06-010

CHARACTERIZATION OF PORCINE HEART VALVE GRAFTS OBTAINED BY TWO DIFFERENT PROCESSES OF DECELLULARIZATION AND IN VITRO BIOCOMPATIBILITY TESTING WITH HUMAN CELLS.

Leitolis, A.(1); Heuschkel, M.A.(2); Roderjan, J.G.(3); Suss, P.H.(4); Da Costa, F.D.A.(4); Stimamiglio, M.A.(5); Correa, A.(5);
(1) UFPR; (2) ICC/Fiocruz; (3) UTFPR; (4) PUCPR; (5) ICC;

Introduction. Heart valves are structures responsible for unidirectional blood flow maintenance into the heart. Changes in valvar functionality may occur after pathological conditions and aging resulting in heart valve disease. The main clinical therapy for this disorder is valve replacement; a procedure performed almost 300.000 times each year. The prosthetic replacement can be performed with biological models (porcine, bovine and human); however those tissues may induce immune responses due to cellular content. Thereby, decellularization is a tissue engineering approach to eliminate allogeneic cells. This method has been tested to improve the biocompatibility of biological prostheses. The present study evaluated the decellularization efficiency of two protocols performed in porcine pulmonary valves and analyzed the in vitro biocompatibility of these grafts. Methods. Porcine pulmonary valves obtained from a slaughterhouse were decellularized with 0.1% SDS solution (Group 1) or 0.1% SDS solution followed by incubation with hypertonic buffer (10 mM Tris-HCl and 2.5 M NaCl) and 20 IU/ml Benzonase (Group 2). The treated heart valves then were subjected to histological, cytotoxicity, DNA and scanning electron microscopic analyses to confirm decellularization and extracellular matrix (ECM) structure. The biocompatibility was verified through the co-culture of porcine tissue with mesenchymal stem cells and valvular interstitial cells (VIC). Results and Discussion. The decellularized tissues showed no histological evidence of cells in the cusps and conduit regions, however, DNA remnants were verified mainly in tissues from group 1. The content and arrangement of elastin, collagen and GAG were not altered in group 1, but a marked reduction in collagen and GAG was observed in group 2. Tissues from both groups were non-cytotoxic. The biocompatibility assays confirmed that cusps from both groups worked as a scaffold for stem cells and VIC. However, cell repopulation into decellularized valve fragments was more effective in scaffolds from group 1. Conclusion. The protocol used in group 2 resulted in a greater decellularization of the porcine heart valves than the protocol of group 1. Nevertheless, protocol #2 was accompanied by ECM impairment and a less effective cell repopulation. Further analyses of immunological responses induced by treated tissues are being conducted to verify which protocol generated a more advantageous graft.