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Relationships between regional myocardial wall stress and bioenergetics in hearts with left ventricular hypertrophy

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Abstract

This study utilized porcine models of postinfarction LV remodeling (MI: n=8) and concentric LVH secondary to aortic banding (AoB: n=8) to examine the relationships between regional myocardial contractile function (tagged MRI), wall stress (MRI and LV pressure), and bioenergetics (P-31 MR spectroscopy). Physiological assessments were conducted at a 4 week time point after myocardial infarction or aortic banding surgery. Comparisons were made with size matched normal animals (normal: n=8). Both myocardial infarction and aortic banding instigated significant LV hypertrophy. Ejection fraction was not significantly altered in the AoB group, but significantly decreased in the MI group ($p < 0.01$ vs. normal and AoB). Systolic and diastolic wall stresses were approximately two times greater than normal in the infarct region and border zone. Wall stress in the AoB group was not significantly different from normal hearts. The infarct border zone demonstrated profound bioenergetic abnormalities, especially in the subendocardium, where the ratio of phosphocreatine to adenosine triphosphate (PCr/ATP) decreased from 1.98 ± 0.16 (normal) to 1.06 ± 0.30 (MI, $p < 0.01$). The systolic radial thickening fraction and the circumferential shortening fraction in the anterior wall were severely reduced (MI, $p < 0.01$ vs Normal). The radial thickening fraction and circumferential shortening fraction in the AoB group were not significantly different from normal. The severely elevated wall stress in the infarct border zone was associated with a significant increase in chemical energy demand and abnormal myocardial energy metabolism. Such severe metabolic perturbations cannot support normal cardiac function, which may explain the observed regional contractile abnormalities in the infarct border zone.

Keywords

myocardial infarction; LV remodeling; hypertrophy; wall stress; metabolism

INTRODUCTION

Postinfarction left ventricular (LV) remodeling often leads to the evolution of heart failure. However, the mechanisms contributing to the transition from compensated ventricular remodeling to congestive heart failure (CHF) remain unclear. Tethering of the border zone BZ to the infarct causes an increase in the radius of curvature of the surrounding viable myocardium, thereby increasing wall stress and stress associated energy demands in viable myocardium in the infarct (BZ) (7). Using a porcine model of postinfarction LV remodeling, we previously observed significant bioenergetic abnormalities in remote, noninfarcted myocardial regions in animals with overt CHF, while only modest alterations were observed

in the remote myocardial regions of animals undergoing compensated LV remodeling (7). We have recently reported that the abnormal myocardial bioenergetic state, as represented by high-energy phosphate (HEP) content and the phosphocreatine (PCr) to adenosine triphosphate (ATP) ratio, is markedly more severe in the infarct border zone compared to remote myocardial regions in the same hearts (7). We hypothesized that over time, the bioenergetic and contractile abnormalities of the border zone, which are triggered by elevated regional wall stress, may extend laterally and eventually involve the entire left ventricle thereby leading to global LV dysfunction and the development of congestive heart failure. If this hypothesis is correct, then hearts with concentric LVH containing a small chamber cavity and thicker LV wall should have normal regional LV wall stress and relatively less severe abnormalities in myocardial bioenergetics. In the present study, we therefore compared a porcine model of eccentric left ventricular hypertrophy (LVH) secondary to a myocardial infarction to a porcine model of concentric LVH secondary to aortic banding. Myocardial wall stress and contractile function were assessed with cine and tagged MRI; myocardial bioenergetics was assessed with ^{31}P NMR spectroscopy.

METHODS

All experiments were performed in accordance with the animal use guidelines of the University of Minnesota, and the experimental protocol was approved by the University of Minnesota Research Animal Resources Committee. The investigation conformed to the *"Guide for the care and use of laboratory animals"* published by the National Institutes of Health (NIH publication No 85-23, revised 1985).

Induction of Myocardial Infarction

Details of the animal model of postinfarction LV remodeling have been described previously (9,24). Briefly, young Yorkshire swine (45 days old, ~10 kg) were anesthetized with pentobarbital (30 mg/kg iv), intubated, and ventilated with a respirator with supplemental oxygen. Arterial blood gases were maintained within the physiological range by adjustments of the respiratory settings and oxygen flow. A left thoracotomy was performed, and 0.5 cm of the LAD distal to the second diagonal branch was dissected free and totally occluded with a ligature. After coronary ligation, animals were observed in the open-chest state for 60 min. When ventricular fibrillation (VF) occurred, electrical defibrillation was performed immediately. The chest was then closed, but if the heart was dilated, the pericardium was left open. The animals were given standard postoperative care, including analgesia, until they ate normally and resumed full activity. Animals returned to the laboratory 3.5 weeks and 4 weeks later for MRI and ^{31}P -MR spectroscopic studies, respectively.

