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Mussel Inspired Protein-Mediated Surface Modification to Electrospun Fibers and their Potential Biomedical Applications

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Mussel inspired protein-mediated surface modification to electrospun fibers and their potential biomedical applications

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Abstract: Mussel inspired proteins have been demonstrated to serve as a versatile biologic adhesive with numerous applications. The present study illustrates the use of such Mussel inspired proteins (polydopamine) in the fabrication of functionalized bio-inspired nanomaterials capable of both improving cell response and sustained delivery of model probes. X-ray photoelectron spectroscopy (XPS) analysis confirmed the ability of dopamine to polymerize on the surface of plasma-treated, electrospun poly(ε-caprolactone) (PCL) fiber mats to form polydopamine coating. Transmission electron microscopy (TEM) images demonstrated that self-polymerization of dopamine was induced by pH shift and that the thickness of polydopamine coating was readily modulated by adjusting the concentration of dopamine and reaction time. Polydopamine coatings were noted to affect the mechanical properties of underlying fiber mats, as mechanical testing demonstrated a decrease in elasticity and increase in stiffness of polydopamine-coated fiber mats. Polydopamine coatings were also utilized to effectively

immobilize extracellular matrix proteins (i.e, fibronectin) on the surface of polydopamine-coated, electrospun fibers, resulting in enhancement of NIH3T3 cell attachment, spreading and cytoskeletal development. Comparison of release rates of rhodamine 6G encapsulated in coated and uncoated PCL fibers also confirmed that polydopamine coatings modulate the release rate of loaded payloads. We further demonstrate the significant difference of rhodamine 6G adsorption kinetics in water between PCL fibers and polydopamine-coated PCL fibers. Taken together, polydopamine-mediated surface modification to electrospun fibers may be an effective means of fabricating a wide range of bio-inspired nanomaterials with unique properties for use in tissue engineering, drug delivery, and advanced biomedical applications.

Key Words: polydopamine coating; electrospun fibers; cell adhesion; sustained release; adsorption

INTRODUCTION

Prior research has demonstrated that specialized adhesive proteins enable mussels to adhere to a variety of materials with great affinity under aqueous conditions.[1] Reports suggest that repeated 3,4-dihydroxy-L-phenylalanine-lysine sequences present in mussel foot proteins may contribute to the unique strength and resilience of this marine adhesive.[2] Towards this end, polymerization of dopamine, a catecholamine moiety containing similar adhesive sequences, may provide a unique substrate useful in functionalizing and/or modifying a variety of biomaterial surfaces. Presently, polydopamine has been used to modify the surface of various materials including metals, oxides, ceramics, semiconductors, carbons, polymers and even living cells to great effect.[3-5] Specifically, the polydopamine coating has been demonstrated to provide a versatile platform for secondary immobilization of many functional molecules.[6-8] Yet, despite this

increased interest, few studies have examined polydopamine functionalization of electrospun, nanofabricated biomaterials.

Electrospinning is an enabling technique which can produce fibrous materials on the order of nanometers from a rich variety of materials including polymers, composites, and ceramics.[9] Resulting electrospun matrices possess a high surface area to volume ratio, flexibility in surface functionalities, and superior mechanical performance optimal for numerous applications in tissue engineering, drug delivery and filtration.[10] Surface modification of electrospun fibers plays a central role in biomedical applications as the fiber surface provides a direct interface to adherent cell populations and local tissues. Current approaches for the functional decoration of fibers include use of di-NH_2-PEG and heparin linkers, EDC/NHS chemistry, layer-by-layer assembly, and surface binding peptides.[12-14] Unfortunately, existing methods present significant limitations which preclude widespread adoption, including the need for specific modifiers, time-consuming and complex processing steps, and limited substrate versatility,

Development of a simple and versatile method of functionalizing electrospun fibers remains an ongoing area of interest in the biomedical community. One recent study proposed the concept of modifying the surface of electrospun PCL fibers with polydopamine for use in engineering novel vascular grafts.[15] While demonstrating the potential of this unique approach, this prior study did not thoroughly explore that ability to control coating thickness and uniformity, or the effect of surface coating conditions (e.g., the effect of surface hydrophilicity, and different surface compositions). Prior studies have also overlooked the effect of polydopamine coatings on the release of encapsulated or conjugated molecules loaded into/onto functionalized fiber materials. Examination of the effect of polydopamine coating on the kinetics of drug delivery via electrospun fiber scaffolds serves as an important milestone in evaluating the clinical potential of

such coatings and devices. The primary objective of the present study was to develop a method for modifying electrospun fibers with uniform coatings of polydopamine of controllable thickness. Additionally, the present study aimed to examine the secondary immobilization of biomacromolecules on polydopamine-coated fibers as means of improving the biomaterial interface and explore the biomedical potential of resulting materials. PCL and poly($_L$-lactide) (PLA) were selected as core materials for fiber fabrication due to their noted biocompatibility, prolonged resorption, ease of processing, and demonstrated use in biomedical applications.[16,17]

MATERIALS AND METHODS

Fabrication of electrospun fibers

Fibers were produced utilizing a standard electrospinning setup, described previously.[18-20] PCL (Mw=80 kDa, Sigma-Aldrich, St. Louis, MO) was dissolved in a solvent mixture consisting of dichloromethane (DCM) and N, N-dimethylformamide (DMF) (Fisher Chemical, Waltham, MA) with a ratio of 4:1 (v/v) (at a concentration 10% (w/v)). PLA (Mw=85-160 kDa, Sigma-Aldrich) was dissolved in a solvent mixture consisting of DCM and DMF with a ratio of 3:2 at a concentration of 5%. For the fabrication of rhodamine 6G (Sigma-Aldrich)-loaded PCL fibers, 5% (w/w) rhodamine 6G was mixed with the polymer solution as described previously.[21] Solutions were pumped at a flow rate of 0.5 mL/h using a syringe pump while a potential of 12 kV was applied between the spinneret (a 22-gauge needle) and a grounded collector located 12 cm apart from the spinneret. Various collectors were employed to generate fiber assemblies of different orders / patterns. Random fibers were directly collected using cover glass slips. A stainless steel frame containing an air gap (2 cm × 10 cm) was used to obtain uniaxially-aligned fiber samples. Subsequently, aligned fibers were transferred from the collector and adhered to

cover glass slips. Fiber materials were then treated with a plasma cleaner (PDC-32G, Harrick Plasma, Ithaca, NY) for 8 min at a medium setting. Subsequently, fiber materials were immersed in either 0.2 mg/mL or 2 mg/mL dopamine·HCl (10 mM) in Tris buffer (pH 8.5) for either 0.5 h, 4 h or 72 h, respectively. Polydopamine-coated fiber materials were then washed with DI water to remove excess monomer.

Morphological characterization

The morphology and structure of polydopamine-coated and un-coated fiber samples were characterized by scanning electron microscopy (SEM) (FEI, Nova2300, Oregon). To avoid charging, polymer fiber samples were fixed on a metallic stud with double-sided conductive tape and coated with platinum for 40 seconds in vacuum at a current intensity of 40 mA using a sputter coater. SEM images were acquired at an accelerating voltage of 15 kV. TEM (FEI Spirit) was additionally utilized to acquired images of solvent-extracted samples which were mounted on carbon-coated copper grids.

Characterization of surface chemistry

Fiber surface chemistry was examined by AXIS His X-ray photoelectron spectroscopy (Kratos Analytical Inc., NY) and associated curve fitting software. For all samples, a survey spectrum was recorded over a binding energy range of 0-1100 eV using a pass energy of 80 eV. In all cases, the survey spectra recorded the presence of oxygen (O1s 533 eV), Carbon (C1s 285 eV), and Nitrogen (N1s 399 eV) at the surface.

