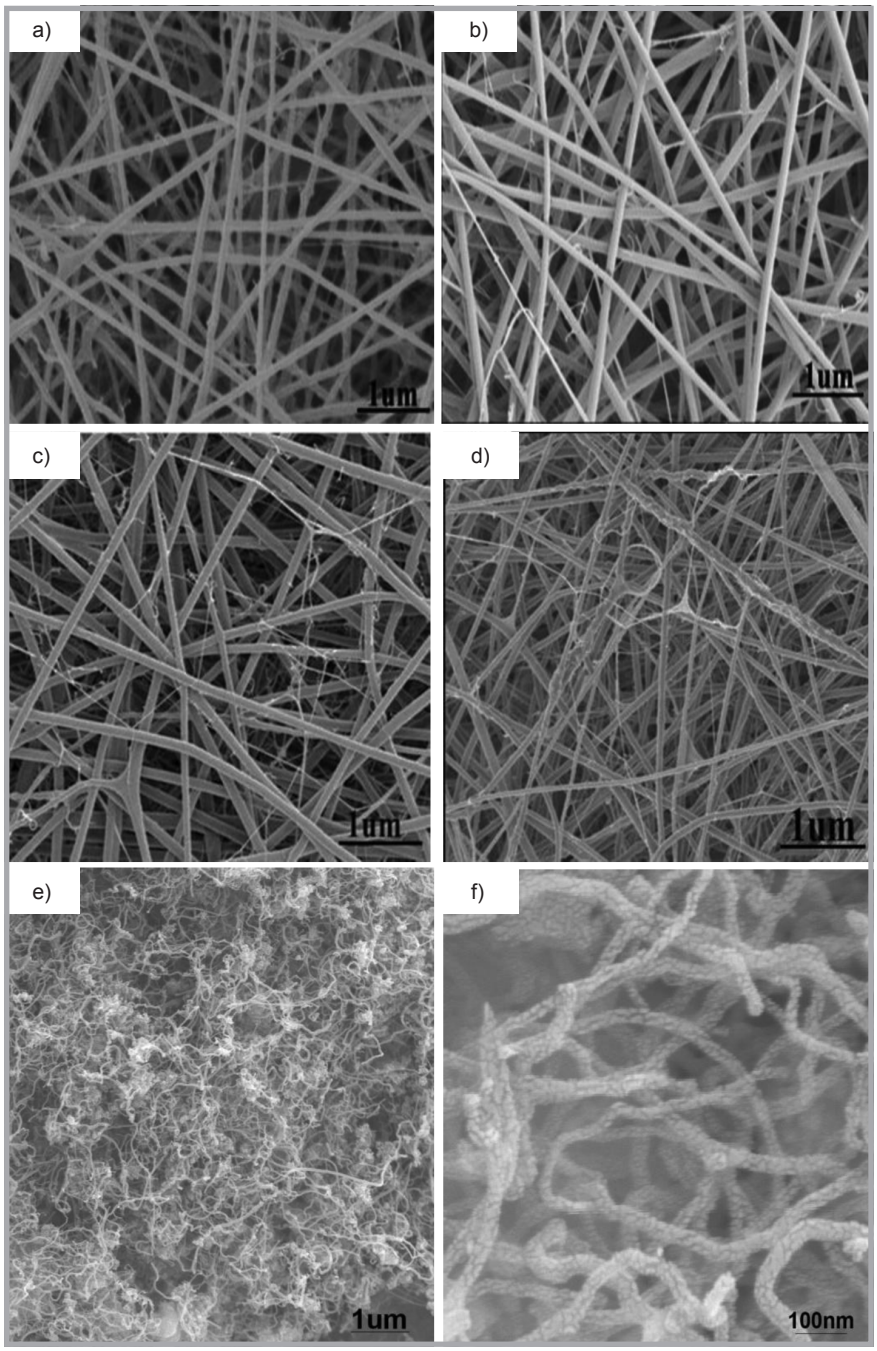

Figure 1. *SEM images of the MWNTs/silk composites with several amounts of MWNTs in the spinning solution: a) with 0.5% MWNTs, b) with 1.0% MWNTs, c) with 1.5% MWNTs, d) with 2.0% MWNTs, e) and (f) MWNTs.*

