ENGINEERING CARDIAC TISSUE

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Introduction

There are currently 5.7 million Americans living with heart failure and 670,000 new cases diagnosed each year. One reason for heart failure is weakened, dead or scarred tissue on the heart as a result of myocardial infarction. Heart transplant could be a useful therapy, but the effects of rejection, the need to take immunosuppressant drugs and the dire lack of available tissue are all deterrents to transplantation. A solution to some patient's transplant needs lies in the creation of a tissue graft that could be placed over the injured heart tissue to help repair the patient's original heart. Bioartificial tissues created with a patient's own cells are a solution to creating a graft of heart tissue that would avoid rejection.

Motivation and Methods

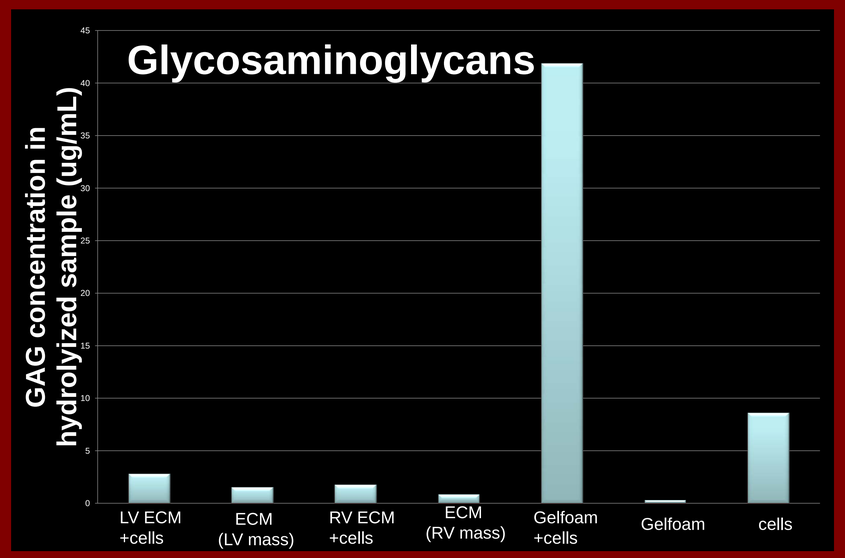
Objective: Characterize cardiac derived cells on different scaffolds, both biological (extracellular matrix) and artificial (gelfoam), that could be used for a therapeutic tissue graft.

- Rat heart extracellular matrix obtained by cell removal with SDS. (fig. 1)
- Left and right ventricle patches dissected or gelfoam.
- Neonatal cardiac-derived cells isolated from neonatal rats using a commercially available kit. (Worthington)
- Cell suspension seeded onto the adherent extracellular matrix, gelfoam, and tissue culture dish for control.
- Cultures kept in carbon dioxide incubator; media changed and cultures observed once daily for 8 days.
- On day 8, cultures collected, RNA extracted from half for qPCR, NaOH hydrolysis of other half for glycosaminoglycans and DNA assay.
- Gene expression will be compared between conditions after qPCR is run for cardiomyocyte, endothelial and coupling genes.

*Rat heart and neonatal cardiac derived cell isolations performed by Stefan Kren.

Results

- Preliminary data on GAG assay gave insight on scaffold conditions and GAG contribution by cells over growth period.
- GAG in gelfoam incubated with cells is far greater than gelfoam alone or matrix with cells.
- ECM and gelfoam controls normalized by mass.
- Final results will be established when quantitative PCR is complete and DNA and GAG assays are completed on a second round of samples.



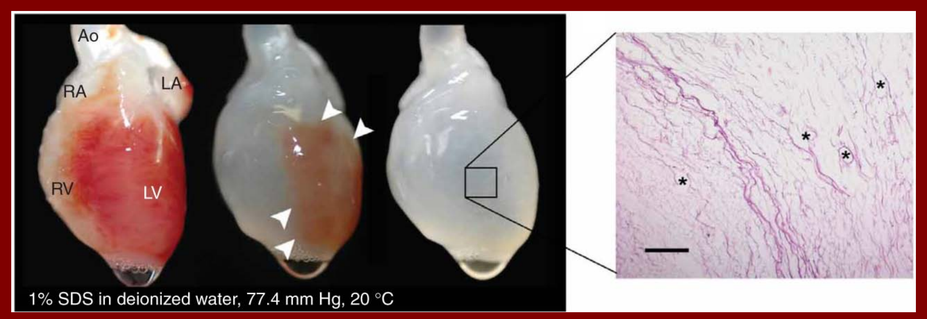


Figure 1. Three stages of rat heart decellularization. The third stage is the resulting extracellular matrix. [1]

Discussion and Conclusions

- More GAGs present in cells on gelfoam and plain cell cultures than in cells on ECM; most of which was produced in culture over growth period.
- For each of the conditions, qPCR will include quantitative measurement of the expression of these genes :
  - Connexin 43
  - Platelet endothelial cell adhesion molecule (CD31)
  - Myosin heavy chain alpha
  - Gap junction protein 1, cardiac troponin T
- Further insight could be obtained from analysis of transcriptional changes of specific target proteins by Western Blot.
- Insight obtained from this experiment will provide insights on the effect of decellularized extracellular matrix on cells. Insights into cell mechanisms will help optimize the engineering of a cardiac patch, which could be an alternative to some heart transplants. Patches may become an alternative to transplant for patients with heart failure.

References

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