Echocardiographic criteria for detection of postinfarction congestive heart failure in rats

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Sjaastad, Ivar, Ole M. Sejersted, Arnfinn Ilebekk, and Reidar Bjørnerheim. Echocardiographic criteria for detection of postinfarction congestive heart failure in rats. J Appl Physiol 89: 1445–1454, 2000.—We evaluated postinfarction myocardial function in rats and determined echocardiographic criteria for congestive heart failure (CHF) using high performance echocardiography. Extensive myocardial infarction (MI) was induced in rats by left coronary occlusion. Sham-operated animals served as controls. Five weeks later, high-frame rate (≈200 Hz), fully digitized, shallow-focus (10–25 mm), two-dimensional, M-mode and Doppler echocardiography was performed. A J-tree cluster analysis was performed using parameters indicative of CHF. Reproducibility was examined. The cluster analysis joined the animals into one Sham and two MI clusters. One of the MI clusters had clinical characteristics of CHF and elevated left ventricular end diastolic pressure. Among the echocardiographic variables, only posterior wall shortening velocity separated the failing and nonfailing MI clusters. We conclude that, by high frame rate echocardiography, it is possible to obtain high-quality recordings in rats. It is feasible to distinguish MI rats with CHF due to myocardial dysfunction from those without failure and to perform longitudinal studies on myocardial function.

Congestive heart failure (CHF) is a serious outcome of myocardial infarction (MI) (1). A CHF diagnosis should be based on symptoms, clinical signs, and echocardiographic findings according to the recommendations presented by the American Society of Echocardiography (25) or the guidelines from European Society of Cardiology (2). Animal models have been used to study various aspects of CHF (e.g., cardiomyopathic rodents (26), pacing-induced failure in dogs (7), salt-sensitive rats (13, 14), hypertensive rats (20), and postinfarction failure in rats (3, 6, 15)). Postinfarction animal models have been extensively studied because myocardial ischemia and infarction are frequent causes of CHF in humans. However, not all postinfarction hearts undergo transition to CHF, and it is generally acknowledged that left ventricular (LV) dilatation occurs mainly after large transmural infarctions (21, 22). In many studies in rats, echocardiographic validation of the model has not been performed, and, consequently, there are no data that allow assessment of the degree of contractile failure. In studies of possible cellular dysfunction in heart failure after MI, it is clearly essential to select animals that have a safe diagnosis of CHF.

Traditionally, because it reflects preload, left ventricular end diastolic pressure (LVEDP) has been used as a main criterion for CHF in rats. However, it is necessary to cannulate one of the carotid arteries to measure LVEDP, and this prevents longitudinal studies. In addition, LV catheterization may cause damage to the aortic valves, and introduction of a catheter into the LV cavity might significantly affect cardiac performance. Accordingly, LVEDP should not be used as the main criterion of CHF (2). Echocardiography represents an alternative approach. There are several possible noninvasive echocardiographic variables that may be used to verify a diagnosis of CHF, such as left ventricular diameter in end-diastole (LVDd), fractional shortening (FS), or left atrial diameter (LAD). However, our laboratory has previously found that LVDd may increase without a concomitant elevation of LVEDP and CHF (Sjaastad, unpublished observations). For this reason, it is important to validate the echocardiographic criteria.

In experiments on papillary muscle strips (28) and isolated cardiomyocytes (12) from CHF rats in overt failure, it has been shown that shortening velocity is substantially reduced. On this basis, we hypothesized that LV wall shortening velocity may be a valid and reliable criterion for CHF. Because infarction in many cases also involves a large part of the anterolateral LV wall, the LV posterior wall may give the most reliable estimate of shortening velocity. In the present study, we used a commercially available, high-frame rate, fully digitized echocardiograph with modified software to allow high-precision recordings. We compared the sensitivity of various variables in detecting and separating CHF, as a group, among postinfarction rats.
Cluster analysis was performed to join the animals into clusters with similar characteristics, and we examined whether the animals in the clusters also had clinical signs of CHF. We looked for the echocardiographic measure that best separated the various clusters, and that measure became the optimal criterion for distinguishing failing rats from nonfailing and sham-operated (Sham) rats.

**METHODS**

The animals were cared for according to the Norwegian Animal Welfare Act, which conforms to the National Institute of Health guidelines (NIH publication No. 85–23, revised 1996). Two animals were kept in each cage, and all animals were housed in a temperature-regulated room that was equipped with an automatic 12:12-h light-dark cycle.

