Enhanced *in vivo* Cell Infiltration and Vascularisation by Dermal Fibroblasts/Novel PLLA-Collagen Scaffold Constructs for Abdominal Wall Replacement

Fanrong Pu¹, Nicholas P. Rhodes¹, Yves Bayon², and John A. Hunt¹

Corresponding author: Fanrong Pu; e-mail: frpu@liv.ac.uk

¹UKCTE, Clinical Engineering, School of Clinical Sciences, University of Liverpool, Liverpool, UK and ²Sofradim Production, 01600 Trevoux, France

**Introduction**

Dermal fibroblasts constitute a potential autogenous source of cells for abdominal wall tissue engineering where they play a key role in extracellular matrix remodelling and new blood vessel formation. In this study, it is demonstrated that dermal fibroblasts seeded in a novel PLLA-collagen scaffold enhanced cell infiltration and vascularisation *in vivo*. It is envisaged that this approach could be a viable strategy for the repair of human hernias.

**Materials and Methods**

The PLLA-collagen composite scaffolds used had a unique structure with a collagen sponge formed in the space of a mechanically stable knitted mesh of PLLA (Fig. 1a, b). Scaffolds 5cm in diameter were seeded with normal human dermal fibroblasts at a density of $10^6$/cm². Cell-loaded scaffolds were placed into a perfusion bioreactor designed by Applikon and operated as a culture system for 7 days. For *in vivo* studies, both acellular and cell-loaded constructs (1 cm²) were implanted subcutaneously in Wistar rats. The implants were removed at day 2 and 7 and histopathology performed using both tinctural and immuno-histochemical staining to analyse the effect of cell-loading on the cell infiltration and vascularisation of scaffolds compared to the controls.

**Results**

The *in vitro* studies demonstrated that the PLLA-collagen scaffolds supported cell proliferation, confirmed by cell viability assay and SEM observation (Fig. 1c, d). It was further facilitated by the flow perfusion system, which was shown to increase cell proliferation and homogeneous distribution within the PLLA-collagen scaffolds (Fig. 2). A highly cellularised 3D-tissue construct was formed by 7 days incubation in perfusion culture *in vitro*. The *in vitro* model demonstrated that the constructs with high cellularity resulted in more cell infiltration (Fig. 3a) compared to acellular control (Fig. 3c).

Furthermore, more mature blood vessels were observed on the scaffold seeded with fibroblasts (Fig. 3b, d), demonstrated by immunostaining for the blood vessel marker, smooth muscle actin.

**Discussion and Conclusions**

It was demonstrated that the use of dermal fibroblasts cultured within a porous scaffold *in vitro* in a flow perfusion bioreactor system had a significant effect on an *in vivo* model of the reconstruction of abdominal wall tissue.

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