Induction of Left Ventricular Hypertrophy (LVH) with Aortic Banding

Details of the animal model of left ventricular hypertrophy (LVH) have been described previously (4,20). Briefly, young Yorkshire swine (45 days old, ~10 kg) were anesthetized with pentobarbital (30 mg/kg iv), intubated, and ventilated with a respirator with supplemental oxygen. Arterial blood gases were maintained within the physiological range by adjustments of the respiratory settings and oxygen flow. A right thoracotomy was performed in the third intercostals space, and the ascending aorta (1.5 cm above the aortic valve) was mobilized and encircled with a polyethylene band 2.5 mm in width. While LV and distal aortic pressures were simultaneously measured, the band was tightened until a 70-mmHg peak systolic pressure gradient was achieved across the narrowing. The chest was then closed, pneumothorax was evacuated, and the animals were allowed to recover. LVH occurred progressively as the area of aortic constriction remained fixed in the face of normal body growth. Animals returned to the laboratory 3.5 weeks and 4 weeks later for MRI and ^{31}P -MR spectroscopic studies, respectively.

Tagged- and Cine- MRI Protocol

MRI was performed ~25 days following surgery on a 1.5 Tesla clinical scanner (Siemens Sontata, Siemens Medical Systems, Islen NJ) using a phased-array 4-channel surface coil and ECG gating. Animals were anesthetized with 1% isoflurane and positioned in a supine position within the scanner. The protocol consisted of: 1) localizing scouts to identify the long- and short-axis of the heart, 2) short- and long-axis cine for the measurement of global cardiac function, 3) short-axis imaging with myocardial tagging in 3 slices for the measurement of regional myocardial strain, and 4) delayed contrast-enhancement for the assessment of scar size and position. Steady-state free precession “True-FISP” cine imaging used the following MR parameters: TR = 3.1 ms, TE = 1.6 ms, flip angle = 79°, matrix size = 256 × 120, field of view = 340 mm × 265 mm, slice thickness = 6 mm (4 mm gap between slices) and 16-20 phases were acquired across the cardiac cycle. Global function and regional wall thickness data were computed from the short-axis cine images using MASS (Medis Medical Imaging Systems, Leiden, The Netherlands) for the manual segmentation of the endocardial and epicardial surfaces at both end-diastole (ED) and end-systole (ES) from base to apex.

The tagging preparation consisted of nonselective radiofrequency pulses separated by encoding gradients for spatial modulation of magnetization (SPAMM), resulting in a tag line separation of 6 mm. Three short-axis slices were prescribed at a basal, midventricular, and apical level identical to the cine image positions. At each slice location, two sets of cine images were acquired with tag lines in orthogonal directions with the following scan parameters: TR = 6.5 ms, TE = 2.1 ms, flip angle = 140°, matrix size = 256 × 128, field of view = 320 mm × 320 mm, slice thickness = 6 mm, and a minimum of 14 cardiac phases. Tagged images were acquired for resting conditions only. Tagging data was analyzed using the HARP analysis package (HARP version 2.0, Nael Osman, John Hopkins Medical School) as described elsewhere (14,15). Two-dimensional myocardial strains were assessed offline in 6 circumferential myocardial segments per short-axis slice. Transmural strains were calculated between the reference end-diastolic (ED) and end-systolic (ES) state as the fractional change in length in the circumferential (Ecc) and radial (Err) directions.

Infarct size

At the completion of the open chest MRS and hemodynamic measurements, animals were sacrificed by an overdose of pentobarbital, and the heart explanted. The LV was opened at the lateral wall from base to apex, and a photograph was taken for infarct size measurement. Infarct size was expressed as a percent of LV surface area by an image analysis system (NIH image J program, <http://rsb.info.nih.gov/ij>).

Surgical preparation for open chest MR spectroscopy study

Animals were anesthetized with pentobarbital (loading dose of 30 mg/kg iv, maintenance dose of 4 mg/kg/hr), intubated, and mechanically ventilated with oxygen-supplemented air to maintain arterial blood gases within the physiological range. 3mm outer-diameter polyvinyl chloride (PVC) catheters were inserted into the ascending aorta (Ao), inferior vena cava (IV), and Left Ventricle (LV). Catheters were flushed with heparinized normal saline to prevent clotting, and secured using silk sutures after placement. The Ao catheter was placed via the left internal carotid artery at the level of the cervical vertebrae. Two IV catheters were placed through the left external jugular vein. The heart was exposed via a sternotomy and suspended in a pericardial cradle. The LV catheter was introduced through the apical dimple.

A 28 mm diameter single loop transmit/receive radiofrequency (RF) coil for magnetic resonance spectroscopy was sutured onto the anterior wall of the LV such that most of the coil was directly over the scar and a small portion of the coil was over the infarct border zone. Scar tissue does not contain high energy phosphates, and therefore the entire NMR signal was

obtained from the infarct border zone. To restore the heart to its normal position, the pericardial cradle was released. The animals were placed into a polyethylene-lined Lucite cradle, and the coil leads were soldered to a balanced-tuned circuit external and parallel to the craniocaudal axis. The entire assembly was then positioned within a 4.7 tesla (T) superconducting magnet.