*Induction of myocardial infarction.* Male Wistar rats (Møllerlegard Breeding and Research Center, Skensved, Denmark), weighing ~320 g, were intubated and ventilated on a Zoovent ventilator (Triumph Technical Services, Milton Keynes, UK) with 68% N2O, 29% O2, and 2–3% isofluran (Abbott Laboratories). Through a thoracotomy on the left side, the heart was exteriorized and extensive MI was induced by ligation of the left coronary artery as described by Tønnesen et al. (29). Sham animals were subjected to the same surgical procedure, but the coronary artery was not ligated. Echocardiography was performed 5 wk later, with the rats sedated (Sham) rats.

*Hemodynamic measurements.* A 2-Fr microtip pressure transducer catheter (SPR-407, Millar Instruments) was introduced into the LV through the right carotid artery for measurements of LV systolic pressure and LV pressure-volume relations. The left arm was dissected out to the brachial artery, which was used to introduced into the LV through the right carotid artery for measurements of LV systolic pressure and LV pressure-volume relations. The left arm was dissected out to the brachial artery, which was used to

*Echocardiography.* In vivo heart function was evaluated by echocardiography using a fully digitized Vingmed System Five (GE Vingmed Ultrasound, Horten, Norway) with a 10-MHz linear array transducer specially designed for examination of superficial vessels. The rats were examined while sedated and in the supine position, with the chest closed, and the transducer placed gently in the left parasternal position. High frame rate was achieved by software modification for the transducer placed gently in the left parasternal position. High frame rate was achieved by software modification for the transducer placed gently in the left parasternal position. High frame rate was achieved by software modification for the transducer placed gently in the left parasternal position. High frame rate was achieved by software modification for

*Statistics.* Data are expressed as group means ± SE, unless noted otherwise. Comparisons between groups were made using one-way ANOVA and Tukey’s post hoc test. When equal variance tests failed, a Kruskal-Wallis one-way analysis on ranks was performed with a Dunn’s post hoc test.

By convention (24), inter- and intrascorer reliability was calculated using Spearman’s rank correlation coefficient (25). The average difference between the scorers ±2 SD, corresponding to the mean and “limits of agreement” in the Bland-Altman analysis (2). Average absolute difference between the scorers was expressed as means ±SD. Between-scorer difference was tested with a paired t-test. Differences were considered significant for P < 0.05.

A J-tree cluster analysis of selected echocardiographic and heart weight data indicating CHF was performed using single linkage and Euclidean distances. The cluster analysis compares cases (animals) with each other, using parameters relevant to the phenomenon of interest. Considering all parameters used, the analysis sets up a linkage tree in which a
short linkage distance implies that the cases are similar, whereas cases that are very different have a long linkage distance. This analysis, therefore, provides an unbiased way of grouping data.

RESULTS

Cluster analysis. J-tree cluster-analysis was performed using echocardiographic data for LVDd, LAD, and PWSV, all possibly altered in heart failure. In addition, heart weight, indicating hypertrophy and dilatation, and Sham-MI grouping were used in the analysis that ultimately joined the animals into two clusters (Fig. 1) that correspond to the MI and Sham groups. The MI cluster was joined from two clusters. The animals in the first MI cluster had large transmural anterolateral infarctions, hypertrophied hearts, dilated left ventricles, dilated left atrium, reduced PWSV, low CO, pleural effusion, ascites, tachypnea, and reduced weight gain, all indicative of chronic CHF. The animals in this cluster were dubbed CHF. The other MI cluster had medium-to-large transmural infarctions, slightly increased heart weight compared with Sham, dilated left ventricles, normal-to-moderately dilated left atrium, and slightly reduced PWSV, and CO was reduced but higher than in the other MI cluster. Only one of the animals in this cluster had clinical signs of CHF. The characteristics of the second cluster indicate that the animals did not have CHF, and we labeled these animals as MI nonfailing (MInf, Fig. 1).

Animal characteristics. The average heart weight in the CHF cluster was 35 and 73% higher than in the MIInf and the Sham clusters, respectively (Table 1). There was no overlap between heart weights in the CHF and Sham clusters, and the weights of only two hearts overlapped between CHF and MIInf (Fig. 2A), whereas there was more overlap of heart weights between MIInf and Sham. In the CHF cluster, body weight and weight gain were significantly lower than in MIInf and Sham (Table 1). Heart-to-body weight ratios were significantly increased in CHF compared with both MIInf and Sham (Table 1). The infarct zone comprised most of the free wall of the left ventricle in CHF (5.31 ± 0.18 arbitrary units). In MIInf, the average infarct size was smaller (3.70 ± 0.52, P < 0.01; Fig. 2).

