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A Color-Detectable Vitamin C Controlled-Release System Fabricated Using Electrospinning

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A Color-Detectable Vitamin C Controlled-Release System Fabricated Using Electrospinning

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Abstract: This study develops a vitamin C controlled-release system trackable via color changes as a function of vitamin C release. The system is composed of coaxial microfibers prepared via coaxial electrospinning, with a core of poly(ethylene oxide) (PEO) incorporating vitamin C, and a shell composed of polycaprolactone (PCL) containing polydiacetylene (PDA) as the color-changing material. The shell thickness was controlled by adjusting the amount of PCL ejected during electrospinning, allowing the regulation of the release rate of vitamin C. When vitamin C added to PEO penetrated the PCL layer, the color of PDA changed from blue to red, indicating a color change. The results of this study can be applied to devices that require immediate detection of vitamin C release levels.

Keywords: vitamin C; controlled-release system; electrospinning; polydiacetylene

1. Introduction

Vitamin C, also known as L-ascorbic acid, is a water-soluble vitamin that plays a crucial role in the development of various organs in the body. It is involved in collagen synthesis and skin formation, and it acts as an antioxidant by preventing the oxidation of various substances within the body. However, vitamin C is prone to oxidation owing to its antioxidant properties. Therefore, research and applications involving vitamin C require significant effort to stabilize it and prevent its degradation [1–3].

Encapsulation using polymeric materials is commonly employed for stabilizing unstable organic substances [4–6]. Using widely known techniques such as solvent evaporation, spray drying, and electrospinning have been widely used for this purpose. In solvent evaporation, the target organic substance solution is dispersed in a larger volume of water by intense stirring, and the polymer solution is slowly added dropwise. During this process, the organic solvent evaporates, encapsulating the organic substances within the polymer matrix. Although this method provides clean encapsulation, it presents challenges when handling substances that completely dissolve in water, making it less suitable for water-soluble compounds such as vitamin C [7–10].

Spray drying involves spraying an aqueous solution containing the target organic substance and the polymer material into air. Encapsulation occurs when the solvent evaporates. Although this method allows the encapsulation of water-soluble organic substances, the resulting microcapsules often possess rough surfaces, leading to inferior encapsulation compared with the solvent evaporation method [11,12].

Electrospinning produces microfibers by subjecting a polymer solution to a high-voltage electric field. Encapsulation can be achieved by incorporating a target organic substance into a polymer solution. The fibers formed by electrospinning have diameters ranging from tens of nanometers to several micrometers, resulting in structures with large surface areas. Consequently, when the electrospun microfiber was used in controlled release experiment, an initial burst-release phenomenon occurs, which is characterized by a significant release of the encapsulated substance during the initial stages. This phenomenon has been observed in various organic substances and polymers [13,14].

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To achieve complete encapsulation, this study employed a coaxial electrospinning system, in which the encapsulating material forms the outer shell of the coaxial structure. By controlling the amount of polymer material ejected during electrospinning, the thickness of the outer shell can be adjusted, enabling the regulation of the release rate of the encapsulated substance.

Polydiacetylene (PDA) is a conjugated polymer in which double and triple bonds are alternately conjugated. PDA exhibits color changes in response to external stimuli, making it suitable for various sensor applications. Previous studies have reported the development of sensor systems using PDA [15–19].

This study aimed to manufacture a system that incorporates PDA into the outer part of a coaxial electrospinning system. By modulating the release of vitamin C, this system exhibits a color change from blue to red, enabling visual tracking of the release process. This study focused on developing a visually trackable vitamin C controlled-release system. Coaxial electrospinning was employed to fabricate coaxial microfibers in which poly(ethylene oxide) (PEO) and vitamin C form the core fiber, and polycaprolactone (PCL) with PDA form the shell fiber. The release rate of vitamin C was regulated by adjusting the amount of PCL injected during electrospinning. The color change from blue to red resulting from the passage of vitamin C through the PCL layer can be utilized in devices requiring immediate detection of vitamin C penetration.