Mechanical analysis

Fiber mats were cut into sections and fixed onto a paper frame. Gauge length and width was set to 10 mm and 5 mm according to the frame size and the thickness of each sample, pre-determined by light microscope. Samples were mounted on a nano tensile tester (Nano Bionix, MTS, USA) and the edge of frame was cut before testing the fiber samples. Ten samples were stretched to failure at a low strain rate of 1%/sec at room temperature while related displacement and force values were recorded.

Fibronectin conjugation

Conjugation of fibronectin to coated and un-coated fiber material was performed as discussed in prior studies.[6,8] Specifically, polydopamine-coated fiber materials were transferred into fibronectin solution (50 μg/mL sodium phosphate buffer, pH 7.8) for 12 hr. Functionalized fiber materials were then washed three times with phosphate buffered saline (PBS) prior to cell culture.

Cell culture

NIH3T3 cells were cultured in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% fetal bovine serum (FBS, Invitrogen) and 1% gentalmycine/streptomycin (Invitrogen) at 37°C in an atmosphere of 95% air / 5% CO_2. Cell culture medium was replaced every 2 days. Prior to cell seeding on fiber scaffolds, cells were trypsinized and counted. Around 1×10^4 cells were seeded on each fiber sample. After incubation for 2 h, 4 h and 24 h, live cells were stained using Calcein AM (Invitrogen) and F-actin was labeled with Alexa Fluor 488® phalloidin (Invitrogen). Fluorescent images were taken using a fluorescence microscope (Zeiss, Thornwood, NY, USA).

In vitro release of Rodamine 6G

Around 20 mg fiber samples were immersed in water and 70% ethanol. The solution was kept at constant temperature of 37.2 °C. At given time intervals, the solution was withdrawn and kept for analysis via microplate reader (Thermo Scientific, Waltham, MA, USA).

Adsorption kinetics

Adsorption kinetics of rhodamine 6G to fibers in water was tested using similar amounts (20 mg) of un-coated (PCL) fibers and polydopamine-coated PCL fibers (coating in 0.2 mg/mL dopamine at pH 8.5 for 4 h). One μg/mL of rhodamine 6G was prepared in water in two scintillation vials, with a total volume of 10 mL in each vial. Following fiber sample submersion into the solution, 150 μL of solution was collected at each time point and placed into a 96 well plate. After the solution was collected, 150 μL of water was added into the scintillation vials to keep the total volume constant at 10 mL. Samples of the solution were collected every minute for the first 12 minutes and then at 27, 117, 297, 1380, 1920, and 3300 minutes. A microplate reader was then used to measure the fluorescent intensity of each sample collected, as calculated by calibration concentrations of rhodamine 6G in water or 70% ethanol (19.5 ng/mL - 10 μg/mL) present on each 96-well plate. Adsorption of rhodamine 6G in 70% ethanol was assessed using the same procedure as that using water, except that the times for solution collection altered. Samples of ethanol solution were collected every minute for the first 12 minutes and then at 27, 117, 237, 1320, 1860, and 3240 minutes, respectively.

RESULTS AND DISCUSSION

In order to examine the effect of surface property on polydopamine coating, we performed *in situ* polymerization of dopamine at concentrations of 0.2 mg/mL (pH 8.5) for 4 h or a concentration of 2 mg/mL (pH 8.5) for 72 h on electrospun fibers with and without prior plasma treatment. In the absence of plasma pre-treatment, polydopamine coatings were observed to be fragmented and uneven upon SEM imaging of polydopamine casts following PCL extraction (data not shown). Fig. 1A shows a SEM image of polydopamine-coated PCL fibers where PCL fibers were plasma treated prior to functionalization in 0.2 mg/mL dopamine at pH 8.5 for 4 h. Fiber morphology was similar between polydopamine-coated and un-coated samples, with both fiber populations demonstrating consistent fiber diameters and limited surface roughness (Fig.1A and inset). Fig. 1B shows a SEM image of polydopamine-coated PCL fibers where PCL fibers were plasma treated prior to functionalization in 2 mg/mL dopamine at pH 8.5 for 72 h. Larger fiber diameters noted in Fig. 1B indicated increased thickness of polydopamine deposition on underlying PCL fibers, resulting from an increased rate and duration of dopamine polymerization. Fig. 1C and 1D show the corresponding TEM images of samples observed in Fig. 1A and 1B following PCL extraction. Resulting TEM images, further demonstrate the selective presence of even polydopamine coatings formed on the surface of plasma-treated PCL fibers. Insets in Fig. 1C and 1D highlight the visual appearance of respective polydopamine-coated PCL fiber dissolved in DCM. Brown coloration observed in both solutions represent the presence of intact polydopamine casts, formed through uniform polydopamine coating on fiber substrates, remaining after dissolution of underlying PCL fibers. TEM images further confirm that polydopamine coatings ranged in thickness from around 12 nm to 70 nm for these two scenarios. Together, the present data suggests that the thickness of polydopamine coating on PCL fibers can be reliably controlled by adjusting the dopamine concentration and reaction time

utilize during polymerization. Examination of polydopamine coating on plasma-treated PLA fibers was further examined to determine the versatility of the polymerization mechanism across various substrates (Fig. 2). Comparison of PCL and PLA fiber samples coated with polydopamine in 0.2 mg/mL dopamine at pH 8.5 for 4 h demonstrated similar fiber morphologies, suggesting successful coating of both fiber substrates (Fig. 2A and 2B). We observed the cracks of polydopamine coating on the PLA fiber surface after coating in 2 mg/mL dopamine for 72 h (Fig. 2C). Fig. 2D shows a TEM image of polydopamine nanotube casts following extraction of underlying PLA fibers, further indicating the successful formation of continuous, even polydopamine coatings (28 nm thick) on PLA fibers.

Plasma surface treatment has been utilized to improve surface hydrophilicity, increase surface energy and develop functional groups with a variety of polymer biomaterials.[22] It is well known that oxygen or oxygen containing gas plasma can react with a wide range of polymers to produce a variety of reactive functional groups, including C-O, C=O, O-C=O and CO_3, on the material surface.[23] Previous studies also suggest that weak-chemical bonds present in PCL are replaced by highly reactive carbonyl (-CO-), carboxyl (-COOH) and hydroxyl (-OH) groups, resulting in profound biochemical activation of the material following treatment.[24,25] In the present work, plasma treated fiber mats were noted to be increasingly hydrophilic and demonstrated superior wettability confirming the effect of the treatment. As a result, dopamine molecules in solution were better able to form covalent bonds with functional groups (-CO- ,–COOH) on the surface of plasma-treated fibers in short periods of time. Induced polymerization of dopamine occurring on the surface of fibers further encouraged the forming of an even coating of polydopamine, controllable through manipulation of polymerization kinetics. Demonstration of successful deposition of polydopamine on a variety of substrates (i.e., PCL and PLA) further

demonstrates the versatility and wide-spread application of the present method of functionalization.

In order to further confirm polydopamine deposition on plasma-treated fibers, XPS analysis was performed on un-coated PCL fiber and polydopamine-coated PCL fiber samples. As expected, no N1s peak appeared upon analysis of PCL fiber sample (Fig. 3A). In contrast, N1s peaks of increasing size were observed in survey spectra for PCL fiber samples coated with polydopamine in 0.2 mg/mL dopamine at pH 8.5 for 4 h and 2 mg/mL dopamine at pH 8.5 for 72 h (Fig. 3 B-D), confirming the presence of increasingly thick polydopamine coatings on fiber substrates. The element composition of the fiber material surface was further characterized as indicated in Table 1. Here the content of N increased with increasing the concentration of dopamine and reaction time, indicating progressively increased deposition of polydopamine on fibers.

