Direct Fabrication by Manual Brushing of Suspended Microscale Fibers for Cell Culture Scaffolds

Scott M. Berry[1], Santosh Pabba[3], Jessica L. Fernades[3], Jeremy M. Rathfon[4], Khaled A. Aamer[4], Gregory N. Tew[4], Andrea S. Gobin[2], Robert W. Cohn[3], and Robert S. Keynton[1,2]
Departments of Mechanical Engineering[1], and Bioengineering[2], and ElectroOptics Research Institute and Nanotechnology Center[3], University of Louisville; Department of Polymer Science[4], University of Massachusetts

Statement of Purpose: A major obstacle in the application of therapeutic angiogenesis is the ability to assess cell growth *a priori* in a controlled *in vitro* environment. The objective of this research is to fabricate arrays of nano-porous fibers composed of amphiphilic and biodegradable poly(L-lactic acid-b-ethylene oxide-b-L-lactic acid) tri-block copolymer that are ideal as scaffolds for endothelial cell culture.

Methods: Past research by our group has demonstrated the ability to rapidly create parallel arrangements of fibers by manually brushing solvated polymer solutions over arrays of microfabricated pillars[1]. Microfabricated pillars were produced in a glass or silicon substrate using a dicing saw (DAD 321, Disco, Tokyo, Japan) and then wet-etched in KOH or deep reactive ion etched (DRIE) using the Bosch process to produce pillars with tip radii <500 nm and 1 μm, respectively. Fibers are then drawn by brushing an applicator with the desired polymer on one end across the pillars (Fig. 1). The suspended filaments of polymer solution thin, via surface tension-driven necking, and dry to yield solid micro- and nanoscale fibers. This process has proven to be compatible with several different polymers and polymer-based composites including poly(methyl methacrylate), poly(vinyl acetate), poly(ethylene oxide), poly(vinyl chloride), poly(styrene-b-methyl methacrylate) di-block copolymer, as well as silver nano-particle and carbon nanotube-doped PMMA[2]. The brush-on process has successfully produced fibers with diameters ranging from 10 nm to 20 μm[1,2]. Recently, this technique was utilized to draw fibers from P(LLA-EO-LLA) tri-block copolymer, a biodegradable and mechanically robust hydrogel[3] (Fig. 2).

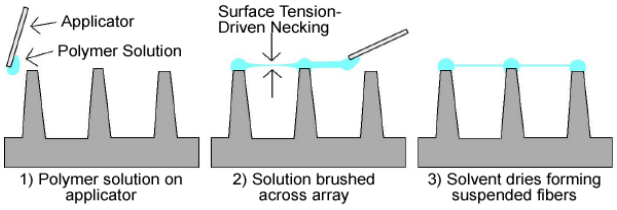

Figure 1: Schematic of brush-on fabrication of fibers on a microfabricated pillar array

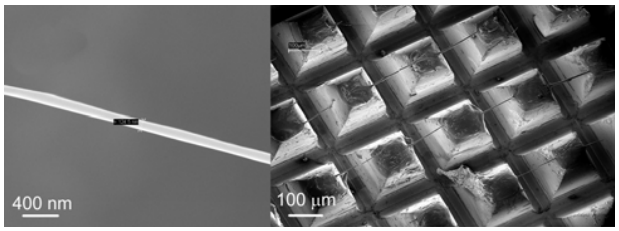

Figure 2 – Untreated P(LLA-EO-LLA) fibers fabricated with brush-on method.

Results/Discussion: Suspended P(LLA-EO-LLA) fibers with diameters ranging from 125 nm to >50 μm have been fabricated using the brush-on method (Fig. 1). The solution used in these trials was a 15% concentration (wt. %) of 136.4 kDa P(LLA-EO-LLA) in chloroform. Suspended, porous fibers have been created by annealing the brushed-on fibers at 140°C for 30 minutes then exposing them to water for 1 minute (Fig. 3c, d). Initial studies show that the pores formed are elongated and circumferentially oriented. These pores have a 3.8:1 circumferential-to-longitudinal aspect ratio, with an average circumferentially oriented distance of 393.8 ± 448.5 nm and axial oriented distance of 103.9 ± 55.5 nm. Fibers were also found to be porous after treatment with water alone (Fig. 3a) or after annealing at 140° C and a 1 minute exposure to isopropyl alcohol (IPA) (Fig. 3b). The pores in the fibers treated only with water were found to be unaligned and measured between 50 and 950 nm (avg. 258 ± 185nm). This ability to control surface morphology will enable the investigation of the influence of fiber structure on cell growth. Additionally, the porous ultrastructure will facilitate the delivery of cellular growth factors during angiogenesis.

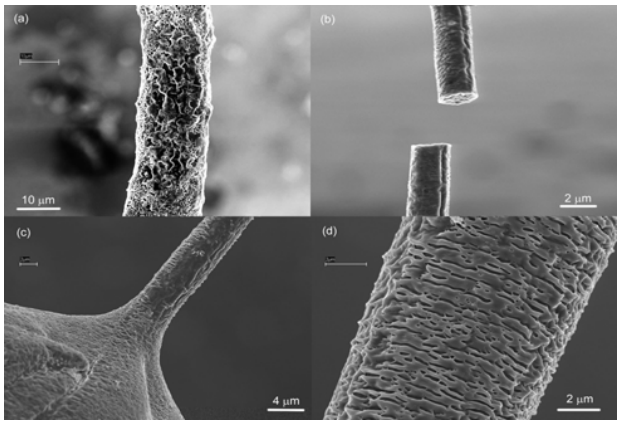

Figure 3: SEM images of P(LLA-EO-LLA) fibers fabricated by brush-on method and treated with a) water only, b) heat and IPA, and c) and d) heat and water.

Conclusions: Suspended micro- and nanofibers composed of a novel biodegradable polymer have been fabricated using a new fabrication process. Additionally, nano-porosity was induced through post-treatment to produce fibers with a unique combination of properties appropriate for an endothelial cell culture scaffold.

References:

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