2. Experimental

2.1. Materials and Equipment

PCL (Mn 80,000), 10,12-pentacosadiynoic acid (PCDA), PEO (Mv 900,000), L-ascorbic acid, *N*-hydroxysuccinimide (NHS), *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide (EDC), ethylenediamine, dimethylformamide (DMF), chloroform, and dichloromethane (DCM) were purchased from Sigma-Aldrich. The ultraviolet (UV) spectra were obtained using a Shimadzu UV-1800 spectrophotometer (Kyoto, Japan). The ^{1}H NMR spectra were obtained using a Bruker DRX 300 spectrometer (Billerica, USA). The coaxial fibers were fabricated using NanoNC ESR 100D electrospinning equipment (Seoul, Korea), and the electrospun fiber mats were collected on a NanoNC-DC90H drum-type collector (Seoul, Korea) with a 94.5-mm diameter. The coaxial nozzle specifications for the electrospinning process are listed in Table 1. The fibers were imaged using an Olympus BX53MRF-S optical microscope (Tokyo, Japan). The dialysis bag purchased from Sigma-Aldrich had a molecular weight cut-off of 14,000.

Table 1. Specifications of the electrospinning nozzle.

Type of nozzle	Position	Gauge (G)	Diameter (mm)	
			ID	OD
DC	core	25	0.26	0.52
	shell	18	0.92	1.28

ID: inner diameter; OD: outer diameter.

2.2. Synthesis of N-(2-aminoethyl)pentacosa-10,12-diynamide (AEPCDA)

AEPCDA was synthesized as described in previous publications [20–22]. PCDA (3.75 g, 10.0 mmol), EDC (2.88 g, 15.0 mmol), and NHS (1.50 g, 13.0 mmol) were agitated in 20 mL of DCM at 30°C for 3 hours to synthesize PCDA-NHS. DCM was evaporated under vacuum, and the residue was purified by extraction with ethyl acetate. PCDA-NHS (3.73 g, 7.91 mmol) was obtained as a white solid (yield: 79.1%). PCDA-NHS (2.50 g, 5.29 mmol) in 50 mL of DCM was added very slowly to ethylenediamine (1.00 g, 16.6 mmol) in 100 mL of DCM to synthesize the AEPCDA. The reaction continued for 6 hours at 30°C, then the precipitate was filtered out and DCM in the residual filtrate was evaporated under vacuum. The product was extracted from the residue using DCM, and the solution was washed with brine twice. The product was purified via recrystallization with DCM. AEPCDA (1.21 g, 2.91 mmol) was obtained as a white solid (yield: 55.0 %).

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^{1}H NMR ($CDCl_3$): 0.90 (t, 3H), 1.28 (s, 26 H), 1.45-1.54 (m, 6H), 2.15 (t, 2H), 2.25 (t, 4H), 2.81 (m, 2H), 3.37 (m, 2H), 4.58 (s, 2H), 6.45 (s, 1H).

2.3. Formation of PDA Vesicle Solution

AEPCDA (20.8 mg, 0.500×10^{-4} mol) was dissolved in 10 mL of chloroform in a 100 mL round-bottom flask. Chloroform was evaporated under vacuum, leaving AEPCDA as a thin membrane at the bottom of the flask. After completely evaporating of chloroform, 50 mL of water was added to the flask. The mixture was sonicated at 70°C for 30 minutes to prepare a 1.00 mM AEPCDA vesicle solution. AEPCDA was photopolymerized by transferring the solution to a Petri dish and subjecting it to UV irradiation (254 nm) for 30 seconds.

2.4. Electrospinning

Electrospinning was performed using a coaxial nozzle. The applied voltage was 14.0 kV, and the distance between the nozzle and the collector was 15.0 cm. The solution for core formation was prepared as follows: 0.350 g of PEO was dissolved with rapid stirring (1,500 rpm) and 150 mg of vitamin C was added. After evaporating the solution in a rotary evaporator, 10.0 mL of $CHCl_3$ was added to the flask and processed for 15 hours in a bath sonicator to produce the core-forming solution. The shell-forming solution was prepared by adding 1.20 g of PCL to a mixture of 8.0 mL DCM and 2.0 mL DMF as solvent, and rapidly stirring the mixture (1,500 rpm). To observe the color change using PDA, 20 mg of AEPCDA was added to the PCL solution in five 4.0 mg portions during the preparation of the shell-forming solution.

2.5. Vitamin C Release

The vitamin C release experiment used a mat composed of electrospun fibers with a PEO core containing vitamin C and a PCL shell. Initially, electrospinning was conducted for 1 h to prepare the fibers, which were then collected and placed in a dialysis bag. The release experiment was performed in 1,000 mL of distilled water. The amount of released vitamin C was measured by determining the absorbance at 266 nm in the UV spectrum.

