Modular Tissue Containing EC and MSC formed in a Microfluidic Perfusion Chamber

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Introduction
Using a modular approach, vascularized tissue can be created by embedding functional cells within submillimeter-sized collagen cylinders (modules) while the outside surfaces are seeded with endothelial cells (EC). The void spaces created by randomly packing the modules into a container form EC-lined channels through which perfusion occurs. When mesenchymal stem cells (MSC) are embedded within the EC-coated modules, we hypothesize that the shear stress (and mechanical loading) produced by perfusion will induce MSC to differentiate into smooth muscle-like cells that will migrate to a pericyte position and support long term EC survival.

Materials and Methods
Module perfusion chambers were produced via photolithography with SU-8 photoresist (Microchem) and Sylgard 184 silicone elastomer (Dow Corning). Collagen (Sigma) modular cylinders were fabricated as described in [1]. Module surfaces were seeded with rat aortic EC (VEC Technologies). Embedded MSC were isolated from rats [2]. Modules were loaded into chambers and perfused with a 1:1 mixture of MCDB-131 complete medium (VEC Technologies) and DMEM with 10% FBS (Sigma) and 1% penicillin/streptomycin (Gibco) for up to 21 days at 0.5 mL/min. At days 7, 14 and 21, modules were removed from chambers and analyzed using histology.

Results
As illustrated in Figure 1, the flow-conditioned EC influenced MSC to differentiate into smooth muscle actin (SMA)-positive cells, which migrated to a supporting position below the EC. This was not observed in static controls. As the MSC migrated, they created new matrix which included the deposition of proteoglycans. EC were found on the surface of static control modules throughout the time course, however, by day 21, the majority of EC were lost with perfusion, despite having SMA-positive MSC present in a supporting position. This loss may be related to timing in that the MSC were unable to differentiate and migrate quickly enough to promote EC retention. With the EC gone, the migrated MSC remained near the module surfaces within their deposited proteoglycan matrix.

Discussion and Conclusions
MSC were used with the intent to aid in the long term survival of EC in a modular assembled tissue. The inability to prevent the eventual loss of EC despite observed changes in the MSC may require further optimization of the co-culture conditions or the way in which the MSC are deployed.

References

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Disclosures
Nothing to disclose.