Investigation and Prevention of Clogging During Electrospinning of Zein Solution

Kobsak Kanjanapongkul, Saowakon Wongsasulak, Tipaporn Yoovidhya

Department of Food Engineering, Faculty of Engineering, King Mongkut's University of Technology, Thonburi, Bangmod, Tungkru, Bangkok 10140, Thailand

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ABSTRACT: Electrospinning is the only known technique for fabrication of long non-woven ultrafine-fibers. However, a number of limitations, including low productivity and clogging of a spinneret, substantially paralyses industrial scale up of the process. Clogging is indeed an unavoidable phenomenon during electrospinning of biopolymers and the exact reason for clogging is still unclear. The aim of the present study was thus to investigate the clogging phenomenon via the study of a solution of zein/ethanol, which was used as a model biopolymer system. The gel-like substance causing clogging at the spinneret was collected and its infrared spectrum and rheological properties were determined via Fourier transform infrared (FTIR) spectrophotometry and rheometry, respectively. The results indicated that clogging was due to solvent evaporation, which led to formation of a highly viscous semi-solid at the spinneret. Moreover, the results also revealed that the applied voltage and polymer concentration were the key parameters affecting clogging. A means to help avoid clogging and hence an ability to continuously perform electrospinning was also proposed and tested. © 2010 Wiley Periodicals, Inc. J Appl Polym Sci 118: 1821–1829, 2010

Key words: biopolymers; electrospinning; film; nanotechnology; zein

INTRODUCTION

Due to a wide range of electrospinning applications, the number of published works on the topic has increased rapidly during the past decade. Nevertheless, at least two main problems with electrospinning still exist and need to be solved. The first problem is related to a low productivity of the process. As production of electrospun fiber is at a nanoscale, the rate of production for a single spinneret is only ~ 100 mg/h. Although some innovative techniques have been developed to increase the production capacity by up to even ten folds compared with the capacity of a conventional electrospinning process,[1–5] another problem, which is seldom mentioned in many previous works, still exists. This second important problem of electrospinning is related to clogging.[6–10] When clogging occurs, a droplet at the needle tip becomes a gel-like substance; electrospinning jet is interrupted and stops. This interruption leads naturally to a decreased productivity of the process. Clogging also affects the product quality in an adverse fashion.[11]

Solvent evaporation and viscosity of the solution have been noted to have strong impacts on clogging. Clogging often occurs when high-volatility solvents are used.[6–10] High volatility of a solvent accelerates solvent evaporation, thus increases the likelihood of clogging. Besides solvent evaporation, high viscosity of an electrospinning solution is also one of the key contributors to clogging.[6] This is because applied electric field cannot overcome the viscous drag force. During electrospinning, evaporation of solvent is expected. As a result, concentration at the droplet surface increases. The higher the concentration, the higher the solution viscosity is.[12,13] If the viscosity is too high, it is possible that, the higher the solution viscosity is the applied electrical force would not be adequate to overcome the viscous drag force at the droplet-air interface, leading to clogging at the spinneret.

In this work, zein, a protein from corn, was selected as a model biopolymer. As zein provides many advantages in such terms as biocompatibility, biodegradability, electrospinnability, and film forming ability, it is considered a potential raw material for bioengineering applications.[14–19] Zein solution is normally prepared in aqueous ethanol as ethanol offers some advantages including excellent solubility

Correspondence to: T. Yoovidhya (tipaporn.yoo@kmutt.ac.th).

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of zein and its non-toxic property. Nevertheless, rapid clogging of ethanolic zein solution during electrospinning was observed. This is because of the high volatility of ethanol, resulting in a rapid increase of the viscosity of the zein solution.[20]

The objective of this work was to investigate clogging of zein solution during electrospinning as well as to develop a technique to prevent clogging. The properties of a gel-like substance collected after clogging and the effects of process parameters on clogging were studied.

MATERIAL AND METHODS

Zein solution preparation

Zein powder (Sigma Z3625) was purchased from Sigma-Aldrich (St. Louis, MO) and used as received. Ethanol was supplied by Earth Chemical Lab (Bangkok, Thailand) and was diluted with distilled water to 90% v/v. Zein was dissolved in aqueous ethanol at 26°C ± 1°C under constant magnetic stirring for 1 hour. The solution concentration was 31% wt. Before loading into a syringe, the solution was left until no air bubble in the liquid phase was observed.

Characterization of gel-like substance collected after clogging

Gel-like sample collection

Zein solution was loaded into a 10-mL glass syringe (inner diameter of 15 mm) equipped with a stainless steel needle. The syringe was then placed on a syringe pump (New Era, NE-1000, Wantagh, NY). The needle tip was connected to a high voltage D.C. power supply (Gamma High Voltage, ES30P-5W, Ormond Beach, FL). The ground electrode was connected to a stainless steel collector plate. The distance between the needle tip and the collector plate was maintained at 12 cm. The solution was electrically charged at 21 kV. Once clogging was observed, the gel clump was collected from the needle tip for further analysis. The thickness of the surface layer of the gel clump was examined under a light microscope (Olympus, CH-2, Tokyo, Japan).