Table 1. *Diameters of MWNTs/silk nanofiber.*

Fibers with MWNTs, w%	Average diameter, nm
0.5	86
1.0	105
1.5	105
2.0	80

Transmission electron microscopy (TEM) was performed using a Philips CM 200 unit, operated at an acceleration voltage of 120 kV. TEM was used to investigate the internal structural details of the MWNTs/silk composites [18]. ***Figure 3*** (see page 42) shows TEM images of MWNTs/silk spun nanofibers containing 0.5, 1.0, 1.5 and 2.0 wt.% MWNTs, respectively. The TEM images showed that MWNTs were embedded and well aligned along the fiber axis. Furthermore, it should be noted that the MWNTs appear to be well dispersed within the individual nanofibers after the electrospinning process. Some entangled MWNTs appear in ***Figure 3.d***, since a fluid blocked the outlet spinneret as a result of the high concentration of MWNTs in the fluid.

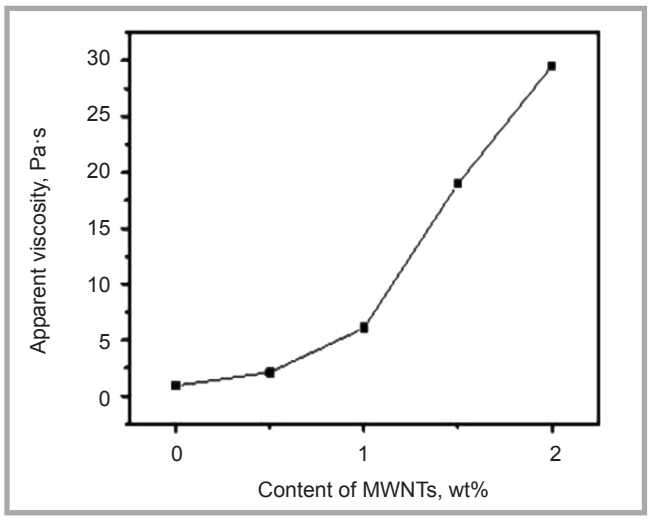

Figure 2. *Relationship between the apparent viscosity of the spinning dope and content of MWNTs.*

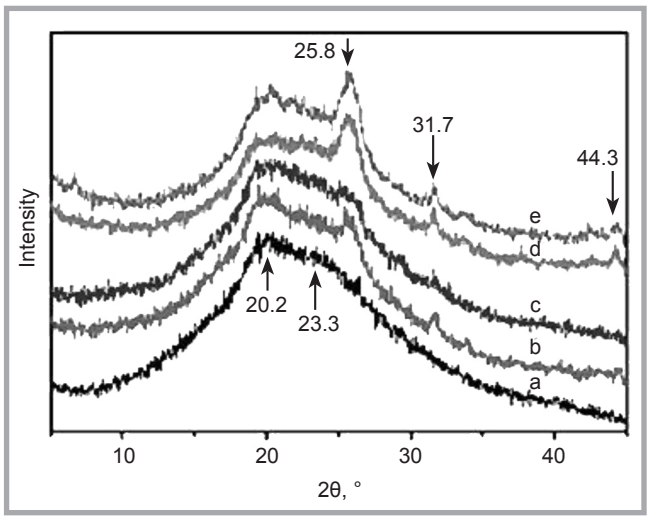

Figure 4. *XRD patterns of MWNT – reinforced and as-spun silk nanofibers: a) electrospun silk nanofibers, b) 0.5% MWNT reinforced nanofibers, c) 1.0% MWNT reinforced nanofibers, d) 1.5% MWNT reinforced nanofibers, and e) 2.0% MWNT reinforced nanofibers.*

An X-ray diffraction (XRD, PAN analytical X' Pert Pro M PD System, The Netherlands,is a useful tool for the investigation of the secondary structure of proteins, and it has already given important insights into the structure of native silk filaments of both spiders and silkworms [18]. As it is known that the secondary structures of silk proteins have the characteristics of silk I, silk II and random coil structures [19].

Figure 4 shows the X-ray diffractograms of five samples reinforced with nanofibers of 0.5%, 1.0%, 1.5% and 2.0% MWNTs and pure electrospun silk nanofiber. All samples show a characteristic peak at 20.2° and 23.3°, indicating a β-sheet structure. We noticed that the electrospun fibers showed a very weak peak at 25.8° (silk I), getting stronger with an increase in MWNT content. Indeed a series of new peaks appeared simultaneously in the samples containing MWNTs at 31.7°and 44.3° which belonged to the MWNTs, indicating that some molecular bonds may connect silk and MWNTs. However, the similar shape of the four curves indicated that increasing the MWNT content had a limited influence on the structure of the electrospun silk fibers.

