ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Suitable Electrospinning Condition for Gallic Acid-loaded Cellulose Acetate Fiber

Manisara Phiriyawirut[1]*, Thawatchai Phachamud[2]

[1] Department of Tool and Materials Engineering, Faculty of Engineering, King Mongkut's University of Technology Thonburi, Bangkok, 10140, Thailand.
[2] Department of Pharmaceutical Technology, Faculty of Pharmaceutical, Silpakorn University, Nakorn Pathom, 73000, Thailand.

ABSTRACT

In this research, gallic acid-loaded electrospun cellulose acetate (CA) fibers were prepared. The average diameters of gallic acid-loaded CA fibers ranged between 702–1280 nm. Electrospun CA fibers containing 2.5% gallic acid had smooth surfaces, but gallic acid flakes were observed on the fiber surfaces with increasing gallic acid content. The effect of electrospinning processing parameters such as electrostatic potential and electrospinning distance on fiber morphology was also investigated. However, it exhibited less significant effect than gallic acid concentration. To obtain the desired gallic acid-loaded CA electrospun fiber with smooth and uniform the suitable electrospinning condition should be: electrical potential 12 kV and the electrospinning distance of 12.5 cm.
Keywords*: Electrospinning, gallic acid, cellulose acetate*

Corresponding author

ISSN: 0975-8585

INTRODUCTION

Gallic acid (3,4,5-trihydroxybenzoic acid) is a natural phenolic compound mostly found in gallnuts, sumac, witch hazel, grapes, oak bark, and green tea. It has several biological activities including antioxidant, antityrosinase, antimicrobial, anti-inflammatory and anticancer activities [1, 2 , 3]. Due to its promising antioxidant activity, gallic acid is added to various skin care products in the form of natural herbal extracts. It is also used as a standard substance in many antioxidant assays. Gallic acid has been shown to possess scavenging activities against several radicals: for example, superoxide anions, hydroxyl radicals, and singlet oxygen or peroxyl radicals. It is also able to protect cells from damage induced by UV-B or ionizing irradiation [4]. Additionally, gallic acid has shown an antibacterial property against *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and especially *Klebsiella pneumoniae* [5]. These biological activities have indicated the potential use of gallic acid as an active compound for skin or transdermal delivery systems. However, the main limitation of gallic acid is its poor water solubility (11.5 mg/mL) [6, 7]. In order to overcome this problem, electrospun fiber mats could be used as a drug delivery carrier of gallic acid; their high surface area should improve the solubility or release of gallic acid.

The electrospinning technique is a simple and versatile method which utilizes a high voltage source to inject a charge of a certain polarity into a polymer solution or melt and then accelerate it toward a collector of opposite polarity. As the electrostatic attraction between the oppositely charged liquid and collector and the electrostatic repulsions between like charges in the liquid become stronger, the leading edge of the solution changes from a rounded meniscus to a cone (the Taylor cone) [8]. A fiber jet is eventually ejected from the Taylor cone as the electric field strength exceeds the surface tension of the liquid. The fiber jet travels through the atmosphere allowing the solvent to evaporate, thus leading to the deposition of solid polymer fibers on the collector. The capacity to easily produce materials at this biological size scale has created a renewed interest in electrospinning for applications in drug delivery. Various polymers have been used as the material for fabrication into nanofiber with this technique such as poly lactide-glycolide, polycaprolactone, poly(ethylene oxide), polyvinyl alcohol, collagen, silk protein [9] and cellulose derivatives.

Cellulose acetate (CA) is one of the most important organic esters derived from cellulose. It is widely used in plastics and as fibers and membranes in many industrial applications. Cellulose acetate is manufactured by reacting cellulose with acetic anhydride using sulfuric acid as a catalyst. Liu and Hsieh [10] reported the preparation of ultrafine CA fiber mats as well as regenerated cellulose membranes by electrospinning. They found that the most suitable solvent system for preparing the CA solutions for electrospinning was 2:1 v/v acetone/dimethylacetamide (DMAc), as this mixture allowed the resulting CA solutions (i.e. 12.5–20.0 wt%) to be electrospun into fibers with average diameters ranging between ~100 nm and ~1 mm. The application of electrospun CA fiber mats as carriers for topical/transdermal delivery of drugs has been reported [11-13]. Taepaiboon et al. [11] developed electrospun CA fiber mats as carriers for topical/transdermal delivery of vitamin A acid (Retin-A) and vitamin E.

