PP-075 MSCs differentiation in decellularized rat and nonhuman primate hearts

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Objectives

To describe results of recellularization of decellularized rat and nonhuman primate hearts with mesenchymal stem cells (MSCs).

Materials and Methods

A shortened detergent-enzymatic protocol using sodium deoxycholate 4% and DNAse was used to decellularize 10 adult male Lewis rat and 3 non-human primate hearts. Recellularization using 90 million autologous MSCs in total was completed via infusion into the aorta of the heart scaffold in equal portions over 5 days. The entire procedure took 18 days. To evaluate the morphology, viability and metabolic activity of the cells seeded on the heart scaffold, MTT testing, live/dead cell staining and IVIS testing were conducted according to manufacturer's instructions.

Results

Histological data obtained after whole organ recellularization demonstrated cell penetration through the basement membrane of blood vessels and were found attached to the inner surface of the heart chambers, coronary vessels. MTT testing showed metabolic activity of the reseeded cells. Live/dead cell staining revealed that a majority (90%) of the reseeded cells remain viable during prolonged cultivation in the scaffold (Calcein positive staining) with approximately 10% dying (Ethidium homodimer I positive staining). *In vivo* bioluminescence imaging also shows the presence of viable cells throughout the entire heart, and concentrated in the coronary vessels. Immunostaining of the reseeded MSCs demonstrated a positive reaction with Ki-67, α-SMA, connexin-43, VEGF and von Willebrand factor antibodies.

Conclusion

Successful recellularization of decellularized rat and nonhuman primate hearts with MSCs demonstrates the nontoxicity of the decellularization protocol to MSCs and its promotion of cell attachment.