Fabrication and Characterization of Synthetic Hydrogel Fibers

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Statement of Purpose: Polymer fibers are widely used in textile biomaterials and tissue engineering scaffolds. Conventional fiber extrusion processes utilize heat or solvent, both of which are incompatible with the direct incorporation of bioactive molecules during the fabrication process. Hydrogel fibers have been created by polyelectrolyte complexation of naturally-derived anionic/cationic polymers and shown to support direct encapsulation of cells and bioactive molecules.[1,2] The use of synthetic materials and covalent crosslinking offers the potential for improved control over degradation and mechanical properties. Celessi first described a tandem gelation process combining reverse thermal gelation and covalent crosslinking for the formation of synthetic hydrogel microspheres.[3] The objective of this study was to investigate the application of this principle to the formation of synthetic hydrogel fibers and characterize their physical/chemical properties.

Methods: Tetronic T1107 was acrylated (T1107 ACR) by reaction with acryloyl chloride in the presence of triethylamine. T1107 ACR was dissolved at 25.6 % (w/v) in pH 7.4 50 mM PBS. Dithiothreitol (DTT, 47.3 mg/ml)) was prepared in PBS and used as a model multi-functional thiol crosslinker. For fiber formation, cooled (4 °C) solutions of T1107 ACR and DTT were combined (9:1 volume ratio and 1:1 acrylate:thiol ratio), vortexed, centrifuged to remove air bubbles, and transferred to 3 mL syringe (BD) with 21 or 25 gauge needle. The extrusion apparatus consisted of syringe pump (Model 100, KD scientific) mounted above a PBS collection bath (37 °C) on a custom made rotating platform controlled by LabView software. Extrusion parameters were: 21 gauge needle: flowrate 35 ml/hr, bath rotation 3.8 rpm; 25 gauge needle: flowrate 20 ml/hr, bath rotation 5.1 rpm. During extrusion, fibers were instantly formed by reverse thermal gelation and allowed to sit in the bath for 15 minutes for covalent crosslinking. Hydrogel discs of similar composition were crosslinked between glass slides for 15 minutes at 37°C. Both fibers and discs were placed in pH 7.0 50mM PBS solution at 37°C overnight for full crosslinking. The mass swelling ratio was calculated as the wet weight/dry weight of fibers and discs. Tensile testing was performed on wet and dry (lyophilized) fibers. Due to their low mechanical properties, both ends of wet fibers were glued to a glass support using cyanoacrylate, loaded with small paperclips of known mass, and the deformation manually recorded using a ruler. Dry fiber tensile tests were done using MTS Synergie 100 with 10 N load cell. For cellular adhesion study, 1 μmol/ml acrylate-PEG-GRGDS was added to the T1107 ACR solution prior to mixing and extrusion. After extrusion and overnight swelling, fibers were transferred to 24 well plates, sterilized with 70% ethanol, and seeded with NIH 3T3 fibroblasts (ATCC).

Results: The mass swelling ratio of fibers extruded from 25 gauge needles was significantly greater than 21 gauge or disc controls (Table I). Diameter varied depending on needle gauge and significantly decreased after drying.

	21 Gauge	25 Gauge	Disc
Swelling ratio	8.48 ± 1.11	10.79 ± 1.22	7.88 ± 0.87
Wet diameter (μm)	1165 ± 48.5	579 ± 70.6	-
Dry diameter (μm)	388 ± 32.7	233.2 ± 50.7	-

Table 1. Mass swelling ratio, wet and dry diameter of two different types of fibers and disc.

Fiber tensile properties are shown in Table 2. Dry fibers exhibited approximate 3 and 2 orders of magnitude higher values of elastic modulus and ultimate stress relative to wet, respectively. All fibers were highly elastic exhibiting strain at failure of over 100%. Elastic moduli and ultimate stress of fibers extruded through 25 gauge needles was significantly lower than fibers prepared using 21 gauge needles in both wet and dry conditions.

	21 Gauge		25 Gauge	
(n=6)	Wet	Dry	Wet	Dry
Elastic modulus (kPa)	11.23 ± 3.08	22165 ± 8180	3.37 ± 1.26	9721 ± 5324
Ultimate stress (kPa)	11.30 ± 1.02	2629 ± 594	6.79 ± 2.79	1635 ± 273.9
Strain at failure	1.02 ± 0.13	2.29 ± 0.91	2.15 ± 0.71	2.12 ± 1.52

Table 2. Mechanical (tensile) properties of wet and dry fiber extruded from two different needle sizes.

Fibers incorporating RGD peptides supported fibroblast adhesion and spreading, while no cell adhesion was observed on controls without RGD (data not shown).

Conclusions: The mass swelling data indicates that fibers extruded through 21 gauge needles crosslinked with comparable efficiency to disc controls, despite being extruded into a large volume collection bath. The increased swelling and reduced mechanical properties of fibers made using 25 gauge needles suggests that as the fiber diameter decreases, diffusive loss of reactants during covalent crosslinking in the collection bath increases, resulting in reduced crosslinking density. Current studies are continuing to explore the biological applications of these materials, including cell encapsulation and sustained release of drugs and biomolecules.

References: 1. Wan A. C. A. J Biomed Mater Res 2003;71A:586. 2. Kim E. K. F. Biomaterials 2006; 27:6111. 3. Cellesi F. Macromol Chem Phys 2002;203:1466.

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