Early Detection of Transplant Rejection by *In Vivo* ^{19}F MRI

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Introduction

Diagnosis of transplant rejection requires endomyocardial biopsy and entails risks. Here, we validated a ^{19}F MRI approach for the early detection of organ rejection by noninvasive visualization of the macrophage host response. We employed biochemically inert emulsified perfluorocarbons (PFCs), which were reported to be preferentially phagocytized by monocytes and macrophages[1,2]. Isografts from C57BL/6 and allografts from C57BL/10A mice were heterotopically transplanted into C57BL/6 recipients. In this model, the transplanted heart undergoes predictable progressive rejection, leading to graft failure after two weeks[3].

Methods

Experiments were performed at a vertical 9.4 T Bruker DRX Wide Bore NMR spectrometer operating at frequencies of 400.13 MHz for ^{1}H and 376.46 MHz for ^{19}F measurements using a Bruker Microimaging unit (Mini 0.5) equipped with an actively shielded 57-mm gradient set (200 mT/m maximum gradient strength, 110 µs rise time at 100% gradient switching) and a ^{1}H/^{19}F 30-mm birdcage resonator. Heterotopic abdominal heart transplantation was carried out as previously described[4]. At various times after surgery 500 µl of a perfluoro-15-crown-5 ether (15C5) emulsion (10%, particle size 130 nm) was injected into the tail vene of the mice. In order to monitor the time course of PFC accumulation within the grafted heart, anatomically corresponding ^{1}H and ^{19}F MR images were acquired with the following parameters: FOV 3x3 cm^2, ^{1}H: ECG- and respiratory-triggered FLASH, slice thickness 1 mm, matrix 256x256, acquisition time 1 min, ^{19}F: RARE (RARE factor 64), slice thickness 2 mm, matrix 128x128, acquisition time 20 min. Image acquisition was triggered to cardiac motion of the donor heart by recording its ECG via the hind paws.

Results and Discussion

To exclude that inflammatory processes by the surgical intervention may mask the immune reaction induced by rejection, we first compared genetically identical isografts with allografts. As can be seen from Fig. 1A, six days post transplantation the ^{1}H images show a similar position and anatomy of the graft in the abdomen for both cases. However, the corresponding ^{19}F images matched a signal pattern in shape of the ventricular walls only in the allograft (Fig. 1A bottom). Merging of these images confirms the localization of PFCs within the left and right ventricle. In contrast, isografts – where no rejection occurs – showed ^{19}F signals only at the site of vessel anastomosis (Fig. 1A top). ^{1}H/^{19}F MRI enabled us to detect the initial immune response not later than 3 days after surgery (Fig. 1B), when conventional parameters did not reveil any signs of rejection (Fig. 2C). In allografts, the observed ^{19}F signal strongly increased with time correlating with the extent of rejection. In separate experiments, rapamycin was used to demonstrate the ability of ^{19}F MRI to monitor immunosuppressive therapy. Histologic analysis of the number of CD11b-positive (as marker for monocytes/macrophages) cells found in the myocardium of the graft confirmed that ^{19}F signals correlate with the quantity of infiltrating immune cells. In conclusion, PFCs can serve as positive contrast agent for the early sensitive detection of transplant rejection by MRI, permitting high spatial resolution and an excellent degree of specificity due to lack of any ^{19}F background.

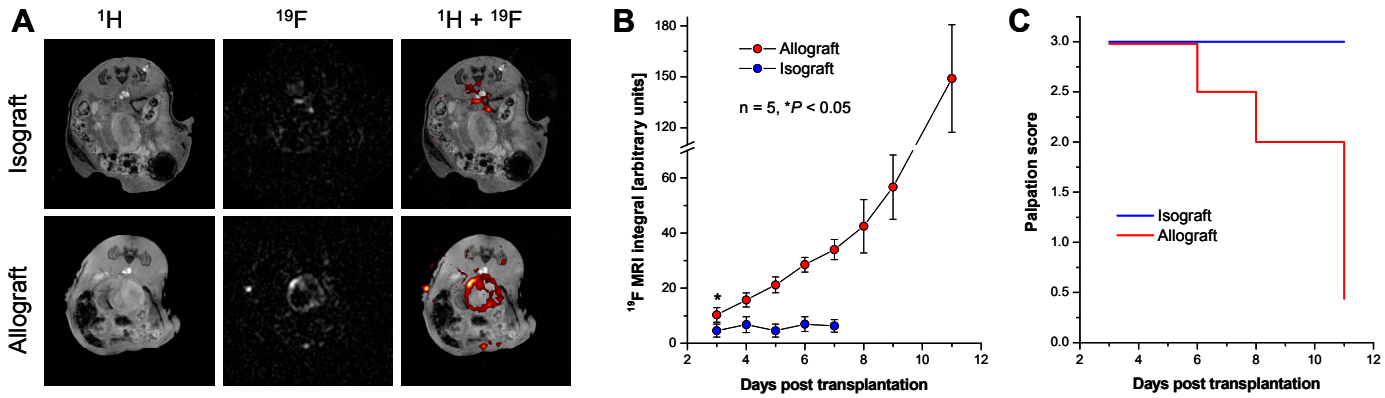

Fig. 1: (A) Anatomical matching ^{1}H and ^{19}F MR images acquired 6 days after transplantation of an iso- and allograft, respectively. For merging of data sets the hot iron colour lookup table was applied to ^{19}F images. (B) Temporal development of detected ^{19}F signals in iso- and allgrafts, which precede conventional parameters of rejection (C, palpation score).

References

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