3. Results and Discussion

Commercially available PCDA is the most commonly used diacetylene compound in sensing research utilizing PDA. It has a diacetylene group in the middle and a carboxylic acid functional group at the end, with a carbon chain length of 25. Prior to entering this study, PCDA was sonicated in water to form a vesicle solution. However, when this solution was used to detect vitamin C, no color changes were observed. Therefore, a synthesis experiment was conducted to convert the carboxylic acid functional group at the end of PCDA into an amine group. PCDA-NHS was synthesized by reacting PCDA with EDC and NHS. Subsequently, AEPCDA was synthesized by reacting the thus formed PCDA-NHS with ethylenediamine. This process is illustrated in Figure 1.

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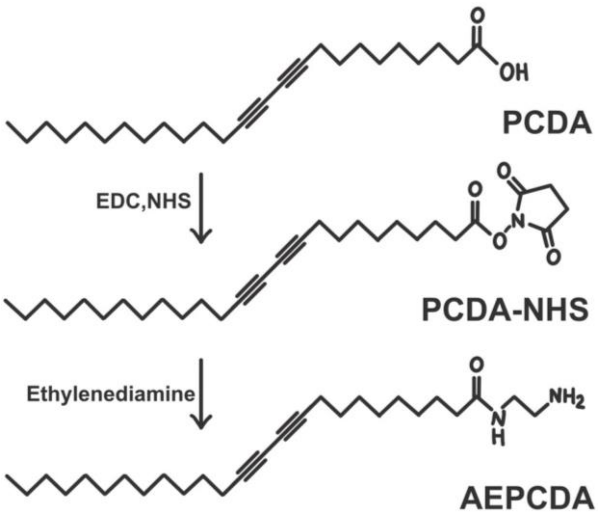

Figure 1. AEPCDA synthesis procedure.

3.1. Vitamin C Detection Using AEPCDA Vesicle Solution

First, the ability to detect vitamin C using AEPCDA synthesized in this study was assessed using the AEPCDA vesicle solution**.** When the vesicles were prepared using AEPCDA, a color change from blue to red was observed as vitamin C penetrated from the external environment to the inside of the vesicle, as shown in Figure 2. The results of this experiment are shown in Figure 3.

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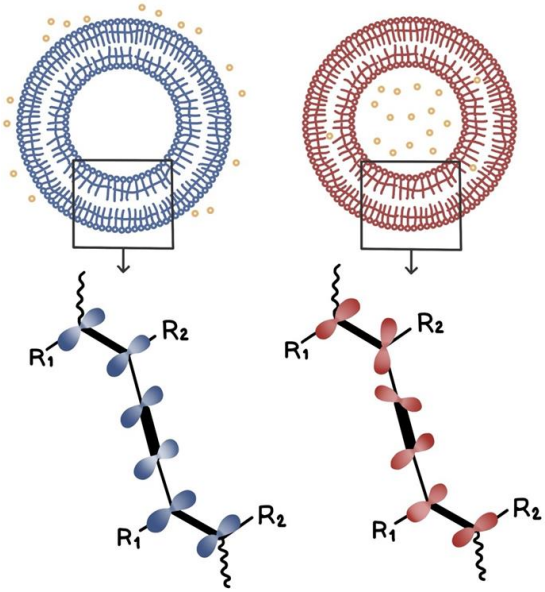

Figure 2. The change in vesicle color from blue to red upon vitamin C penetration from the outside to the inside of the vesicle.

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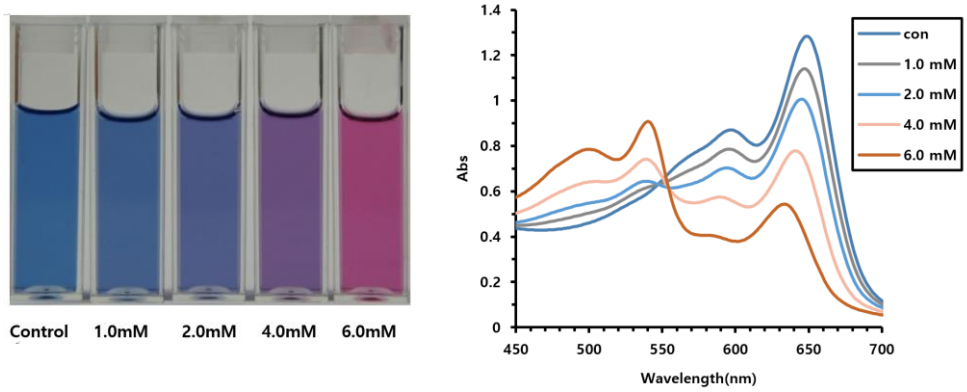

