Alterations in Myofiber Architecture in Response to Left Ventricular Pressure Overload are Associated with the Upregulation of Genes Encoding for Cell Adhesion and Matrix Remodeling

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Target Audience: Scientists/clinicians interested in MRI of the Heart and Diffusion Tensor MRI (DTI).

Purpose: Pressure overload causes the left ventricle (LV) to undergo compensatory hypertrophy in order to normalize its wall tension. The hypertrophic response, however, becomes maladaptive and leads to a reduction in LV compliance and ultimately to heart failure. We hypothesized that left ventricular hypertrophy (LVH) would be associated with pathological changes in myofiber architecture. We further hypothesized that these changes would be associated with the induction of genes responsible for cell-cell and cell-matrix adhesion in the myocardium.

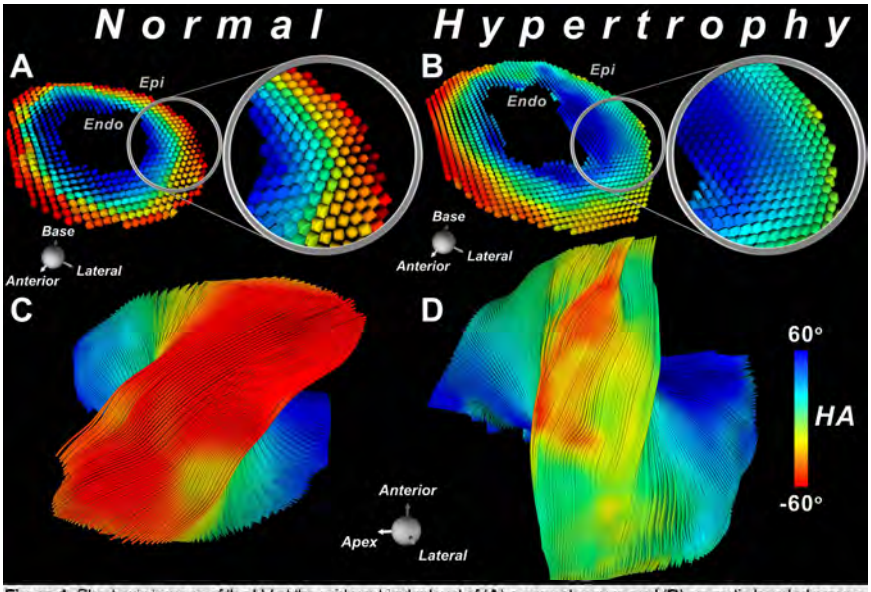

Figure 1: Short axis images of the LV at the mid-ventricular level of (**A**) a normal mouse and (**B**) an aortic-banded mouse. The supertoroidal glyphs are color-coded by myofiber helix angle (HA). Myofibers in the LV free wall have undergone a rightward shift. This is most striking in the subepicardium, where the fibers in the aortic-banded mouse have transitioned from red to green. Tractography of myofibers in the LV free wall in (**C**) a normal mouse and (**D**) an aortic-banded mouse. The rightward (positive) shift in myofiber HA is clearly demonstrated.

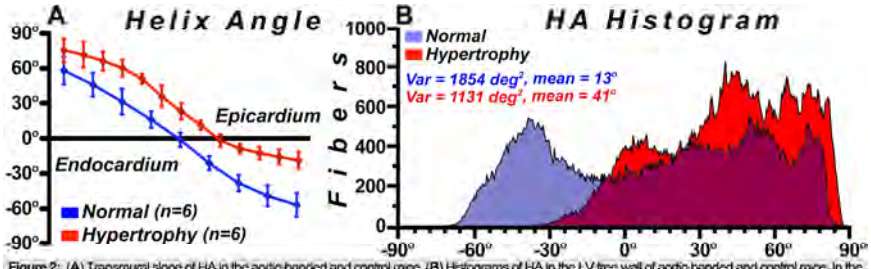

Figure 2: (**A**) Transmural slope of HA in the aortic-banded and control mice. (**B**) Histograms of HA in the LV free wall of aortic-banded and control mice. In the LV free wall of the aortic-banded mouse, circumferential fibers and those with a positive HA predominate, reflecting a rightward shift.

