Research Article

Electrospun Polycaprolactone Fiber Mats as Carriers for Resorcinol

Pimpawan Parana,Tidarat Bandhitsing and Patcharaporn Thitiwongsawet*

Department of Chemical Engineering, Faculty of Engineering, Thammasat University,Rangsit Campus,Khlong Nueng, Khlong Luang, Pathumthani, 12120, Thailand

Abstract

Resorcinol (RC)-loaded electrospun polycaprolactone (PCL) nanofibers were prepared by electrospinning from 12% and 15% w/v PCL solutions containing either 2.5 or 5 wt.% RC (based on the weight of PCL). The mixture of 1:1 v/v dichloromethane: *N*,*N*-dimethylformamide was used as solvent. The average diameters of RC-loaded PCL nanofibers prepared from 12%PCL/2.5%RC, 12%PCL/5%RC, and 15%PCL/2.5%RC were 137, 147, and 185 nm, respectively. The amounts of water retention and the release characteristics of RC from nanofibers in an acetate buffer (pH 5.5) were investigated and compared with the corresponding cast-films fabricated by the solvent casting method. The amount of water retention of the fiber mats was about 240-290%, which was 3 times higher than that of films (i.e. 80-90%). The study of release characteristics of RC from fiber mats and films was carried out by the total immersion method in an acetate buffer at 32°C for 48 hr. A burst release of RC at the initial time followed by a gradual release to reach a plateau was observed for both fibers and films. The maximum amounts of RC released from fiber mats and films were about 75-90% and 40-50%, respectively. An analysis based on various models of release kinetics including the Korsmeyer-Peppas, the zero and the first order models was studied. From the Korsmeyer-Peppas model, the exponents of release (n) were 0.37-0.44 for fibers and 0.33-0.34 for films. From all models, RC-loaded PCL fiber mats exhibited higher rate constant (k) than that of films. Moreover, a rate constant (k) from the fibers with smaller diameters was higher than that from the fibers with larger diameters.

Keywords: polycaprolactone; electrospinning; resorcinol; drug delivery.

1. Introduction

Electrospining is a technique for producing ultrafine fibers by applying high electrostatic potential to a polymer solution or melt to generate a charged jet [1, 2]. The charged jet is then travelled to a collector at the same time that a solvent is evaporated. The polymer fibers are collected on a collector. According to the physical uniqueness of electrospun fibers, e.g., high porosity, high surface area to mass ratio, they have been proposed as an ideal material in Many applications such as filtration, composite reinforcement, tissue engineering scaffolds and delivery of drugs. Electrospun fibers have widely been used as carriers for drug or chemical substances [3-5]. The advantage of electrospun fibers is the highly porous structure which exhibits a large surface area that could allow drug molecules to diffuse out more conveniently than from the corresponding films [5].

Resorcinol (RC), or 1,3-dihydroxybenzene, is an active ingredient used in acne treatments. It is used as an external pharmaceutical substance for remedes for acne and related skin diseases

***Correspondence :** andtpatchar@engr.tu.ac.th

[6]. The antioxidant property of RC has been reported in which the radical-scavenging activity of RC was found to be equivalent to that of (-)-epigallocatechin gallate in green tea [7]. It has also been reported to inhibit tyrosinase, which produces melanin pigment [8]. The chemical structure of RC is shown in Figure 1.

Polycaprolactone(PCL),a based polymer used in this work, is a biodegradable and biocompatible polymer that has widely been used in tissue engineering and drug delivery applications. Successful electrospinning of PCL has been reported from various solvents [9, 10]. A number of studies on drugs release based on PCL fibers matrix have been reported [4, 11-14]. Naproxen, a non-steroidal anti-inflammatory drug (NSAID), was complexed with cyclodextrin and incorporated in electrospun PCL fibers, and the release behaviors of the drug were reported [4]. The release characteristics of diclofenac sodium, a kind of NSAID, from electrospun PCL fibers were also reported [11]. Various kinds of herbs or synthetic herbs including tannic acid [12], curcumin [13], and crude bark extract from *Tecomella undulate* plants [14] were loaded in electrospun PCL fibers and were studied for release behaviors and related actions of drugs such as the antibacterial, antioxidant, and anti-inflammatory properties.