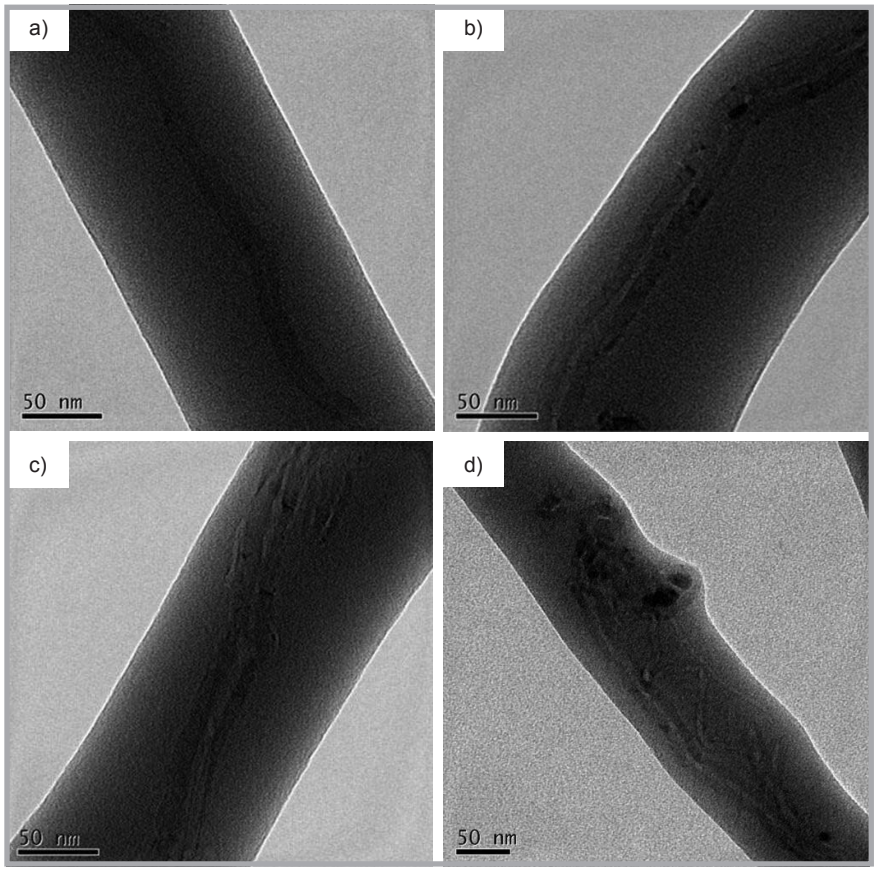

Figure 3. *TEM images of the MWNTs/silk composites with several amounts of MWNTs in the spinning solution: a) with 0.5% MWNTs, b) with 1.0% MWNTs, c) with 1.5% MWNTs, d) with 2.0% MWNTs.*

The thermal performance of the composites was examined by means of a TG-DTA (PE-S II) (PE company of America). There are five DTG curves plotted in ***Figure 5***. The thermal decomposition behaviour of MWNT-reinforced silk nanofibers was changed compared to as-spun silk nanofibers, although their two decomposition peaks occurred in the range of 200-400 °C. A prominent decomposition peak of as-spun regenerated silk nanofibers appeared at 298.5 °C corresponding to the thermal decomposition of the silk II structure [12]. There was a considerably weak decomposition peak at 219.7 °C attributed to electrospun silk nanofibers with a mainly random coil and silk I conformations. While the decomposition temperature of MWNT reinforced nanofibers variedirregularly in the process of initial decomposition and extended to higher temperature gradually as the MWNT content increased in the second decomposition process.

