Dobutamine-Stress Magnetic Resonance Microimaging in Mice
Acute Changes of Cardiac Geometry and Function in Normal and Failing Murine Hearts

Frank Wiesmann, Jan Ruff, Stefan Engelhardt, Lutz Hein, Charlotte Diennesch, Andrea Leupold, Ralf Illinger, Alex Frydrychowicz, Karl-Heinz Hiller, Eberhard Rommel, Axel Haase, Martin J. Lohse, Stefan Neubauer

Abstract—The aim of this study was to assess the capability of MRI to characterize systolic and diastolic function in normal and chronically failing mouse hearts in vivo at rest and during inotropic stimulation. Applying an ECG-gated FLASH-cine sequence, MRI at 7 T was performed at rest and after administration of 1.5 μg/g IP dobutamine. There was a significant increase of heart rate, cardiac output, and ejection fraction and significant decrease of end-diastolic and end-systolic left ventricular (LV) volumes (P<0.01 each) in normal mice during inotropic stimulation. In mice with heart failure due to chronic myocardial infarction (MI), MRI at rest revealed gross LV dilatation. There was a significant decrease of LV ejection fraction in infarcted mice (29%) versus sham mice (58%). Mice with MI showed a significantly reduced maximum LV ejection rate (P<0.001) and LV filling rate (P<0.01) and no increase of LV dynamics during dobutamine action, indicating loss of contractile and relaxation reserve. In 4-month-old transgenic mice with cardiospecific overexpression of the β1-adrenergic receptor, which at this early stage do not show abnormalities of resting cardiac function, LV filling rate failed to increase after dobutamine stress (transgenic, 0.19±0.03 mL/ms; wild type, 0.36±0.01 mL/ms; P<0.01). Thus, MRI unmasked diastolic dysfunction during dobutamine stress. Dobutamine-stress MRI allows noninvasive assessment of systolic and diastolic components of heart failure. This study shows that MRI can demonstrate loss of inotropic and lusitropic response in mice with MI and can unmask diastolic dysfunction as an early sign of cardiac dysfunction in a transgenic mouse model of heart failure. (Circ Res. 2001;88:563-569.)

Key Words: heart failure ■ chronic ischemic heart disease ■ remodeling ■ MRI ■ animal models

Transgenic (TG) mouse models offer the unique opportunity to investigate the pathophysiological consequences of augmented, deleted, or mutated specific genes and their dependent gene products. To analyze the resulting phenotype of such models, successful miniaturization of the well-characterized isolated perfused heart preparation has been demonstrated using either the working heart or Langendorff heart preparation.1–3 However, the isolated heart is unsuitable for the study of the complex pathophysiology of the intact cardiovascular system. In particular, the overall influence of neurohumoral factors on functional parameters can only be studied in vivo and is of major relevance when investigating the consequences of genetic alterations. Hence, reliable measurement tools are required to allow for in vivo studies of murine cardiac morphology and function. The main limitation of in vivo hemodynamic measurements with high-fidelity left ventricular (LV) micromanometer catheters4 is their invasive nature, resulting in the animal’s death at study end. Because cardiovascular disease is a dynamic process characterized by periods of compensation, transition, and decompensation, accurate and reproducible analytical techniques that can be repeated over time are required. With echocardiography, quantification of LV mass5,6 and detection of functional changes during pharmacological stress7,8 may be feasible in mice. Assessment of LV functional changes in mouse models of heart failure by echocardiography has been reported9–11 and can currently be performed with high frame rates of 100 per second. However, echocardiographic measures are based on geometric assumptions, which may no longer be valid when the ventricle undergoes asymmetrical shape changes during remodeling.12

MRI as an intrinsically 3-dimensional method allows for volumetric quantification without relying on geometric models.13 This renders the magnetic resonance (MR) method uniquely suited for the assessment of volumetric and func-
tional changes in hearts with shape distortions. We and others recently demonstrated that high-resolution MRI can visualize the murine cardiovascular anatomy with great detail. The purpose of the present study was to assess physiological changes in LV geometry and function in vivo during β-adrenergic stress in mice by MRI. We asked whether MRI allows the detection of changes of both contraction and relaxation, and thus, of systolic as well as diastolic properties of the left ventricle. The MR technique was then applied to the surgical model of chronic myocardial infarction (MI) in mice to study the effects of acute β-adrenergic stimulation on LV morphology and dynamics. Furthermore, MRI was performed in a TG mouse model of β1-adrenergic receptor overexpression. As a result of 15-fold myocardial overexpression of the β1 receptor, these mice develop progressive LV hypertrophy and myocardial fibrosis, eventually resulting in overt signs of heart failure at age 9 months. Because cardiac morphology and resting function in these mice at early age are normal, we hypothesized that dobutamine-stress MRI might allow the unmasking of a loss of contractile of relaxation reserve as an early indicator of developing heart failure.