In this work, RC-loaded PCL fibers were prepared by electrospinning. The morphology and the sizes of electrospun fibers were examined. Amounts of water retention and release behaviors of RC from both electrospun fibers and cast-films were investigated. Finally, the analysis of the kinetics of release was studied.

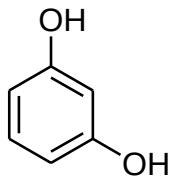

Fig.1. Chemical structure of RC.

2. Experiment setup

2.1 Materials

PCL (a number-averaged molecular weight of 80,000 $g.mol^{-1}$) was purchased from Sigma-Aldrich (USA). RC was purchased from Acros Organics (USA). The chemical structure of RC is shown in Figure 1. Dichloromethane (Carlo Erba, Italy) and *N*,*N*-dimethylformamide(Lab-ScanAsia, Thailand) were used as solvents for PCL.

2.2 Preparation of solutions and electrospinning process

A 12% w/v PCL solution was prepared in a solvent mixture of 1:1 v/v dichoromethane/ *N*,*N*-dimethylformamide. RC powder was added into a 12% w/v PCL solution at the concentrations of 2.5, 5, 7.5, and 10 wt.% (based on the weight of PCL). A 15% w/v PCL solution was also prepared. RC was added into a 15% w/v PCL solution at the concentration of 2.5 wt % (based on the weight of PCL). RC-loaded PCL fiber mats were prepared by an electrospinning process. The obtained eletrospun fibers are denoted as EF specimens. A Gamma High-Voltage Research ES30P-5W power supply was used to generate high DC potential. The applied potential was 18 kV. The polarity of the electrode was positive. The distance between the nozzle and the collector was 15 cm. The diameter of the nozzle was 0.91 mm. A rotating drum was used as a collector with a surface velocity of about 0.5 m/s. The feed rate of the solution was controlled to about 1 mL/hr. The shear vicosity of the solutions was measured by using a Cannon-Fenske viscometer. PCL films containing 2.5 and 5 wt. % RC (based on the weight of PCL) were also prepared for comparative studies by the solvent-casting method. The obtained films are denoted as CF specimens. The thickness of both the EF and the CF specimens was controlled to about 50 μm. The weight of the EF specimens was in the range of 5 – 8 mg whereas the weight of the CF specimens was about 7 – 10 mg.

2.3 Characterization of EF and CF specimens

The morphological appearance of the EF specimens was observed with a JEOL JSM-5200 scanning electron microscope (SEM). The average diameter of the fibers was measured from the SEM images, using the SemAphore 4.0 software. The amounts of water retention of both the EF and the CF specimens were determined after submersion in an acetate buffer solution (pH = 5.5) at 32°C for 48 hr according to the following equation:

$$Water\ retention\ (\%) = \frac{M - M_i}{M_i} \times 100 \quad (1)$$

where, M is the weight of each sample after submersion in acetate buffer for 48 hr and M_i is the weight of the sample before submersion.

2.4 Release of RC from EF and CF specimens

Preparation of acetate buffer solution. An acetate buffer solution (pH 5.5) was used as media in this study to simulate the human skin pH condition. Fifteen grams of sodium acetate was dissolved in distilled water followed by slowly adding of 1.5 mL of glacial acetic acid yielding a 100 mL buffer solution.

RC-release assay. Each EF and CF specimen was cut into a square shape with a dimension of 2 cm×2 cm and was submerged in 40 mL of an acetate buffer solution at the physiological temperature of human skin of 32°C. At a specified submersion time ranging between 0 to 48 hr, 3.0 mL of released media was withdrawn. An equal amount of freash media was refilled. The withdrawn solution was diluted with a buffer solution and was measured for absorbance to determine the amount of released RC by using a Shimadzu UV-1201 UV-vis spectrophotometer at the wavelength of 275 nm. The obtained data were calculated against the calibration curve to determine the cumulative amounts of RC released at each immersion time point. The experiments were carried out at least in triplicates.

