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The relevance of high strains in tissue engineering of heart valves

A. Mol¹, S.P. Hoerstrup³, C.V.C. Bouten¹, J.F. Visjager², C. Guenter³, G. Zünd³, M.I. Turina³, F.P.T. Baaijens¹

1. Eindhoven University of Technology, Department of Biomedical Engineering

2. Swiss Federal Institute of Technology Zurich, Department of Materials

3. University Hospital Zurich, Clinic for Cardiovascular Surgery

Introduction

Tissue engineered heart valves (TEHV) show promising results in animal studies when placed in pulmonary position [1,2]. They, however, still lack mechanical strength to resist stresses in the aortic position. This study aims at improving the load-bearing properties of TEHV by applying cyclic strains on tissue engineered constructs in order to improve the conditioning protocol which will be applied in the bioreactor.

Material and methods

- Cells: human saphenous vein cells p5-6
- Scaffold: PGA/P4HB
- Culturing: in a custom-made straining device (fig. 1) under increasing cyclic strain (max. 7-10%) up to 21 days.

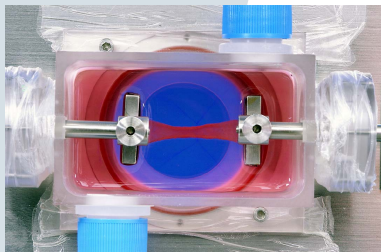


Figure 1 Photograph of the culture chamber of the straining device containing a tissue engineered construct.

- Qualitative analyses: histology, SEM, TEM and visualization of cell orientation and viability by CLSM using the viable CTG/PI staining.
- Quantitative analyses: DNA, GAG, hydroxyproline and uniaxial tensile tests.

Results

Qualitative analyses showed more pronounced and organized tissue formation in strained constructs versus unstrained controls (fig. 2).

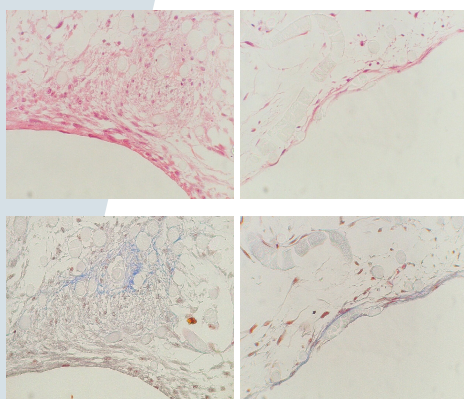


Figure 2 Top: HE 40X, bottom: M. Trichrome 40X of strained (left) constructs versus unstrained (right) constructs.

Large strains resulted in a smooth and dense surface layer (fig 3).

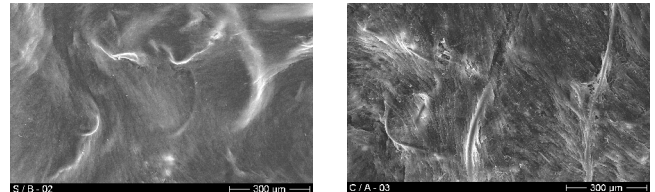


Figure 3 Scanning Electron Microscopy of the surfaces of strained (left) constructs versus unstrained (right) constructs.

DNA, GAG and collagen content as well as mechanical properties (fig. 4) were significantly increased in strained constructs at strain levels exceeding 9%.

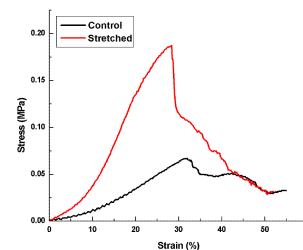


Figure 4 Load-bearing properties of strained constructs versus unstrained constructs.

The constructs displayed high viability throughout the culture period and the cells were observed to orient predominantly in the direction of the applied strain, whereas unstrained controls showed more random orientations (fig. 5).

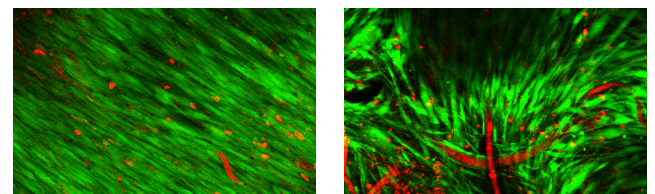


Figure 5 Cell orientation and viability visualized by Confocal Scanning Microscopy of strained (left) constructs versus unstrained (right) constructs.

Conclusions

Mechanical conditioning using cyclic strain represents a promising method to improve the load-bearing properties of TEHV. We conclude that large strains are beneficial in early tissue development, whereas shear stress appears critical prior to implantation to enable the production of living heart valve replacements capable of withstanding the systemic hemodynamic stresses in aortic position.

References

1. Sodian R et al. Circulation 2000; 102(19 suppl III): 22-29
2. Hoerstrup SP et al. Circulation 2000; 102(19 suppl III): 44-49