Inter- and intrascorer reliability. To assess inter- and intrascorer reliability, echocardiographic data from a subset of 10 animals were analyzed by two persons under the same physical conditions (i.e., in the same room and illumination). One scorer analyzed the data twice, on separate days, to evaluate the intrascorer reliability. Both the intra- and interscorer reliabilities were acceptable with a mean difference <10% (Table 2), and no significant differences were detected. Thus analysis by one person who was unaware of the hemodynamic data seemed to be satisfactory to obtain reliable results.

LV and left atrial dimensions. The interventricular septum (IVS) was thinner in CHF and MIInf than in Sham (Table 3) because the infarct area in some animals was included in the measurements due to extensive infarctions that comprised most of the anterior wall. LVDd was 38 and 27% larger in CHF and MIInf, respectively, than in Sham (P < 0.01), and the difference was greatly increased with regard to LVDs because FS was reduced by 76 and 38% in CHF compared with SHAM and MIInf, respectively. M-mode and long axis recordings of the left ventricle acquired in diastole and systole are shown in Figs. 3 and 4. The beams were focused on the left ventricular posterior wall (LVPW), leaving the scarred anterior wall slightly out of focus. In diastole, the LVPW was significantly thicker than MIInf and CHF than in Sham (Table 3). A linear fit was made to the maximal PWSV in M-mode using the endocardial border of the LVPW. We found reduced shortening velocity in CHF compared with the shortening velocity of both MIInf and Sham (P < 0.01), and there was no overlap between results in CHF and MIInf (Fig. 2C).

CHF is usually associated with a dilatation of the left atrium (21). We found a 73% increase in LAD measured in M-mode in CHF compared with Sham (Table

Table 1. Animal characteristics

<table>
<thead>
<tr>
<th></th>
<th>Sham</th>
<th>MIInf</th>
<th>CHF</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of animals</td>
<td>14</td>
<td>10</td>
<td>16</td>
</tr>
<tr>
<td>Body wt, g</td>
<td>410 ± 5</td>
<td>402 ± 7</td>
<td>381 ± 6*†</td>
</tr>
<tr>
<td>Body wt gain, g</td>
<td>87 ± 4</td>
<td>81 ± 5</td>
<td>61 ± 6*†</td>
</tr>
<tr>
<td>Heart wt, g</td>
<td>1.28 ± 0.04</td>
<td>1.64 ± 0.06‡</td>
<td>2.22 ± 0.07</td>
</tr>
<tr>
<td>Heart wt/body wt, g/kg</td>
<td>3.1 ± 0.1</td>
<td>4.2 ± 0.2‡</td>
<td>5.4 ± 0.2‡†</td>
</tr>
</tbody>
</table>

Values are means ± SE. Sham, sham-operated cluster; MIInf, nonfailing myocardial infarction cluster; CHF, congestive heart failure cluster. *CHF significantly different from Sham (P < 0.01); ‡CHF significantly different from MIInf (P < 0.01); †Sham significantly different from MIInf (P < 0.01).
but values in MI_{inf} overlapped with values from both groups (Fig. 2D). M-mode recordings of aorta and LAD are shown in Fig. 4. In Fig. 5, the relationship between LAD and LVDd is shown. The relationship is not linear, and the data indicate that increased LVDd is not necessarily associated with CHF, unless LAD is increased at the same time. Aortic diameter was not significantly different between the groups (Figs. 3, 4, and 5).

**Doppler data.** Peak mitral flow velocity was not significantly different between the groups (Table 4). In CHF, the peak LVOT flow velocity was 28–29% lower and CO was 42% lower than in both MI_{inf} and Sham. The average CO values calculated from LVOT and RVOT were not significantly different, being 20.14 ± 0.32 ml/min, with all three groups included. Doppler tracings from LVOT, RVOT, and the mitral valves are shown in Fig. 6.

