Ultra-Rapid Pre-Fabrication of Biomimetic Collagen 'Tissues': Generating Meso-Scale Structure with Hyaluronan.

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INTRODUCTION:

Plastic compaction of hyper-hydrated native collagen gels (Brown et al 2005) represents a radical and new approach to the fabrication of biomimetic tissues & templates (as opposed to biomaterials-based tissue-equivalents). It allows the *rapid engineering* of meso-structure by directional water removal. The deformation is plastic (ie constructs retain their new structure) since the collagen has little inherent swelling potential. However, a further level of biomimesis could be achieved by local incorporation of osmotically active (swelling) macromolecules – a common example being hyaluronan. In this study, the GAG, hyaluronan, was locally incorporated into high density, ***cell-free,*** compressed collagen constructs with the aim both of identifying new techniques for rapid tissue engineering and as a model to test how connective tissue cells might utilise such mechano-osmotic fluid flow, in vivo.

METHODS:

Freeze-dried hyaluronan was applied to localised areas of freshly prepared Plastic Compacted (PC) collagen sheets (~50um thick), particularly along the short edges. Constructs were immediately formed into spirals by routine rolling from the short edge, ie., leaving a strip of hyaluronan at the core of the spiral. Spiral constructs were then incubated in water for periods from 1 min. to 3hrs at which point they were fixed for scanning electron microscopy.

RESULTS

welling of the core region was rapid, within the first 10 -30 min and largely complete in 1-2h. Over this time the thickness of outer collagen layers appeared to reduce dramatically, forming a clear and continuous channel along the full-length of the construct – essentially producing a tube structure (Fig. 1).

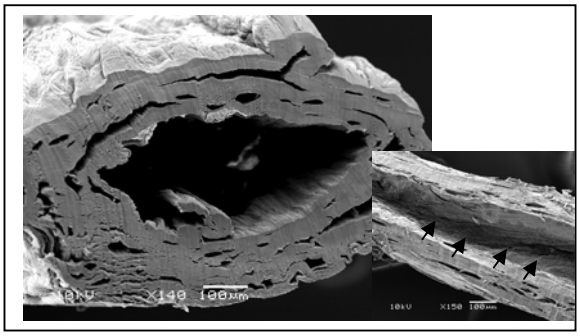

Fig. 1: SEM (main picture) of the core channel in TS. formed within a spiral collagen PC construct by hyaluronan, post compaction re-swelling. INSERT: LS view showing the full length channel (arrows).

DISCUSSION & CONCLUSIONS:

The prediction here was that hyaluronan would re-swell those areas of the PC collagen construct where it was localised, leading to cavity formation. This was indeed the experimental result, representing a rapid and localisable technique for inducing new structures and interfaces within the constructs, in this case as a tube. However, it was also found that the collagen layers themselves were further compressed, presumably by additional loss of fluid to the swelling polysaccharide gel. This presents the intriguing possibility that cells might *naturally* use a timed secretion of such GAG molecules to manipulate local tissue structure, and to form interfaces between fibril bundles. Local structural re-swelling represents important new biomimetic approach for engineering tissues and a model for investigation of natural cell systems.

REFERENCES:

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