Mechanical property of un-coated and polydopamine-coated fibers was additionally examined as it plays an important role in various applications. Mechanical property represents an important consideration in assessing the biocompatibility of modified fiber matrices, as forces exerted on the fibers may result in permanent deformation or even failure during the service lifetime of the fibers.[26] Tensile testing was performed for all fibers samples using a nano tensile tester (Fig. 4). Comparison of un-coated PCL fibers and polydopamine-coated PCL fibers demonstrated that polydopamine coating resulted in material with a higher Young's modulus and lower terminal strain. The present data suggests that thicker coating of polydopamine may influence the mechanical behavior of fiber matrices and lead to materials that are less elastic and more rigid.

Promotion of robust cellular adhesion is critical for the chronic success of implantable biomaterials, such as fiber matrices. Cellular adhesion to polymeric biomaterials is primarily mediated by integrins which are covalently bound to specific motifs present on the biomaterials.[27] Polydopamine has previously been demonstrated to promote cell adhesion on various substrates.[28-30] In addition, polydopamine coatings have been shown to serve as an ideal linker for secondary immobilization of functional biomacromolecules, thereby enabling superior biocompatibility of underlying substrates.[6-8] In the present study fibronectin, an extracellular matrix protein, was immobilized on the surface of polydopamine-coated PCL fibers through a secondary reaction.[6] Adhesion and migration of NIH 3T3 fibroblasts to both un-coated PCL fibers, polydopamine-coated PCL fibers, and fibronectin-conjugated polydopamine-coated PCL fibers was assessed as a measure of cellular integration and biocompatibility. We chose NIH3T3 cells because their adhesive properties like integrin expression pattern and spreading have been well documented previously.[31] And they are also commonly used in studies of cell functions and to demonstrate the critical roles of cytoskeletal components in cell adhesion.[32] Fibroblasts themselves are related to the applications in tissue engineering like connective tissue regeneration (e.g., tendon) and wound healing. Fig. 5 shows live cell staining on seeded fiber samples after incubation for 2, 4 and 24 h. Cells seeded on the PCL fibers were noted to maintain a round morphology over a 2 h incubation period, demonstrating limited interaction and adhesion to the material. In contrast, cells seeded on polydopamine-coated PCL fibers began to demonstrate limited spreading and formation of cellular processes within 2 h of plating. Fibronectin-conjugated, polydopamine-coated PCL fibers were demonstrated to promote robust cellular adhesion and spreading of plated cells over the same period, demonstrating robust cellular interaction and adhesion to the functionalized, coated fiber matrix. In the latter two

cases, cells appeared to be biologically attached instead of physically adsorbed, demonstrating superior interaction between plated cellular populations and polydopamine-coated PCL fibers. After 4 h, cells cultured on PCL fibers demonstrated minimal change in morphology, while cells cultured on polydopamine-coated PCL fibers showed protrusion of cellular processes and spindle morphologies. Cells cultured on fibronectin-immobilized fiber samples demonstrated superior spreading and adhesion compared the other materials. After incubation for 24 h, a small proportion of cells seeded on un-coated PCL fibers showed spindle morphology and with many remaining in a round morphology. In contrast, the vast proportion of cells seeded on polydopamine-coated PCL fibers exhibited robust adhesion and spreading. Cells cultured on fibronectin-conjugated, polydopamine-coated PCL fibers showed excellent spreading, further confirming the improved biocompatibility of the functionalized material. Projected area per cell calculations increased in the order of un-coated PCL fibers, polydopamine-coated PCL fibers, and fibronectin-conjugated, polydopamine-coated PCL fibers. Comparison of cell morphology evolution on uniaxially-aligned fiber matrices with similar coatings demonstrated similar trend (Fig. S1).

Cell adhesion typically progresses through stages of substrate attachment, spreading, and cytoskeletal development.[33] In order to examine the cytoskeletal development of cells seeded on various fiber samples F-actin staining was performed (Fig. 6). Few cells attached to un-coated PCL fibers displayed elongated actin bundles following 24 h of incubation. Cells adherent on polydopamine-coated PCL fibers demonstrated actin bundles after only 4 h of incubation. In contrast, cells adhered to fibronectin-conjugated, polydopamine-coated PCL fibers present evident F-actin bundles and stress fibers after only 2 h of incubation, revealing more rapid cellular adhesion and maturation on the functionalized substrates. Evaluation of F-action staining

in cells adherent to aligned fiber matrices with similar coatings demonstrated similar trends (Fig. S2). Together, polydopamine coating and fibronectin conjugation not only enhanced cellular adhesion and spreading on fiber materials, but also accelerated the development of cell cytoskeleton.

Electrospun fibers can be used to construct tissue engineering scaffolds capable of incorporating bioactive molecules and, thereby, controlling biological function.[9] However, encapsulation of hydrophilic agents within fiber drug delivery systems has proved problematic, with high incidence of large scale burst release.[34,35] One previous study demonstrated that polydopamine coating can be as a barrier to slow the release of DNA plasmid from porous polymer scaffolds.[36] In order to examine the effect of polydopamine coating on the release of encapsulated hydrophilic agents, the release of loaded rhodamine 6G was examined from various samples under different conditions *in vitro* (Fig.7). We chose Rhodamine 6G as a molecular probe because it is fluorescent and easy to detect. Other small hydrophilic molecules/drugs may exhibit similar release profiles as Rhodamine 6G. Polydopamine coatings were observed to effectively inhibit initial burst release of rhodamine 6G in water, as increasing coating thickness resulted in reduced burst release and lower rates of release thereafter. However, release rates were largely unchanged by different thicknesses of polydopamine coating upon use of 70% ethanol solution. Together, these results suggest that polydopamine coating can be used to tailor release profiles of hydrophilic agent from fiber matrices in aqueous environments. In addition, the releasing medium may also be used to regulate the release rate of encapsulated drugs. It was reported polydopamine coating exhibited unidirectional permeability to rhodamine 6G.[37] But the release period they examined less than 180 h could be too short for the diffusion of rhodamine 6G molecules across the coating. In total, the present work demonstrated polydopamine-coating

can modulate the release of rhodamine 6G from PCL fibers in water, offering a unique method of designing tailored drug delivery mechanisms for *in vivo* use.

Direct encapsulation of drugs in fiber substrates during electrospinning has been suggested to result in a loss of bioactivity due to the presence of harsh solvents (e.g., organic solvent and high electric field). Owing to the large surface area to volume ratio, drugs may also be readily loaded into electrospun fiber matrices by adsorption post-fabrication. In order to examine the effect of polydopamine coating on agent loading, fabricated fiber matrices were placed into solutions containing either water or 70% ethanol and rhodamine 6G, respectively. Fig. 8A shows the kinetics of rhodamine 6G adsorption to un-coated PCL fibers and polydopamine-coated PCL fibers in water. Adsorption equilibrium was reached in 5 minutes for polydopamine-coated PCL fibers, while adsorption equilibrium was not reached until 1400 minutes for un-coated PCL fibers. The loading capacity in water for un-coated PCL fibers was also much lower than polydopamine-coated PCL fibers after the first 10 minutes (Fig. 8B). In contrast, both PCL fibers and polydopamine-coated PCL fibers showed very similar kinetics of rhodamine 6G loading in 70% ethanol (Fig. 9). Compared to the loading kinetics in different media, the loading capacity in water appeared to be much larger (about 2 times) than that in 70% ethanol. Polydopamine coating could exhibit zwitterionicity: at high pH the coating has a net negative charge which excludes anions and passes cations; at low pH it is positively charged and excludes cations but pass anions.[38] Polydopamine coating could be used as a wall for selective permeation of certain ions or charged molecules by manipulation of pH value. Therefore, it is possible to develop a fiber-based system where the loading and release rates of certain ions or charged molecules could be regulated by adjusting pH value in the loading and release medium.