Moreover, Suwantong et al. [13] studied the use of electrospun CA fiber mats as carriers of curcumin, an herbal compound found in the plant *Curcuma longa* L.

The parameter affecting electrospinning of polymer solution may be broadly classified into polymer solution parameters, processing conditions which include the electrostatic potential, electrospinning distance (distance between tip of nozzle and collector device), temperature and ambient conditions. It was found that it is possible to create nanofibers with different morphology by varying the parameters [14]. The properties of the polymer solution have the most significant influent in the electrospinning process and the resultant fiber morphology [15]. The surface tension, the viscosity and the electrical properties of electrospinning solution have an effect on the diameter of the electrospinning fiber. Even through the processing condition have a certain influence in the fiber morphology less significant than the solution parameters, it is the external factors exerting on the electrospinning jet [16, 17].

The aim of this study is to study the effect of gallic concentration, electrical potential and electrospinning distance, distance between tip of nozzle and collector device, on the diameter and number per area o the gallic acid-loaded CA electrospun fiber mats fabricated with electrospinning technique.

MATERAILS AND METHODS

Materials

Cellulose acetate (CA; white powder; Mw = 30,000 Da; acetyl content = 39.7 wt%; degree of acetyl substitution 2.4) was purchased from Sigma–Aldrich (Switzerland). Acetone and *N, N*-dimethylacetamide (DMAc), were purchased from Labscan (Asia), Thailand. Gallic acid was purchased from Fluka.

Methods

Fiber mat preparation by electrospinning

An initial weighed amount of CA powder was dissolved in 2:1 v/v acetone/DMAc to obtain a 17% w/w CA solution, as mentioned in the literature [23]. Gallic acid-loaded CA solutions were prepared by dissolving the same amount of CA powder and gallic acid in amounts of 2.5–10 wt% (based on the weight of CA powder) in the acetone/DMAc mixture.

Electrospinning of the as-prepared solutions was carried out by connecting the emitting electrode of positive polarity from a Gamma High-Voltage Research ES30PN/M692 high voltage DC power supply to the solutions contained in a standard 5-ml syringe, the open end of which was attached to a blunt gauge-20 stainless steel needle (OD = 1.2 mm), used as the nozzle, and the collection screen was covered with aluminum foil to used as the fiber collection device. The

electrospinning was performed at room temperature. Electrostatic potential was controlled between 12, 17 and 22 kV and electrospinning distance was between 12.5, 15 and 17.5 cm for proper electrospinning conditions.

Characterizations and testing

Prior to electrospinning, the as-prepared solutions were measured for their viscosity and conductivity using a VISCO STAR Plus viscometer (Fungilab, Spain) and a CON 6/TDS 6 conductivity/TDS meter (Oakton Instruments, USA). Measurements were carried out at 25 °C, and average values for each solution were calculated from at least three measurements.

Morphological appearance of the neat and gallic acid-loaded electrospun CA fiber mats was observed using a JEOL (Japan) JSM-6380LV scanning electron microscope (SEM). The fiber mat samples were sputtered with a thin layer of gold prior to SEM observation. Diameters of the individual fibers in the as-spun fiber mats were measured directly from the SEM images using Semaphore 4.0 software (n=30).

A oneway ANOVA was used to compare the means of different data sets, and statistical significance was accept at 0.05 confidence level.

RESULTS AND DISCUSSION

Table 1 Shear viscosity and electrical conductivity of neat and gallic acid-loaded CA solutions.

Gallic acid content (%w/w)	Viscosity (cP)	Conductivity (µS)
0	498.5	4.45
2.5	542.2	4.32
5	686.2	4.28
7.5	832.1	4.13
10	973.2	4.04

Due to the limited solubility of gallic acid in CA solution, the CA solution containing 2.5–5% gallic acid was yellowish-clear; but when the content of gallic acid was increased to 7.5–10%, a colloidal solution was achieved and the color of the solution turned more yellow with increasing gallic acid content. Prior to electrospinning, both neat and gallic acid-loaded CA solutions were measured for their viscosity and conductivity; the results are summarized in Table 1.

The presence of gallic acid in the base CA solution was responsible for the observed increase in the viscosity values; but the solution conductivity was found to slightly decrease with increasing gallic acid content.