3. Results and Discussions

3.1 Electrospinning of RC-loaded PCL solutions

The as-prepared 12% w/v PCL solutions with the addition of 2.5, 5, 7.5, and 10% RC (based on the weight of PCL) were electrospun under an electrostatic field of 18 kV/15 cm. The morphological appearance of the EF specimens obtained from 12% PCL/2.5% RC and 12% PCL/5% RC was round with a smooth surface as shown in Figures 2a and 2b. However, fibers with a rough surface and appearance of RC aggregates were observed from the electrospinning of 12%PCL/7.5%RC and 12%PCL/10%RC (SEM images are not shown here). This could be because the concentrations of RC were too high. Some amounts of RC could not be completely dissolved. Therefore, the EF specimens obtained from 12%PCL/7.5% RC and 12% PCL/10% RC were not used in the release study. The average diameters of the fibers obtained from 12%PCL/2.5%RC and 12% PCL/5% RC were 137±32 nm and 147±40 nnm, respectively. Therefore, here after, the obtained electrospun fibers are denoted as EF2.5RC/137nm and EF5RC/147nm, respectively, according to the amounts of RC and the sizes of the resulted fibers. Upon an addition of RC into the PCL solution (i.e. 2.5% and 5%RC), the viscosities of solutions were increased from 149 to 157 cSt (see Table 1). The viscosity of a solution is an important parameter controlling the size of the fibers. The increase in the viscosity indicated increase in the viscoelastic force. The increase in viscoelastic force is against the electrostatic and Coulombic repulsion forces. Therefore, the diameters of the resulted fibers were larger [15].

An attempt to increase the size of the fibers was done by increasing the concentration of the PCL solution or by increasing the viscosity of the solution. The 15% w/v PCL solution with an addition of 2.5% RC (base on the weight of PCL) was prepared and electrospun under the same condition. The viscosity of the solution was 314 cSt. The average diameter of the fibers was 185±52 nm (see Table 1). Electrospun fibers from 15%PCL/2.5%RC is denoted as EF2.5RC/185nm here after. The SEM image is shown in Figure 2c. Obviously, the size of the fibers increased as the viscosity of the solution increased. However, an addition of more than 2.5% RC into 15% PCL solution yielded a solution with viscosity that was too high and could not be electrospun. Therefore, only 3 types of the EF specimens including EF2.5RC/137nm, EF5RC/147nm, and EF2.5RC/185nm were used in the release study. Moreover, the films prepared through the solvent-casting process were used in a comparative study with the EF specimens. The films were prepared from 12%PCL/2.5%RC and 12%PCL/5%RC solutions. The obtained cast-films are denoted as CF2.5RC and CF5RC, respectively. The surface of the films were smooth. However, the SEM images of the films are not shown.

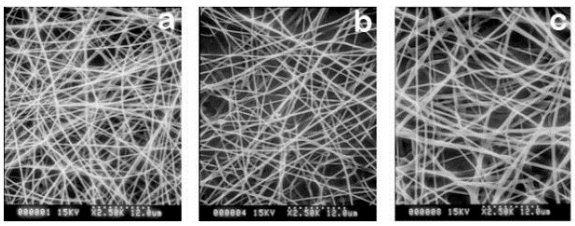

Fig.2. Selected SEM images of a)EF2.5RC/137nm b) EF5RC/147nm and c) EF2.5RC/185nm.

3.2 Water retention behavior of EF and CF specimens

The EF and CF specimens were characterized to determine the degree of water retention after submersion in an acetate buffer (pH 5.5) at 32°C for 48 hr. Figure 3 shows the percentage of water retention of all specimens including EF2.5RC/137nm, EF5RC/147nm, /185nm, CF2.5RC, and CF5RC. They were 292±3, 281±3, 240±3, 87±3, and 85±6%, respectively. Evidently, the amounts of water retention of the EF specimens were greater than those of the CF specimens. This could be due to the highly porous structure and a much greater surface area of fibers in comparison to those of films [5, 16]. Among the EF specimens, the amount of water retention of EF2.5RC/137nm was greater than those of EF5RC/147nm and EF2.5RC/185nm, respectively. Apparently, the smaller the fibers were, the greater the water retention was. The smaller fibers had the greater surface area per volume or mass ratio. Therefore, they held water better in a porous structure. The amount of water retention of these specimens will be discussed along with the results of the release characteristics of RC in the following section.

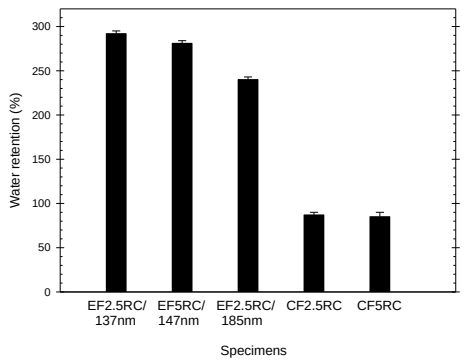

Fig.3. Water retention of EF and CF specimens.