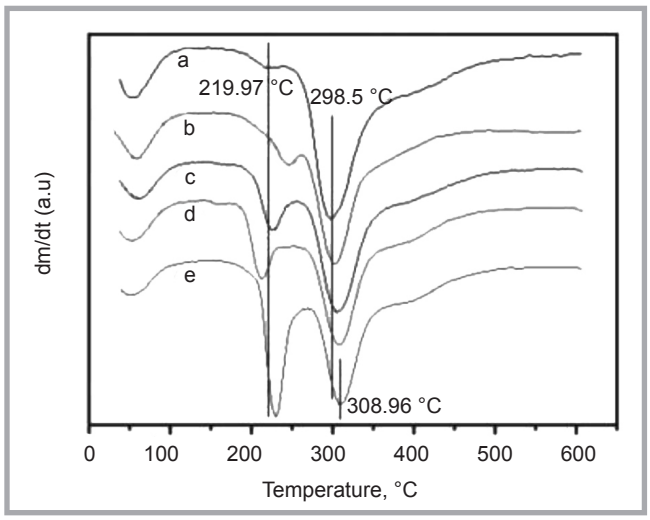

Figure 5. *DTG curves of MWNT reinforced and as-spun silk nanofibers: a) electrospun silk nanofibers, b) 0.5% MWNT reinforced nanofibers, c) 1.0% MWNT reinforced nanofibers, d) 1.5% MWNT reinforced nanofibers, and e) 2.0% MWNT reinforced nanofibers.*

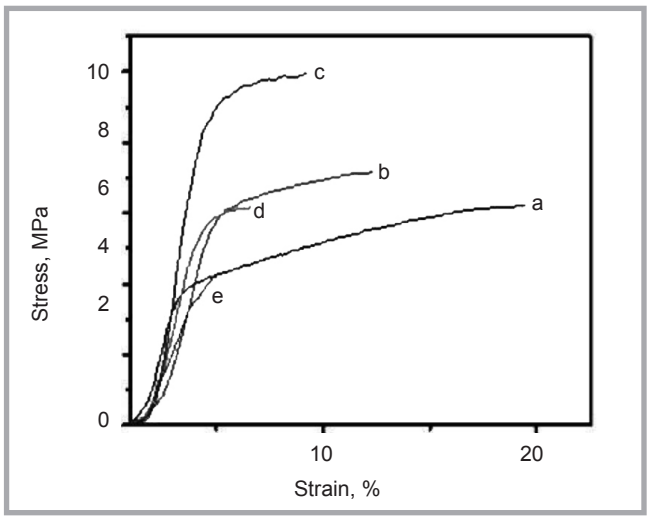

Figure 6. *Stress-strain curves of electrospun random silk mats: a) without MWNTs and b), c), d), e) with 0.5%, 1%, 1.5%, 2% MWNTs, respectively.*

The nanofibers with 2.0% MWNTs had a main decomposition peak at 308.96 °C, which indicated that the thermal stability increased compared to as-spun nanofibers. Interestingly the peak of 2.0% MWNT reinforced nanofibers at about 224 °C was more obvious compared with that at 308.96 °C, which indicated that more entangled aggregates in the nanofibers resulted in molecular chains being placed much more irregularly.

The results obtained above clearly present that introducing MWCNTs into silk fibroin fibers had no significant effect on the structure of the composite fibers compared to silk fibroin fibers. Generally CNTs have been used as a reinforcing agent, which result in high performance polymer nanocomposite fibers, as mentioned in previous studies [20]. To confirm the influence of the incorporation of MWNTs into silk nanofibers, the mechanical properties of the composites were investigated. For random a mat, strips measuring 4×0.5 cm were glued on a paper frame and then mounted on a universal test machine (Model no. 3365, Instron, USA) and the average tensile properties from five samples were measured [21]. The displacement was converted to strain by dividing it by the gauge length. The load on the strips was converted to Newtons. The tensile stress in MPa was calculated using the following ***Equation (1)***:

$$\text{Tensile stress} = \frac{F}{\text{WT}},\ \text{MPa} \qquad (1)$$

F – force, N; W – width, mm;
T – thickness, mm.

The thickness of the sample was measured through an electronic screw micrometer gauge (accuracy of 0.01 mm) and the average thickness from 10 different positions of each sample was measured.

Figure 6 shows the stress-strain curves for MWNT reinforced silk fibers and as-spun silk fibers. It can be evidently seen that there are different mechanical performances between the as-spun silk fibers and MWNT reinforced fibers from the figure. Detailed data are summarised in ***Table 2***. Both the tensile strength and Young's modulus were significantly increased by introducing different contents of MWNTs, except for nanofibers with 2% MWNTs. Incorporating 1.0% MWNTs into silk nanofibres, The Young's modulus and tensile strength of the composite increased from 32.5 ± 4.27 MPa to 107.46 ± 9.15 MPa and increased from 6.26 ± 0.8 MPa to 9.94 ± 1.2 MPa, respectively. The increased mechanical performance of 1.0% MWNT/silk composite were thought to result from the good dispersion and alignment of MWNTs along the fiber axis within silk nanofibers (***Figure 3***). The elongation at break, however, indicated the flexibility of materials, which decreased with MWNTs incorporated [10]. The random silk mats showed an elongation at break of 19.26 ± 1.2%, while with 1.0% MWNTs, it decreased to 9.25 ± 1.5%, also declining with an increase in MWNT content. In addition, both the tensile strength and elongation at break of the 2.0% MWNT/silk composite were significantly decreased, which was thought to be due to the aggregation of MWNTs. Accordingly it was concluded that the addition of a little amount of MWNTs enhanced the mechanical properties of MWNT/silk nanofiber.