**Materials and Methods**

**Mouse Preparation**

Fifteen male C57BL/6 mice (body weight, 20.6 ± 2.0 g) were investigated by MRI. Mice were anesthetized with inhaled isoflurane (1.5 vol % at 1 L/min oxygen flow) via a nose cone. The ECG trigger signal was taken from a homebuilt ECG unit. During the experiment, the mouse was positioned supine on a nonmagnetic warming pad to maintain constant body temperature throughout the MR study. MRI was performed in 12 mice with MI 2 weeks after ligation of the left arterial descending coronary artery (LAD). For comparison of measurements, MR studies were also performed in 6 sham-operated mice. Furthermore, MRI was performed in TG mice with β1-adrenergic receptor overexpression (TG, n = 4) and corresponding littermates (wild-type [WT], n = 5) at age 4 months (generation of the TG mouse line is described in detail in Reference 19). All experimental animal procedures were in accordance with institutional guidelines and were approved by local authorities.

**Coronary Artery Ligation**

MI was induced by LAD ligation. Mice were anesthetized with isoflurane (1.5 vol % with 1 L/min oxygen flow), and the trachea was intubated with a steel tube with outer diameter 1.1 mm. Artificial ventilation with positive airway pressure (stroke volume, 1.0 to 1.5 mL; ventilation rate, 60 to 80 per minute) was initiated, and a left-sided thoracotomy in the fourth intercostal space was performed. The intercostal muscles were then transected. After opening the pericardial sack, the left atrial appendage and the left main coronary artery were clearly identifiable. Immediately distal to the bifurcation of the left main coronary artery, the LAD was ligated using anatraumatic needle and a 6-0 silk thread. After ligation, successful infarction was immediately evident by pale discoloration of LV myocardium due to ischemia. At the end of the operation, the thorax was closed and tracheal tubes were disconnected from the ventilator, allowing for free breathing. Animals recovered and were extubated within 30 minutes after the end of the operation. For sham operation, a control group of mice underwent an identical surgical procedure with the exception that the LAD was not ligated. Perioperative survival rates of mice after LAD ligation and sham operation were 65% and 100%, respectively.

**In Vivo MRI**

Experiments were performed on a 7.05-T MR scanner (Bruker). The scanner was equipped with a microscopy gradient system capable of 870 mT/m maximum gradient strength and 280 μs rise time at maximum gradient switching. For NMR signal transmission and reception, a 10- rung birdcage probehead (Bruker) with inner diameter of 35 mm was used. For exact ECG triggering, an ECG trigger unit was used, allowing for multiple filtering of the original surface ECG signal to sufficiently isolate the QRS signal from noise generated by the magnet and the gradient coils. This trigger unit allowed for free choice of signal derivative and fine adjustment of trigger level. Hence, the trigger point was set on the ascending limb of the R wave, resulting in initiation of MR data acquisition within the isovolumetric period of LV contraction. Furthermore, to guarantee capture of end diastole, the quality of ECG triggering was checked by overlapping data acquisition, whereby cine data acquisition was started after a time delay of 50 ms from QRS with data acquisition deliberately beyond one cardiac cycle. Comparison of the LV slice volumes of the cine frame with the largest LV cavity and the first cine frame after triggering on the R wave was used for the decision on whether the set trigger level was kept or adjusted.

Dynamic imaging was performed using an ECG-triggered fast gradient echo (FLASH) cine sequence with the following parameters: echo time, 1.5 ms; repetition time, 4.3 ms; field of view (30 mm²); acquisition matrix, 256 × 256; and slice thickness, 1.0 mm. MR data acquisition was performed in multiple contiguous short-axis slices as previously described.