Hemodynamic measurements

Aortic and LV pressures were measured using pressure transducers positioned at mid-chest level and recorded on an 8-channel recorder (11,20,22,23). Ventilation rate, volume and inspired oxygen content were adjusted to maintain physiologic values for arterial PO₂, PCO₂ and pH. Aortic and LV pressures were monitored continuously throughout the study (11,20,22,23). Hemodynamic measurements were acquired simultaneously with the spectra.

Spatially localized ³¹P-MR spectroscopy technique

Spatially localized ³¹P NMR spectroscopy was performed using the RAPP-ISIS/FSW method (rotating-frame experiment using adiabatic plane-rotation pulses for phase modulation (RAPP)-imaging-selected in vivo spectroscopy (ISIS)/Fourier series window (FSW) method) (11,20,22,23). Detailed experiments documenting voxel profiles, voxel volumes and spatial resolution attained by this method have been published previously. In this application of RAPP-ISIS/FSW, the signal origin was first restricted to a 12*12 mm two-dimensional column perpendicular to the LV wall. The signal was later localized into three well resolved and five partially resolved layers along the column and hence, across the LV wall. Localization along the column was based on B1 phase encoding and employed a 9-term Fourier series window as previously described (11,20,22,23). Whole wall spectra were obtained with the image-selected in vivo (ISIS) technique, defining a column 12*12 mm² perpendicular to the heart wall. The calibration of spectroscopic parameters was facilitated by placing a polyethylene capillary filled with 15 µl of 3 M/L phosphonoacetic acid into the inner diameter of the surface coil. This phosphonoacetic acid standard was used only for calculating the 90 degree pulse length of the RAPP-ISIS method (11,20,22,23). The position of the voxels relative to the coil was set according to the B1 strength at the coil center which was experimentally determined in each case by measuring the 90° pulse length for the phosphonoacetic acid standard contained in the reference capillary at the coil center. NMR data acquisition was gated to the cardiac and respiratory cycles using the cardiac cycle as the master clock to drive both the respirator and the spectrometer as previously described (11,20,22,23). The surface coil was constructed from a single turn copper wire 28 mm in diameter with each side of the coil leads soldered to a 33 pF capacitor. Complete transmural data sets were obtained in 10-minute time blocks using a repetition time of 6-7 seconds to allow for full relaxation for ATP and inorganic phosphate (Pi) and approximately 95% relaxation of the PCr resonance (11,20,22,23). The ratios of PCr to ATP (PCr/ATP) were calculated for each transmurally differentiated spectra set as previously described (11,20,22,23). All resonance intensities were quantified using integration routines provided by the SISCO software.

Data analysis

Data was analyzed with one-way analysis of variance for repeated measurements. A value of p<0.05 was considered significant. When significant results were found, individual comparisons were made using the Bonferroni Correction.

RESULTS

Anatomic data

Table 1 summarizes the anatomic data from 8 normal animals, 8 animals with post infarction LV remodeling (MI group), and 8 animals with aortic banding (AoB group). Infarct size in the

MI group was calculated and expressed as the ratio of scar surface area to LV surface area (SSA/LVSA). LAD occlusion produced an infarct on the anterior wall, which encompassed $13 \pm 3\%$ of the LV. Myocardial infarction and aortic banding instigated significant LV hypertrophy, as assessed by the LV weight to body weight ratio ($p < 0.05$, Table 1). The LV weight to body weight ratio increased from 2.6 ± 0.1 g/kg (normal) to 3.3 ± 0.3 g/kg and 3.7 ± 0.2 g/kg in MI and AoB, respectively ($p < 0.05$, Table 1).

MI hearts were characterized by a thin wall and dilated LV, however the total LV mass (determined during post-mortem assessments) of dilated hearts was still greater than normal hearts. These observations are in agreement with earlier reports that disaggregated myocytes from dilated MI hearts were characterized by elongated myocytes with the same or slightly reduced cross-sectional areas (11). Although AoB hearts tended to have a higher LVW/BW ratio, the severity of myocyte hypertrophy was likely similar between the two groups considering the 13% loss of LV mass in the MI hearts.

LV Ejection Fraction and Hemodynamics

LV ejection fraction (EF) was measured from CINE MRI images. After 4 weeks, EF was not significantly decreased from normal in the AoB group (Normal: $55.3 \pm 3.1\%$ vs. AoB: $49.5 \pm 2.1\%$) but was significantly decreased in the MI group (MI: $30.4 \pm 2.3\%$, $p < 0.01$ vs. normal and AoB, Table 2, Figure 1A). Systemic hemodynamics variables were not statistically different between the three groups of animals. LV systolic pressure was observed to be elevated in AoB animals, however this observation was not statistically significant. Importantly, LV end diastolic pressure was not elevated in either group, indicating both groups of animals were still undergoing compensated LV hypertrophy 4 weeks after MI or aortic banding.

