Long Term in vivo Study of Rapidly Degradable Synthetic Arterial Grafts

Robert A Allen,[1] Wei Wu,[1,2] Mingyi Yao,[3] Debaditya Dutta,[4,5] Xinjie Duan,[6] Timothy N Bachman,[3] Hunter C Champion,[3,5,7] Donna B Stolz,[8] Anne M Robertson,[6,9] Kang Kim,[1,4,5,9] Jeffrey S Isenberg,[3,7] Yadong Wang[1,9]

[1]Department of Bioengineering; [2]Qindu Hospital, Fourth Military Medical University, Xian, China; [3]Vascular Medicine Institute; [4]Center for Ultrasound Molecular Imaging and Therapeutics; [5]Heart and Vascular Institute; [6]Deptartment of Mechanical Engineering and Materials Science; [7]Division of Pulmonary, Allergy and Critical Care Medicine, Department of Medicine; [8]Department of Cell Biology and Physiology; [9]McGowan Institute for Regenerative Medicine; [1,3-8]University of Pittsburgh, UPMC, and University of Pittsburgh School of Medicine

Statement of Purpose: Patients without suitable autografts need small artery grafts for bypass, hemodialysis, or vascular repair.[1] This large clinical need remains unmet because clinically available vascular grafts fail rapidly at diameters less than 6 mm.[2] To address this clinical need, our group developed a cell-free small artery graft based on a fast-degrading elastomer. (**Fig. 1A**) This design is attractive because it can be available off-the-shelf at a fraction of the cost of tissue engineered grafts. Our group recently reported that these grafts transformed into 'neoarteries' (artery-like tissues) in rats with little residual material present at 3 months.[3] The objective of this study is to evaluate the *in vivo* performance of these artery grafts at 1 year post-implant.

Methods: Small artery grafts were made from porous tubes of poly(glycerol sebacate) (PGS) reinforced with a 15 μm-thin sheath of polycaprolactone (PCL) nanofibers as previously described.[3] Grafts were implanted as 8 to 10 mm long interposition grafts in the abdominal aortas of male Lewis rats. Grafts were soaked in 2 mg/ml heparin prior to implant, but rats received no additional anticoagulation or anti-platelet treatment. At 1 year post-implant, grafts were characterized in patency, tissue architecture, dynamic compliance, and vasomotor responsiveness. Dynamic compliance was measured *in vivo* by simultaneously measuring graft inner diameter using ultrasound and aortic pressure using an implanted manometer.

Results: *Patency* was 80% (4/5). *Gross remodeling:* grafts remodeled into 'neoarteries' with similar gross appearance to native rat aortas (**Fig. 1B**). Patent neoarteries showed no sign of stenosis, dilation, or aneurysm (**Fig. 1C**). Neoartery *tissue architecture and cellular organization* resembles the trilayered structure of native arteries (**Fig. 1D-E**). Of particular interest, neoarteries are innervated with perivascular nerves in their adventitia (**Fig. 1F**), similarly to native arteries. This is the first report of perivascular nerves infiltrating into the neo-adventitia of a vascular graft. Residual graft material is undetectable, indicating complete or near-complete graft resorption. *Extracellular matrix organization:* Neoartery elastin and collagen is oriented circumferentially (perpendicular to the direction of blood flow), similar to native aortas (**Fig. 1G,H**). Neoarteries were negative for von Kossa staining, suggesting an absence of calcification. *Dynamic compliance* was statistically the same between neoarteries and native aortas. (**Fig. 1I**). *Vasomotor response:* Neoarteries responded to a range of vasoconstrictors and vasodilators (**Fig. 1J**). Importantly, neoarteries demonstrated robust vasodilation in response to acetylcholine (Ach), suggesting a healthy endothelial lining. Neoarteries had decreased vasoconstriction but increased vasodilation compared with native aortas.

Conclusions: Taken together, these results demonstrate this graft design leads to functional arteries with – perivascular nerve-like structures, physiologic vasoactivity, and mechanical and biochemical properties highly resembling native arteries.

References:

[1] Baguneid MS. Br J Surg. 2006;93:282-290.
[2] Kannan RY. J Biomed Mater Res B. 2005;74:570-81.
[3] Wu W. Nat Med. 2012;18:1148-1153.

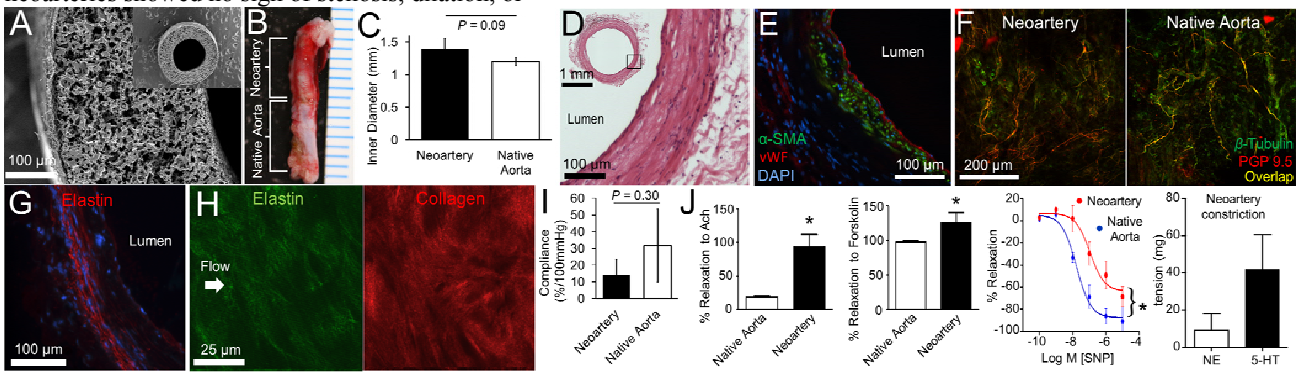

Figure 1. A. SEM image of transverse section of graft. Inset: Lower magnification. **B.** Gross morphology of explanted neoartery. **C.** Neoartery inner diameter. **D.** H&E of transverse section of middle of neoartery. Inset: lower magnification. **E.** Immunofluorescence for vascular cells in transverse section of middle of neoartery. **F.** *En face* confocal imaging of perivascular nerves (yellow) in neoartery adventitia. **G.** Immunofluorescence for elastin in transverse section of middle of neoartery. **H.** *En face* multiphoton imaging of elastin and collagen autofluorescence in the media layer of neoarteries. **I.** *in vivo* dynamic compliance. **J.** Vasomotor response of neoarteries measured by wire myography. Ach: acetylcholine (10 μM), SNP: sodium nitroprusside, NE: norepinephrine (100μM), 5-HT: serotonin (100μM). *$P < 0.05$