ISSN: 0975-8585

Effect of gallic acid content

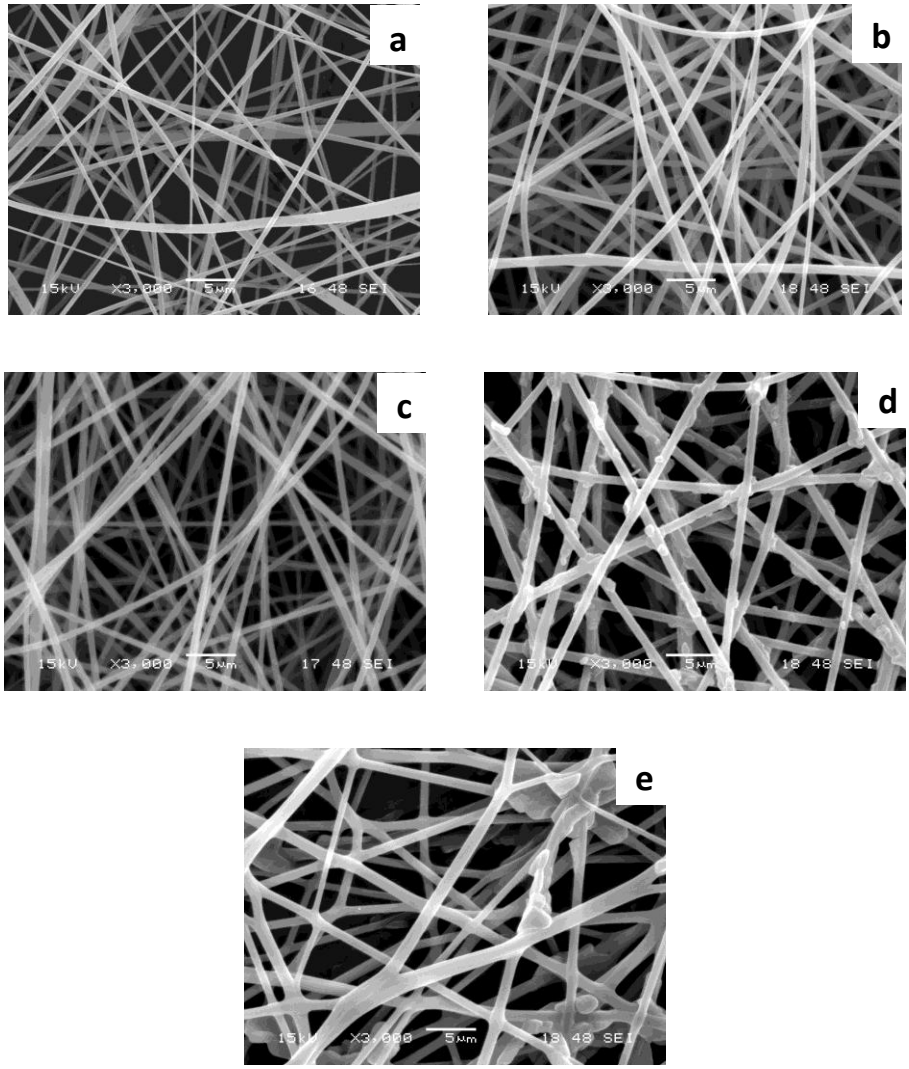


Fig 1. Scanning electron micrographs of neat and gallic acid-loaded electrospun CA fiber mats at various gallic acid contents for (a) 0% (b) 2.5% (c) 5% (d) 7.5% and (d) 10%. Note: applied electric field = 12 kV/12.5 cm (at magnification of 3000).

Electrospinning of the CA solution containing 2.5-10% gallic acid was carried out. Selected SEM images of the electrospun fibers from these solutions are shown in Fig. 1.

Clearly, cross-sectionally round fibers were obtained. The diameters of the neat CA fibers were ~539 ± 213 nm, while those of the gallic acid-loaded CA fibers ranged between ~701 ± 162 and 1057 ± 298 nm, depending on the initial amount of as-loaded gallic acid (see Table 1). Neat CA fibers were smaller than the gallic acid-loaded CA fibers because of the lower solution viscosity and higher electrical conductivity, which accelerate the stretching of the polymer during electrospinning. The addition of gallic acid to the CA solution affected its viscosity and

resulted in increasing fiber diameters. Normally, solution viscosity increases with increasing solution concentration, according to a power-law relationship; an increase in solution viscosity should thus result in the formation of fibers with greater diameters [15, 19].