MRI Analysis of LV Wall Thickness

LV structure was assessed in 3 horizontal short axis rings from the midventricular level of the LV. Each ring was divided into 6 regions according to the coronary artery perfusion pattern as depicted in Figure 2. Papillary muscle tissue was avoided in wall thickness measurements. Short axis cine MRI analyses revealed that animals in the MI group developed significant diastolic wall thinning in the anterior region (Table 3, Figure 1B; $p < 0.01$ vs. Normal). Systolic wall thickness was decreased throughout the entire LV ring, being most severe in the anterior wall (Table 3, Figure 1C; $p < 0.01$ vs. Normal). Both systolic and diastolic thickness were increased in the AoB group (Table 3, Figure 2; $p < 0.05$ vs. Normal).

MRI Measured LV Function

Regional myocardial contractile function was assessed by calculating the radial thickening fraction and circumferential shortening fraction (tagged MRI) in the 6 different segments from 3 horizontal short axis rings from the midventricular level of the LV. Radial thickening fraction was calculated by the equation $TF\% = (EST - EDT) / EDT \times 100\%$, where EST and EDT correspond to end systolic thickness and end diastolic thickness, respectively. Circumferential shortening fraction was calculated by the equation $SF\% = (l_s - l_d) / l_d \times 100\%$ where l_s and l_d correspond to systolic and diastolic lengths along the circumferential direction.

The analysis revealed that animals in the MI group developed significant contractile dyskinesia in the anterior region of the LV, where the infarct region and border zone were localized. The radial thickening fraction in the anterior wall was reduced by almost 98% from $40.5 \pm 3.1\%$ (normal) to $0.2 \pm 1.8\%$ (MI, $p < 0.01$, Table 4, Figure 1D). The reduction in radial thickening fraction was much less severe in myocardial regions remote from the scar (posterior wall: $36.7 \pm 3.1\%$ (normal) vs. $25.4 \pm 1.5\%$ (MI), $p < 0.05$, Table 4, Figure 1D). Moreover, negative radial thickening fractions were observed in a number of pigs in the MI group, indicating that infarct region and border zone were “bulging” outwards during systole and not contributing to

ventricular contraction. The circumferential shortening fraction was decreased in the anterior wall ($-16.7 \pm 0.8\%$ (normal) to $-5.4 \pm 0.1\%$ (MI, $p < 0.01$, Table 4, Figure 1E) but was not significantly changed in remote myocardial regions in the posterior wall. The radial thickening fraction and circumferential shortening fraction in the AoB group were not significantly different from normal.

Wall Stress

LV Wall stress (σ) in different LV segments was calculated according the Laplace law, as previously described, using the following equation: $\sigma = PR/(2T)$ (16), where P corresponds to LV pressure measured with a pressure transducer in a fluid filled catheter, R corresponds to chamber radius and T corresponds to wall thickness measured from CINE MRI images in the particular region of the LV. Systolic and diastolic wall stresses were approximately two times greater than normal in the infarct region and border zone (systolic wall stress: 157 ± 11 mmHg [Normal] vs. 297 ± 17 mm Hg [MI, $p < 0.01$, Table 5, Figure 1F], diastolic wall stress: 11 ± 1 mm Hg [normal] vs. 25 ± 3 mm Hg [MI, $p < 0.01$], Table 5, Figure 1G). Systolic and diastolic wall stresses in remote myocardial regions along the posterior wall were also elevated, but to a lesser extent. Wall stress was not elevated in the AoB group.

Myocardial Bioenergetics

Figure 3A illustrates typical transmurally differentiated ^{31}P NMR spectra obtained from a normal heart, the infarct border zone of an MI heart, and the anterior wall of an AoB heart. The infarct border zone demonstrated profound bioenergetic abnormalities, especially in the subendocardium, where the ratio of phosphocreatine to adenosine triphosphate (PCr/ATP) decreased from 1.98 ± 0.16 (normal) to 1.06 ± 0.30 (MI, $p < 0.01$, Table 6, Figure 3B). It should be noted that although the PCr/ATP ratio was not measured in remote myocardial regions, in previous experiments using the same animal model, we observed only modest alterations in the PCr/ATP ratio in remote myocardium of animals with compensated post-infarction LV remodeling (7, 24). The subendocardial PCr/ATP ratio was moderately decreased in the AoB group (PCr/ATP: 1.62 ± 0.08), however, the bioenergetic abnormalities were not nearly as severe as in the infarct border zone (Table 6, Figure 3B). Correlations between the regional wall stress and border zone myocardial bioenergetics are depicted in Figure 4. Elevated wall stress was associated with increased regional energy demand and bioenergetic abnormalities (Figure 4, Panels A and B).

Because a mature LV scar is very thin, an obvious question concerns the partial volume effect of the thinned LV scar. Due to the relatively low signal to noise ratio of ^{31}P -MR spectroscopy, high-energy phosphate (HEP) levels in the scar tissue are too low to be detected within the defined sample region of interest (defined by the ISIS method as $17 \times 17 \text{ mm}^2$). Consequently, the HEP signals obtained from the surface coil positioned adjacent to the infarct represent border zone myocardium with essentially no contribution from the scar tissue.