MWNTs were introduced into electrospun silk fibers to improve the mechanical properties as well as the electrical conductivity of the composite. To measure the electrical conductivity, we used a ZC-90G high insulation resistance meter purchased from the Shanghai Yuanzhong electronic instrument factory (China). The samples were fixed on the equipment and the two probes of the machine were lowered onto the samples. The length, width and thickness of the sample were noted. The distance between the probes (probe space) was 2 cm. The known amount of resistance was

Table 2. *Mechanical properties of electrospun random silk mats and silk/MWNT mats.*

Fibers with MWNTs, w%	Young's modulus, MPa	Tensile strength, MPa	Elongation at break, %
0	32.50±4.27	6.26±0.8	19.26±1.2
0.5	58.53±1.97	7.20±0.6	12.30±0.8
1.0	107.46±9.15	9.94±1.2	9.25±1.5
1.5	91.75±10.56	6.12±0.9	6.67±0.6
2.0	85.94±15.51	4.28±0.4	4.98±0.9

Table 3. *Electrical conductivity of the electrospun random silk mats and silk/MWNT composite mats.*

Fibers with MWNTs, w%	Electrical conductivity of nanofibers, s/cm
0	5.04×10^{-14}
0.5	8.66×10^{-6}
1.0	1.20×10^{-4}
1.5	8.61×10^{-8}
2.0	7.15×10^{-9}

read on the viewing screen. The conductivity in Siemens/cm (S/cm) of the sample was measured using the following ***Equation (2)***.

$$\sigma = \frac{L}{\mathrm{RWT}},\ \mathrm{S/cm} \qquad (2)$$

L – lengt, cm; R – resistance, Ω;
W – width, cm; T – thickness, cm.

Electrospun silk nanofibers and nanofibers with 1% MWNTs had a conductivity of 5.04×10^{-14} S/cm and 1.2×10^{-4} S/cm, respectively, shown in ***Table 3***. The silk fibroin is the least conductive material. MWNTs played a role in boosting the electron conduction path. Consequently there is the possibility of "bridging" between the silk phases with an increase in MWNT loading. As a result the effective concentration of charge, the carrier increases which, in effect, reduces the hopping distance of the conduction electron and favours the percolation phenomenon [18]. The MWNTs incorporated in silk nanofibers at different concentrations were confirmed by TEM (***Figure 3***). However, with an increasing content of MWNTs, some aggregates resulted in a different electron conduction path to decrease the electrical conductivity.

Conclusions

An electrospinning process was used to fabricate silk fibroin nanocomposite fibers where MWNTs were embedded and well aligned. With a gradual increase in MWNT content, the diameter of the fibers has no obvious change. The results of XRD and thermal analysis indicated that the addition of MWNTs in silk fibroin fibers had no significant effect on the structure of the composite fibers compared to silk fibroin fibers. The silk nanofibers were significantly reinforced and achieved improved electrical conductivity by introducing a small amount of MWNTs, whereas the content of MWNTs is best with less than 1.0%. For tissue engineering of a load bearing tissue, which also requires electrical conductivity for cell growth, such as nerve cells [14], functional silk/MWNT nanocomposites should be expanded in their application for the repair of nerve injuries [13].

Acknowledgements

The authors would like to thank for the support of the Graduate Student Innovation Project of Jiangsu Province (KYZZ15_0327), China National Textile and Apparel Council (J201405).

