

Endothelial and smooth muscle cell interactions with a PCL-PU composite vascular scaffold with potential for bioactive release.

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INTRODUCTION: We have developed a polycaprolactone (PCL) – polyurethane (PU) composite scaffold material (Figure 1) which is a promising alternative for small diameter vascular grafts. This scaffold could provide increased compliance and thus reduce mismatch with native blood vessels. The scaffold also provides a favourable luminal surface for endothelial cell (EC) attachment, as well as a porous anti-luminal surface for smooth muscle cell (SMC) attachment and migration.

In this study, we examined how human umbilical venous endothelial cells (HUVECs) adhered to the luminal surface of the prosthesis and, for an improved functional vascular graft, SMCs were seeded to the anti-luminal surface of the prosthesis.

METHODS: The morphology and phenotype of HUVECs and SMCs was assessed on the scaffolds using environmental scanning electron and immunofluorescence microscopy. Endothelial functional behavior assays were used to assess the release of nitric oxide and von Willebrand factor (vWF) under stimulation.

The demonstration that sustained release of therapeutic proteins is possible has stimulated the development of new implantable polymers and devices which controlled delivery of growth factors (3). The scaffolds were loaded with 0.67% and 0.5% (w/w) lyophilised trypsin (25kDa) (a “model” growth factor) to assess protein release and activity in vitro over a period of 4 days.

RESULTS: The HUVECs demonstrated strong cell attachment to the scaffold, approx. 60% compared to tissue culture plastic (TCP). After 7 days of culture, the cells exhibited endothelial markers and had become confluent on the luminal surface of the scaffold. When stimulated, the HUVECs released levels of nitric oxide and vWF that were comparable TCP standard conditions in vitro.

SMC showed strong cell attachment to the PU (or anti-luminal) surface of approx. 50% this could be increased slightly by the absorption of fibronectin (FN) or fibrillin-1 RGD fragment PF8.

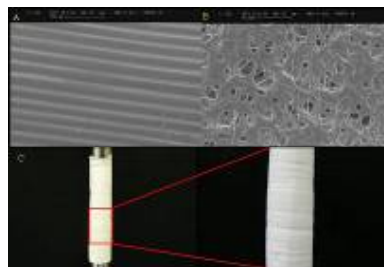


Figure 1 Scanning electron microscopy (SEM) of the 2-ply scaffolds luminal (A) and anti-luminal (B) surface topography. Luminal surface (50x) shows originated PCL fibres with a fibre to fibre gap of approx. 1-5µm. The Anti-luminal surface (400x) shows a highly porous topography with a pore size ranging from 10-30µm. (C) Digital picture of the tubular 2-ply scaffold

Release of total trypsin was only detectable at 24 hours, with a release of approximately 55%. However active trypsin release was confirmed at 24 and 48 hours due to an increase of absorption at 258nm.

DISCUSSION and CONCLUSIONS: The favourable luminal surface for HUVECs attachment coupled with the functionality of the cells and the high attachment of SMCs on the anti-luminal surface, combined with the possibility of active local release of growth factors to the newly regenerating tissue, recommend this scaffold in vascular and other soft tissue engineering.

REFERENCES: [1] Williamson et al. *Biomaterials* 2004;25:5053-60 [2] Kakisis et al. *Journal of Vascular Surgery* 2005;41:349-354 [3] Williamson et al. *Biomaterials* 2006;27:3608-3616

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