Research Article

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Temperature effects on electrospun chitosan nanofibers

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Abstract: Effects of the temperature of chitosan (CS) solutions as well as the temperature of the chamber on an electrospinning process were investigated. CS with a low molecular weight was dissolved in the solvent of trifluoroacetic acid/dichloromethane (70/30 v/v) at a concentration of 80 mg/mL for electrospinning. Both CS solution and chamber temperatures strongly affected the morphology of electrospun CS nanofibers. At the solution temperature and chamber temperature of 32°C, uniform CS nanofibers with an average diameter of 200 nm could be obtained. Although the chamber temperature is generally regarded as an unimportant parameter in the electrospinning of polymers, the experimental results demonstrated its critical effect on the electrospinning of CS.

Keywords: chitosan, electrospinning, nanofibers, temperature effects

1 Introduction

Chitosan (CS) is a biodegradable, biocompatible, and nontoxic polymer that is abundant in nature [1–5]. Besides the above advantages of CS, nanofibrous CS substrates can provide a high surface area for adsorption and cell attachment [1,6]. Thus, CS nanofibers should have a high potential use for tissue-engineering scaffolds as well as adsorption processes [1,2,7]. However, electrospinning of CS is limited due to its high viscosity, the limitation of solvents, and the positive charges of amino groups [8]. Although there are a few reports about the electrospinning of pure CS, important operational parameters for the electrospinning of CS have not been investigated thoroughly. Ohkawa et al. (2004) reported the first successful preparation of pure CS fibers by the electrospinning technique. By using trifluoroacetic acid (TFA) as the solvent, homogeneous CS nanofibers were successfully prepared [9]. High volatility of TFA plays a crucial role in the formation of uniform nanofibers. Moreover, TFA can block the positive charges of the amino groups on CS, which can decrease the electrostatic forces and enable the production of continuous fibers [9,10]. Therefore, the effectiveness of the TFA fostered further studies on electrospinning CS with various molecular weights where TFA was utilized as the solvent [11,12].

Electrospinning can create very fine fibers from a liquid by using an electrostatic force. When the electrical field is applied, charges on the surface of a polymer solution repulse each other. Meanwhile, surface tension causes forces opposite to the electrical force. With counteraction between mutual repulsive force between charges and the force from surface tension, the liquid is bended to form an electrospun jet. The eruption of the liquid has a conical shape, called as the Taylor cone [13–16]. Then, the jet is continuously elongated, and the solvent is also evaporated to form fibers if the entanglement force in the liquid is high enough to prevent the breaking of the stream. Finally, fibers are deposited on

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the collector. Electrospinning is a simple process to produce micro- and nanofibers. However, the effects of parameters are very complicated [13,17]. The electrospinning parameters are usually divided into three groups: the properties of a polymer solution (e.g., polymer concentration, polymer molecular weight, the type of solvent); operational parameters (e.g., the applied voltage, the distance between the needle tip and collector, feeding rate); and ambient conditions (e.g., temperature, humidity, velocity of air chamber) [13,17,18].

While various studies focused on polymer solution and operational parameters, ambient parameters are generally considered to contribute an insignificant effect to the electrospinning process. However, it should be noted that an increase in the temperature of a polymer solution might lead to a decrease in the viscosity of that solution and an increase in the evaporation of the solvent [19]. Thus, an increase in temperature of a polymer solution could lead to a decrease in diameters of electrospun fibers. Therefore, there are several studies on the effects of the polymer solution temperature on electrospinning. The results show that increasing the temperature is often beneficial to the process of electrospinning [19–22]. In addition, melting electrospinning is also an attractive issue because there is no need to use solvents for electrospinning [23–25].

Although uniform CS nanofibers could be prepared by using the TFA/dichloromethane (DCM) as the solvent, the temperature effect of the CS solution has not been investigated. Also, it is questionable whether the temperature of the electrospinning chamber might have a significant effect on the evaporation of the solvent. To address this knowledge gap, this research work investigated the effects of both solution temperature and chamber temperature on electrospun CS nanofibers.

Rigorous experimentation on the impact of various types of solvents on the nanofiber morphology was also conducted to reconfirm the effectiveness of TFA/DCM for the successful electrospinning of pure CS nanofibers.

2 Materials and methods

2.1 Materials

Ascorbic acid, glycolic acid, malic acid, succinic acid, TFA, ethanol, hexane, tetrahydrofuran (THF), and CS with a low molecular weight were purchased from Sigma-Aldrich. DCM was obtained from TEDIA.