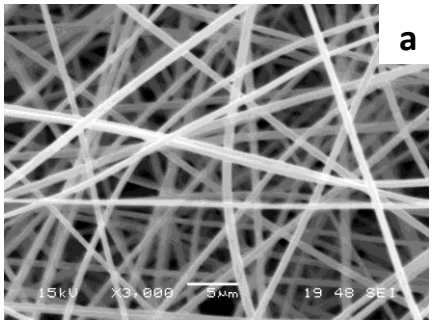
Interestingly, the aggregation of gallic acid was observed on the surfaces of these fibers, especially with gallic acid content of 7.5–10%, implying that the as-loaded gallic acid was not perfectly incorporated within the fibers. Gallic acid has poor water solubility [7]; hence its solubility in the electrospinning solution was limited. During the rapid evaporation of the solvent, phase separation took place quickly between the drug and CA. Therefore, a substantial portion of the drug aggregate migrated to the fiber surface.

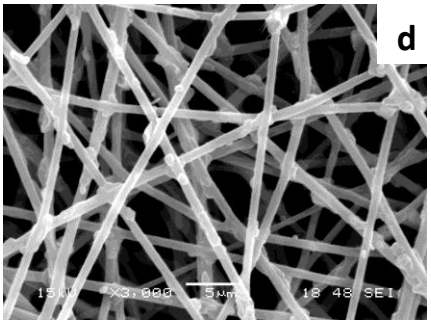
Effect of electrostatic potential

Table 2. Effect of gallic acid concentration and electrostatic potential on number of fiber/area of gallic acid-loaded CA electrospun fiber prepared at the electrospinning distance of 12.5 cm.

Gallic acid content (%w/w)	Electrical potential (kV)	Fiber diameter (nm)	Number of fiber (/100 μm^2)
0	12	539±213	17
	17	566±217	22
	22	671±234	22
2.5	12	702±162	23
	17	726±155	19
	22	686±156	19
5	12	716±160	18
	17	1280±243	10
	22	673±423	11
7.5	12	935±142	12
	17	1141±390	7
	22	1220±866	8
10	12	1057±298	11
	17	1443±307	8
	22	-	-

- : no fiber formation





ISSN: 0975-8585

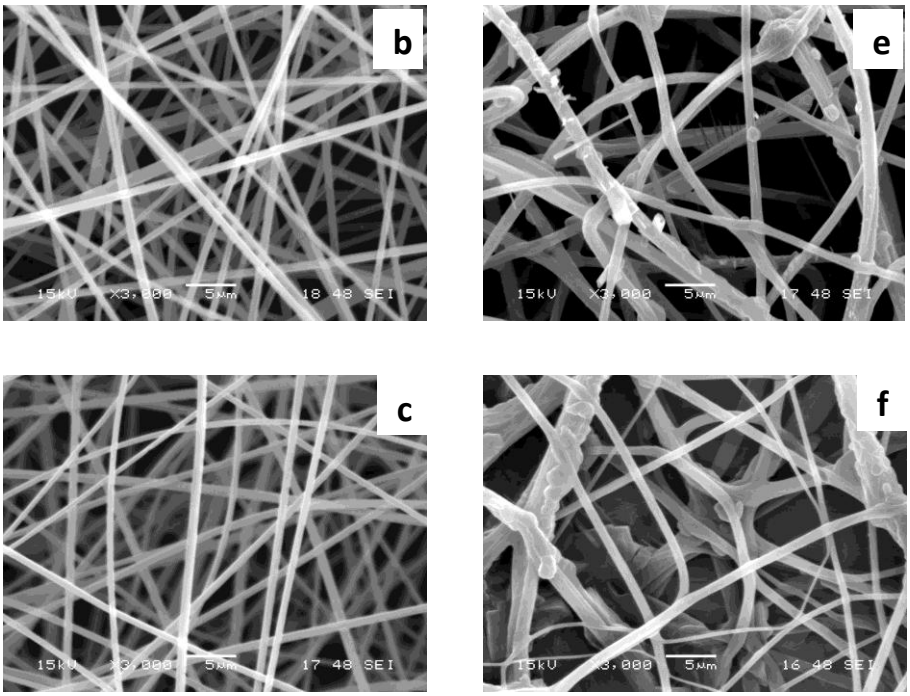


Fig 2. Scanning electron micrographs of gallic acid-loaded electrospun CA fiber mats a-c) 2.5% and d-f) 7.5% loading content, using the electrospinning distance of 12.5**cm at different electrostatic potential of a, d)** 1**2 kV, b, e) 17 kV and c, f)** 2**2 kV (at magnification of 30**0**).**

