Electrospun Vascular Scaffolds as a Bypass Graft: A Feasibility Study

Bryan W. Tillman[1,2], Sang Jin Lee[1], Saami K. Yazdani[1], Anthony Atala[1], James J. Yoo[1]

[1] Wake Forest Institute for Regenerative Medicine and [2] Department of Surgery, Section on Vascular and Endovascular Surgery

Introduction

Nearly 500,000 peripheral bypass grafts are performed annually. Many of these patients require the use of prosthetic grafts to allow for reconstruction of diseased vessels. Despite their utility in the absence of autologous vessel replacement, permanent prosthetic materials present a lifetime risk of thrombosis and infection. In contrast to prosthetic materials, an engineered vascular conduit composed of a patients own tissue, particularly antithrombogenic endothelial cells would resist the risks of blood clots. Clinically, a decreased incidence of infection is observed in vascular reconstruction with autologous vein compared to permanent prosthetic graft materials and supports the concept that cell-seeded engineered constructs may similarly resist infection common in prosthetic materials. Despite advances in cell sourcing that have enabled generation of specific vascular component cell types, the scaffolds onto which these cells are seeded warrant further development. Scaffolds fabricated using methods derived from conventional manufacturing methods have a well defined composition and virtually unlimited size specifications, but lack the microarchitecture necessary for cell adherence, penetration and growth. The advent of electrospinning technology has enabled generation of a nanofiber based scaffold that possess the microarchitecture to facilitate cell growth but also allows for directed composition to facilitate controlled degradation as the scaffold remodels. Our initial prototype fabricated with PLGA based polymer, collagen and elastin showed adequate biocompatibility and mechanical characteristics for vascular applications. However, the electrospun graft failed to withstand physiologic flow and pressure when placed under a bioreactor system. We fabricated a reinforced vessel scaffold using poly(ε-caprolactone) (PCL) and collagen, which are designed to resist physiologic pressure and flow. *In vitro* testing in our laboratory has revealed that a mixture of polycaprolactone and collagen imparts the cell conducive properties of collagen with the longer term tensile properties of the biodegradable polymer polycaprolactone. We hypothesize that vessel conduits based upon an electrospun scaffold will be durable in the *in vivo* environment.

Methods

Electrospun vascular grafts composed of a blended polycaprolactone and collagen were fashioned with a diameter of 5 mm. The grafts were implanted as an aortoiliac interposition of New Zealand white rabbits using a proximal end to side anastomosis and a functional end to end iliac anastomosis with nonabsorbable suture material. The implanted vascular grafts were followed by ultrasound weekly for one month to evaluate graft diameter as well as patency. Representative animals underwent contrast enhanced CT scan or subtraction angiography at 4 weeks after implantation. Mechanical testing of explanted specimens was carried out using an Instron material testing system. Tissue specimens of pre-implant and post-implant (n = 4) were cut into strips of 8 mm x 5 mm for circumferential testing. Specimens were tested with a crosshead speed of 5.0 mm/min and peak stress at rupture was calculated and plotted. Histology was conducted on midgraft sections.

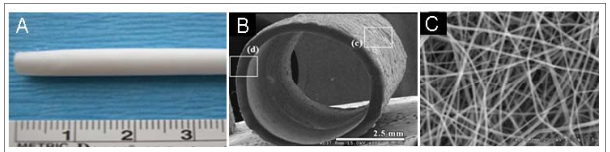

Figure 1: PCL-collagen electrospun in a tubular format (A,B) and enlarged electron microscope image (C).

Results

In vitro testing has previously demonstrated that electrospun scaffolds can be fashioned as tubular conduits (Fig. 1) that are durable under physiologic pressures. Subsequently, development of a rabbit model of aortoiliac replacement (Fig 2a) has been used for *in vivo* implantation of scaffolds (Fig. 2B). The electrospun graft material exhibited good suture retention and hemostasis at suture sites. Duplex ultrasonography illustrated a relatively stable graft diameter over time (Fig. 3A,B) for the duration of the one month study period. Overall 7 of 8 grafts (87.5%) remained patent despite the absence of endothelial cells. CT scan confirmed the absence of aneurysm or pseudoaneurysms of implanted grafts (Fig. 3C). Further, grafts showed incorporation with adjacent tissues. Biomechanical testing indicated that PCL/collagen grafts retained nearly 50% of their strength profile at one month (Fig. 4). Histology confirmed absence of significant inflammatory infiltrate (Fig. 5).

