ABSTRACT SUBMISSION FORM

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SEX + GENDER

Exploring the role of sex and gender on health research

CHILD HEALTH RESEARCH DAYS

CHRD 2020: Abstract Submission Form

Submitter Name
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Title
Aprotinin and fibrinogen improve the mechanical stability of a 3D bioprinted model of skeletal muscle

Background
Despite shortcomings, 2D cell culture remains an essential tool for biomedical research. We are developing a 3D bioprinted model of skeletal muscle that can more accurately recapitulate health and disease conditions, potentially making it a superior model for disease research. However, contractile forces generated by the differentiating muscle cells are sufficient to tear the tissue apart.

Objective
Improve the mechanical stability and contractile phenotype of bioprinted skeletal muscle tissue with fibrinogen and the anti-fibrinolytic agent aprotinin.

Methods
C2C12 skeletal myoblasts were bioprinted in a 'bioink' comprised of 1 mg/mL collagen, 0.25% alginate and 5-20 mg/mL fibrinogen. The muscle ring was encapsulated within an acellular support structure containing 1% alginate. After printing, tissues were thrombin treated to polymerize fibrinogen and maintained with or without 20 mg/mL aprotinin for 3 days. Tissues were then differentiated for 7 days with or without aprotinin and assessed for histology (gross morphology, actin staining) and contractile phenotype (qPCR for contractile markers).

Results
Bioprinted muscle cultured without aprotinin began to visibly degrade by day 2, with the 5 mg/mL condition having poor structural integrity and weak myotube formation. Culturing with aprotinin resulted in a dramatic improvement in tissue structure at all fibrinogen concentrations, with extensive formation of elongated and multinucleated myotubes consistent with in vivo tissue. RNA yields were 3-7 fold higher in the aprotinin groups ($p<0.05$), yet markers for contractile phenotype were not significantly different when normalized against reference genes. This indicates aprotinin does not improve individual cell differentiation, but

significantly increase the yield of differentiated cells.

Conclusion
Culturing bioprinted skeletal muscle in aprotinin dramatically improved tissue structure and myotube formation, to the point that our tissues are now suitable for performing contraction assays. This represents an important development towards our goal of creating an experimental model to improve muscle disease research.

Theme:
Basic Science

Do you have a table/figure to upload?
No

Are you willing to participate in Goodbear's Den?
Yes

Presenter Status:
Non-Trainee

What was your role in the project?
Design

Authors

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