Imaging of Atherosclerosis In Vivo Using a Magnetic Resonance Contrast Probe Molecularly Targeted to Matrix Metalloproteinases (MMPs).

S. Amirbekian[1], J. S. Aguinaldo[2], V. Amirbekian[3], M. Sirol[2], F. Hyafil[2], E. Vucic[2], V. Mani[2], E. Lancelot[4], C. Corot[4], Z. A. Fayad[5]

[1]Imaging Science Laboratories - Depts. of Radiology & Cardiology, Emory University School of Medicine - Mount Sinai School of Medicine, New York, New York, United States, [2]Imaging Science Laboratories, Mount Sinai School of Medicine, New York, New York, United States, [3]Imaging Science Laboratories - Depts. of Radiology & Cardiology, Johns Hopkins University School of Medicine - Mount Sinai School of Medicine, New York, New York, United States, [4]Guerbet, Paris, France, [5]Director - Imaging Science Laboratories - Depts. of Radiology & Cardiology, Mount Sinai School of Medicine, New York, New York, United States

Introduction and Purpose: Matrix Metalloproteinaes (MMPs) play an important role in the pathogenesis of atherosclerosis and vascular remodeling. Most notably, over-expression of MMPs is associated with plaque rupture and instability. MMPs are often highly expressed at the shoulder regions of plaques that rupture. P947 (Guerbet) is a short peptide (5 residues) ligand for MMPs. P947 has a DOTA-Gadolinium chelate attached to it, making it into an MRI contrast agent. Here we investigated the in-vivo efficacy of P947 in detecting atherosclerotic plaque using ApoE KO mice.

Methods: 15-month-old ApoE KO mice (n=14) underwent in vivo MRI of the abdominal aorta using a 9.4T MRI system. Pre-contrast-enhanced (CE) and post-CE MRI was performed at 1, 2, 3, and 22 hours post injection using a T1W black blood sequence. P947 (100 umol/kg) was injected via the tail vein. As a powerful control (n=4), we used the peptide contrast agent P1135, which is P947 with the peptide sequence completely scrambled. By scrambling the peptide sequence specificity for MMPs is lost. As a second control, another group of ApoE KO mice (n=5) were injected with the nontargeted Gd-chelate Gd-DOTA (Dotarem, Guerbet). After MRI, the aortas were isolated, fixed and immunohistochemistry was performed for MMPs. MMP Zymography was performed to show expression of MMPs in imaged sections.

Results: In ApoE mice that were injected with P947 there was heterogeneous enhancement seen on MRI, with a significant increase in the contrast-to-noise ratio (CNR) post-contrast-enhanced images (1, 2 and 3 hrs post-injection). Please See Figure 1. In the Gd-DOTA control group, there was no significant enhancement of the wall. The ratio of the post to the pre contrast signal intensity of the wall, normalized to an externally placed standard, with P947 was 2.25 ± 0.19 (125% enhancement) in ApoE-KO mice at 1 hr, 1.74 ± 0.14(74%) at 2 hrs, 1.31 ± 0.13(31%) at 3 hrs, and 1.18 ± 0.06 (18%) at 24 hrs. The ratio of the post to the pre contrast signal intensity of the wall, normalized to muscle, with P947 was 1.91 ± 0.19(91% enhancement) in the ApoE-KO mice at 1 hr, 1.52 ± 0.14(52%) at 2 hrs, 1.22 ± 0.13(22%) at 3 hrs, and 1.14 ± 0.06 (14%) at 24 hrs. Using P1135 (P947 scrambled) we saw enhancements of only 1.31 ± 0.14(31% enhancement) in the ApoE-KO mice at 1 hr, 1.19 ± 0.11(19%) at 2 hrs, 1.09 ± 0.1(9%) at 3 hrs, and 1.07 ± 0.05 (7%) at 24 hrs. For a summary of the results see the Chart Below. MMP Zymography demonstrated significant MMP activity in imaged sections (controls were negative). Please see Figure 1.

Conclusions: Targeting MMPs with P947 showed highly significant MRI enhancement of aortic atherosclerotic plaque in Apo E KO mice. P947 may be useful in in-vivo noninvasive detection of atherosclerosis and assessment of plaque stability using MRI.

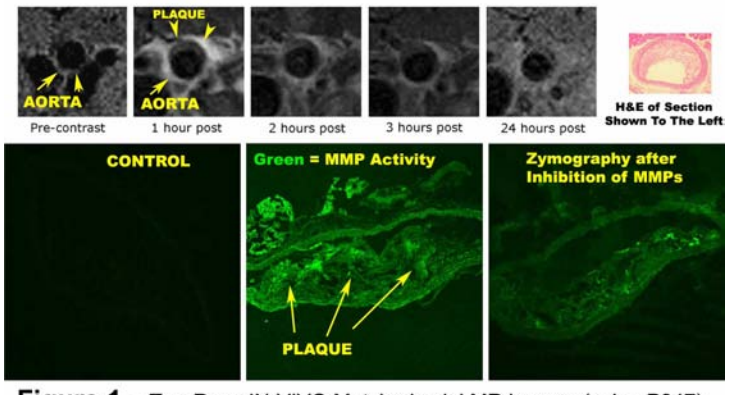

Figure 1 – ***Top Row***: IN-VIVO Matched axial MR images (using P947) corresponding to the H&E section shown of the abdominal aorta of an ApoE KO mouse. ***Bottom Row:*** MMP Zymography – Left Panel is a control MMP Blank Section; Middle Panel is MMP targeted Zymography; Right Panel is MMP targeted Zymography following inhibition of MMPs with EDTA.