Determination of zein concentration of gel-like substance

Three samples of the gel-like substance were collected after clogging. The solvent was evaporated from each sample using a hot-air oven (Memmert, model 800, Schwabach, Germany) at 105°C over night. The zein concentration in each sample was calculated using eq. (1):

$$\text{Zein concentration } (\%\text{w}) = W_a/W_b \quad (1)$$

where W_b (g) and W_a (g) are the sample masses before and after drying, respectively. All experiments were performed in triplicate.

Rheological measurement of zein solution

To study an increase in the viscosity of the solution during clogging, fresh zein solution was prepared in an open-top container and then electrically charged at 21 kV (the same voltage used in the electrospinning experiment). The solvent was allowed to evaporate from the sample until the solution had the same solid fraction as the gel-like substance. The apparent viscosity of the sample was then measured using a rotational viscometer (Haake, VT500, Karlsruhe, Germany) fitted with SV1 concentric cylindrical sensors. The shear stress of the sample was recorded as a function of the shear rate from 0 to 100 s^{-1} at ambient temperature (26°C ± 1°C) and fitted to the Ostwald-de-Waele power law model[20,21]:

$$\sigma = K\,\dot{\gamma}^n \quad (2)$$

where σ (Pa) is the shear stress, $\dot{\gamma}$ (s^{-1}) is the shear rate, K (Pa s^n) is the consistency coefficient and n is the flow-behavior index. All experiments were performed in triplicate.

Fourier transform infrared spectroscopy analysis of gel-like substances

Functional groups and amide structure conformations of the gel-like sample periodically collected during electrospinning (0–35 s) were analyzed using a Fourier-transform infrared (FTIR) spectrometer (Perkin–Elmer, Spectrum Spotlight 300, Waltham, MA). The samples were prepared in the form of KBr pellets. In brief, each collected gel-like sample was firstly vacuum-dried at 40°C for 7 h to completely remove the solvent. Then, 2 mg of the dried sample was mixed with 200 mg of spectroscopy-grade KBr and the mixture was then pressed into a 12 mm-disk using a pressing die. A DTGS-KBR detector was used to collect the spectra of the sample disk within a range of 4000–400 cm^{-1} with a 4 cm^{-1} resolution and 10 scans. Duplicate spectra were obtained from independent experiments. The average spectrum was taken for subsequent analysis.

Determination of clogging time

A video camera (JVC, GZ-MG630AAG, Tokyo, Japan) was used to record the transformation of a droplet at the spinneret to a gel-like pocket entrapping the electrospining solution. A series of images was extracted from the video file with the minimum frame rate of five images per second. Clogging was

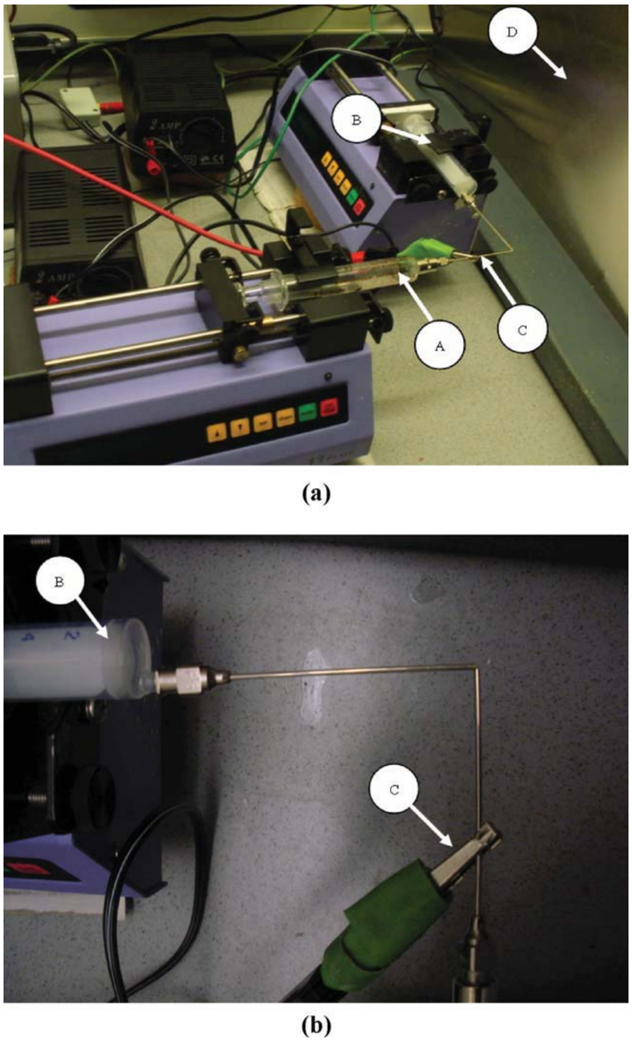

Figure 1 (a) Modified electrospinning setup. A and B: syringes containing polymer solution and solvent, respectively; C: electrode; D: collector plate. (b) Top view picture to show the positions of the needle tips. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

identified when the electrospinning process could no longer continue due to an interruption by a gel clump formed at the spinneret. The clogging time (which was defined as the time from the beginning of electrospinning to the time that clogging was observed) was determined from the extracted images. All experiments were done in triplicate.

