Cellular imaging of rat aortic endothelial cell monolayer using a 3T whole body scanner

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Introduction

Small calibre vascular grafts are seeded with a monolayer of endothelial cells to improve the biocompatibility and patency of the graft. The acceptance of the seeded endothelial cells is influenced by diverse factors like, attachment to the vascular matrix, shear stress by the blood flow, inflammation processes and immune responses, especially if non-autologous endothelial cells are used. The aim of this study is to test the feasibility of cellular MRI for monitoring the monolayer of seeded endothelial cells to get information about their survival under *in vivo* conditions.

Methods

Isolated rat aortic endothelial cells were labeled with small iron oxide particles (VSOP C200, Ferropharm GmbH, Teltow) at a concentration of 1,5 mM for 18 h at 37°C as described elsewhere for other cell types [1]. After several washing steps the labeled as well as unlabeled control endothelial cells were seeded onto the luminal side of vascular grafts which were trypsin treated for decellularization. VSOP uptake could be detected either by luminal surface staining or as well in cryostat sections of the vascular grafts by Prussian blue staining. Seeded vascular grafts were transplanted in a rat interposition model into the region of the abdominal aorta.
All measurements with aortic grafts *in vitro* and *in vivo* were performed on a 3T whole body scanner (MEDSPEC30/100, BRUKER) utilizing a CSA [2] as planar transmit/receive coil. A 3D gradient echo sequence (TE = 8 ms) was used to image a 256x128x128 matrix with 500 µm isotropic voxel resolution. The overall scan time was about 6 minutes. The anesthetized rats where imaged in supine position.

Results

Labelling monolayer endothelial cells with VSOP's resulted in a considerable enhancement of MR contrast compared to unlabeled cells as shown in Fig.1. Subsequent histological examination confirmed that iron labeled cells form a monolayer on the luminal surface of the vascular graft as shown by the strong staining of the luminal graft surface (s. Fig. 2). Furthermore, in grafts seeded with unlabeled endothelial cells no iron staining could be detected (s. Fig. 2). After MR imaging the aortic grafts are still suitable for further transplantation. In a first attempt we transplanted a synthetic graft seeded with VSOP labeled endothelial cells in a rat. The corresponding *in vivo* MR images of the abdominal region exhibit sufficient contrast for follow up monitoring of cell survival.

Conclusion

Labeling of endothelial cells with VSOP's is sufficient to detect a monolayer of cells on the luminal side of a vascular graft using a 3T whole body MR scanner. Hence, the described method is suitable to monitor the fate of transplanted endothelial cells as part of a vascular small diameter graft.

References:

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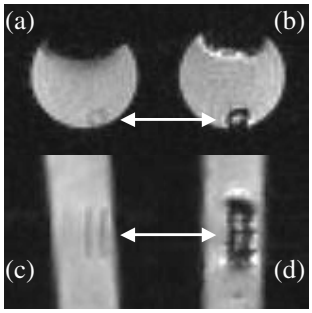

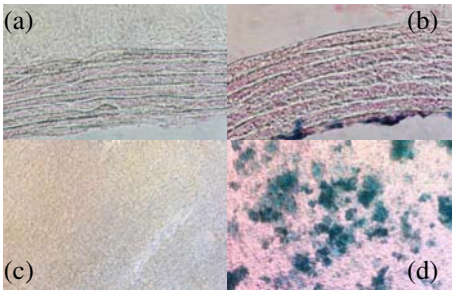

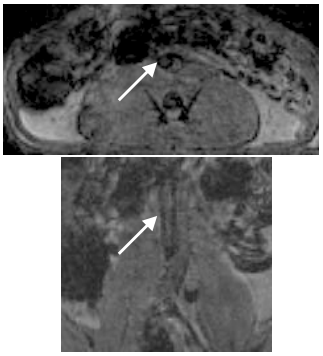

Fig.1 (left): Images of rat vascular grafts within tubes with culture medium derived from 3D MR data set. (a) non labeled endothelial cells, axial view. (b) VSOP labeled cells, axial view. (c) non labeled cells, coronal view. (d) VSOP labeled cells, coronal view.
Fig.2 (middle): Cryostat sections (a), (b) (100x) or luminal surfaces (c), (d) (200x) of decellularized vascular aortic grafts seeded with unlabeled (a), (c) or VSOP labeled (b), (d) rat aortic endothelial cells *in vitro* stained with Prussian Blue for detection of iron. Areas of Prussian blue-positive endothelial cells were seen on the inner surface of the grafts.
Fig. 3 (right): *in vivo* images of a synthetic vascular graft seeded with VSOP labeled rat aortic endothelial cells, axial (top) and coronal (bottom) views were derived from a 3D MR data set with 500 µm isotropic resolution.