(1) (2)

Figure 3. Vitamin C detection using AEPCDA vesicle solution. 1) Photographs of color change, 2) UV spectrum of the vesicle solution.

From the image in Figure 3, it can be observed that color changes can be detected from a vitamin C concentration of 2.0 mM. At 4.0 mM, a color change to purple, while at 6.0 mM, a color change to red completely. Furthermore, in their UV spectra, the blue UV absorption between 630 and 650 nm decreases with increasing vitamin C concentration, while the red absorption near 540 nm gradually increases.

No color changes were detected in our preliminary experiment using PCDA. Conversely, color changes from blue to red were detected using AEPCDA. This is presumably induced by the strong interactions between the acid functional group of vitamin C molecules and the amine group at the end of AEPCDA.

3.2. Detecting Vitamin C Release from Microfiber Mat

In this experiment, vitamin C release was detected using a mat of coaxially electrospun fibers with a vitamin C-containing PEO core and an outer shell of PCL containing PDA as the sensing material undergoing the color change. Thus, when vitamin C in the inner part is released into the outer part (PCL) of the fiber, it encounters PDA, resulting in the color change.

Figure 4 shows the electrospinning device with coaxial nozzle, and the experimental process in this study.

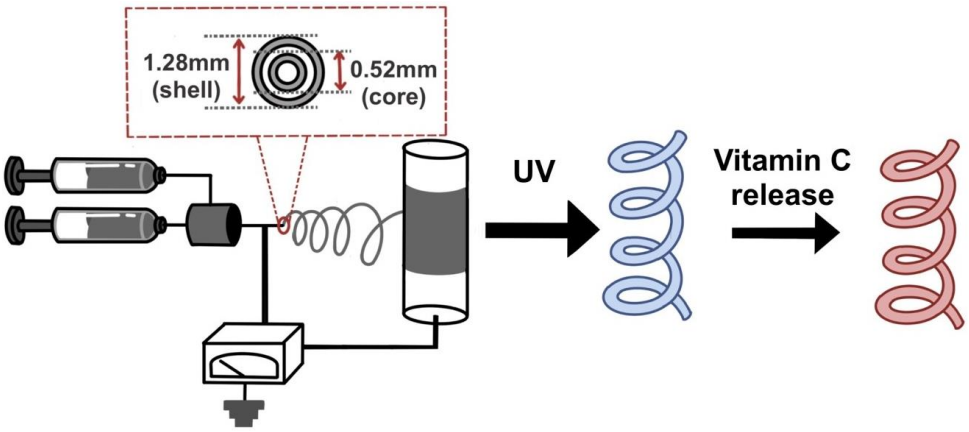

Figure 4. Electrospinning device and coaxial nozzle, along with the experimental process.

Three coaxial microfiber types were used in this experiment, and the electrospinning conditions and fiber thicknesses summarized in Table 2. The morphology of the formed fibers was observed using a microscope at magnifications of 200x and 1000x, and the results are shown in Figure 5.

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Table 2. Electrospinning conditions* and average diameter of the formed fiber.

	Flow rate (shell)	Average diameter
a	1.5 mL/h	3.9 ± 0.6 μm
b	2.0 mL/h	4.8 ± 0.6 μm
c	2.5 mL/h	5.7 ± 0.7 μm

*Flow rate (core) 0.50 mL/h.