In principle, the deeper voxels (i.e. more distant from the epicardial surface) contain contributions from LV cavity blood because of partial volume effects in which the voxel contains both the LV wall and the LV chamber. This is recognizable by the presence of 2,3-DPG resonances in the ~ 3 ppm region of the spectra (7,21). The presence of both blood and cardiac muscle in the same voxel has the potential to distort ATP levels and PCr/ATP ratios because blood contains ATP but not PCr. The ATP contribution from blood to the endocardial spectrum PCr/ATP has been previously examined and found to be trivial (7,21). In the present study, the contribution of blood in the NMR region of interest might be greater because of the thinner wall in the infarct border zone. To assess this possibility, the blood ATP contribution was examined with a phantom filled with fresh heparinized blood using the identical spectrometer setup. Prominent resonance peaks of 2,3 diphosphoglycerate (2,3 DPG) appeared

at ~ 3 PPM. No ATP resonance was detected, demonstrating that the contribution of LV cavity blood ATP to the endocardial PCr/ATP ratio was negligible.

Correlations between Regional Variables

Correlations between the 3 regional variables (anterior wall stress, border zone myocardial bioenergetics, and anterior wall contractile function) are depicted in Figure 4. Elevated wall stress increases the regional energy demand and results in bioenergetic abnormalities (Figure 4, Panels A and B). Abnormalities in the 3 regional variables were found to be significantly linearly related. The observed relationships between alterations in regional wall stress, abnormalities in myocardial bioenergetics, and the impaired regional contractile function suggest a “vicious cycle” that is initiated by the elevated wall stress in the overstretched cardiomyocytes within the infarct region and border zone.

DISCUSSION

The main finding of the present study is that the severely elevated wall stress in the infarct border zone was associated with a significant increase in chemical energy demand and abnormal myocardial energy metabolism. Such severe metabolic perturbations cannot support normal cardiac function, which may explain the observed contractile abnormalities. In addition, the dysfunctional cardiomyocytes in the infarct border zone are overstretched, which acts to induce strain-mediated apoptotic signaling systems (21,22). These data support the hypothesis that a progressive radial expansion of functional and bioenergetic abnormalities from the infarct border zone to the entire LV contributes to the onset of severe LV dysfunction in hearts with postinfarction LV remodeling and the evolution of congestive heart failure.

Comparison between Concentric and Eccentric LVH

In post-infarction LV remodeling, it is hypothesized that the evolution of heart failure may be related to the progressive expansion of contractile and bioenergetic dysfunction from the region of viable myocardium that surrounds the infarct border zone to the entire left ventricle (7). As a consequence of being mechanically “tethered” to the infarct, the viable myocardial tissue in the border zone increases its radius of curvature, thereby increasing its wall stress and stress-associated energy demands (5,10,18). Since the border zone is also mechanically tethered to myocardial tissue which is more distal to the infarct region, the progressive dysfunction in border zone may adversely affect function of the myocardial tissue to which it is connected leading to a lateral expansion of functional and bioenergetic abnormalities throughout the LV wall.

In the MI group of the present study, permanent LAD ligation caused a $13 \pm 3\%$ infarct on the anterior wall of the LV, resulting in eccentric hypertrophy with chamber dilatation and severe wall thinning. Although this level of myocardial infarction resulted in compensated post-infarction LV remodeling, significant functional and bioenergetic alterations were observed (Tables 4, 5, and 6). Pigs in the MI group developed contractile abnormalities and elevated wall stress throughout the LV. However, the abnormalities were most severe in the infarct region and border zone (Figures 1 and 3). Interestingly, the abnormalities myocardial wall stress and myocardial bioenergetics were found to be related (Figure 4), implying that the detrimental cascade of events may be initiated by the elevated wall stress in overstretched cardiomyocytes within the infarct region and border zone.

This study also utilized a porcine model of concentric hypertrophy for comparison with post-infarction hearts, in order to determine whether the increased BZ wall stress causes regional myocardial bioenergetic abnormalities. Concentric LVH was induced by aortic banding, and resulted in a significant increase in LV wall thickness. In pigs with concentric LVH, the thicker

LV wall acted to normalize LV wall stress (Figure 1). Interestingly, the severity of hypertrophy was similar between the hearts of concentric- and eccentric- LVH (Table 1, LVW/BW), however, the bioenergetic abnormalities which were manifested by the decrease of PCr/ATP ratio, were much more severe in the BZ of the MI hearts (Table 6). This data further supports the concept that regional myocardial wall stress contributed to the myocardial bioenergetic abnormalities in hearts with post infarction LV remodeling (Figure 4). These data also imply that different mechanisms contribute to the transition from compensated LVH to CHF in hearts with eccentric LVH than hearts with concentric LVH.

It should be noted that the correlation among bioenergetics with these estimates of wall stress and direct measures of contractile function do not prove that impaired energetics causes regional dysfunction. There are likely other differences among these eccentric and concentric LVH models that can also affect contractility, including calcium handling, myocardial blood flow, and molecular remodeling.