**Hemodynamic data.** Table 5 shows that the contractility parameter (LVdP/dt_{max}/IP) averaged 77 ± 6 s^{-1} in the CHF cluster, 121 ± 6 s^{-1} in MI_{inf} and 154 ± 10 s^{-1} in Sham (P < 0.001 between all groups). Values from MI_{inf} in some cases overlapped with values from CHF and Sham (Fig. 7A). Figure 7 also shows the relationship between LVdP/dt_{max}/IP and PWSV. Even though LVdP/dt_{max}/IP does not distinguish between MI_{inf} and CHF, high values of LVdP/dt_{max}/IP are seen in animals with high PWSV.

In CHF rats, LV systolic pressure was 25% lower than in both MI_{inf} and Sham (Table 5; P < 0.01). However, LVEDP was substantially higher in CHF compared with both Sham and MI_{inf} (P < 0.001). Our laboratory has previously used LVEDP $\geq$ 15 mmHg as a cutoff value for inclusion in studies on CHF (12, 29). In Table 6 the CHF, MI_{inf} and SHAM clusters are grouped using the criteria LVEDP $>\leq$ 15 mmHg. All rats in the CHF cluster had LVEDP $>\leq$ 15 mmHg, whereas all rats in the MI_{inf} and Sham clusters had LVEDP $<\leq$ 15 mmHg. These results indicate that there is a high correspondence between pressure measurements indicating failure and the parameters used in the cluster analysis and that the risk of type I or II errors is acceptably small when using echocardiographic data.

**DISCUSSION**

Echocardiography can offer important information on cardiac function in rodents. We used a novel, fully digital echocardiographic system with slightly modified software to give sufficient time resolution to allow reliable measurements both in 2D, M-mode, and Doppler. The J-tree cluster analysis, based on selected echocardiographic data and heart weight, allowed us to separate Sham and MI rats and, furthermore, to dis-
Distinguish between two subgroups within the MI cluster, one with and one without signs of failure. The CHF cluster had the clinical, hemodynamic, and echocardiographic characteristics typical for animals in overt heart failure, whereas the other MI cluster lacked these characteristics. Analysis of the echocardiographic data showed that PWSV was lower in all CHF animals compared with MI<sub>inf</sub> animals, whereas the

Table 3. Left ventricular, atrial, and aorta dimensions

<table>
<thead>
<tr>
<th></th>
<th>Sham M-mode</th>
<th>Sham 2D SAX</th>
<th>MI&lt;sub&gt;inf&lt;/sub&gt; M-mode</th>
<th>MI&lt;sub&gt;inf&lt;/sub&gt; 2D SAX</th>
<th>CHF M-mode</th>
<th>CHF 2D SAX</th>
</tr>
</thead>
<tbody>
<tr>
<td>IVS, mm</td>
<td>1.9 ± 0.1</td>
<td>1.8 ± 0.1</td>
<td>1.0 ± 0.1‡</td>
<td>1.3 ± 0.1</td>
<td>1.1 ± 0.1*</td>
<td>1.2 ± 0.1</td>
</tr>
<tr>
<td>LVD&lt;sub&gt;d&lt;/sub&gt;, mm</td>
<td>7.4 ± 0.2</td>
<td>7.0 ± 0.2</td>
<td>9.4 ± 0.3‡</td>
<td>8.8 ± 0.2</td>
<td>10.2 ± 0.1*†</td>
<td>9.9 ± 0.2</td>
</tr>
<tr>
<td>FS, %</td>
<td>54 ± 1</td>
<td>50 ± 2</td>
<td>21 ± 2‡</td>
<td>20 ± 2</td>
<td>13 ± 1*†</td>
<td>11 ± 1</td>
</tr>
<tr>
<td>PWD, mm</td>
<td>1.85 ± 0.06</td>
<td>1.75 ± 0.04</td>
<td>2.00 ± 0.06‡</td>
<td>2.06 ± 0.05</td>
<td>2.21 ± 0.09*†</td>
<td>2.23 ± 0.05</td>
</tr>
<tr>
<td>PW&lt;sub&gt;d&lt;/sub&gt;, mm</td>
<td>2.93 ± 0.11</td>
<td>2.80 ± 0.10</td>
<td>40 ± 4‡</td>
<td>28 ± 1*†</td>
<td>27.9 ± 0.11</td>
<td></td>
</tr>
<tr>
<td>LAD, mm</td>
<td>4.9 ± 0.1</td>
<td>5.9 ± 0.3‡</td>
<td>8.5 ± 0.2*†</td>
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</tr>
<tr>
<td>PWSV, cm/s</td>
<td>4.12 ± 0.18</td>
<td>3.40 ± 0.17‡</td>
<td>2.12 ± 0.09*†</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Aorta, mm</td>
<td>2.77 ± 0.06</td>
<td>2.74 ± 0.06</td>
<td>2.64 ± 0.06</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE. IVS, interventricular septum; LVD<sub>s</sub>, left ventricular diameter at systole; FS, fractional shortening; PWD<sub>d</sub>s, posterior wall thickness in diastole and systole, respectively; 2D SAX, two-dimensional short axis. Statistics are given for M-mode data. *CHF significantly different from Sham (P < 0.01); †CHF significantly different from MI<sub>inf</sub> (P < 0.01); ‡Sham significantly different from MI<sub>inf</sub> (P < 0.01).