2.2 Methods

2.2.1 Electrospinning of CS

In this study, we developed a customized electrospinning setup, as shown in Figure 1. There are three main components in the electrospinning equipment: a syringe pump (KD Scientific, KDS 100) with a metallic needle (0.65 × 32 mm), where the temperature of the solution is controlled; a high voltage generator; and a collector covered by an aluminum foil.

The solution temperature was controlled by surrounding the needle and the syringe with a tube that allowed a continuous flow of water from a temperature-controlled water bath (as illustrated in Figure 1, Section I). An electric heater attached on both sides of the chamber was used to warm up the chamber to the desired chamber

Figure 1: Electrospinning setup.

temperature measured by a thermocouple that was placed close to the workspace of the collector (as illustrated in Figure 1).

In this study, CS solutions were prepared for electrospinning using various solvents with different levels of volatility. First, low-volatility acids (i.e., ascorbic acid, glycolic acid, malic acid, succinic acid) were utilized to produce CS solutions of 2–5% concentration for electrospinning. Second, acetic acid, a moderate-volatility acid, was also used to produce CS solutions of 5–7% concentration for electrospinning. Co-solvents (i.e., ethanol, hexane, THF) were also added into the solution to analyze their effect on electrospinning products. Finally, CS was dissolved by strong and high-volatility TFA with DCM as the co-solvent, as described by the previous report for preparing pure CS nanofibers [9]. CS was dissolved in TFA/DCM (70/30 v/v) at a concentration of 80 mg/mL. After stirring for 12 h, the CS solution was placed in a 3 mL syringe for

electrospinning. The distance and the applied voltage between the tip and the collector were 120 mm and 17 kV, respectively. The flow rate of CS solution was fixed at 0.2 mL/h. The electrospinning experiments were carried out at a humidity of 70–75% for 45 min. To study the temperature effects of the solution, the solution temperatures of 20°C, 22°C, 27°C, and 32°C were observed under experimentation after the chamber temperature had been fixed at 32°C. To investigate the chamber temperature, the experiments were conducted at the chamber temperatures of 24°C, 29°C, 32°C, and 37°C after the solution temperature had been controlled at 32°C.

2.2.2 Characterizations of electrospun CS nanofibers

Surface morphology of the CS nanofibers was observed by a scanning electron microscope (SEM, JSM-6390LV, JEOL, Japan) at an accelerating voltage of 20 kV after gold coating.

Figure 2: SEM images of electrospun CS with 30 mg/mL of CS concentration in various solvents: (a) ascorbic acid, (b) glycolic acid, (c) malic acid, and (d) succinic acid.

From the SEM image results, the diameters of CS fibers and the distribution of fiber diameters were analyzed by ImageJ software. A hundred fibers were randomly selected from different areas of a SEM image to determine the mean diameter and its standard deviation of the obtained nanofibers.

3 Results and discussion

3.1 Effects of the solvent

Besides TFA, various solvents for CS including low-volatility acids such as ascorbic acid, glycolic acid, malic acid, and succinic acid; and a moderate-volatility acid (i.e., acetic acid and its mixtures with various co-

solvents) were experimented in this study. Figure 2 shows the SEM images of electrospinning CS using solvents of the former type. No uniform structure could be observed. It was known that TFA could change the viscosity of a CS solution and the interaction between the amino groups of CS [9], resulting in the electrospinnability of pure CS. For ascorbic acid, succinic acid, and malic acid, they increased the viscosity of a CS solution, revealing the enhancement in the entanglement force between the polymer chains of CS [26]. Thus, no homogeneous and continuous fibers could be formed by using these solvents.

Figure 3 shows the SEM images of electrospun CS with acetic acid and the mixture of acetic acid with co-solvents including hexane, ethanol, and THF. These co-solvents were chosen due to their low surface tension, low dielectric constant, and high volatility (Table 1). It was found that the bead-in-string morphology existed,

Figure 3: SEM images of electrospun CS with 70 mg/mL of CS concentration in various co-solvents: (a) acetic acid, (b) acetic acid and hexane, (c) acetic acid and ethanol, and (d) acetic acid and THF.