CONCLUSIONS

The present study demonstrates a simple and versatile approach to the surface modification of electrospun fibers with a controllable layer of polydopamine using *in situ* aqueous polymerization. We have further conjugated fibronectin on polydopamine-coated fibers through a secondary immobilization. NIH3T3 cell culture demonstrated polydopamine coating and fibronectin-conjugation enhanced cellular adhesion and spreading on the material, and accelerated cytoskeletal maturation *in vitro*. Further, polydopamine coatings were further demonstrated to regulate the release and loading rates of bioactive materials from fiber matrices and media, respectively. Together, these results suggest that polydopamine-mediated surface modification to electrospun fibers could render a wide range of novel fiber materials with multiple uses in tissue engineering and controlled drug delivery applications.

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Table 1. XPS analysis

Sample	Element	Atomic conc%
S1	C	76.1
	N	0
	O	23.9
S2	C	74.3
	N	1.5
	O	24.2
S3	C	74.2
	N	5.4
	O	20.4

S1: PCL nanofibers.

S2: coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 4 h.

S3: coating of polydopamine with 2 mg/mL of dopamine at pH 8.5 for 72 h.

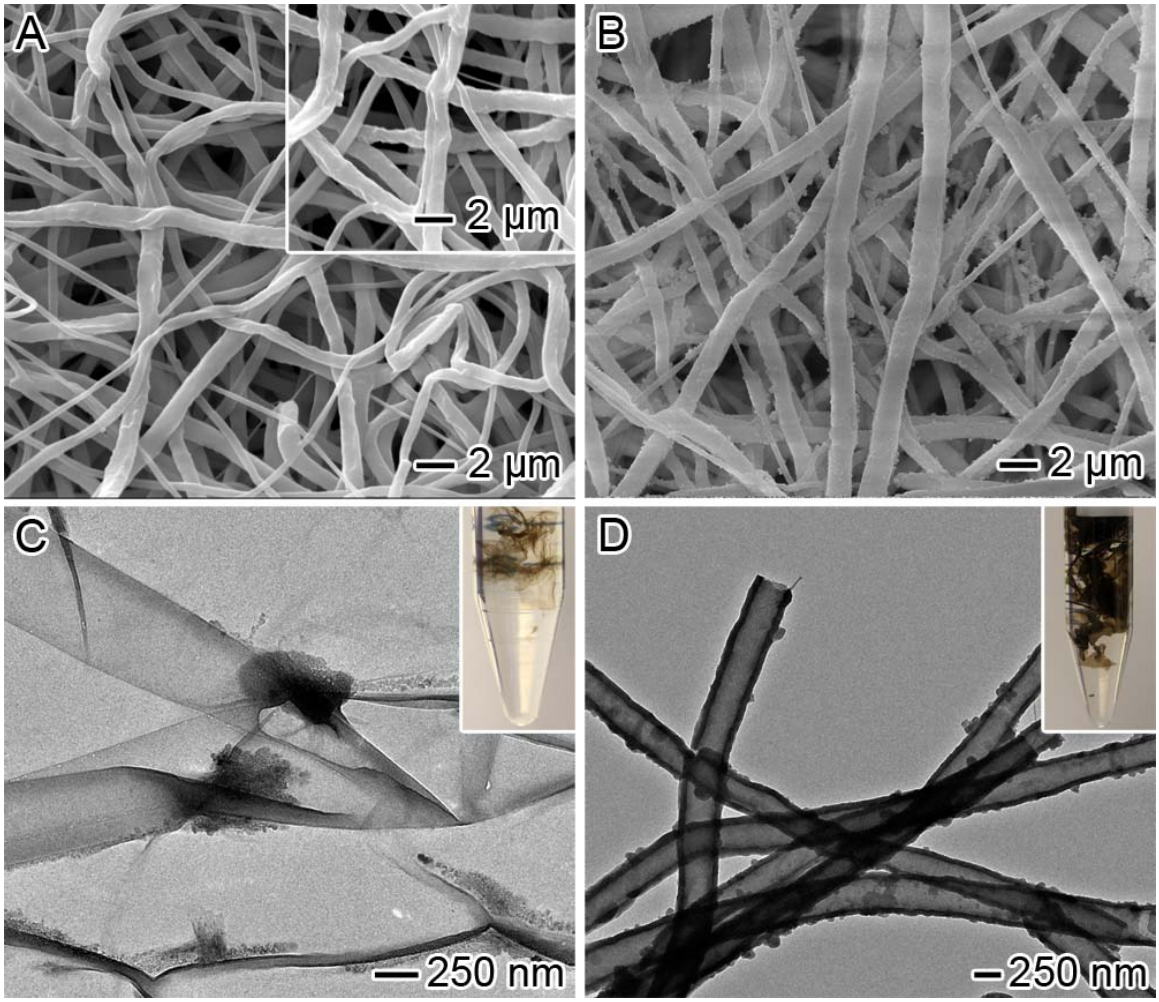

FIGURE 1. (A, B) SEM images of PCL fibers after coating of polydopamine with 0.2 mg/mL and 2 mg/mL dopamine at pH 8.5 for 4 h and 72 h, respectively. Inset in (A): Pristine PCL fibers. (C, D): TEM images of samples in (A) and (B) and the PCL core was removed by soaking the sample in DCM. Insets: photographs of samples in (A) and (B) when they were soaked in DCM.