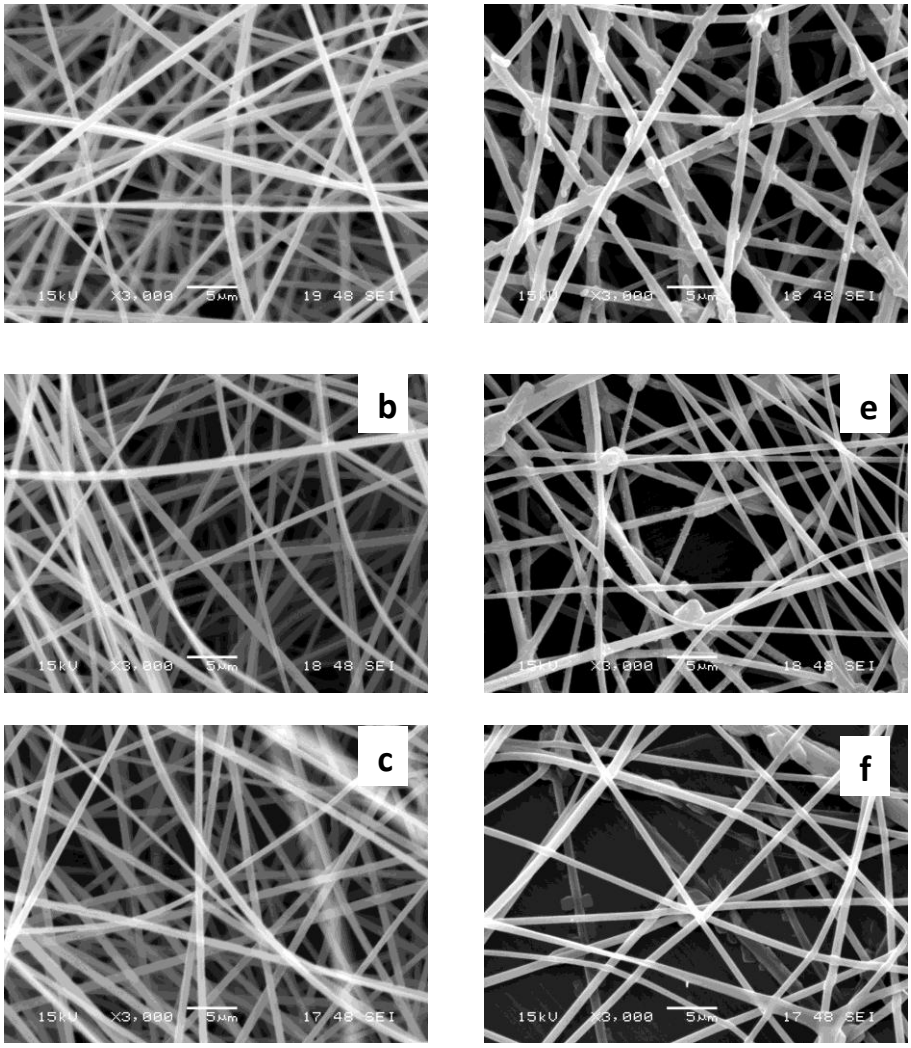
The effect of electrical potential on diameter and number per area of the gallic acid-loaded CA fiber were shown in Fig. 2 and Table 2. For CA fiber and 2.5% gallic acid-loaded CA fiber, the diameter and number of fiber per area were rather similar for the fibers prepared with different electrical potential of electrospinning. As increasing the gallic acid loading content from 5% to 10%, it was found that as increasing electrical potential form 12 to 17 kV, the fiber diameter was increased but it have no trend when electrical potential was reached at 22 kV.

