Fabrication and evaluation of elastomeric hollow fiber membranes as small diameter vascular graft substitutes

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Statement of Purpose: The general consensus in vascular tissue engineering is that a completely successful small diameter vascular graft (SDVG), with diameters less than 6 mm, is still elusive. The challenges to be overcome for SDVGs are the propensity to infection and inflammation, the possibility of restenosis and regression, poor patency rates, and the risk of thrombus formation [1]. As such, key characteristics of an ideal vessel substitute include mechanical compliance, antithrombogenicity, antifouling, tailored degradation profiles, and permeability [2]. Biologically-based grafts take a long time for these grafts to be constructed, and supply and size are limited by amount and selection of extracellular matrix-depositing cells. Polymer SDVGs have presented a promising solution due to their ease of production and customization. Here, we present a biocompatible, biodegradable, elastomeric polyester urethane-based hollow fiber membrane (HFM) as a potential SDGV.

Methods: Commercially-available Estane® polyurethane (PU) pellets and synthesized powdered 4-arm polylactic acid (PLA) were dissolved in dimethylsulfoxide at variable concentrations. HFMs were fabricated by a phase inversion method [3]. In this process, the polymer solution is perfused through a specially-designed spinneret with a tubular nozzle into a coagulant bath, where it continually precipitates as solid HFM (Figure 1). Fabrication parameters of polymer flowrate, water flowrate, polymer stream composition, drop height, and coagulant temperatures were varied. Cross-sectional and longitudinal samples of HFM were observed under SEM. Tensile testing for the HFM was conducted using a 2 kN Instron® ElectroPuls™ E10000 test system. Burst pressure was determined by monitoring the pressure of pumped water into the HFM until failure. Permeability was measured by using a countercurrent flow chamber in which a fluorescent solute was allowed to permeate through the membrane. Samples of HFM (2-cm length) were carefully weighed and placed in 2 mL of HBSS at 37°C for 8 weeks; every week the wet and dry weights were recorded to determine their degradation profiles. Bone marrow stromal cells (BMSCs) and human umbilical vein endothelial cells (HUVECs) were seeded onto the HFMs to determine their compatibility in vitro. For in vivo compatibility, small sections of the HFM were placed into the groin of a Sprague-Dawley rat for 5 weeks and subsequently collected for histological evaluation.

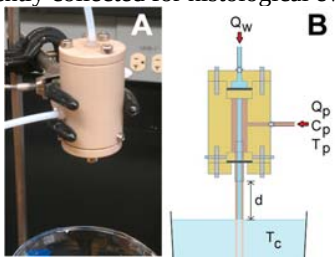

Figure 1. (A) Spinneret photo (B) HFM fabrication

HFM	ID (mm)	OD (mm)	Permeability $\times 10^6$ (cm/s)	Burst Pressure (psi)	Modulus (MPa)	Strength (MPa)	Strain (%)
1	0.467	0.579	2.21±0.85	27.6±2.5*	1.41±0.06*	1.47±0.08*	291.50±32.65*
2	0.788	0.919	0.34±0.02*	34.5±1.2*	1.87±0.07*	1.53±0.14	296.88±39.70
3	0.876	0.974	1.19±0.42	35.6±5.9	1.92±0.40	2.09±0.02*	432.66±53.81
4	0.968	1.124	3.07±1.51	27.6±3.0	3.72±0.61	5.52±1.77	511.71±14.14*
5	1.127	1.288	0.98±0.01*	31.4±1.3	4.07±0.77	3.96±0.62	393.63±35.57

Table 1. Physical properties of fabricated HFM; *: $p<0.05$

Results: HFM had diameters of less than 1.5 mm. A representative HFM shows a highly porous wall (Figure 2A). Results show that the fibers were very elastic, with moduli ranging from 1-4 MPa, strengths from 1-5 MPa, and max strains of 300-500% (Table 1). Permeability of the fibers varied from 0.5-3.5 × 10^{-6} cm/s, while burst pressure varied from 25 to 35 psi. A slow degradation profile was observed for all fiber formulations, with 71 to 78% of the original mass remaining after 8 weeks. Both HUVECs and BMSCs attached and proliferated on the surface of all HFM after 7 days. Fluorescent microscopy confirmed attachment of cells in the inner and outer surfaces of the HFMs (Figure 2B). There were cellular infiltrates into the subcutaneous implants, but no acute or chronic inflammatory response was observed, indicating good compatibility *in vivo* (Figure 2C).

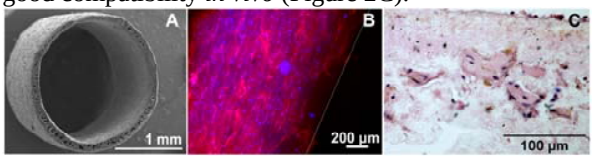

Figure 2. Representative imaging results for HFMs: (A) SEM image of HFM with porous wall (B) Fluorescent image of BMSC attached to HFM after 48 hours (C) Histological cross section of HFM 5 weeks after implantation

Conclusions: Elastomeric HFM fabricated by phase inversion methods matched or exceeded the physical and mechanical properties of native blood vessels or other artificial vascular grafts [4-6]. Porosity of the HFM could allow cellular infiltration and material exchange through its wall. This, combined with a slow, controlled degradation rate, would lead to HFM remodeling into an endothelialized neovessel after implantation. HFM biocompatibility exhibited both *in vivo* and *in vitro* also supports the feasibility of elastomeric HFM as surrogate SDVG. Future studies include development of novel elastomers for HFM synthesis and surgical evaluation of the elastomeric HFM in a rat femoral model.

References

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