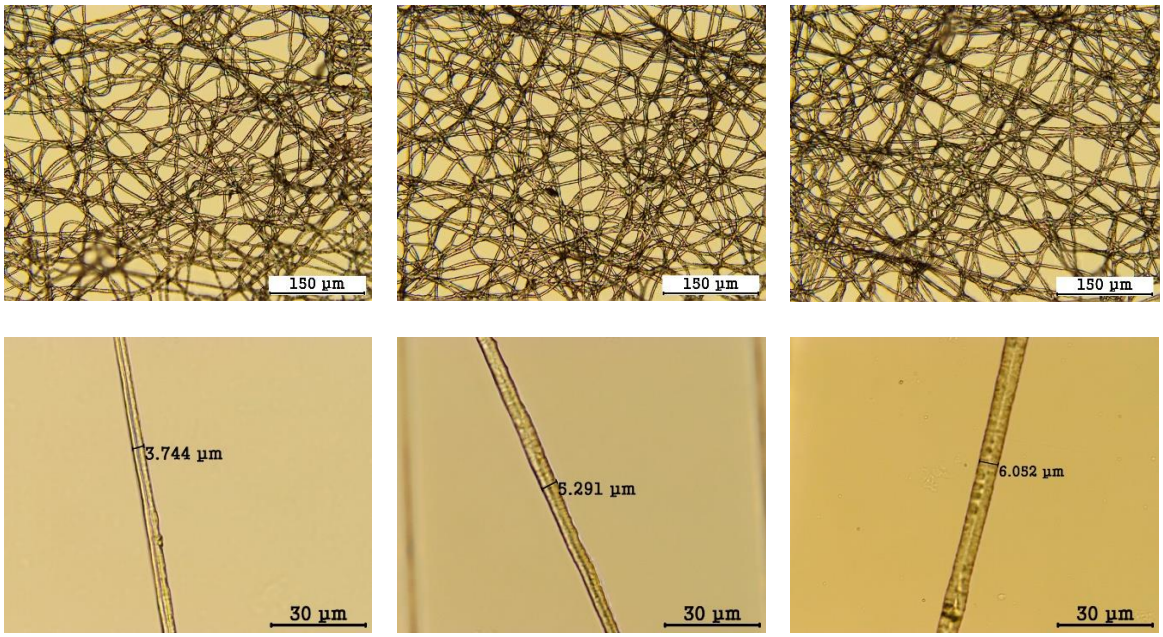

Figure 5. Microscope images of the fibers. a, b, and c refer to the same variables as in Table 2.

The average fiber diameters obtained from Figure 5 for a, b, and c are 3.9 ± 0.6 μm, 4.8 ± 0.6 μm, and 5.7 ± 0.7 μm, respectively. Indicating that increasing the flow rate of the shell portion of the coaxial spinneret results in an increase in the size of the shell portion, leading to an overall increase in the fiber diameter.

The IR spectra of the fibers were obtained from the samples shown in Table 2 (b) and compared with the IR spectra obtained from the basic materials of the fibers: PEO and PCL. The results are shown in Figure 6.

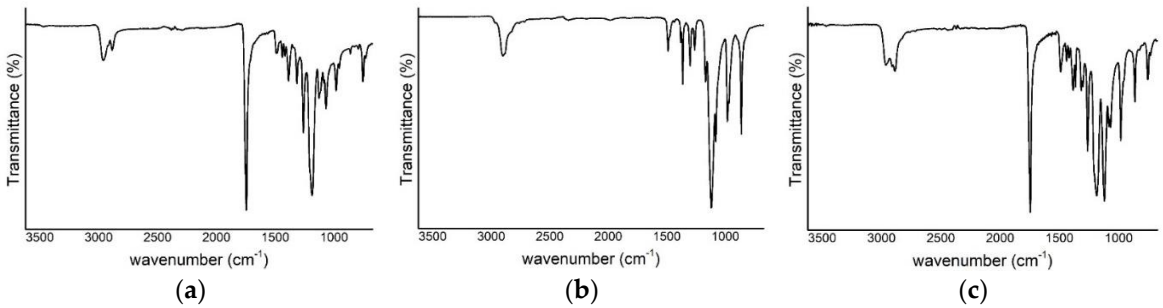

Figure 6. IR spectrum of (a) PCL, (b) PEO, and (c) electrospun fiber with PCL/PEO.

The IR spectrum of PCL in Figure 6(a) includes absorption peaks at 1723 cm^{-1} and 1164 cm^{-1}, indicating absorption by the carbonyl and C–O group, respectively. The IR spectrum of PEO in Figure 6(b) shows an absorption peak at 1098 cm^{-1}, indicating absorption by the C–O group. These absorption peaks are all visible in the IR spectrum of the electrospun fiber with PCL/PEO (Figure 6 (c)), indicating that the fiber was composed of both PCL and PEO.