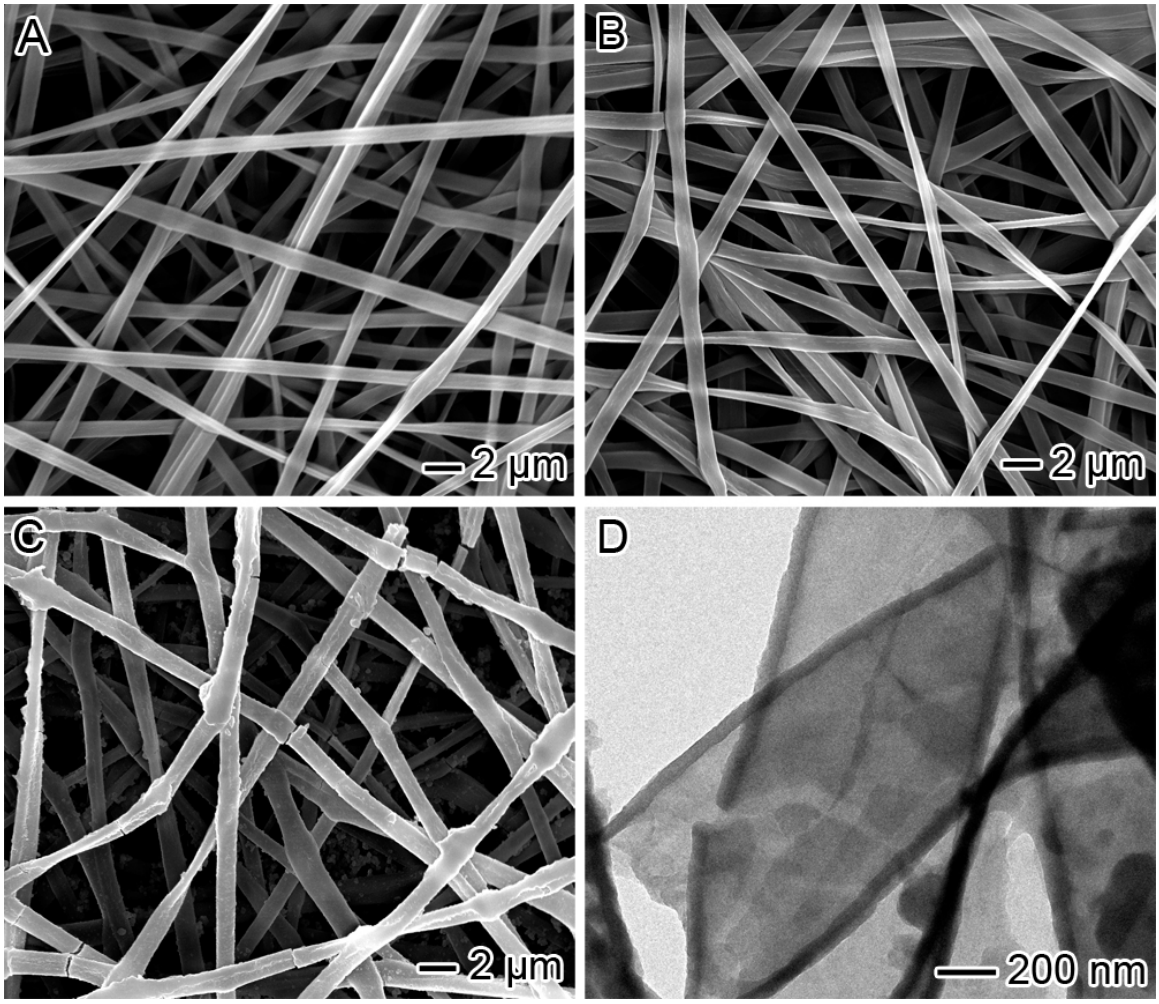

FIGURE 2. (A) SEM images of PLA fibers. (B, C) SEM images of PLA fibers after coating of polydopamine with 0.2 mg/mL and 2 mg/mL dopamine at pH 8.5 for 4 h and 72 h, respectively. (D) TEM image of (C) after they were dissolved in DCM to remove the PLA core.

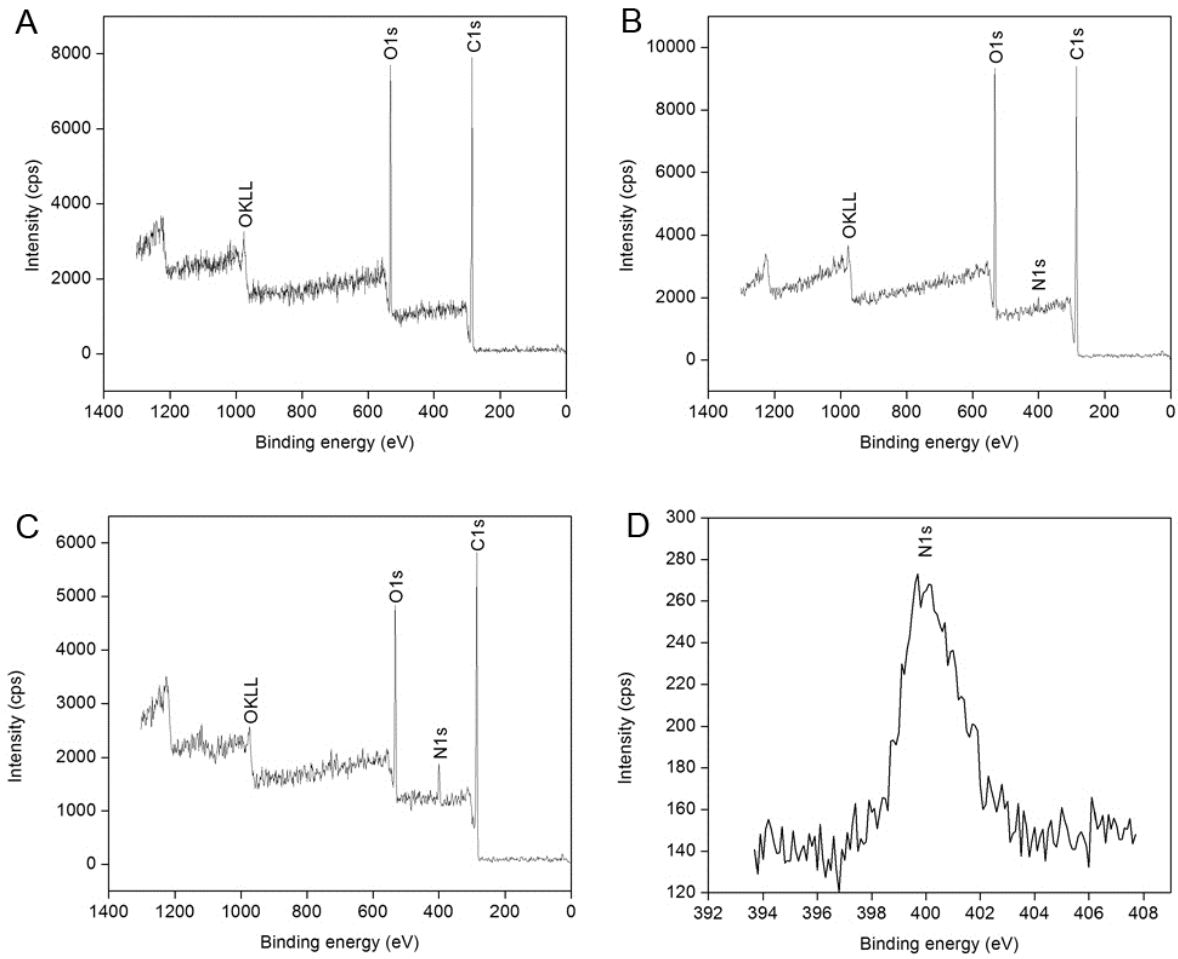

FIGURE 3. XPS analysis of fiber samples. (A) PCL fibers. (B) PCL fibers after coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 4 h. (C) PCL fibers after coating of polydopamine with 2 mg/mL dopamine at pH 8.5 for 72 h. (D) N1s peak for (C).

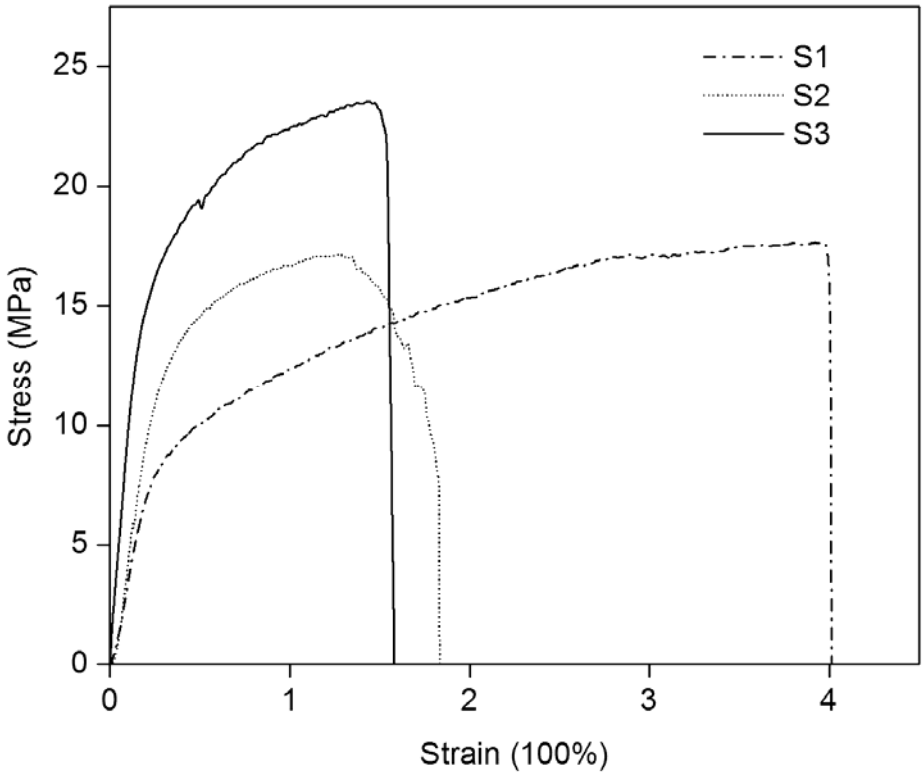

FIGURE 4. Representative stress-strain curves of fiber samples. S1: PCL fibers; S2: PCL fibers after coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 4 h; S3: PCL fibers after coating of polydopamine with 2 mg/mL dopamine at pH 8.5 for 72 h.

