Tissue Engineered Muscle-Tendon Junction 3D Scaffold Development and Evaluation

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Introduction:
The US orthopedic biomaterials market is predicted to grow to nearly $3.7 billion by 2017. Contrary to this large market demand, there is still a lack of reliable biomaterials that can repair complex musculoskeletal tissue such as a muscle-tendon junction (MTJ). Although success has been reported from the tissue engineering of single cell types, there is a growing demand for an appropriate combination of cell types with a gradient structure. The goal of our study is to develop and evaluate 3D tissue engineering scaffolds that mimic the anatomical, mechanical, and physiological characteristics of native MTJ tissue.

Materials and Methods:
Polyethylene terephthalate (PET) was melt spun into 150 denier multifilament yarns with a 12.5µm filament diameter. The ideal structure was selected from ProCAD, a 3D simulation design software. The latest warp knitting technology was applied to fabricate the design of a 3D prototype which has commercial potential. The flow of work includes:

After fabrication of the knitted samples with three distinct structures to mimic the characteristics of the muscle, tendon and the interface of the MTJ, physical and mechanical tests have been performed under dry-conditions. Then collagen was applied on the textile structure to activate the surface. Primary human skeletal muscle cells and dermal fibroblasts will be cultured on the scaffolds for a period of four weeks in a dynamic stretching bioreactor. At four different time points, a systematic evaluation will be conducted, including mechanical tests under wet-conditions, cell viability (MTT assay), histology and immunohistochemistry for MTJ specific marker [2].

Results:
The three component structure has been simulated in 3D using ProCAD software and has been warp knitted as a continuous structure with three different levels of porosity all connected to each other (Figure 1). The overall porosity and pore size distribution of the three structures has been measured using ImageJ image analysis software to qualify the void spaces in cross-sections viewed by optical microscopy. The mechanical test results indicate that fabric structures have superior tensile strength and elongation than normal human tissue in both the muscle and tendon regions. Preliminary biocompatibility studies using an MTT viability assay, scanning electron microscopy (SEM) and laser scanning confocal microscopy (LSCM) after 14-days of culture with human dermal fibroblasts indicated good cell attachment to the scaffold surface and proliferation through the thickness of the construct (>2mm) (Figure 2).

Conclusions and Future Work:
A series of PET prototype knitted structures with a continuous porosity gradient have been successfully developed and show appropriate structural characteristics, mechanical properties and biocompatibility. Future work will involve the use of an equivalent PLLA knitted structure and a dynamic co-culture study involving both smooth muscle cells and fibroblasts, which will be followed by further mechanical testing, histology and immunohistochemistry.

Reference:

Abstract #610
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