3.3 Release of RC from EF and CF specimens

The release characteristics of RC from both the EF and the CF specimens were carried out using the total immersion method. Each specimen was cut into 2cm×2cm square shape and was immersed in an acetate buffer (pH 5.5) at 32°C for 48 hr. The release

profiles are shown in Figure 4, which reports the percentages of the cumulative amounts of RC released from specimen against the time of release. A percentage of the RC released was calculated as the weight of RC released ×100/the weight of the initial RC loaded. The release characteristics of both the EF and the CF specimens were alike a burst release at the initial time followed by a gradual release to reach a plateau. The amounts of the RC released from both the EF and the CF specimens reached the maximum and stabilized in the second hour of release. Even though, the experiments were done up to 48 hr,Figure 4 shows the data only for the first 5 hr because the amounts of release were constant after that. The maximum amounts of the RC released from the EF and the CF specimens were about 80-90% and 40-50%, respectively. Comparatively, all of the EF specimens exhibited greater cumulative released amounts of RC than the CF specimens did. The release kinetics including the Korsmeyer-Peppas, the zero and the first order models were studied.

For the Korsmeyer-Peppas model [17], the following equation was considered:

$$\frac{M_t}{M_\infty} = kt^n \qquad (2)$$

where M_t is the cumulative amount of RC released at a time t, M_∞ is the cumulative amount of RC released at an infinite time (here, at 48 hr), n is an exponent characterizing the mechanism, and k is a rate constant of release. Only the initial time of release was considered for kinetics study. Table 2 shows the values of n and k for each specimen. For the EF specimens, an exponent of release (n) was in the range of 0.372-0.441. For the CF specimens, an exponent of release (n) was in the range of 0.332-0.343. The exponents of release (n) of both the EF and the CF specimens were close to 0.45, which can be categorized as a Fickian diffusion [17]. The rate constants (k) of release from the EF and the CF specimens were in the range of 0.355-0.404 and 0.286-0.290, respectively. The values of the coefficient determination (r^2) for determining how well the data fitted the model is shown in Table 3. Comparatively,the EF specimens exhibited a higher rate constant (k) than the CF specimens did.

For the zero order model, the release kinetics of RC can be characterized using the following equation:

$$C = C_0 + kt \qquad (3)$$

where C is the cumulative amount of RC released at a time t, C_0 is the initial amount of RC, and k is a rate constant of the zero order release. The rate constant (k) for RC released from the EF and the CF specimens were in the range of 4.31-4.67 and 2.21-2.32, respectively (see Table 2). The values of r^2 are shown in Table 3. Comparatively, the EF specimens exhibited a higher rate constant (k) than the CF specimens did.

For the first order model, the release kinetics of RC can be characterized using the following equation:

$$\log C = \log C_0 + \frac{kt}{2.303} \qquad (4)$$

where C is the cumulative amount of RC released at a time t, C_0 is the initial amount of RC, and k is a rate constant of the first order release. The rate constants (k) for RC released from the EF and the CF specimens were in the range of 0.080-0.107 and 0.028-0.029, respectively (see Table 2). The values of r^2 are shown in Table 3. Again, the EF specimens exhibited a higher rate constant (k) than the CF specimens did. Considering the values of r^2 in Table 3, the values of r^2 obtained from the first order were higher than those from the zero order models. This indicated that the released data fitted better with the first order than the zero order model.

From all models, the EF specimens exhibited a higher rate constant than the CF specimens did. The rate and the amount of drug release in media depend on a number of factors, e.g. degree of swelling or amount of water retention [18], amount of the weight

loss of matrix [19], solubility of drug in media [16]. In this work, the rate constants and the maximum amounts of the RC released observed from the EF specimens were greater than those from the CF specimens. This could be due to the highly porous structure of electrospun fibers that provided a greater surface area over that of films. The results were in agreement with the greater amounts of water retention observed for the EF specimens compared to those observed for the CF specimens.