Fig. 3. Examples of long axis views of hearts in end diastole and end systole from the three clusters. The left atrium (LA), posterior wall (PW), LV cavity (LV), and interventricular septum (IVS) are identified. In the frames from MI<sub>inf</sub> and CHF, the infarction (INF) can be seen.
other echocardiographic measurements did not discriminate as well between the two groups.

Cluster analysis. Cluster analysis is used to identify cases or animals with similar characteristics. The variables used in the analysis should be associated with the phenomenon of interest. Echocardiographic data provide information about cardiac function that may allow clear discrimination between Sham, MI_{nf}, and CHF cases. We used the variables LVDd, LAD, and PWSV; in addition, we also included heart weight, reflecting hypertrophy, fibrosis, and dilatation as variables. LVDd is related to LV function, and it has previously been shown that this parameter is altered in CHF, in both rodent (15) and human (5) studies. PWSV has not previously been examined in studies on small animals, but, according to our hypothesis, the reduced shortening velocity found in papillary preparations (28) and in isolated cells (12) from the left ventricle indicate a reduced PWSV. In papillary muscle preparations, there is reduced shortening velocity in MI compared with Sham, indicating that the corresponding echocardiographic parameter may distinguish between MI_{nf} and CHF (I. Sjaastad, I. Orskomedal, J. B. Osnes, and O. M. Sejersted; unpublished data). PWSV is directly related to myocardial function, whereas ejection fraction, FS, LVDd, and LVDs also are influenced by other factors, such as the

![Fig. 4. M-mode tracings of the left ventricle (top) and atrium (bottom). Top shows that the septum/anterior wall was thin and did not contract in CHF, and contraction was depressed in MI_{nf}. Note reduced PW shortening and PWSV in CHF. LV cavity was dilated both in CHF and MI_{nf}, but fractional shortening was more depressed in CHF than in MI_{nf}. Bottom shows M-mode tracings of the aorta (Ao) and LA. LAD was increased in CHF.]

![Fig. 5. Relation between LV diameter and LAD. All three clusters are plotted. ● Sham; ○ MI_{nf}; ■ CHF. Notice that the MI_{nf} and Sham clusters both have small atria, whereas the MI_{nf} and CHF clusters both have large left ventricles.]

Table 4. Doppler data

<table>
<thead>
<tr>
<th></th>
<th>Sham</th>
<th>MI_{nf}</th>
<th>CHF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak mitral flow, ms⁻¹</td>
<td>0.73 ± 0.05</td>
<td>0.79 ± 0.03</td>
<td>0.84 ± 0.06</td>
</tr>
<tr>
<td>Peak LVOT flow, ms⁻¹</td>
<td>1.01 ± 0.06</td>
<td>1.02 ± 0.13</td>
<td>0.73 ± 0.05*†</td>
</tr>
<tr>
<td>Peak RVOT flow, ms⁻¹</td>
<td>0.77 ± 0.05</td>
<td>0.75 ± 0.04</td>
<td>0.54 ± 0.03**</td>
</tr>
<tr>
<td>CO in LVOT, ml/min</td>
<td>117 ± 2.6</td>
<td>116 ± 8</td>
<td>68 ± 6*†</td>
</tr>
<tr>
<td>CO in RVOT, ml/min</td>
<td>119 ± 2.8</td>
<td>112 ± 9</td>
<td>70 ± 4*†</td>
</tr>
<tr>
<td>VTI in LVOT</td>
<td>5.6 ± 0.3</td>
<td>5.3 ± 0.5</td>
<td>3.9 ± 0.3*†</td>
</tr>
<tr>
<td>VTI in RVOT</td>
<td>5.1 ± 0.2</td>
<td>5.1 ± 0.3</td>
<td>3.4 ± 0.1*†</td>
</tr>
</tbody>
</table>