Modified electrospinning set-up

The ultrafine-fibrous zein films were produced using the newly developed technique as well as by a conventional electrospinning method. A modified electrospinning set-up was developed to prevent clogging during electrospinning [Fig. 1(a)]. In brief, one extra syringe pump was used to supply additional amount of aqueous ethanol to the droplet surface at the needle tip to prevent clogging during electrospinning. The volumetric flow rates of the solution and the solvent were controlled independently by pumps 1 and 2, respectively. The modified set-up was designed in such a way that the needle tip of the syringe containing solvent came into direct contact with the needle tip of the syringe containing zein solution to ensure that the additional solvent was supplied directly onto the surface of the droplet of the zein solution [Fig. 1(b)].

Determination of fiber morphology

The films obtained were left at ambient temperature (26°C ± 1°C) overnight before being investigated for their morphology using a scanning electron microscope (SEM) (JEOL, JSM 5800, Tokyo, Japan). The films were vacuum-dried and gold-sputtering coated at a coating condition of 15 mA for about 4 min. The average diameters of the electrospun fibers were determined by the image analysis software Image J (National Institutes of Health, Bethesda, MD).

A light microscope (Olympus, CH-2, Tokyo, Japan) was also employed to investigate the fiber morphology. The electrospinning experiment was the same as that described in "Modified electrospinning set-up" section but with a glass slide fixed on the collector plate. The glass slide was used to collect the fibers for 5 s. Then, the glass slide was removed from the collector plate and the fiber images were taken via the light microscope.

RESULTS AND DISCUSSION

Characterization of the gel-like substance collected after clogging

As mentioned earlier, clogging was observed when zein/ethanol solution was electrospun. The gel-like substance occurring during electrospinning inhibited jet initiation, resulting in clogging [Fig. 2(a)]. Figure 2(b) shows a light microscopic image of the gel-like substance collected from the needle tip. Only the outer surface of the clogging substance became gel-like while the inner part was still liquid. As shown in Figure 2(c), the liquid solution flowed out after the gel-like substance was penetrated by a sharp pin. The zein concentration of the gel-like surface was found to be 0.51 ± 0.03 g zein/g sample. Compared with the zein concentration of 0.31 g zein/g sample of the original solution, this higher concentration indicated a significant loss of solvent due to evaporation during electrospinning. To investigate the change in the viscosity of the solution, zein solution having the same solid fraction as the gel-like sample (concentration of 50% wt) was prepared. Rheological properties, namely, apparent viscosity,

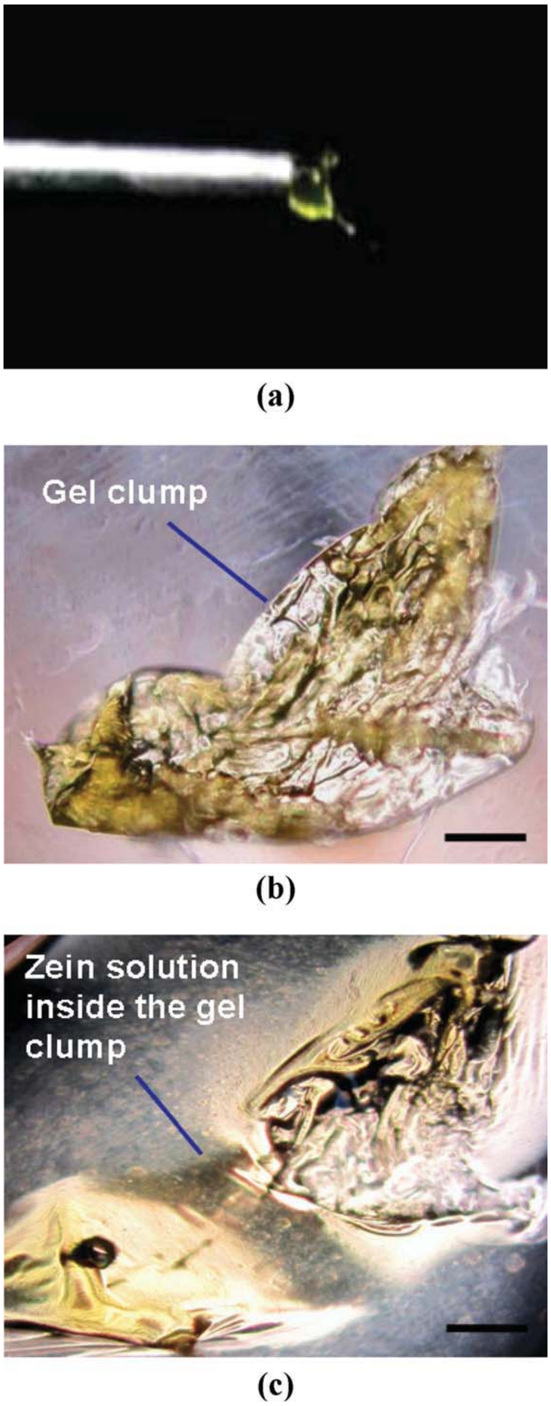

Figure 2 (a) Photo of droplet after clogging. (b) Light microscopic image of the gel-clump collected from the needle tip. (c) Zein solution inside the gel-clump (after breaking the outer surface using a pin). Scale bars correspond to 100 μm. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

