Mechano-dependent Biosynthetic Response of Cell Micro-integrated Elastomeric Scaffolds

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Introduction

This study examines the biosynthetic effects of cyclic mechanical strain placed on a poly (ester urethane) urea PEUU scaffold densely integrated with rat vascular smooth muscle cells with the overall goal of developing an engineered tissue that mimics the load bearing capabilities of native tissue.

- The field of tissue engineering combines the principles of biology and engineering in an effort to create biological substitutes that recapitulate the requisite mechanical and structural properties of healthy native tissues to restore, maintain, or improve tissue function [1].
- Approximately 75,000 patients per year in the US receive prosthetic heart valves, and it is estimated that this number increases to 250,000 worldwide [2].
- Current valve replacement solutions have proven to prolong the lives of those living with vascular disease, but there still remains to be an ideal solution to valve replacement.

Methods

Cell Expansion

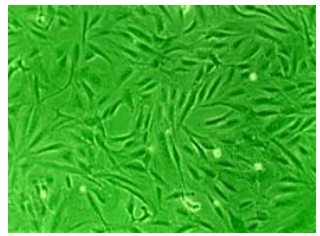

Figure 1: Vascular smooth muscle cells isolated from rat aorta ☐ www.genlantis.com/corp/images/RAOSMC%20cells.jpg

- Cells were expanded onto tissue culture plates under Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum, 2% anti anti and 1% HEPES solution.
- After expansion, cells were incubated at 37 °C and 5% CO_2 for approx. 3-5 days until fully confluent.

Specimen Preparation

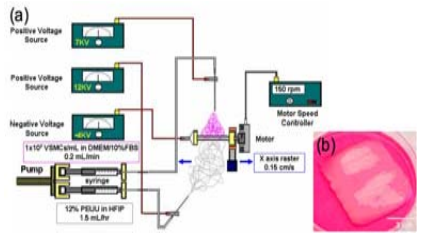

Figure 2: (a) Process of electrospinning the PEUU fibers while concurrently electrospraying cells resulting in (b) a scaffold that exhibits tissue like mechanical properties [3].

- Process involves depositing a solubilized polymer across a voltage gradient onto a rotating mandrel
- Provides a suitable environment for cell proliferation and growth.

Dynamic Conditioning

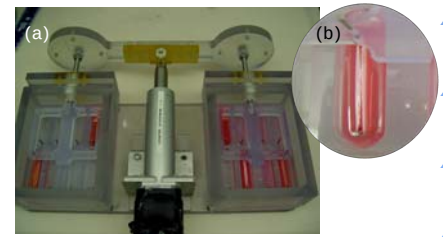

Figure 3: (a) Cyclic tension bioreactor used to mechanically condition specimens. (b) Magnification of one cell loaded with a specimen

- Experimental groups consisted of Day 0 control, Day 7 static, Day 7 15% strain, and Day 7 30% strain. A duty cycle of 1 Hz use for all specimens.
- Specimen were assessed via soluble collagen and proteoglycan DNA production as compared to day 7 static and day 0 control.

Results

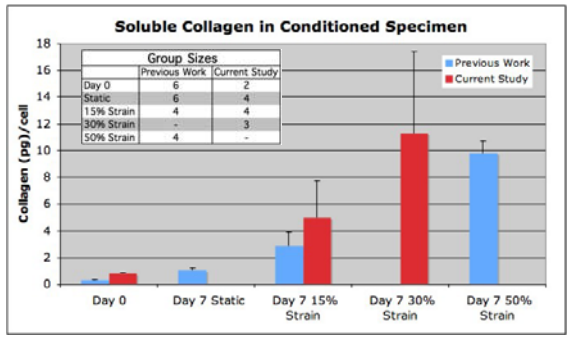

Figure 4: Collagen quantification. Large strain was seen to induce a statistically significant increase in soluble collagen production vs. Day 0 and static controls. Preliminary results (30% strain) suggest the potential for a deformation dependent plateau in matrix synthesis.

- From previous work by Stella et al., a large strain (50%) was seen to induce an increase in soluble collagen per cell.
- From the current study, Day 7 30% specimen showed an increase in soluble collagen compared to day 0 and static specimen.
- Specimen conditioned at 30% strain exhibited a higher collagen production per cell than those conditioned at 50% strain possibly indicating a peak strain for maximum collagen production.

Discussion

- Electrospinning PEUU fibers while concurrently electrospraying viable cells provides the opportunity to:
  - Characterize the mechanical behavior of the scaffold in response to dynamic conditioning
  - Evaluate the scaffold as a functional tissue
- Both the previous and current studies suggest that mechanical strain may induce an increased extracellular matrix production in artificial scaffolds.

Future Studies

- Extend current study to 7 and 14 day endpoints.
- Assess the mechano-dependent biosynthetic response in a simulated physiological environment with a previously developed organ bioreactor.
- It is expected that the mechanical effects of the simulated pulmonary valve environment will cause an increase in extracellular matrix production [4].
- It is our hope that these studies will guide the emergence of new materials and processing methods to develop functional pulmonary valve (PV) tissue surrogates.

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2) Nick Amoroso and Dr. William R. Wagner

References

[1] Langer and Vacanti. Tissue engineering. Science 260, 920-6; 1993.
[2] American Heart Association. 2002.
[3] Stankus et al. Biomaterials v.27(19) pp. 735-44
[4] Stella et al. Society for Biomaterials Annual Meeting, San Antonio, TX, April 2009.