Functional Cardiac Phenotyping of Vasoactive Intestinal Peptide (VIP) deficient mice by MR Microscopy

S. D. Smith[1], A. Szema[2], S. I. Said[2], and H. Benveniste[1,3]

[1]Medical, Brookhaven National Laboratory, Upton, NY, United States, [2]Medicine, Stony Brook University, Stony Brook, NY, United States, [3]Anesthesiology, Stony Brook University, Stony Brook, NY, United States

BACKGROUND: Humans with idiopathic pulmonary artery hypertension lack vasoactive intestinal peptide (VIP) immunoreactivity in pulmonary arteries and benefit from nebulized VIP therapy (1). Mice with deletion of the gene for VIP exhibit spontaneous pulmonary arterial hypertension (PAH) and vessel remodeling in the absence of hypoxemia with pathology showing thickened pulmonary vessels (2). We hypothesized that VIP deficient mice with known spontaneous PAH would also develop a decreased functional capacity of their right ventricle. We tested this hypothesis using MR microscopy to evaluate right and left heart ventricular volumes and corresponding ejection fractions in VIP deficient and wildtype (WT) control mice.

METHODS: A total of twelve male mice were used for the studies; Group 1= Control, C57BL6/J male mice (n=6) and Group 2 = vasoactive intestinal peptide deficient ($VIP^{-/-}$) male mice (n=6). For MR imaging all animals were initially anesthetized with isoflurane and breathing spontaneously. The electrocardiogram (ECG), respiratory rate, heart rate and body temperature was continuously monitored (SA Instruments). MR imaging was performed on a superconducting 9.4T/210 horizontal bore magnet (Magnex) controlled by an ADVANCE console (Bruker). Short axis bright blood views of the heart were obtained with an ECG gated multi-slice 2D-FLASH gradient echo sequence: TR=7.5ms, TE=2.9ms, Flip angle=10^{o}, in-plane-resolution: 0.133x0.133 mm^2, slice thickness: 1mm; interslice gap=0.5mm. The heart rate and consequent R-R interval typically allowed for acquisition of 12 cine frames within each cardiac cycle with the given temporal resolution of 7.2 ms. Image analysis was performed using the Segment (http://segment.heiberg.se). For all studies, end-systole was referred to as the frame with minimal ventricular cavity volume. Right ventricular (RV) and left ventricular (LV) volume measurements extracted from three consecutive short-axis slices.

RESULTS: Three-dimensionally the RV geometry is complex in comparison to the LV which is of a straighter cylinder configuration (Fig. 1). The three mid-ventricular short-axis slices do not cover the full extent of the RV and the RV volumes are therefore somewhat underestimated compared to the LV using this approach.

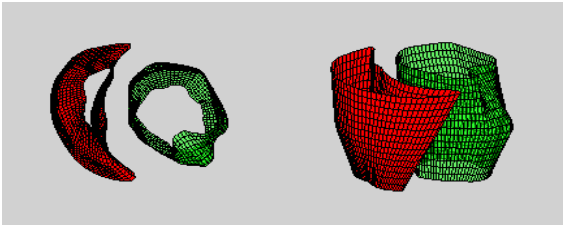

Figure 1: Three-dimensional reconstruction of RV (red) and LV (green) in mid-diastole based on the three short-axis slices using Segment software (ref). Top- and side-views are presented and clearly illustrates the complexity of the RV geometry in comparison to the LV.

Table 1: EDV=enddiastolic volume; ESV=endsystolic volume

Mouse Genotype	EDV (µl)		ESV (µl)		EF (%)	
	LV	RV	LV	RV	LV	RV
WT (n=6)	47.0±7.0	23.6±2.6	15.9±3.7	8.8±3.0	65.7±8.7	63.1±12.4
VIP-/- (n=6)	43.9±7.4	32.0±11.2	16.6±4.4	19.3±10.8**	62.5±4.7	41.9±12.4*

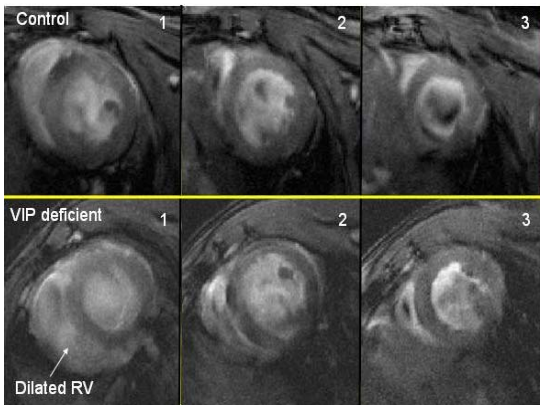

Table 1 shows average EDV, ESV and ejection fractions (EF) of the LV and RV of control and $VIP^{-/-}$ mice and demonstrate that the ESV of $VIP^{-/-}$ mice was 55% larger than that of control mice. In addition the EF of the RV of the $VIP^{-/-}$ mice was also reduced in comparison to control mice. **Figure 2** shows the enlarged end-systolic RV volume in a $VIP^{-/-}$ mouse. Fig. 2 displays two series of MR microscopy short axis cardiac images at the time of end-diastole from a WT control mouse (upper panel) and a $VIP^{-/-}$ mouse (lower panel) and there is clear visual evidence of a dilated RV in the $VIP^{-/-}$ mouse.

CONCLUSION: Our preliminary data demonstrates that $VIP^{-/-}$ mice has RV disease phenotypically expressed as a RV end-systolic volume increase and a reduced RV EF whereas the LV is unaffected when compared to WT controls. Studies are underway to characterize the development of RV pathology in $VIP^{-/-}$ mice over time and to examine the effect of VIP therapy on the RV functional capacity in VIP deficient mice.

References: (1) Petkov et al., JCI, 111:1339-1346;2003; (2) Said et al., Circulation, 115:1260-8; 2007

Acknowledgments: Department of Energy Office of Science and Biological Research (Contract No. DE-AC02-98CHI-886) and the New York State Office of Science, Technology and Academic Research.