consistency coefficient (K) and flow-behavior index (n), calculated using eq. (2), are shown in Figure 3. The average viscosity of the prepared zein solution was found to be about 16 times higher than that of the original zein solution. With the increase in the viscosity, electric force could not overcome viscous drag force; clogging thus occurred.

To investigate whether the functional groups of the gel-like substance and those in typical electrospinning solution were different or not, the two samples, the surface layer of gel clump (C_1) and electrospinning typical solution (C_0), were examined by FTIR. The results are shown in Figure 4. The intensity ratios of the peaks at the regions ~ 2850 (thiol groups of zein) and ~ 3300 cm^{-1} (free —OH group of ethanol) were 0.4 and 0.38, corresponding to the C_1 and C_0 samples, respectively [Fig. 4(a)]. The normalized spectra also showed that the intensity of the peak at 1350 cm^{-1} of C_1 was higher than that of C_0. Figure 4(c,e) show the deconvoluted spectra regions 1730–1430 cm^{-1} for C_0 and C_1, respectively. The deconvoluted peaks at around 1680, 1655, 1630 and 1544 cm^{-1} were attributed to the presence of β-sheets, random coil or α-helix, intermolecular β-sheets, and α-helix structure, respectively.[16,22–26] The obtained figures show no peak shift. This may indicate that no changes occurred in structural conformation of amide I and amide II in zein protein chains. The overall FTIR results indicated that the types of functional group components in C_0 and C_1 were not different; on the other hand, the higher concentration of zein protein in C_1 could be attributed to the increase in the zein concentration at the droplet-air interface of the Taylor cone as a result of solvent evaporation. This was also supported by the 16-time higher viscosity of C_1.

Effects of process parameters on clogging

To investigate the effects of process parameters, namely, ethanol concentration in the solvent, zein concentration, voltage, spinning distance, and solvent feed rate, on clogging (see Table I), an electrospinning experiment was set up. It was found that smooth zein fibers were obtained when the

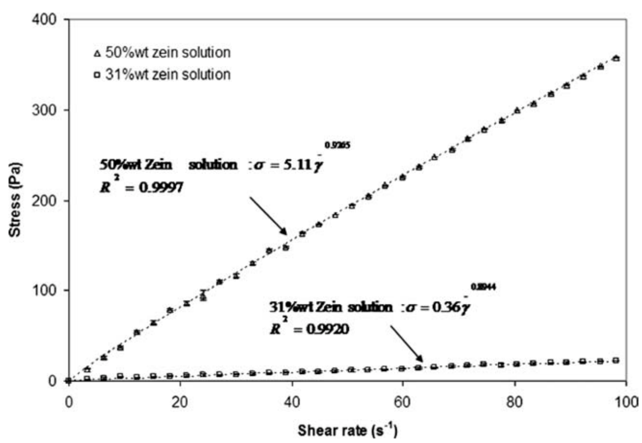

Figure 3 Shear stress–strain behaviors of zein solutions.

