**Tensile Testing of Native and Decellularised Human Aortic and Pulmonary Valve Conduits**

Human cryopreserved aortic (n=12) and pulmonary (n=8) valves were received from NHS Blood and Transplant Tissue Services, Liverpool, UK. Eight aortic and four pulmonary valves remained native and the other four aortic and four pulmonary valves were subject the decellularisation process documented in the manuscript to remove the cellular components. Root wall and leaflet specimens from all the native and decellularised valves were subjected to uniaxial tensile testing to failure using an Instron 3365 (Instron® Corporation) fitted with a 50 N load cell at 10 mm/min under hydrated conditions in phosphate buffered saline solution (PBS) at 37°C. Tissue specimens with 10 mm gauge length and 5 mm width were prepared from the valve root wall in the axial and circumferential directions from each valve, and from the valve leaflet in the circumferential direction. Due to size limitations, the radial specimens from valve leaflets were 3 mm in width with a 6 mm gauge length. Each tissue specimen thickness was measured with a digital thickness gauge J-40 V; having a precision of 0.01 mm.

The measured load was plotted against the extension and maximum load was recorded for all the specimens. Results were represented as mean maximum load ± 95 % confidence limits. An independent sample t-test was employed to compare these values between native and decellularised groups and a p-value of <0.05 was considered to be statistically significant.

The statistical analysis demonstrated no significant difference (P>0.05) in the mean maximum load between the native and decellularised groups for axial and circumferential aortic wall specimens, radial aortic leaflet specimens, and circumferential pulmonary wall and leaflet specimens. However, a significant increase in mean maximum load for the decellularised aortic leaflet specimens in the circumferential direction was observed (P<0.05). Also the mean maximum load for the axial decellularised pulmonary wall specimens and the radial decellularised pulmonary leaflet specimens was significantly reduced. (P<0.05).

 **Results: Tensile Testing of Native and Decellularised Human Aortic Valves**

Figure 1: Load– Extension Graph for the Native (solid line) and Decellularised (dotted line) Axial Aortic Wall Groups

Figure 2: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Circumferential Aortic Wall Groups

Figure 3: Mean Maximum Load for the Native (n=8) and Decellularised (n=4) Aortic Wall Groups – The Error Bars Indicate 95 % Confidence Limits

Figure 4: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Radial Aortic Leaflet Groups

Figure 5: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Circumferential Aortic Leaflet Groups

Figure 6: Mean Maximum Load for the Native (n=8) and Decellularised (n=4) Aortic Leaflet Groups – The Error Bars Indicate 95 % Confidence Limits

\* - Denotes Statistical Significance Difference (P<0.05) Between Native and Decellularised Tissue

**Results: Tensile Testing of Native and Decellularised Human Pulmonary Valves**

Figure 7: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Axial Pulmonary Wall Groups

Figure 8: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Circumferential Pulmonary Wall Groups

Figure 9: Mean Maximum Load for the Native (n=4) and Decellularised (n=4) Pulmonary Wall Groups – The Error Bars Indicate 95 % Confidence Limits

\* - Denotes Statistical Significance Difference (P<0.05) Between Native and Decellularised Tissue

Figure 10: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Radial Pulmonary Leaflet Groups

Figure 11: Load – Extension Graph for the Native (solid line) and Decellularised (dotted line) Circumferential Pulmonary Leaflet Groups

Figure 12: Mean Maximum Load for the Native (n=4) and Decellularised (n=4) Pulmonary Leaflet Groups – The Error Bars Indicate 95 % Confidence Limits

\* - Denotes Statistical Significance Difference (P<0.05) Between Native and Decellularised Tissue