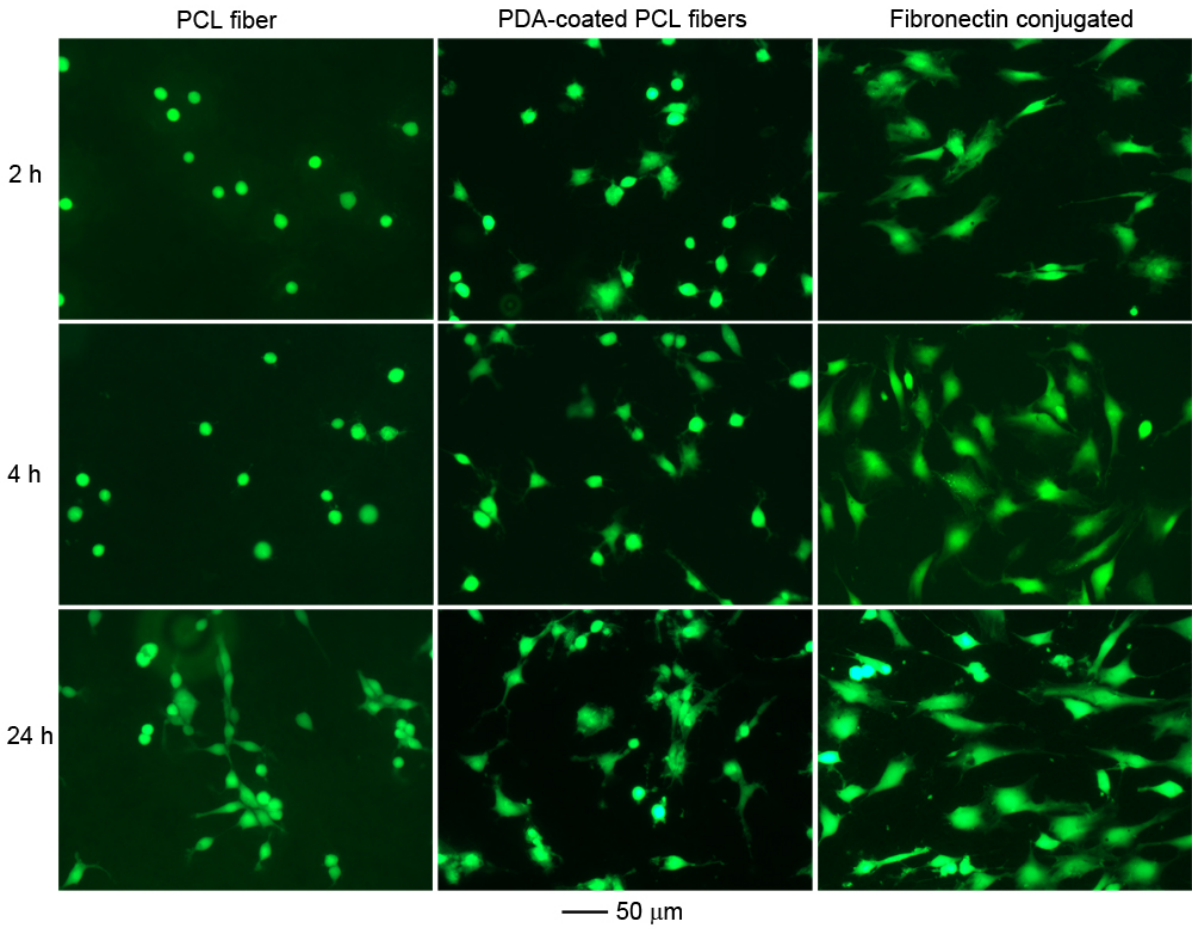

FIGURE 5. Calcein AM staining of live cells cultured on random fiber mats with different surface chemistries.

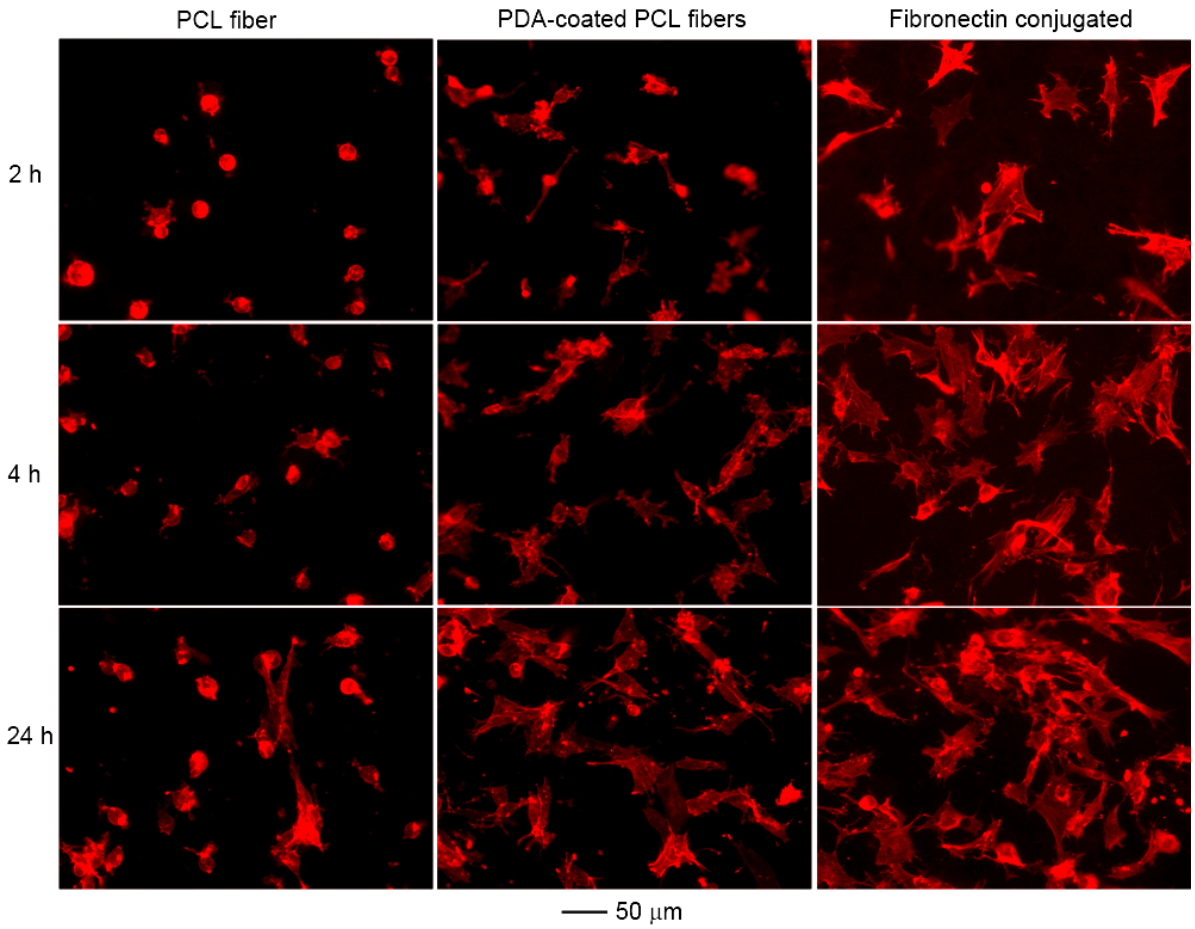

FIGURE 6. F-actin staining of NIH 3T3 cells cultured on random fiber mats with different surface chemistries.