As both the electrical potential and the resultant electric field have an influence in the stretching and the acceleration of the jet, they will have an influence on the morphology of the obtained fibers. In most cases, a higher electrical potential will lead to greater stretching of the solution due to the greater columbic forces in the jet as well as the stronger electric field. These have the effect of reducing the diameter of the fibers [16]. Some report claimed that the spit ability of charged droplets was enhanced owing to the increasing the electrostatic force [20]. But someone mentioned that the increasing the Coulombic repulsion force with increase in the applied electrical potential resulted the decreasing the fiber diameter whereas the increasing the electrostatic force with increase in the applied electric potential promoted the increasing the fiber diameter [21]. Krishnapp and coworker [22] reported that increasing voltage could increase the beads density, which at an even higher electrical potential, the beads will join to form a larger diameter fiber. Typically the increasing spinning electrical potential could elongate the jet and also drawn more solution out of the nozzle [15]. Interestingly, as increasing

ISSN: 0975-8585

electrical potential, wide size distribution of the fiber was obtained. It was due to the unbalanced effect of Coulombic repulsion and other forces which result in the formation of secondary jet during electrospinning.

Effect of electrospinning distance



The effect of electrospinning distance on diameter and number per area of the gallic acid-loaded CA fiber were shown in Fig. 3 and Table 3. Depending on the solution properties, the effect of varying the electrospinning distance may or may not have a significant effect on the fiber morphology. For CA fiber and 2.5% gallic acid-loaded CA fiber, the diameter and number of fiber per area were rather similar for the fibers prepared with different electrospinning distance of electrospinning. As increasing the gallic acid loading content to 5%, it was found that as increasing electrospinning distance form 12.5 to 17.5 cm, the fiber diameter was increased but it have no trend when the gallic loading content was 7.5%. In the opposite way, as the gallic acid loading content was 10%, the diameter of the electrospun fiber was decreased with increasing electrospinning distance.

ISSN: 0975-8585

Table 3. Effect of electrospinning distance on number of fiber/area of gallic acid-loaded CA electrospun fiber prepared at the electrical potential of 12 kV

Gallic acid content (%w/w)	Electrospinning distance (cm)	Fiber diameter (nm)	Number of fiber (/100 μm^2)
0	12.5	539±213	17
	15	769±239	14
	17.5	775±210	17
2.5	12.5	702±162	23
	15	750±195	22
	17.5	759±139	21
5	12.5	716±160	18
	15	565±117	25
	17.5	1053±244	12
7.5	12.5	935±142	12
	15	746±165	12
	17.5	849±320	8
10	12.5	1057±298	11
	15	945±300	10
	17.5	647±160	8

In electrospinning principle, varying electrospinning distance will have direct influence in both fight time and the electric field strength. In circumstance, increasing the distance results in a decrease in the average fiber diameter [23]. The longer distance means that there is a longer fight time for the solution to be stretched before it is deposited on the collector [17]. However, there are cases where at a longer distance, the fiber diameter increases. This is due to the decrease in the electrostatic field strength resulting in less stretching of the fibers on the collector [24]. In some cases, changing the distance has no significant effect of the fiber diameter [25]. Because the difference value of distance was rather small the apparent of this effect on fiber characteristic was not found. Typically, the lower number of fiber per area should be obtained when there was the larger electrospinning distance since the stretched polymer deposited in larger area and thereafter there was the lower repetition of fiber deposition.

From the above result, the suitable condition to prepare the gallic acid-loaded CA electrospun fiber was the using electrical potential 12 kV and the electrospinning distance of 12.5 cm. The obtained electrospun fiber prepared with that condition was continuous and homogeneous in size and dispersion.

CONCLUSION

Gallic acid-loaded electrospun cellulose acetate (CA) fiber mats were prepared by electrospinning technique under a fixed electric field of 12kV/ 12.5cm. CA solution (%17w/v) in 2:1v/v acetone/*N,N*-dimethylacetamide was used as the base spinning solution, into which gallic acid in amounts of 2.5– % 10w/w (based on the weight of CA) was added to prepare the gallic acid spinning solutions. The electrospun fibers from these solutions were found to be

cross-sectionally round with smooth surfaces for low gallic acid loading content, but with aggregated gallic acid flakes observed on the surfaces of fibers for high gallic acid loading content. The gallic acid loading content was the main factor affected the morphological characteristic, diameter, number of fiber per area of the gallic acid-loaded CA electrospun fiber. The average diameters ranged between 702 and 1280 nm, and were found to increase with increasing gallic acid loading content. The effect of electrospinning processing parameters such as electrostatic potential and electrospinning distance on fiber morphology was also investigated. However, it has less significant effect than gallic acid concentration. To obtain the desired gallic acid-loaded CA electrospun fiber with smooth and uniform the suitable electrospinning condition should be: electrical potential 12 kV and the electrospinning distance of 12.5 cm.

