Plasma Surface Modification of Electrospun Poly(ε-Caprolactone) Nanofibers and its Effect on Surface Bioactivity

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Statement of Purpose: The nanoscale dimension and slow degradation rate of electrospun Poly(ε-caprolactone) (PCL) nanofibers make them suitable candidates for tissue-engineering scaffolds[1,2]. Unfortunately, PCL nanofibers have low surface wettabilities that could impair cell binding. To overcome this problem, plasma surface treatment of PCL nanofibers using a radio frequency (RF) glow discharge (RFGD) has been proposed. Our hypothesis is that the RFGD from a mixture of NH_3 and O_2 can incorporate hydrophilic groups onto the PCL surfaces, thereby enhancing the cell-surface interaction of PCL nanofibers. In this presentation, the effects of plasma treatment on the mechanical properties and bioactivities of random and aligned PCL nanofibers will be reported and discussed.

Methods: A 9 wt% PCL solution was prepared by dissolving PCL (Aldrich Chemical Co., Milwaukee, WI) in a 3:1 by volume mixture of chloroform and methanol. The PCL was allowed to dissolve for 24 h and was then drawn into a 10 ml plastic syringe fitted with a 22 gauge blunt-end stainless steel needle. A high power supply was used to deliver 20 kV to the needle. A syringe pump was used to deliver the polymer solution at a pump rate of 0.6 ml/h. Random nanofiber mats (RMs) were deposited onto an electrically-grounded 15 cm x 10 cm aluminum panel set 17 cm from the tip of the needle. For aligned nanofibers, 20 and 40 mesh stainless steel was used as the grounded collector. The PCL mats were then immersed in deionized water for 3 days to remove residual solvent. Samples were then dried overnight at 25°C.

Following drying, PCL nanofiber mats were placed inside a bell jar-type plasma reactor with dimensions of 46 cm in height and 44.5 cm in diameter. The sample was placed between two titanium electrodes of dimensions 17.9 cm x 17.9 cm x 0.08 cm. The reactor was pumped down to less than 1 mTorr (0.133 Pa) pressure. The treatment gas was a 2:1 NH_3 + O_2 mixture held at a pressure of 50 mTorr (6.65 Pa). A 13.56 MHz RF power supply was used to sustain the RFGD at 20 W RF for 8 min.

Surface wettabilities were assessed by measuring the contact angle of a 1 µl sessile deionized water droplet deposited onto the nanofiber surfaces with a contact angle measuring system (AST Products Inc., Billerica, MA). Measurements were taken immediately after treatment. Mechanical measurements were conducted with a TA-HDi Texture Analyzer (Texture Technologies Corp., Scarsdale, NY, USA) using a 50 kg load cell.

Cell culture of the PCL nanofiber mats was performed with the mouse osteoblast MC3T3-E1cell line acquired from American Type Culture Collection (ATCC). Cells were grown on 75 cm^2 culture flasks at 37°C in 5% CO_2 modified α-MEM lacking ascorbic acid (GIBCO), supplemented with 10% fetal bovine serum and antibiotics. The cell medium was changed every 3 days until 90-95% confluence was reached. Cells were passaged with 0.05 wt% trypsin/EDTA (Invitrogen Corp., Carlsbad, CA). Three ml of the cell solution was seeded onto 2 cm x 2 cm nanofiber mats, and the final cell concentration was 5 x 10^4 cells ml^{-1}. An MTT assay was used to assess cell density, and spreading was determined through low vacuum scanning electron microscopy.

Results: Surface contact angles of PCL nanofibers were reduced from ~140° to 0° after plasma treatment, indicating nearly completely wetted surfaces. Figure 1 shows that the plasma treatment did not adversely affect the ultimate tensile stresses of the PCL nanofibers. MTT absorbance data (Fig. 2) showed higher cell densities on all plasma treated groups. SEM images showed cell spreading on the fibers, indicating improved cell-surface interaction.

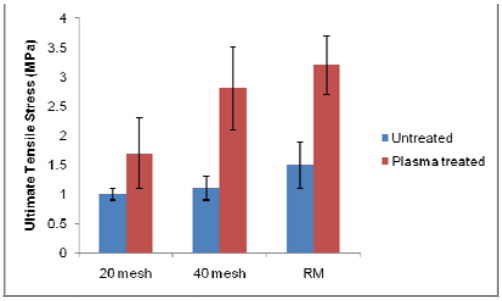

Figure 1. Ultimate tensile stresses for untreated and plasma-treated PCL.

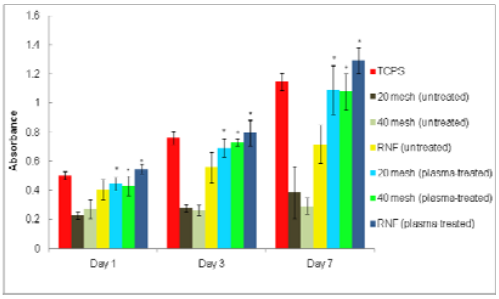

Figure 2. MTT absorbance data for PCL nanofibers (* denotes $p < 0.05$).

Conclusions: Surface treatment of PCL nanofibers using NH_3 + O_2 plasmas produced wettable surfaces that are capable of sustaining enhanced cell growth on both aligned and randomly-arranged PCL nanofibers without reducing their mechanical strength. Plasma treatment is effective in improving PCL nanofibers' biocompatibility.

References:

[1] Liao Y. Polymer, 2008;**49**;5294-5299
[2] Garcia-Giralt N. Journal of Biomedical Materials Research, Part A, 2008;**85A**;1082-1089

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