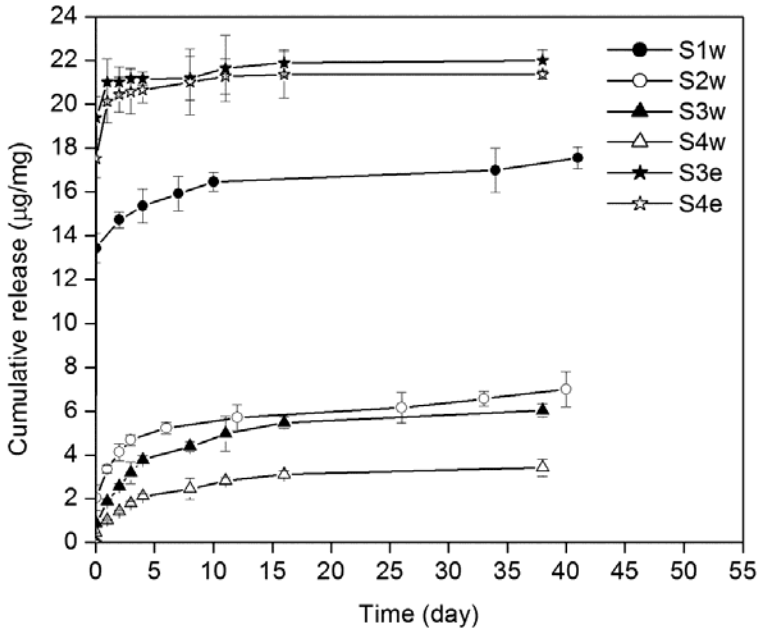

FIGURE 7. Rhodamine 6G release profiles of various samples in water (S1w-S4w) and 70% ethanol (S3e, S4e). S1w: Rhodamine 6G-loaded PCL fibers; S2w: coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 0.5 h; S3w: coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 4 h; S4w: coating of polydopamine with 2 mg/mL dopamine at pH 8.5 for 72h; S3e: the same as S3w; and S4e: the same as S4w. Each data point shown is the average of three samples. The results of Student t-test show that p values (S1w and S2w, S1w and S3w, S1w and S4w, S2w and S4w, S2w and S3e, S2w and S4e, S3w and S3e, S3w and S4e, S4w and S3e, S4w and S4e) are less than 0.01, indicating significant difference.

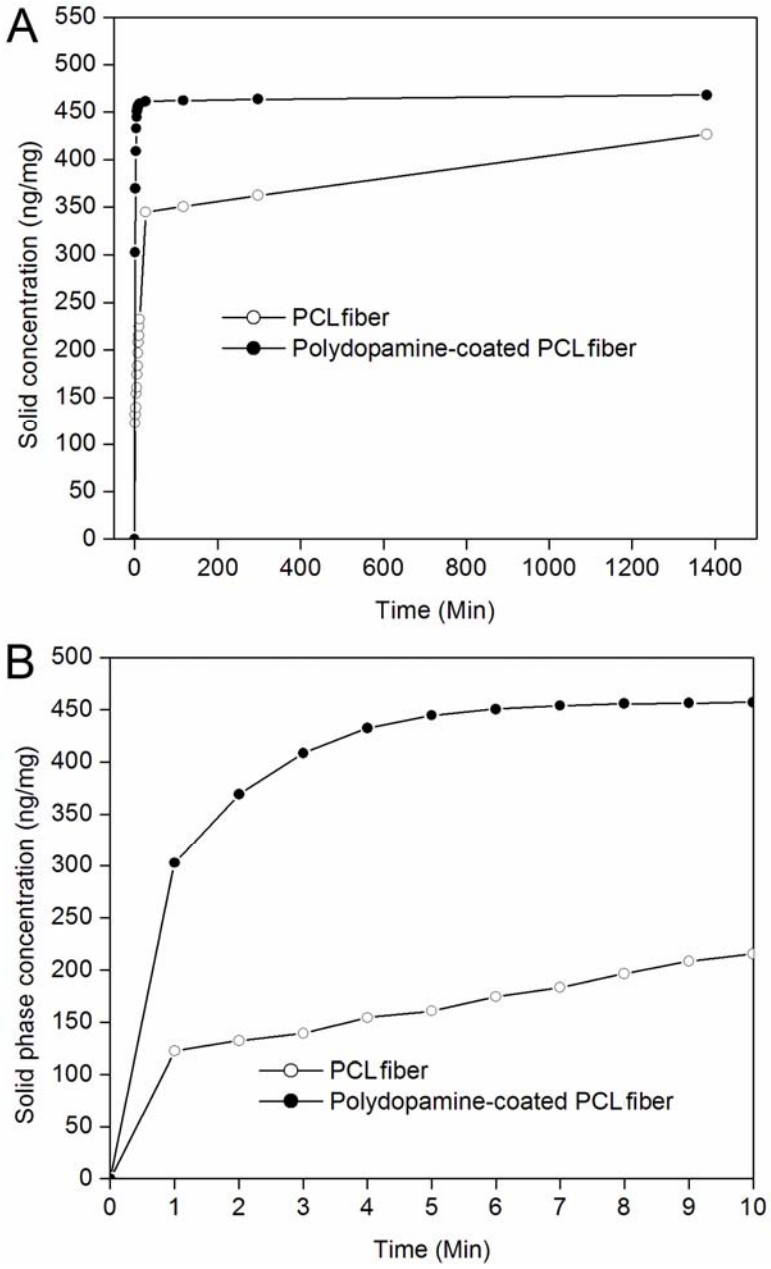

FIGURE 8. (A) Rhodamine 6G adsorption kinetics in water. (B) The close-up view of adsorption curve in (A) for the first 10 mins. Polydopamine-coated PCL fiber: coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 4 h. Each data point shown is the average of three samples.

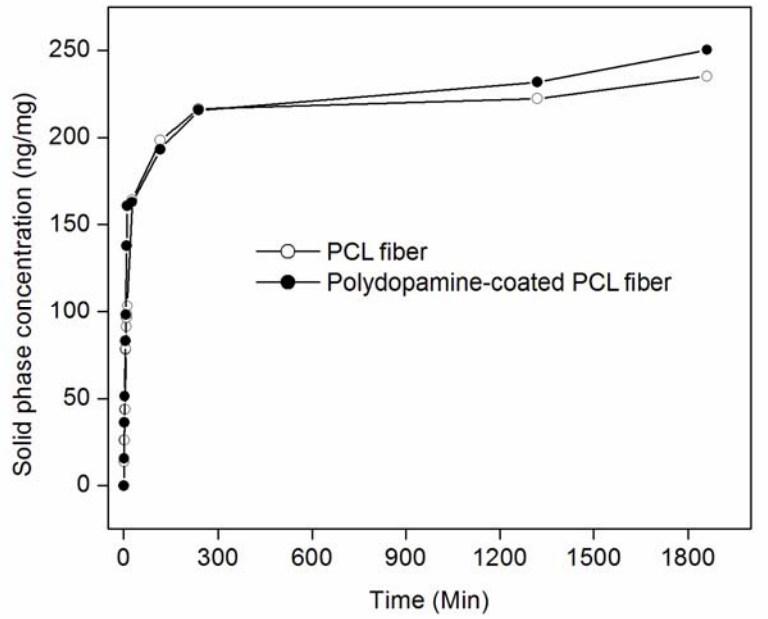

FIGURE 9. Rhodamine 6G adsorption kinetics in 70% ethanol. Polydopamine-coated PCL fiber: coating of polydopamine with 0.2 mg/mL dopamine at pH 8.5 for 4 h. Each data point shown is the average of three samples.

