Imaging of Macrophage Infiltration in an Abdominal Aortic Aneurysm Mouse Model

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Introduction

Ultrasmall Superparamagnetic Iron Oxide (USPIO) imaging contrast agents have been used for labeling atherosclerotic plaque in both human and animal models and acts as an imaging surrogate for macrophage load or activity [1, 2]. Another vascular inflammatory process involving macrophage recruitment is aneurysm formation in the abdominal aorta. Abdominal aortic aneurysms (AAA) can be consistently produced in hyperlipidimic ApoE$^{-/-}$ mice by continuous infusion of Angiotensin-II (Ang-II) [3]. MRI allows for AAA visualization *in-vivo* with sufficient spatial resolution to assess changes in aneurysm size and morphology. In this study USPIO was administered to mice with AAAs to evaluate the ability of MRI to image macrophage distribution in this disease model.

Methods

Osmotic pumps were implanted subcutaneously in ApoE$^{-/-}$ mice for continuous infusion of Ang-II (1000 ng/kg/min). Weekly bright-blood gradient echo scans were performed using a 9.4T MRI scanner to evaluate aneurysm development (TR=15 ms, TE=2.7 ms, 256X256 matrix, 0.12X0.12X1 mm voxels, 10 slices, NEX=8). Once an AAA was detected, animals were given either one or two (spaced 24-hours apart) 1000 µmol/kg doses of the USPIO contrast agent Combidex®. Mice were sacrificed 24-hours post-USPIO administration and the aortas were harvested for ex-vivo imaging (Spin-echo, TR/TE=300/10 ms, 256X256 matrix, FOV=1cm, 10 slices, NEX=4), histology (Cat-S for macrophage, Perls for iron) and iron analysis. To determine area of iron-induced signal reduction, signal intensity was thresholded at two standard deviations below the average intensity of ex-vivo images of a group of 8 AAAs from non-USPIO dosed mice.

Results

Reduced signal intensity was evident in the post-USPIO images (Figure 1). The areas of reduced signal were primarily along the aneurysm shoulder and outer boundaries and corresponded to regions of macrophage infiltration and iron staining detected from immunohistochemistry. Absolute iron content measured as a surrogate for USPIO uptake was only significantly elevated in aortas with aneurysms receiving USPIO (Figure 2A). In vessels without aneurysm formation macrophage infiltration was minimal as was USPIO deposition. The measured iron content in aorta was significantly correlated to the area of signal reduction in the ex vivo images (Figure 2B, r = 0.9, P<0.01). Combined these results indicate that measures of USPIO-induced signal decrease can be used as a marker of macrophage distribution within the remodeled adventitia of this mouse model of aortic aneurysms.

Conclusion

These results demonstrate the feasibility of using a USPIO contrast agent as a surrogate for acute inflammatory processes in development of abdominal aneurysms.

References

1. Kooi et al., *Circulation* 107:2453, 2003
2. Morris *et al., Proc ISMRM, Berlin*, 2007
3. Daugherty *et al., J Clin Invest*, 105:1605, 2000

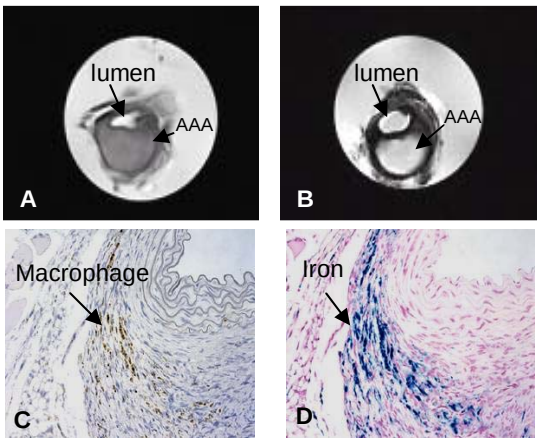

Figure 1 Ex-vivo MRI of abdominal aortic aneurysm with (A) and without (B) USPIO. Histology reveals corresponding areas of (C) macrophage and iron (D).

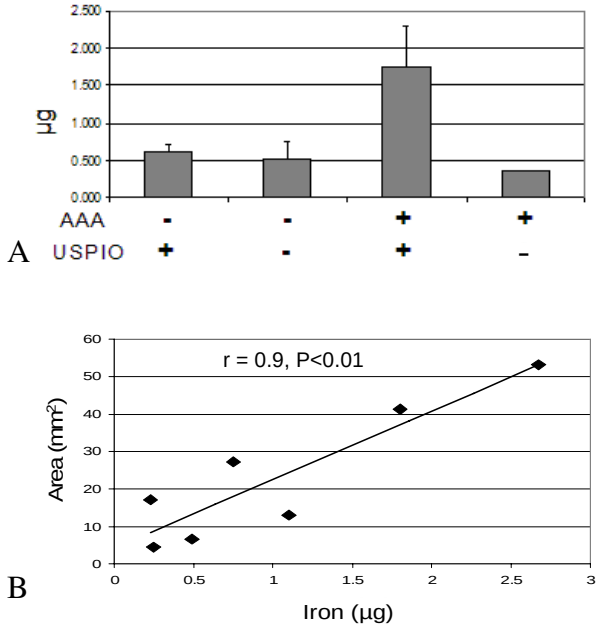

Figure 2 A) Absolute iron content aortas from Ang-II infused apoE mice. B) Area of signal reduction in MRI images is significantly correlated to absolute iron content.