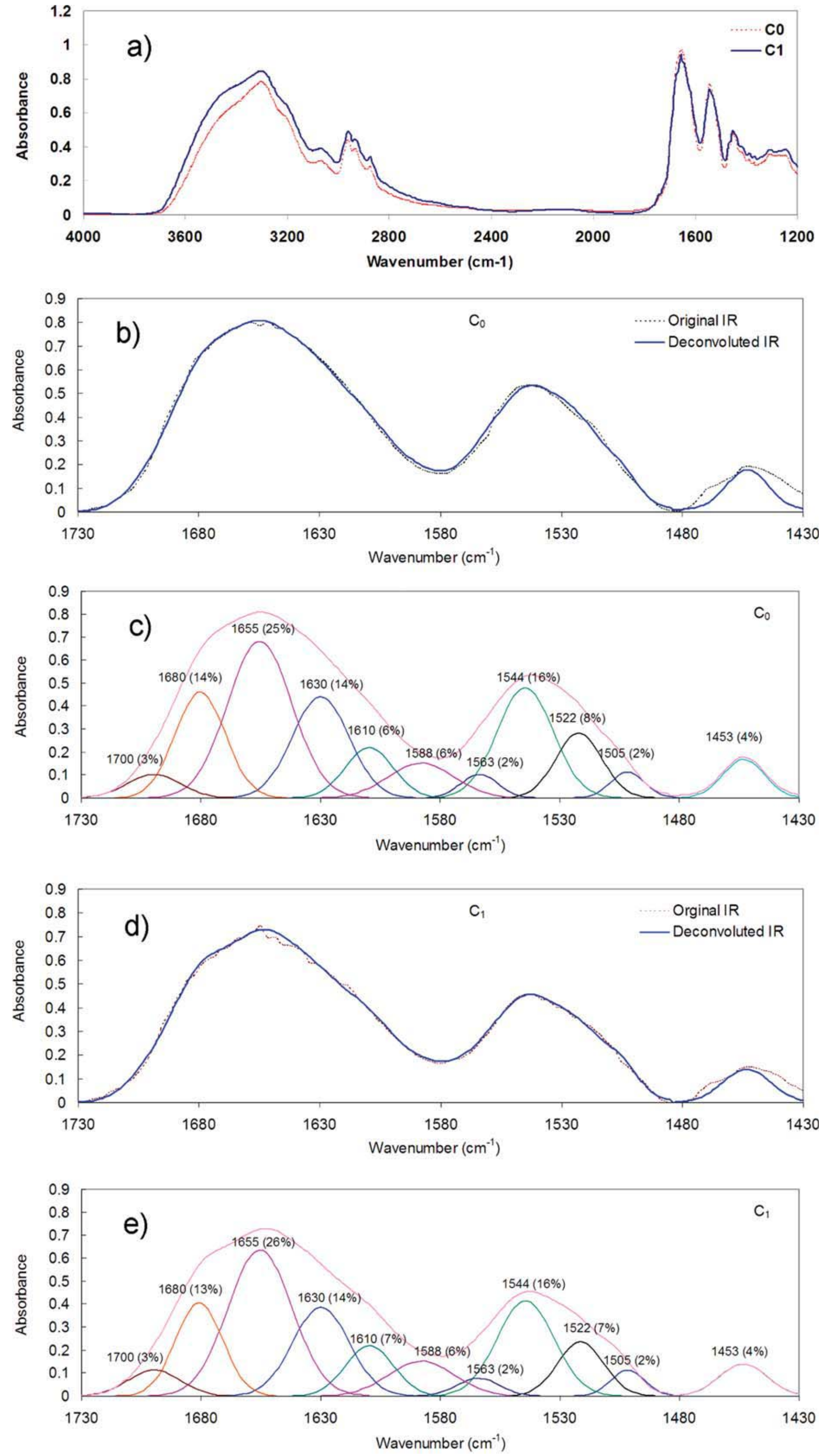

Figure 4 (a) IR spectra of zein solution (C_0) and gel-like surface collected after clogging (C_1) regions 4000-1200 cm^{-1}; (b) original and deconvoluted IR spectra of C_0 in Amide I and Amide II ranges; (c) curve-fitting of b); (d) original and econvoluted IR spectra of C_1 in Amide I and Amide II ranges; (e) curve-fitting of (d). [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

TABLE I
Process Parameters Studied

Process parameter	Value
Ethanol content in solvent (% v/v)	70, 80, 90, 95
Zein solution concentration (% wt)	18, 23, 28, 33, 38
Voltage (kV)	12, 15, 18, 21, 24
Spinning distance (from syringe tip to collector plate) (cm)	6, 9, 12, 15, 18
Solvent feed rate (μL/min)	5, 10, 15, 20,25

electrospinning conditions were as follows: solution concentration of 33% wt, voltage of +12 kV, distance from the tip to collector plate of 12 cm and feed rate of 20 μL/min. The collector plate had dimensions of 30 × 30 cm. The outer and inner diameters of the spinneret are 9 and 8 mm, respectively, and the working temperature was 26°C ± 1°C.

Figure 5 shows a series of images of clogging formation as a function of time. At the beginning of electrospinning [Fig. 5(a,b)], the Taylor cone was stable and the polymer jet was continuously electrospun. However, once clogging started to occur, the cone was no longer stable. A short viscous polymer tail was observed at the end of the cone [pointed by an arrow in Fig. 5(c)] and no more fibers could be produced, resulting in clogging. It was also observed that as the process time increased, the polymer tail was noticeably longer [Fig. 5(d)].

Figures 6a–e show the effect of each process parameter on the clogging time. In Figure 6(a), the clogging time could be prolonged, from 4.0 to 21.5 s, as the ethanol concentration was reduced from 95 to 70%. Reducing the ethanol concentration reduced the volatility of the solvent; evaporation thus became slower and clogging was delayed. Unfortunately,