Furthermore, among the EF specimens, the rate constant from all models of fibers with smaller diameters was higher than that of fibers with larger diameters. For example, the rate constants of the first order model of EF2.5RC/137nm, EF5RC/147nm, and EF2.5RC/185nm were 0.107, 0.093, and 0.080, respectively. The same trends were observed from the Korsmeyer-Peppas and the zero order models. A higher rate constant indicated a faster release of drug. Also, the maximum amount of the RC released from fibers with smaller diameters was higher than that from fibers with larger diameters. The maximum amounts of RC released from EF2.5RC/137nm, EF5RC/147nm, and EF2.5RC/185nm were 87, 82, and 75%, respectively (see Figure 4). These results were in agreement with the amount of water retention mentioned in the previous section. The fibers with smaller diameters that contributed a larger surface area exhibited a higher amount of water retention and therefore contributed to a higher rate constant and maximum amounts of release.

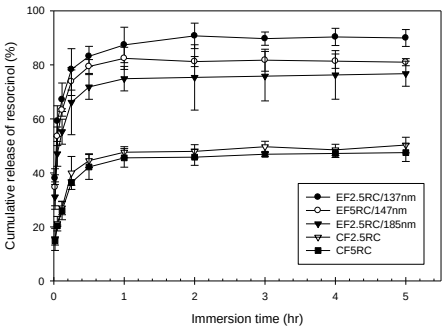

Fig.4. Cumulative release profiles of RC from EF and CF specimens.

Table 1. Viscosity of the as-prepared solutions and average diameters of electrospun fibers.

Solutions	Viscosity (cSt)	Average diameters of fibers (nm)
12PCL/2.5RC	149	137±32
12PCL/5RC	157	147±40
15PCL/2.5RC	314	185±52

Table 2. Values of k of release data of EF and CF specimens for different models of release mechanisms.

Model	k (min^{-1})				
	EF2.5RC /137nm	**EF5RC/147nm**	**EF2.5RC /185nm**	**CF2.5RC**	**CF5RC**
Korsmeyer -Peppas	0.404 (n=0.372)	0.378 (n=0.404)	0.355 (n=0.441)	0.290 (n=0.332)	0.286 (n=0.343)
Zero-order	4.67	4.55	4.31	2.21	2.32
First-order	0.107	0.093	0.080	0.028	0.029

Table 3. Values of r^2 of release data of EF and CF specimens for different models of release mechanisms.

Model	r^2				
	EF2.5RC/ 137nm	EF5RC/ 147nm	EF2.5RC/ 185nm	CF2.5RC	CF5RC
Korsmeyer-Peppas	0.988	0.990	0.991	0.994	0.943
Zero-order	0.890	0.894	0.890	0.909	0.894
First-order	0.961	0.957	0.944	0.933	0.922

4. Conclusions

In this work, RC-loaded PCL nanofibers were successfully prepared by electrospinning from PCL solutions containing either 2.5 or 5 wt.% RC (based on the weight of PCL). The average diameters of fibers were in the range of 130-190 nm. The amounts of water retention of the EF and the CF specimens were about 240-290% and 80-90%, respectively. Upon submersion in an acetate buffer (pH 5.5), both the EF and the CF specimens exhibited a burst release at the initial time followed by a gradual release to reach a plateau. The maximum release amounts from the EF and the CF specimens were about 90% and 50%, respectively. From the kinetics models including the Korsmeyer-Peppas, the zero and the first order models, the EF specimens showed higher rate constants (k) than the CF specimens did. Moreover, the rate constants (k) from the fibers with smaller diameters werre higher than those from the fibers with larger diameters.

5. Acknowledgements

This work was financially supported by the Department of Chemical Engineering, Faculty of Engineering, Thammasat University.

6. References

[1] Doshi, J. and Reneker, D.H., Electrospinning process and applications of electrospun fibers, J Electrostat, Vol.35, pp.151-160, 1995.

[2] Huang, Z.M., Zhang, Y.Z., Kotaki, M. and Ramakrishna, S., A review on polymer nanofibers by electrospinning and their applications in nanocomposites, Compos Sci Technol, Vol.63, pp.2223-2253, 2003.

[3] Fernandes, J.G., Correia, D.M., Botelho, G., Padrão, J., Dourado, F., Ribeiro, C., Lanceros-Méndez, S. and Sencadas, V., PHB-PEO electrospun fiber membranes containing chlorhexidine for drug delivery applications, Polym Test, Vol.34, pp.64-71, 2014.

