Effect of Mesenchymal Stem Cells on the Vascularization of the Artificial Cavity Used as a Site for Islet Transplantation

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Target audience

Researcher in the area of cell transplantation

Purpose

At least 50% or more of pancreatic islets (PI) transplanted by portal vein infusion are estimated to be destroyed within the first two days owing to [1]. Therefore, in order to improve transplantation outcome, artificially created sites for islet transplantation have gained attention in the scientific community. The blood supply feeding of those artificial sites (a polymer composite) is higher in case of implantation into greater omentum than subcutaneously. The maximal blood supply of those biocompatible scaffolds was achieved at 1 week after implantation and then it had decreaseds over time [2]. Therefore there is a need to support neoangiogenesis in implanted scaffolds. The support was provided by isogeneic mesenchymal stem cells (MSC) of adipose tissue. The aim of our study was to verify the effect of MSCs on vascularization of the artificially created cavities intended for islet transplantation implanted subcutaneously or into the greater omentum in a preclinical rat model by dynamic contrast-enhanced magnetic resonance imaging (DCE-MRI).

Subjects and Methods

Two polymeric meshes shaped in rounded scaffolds (24x6 mm) were implanted both subcutaneously and into the greater omentum of Brown-Norway female rats (n=6). $15x10^6$ syngeneic MSCs were injected into the scaffolds in 3 animals (another three animals served as controls). MSC were isolated from epididymal fat and cultured in the DMEM medium for 3-4 weeks. MSC were characterized by specific antigens (CD90+, CD44+, CD29+ and CD105+) and by differentiation kit (Rat Mesenchymal Stem Cell Functional Identification Kit). Production of VEGF (Vascular Endothelial Growth Factor) by MSCs ($5x10^5$) in culture within 24 hours was measured by Quantikine Rat VEGF Immunoassay. On the day of implantation of the scaffolds as well as 1, 3, and 4 weeks later, the anaesthetized animals (1.5% Isoflurane) were scanned at a 4.7 T MR scanner equipped with a resonator coil. To assess the actual blood supply at the transplantation site, we analyzed the changes of the signal intensity observed within the scaffolds after the intravascular administration of a contrast agent Gadofosveset (0.05 ml/100 g) by DCE-MRI. For the DCE-MRI, a 3-dimensional gradient echo sequence was used with 32 repetitions (evolution delay = 5.0 s) and resolution = 0.243x0.469x0.422 mm^3. After the 10^{th} cycle, the contrast agent (60 µl) was injected into the lateral tail vein. The regions of interest were manually outlined and mean pixel intensity within each target structure (omental device, subcutaneous device, and kidney) was assessed for every cycle. The average signal intensity during the first 10 cycles was considered as a basal level. The average signal intensity during the last 10 cycles (plateau phase) was considered as a contrast-enhanced level. To minimize the influence of variability in contrast agent application, all outcomes measured in the implanted devices were normalized to the signal intensity of kidney.

Results

The type of cells used for the experiment was confirmed by detection of specific surface markers by flow cytometry and the ability to differentiate into adipocytes, osteocytes and chondrocytes The significant production of VEGF by our MSCs was also confirmed.

The implanted polymeric devices induced no adverse effects and did not cause any image artifacts. One week after the implantation, the connective tissue adequately penetrated and covered the porous devices at all locations. The penetration of the contrast agent was detected by an increase in signal intensity within the implanted devices. On day of implantation, no signal enhancement was detected in any device. However, over the following weeks, a signal increase in the omental device without MSC was detected (to 34% of the signal of the kidney week 1, 21% week 3, and 14% week 4). Within the subcutaneously implanted devices without MSC a signal increase of 11% (week 1), 10% (week 3), and 7% (week 4) of that detected in the kidney was detected. With MSC use, the signal intensity was higher in scaffolds both in great omentum (week 1: 42%, week 3: 41%, and week 4: 64%) and in subcutaneous (week 1: 23%, week 3: 54%, and week 4: 52%). Results are summarized in figure 1.

Discussion

A blood supply in an artificial site is crucial for PI graft survival. MSCs isolated from adipose tissue demonstrated the considerable supportive effect of neoangiogenesis within polymeric scaffolds. Higher blood supply was verified by DCE-MRI. This method could also indicate the optimal time for islet implantation (in case of MSC use: week 4).

Conclusions

Our results indicate that the artificial device containing the MSCs provides a better blood supply for the transplanted cells and a longer optimal time for PI transplantation.

References

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2. Kriz J, Jirak D, Koblas T, et al: Detection of pancreatic islet allograft impairment in advance of functional failure using magnetic resonance imaging. Transpl Int. 25:250-60, 2011

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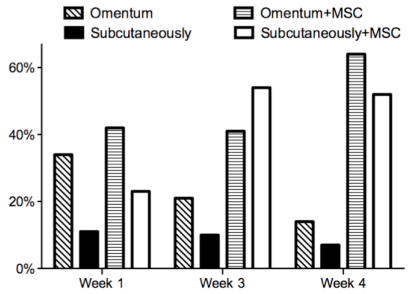

Figure 1: The mean increase of intensity after the contrast agent injection in a polymer composite implanted subcutaneously or into great omentum with/without MSC.