For assessment of LV systolic and diastolic dynamics, the cine in the midventricular short-axis slice was repeated with a higher number of frames, purposely exceeding the cardiac cycle. This allowed for data acquisition beyond late diastole into the next cardiac cycle, offering information on both the LV ejection and filling processes. MR measurements were done at rest and after intraperitoneal bolus injection of the β1-receptor agonist dobutamine (1.5 μg/g body weight).

To investigate changes of LV volumes and function, MR experiments in mice after MI (n = 12) or sham operation (n = 6) were performed. Additionally, acute LV volumetric and functional changes in infarcted murine hearts (n = 8) were assessed by in vivo MRI performed at baseline and after dobutamine injection. For standardized slice localization, measurement of peak LV ejection and filling rate in mice with MI was performed in an end-diastolic midventricular plane, which in all studied mice comprised both infarcted anterior myocardium and contracting remote myocardium.

MR experiments with identical study design were performed in TG mice with cardiac-specific overexpression of the β1-adrenergic receptor (TG, n = 4) and corresponding littermates (WT, n = 5) at an age of 4 months.

**Data Analysis**

For LV mass measurements, epicardial borders were manually delineated. LV cavity volume could be segmented by a thresholding algorithm. Total LV volumes were calculated as the sum of all slice volumes.

For assessment of LV systolic and diastolic dynamics in the dobutamine study, the cavity slice volume was measured in all acquired cine frames and was plotted against the time from onset of the QRS trigger, resulting in a volume-time curve (Figure 1). For quantitative characterization of contraction and relaxation, peak ejection rate (given by the maximum slope [dV/dt] of the systolic limb of the volume-time curve) and peak filling rate (given by the maximum slope [−dV/dt] of the LV filling curve) were calculated. This allowed us to separately assess the dynamics of both LV contraction and relaxation.

**Statistical Analysis**

Statistical analysis was performed using StatView software (Abacus Corp, Inc.). All results are given as mean ± SEM. Comparisons between rest and dobutamine were made using the Student paired t test. For comparison between sham and MI groups, an unpaired t test.
was performed. Differences were considered statistically significant at a value of $P<0.05$.

## Results

### Dobutamine Stress in Normal Mice

The fast-gradient echo MR technique offered high contrast between blood and myocardium, allowing for clear delineation of epicardial and endocardial borders (Figure 2).

There was a significant decrease of both LV end-diastolic volume (EDV) ($-26.0\%, P<0.01$) and end-systolic volume ($-64.7\%, P<0.01$) after dobutamine injection (Figure 3). However, LV stroke volume remained unchanged ($-5.7\%, P=0.16$) (Figure 3c). Ejection fraction increased significantly ($+18.7\%, P<0.01$). Heart rate remained constant throughout scanning at resting conditions but increased significantly ($+32.9\%$) after dobutamine injection (Figure 3f). This resulted in a significant increase of cardiac output ($+23.7\%, P<0.01$) compared with rest (Figure 3e), although stroke volume did not change significantly during dobutamine stress.

LV wall thickness during dobutamine stress was increased both at end diastole ($+23.7\%, P<0.05$) and end systole ($+22\%, P<0.05$) (Table 1). There was no significant change of epicardial diameters during dobutamine either at end diastole (rest, $5.0\pm0.3$ mm; dobutamine stress, $4.9\pm0.2$ mm; NS) or at end systole (rest, $4.5\pm0.3$ mm; dobutamine stress, $4.2\pm0.2$ mm, NS). Endocardial diameters, on the other hand, were significantly smaller during dobutamine stress both at diastole ($-11.8\%, P<0.05$) and, particularly, at end systole ($-52.6\%, P<0.05$) compared with resting conditions. Furthermore, relative end-diastolic wall thickness (in relation to endocardial diameter) was significantly higher under dobutamine stress ($+53.3\%, P<0.05$) (Table 1).

During dobutamine stimulation, there was a significant increase of peak LV ejection rate (rest, $0.49\pm0.05\, \mu\text{L}/\text{ms}$; stress, $0.66\pm0.06\, \mu\text{L}/\text{ms}; P<0.05$) as well as peak LV filling rate (rest, $0.41\pm0.05\, \mu\text{L}/\text{ms}$; stress, $0.67\pm0.05\, \mu\text{L}/\text{ms}; P<0.05$). Comparison of maximal ejection rate with maximal filling rate revealed no significant differences either at rest or during stress.