References

1. Ayutsede J, Gandhi M, Sukigara S, et al. Regeneration of Bombyx mori silk by electrospinning. Part 3: characterization of electrospun nonwoven mat. *Polymer J* 2005; 46(5): 1625-1634.
2. Wu ZL, Zhang P, Gao MX, et al. One-pot hydrothermal synthesis of highly luminescent nitrogen-doped amphoteric carbon dots for bioimaging from Bombyx mori silk–natural proteins. *Journal of Materials Chemistry* B J 2013; 1(22): 2868-2873.
3. Wang Y, Blasioli DJ, Kim HJ, et al. Cartilage tissue engineering with silk scaffolds and human articular chondrocytes. *Biomaterials J* 2006; 27(25): 4434-4442.
4. Wang Y, Kim H J, Vunjak-Novakovic G, et al. Stem cell-based tissue engineering with silk biomaterials. *Biomaterials J* 2006; 27(36): 6064-6082.
5. Meinel A J, Kubow KE, Klotzsch E, et al. Optimization strategies for electrospun silk fibroin tissue engineering scaffolds. *Biomaterials J* 2009; 30(17): 3058-3067.
6. Chao PHG, Yodmuang S, Wang X, et al. Silk hydrogel for cartilage tissue engineering. *Journal of Biomedical Materials Research Part B: Applied Biomaterials J* 2010; 95(1): 84-90.
7. Min B M, Lee G, Kim SH, et al. Electrospinning of silk fibroin nanofibers and its effect on the adhesion and spreading of normal human keratinocytes and fibroblasts in vitro. *Biomaterials J* 2004; 25(7): 1289-1297.
8. Matthews JA, Wnek GE, Simpson DG, et al. Electrospinning of collagen nanofibers. *Biomacromolecules J* 2002; 3(2): 232-238.
9. Iijima S. Helical microtubules of graphitic carbon. *Nature J* 1991; 354(6348): 56-58.
10. Cheng HKF, Pan Y, Sahoo NG, et al. Improvement in properties of multiwalled carbon nanotube/polypropylene nanocomposites through homogeneous dispersion with the aid of surfactants. *Journal of Applied Polymer Science J* 2012; 124(2): 1117-1127.
11. Coleman JN, Khan U, Blau WJ, et al. Small but strong: a review of the mechanical properties of carbon nanotube–polymer composites. *Carbon J* 2006; 44(9): 1624-1652.
12. Hou H, Ge JJ, Zeng J, et al. Electrospun polyacrylonitrile nanofibers containing a high concentration of well-aligned multiwall carbon nanotubes. *Chemistry of Materials J* 2005; 17(5): 967-973.
13. Wei K, Xia J, Kim BS, et al. Multiwalled carbon nanotubes incorporated bombyx mori silk nanofibers by electrospinning. *Journal of Polymer Research J* 2011; 18(4): 579-585.
14. Lovat V, Pantarotto D, Lagostena L, et al. Carbon nanotube substrates boost neuronal electrical signaling. *Nano letters J*, 2005; 5(6): 1107-1110.
15. Clingerman ML, Weber E H, King J A, et al. Development of an additive equation for predicting the electrical conductivity of carbon-filled composites. *Journal of applied polymer science J* 2003, 88(9): 2280-2299.
16. Jeong JS, Jeon SY, Lee TY, et al. Fabrication of MWNTs/nylon conductive composite nanofibers by electrospinning. *Diamond and Related Materials J* 2006, 15(11): 1839-1843.
17. Kang M, Jin HJ. Electrically conducting electrospun silk membranes fabricated by adsorption of carbon nanotubes. *Colloid and Polymer Science J* 2007, 285(10): 1163-1167.
18. Um IC, Kweon HY, Park YH, et al. Structural characteristics and properties of the regenerated silk fibroin prepared from formic acid. *International Journal of Biological Macromolecules J* 2001, 29(2): 91-97.
19. Kang M, Chen P, Jin HJ. Preparation of multiwalled carbon nanotubes incorporated silk fibroin nanofibers by electrospinning. *Current Applied Physics J* 2009, 9(1): 95-97.
20. Lu Q,Wang XL,Lu SZ, Li MZ, Kaplan DL, Zhu HS. Nanofibrous architecture of silk fibroin scaffolds prepared with a mild self-assembly process. *Biomaterials J* 2011, 32(4): 1059-1067.
21. Mercante LA, Pavinatto A, Iwaki LEO, et al. Electrospun polyamide 6/poly (allylamine hydrochloride) nanofibers functionalized with carbon nanotubes for electrochemical detection of dopamine. *ACS Applied Materials & Interfaces J* 2015, 7(8): 4784-4790.
22. Chen D, Liu T, Zhou X, et al. Electrospinning fabrication of high strength and toughness polyimide nanofiber membranes containing multiwalled carbon nanotubes. *The Journal of Physical Chemistry B J* 2009, 113(29): 9741-9748.

Received 13.04.2016 Reviewed 04.11.2016