Post-Infarction LV Remodeling and Border Zone Contractile Dysfunction

Post-infarction LV remodeling often leads to a dilated LV even if the coronary occlusions are treated (2). After an acute myocardial infarction (AMI), the entire LV is exposed to significantly increased wall stresses and chamber dilatation that is most severe in BZ. Cardiomyocyte hypertrophy begins shortly after myocardial infarction to restore normal LV wall stress and cardiac function. However, despite initial compensation, the underlying loss of contractile mass in the infarct region causes a mechanical overstretch on surviving cardiomyocytes in the border zone (13). Overstretch of cardiomyocytes is coupled to increased reactive oxygen species (ROS) production, increased oxidative stress, further chamber dilatation mediated by the rennin-angiotensin system, overexpression of the apoptosis mediating Fas molecule, cardiomyocyte apoptosis, and impairment in force development (1,3). This combination of adverse events consequently worsens the contractile function in the border zone and may instigate the progression to heart failure.

In a study using an ovine myocardial infarction model and echo contrast imaging, Jackson et al. observed that the radial displacement of the infarct region during systole forced the adjacent myocardium to curve outward (8). This resulted in a decreased local radius of curvature and more than doubled computed systolic wall stress in the infarct border zone (8). The elevated wall stress in the infarct border zone of MI pigs in the present study mirrors these findings (Table 5, Figure 2). Because the viable myocardial tissue in border zone is mechanically tethered to the dysfunctional infarct region, early increases in BZ wall stress that result from geometric changes in this region of the myocardium have been implicated as an important mechanism for inciting the development of adverse ventricular remodeling and the subsequent development of congestive heart failure (5,8).

Myocardial Bioenergetics

In the present study, the bioenergetic efficiency in the infarct border zone was assessed by the PCr/ATP ratio. The PCr/ATP ratio reflects the mitochondrial oxidative phosphorylation (mtOXPHOS) regulation, myocardial energy efficiency (6), and extent of LV dysfunction (12,22). Because the creatine kinase reaction is nearly in equilibrium in the in vivo heart, a lower PCr/ATP indicates elevated levels of myocardial free ADP. The phosphorylation potential (ΔG) for hydrolysis of ATP is proportional to the ratio of ATP to ADP and Pi, therefore elevated levels of ADP result in a significantly lower ΔG and less energy made available for each unit of ATP that is utilized (6,17). Consequently, hearts with lower a PCr/ATP ratio are energetically less efficient (6,17).

Using an in-vivo canine model of severe LVH, we reported that severity of the reduction in PCr/ATP is linearly related to the severity of LVH and that failing hearts had the lowest PCr/ATP ratio (23). We also found that a reduced PCr/ATP ratio is linearly related to the decrease in LV ejection fraction in hearts with post-infarction LV remodeling (22). It has been observed clinically that a reduction in the PCr/ATP ratio is a good predictor of mortality in patients with dilated cardiomyopathies (12).

The myocardial bioenergetic efficiency, as reflected by the PCr/ATP ratio, was severely reduced the infarct border zone of pigs in the MI group (Table 6, Figures 3 and 4). These bioenergetic alterations were remarkably more profound than in remote myocardial regions of the same MI hearts or in AoB hearts with concentric hypertrophy (Table 6, Figures 3 and 4). Moreover, the reduction in the border zone PCr/ATP ratio was more severe than what we previously reported in remote myocardial regions of failing hearts (24) suggesting that the spared myocardium in the infarct border zone had a severely reduced energetic capacity, operated at a very low energetic state, and likely are most vulnerable to oxidative stresses.

Limitations

The material properties of the LV scar can significantly impact wall stress calculation in the IZ (5,18,19). Four weeks after AMI, the necrotic AMI tissue changes to a thinned mature scar. The scarring could significantly reduce extensibility because of the increased stiffness LV scar, and consequently reduces wall stress of IZ as well as the BZ (5,18,19). Because of the material properties of the stiffness of the infarct have not been considered, the wall stress in IZ could be overestimated. However, the focus of the present study is the BZ, and an overstretched BZ myocytes (11) is likely resulted from the increase of BZ wall stress. The material properties of IZ and BZ are beyond the scope of the present study. Future experiments are warranted incorporating material property measurements into the regional wall stresses alterations to provide insights in further understanding of a ventricle with postinfarction LV remodeling.

Conclusion

The present study demonstrates that in postinfarction LV remodeling, the left ventricle should not be considered as a homogeneous organ. The functional and bioenergetic abnormalities observed in the infarct border zone of heart undergoing compensated LV remodeling were much more severe than what has been observed in remote myocardium (24). The detrimental cascade of events leading to metabolic perturbations and contractile dysfunction was likely initiated by the elevated wall stress in the overstretched cardiomyocytes within the infarct region and border zone. The data indicates that a new heart failure prevention therapeutic target may start with the prevention of systolic LV bulging and reduction of the elevated wall stress in the infarct region and border zone to prevent the abnormal myocardial bioenergetics and border zone extension.