Supplementary Materials

Mussel inspired protein-mediated surface modification to electrospun nanofibers and their potential biomedical applications

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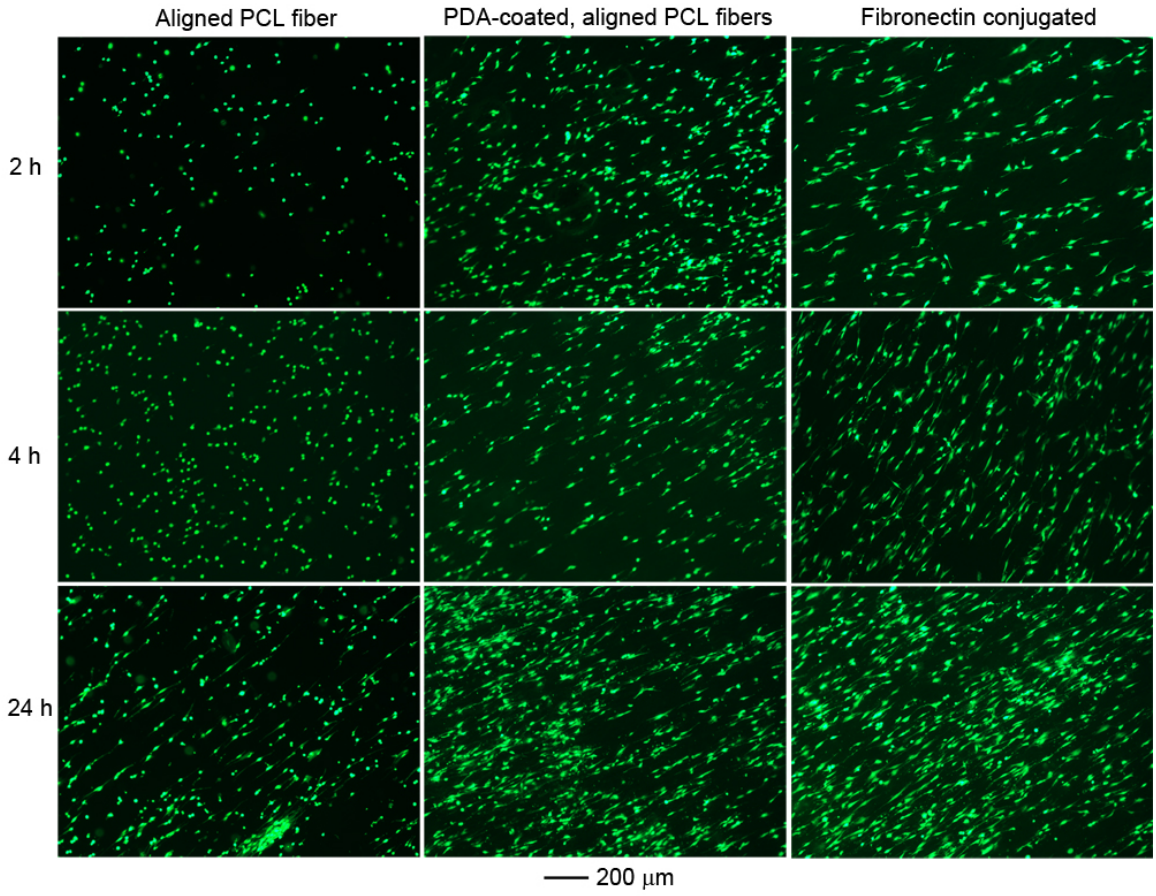

FIGURE S1. Calcein AM staining of live cells cultured on aligned nanofibers with different surface chemistries.

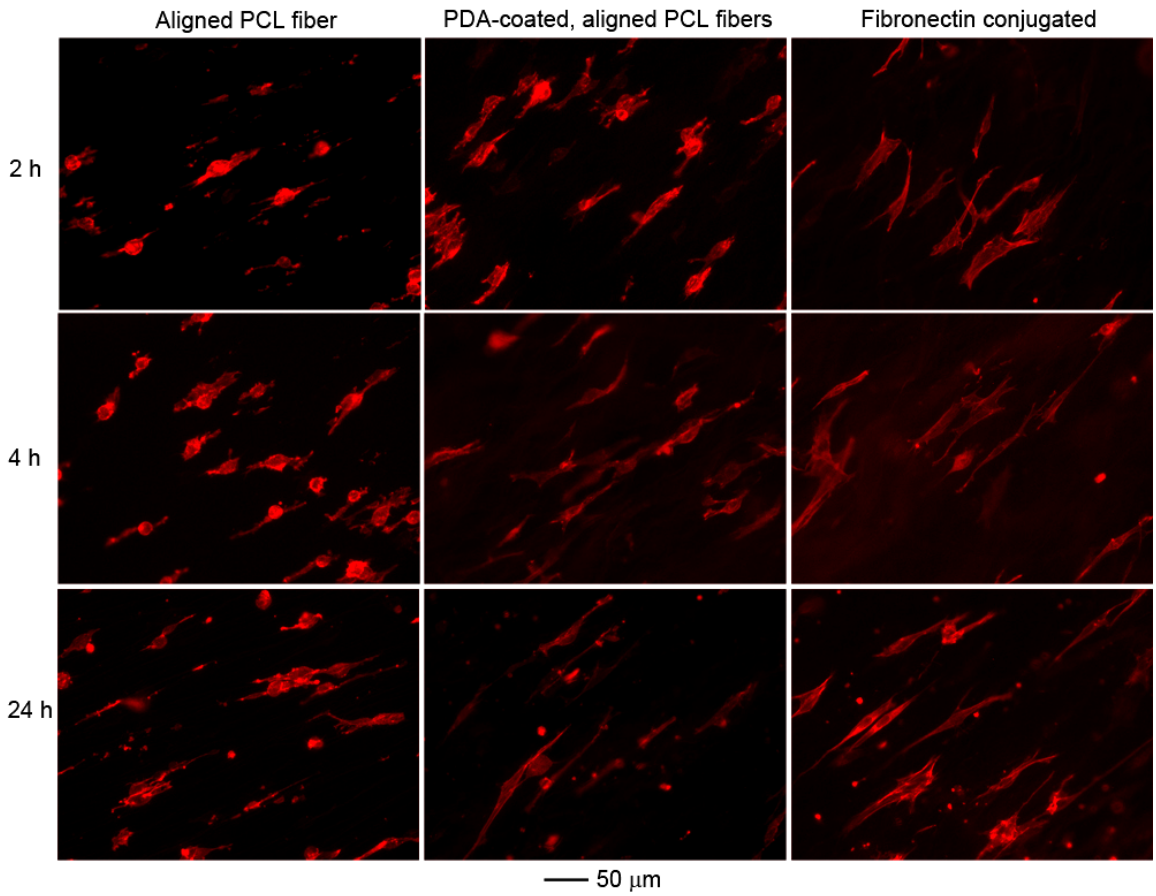

FIGURE S2. F-actin staining of NIH 3T3 cells on aligned nanofibers with different surface chemistries.