MRI-derived LV mass correlated well with LV mass at autopsy (LV mass$_{\text{autopsy}}=1.059\times$LV mass$_{\text{MRI}}-0.957$; $r=0.96$, $P<0.0001$). Bland-Altman analysis revealed high agreement between MRI-derived and autopsy LV mass (mean difference, $3.3\pm1.4$ mg) with narrow limits of agreement ($\pm2$ SD, $\pm11.4$ mg), indicating high accuracy of the MR volume quantification.

## Morphological and Functional Changes in Infarcted Hearts

MRI in mice 2 weeks after MI revealed marked thinning of the LV anterior wall in diastole (MI, $0.49\pm0.07$ mm; sham, $0.93\pm0.03$ mm) with complete absence of systolic thickening (for end-systolic anterior wall thickness, MI, $0.49\pm1.0$ mm; sham, $1.70\pm0.17$ mm). Furthermore, cine MRI revealed clear akinesia or even dyskinesia of the infarcted myocardium at systole (Figure 4). (Online movies can be viewed in the data supplement available at http://www.circresaha.org.) Body weight and LV mass were identical in mice 2 weeks after MI and sham operation (Table 2). Infarcted hearts revealed gross LV dilatation both at diastole and systole compared with sham ($P<0.01$ and $P<0.001$, respectively). LV stroke volume was preserved, but LV ejection fraction significantly decreased in infarcted mice ($29.4\pm4.2\%$) versus sham ($57.8\pm3.0\%$). Furthermore, there was formation of an apical aneurysm with marked LV dilatation (Figure 4).

Starting from a similar heart rate at rest (MI, $444\pm16$ bpm; sham, $420\pm16$ bpm; NS), intraperitoneal dobutamine administration resulted in a significant increase of heart rate in both
groups (MI, 510±10 bpm; sham, 531±26 bpm; NS) after a mean interval of 12±2 minutes. After initiation of scanning, heart rate was stable during the entire MR data acquisition with no changes at midtime (MI, 515±6 bpm; sham, 532±27 bpm; NS) and at the end of MR scanning (MI, 523±6 bpm; sham, 550±6 bpm; NS). During stress, only the remote portion of the myocardium showed an increase in end-diastolic (+11%, P<0.05) and end-systolic (+15.3%, P<0.05) thickness. The infarcted myocardium, however, did not increase wall thickness during dobutamine stress and remained akinetic or even dyskinetic in long-axis cine views, clearly indicating the absence of contractility within the scarred tissue.

In mice with MI, there was a significantly reduced maximum LV ejection rate (MI, 0.17±0.02 μL/ms; sham, 0.37±0.01 μL/ms; P<0.001) and LV filling rate (MI, 0.28±0.03 μL/ms; sham, 0.43±0.04 μL/ms; P<0.01). In the infarcted hearts, dobutamine stress did not induce significant changes in LV contraction or relaxation dynamics (for dV/dt, MI, 0.17±0.01 μL/ms; sham, 0.42±0.01 μL/ms [P<0.001]; for −dV/dt, MI, 0.23±0.03 μL/ms; sham, 0.47±0.03 μL/ms [P<0.001]), indicating a complete loss of contractile and relaxation reserve in the infarcted hearts.

Effects of Inotropic Stimulation in Mice With β1-Adrenergic Receptor Overexpression

MRI at rest revealed no significant differences for end-diastolic, end-systolic, and stroke volumes between TG and WT mice (Table 3). Furthermore, ejection fraction, cardiac output, and heart rate were similar, and mean LV wall thickness did not differ between TG and WT in either diastole or systole. LV mass was increased by 19% in TG mice. Whereas there was no significant difference for LV ejection and filling rates between TG and WT mice at rest, MRI clearly revealed a significant decrease of LV filling rate in TG mice during inotropic stimulation (TG, 0.19±0.03 μL/ms; WT, 0.36±0.01 μL/ms; P<0.01), indicating diastolic dysfunction. This change was seen despite a higher dobutamine-stimulated heart rate in the TG animals (TG, 560±9 bpm; WT, 515±19 bpm).