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Figure 1. Myocardial Structure and Contractile Function

Global myocardial function was assessed by the ejection fraction (EF) (A). Myocardial wall thickness was assessed during diastole (B) and systole (C). Regional myocardial function was assessed throughout LV ring by measuring radial thickening fraction (TF) (D), circumferential shortening fraction (SF) (E), systolic wall stress (F), and diastolic wall stress (G). AN=Anterior Wall; AP=Anterior Papillary Region; LAT= Lateral Wall; PP=Posterior Papillary Region; PO=Posterior Wall; SP=Septal Wall; * p,0.01 vs normal; †p<0.05 vs. Normal; #p<0.01 vs MI; ^p<0.05 vs MI. The infarct region and border zone were localized in the AN segment.

Figure 2. Regional Analysis by MRI

LV structure was assessed in 3 horizontal short axis rings from the central portion of the LV. Each ring was divided into 6 regions according to the coronary artery perfusion pattern. In the MI group, the scar and border zone were localized in the anterior region of the LV. AN=Anterior Wall; AP=Anterior Papillary Region; LAT= Lateral Wall; PP=Posterior Papillary Region; PO=Posterior Wall; SP=Septal Wall.

Figure 3. ^{31}P -NMR Spectroscopy

A. Representative transmurally differentiated ^{31}P NMR Spectra obtained from the subendocardium (ENDO) and subepicardium (EPI) of a normal heart (normal), an infarcted heart (MI) and a heart with aortic banding (AoB). Vertical scale was adjusted in each spectrum for optimal visualization of the resonance peaks. Therefore, only the PCr/ATP ratios are compared in this figure. Resonance peaks correspond to 2, 3 diphosphoglycerate (2, 3 DPG) from the erythrocytes in the ventricular cavity; inorganic phosphate (Pi), Phosphocreatine (PCr), and the three phosphates on ATP. B. The PCr/ATP ratio was calculating by integrating the area under the PCr and ATP γ peaks. The PCr/ATP ratio was severely decreased in the infarct border zone of MI hearts, especially on the subendocardial surface, while only a moderate depletion in the PCr/ATP ratio was observed in the AoB group. * p,0.01 vs normal; #p<0.01 vs MI.

Figure 4. Correlation between Regional Variables

Correlations between the anterior wall stress and border zone myocardial bioenergetics. The observed relationships between alterations in regional wall stress and abnormalities in myocardial bioenergetics suggest that elevated wall stress increases the regional energy demand and results in bioenergetic abnormalities (Panels A and B). The observed functional and bioenergetic heterogeneity also suggests that the underlying mechanism behind the progression from compensated post-infarction LV remodeling to congestive heart failure involves the lateral expansion of functional and bioenergetic abnormalities from the infarct border zone to the entire myocardium.

Table 1

Anatomical Data

	BW (kg)	LV (g)	RV (g)	LV/BW (g/kg)	RV/BW (g/kg)	Infarct Size (SSA/LVSA)
Normal (n=8)	45 ± 5	118 ± 16	40 ± 3	2.6 ± 0.1	0.9 ± 0.1	0
MI (n=8)	44 ± 8	146 ± 9 [†]	51 ± 6	3.3 ± 0.3 [†]	1.3 ± 0.2 [†]	0.13 ± 0.3
AoB (n=8)	42 ± 4	151 ± 16 [†]	52 ± 4 [†]	3.7 ± 0.2 [†]	1.3 ± 0.2 [†]	0

Values presented as mean±SEM; n=Number of Pigs; BW=Body Weight, LV=Left Ventricular Weight, RV=Right Ventricular Weight; SSA=Scar Surface Area; LVSA=LV Surface Area (by post-mortem examination)

[†] p<0.05 vs Normal

Table 2

LV Ejection Fraction and Hemodynamics

	Ejection Fraction (%)	HR (bpm)	Ao-S (mmHg)	Ao-D (mmHg)	LVSP (mmHg)	LVEDP (mmHg)
Normal	55.3 ± 3.1	110 ± 5	100 ± 8	76 ± 12	114 ± 12	4.2 ± 1.2
MI	30.4 ± 2.3 [*]	103 ± 8	108 ± 5	78 ± 5	109 ± 5	6.2 ± 1.5
AoB	49.5 ± 2.1 [#]	117 ± 8	113 ± 5	84 ± 7	136 ± 8	5.3 ± 1.0

Values presented as mean±SEM; HR=Heart Rate; Ao-S = Aortic Systolic Pressure; Ao-D=Aortic Diastolic Pressure; LVSP=LV Systolic Pressure; LVEDP=LV Diastolic Pressure