ACKNOWLEDGEMENTS

The financial support was provided from the Research, Development and Engineering (RD&E) Fund through National Nanotechnology Center (NANOTEC), National Science and Technology Department Agency (NSTDA), Thailand (Project No. NN-B-22-TT2-17-51-22). The authors would like to acknowledge the facility support from Department of Tool and Materials Engineering, Faculty of Engineering, King Mongkut`s University of Technology Thonburi and Faculty of Pharmacy, Silpakorn University, Thailand. The authors would like to thank to Mr. Chaloemphon Chumpradid, Mr. Panuwat Tanasanskulwong, and Mr. Teeranan Wandee for their help.

REFERENCES

[1] Krogh R, Yunes RA, Andricopulo AD, II Farmaco 2000; 55: 730-35.
[2] Lu Z, Nie G, Belton PS, Tang H, Zhao B. Neurochem Inter 2006; 48: 263-74.
[3] Shahrzad S, Aoyagi K, Winter A, Koyama A, Bitsch I. J Nutr 2001; 131: 1207-10.
[4] Sawa T, Nakao M, Akaike T, Ono K, Maeda H. J Agric Food Chem 1999; 47: 397-402.
[5] Rodríguez Vaquero MJ, Alberto MR, Manca de Nadra MC. Food Control 2007; 18: 93-101.
[6] O'Neil MJ, Smith A, Heckelman PE, Obenchain JR, Gallipeau JAR, D'Arecca MA, eds. Gallic acid. In: The Merck index 13th ed. Merck research laboratories, NJ, 2001, pp 261.
[7] Hansen CM, Just L. Ind Eng Chem Res 2001; 40: 21-25.
[8] Reneker DH, Chun I. Nanotechnology 1996; 7: 216.
[9] Won HP, Lim J, Dong IY, Hudson S. Polymer 2004; 45: 7151-57.
[10] Liu H, Hsieh YL. J Polym Sci Polym Phy 2002; 40: 2119-29.
[11] Taepaiboon P, Rungsardthong U, Supaphol P, Eur J Pharm Biopharm 2007; 67: 387-97.
[12] Tungprapa S, Jangchud I, Supaphol P. Polymer 2007; 48: 5030-41.
[13] Suwantong O, Opanasopit P, Ruktanonchai U, Supaphol P. Polymer 2007; 48: 7546-57.
[14] Ramakrishna S, Fujihara K, Teo WK, Lim TC, Ma Z. An introduction to electrospinning and nanofibers, World Scientific, Singapore, 2005, pp. 90-116.
[15] Mit-uppatham C, Nithitanakul M, Supaphol P. Macromol Chem Phys 2004; 205: 2327-38.

[16] Lee JS, Choi KH, Ghim HD, Kim SS, Chun DH, Kim HY, Lyoo Ws. J Appl Polym Sci 2004; 93: 1638-46.
[17] Reneker DH, Yarin AL, Fong H, Koombhongse S. J Appl Phys 2000; 87: 4531-47.
[18] Taepaiboon P, Rungsardthong U, Supaphol P. Nanotechnology 2006; 17: 2317-29.
[19] Mit-uppatham C, Nithitanakul M, Supaphol P. Macromol Symp 2004; 216: 293-99.
[20] Ding B, Kim HY, Lee SC, Lee DR, Choi KJ. F Fibers Polym 2002; 3: 73-9.
[21] Supaphol P, Chuangchote S. J Appl Polym Sci 2008; 108: 969-78.
[22] Krishnapp RVN, Desai K, Sung CM. J Mater Sci 2003; 38: 2357-65.
[23] Ayutsede J, Ganhi M, Sukigara S, Micklus M, Chen HE, Ko F. Polymer 2005; 46: 1625-34.
[24] Zhao SL, Wu XH, Wang LG, Huang Y. J Appl Polym Sci 2004; 91: 242-46.
[25] Qiang L, Jia Z, Yang Y, Wang L, Guan Z. Inter Conf Solid Dielectrics 2007, 215-8.