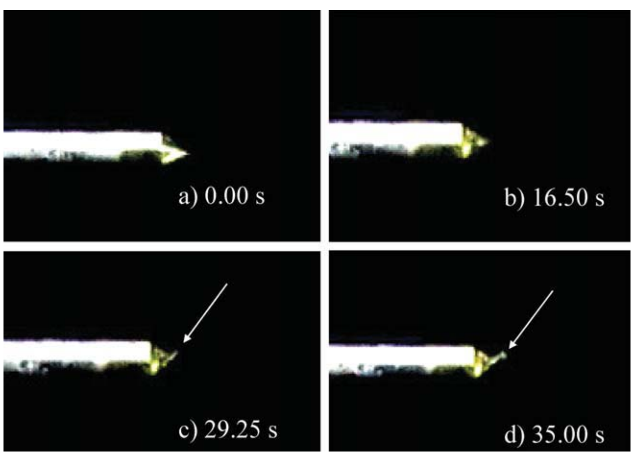

Figure 5 A series of images collected as a function of time. The electrospinning conditions were as follows: voltage = 12 kV, spinning distance = 12 cm, solution flow rate = 20 μL/min, zein concentration = 23% w/w, ethanol concentration = 90% v/v. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

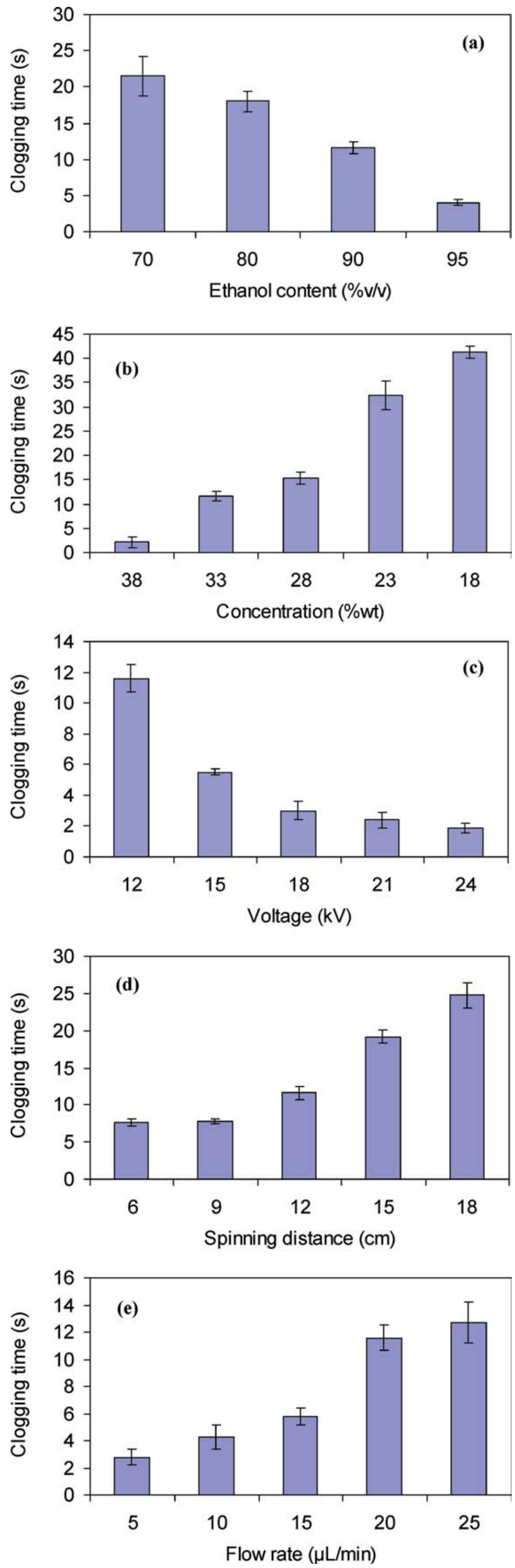

Figure 6 Influence of (a) ethanol content; (b) zein concentration; (c) voltage; (d) spinning distance; and (e) solution flow rate on clogging time. The process conditions used

reduced ethanol concentration resulted in an increase in the solution surface tension. It is known that a polymer solution having high surface tension tends to produce non-uniform fibers with beads,[27] which are undesirable. In our case, the bead-on-fibers were produced when an ethanol content was reduced to 70%.

Reducing the zein concentration resulted in a decrease in the solution viscosity and thus reducing the clogging frequency.[6,8] The minimum electrospinnable concentration of zein solution has been reported to be around 18–23% wt.[16,17,19] Therefore, the minimum zein solution concentration was set at 18% wt ($\sigma = 0.24\dot{\gamma}^{0.62}$). At this concentration, clogging occurred after 41.3 s [Fig. 6(b)]. To find out whether clogging would occur or not when the zein concentration decreased from an electrospinning level to electrospraying level (<18% wt), 16 and 10% wt zein solutions were also tested (data not shown). It was found that clogging was still observed in both cases, but at a later time (about 70 s in the case of 10% wt solution). When the zein concentration increased to 38 wt %, clogging occurred rapidly, within only 2.2 s. However, there is a limit in reducing the solution concentration since it has been reported that non-uniform fibers are generally produced when the solution concentration is too low.[28,29] In the present case, smooth and uniform fibers were produced from zein solution having concentrations of 28 wt % or higher. Within this concentration range, clogging was observed within 15 s or less.

