Preliminary optimization of supercritical decellularization

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INTRODUCTION

Organ transplantation comes with many risks, such as organ rejection, lifelong immune suppression, and drastically reduced life expectancy. Using an extracellular matrix (ECM) derived tissue scaffold to engineer a new personalised organ is a promising alternative that can minimize these risks due to the biochemical and biophysical properties of the ECM [1]. Current decellularization methods used to generate ECM scaffolds utilize a combination of chemical and biological agents that disrupt the ECM architecture and surface structure and may leave residual detergents and toxicity behind [2]. Herein, supercritical carbon dioxide was investigated for its decellularization efficacy as a non-toxic and safe alternative.

MATERIALS AND METHODS

Pig liver (1g) was exposed to supercritical carbon dioxide ($scCO_2$) in a 20ml autoclave at ~2900psi; 37°C for varying durations, with or without 24 hours of deionised water (dH_2O) washes. Residual DNA content was used as a metric of decellularization. DNA concentration (ng/ml) was determined using the Quant-iT Pico Green (Invitrogen) assay kit and normalized to dry tissue weight.

RESULTS

A 5 hour exposure to $scCO_2$ (batch-system) reduced liver DNA content compared to controls ($p = 0.014$). Less than 5 hours scCO2 exposure (15mins, 1hr or 2hrs) was insufficient to reduce DNA content. However, short exposures to $scCO_2$ (15mins or 1hr) reduced DNA content when followed by 24 hours agitation in dH_2O ($p < 0.01$).

CONCLUSION

The results herein show that $scCO_2$ has the ability to reduce cellular content, required for effective decellularization. Future studies will investigate other $scCO_2$ conditions to improve the level of DNA reduction.

REFERENCES

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