Development of Automated Loom for Woven Tissue Engineering Test Systems

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Statement of Purpose:

Tissue engineering has been defined as the application of multidisciplinary principles toward treatment development for regeneration of functional tissue. These treatments involve the combination of cells with scaffolds. Many biodegradable synthetic polymers such as polyglycolide (PG), polylactide (PL), poly-lactide-coglycolide (PLG), and polycaprolactone (PCL) have been used in scaffold development for support of tissue regeneration.²

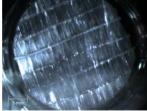
One area of focus in polymeric scaffold engineering has been in surgical mesh development. Surgical mesh efficacy is related to mesh composition, filament structure, and amount of synthetic material.³ Therefore, researchers have focused on the design and fabrication of reproducible, bioactive and absorbable 3D scaffolds with tailored properties.⁴ The goal of this work was the development of an automated loom capable of creating woven scaffolds with variable properties for the evaluation of mesh combinations in vitro. Loom effectiveness was evaluated by assessing loom capability to create meshes with defined structure and ability to support cell attachment.

Methods:

An automated loom was constructed with an extruded aluminum frame of 1.2 m x 0.6 m dimensions. Movement of loom components was accomplished pneumatically with laboratory house air. Electronic components were controlled via National Instruments LabVIEW (National Instruments). The weave configurations tested were 50/50 and 75/25 compositions in an over/under pattern. Woven meshes consisted of the following three synthetic polymer combinations of warp and weft fibers: polylactide (PL; Natureworks LLC, 2003d biopolymer, ~ 228,000 Da):PL, PL:poly-l-lactide-co-caprolactone (PLCL; Purac, Purasorb PLC 7015, ~ 154,500 Da), PLCL:PLCL. Meshes were woven on the automated loom and stored under vacuum until cell studies were conducted. Prior to cell culture, meshes were cleaned in 75% ethanol, then treated with ultraviolet radiation for 24 hr. D1 murine mesenchymal stem cells (ATCC, passage 5-8) were cultured on each mesh in non-treated 12-well plates (Corning). Cells were cultured in Dulbecco's Modified Eagle's Medium (DMEM; Atalanta Biologics) supplemented with 10% fetal bovine serum (FBS; Life Technologies), antibiotic-antimycotic (Life Technologies), and fungizone (Life Technologies). Each mesh was cultured with D1 cells for 7 days, with the first 24 hours of culture on an orbital shaker at 100 rpm. Culture medium was changed every 3 days. Cell attachment was assessed qualitatively using Live/Dead® Cell Viability Assays (Life Technologies).

Results:

The automated loom facilitated the development of meshes with tunable weave configuration and material type. The stereoscope image in Figure 1a illustrates one of the PL:PLCL meshes constructed with a 50:50 weave configuration. Cell attachment results indicated a significant number of viable cells after 1 week in culture. However, meshes containing PL weft fibers had more dead cells than the other material combinations. There was no significant difference in cell attachment with respect to warp fiber material. Figure 1b shows the Live/Dead® results of a cellular PL:PL mesh after 1 week in culture. Cell affinity results for 75:25 weave configurations were inconclusive due to 3D imaging limitations using the fluorescent Live/Dead® system. The close proximity of weft fibers in this configuration compounded fluorescence inconsistencies and inhibited cell identification.



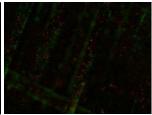


Figure 1: A) Stereoscope image of a 50:50 PL:PLCL mesh B) Live/Dead® image of 50:50 PL:PL mesh at 50x total magnification. Red indicates dead cells. Green indicates live cells.

From the results gathered through cell attachment / affinity studies of multiple mesh constructions, it can be concluded that the automated loom is able to create woven tissue engineering scaffolds with variable properties for in vitro study. The ability to vary material composition will lead to the ability to tailor tissue engineering meshes to specific anatomical locations, targeting specific mechanical properties or degradation rates. Manipulation of weave configuration will lead to the ability to affect pore size, fluid transport, and the amount of material in the construct. All of these variables have been shown to affect biologic response in vivo.³ Quantitative techniques to compare cell attachment should be employed in the future to more closely compare specific combinations and eliminate the ambiguity of assessment via qualitative fluorescence.

Acknowledgements

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