ISAKOS

International Society of Arthroscopy, Knee Surgery and Orthopaedic Sports Medicine

9th Biennial ISAKOS Congress • May 12-16, 2013 • Toronto, Canada

Paper #24

Evaluation Of Current Biologic Carriers For Mesenchymal Stem Cell Application In Rotator Cuff Surgery

Knut Beitzel, MD, MA, GERMANY
Mary Beth McCarthy, BS, USA
Ryan P. Russell, MA, USA
Andreas B. Imhoff, MD, Prof, GERMANY
Robert A. Arciero, MD, USA
Augustus D. Mazzocca, MS, MD, USA

Unoversity of Connecticut Health Center
Farmington, Connecticut, USA

Summary:
Differences comparing the various types of ECMs feasible for MSC application existed indicating that surgeons should be aware of the particular properties of each ECM.

Abstract:
Background: Application of human mesenchymal stem cells (MSCs) is considered one of the future key factors for improving the healing environment in rotator cuff repair (RCR). MSC´s potential for self-renewal and ability to differentiate into bone, tendon and cartilage may enhance tendon to bone healing. However a practicable biological carrier (extracellular matrix - ECM) is needed to localize and maintain a high concentration of cells at the site of the repair zone. The purpose of this study was to examine the reaction of human MSCs in culture to currently available scaffold materials compared to native tendon tissue as a control. The hypothesis was that currently available ECMs would be suitable for cell application but demonstrate significant differences in MSC adhesion, proliferation, and viability.

Methods: Human bone marrow aspirated from the proximal humerus was used to isolate and culture MSCs. MSCs were defined by 1) their colony forming potential; 2) their ability to differentiate into tendon, cartilage, bone and fat tissue; and 3) by FACS analysis (CD73+, CD90+, CD105+; CD45-). ECM-Samples (5x5mm2) were taken from fresh frozen human RC tendon, human highly cross-linked collagen membrane (Flexigraft®), porcine non-crosslinked collagen membrane (Mucograft®), a human platelet rich fibrin matrix (PRF-M) and a fibrin matrix based on platelet rich plasma (ViscoGel®). Each sample was soaked for 30 min. in the MSC solution (450x103cells/0.1ml) and then transferred into control media. Cells were analyzed for adhesion (24h); thymidine assay (disintegrations per minute) was obtained to examine cell proliferation (96h) and live/dead stain (calcein-AM/ethidium homodimer-1) was evaluated with confocal-microscopy (Zeiss LSM510) for viability (168h). Histology (H&E) was performed after 21 days and the unloaded scaffolds were scanned with electron microscopy to calculate pore sizes and visualize microstructure. ANOVA & Tukey post hoc tests were performed for statistical analysis.

Results: From cell colonies isolated MSCs were successfully differentiated into bone, tendon, cartilage and adipogenic cell lines. FACS analysis showed MSCs to be over 98.4% positive for the characteristic MSC markers CD73, CD90 and CD105. These cells were also less than 0.05% positive for CD45, indicating the cell population was almost exclusively MSCs. A significantly greater number of cells adhered to both Mucograft® and PRF-M. Cell activity (proliferation) was significantly higher in the Mucograft® compared to PRF-M and Viscogel®. There were no significant differences found in the results of the Live/Dead assay. The morphology of native human rotator cuff tissue was found on average to have a porosity range of 20-30% using a porosimeter. The non-crosslinked collagen ECM was shown to have a more open porous structure with porosity in the 60-70% range.Porosity of the highly crosslinked collagen ECM and fibrin matrices was found to be in the same range as the native rotator cuff tissue

Paper #24

Conclusion: Differences comparing the various types of ECMs existed indicating that surgeons should be aware of the particular properties of each ECM. This allows selection of ECMs based on their specific features (e.g. highly crosslinked for more biomechanical properties vs. non-crosslinked for their biological properties). Results of this study may be utilized as a basis for further animal or clinical studies on the application of mesenchymal stem cells for improved healing in RC-repair.