Stem Cell Therapy in Heart Failure: Application Strategies for the CardioCel® Matrix.

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Introduction:

Repair of tissue through provision of a bioscaffold and the addition of cells to repopulate and replace this initial scaffold would be anticipated to offer a superior, long lasting implant that remodels to become native tissue. In addition to using an appropriate scaffold, stem cells are recognised as important in tissue repair and regeneration and are believed to act through mechanisms including the recruitment of cells to the area for repair. However, for cardiac repair, delivery and retention of sufficient cells has proved difficult. Studies have demonstrated that the properties and composition of the scaffold on which stem cells are seeded has a direct effect on the behaviour of the stems cells and the type of new tissue formed.

In the present study we have examined a novel tissue engineering product, CardioCel®, derived from acellular pericardium using a tissue stabilisation technology, ADAPT® (Neethling WML and Hodge AJ, US 2006/0193885 A1), that has the potential to aid tissue repair in the heart, through delivery of adult mesenchymal stem cells in models of heart failure. The CardioCel® biomaterial is fully compatible with the human body. The engineering process also provides the ability to control the porosity and tissue properties enabling design for use as bioscaffolds for the delivery of stem cells. Preliminary studies, as part of animal studies and a Phase II human clinical trial for CardioCel® as a cardiovascular patch have indicated the reduction of calcification post implantation compared to other substrates.

Methods:

Collagen samples: Samples CardioCel® biomaterial were kindly provided by Celxcel (part of the Allied Healthcare Group, Perth, Western Australia). A control sample of glutaraldehyde stabilised pericardium was obtained from a local supplier.

Cell biology: Samples of CardioCel® were seeded with human bone marrow-derived mesenchymal stem cells (MSC) (Millipore) at cell densities from 1 x 10^4 to 1 x 10^5 cells per cm^2 under standard static culture conditions. The MSC were monitored to assess the degree of cell attachment and cell viability using standard cell counting and red/green viability tests after 7 d. Morphology was monitored at 7 d using 5 x 10^4 seeded MSCs and proliferation monitored up to 28 d using 1 x 10^4 seeded MSCs. Quantitation of cell numbers was determined using an MTS assay. Cellular morphology was examined using SEM and phalloidin staining. Collagen synthesis was assessed using 5 x 10^4 seeded MSCs after 70 d by immuno-histology using anti-collagen MAb's (5D8-G9/Col1; 2G8-B1/Col3) and peroxidase detection.

Results:

The MSC seeded samples of CardioCel® showed excellent cell survival at 1 d when compared with a similarly seeded control sample. Similarly, at 7 d there was still excellent MSC viability, whereas the control GA-treated tissue showed limited cell survival (Fig. 1).

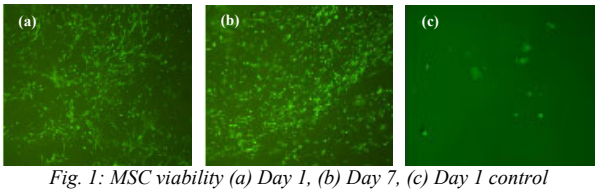

Fig. 1: MSC viability (a) Day 1, (b) Day 7, (c) Day 1 control

The proliferation of MSCs on CardioCel® was followed over 28 d. This showed significant proliferation up to 14 d, which continued at a lower rate up to 28 d (Fig. 2).

Cell morphology was shown after 7 d by SEM. Cells were spreading across the surface of the CardioCel®, while some showed penetration into the collagen matrix (Fig. 3b). Phalloidin also showed cell spreading (Fig. 3b).

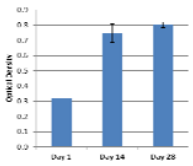

Fig. 2: MSC proliferation

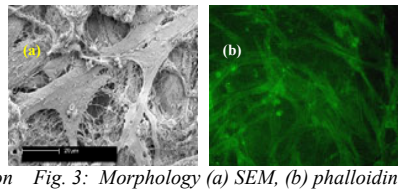

Fig. 3: Morphology (a) SEM, (b) phalloidin

New collagen synthesis associated with the MSC's after 70 d was shown by immunohistology. For example, the new human type III collagen was readily seen, through use of a type and species specific MAb (Fig. 4a). An isotype control showed no staining (Fig. 4b).

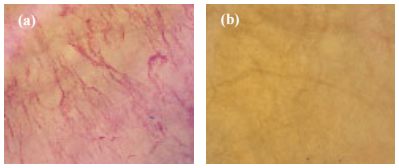

Fig. 4: New collagen production after 70 d MSC proliferation (a) Type III collagen (b) isotype control

Conclusions:

The present data indicate the excellent properties of CardioCel®. It provides an ideal scaffold for delivery of stem cells. The ADAPT® technology shows the potential for medical professionals to use regenerative products instead of synthetic products currently used in soft tissue repair. The present studies have considered the use of CardioCel® as a cardiac patch, but the material also has potential for other biomedical products.

Statement: None of the CSIRO Authors have any financial interest in Allied Healthcare Group, Celxcel or CardioCel®. The University author is an inventor of the ADAPT® process.