Methods: DTI was performed in a mouse model of LVH (aortic-banded mice, n=6) and in healthy controls (n=6) on a horizontal bore 9.4T scanner (Bruker) equipped with a 1500 mT/m gradient insert. The banded mice were imaged 4 weeks after thoracic aortic banding and the presence of LVH was confirmed by cine MRI. A motion-compensated version of the Stejskal-Tanner sequence was used and imaging was performed in mid-systole with a fat-suppressed spin echo EPI sequence. Key parameters included: matrix 70x70x28 interpolated to 128x128x51, isotropic resolution 156 μm, b-value of 580 s/mm^2, 24 diffusion encoding directions, one b=0 s/mm^2 image, and 2 averages.[1] DTI of the heart was performed *in vivo* with the LV, therefore, exposed to physiological loading conditions. Gene expression analysis was performed in a comparable group of aortic-banded mice (n=5) one week after banding and in age-matched controls (n=5). Analysis of 31,556 genes was performed with the Affymetrix mouse array. Principal component analysis and noise filtering were used to improve data accuracy.

Results: Mice with LVH due to aortic banding consistently showed a rightward (positive) shift in myofiber orientation in the free wall of the LV. This can be seen in the short axis images shown in Figure 1A-B, where supertoroidal glyphs have been color-coded by myofiber helix angle (HA). The myofibers in the subepicardium have developed a more circumferential orientation, while those in the subendocardium are more oblique. The rightward shift in fiber architecture in the banded vs. control mice was revealed by tractography, with myofibers in the banded mice being more circumferential (Figure 1C-D). Likewise, the slope of transmural HA (Figure 2A) and HA histograms (Figure 2B) demonstrated a nearly 30-degree positive shift and a reduction in variance. Fiber architecture in the septum remained largely unchanged. Aortic banding was associated with significant differential expression of 890 genes. Gene expression was increased by >2.85-fold in 48 genes. The upregulation of genes with the potential to influence myofiber orientation was prominent (Figure 3). Over 1/3 (18/48) of the genes with marked changes in expression were related to cell-cell adhesion, cell-matrix adhesion, and the structure of the extracellular matrix (ECM).

Gene	FC
THBS4	8.16
NPPA	5.07
MYH7	5.02
LOX	4.92
COL8A1	4.83
FMOD	4.72
MFAP5	4.21
COL12A1	3.80
THBS1	3.74
ACTA1	3.60
TIMP1	3.56
ITGBL1	3.53
MFAP4	3.52

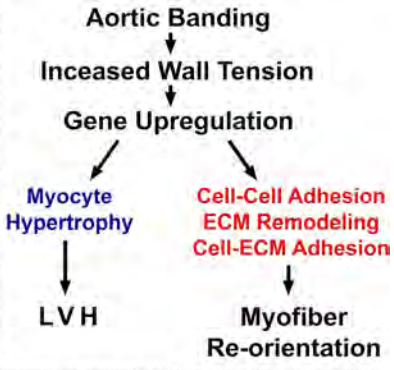

Figure 3: Changes in gene expression in the myocardium of aortic-banded mice. Selected genes are listed in descending order by magnitude of the expression change, and are color-coded by pathway. The upregulation of genes associated with myofiber orientation (red) was more prominent than the upregulation of genes associated with cardiomyocyte hypertrophy (blue). *FC = fold change, ECM = extracellular matrix.*

Discussion: LVH due to pressure overload is associated with a marked rightward shift in myofiber orientation in the free wall of the LV. This shift is accompanied by a pronounced upregulation of genes in the LV encoding for structural changes in the extracellular matrix and cell-matrix adhesion. This suggests that the increased wall tension imposed by pressure overload, particularly in the LV free wall, causes the upregulation of genes that lead to myofiber realignment, resulting in fiber orientation that is more circumferential, and which may impact myocardial mechanics.[2]

Conclusion: Pressure overload of the LV induces a gene program that results in marked changes in myofiber orientation. While initially adaptive, these changes may reduce LV compliance and the efficiency of myocardial contraction, and likely contribute to the subsequent development of heart failure.

References: 1) Sosnovik DE, et al. Circulation 2014; 2) Pluijmert M, et al., AJP-Heart Circ Physiol. 2014.