It can be seen in Figure 6(c) that an increase in the voltage reduced the clogging time. Barrero et al.[30] indeed derived mathematically the velocity profile of liquid in the Taylor cone and found that the velocity was proportional to the tangential electrical stress at the gas–liquid interface and that the tangential electrical stress increased with the electric field strength. Thus, increasing the voltage increased the velocity of the liquid in the droplet; as a result, evaporation increased and clogging was accelerated.

Figure 6(d,e) show, respectively, the effect of the distance between the needle tip to the collector plate (spinning distance) and the effect of flow rate on the clogging time. A decrease in the electrospinning distance from 18 cm to 6 cm shifted the clogging time from 25 s to 8 s. Thus, clogging might be delayed by adjusting the spinning distance; however, there was again a limit to which clogging could be delayed. In this case clogging could be delayed to 25 s when the spinning distance increased to 18 cm. Similar to the spinning distance, increasing the flow rate also delayed clogging time by several seconds. Changing the flow rate resulted in different Taylor cone shapes. Increasing the flow rate would decrease the cone half-angle, thus reduced specific surface area (i.e., the ratio between the surface area and volume) of the cone, resulting in a decrease in the evaporation rate (per volume).

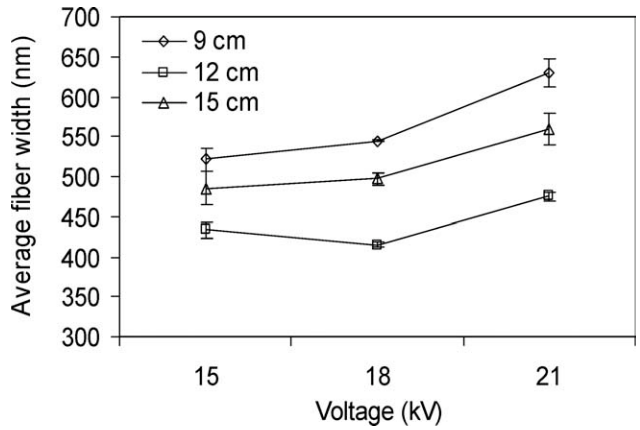

Figure 7 Average fiber width at different process conditions. Concentration and flow rate of zein solution were 31% wt and 20 μL/min, respectively.

Electrospinning with newly developed technique to prevent clogging

As mentioned earlier, the clogging problem might be solved by preventing the loss of solvent at the droplet-air interface. Simply adjusting the process parameters, e.g., solution concentration or voltage, could not completely prevent clogging as the loss of solvent at the air-droplet interface still excessively existed. Therefore, a new electrospinning experimental set-up was proposed (Fig. 1). To prevent the loss of solvent at the droplet-air interface, additional solvent was supplied directly onto the surface of the droplet of the zein solution. From our preliminary study, when the amount of the added solvent balanced with that of the evaporated solvent, clogging did not occur. If the solvent feed rate was too low, clogging would still occur. On the other hand, if the solvent feed rate was too high, there would be a number of small droplets of solvent sputtered onto the film on the collector plate. The flow rate of the zein solution was fixed at 20 μL/min, while the voltage was varied at 15, 18, and 21 kV; the spinning distance was varied at 9, 12, and 15 cm. The solvent feed rate that corresponded to the solution feed rate of 20 μL/min was found to be 4 μL/min. At all these conditions, clogging was not observed along the whole test period of 8 h.

To compare the morphology of the film produced by the newly developed technique with that produced by the conventional electrospinning technique (i.e., no supply of additional solvent), one optimum condition was chosen from the above nine tested conditions. The selected condition was the one that produced the fiber with smaller diameter and no beads. Figure 7 shows the average diameters of the