* p<0.01 vs. Normal

[†] p<0.05 vs. Normal

[#] p<0.01 vs. MI

Table 3

Wall Thickness

	AN (IZ) (mm)	AP (BZ) (mm)	LAT (mm)	PP (mm)	PO (mm)	SP (BZ) (mm)
End Systolic Wall Thickness						
Normal	7.5 ± 0.7	7.7 ± 0.7	7.7 ± 0.6	8.1 ± 0.6	8.3 ± 0.6	8.1 ± 0.7
MI	4.4 ± 0.1 [*]	5.3 ± 0.2 [*]	6.7 ± 0.3 [†]	6.7 ± 0.2 [†]	7.0 ± 0.1 [†]	6.2 ± 0.2 [*]
AoB	9.2 ± 0.6 ^{†#}	9.1 ± 0.5 ^{†#}	9.7 ± 0.5 ^{†#}	10.3 ± 0.5 ^{†#}	9.4 ± 0.7 ^{†#}	9.1 ± 0.6 [#]
End Diastolic Wall Thickness						
Normal	5.6 ± 0.6	5.5 ± 0.6	5.3 ± 0.4	5.6 ± 0.4	6.3 ± 0.5	6.5 ± 0.6
MI	4.3 ± 0.1 [†]	4.8 ± 0.2	5.2 ± 0.2	5.2 ± 0.1	5.4 ± 0.1	4.9 ± 0.1 [†]
AoB	7.1 ± 0.4 ^{†#}	6.9 ± 0.4 ^{†#}	7.4 ± 0.3 ^{†#}	7.5 ± 0.3 ^{†#}	7.9 ± 0.7 ^{†#}	7.2 ± 0.9 ^{†#}

Values presented as mean±SEM; Values presented as mean±SEM; AN=Anterior Wall; AP=Anterior Papillary Region; LAT = Lateral Wall; PP=Posterior Papillary Region; PO=Posterior Wall; SP = Septal Wall

- * p < 0.01 vs Normal
- [†] p < 0.05 vs Normal
- [#] p < 0.01 vs MI

Table 4

Regional Myocardial Contractile Function

	AN (IZ) (%)	AP (BZ) (%)	LAT (%)	PP (%)	PO (%)	SP (BZ) (%)
Radial Thickening Fraction						
Normal	40.5 ± 3.1	45.9 ± 4.3	44.8 ± 3.1	44.2 ± 2.3	36.7 ± 3.1	40.0 ± 5.0
MI	0.2 ± 1.8*	7.1 ± 1.2*	25.5 ± 1.2*	27.3 ± 1.1*	25.4 ± 1.5 [†]	19.5 ± 1.9*
AoB	28.3 ± 4.5 ^{†#}	32.8 ± 4.0 ^{†#}	36.1 ± 5.3	32.6 ± 4.8 [†]	29.4 ± 4.6	30.8 ± 5.3 [^]
Circumferential Shortening Fraction						
Normal	-16.7 ± 0.8	-12.8 ± 0.8	-14.3 ± 1.0	-14.5 ± 0.9	-12.7 ± 1.9	-13.0 ± 1.6
MI	-5.4 ± 0.1*	-10.0 ± 0.4	-12.3 ± 0.7	-15.2 ± 0.7	-13.4 ± 0.8	-14.3 ± 0.6
AoB	-14.7 ± 1.3 [#]	-14.6 ± 0.9	-12.1 ± 1.4	-13.9 ± 1.2	-14.0 ± 1.1	-13.7 ± 1.0

Values presented as mean±SEM; AN=Anterior Wall; AP=Anterior Papillary Region; LAT = Lateral Wall; PP=Posterior Papillary Region; PO=Posterior Wall; SP = Septal Wall

* p < 0.01 vs Normal

[†] p < 0.05 vs Normal

[#] p < 0.01 vs MI

[^] p < 0.05 vs MI

Table 5

Wall Stress

	AN (IZ) (mm Hg)	AP (BZ) (mm Hg)	LAT (mm Hg)	PP (mm Hg)	PO (mm Hg)	SP (BZ) (mm Hg)
Systolic Wall Stress						
Normal	157±10	153±11	155±11	146±9	141±7	144±9
MI	297±17*	248±15*	206±15*	196±9*	186±7*	211±9*
AoB	162±6 [#]	165±8 [#]	154±4 [#]	149±7 [#]	158±7 [#]	159±10 [#]
Diastolic Wall Stress						
Normal	11±1	11±1	11±1	11±1	9.5±0.6	9±1
MI	25±3*	23±3*	21±2*	21±2*	20±2*	22±2*
AoB	10±2 [#]	10±2 [#]	10±2 [#]	9±1 [#]	9±1 [#]	9±1 [#]

Values presented as mean±SEM; AN=Anterior Wall; AP=Anterior Papillary Region; LAT = Lateral Wall; PP=Posterior Papillary Region; PO=Posterior Wall; SP = Septal Wall

* p < 0.01 vs Normal

[#] p < 0.01 vs MI

Table 6³¹P NMR Spectroscopy

PCr/ATP		
	Epi	Endo
Normal	2.12 ± 0.25	1.98 ± 0.16
MI (BZ)	1.25 ± 0.10 [*]	1.06 ± 0.11 [*]
AoB	1.62 ± 0.08 ^{*#}	1.62 ± 0.13 ^{*#}

Values presented as mean±SEM; BZ=Infarct Border Zone; PCr=Phosphocreatine; ATP = Adenosine Triphosphate Epi = subepicardium; Endo=subendocardium

^{*}
p<0.01 vs Normal

[#]
p<0.01 vs. MI (BZ)