[4] Canbolat, M.F., Celebioglu, A. and Uyar, T., Drug delivery system based on cyclodextrin-naproxen inclusion complex incorporated in electrospun polycaprolactone nanofibers, Colloid Surface B, Vol.115, pp.15-21, 2014.

[5] Thitiwongsawet, P. and Supaphol, P., Carbendazim-loaded electrospun poly(vinyl alcohol) fiber mats and release characteristics of carbendazim therefrom, Polymer Adv Tech, Vol.22, pp.1366-1374, 2011.

[6] Rivera, A.E., Acne scarring: A review and current treatment modalities, J Am Acad Dermatol, Vol.59, pp.659-676, 2008.

[7] Hiasa, M., Kurokawa, M., Ohta, K., Esumi, T., Akita, H., Niki, K., Yagi, Y., Echigo, N., Hatakeyama, D. and Kuzuhara, T., Identification and purification of resorcinol, an antioxidant specific to Awa-ban

(pickled and anaerobically fermented) tea, Food Res Int, Vol.54, pp.72-80, 2013.

[8] Stratford, M.R., Ramsden, C.A. and Riley, P.A., Mechanistic studies of the inactivation of tyrosinase by resorcinol, Bioorgan Med Chem, Vol.21, pp.1166–1173, 2013.

[9] Theron, S.A., Zussman, E. and Yarin, A.L., Experimental investigation of the governing parameters in the electrospinning of polymer solutions, Polymer, Vol.45, pp.2017-2030, 2004.

[10] Wutticharoenmongkol, P., Sanchavanakit, N., Pavasant, P. and Supaphol, P., Preparation and characterization of novel bone scaffolds based on electrospun polycaprolactone fibers filled with nanoparticles, Macromol Biosci, Vol.6, No.1, pp.70-77, 2006.

[11] Kanawung, K., Panitchanapan, K., Puangmalee, S., Utok, W., Kreua-ongarjnukool, N., Rangkupan, R., Meechaisue, C. and Supaphol, P., Preparation and characterization of polycaprolactone/diclofenac sodium and poly(vinyl alcohol)/tetracycline hydrochloride fiber mats and their release of the model drugs, Polym J, Vol.39, No.4, pp.369-378, 2007.

[12] Thitiwongsawet, P., Wichapeng, N. and Waiyaroj, A., Electrospun polycaprolactone fiber mats as carriers for tannic acid, Adv Mat Res, Vol.774-776, pp.896-900, 2013.

[13] Merrell, J.G., Mclaughlin, S.W., Tie, L., Laurencin, C.T., Chen, A.F. and Nair, L.S., Curcumin-loaded poly(epsilon-caprolactone) nanofibres: diabetic wound dressing with anti-oxidant and anti-inflammatory properties, Clin Exp Pharmacol Physiol, Vol.36, pp.1149-1156, 2009.

[14] Suganya, S., Senthil Ram, T., Lakshmi, B.S. and Giridev, V.R., Herbal drug incorporated antibacterial nanofibrous mat fabricated by electrospinning: An excellent matrix for wound dressings, J Appl Polym Sci, Vol.121, pp.2893-2899, 2011.

[15] Deitzel, J.M., Kleinmeyer, J., Harris, D. and Beck Tan, N.C., The effect of processing variables on the morphology of electrospun nanofibers and textiles, Polymer, Vol.42, pp.261-272, 2001.

[16] Taepaiboon, P., Rungsardthong, U. and Supaphol. P., Drug-loaded electrospun mats of poly(vinyl alcohol) fibres and their release characteristics of four model drugs, Nanotechnology, Vol.17, No.9, pp.2317-2329, 2006.

[17] Peppas, N.A. and Khare, A.R., Preparation, Structure and Diffusional Behavior of Hydrogels in Controlled Release, Adv Drug Deliver Rev, Vol.11, pp.1-35, 1993.

[18] Rujiravanit, R., Kruaykitanon, S., Jamieson, A.M. and Tokura, S., Preparation of Crosslinked Chitosan/Silk Fibroin Blend Films for Drug Delivery System, Macromol Biosci, Vol.3, pp.604-611, 2003.

[19] Zeng, J., Aigner, A., Czubayko, F., Kissel, T., Wendorff, J.H. and Greiner, A., Poly(vinyl alcohol) nanofibers by electrospinning as a protein delivery system and the retardation of enzyme release by additional polymer coatings, Biomacromolecules, Vol.6, pp.1484-1488, 2005.