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Decellularisation barotrauma in rat liver scaffolds compromises scaffold quality and recellularisation capacity

Abstract: Liver transplantation remains the only life-saving treatment for end-stage liver failure, but is limited by organ shortage, and transplanted patients face significant immunosuppression-related pathologies. Bioengineering neo-organs by repopulation of decellularised scaffolds with candidate cells offers the possibility of recipient-specific, immunosuppression-free transplantation. Scaffold preparation requires cell removal whilst minimising damage to extra-cellular matrix (ECM), which provides biophysical/biochemical cellular fate-defining instructions.

Portal vein perfusion with decellularisation solution is an established method of generating liver scaffolds, but flow rate effects on scaffold characteristics are poorly defined. We therefore studied effects of decellularisation flow rates on scaffold properties by light-microscopy, immunofluorescence, residual DNA, glycosaminoglycan and hepatocyte growth factor content, structural ECM proteins, and scaffold recellularisation quality with vascular progenitor stem cells.

We show that decellularisation solution flow rate is a determining factor of scaffold physical and biochemical quality and report the appearance of previously undescribed disruptions within scaffold ECM with 3D structure consistent with false passages distinct from vascular lumina. Compared to sub-physiological flow rates (5mL/min), disruptions were 20-fold more frequent ($p=0.0022$) at physiological and higher flow rates (15 and 30 mL/min). Disruptions resulted in poor decellularisation with 5-fold higher levels of remnant DNA ($p=0.0045$), out-with established quality criteria. Disruptions were also associated with poor scaffold recellularisation, with 2-fold reduction in sinusoidal repopulation ($p=0.0066$), 7-fold imbalance in peripheral to central scaffold cellular engraftment ($p=0.0049$), 3-fold and 4-fold reduction in portal and hepatic venous recellularisation efficiency ($p=0.0001$).

This is the first report demonstrating barotrauma to ECM resulting from flow rate variations, with associated poor decellularisation and recellularisation parameters. These results inform future decellularisation techniques to optimise scaffold repopulation with the objective of regenerative bioengineering.

Keywords: Scaffolds, Biomaterial-cell interaction, Materials structure, Stem cells, Vascular, Perfusion

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Biography: Zeeshan Afzal is a specialty registrar in the run-through General Surgery training program at Addenbrooke's Hospital, University of Cambridge, and a member of the Royal College of Surgeons of England. He completed his medical degree at the University of Leicester, followed by further clinical and surgical training in Cambridge.

He is currently working in the Department of Transplant Surgery at Addenbrooke's Hospital and is pursuing a PhD at the University of Cambridge, focusing on bioengineering liver tissue using bone marrow progenitor cells. Zeeshan aspires to become a consultant in Hepato-Pancreato-Biliary and Transplant Surgery, with a special interest in regenerative medicine.

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