Small-Diameter Biodegradable Vascular Grafts Mechanical Characterization and Tissue Interactions

Anthony J Melchiorri,[1] Zachary R Brandes,[1] Narutoshi Hibino,[2] John P Fisher.[1]

[1]Fischell Department of Bioengineering, University of Maryland, College Park, MD. [2]Department of Cardiovascular Surgery, Children's National Medical Center, DC.

Statement of Purpose: Insufficient mechanical properties and lack of cellular attachment and ingrowth in tissue engineered vascular grafts (TEVGs) can lead to issues of noncompliance, intimal hyperplasia, disturbed flow, and aneurysmal graft failure.[1,2] Mechanical properties of TEVGs must be maintained as tissue ingrowth takes place. Thus, it is important to consider the mechanical properties of a graft as they change during degradation, while maintaining cellular attachment and proliferation. A biodegradable polymer felt/polymer sealant graft fabrication methodology was adopted here.[3] Different polymer concentrations and solvents were used to fabricate TEVGs, analyzing mechanical properties, endothelial cell attachment, and *in vivo* functionality.

Methods: Graft fabrication consisted of rolling 5x7 mm woven poly(glycolic-co-lactic acid) felt (PGLA) (Biomedical Structures, Warwick, RI), followed by saturation with a poly(DL-lactide-co-caprolactone) (50:50 ratio) (P(LA/CL)) solution. This study tested four groups: grafts sealed with a 10% w/v solution of P(LA/CL) in acetic acid, 15% in acetic acid, and 10% and 15% in 1,4-dioxanes. We predicted the lower boiling point of 1,4-dioxanes would enable the solvent to evaporate faster than acetic acid, improving sealing of the graft. Higher concentrations of P(LA/CL) were predicted to increase mechanical strength via increase P(LA/CL) deposition. Grafts were frozen and freeze-dried. Graft dimensions were observed via scanning electron microscopy. Axial ultimate tensile strength (UTS) and the elastic modulus were found using a tensiometer. Circumferential strength was also determined with the tensiometer and used to approximate burst pressure via Laplace's Law: $P=(UTS*t)/r$, where P = internal pressure, UTS = ultimate tensile strength, t = wall thickness, and r = radius. Degradation studies were performed at 37°C to asses mechanical properties over time. Grafts were cut to fit a 96-well tissue culture plate and sterilized. Human umbilical vein endothelial cells (HUVECs) were seeded at a concentration of 5 x 10^3 cells/well. After 4 hrs, 1 day, 3 days, and 7 days, a Live/Dead assay was performed to assess cell attachment and proliferation via microscopy. Grafts were implanted as inferior vena cava interposition grafts in a nude mouse model. H&E histology staining was performed on the grafts after excision 2 weeks post-implantation.

Results: Burst pressures were found to be 1025.4 ± 490.7 mmHg for 10% P(CL/LA) in acetic acid, 976.6 ± 342.9 mmHg for 10 % P(CL/LA) in dioxane, 1217.3 ± 435.5 mmHg for 15% in acetic acid, and 1084.1 ± 385.3 mmHg for dioxane. Figures 1 and 2 show the changes in mechanical properties over time due to *in vitro* degradation. Figure 3 demonstrates cell attachment and proliferation, while Figure 4 displays histological samples of explanted grafts from the mouse model.

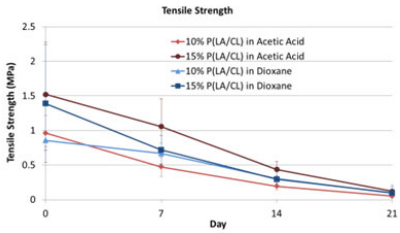

Figure 1: Tensile strength of grafts over time during *in vitro* degradation

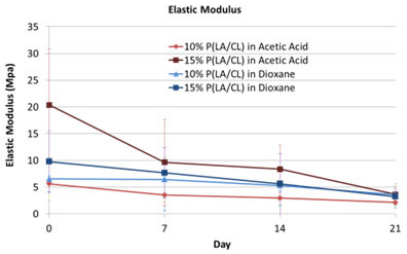

Figure 2: Elastic modulus of grafts over time during *in vitro* degradation

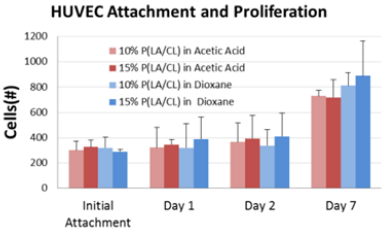

Figure 3: HUVEC attachment and proliferation on various grafts

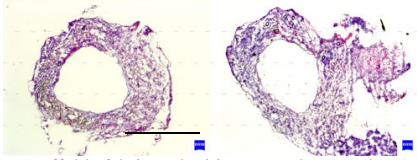

Figure 4: Scaffolds fabricated with 10% and 15% w/v P(LA/CL) in dioxane exhibit tissue ingrowth and patency *in vivo*. Scale bar is 1mm.

Conclusions: While there were no observable significant differences between the mechanical properties of the grafts despite differences in P(LA/CL) solutions, UTS and elastic modulus of the grafts were comparable to mechanical properties of native vessels.[4] Observed burst pressures were also above physiological pressure ranges.[3] Grafts experienced rapid degradation in an *in vitro* environment, virtually losing all mechanical strength by three weeks of degradation. Cell attachment and proliferation was ubiquitous in all graft groups. Importantly, HUVECs did proliferate over time, demonstrating cytocompatibility. Histological analysis of explanted grafts showed good patency and tissue ingrowth in all the grafts. These grafts appear to provide a good platform for future vascular graft modifications and studies.

References: 1) Greenwald SE, Berry CL. *J Pathol*. 2000:190:292-299. **2)** Nieponice A. *Tissu Eng A*. 2010:16:1215-1223. **3)** Roh J. *Biomaterials*. 2008:29:1454-63. **4)** Sell SA. *Biomed Mater*. 2006:1:72-80.