Pro-angiogenic and Anti-inflammatory Biomaterial Therapies for Peripheral Artery Disease

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Statement of Purpose: Peripheral artery disease (PAD) is characterized by the formation of arterial plaques, resulting in ischemia in extremities. Pro-angiogenic biomaterial therapies are a promising strategy for restoring blood flow and rescuing limbs from amputation.[1] For tissue regeneration in the context of PAD, the formation of highly vascularized tissue with minimal inflammatory response is desired, indicating the urgent need to consider simultaneous promotion of both anti-inflammatory and pro-ngiogenic responses. Therefore, we have developed combinatorial polymeric scaffolds to locally deliver functional peptides (i.e., pro-angiogenic C16 and anti-inflammatory Ac-SDKP) to the surrounding tissue of an occluded vessel.

Methods: Combinatorial copolymers of *x*% polyethylene glycol (PEG)-*y*% poly(ε-caprolactone) (PCL), -*z*% carboxylated PCL (CPCL) (*x, y, z*%: molar ratio) were fabricated into porous scaffolds by salt leeching. Pores (>80% v/v) of the scaffold were filled with collagen mixed with functional molecules, 75μg peptides, or 100ng lipopolysaccharide (LPS)/scaffold. As a model of PAD, the right femoral arteries of A/J mice were ligated and dissected; and peptide and collagen-filled scaffolds were implanted on top of the site of ligations before closing the wound. Hind limb perfusion was measured by laser Doppler perfusion imaging (LDPI) at 0, 3, 7, and 14 days post ligation. Perfusion values for the ischemic hind limb were normalized to the control limb (i.e., the non-ligated left leg of the same mouse) at each time point. Fluorescent microangiography was performed to visualize functional vessels in the scaffolds. After mice were sacrificed, the implanted scaffolds were harvested; and Vybrant phagocytosis assay (Life Technologies, Grand Island, NY) was performed to visualize and quantitate phagocytic cells in the harvested scaffolds.

Results: Our results revealed clear interdependency between angiogenesis and inflammation. When compared to no peptide control, both the number of phagocytic cells and the formation of functional blood vessels increased in the presence of pro-inflammatory LPS or pro-angiogenic C16 but decreased in the presence of anti-inflammatory Ac-SDKP (Figure 1A-C), indicating the intrinsic interconnectivity between angiogenesis and inflammation.[2] Interestingly, when the two types of peptides were co-treated, pro-angiogenic activities were promoted while limiting the number of destructive phagocytic cells, suggesting a promising therapy for PAD to increase angiogenesis without promoting detrimental inflammation. LDPI also revealed that the co-treatment of the two peptides increased blood perfusion of the hind-limb over 14 days compared to no peptide treatment or anti-inflammatory Ac-SDKP treatment (Fig. 1D). These findings validate the potential therapeutic efficacy of the dual peptide delivery to promote collateral vessel formation and restore blood flow to the ischemic limb.

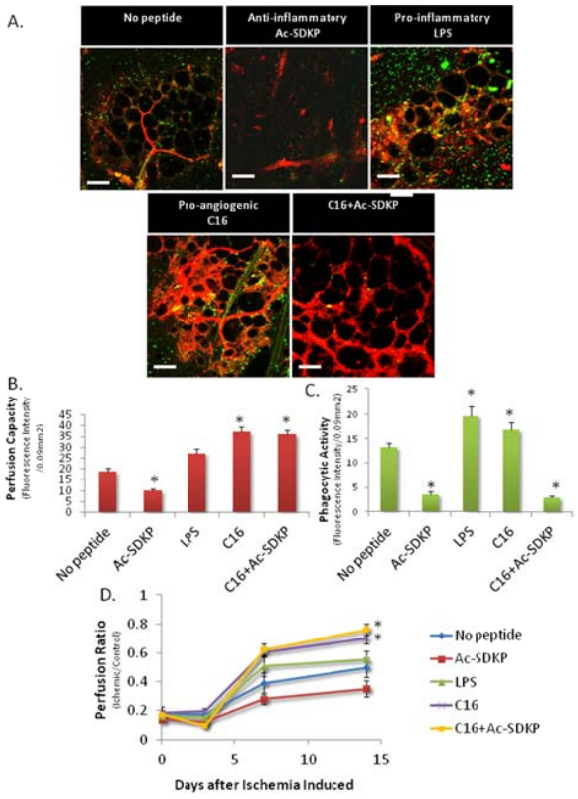

Figure 1. A) Confocal images of blood vessels (red) and phagocytic cells (green) in explanted scaffolds. B) Quantification of perfusion and C) phagocytic activity. D) Perfusion ratio of the ischemic/control hind limb as measured by LDPI 0, 3, 7, and 14 days after femoral artery ligation. B-D) *p<0.05 vs. no peptide control.

Conclusions: This study suggests that combinatorial polymer scaffolds loaded with C16 and Ac-SDKP peptides can be considered as a promising therapeutic means for PAD. Despite the interdependency between the two types of host responses, pro-angiogenic and anti-inflammatory processes were activated through co-treatment of the peptides. On-going works utilize injectable scaffolds made from the combinatorial polymer library for a minimally invasive treatment of the peptide therapeutics.

Acknowledgments: This work was funded by NSF 1006558, NIH HL091465, and AHA pre-doctoral fellowship 12PRE12070196.

References: [1] Pacilli A et al., Therapeutic Angiogenesis for PAD. Ann Vasc Surg. 2010;24(2):258-68. [2]Imhof BA, Aurrand-Lions M. Angiogenesis and inflammation face off. Nat Med. 2006; 12(2):171-2.