A novel test bed for 3D cell recruitment under hemodynamic conditions

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Introduction

Recently, in situ tissue engineering has emerged as a new approach to obtain autologous, living replacement tissues with off-the-shelf availability [1]. The method is based on the use of a ‘smart’ degradable scaffold that is capable of repopulation with host cells in situ and subsequent tissue formation. For cardiovascular grafts, this repopulation (with circulating cells from the blood) is hypothesized to be highly influenced by the hemodynamic environment (Fig. 1).

The aim of this study is to develop an in vitro test bed to study cell recruitment in 3D scaffolds under physiological hemodynamic conditions.

Cross-flow chamber

A cross-flow chamber for 3D scaffolds was developed in-house. Computational validation of the chamber demonstrates a fully developed flow in the region of interest with a homogeneous stress distribution at the scaffold surface (Fig. 2 A-C). For pulsatile flow conditions, the flow profile remained strictly within the laminar regime.

Cell recruitment

A feasibility study was performed to study cell recruitment in a 3D electrospun scaffold under dynamic conditions. Human peripheral blood mononuclear cells (PBMC) were labeled with Cell Tracker Green (CTG) and run through the fluidic system at physiological flow rates and pressures. Results show a gradual increase in cell density in the scaffold over time (Fig. 4). Cell infiltration into the scaffold was predominantly dependent on the applied pressure difference.

Figure 1. Hypothesized influence of physiological wall shear stress ($\tau$) and transmural pressure difference ($\Delta p$) on cell recruitment from the bloodstream (A). Schematic of the translation into an in vitro model system (B).

Figure 2. Design of the cross-flow chamber (A) with predicted shear stress distribution at the scaffold surface (B). For steady-state flow, the flow at the scaffold is fully developed with a parabolic profile in height and minimal wall effects in transverse direction (C). Snapshot of fluorescent microbeads in flow (scale = 500 µm) (D).

Figure 3. Image (A) and schematic (B) of the fluidic system with the cross-flow chamber incorporated. Representative examples of the generated pressure differences over the scaffold (C) and corresponding flow rates in the fluidic channel (D).

Figure 4. Representative images of a flow experiment revealing a gradual recruitment of CTG-labeled PBMC into the scaffold over time.

Conclusion

Recruitment of circulating cells is dependent on a range of parameters (e.g. scaffold architecture and incorporated biochemical stimuli). To study this, it is critical to take the hemodynamic environment into account. We successfully developed and validated a fluidic system that provides a valid screening platform to study cell recruitment in 3D scaffolds under hemodynamic conditions in vitro.