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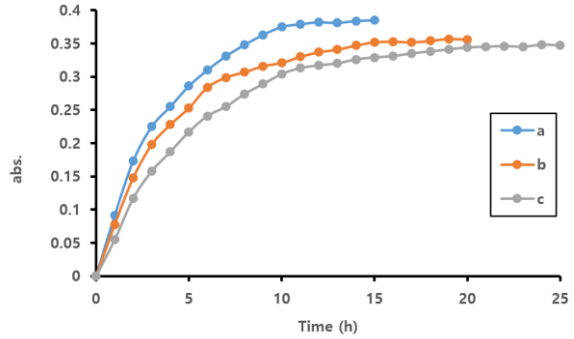
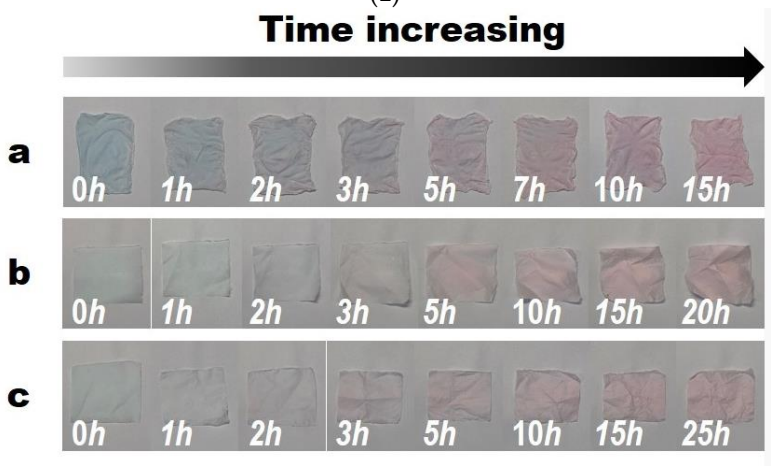
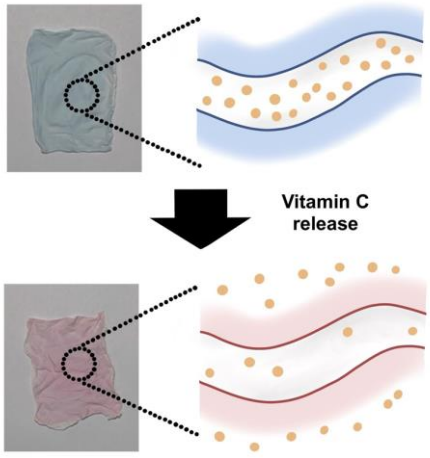
The release experiments were conducted, and the concentrations of released vitamin C over time are plotted in Figure 7. Additionally, the color changes resulting from the release of vitamin C at each point are shown in Figure 7.



(1)



(2)



(3)

Figure 7. (1) Changes in the concentration of released vitamin C with time, (2) color change of the fiber-mat due to the increase in the amount of penetrating vitamin C, (3) the process of color change occurring. a, b, and c refer to the same variables as in Table 2.

From Figure 7, it is evident that the release rate can be controlled by varying the shell thickness. In case a, nearly all the releasable vitamin C is released after 10–11 hours. In contrast, this occurs after 15–16 hours and 21–22 hours in case b and c, respectively. Additionally, the final amount of released vitamin C differs slightly in the order of a > b > c. This is attributed to the presence of captured vitamin

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C that is difficult to release, with quantities of captured vitamin C being in the order c > b > a. Furthermore, the color changes observed at each point in Figure 7(2) show that the color changes from blue to red as time progresses.

4. Conclusions

In this study, a new system that monitors vitamin C release through color change was prepared by electrospinning. Coaxial nozzles were used in electrospinning to prepare fibers with core–shell structures. The core was composed of PEO incorporating vitamin C, and the shell was composed of PCL containing polydiacetylene as the indicator undergoing color change upon contact vitamin C. Polydiacetylene was synthesized using PCDA derivatives in which the end functional group of PCDA was changed to an amine group. The obtained polydiacetylene exhibited a color change from blue to red upon direct contact with vitamin C. Similarly, the release experiments using fiber mats prepared using coaxial nozzles revealed a color change from blue to red upon vitamin C release.

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Conflicts of Interest: The author declares no conflicts of interest.

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