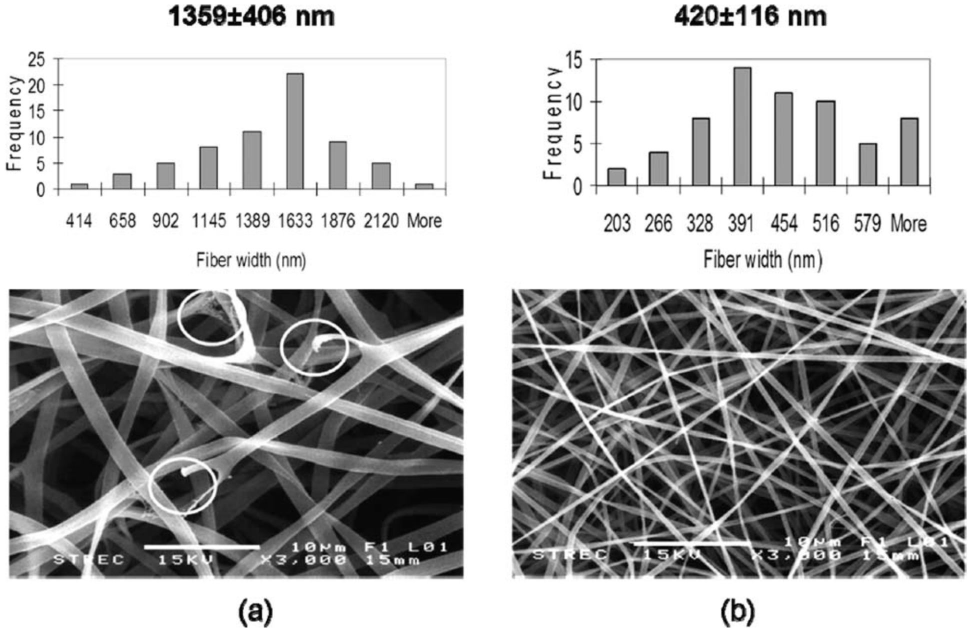

Figure 8 Morphology of ultrafine-fibers produced under optimum condition using (a) conventional electrospinning technique; (b) newly developed technique. The number given in each histogram was the average value ± standard deviation.

fibers obtained at different process conditions. Typically, fibers collected at a longer distance were thinner as the flying time was longer. However, at too long distance, the electric field strength would be too weak and the stretching force would be low and fibers would again become thicker. At a distance of 12 cm the fiber diameter was smaller compared with those of fibers collected at 9 and 15 cm. This could be due to the balance between the flying time and the stretching by the electrical repulsive force at a distance of 12 cm. Increasing the voltage noticeably increased the average fiber diameter. Using the voltage of 18 kV resulted in the fibers with the smallest average diameter. At the end the following electrospinning condition was selected: voltage of 18 kV, spinning distance of 12 cm and solution feed rate of 20 μL/min. This process condition was also used to produce zein film using the conventional electrospinning technique.

A selected SEM image of the film produced by the conventional electrospinning technique is shown in Figure 8(a). The fibers were ribbon-like with an average diameter (or fiber width) of 1,359 ± 406 nm. Discontinuities of the fibers were also observed (white circles). The fibers produced using the conventional electrospinning technique were short, while long continuous fibers were obtained using the newly developed technique (Fig. 9). Moreover, clogging was observed more frequently (every about 10 s) during the production of the film using the conventional technique. This led to fiber discontinuity and caused the film produced by the

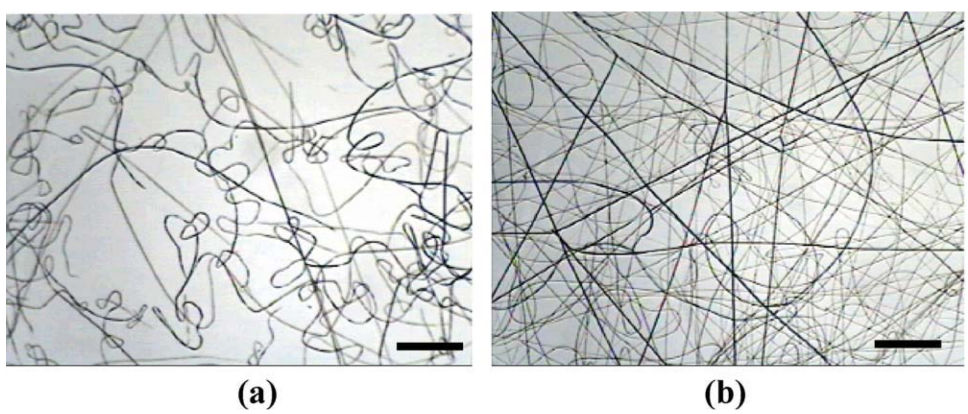

Figure 9 Selected light microscopic images of zein ultrafine-fibers produced using (a) conventional electrospinning technique; (b) newly developed technique (Scale bar is 50 μm). [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

conventional technique to be very fragile. On the other hand, the newly developed technique could produce long smooth ribbon-like fibers [Fig. 8(b)] with an average diameter of 417 ± 110 nm. This is probably because the additional amount of solvent reduced the viscosity of the droplet and facilitated stretching of the ultra-thin jet during its flight.

CONCLUSION

The present work investigated the clogging phenomenon during electrospinning of zein/ethanol solution. Better understanding of clogging was obtained through a combination of the studies of the properties of the gel-like substance collected after clogging as well as of the effects of process conditions on the clogging time. The solid fraction and the viscosity of the gel-like sample increased by 1.7 and 16 times, respectively, compared to those of the fresh zein solution. However, similarity of the IR spectra of the gel-like sample and of the zein solution revealed that no chemical change occurred during clogging. Based on all the obtained results, it was suggested that clogging occurred because of the solvent loss at the droplet-air interface due to solvent evaporation. This suggestion was supported by the success in clogging prevention by providing additional solvent onto the surface of the droplet at the needle tip. Besides, the average fiber diameter fabricated by the newly developed technique was reduced by approximately 3 times with a much smaller standard deviation. Due to its simplicity and practicality, the improved method is expected to be very useful for fabrication of electrospun ultrafine-fiber and would be applicable not only for zein solution but also for other polymer solutions as well.

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