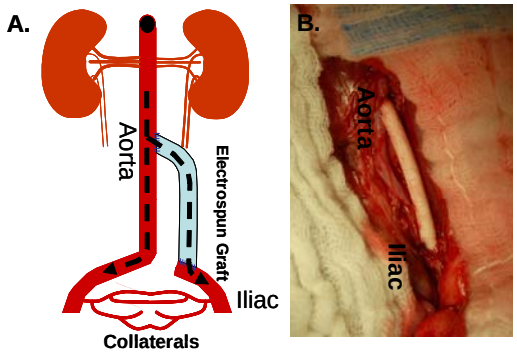

Figure 2: (A) Aortoiliac bypass model: an end to side anastomosis supplies blood to the left extremity, pelvic collaterals provide blood in the event of graft occlusion. (B) Operative photo of an implanted electrospun graft in the aortoiliac configuration.

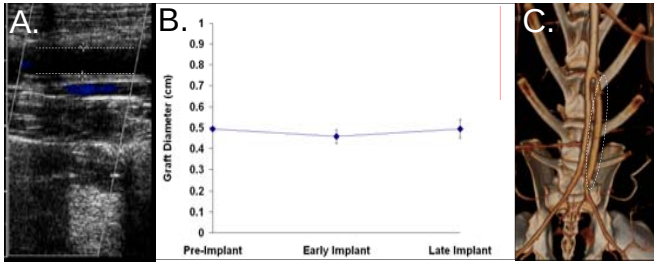

Figure 3: Ultrasonography used to assess graft diameter (A) at an early timepoint (1-2 weeks) and a late timepoint (4 weeks) indicates stable graft diameter over the duration of the one month study (B). Contrasted CT scan at 1 month (C) confirms absence of aneurysm or pseudoaneurysm in the graft.

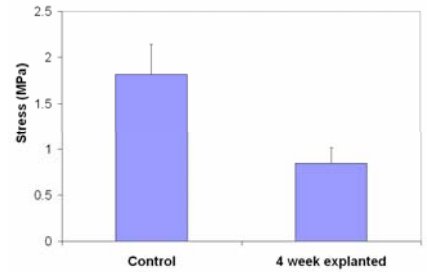

Figure 4: Biomechanical testing of explanted grafts compared to pre-implant controls (n=4) for each group reveals nearly 50% of strength remains at 1 month.

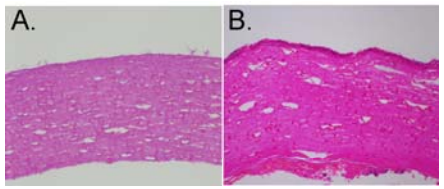

Figure 5: H&E histology of pre-implant electrospun graft (A) and 4 week explanted specimen reveals an absence of inflammatory infiltrate (B) adjacent to the graft on H&E histology.

Conclusions

Fabrication of vascular grafts using electrospinning techniques has become attractive due to the ability to control composition and configuration. In this study we investigated whether the new prototype would withstand normal physiologic vascular conditions in vivo. We examined the biomechanical properties, surgical handling and host tissue response of electrospun grafts when placed in continuity with the vascular circuit. This study demonstrates that electrospun grafts, composed of PCL and collagen, are able to maintain their structural integrity under normal physiologic vascular conditions in a rabbit model. Further studies are being performed to assess the long term efficacy of these grafts for arterial replacement as well as to examine the role of cell seeding with endothelial and smooth muscle cells to promote patency and remodeling, respectively.

References

Lee, S. J., Yoo, J. J., Lim, G., Atala, A., and Stitzel, J. (2007) In vitro evaluation of electrospun nanofiber scaffolds for vascular graft application, Journal of Biomedical Materials Research Part A in press.

Stitzel, J., Liu, J., Lee, S. J., Komura, M., Berry, J., Soker, S., Lim, G., Van, D. M., Czerw, R., Yoo, J. J., and Atala, A. (2006) Controlled fabrication of a biological vascular substitute, Biomaterials 27, 1088-1094.

Chew, S. Y., Wen, Y., Dzenis, Y., and Leong, K. W. (2006) The role of electrospinning in the emerging field of nanomedicine, Curr. Pharm. Des 12, 4751-4770.

Acknowledgements

We thank Cindy Andrews and Mandy Lockard for technical assistance