Table 1. Comparison of LV Wall Thickness and Thickening and LV Diameters in Normal Mice at Rest and During Dobutamine Stress (n=15)

<table>
<thead>
<tr>
<th></th>
<th>Rest</th>
<th>Dobutamine</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>ED wall thickness, mm</td>
<td>0.76±0.04</td>
<td>0.94±0.06</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>ES wall thickness, mm</td>
<td>1.31±0.09</td>
<td>1.60±0.06</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Systolic wall thickening, mm</td>
<td>0.56±0.06</td>
<td>0.66±0.07</td>
<td>NS</td>
</tr>
<tr>
<td>Systolic wall thickening, %</td>
<td>74±7</td>
<td>75±11</td>
<td>NS</td>
</tr>
<tr>
<td>ED epicardial diameter, mm</td>
<td>5.0±0.3</td>
<td>4.9±0.2</td>
<td>NS</td>
</tr>
<tr>
<td>ES epicardial diameter, mm</td>
<td>4.5±0.3</td>
<td>4.2±0.2</td>
<td>NS</td>
</tr>
<tr>
<td>ED endocardial diameter, mm</td>
<td>3.4±0.1</td>
<td>3.0±0.2</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>ES endocardial diameter, mm</td>
<td>1.9±0.1</td>
<td>0.9±0.2</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Relative ED wall thickness, %</td>
<td>45±4</td>
<td>69±7</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>ED LV long-axis length, mm</td>
<td>6.9±0.3</td>
<td>6.2±0.2</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>ES LV long-axis length, mm</td>
<td>3.9±0.3</td>
<td>5.6±0.3</td>
<td>NS</td>
</tr>
</tbody>
</table>

ED indicates end-diastolic; ES, end-systolic.
Discussion

With the increasing importance of TG mouse models in cardiovascular research, molecular biologists and geneticists are demanding methods for accurate assessment of cardiac morphology and function. This study demonstrates the potential of high-resolution MRI to visualize acute changes of cardiac geometry and function in the intact and failing mouse heart during adrenergic stimulation.

Cardiac anatomy and physiology place high demands on any potential imaging method. The small size of the mouse heart as well as the high basal heart rate require the highest spatial and temporal resolution. These demands of the mouse heart can be fully met by the presented MR method. We chose a nominal in-plane image resolution (117 pixels, resulting in 850 pixels within the LV wall of a midventricular short-axis slice. Although the chosen slice thickness of 1 mm in the murine studies does not compare favorably with the 5- to 7-mm slice thickness commonly used in humans, it represents a compromise between total imaging time and sufficient signal-to-noise ratio.

Whereas previous cine MRI studies in mice reported by Kubota et al. and Franco et al. showed a limited temporal resolution of 24 and 39 ms, respectively, the presented MR microimaging method allows for very short acquisition times as a result of rapid gradient performance. Hence, in this study, robust image acquisition with acquisition windows of 4.3 ms per cine frame (corresponding to a frame rate of 233 per second) was feasible even up to peak heart rates of 750 bpm (Figure 2).

Achievement of short total scan times is particularly necessary in light of the anesthesia, which at present is unavoidable for in vivo MRI in the mouse. Many anesthetics cause respiratory depression and negative chronotropic and inotropic effects. In this study, inhalative anesthesia with isoflurane was chosen, which is easy to administer and to...

$\approx$850 pixels within the LV wall of a midventricular short-axis slice. Although the chosen slice thickness of 1 mm in the murine studies does not compare favorably with the 5- to 7-mm slice thickness commonly used in humans, it represents a compromise between total imaging time and sufficient signal-to-noise ratio.

Whereas previous cine MRI studies in mice reported by Kubota et al. and Franco et al. showed a limited temporal resolution of 24 and 39 ms, respectively, the presented MR microimaging method allows for very short acquisition times as a result of rapid gradient performance. Hence, in this study, robust image acquisition with acquisition windows of 4.3 ms per cine frame (corresponding to a frame rate of 233 per second) was feasible even up to peak heart rates of 750 bpm (Figure 2).