Table 1: Physical properties of the main solvents for the electrospinning of CS^a

Solvent	Surface tension (mN m ⁻¹ , at 20°C)	Relative density (g/cm ³ , at 25°C)	Viscosity (cP, at 25°C)	Vapor pressure (hPa, at 25°C)	Boiling point (°C, at 760 mm Hg)	Dielectric constant at 20°C
TFA	72.50	1.479	0.813	158	71.80	8.55 ϵ_0
DCM	26.50	1.317	0.413	584	39.75	8.93 ϵ_0
Acetic acid	27.60	1.049	1.155	20.79	118	6.15 ϵ_0
THF	27.30	0.880	0.480	255	65	7.60 ϵ_0
Ethanol	22.10	0.787	1.095	88.44	78.29	25.30 ϵ_0
Hexane	18.43	0.659	0.297	100	69	1.89 ϵ_0

^a The values of physical properties refer to the Safety Data Sheet of Sigma-Aldrich for the solvents.

and the homogeneity of CS nanofibers was not improved using different co-solvents. These factors agreed well with the previous reports in ref. [9,27], which supported the use of TFA as a high-volatility solvent to suppress the positive charges of CS for the successful electrospinning of pure CS.

3.2 Effects of the solution temperature

Figure 4 shows the SEM images of the electrospun fibers when the solution temperatures were varied from 20°C, 22°C, 27°C, and 32°C. Remarkably, morphology change from bead-connected fibers to uniform fibers was

Figure 4: SEM images of electrospun CS with 80 mg/mL of CS concentration in the solvents TFA/DCM (70/30 v/v) at different temperatures of the solution: (a) 20°C, (b) 22°C, (c) 27°C, and (d) 32°C.

observed, following the increase in solution temperature. This finding could be very useful for the formation of uniform electrospun fibers and was consistent with previous research works on the electrospinning of CS/poly(acrylamide) [20], CS/polyethylene oxide [19], and nylon 6 [21].

Because the high viscosity of CS solution may limit the electrospinning process, a high concentration of CS with a low molecular weight was used as a trade-off between lowering viscosity (i.e., using low-molecular weight CS) and allowing the successful electrospinning of CS nanofibers (i.e., increasing concentration to provide enough CS material). Therefore, an increase in the solution temperature might lead to a decrease in the viscosity of CS solution. As a consequence, the entanglement force could be reduced and the electrospinning process could get accelerated to produce uniform CS nanofibers.

The solution temperature increase could also result in an increase in chain mobility and repulsive

interaction among polymer chains. Thus, increasing solution temperature could partially promote the formation of fibers and limit the formation of beads. These tendencies could have facilitated the formation of uniform electrospun fibers.

3.3 Effect of the chamber temperature

Figure 5 shows the SEM images of the electrospun CS fibers at various chamber temperatures (24°C, 29°C, 32°C, and 37°C). Uniform CS nanofibers were obtained at the chamber temperature of 32°C. An increase in the chamber temperature could have resulted in a higher evaporation rate of the solvent; thus, the electrical forces could be reduced during the electrospinning process. However, at a higher chamber temperature (i.e., 37°C), the electrospinning jet was solidified, which resulted in

Figure 5: SEM images of electrospun CS with 80 mg/mL of CS concentration in the solvents TFA/DCM (70/30 v/v) at different temperatures of the chamber: (a) 24°C, (b) 29°C, (c) 32°C, and (d) 37°C.

Figure 6: Diameter distribution of electrospun CS nanofibers using 80 mg/mL of CS concentration in the solvents TFA/DCM (70/30 v/v) at the optimal chamber and solution temperature of 32°C.

the production of inhomogeneous fibers. Therefore, chamber temperature should be considered as a crucial electrospinning parameter for obtaining uniform CS nanofibers.

Figure 6 shows the distribution of the CS nanofiber diameters obtained under the optimal temperature conditions. The diameter of the obtained CS nanofibers had a mean value of about 200 nm with a standard deviation of 86 nm.

4 Conclusion

Uniform nanofibers with an average diameter of 200 nm were successfully obtained from pure CS by the electrospinning technique. The TFA/DCM (70/30 v/v) solvents were the best choice for the electrospinning process. The temperature of the CS solution and the chamber had significant effects on the morphology of CS nanofibers. The proper temperatures of the solution and the chamber were 32°C for preparing pure CS nanofibers, which would be a potential factor for various applications in biomaterials, adsorbents, and carriers.

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Conflict of interest: The authors declare that there is no conflict of interest.

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