Achievement of short total scan times is particularly necessary in light of the anesthesia, which at present is unavoidable for in vivo MRI in the mouse. Many anesthetics cause respiratory depression and negative chronotropic and inotropic effects. In this study, inhalative anesthesia with isoflurane was chosen, which is easy to administer and to...
control and has short onset and termination times. Whereas robust ECG triggering is crucial for cardiac MR volumetry, correction for breathing motion is not essential for cine FLASH MRI in the mouse. Because of the murine breathing pattern under isoflurane with long standstill of breathing at end expiration and because of the compensating effects of multiple signal averaging,21 MRI showed high accuracy in the volumetric validation. Hence, temporal averaging of the respiratory cycle showed only minor influence on measurement accuracy. Further advantages of isoflurane are its relatively low degree of negative inotropic and chronotropic effects. In previous MRI studies in mice, mean heart rates of 356 bpm were reported by Franco et al11 for intraperitoneal tribromoethanol (Avertin). Kubota et al9 described a reduced mean heart rate of 218 bpm with intraperitoneally administered pentobarbital. Hence, by using isoflurane, MRI in mice could be performed closer to physiological conditions (baseline heart rate, 417±16 bpm; baseline ejection fraction, 69±2%). One limitation in this study is the calculation of LV peak ejection and filling rates from one midventricular slice volume only. Although it would be ideal to follow the volume-time relationship of the entire left ventricle, such a volume only. Although it would be ideal to follow the peak ejection and filling rates from one midventricular slice could be performed closer to physiological conditions (baseline heart rate, 417±16 bpm; baseline ejection fraction, 69±2%). One limitation in this study is the calculation of LV peak ejection and filling rates from one midventricular slice volume only. Although it would be ideal to follow the volume-time relationship of the entire left ventricle, such a measurement would prohibitively prolong the total duration of anesthesia.

Effects of Dobutamine on the Murine Heart
Whereas in previous echocardiographic studies in TG mice with cardiac contractile failure only changes of LV diameters and global systolic function were described,8,22 the main purpose of this study was to demonstrate the feasibility of MRI during inotropic stimulation to separately quantify systolic and diastolic LV performance.

The β-adrenergic effect of dobutamine was detected after a mean interval of 12±2 minutes after application by a significant increase of heart rate, which lasted for ≈25 minutes. This is in good agreement with other data.8 MR data acquisition during dobutamine action was completed within 13 minutes (because of the higher heart rate during stress) without any changes of heart rate during MR data acquisition. Consistent with effects of β-adrenergic stimulation in humans,23 there was a significant decrease of both EDV and end-systolic volume. However, we could not detect significant changes in stroke volume during dobutamine action, in contrast to human physiology, in which stroke volume can be augmented during β-adrenergic stimulation.24,25 In parallel, LV wall thickness increased both at diastole and systole, whereas systolic wall thickening did not change during dobutamine action. Hence, under adrenergic stimulation, the mouse heart initiates its contraction from a lower diastolic volume level (with a partially precontracted LV wall) to a lower systolic volume level (with further thickening of the LV wall) compared with rest (Figure 2), although the absolute volumetric change remains similar. These geometric changes are also represented by the increase in relative wall thickness as a measure of the ratio between wall thickness and LV cavity diameter (Table 1). Furthermore, during inotropic stimulation the diastolic shape of the left ventricle changes from an ellipsoidal to a more spherical one, as attested to by a significant reduction of diastolic LV long axis length (Table 1).

### Table 4. Differences in Regional LV Wall Thickness and Thickening in Mice With Myocardial Infarction at Rest and During Dobutamine Stress (n=8)

<table>
<thead>
<tr>
<th></th>
<th>Rest</th>
<th>Dobutamine Stress</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>ED wthremote, mm</td>
<td>0.32±0.04</td>
<td>0.34±0.03</td>
<td>NS</td>
</tr>
<tr>
<td>ES wthremote, mm</td>
<td>0.41±0.04</td>
<td>0.41±0.02</td>
<td>NS</td>
</tr>
<tr>
<td>Systolic wall thickening_{remote} %</td>
<td>30.6±9.1</td>
<td>22.1±9.8</td>
<td>NS</td>
</tr>
<tr>
<td>ED wthremote, mm</td>
<td>1.23±0.05</td>
<td>1.36±0.01</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>ES wthremote, mm</td>
<td>1.64±0.04</td>
<td>2.05±0.07</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Systolic wall thickening_{remote} %</td>
<td>34.3±6.4</td>
<td>50.7±6.5</td>
<td>NS</td>
</tr>
</tbody>
</table>

Abbreviations as in Table 2 legend.

β-Adrenergic Stress in Overt and Latent Murine Heart Failure
Consistent with postmortem morphology,26 infarcted hearts showed severe LV dilatation with formation of an apical aneurysm in MR images. Furthermore, because of the gross dilatation of the left ventricle, turbulent motion of blood within the ventricle was observed in the MR images, appearing as whirling lines in the apical portion of the LV (Figure 4).

Whereas the changes of overall LV geometry and global function were evident at rest, application of dobutamine in addition revealed a reduced response of the remote intact myocardium in mice with MI. This might represent a loss of contractile reserve in the noninfarcted tissue. However, some inotropic response cannot be excluded given the reduction of end-diastolic LV size during inotropic stimulation.

During dobutamine stimulation, the intact basal portion of the myocardium showed increase of wall thickness both at diastole and systole, which might reflect a precontracted level of the remote LV myocardium during β-adrenergic stimulation. The improvement of systolic wall thickening during dobutamine, however, did not reach statistical significance (Table 4). This reflects the remodeling process of the myocardium of infarcted hearts, with reduced function of the noninfarcted, remote myocardium.27,28 As expected, MRI myocardium of infarcted hearts, with reduced function of the noninfarcted, remote myocardium.27,28 As expected, MRI during dobutamine did not detect an improvement in wall motion or thickness in the infarcted area of the myocardium. The theoretical chance of a diminished response in the failing hearts due to a decreased absorption of IP dobutamine can be excluded, because there was a similar increase in heart rate in both MI and sham mice, representing similar chronotropic response to dobutamine.

Major changes in the dynamics of the LV contraction and relaxation processes were also seen from volume-time curves derived from cine MR imaging. At baseline, infarcted hearts started from a much higher EDV than sham-operated hearts, representing LV dilatation. LV peak ejection and filling rate were significantly slower in MI compared with sham mice, demonstrating the severe impairment of contraction and relaxation processes. Under β-adrenergic receptor stimulation, infarcted hearts showed no increase in contraction and relaxation rate compared with hearts at rest, indicating complete loss of inotropic and lusitropic reserve. Hence, it is concluded that the chronically infarcted murine heart preserves its LV stroke volume by working at its performance limits. However, some inotropic responses cannot be ex-
cluded, given the reduction of end-diastolic LV size during inotropic stimulation.

Studies in TG mice allowed us to show that MRI under dobutamine stimulation can actually reveal diastolic dysfunction in animals that appear normal under resting conditions. In mice with myocardial overexpression of the β1-adrenergic receptor, LV geometry and volumes as well as LV function represented by ejection fraction and cardiac output were identical for TG and WT mice at age 4 months. Because the only difference detected at that early age was a significantly increased LV mass in the TG mice, we hypothesized that high-resolution MRI during inotropic stimulation might show changes in the ventricular ejection and filling dynamics and thus allow us to noninvasively detect an early stage of heart failure by unmasking impaired inotropic and lusitropic response to pharmacological stress. Indeed, under dobutamine, compared with WT mice, we observed a significantly lower maximal LV filling rate (−dV/dt) in β1 receptor–overexpressing mice. This demonstrates early loss of relaxation reserve in this model and highlights the potential of dobutamine-stress MRI in mice to detect early stages of LV dysfunction.

In conclusion, dobutamine-stimulation MRI is a powerful tool for the characterization of the cardiovascular phenotype in mice and may allow novel insight into the consequences of genetic alterations for the development of heart failure.

Acknowledgments

This study was supported by grants from the Deutsche Forschungsgemeinschaft (Grant Wi-1510/2-1) and Sonderforschungsbereich 355 “Pathophysiologie der Herzinsuffizienz,” Teilprojekt A1, A6, C9, and C10, and by British Heart Foundation Project Grant PG/2000039. We thank Sabine Voll and Titus Lanz for technical assistance.

References


Dobutamine-Stress Magnetic Resonance Microimaging in Mice: Acute Changes of Cardiac Geometry and Function in Normal and Failing Murine Hearts
Frank Wiesmann, Jan Ruff, Stefan Engelhardt, Lutz Hein, Charlotte Dienesch, Andrea Leupold, Ralf Illinger, Alex Frydrychowicz, Karl-Heinz Hiller, Eberhard Rommel, Axel Haase, Martin J. Lohse and Stefan Neubauer

_Circ Res._ 2001;88:563-569
doi: 10.1161/01.RES.88.6.563

_Circulation Research_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2001 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/88/6/563

Data Supplement (unedited) at:
http://circres.ahajournals.org/content/suppl/2001/03/20/88.6.563.DC1

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in _Circulation Research_ can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to _Circulation Research_ is online at:
http://circres.ahajournals.org/subscriptions/