Values are means ± SE. LVOT, left ventricular outflow tract; RVOT, right ventricular outflow tract; CO, cardiac output; VTI, velocity time integral. *CHF significantly different from Sham (P < 0.01); †CHF significantly different from MI_{nf} (P < 0.01); ‡Sham significantly different from MI_{nf} (P < 0.01).
LV geometry. It is thus reasonable to assume that
PWSV correlates better than other echocardiographic
variables with in vitro myocardial parameters. LAD, a
variable also used in the present study, dilates later
than the LV, and seems to be a more valid measure of
CHF than LVDd or FS. Hypertrophy of the ventricles
and the atria and formation of LV scar tissue contrib-
ute to increased heart weight.

The outputs of the cluster analysis are clusters with
cases that have similar characteristics, but the analy-
ysis does not offer a statistical interpretation of the

Table 5. Hemodynamic data

<table>
<thead>
<tr>
<th></th>
<th>Sham</th>
<th>MIaf</th>
<th>CHF</th>
</tr>
</thead>
<tbody>
<tr>
<td>LVSP, mmHg</td>
<td>111 ± 5</td>
<td>111 ± 8</td>
<td>83 ± 2 †‡</td>
</tr>
<tr>
<td>LVEDP, mmHg</td>
<td>4.2 ± 0.9</td>
<td>6.8 ± 0.8 ‡</td>
<td>24.8 ± 1.3 ††</td>
</tr>
<tr>
<td>Heart rate, beats/min</td>
<td>347 ± 11</td>
<td>361 ± 12</td>
<td>323 ± 10 ††</td>
</tr>
<tr>
<td>LVDp/dt_max, mmHg/s</td>
<td>9524 ± 1023</td>
<td>7440 ± 492 ‡‡</td>
<td>4540 ± 427 ††</td>
</tr>
<tr>
<td>IP, mmHg</td>
<td>80 ± 4</td>
<td>62 ± 3</td>
<td>58 ± 2</td>
</tr>
<tr>
<td>LVDp/dt_max/IP, s⁻¹</td>
<td>154 ± 10</td>
<td>121 ± 6 ‡</td>
<td>77 ± 6 ††</td>
</tr>
</tbody>
</table>

Values are means ± SE. LVSP, left ventricular systolic pressure; LVEDP, left ventricular end diastolic pressure; LVDp/dt_max, maximal value of the first derivative of the left ventricular pressure; IP, instantaneous left ventricular pressure. †CHF significantly different from Sham (P < 0.01); ‡CHF significantly different from MIaf (P < 0.01); ††Sham significantly different from MIaf (P < 0.01).

Echocardiographic method. Echocardiographic char-
acteristics of the postinfarction model in rats have been
described by others recent years (3, 6, 15); however,
these studies used equipment with technical limita-
tions. The depth of interest is ~20 mm in rats, and this
distance is too short to allow ordinary cardiac trans-
ducers to focus on the relevant area. To achieve a
suitable working distance, previous investigators used
high frequency vessel transducers. However, the frame
rate has been too slow to truly sample systolic and
diastolic frames in 2D (3, 15). Other investigators
studying rat models of failure have used intravascular
transducers in the thoracic cavity, which give high
spatial resolution (8, 16, 27). These transducers have
limited ability to switch orientation; they do not have
Doppler; the frame rate is low; they cannot be used in
chronic studies with multiple measuring points; and
they may interfere with normal blood flow by compres-
sion of the intrathoracic content. The fully digitized
echocardiography used in the present study had its
software modified to allow recordings in 2D at ~200
Hz. This allows sampling of true diastolic and systolic
data. The researcher has to justify an interpretation of
the clustering, which, in this case, seems to be MI or
not (clusters Sham and MI), and CHF or not (clusters
CHF and MIaf).
frames. In addition, software modifications and the use of a 10 MHz, specially designed vessel transducer, allowed for a short focus distance (10–25 mm). These improvements allowed high quality recordings in 2D. The analysis method was also improved. Our sampling system is fully digitized, a fact that allows offline data analysis and the opportunity to adjust the gain and time scale to optimize resolution. The software allows almost all available options for online analysis on the scanner. Adjustment of gain and time scale during post hoc analysis may reduce reproducibility compared with conventional analysis of printouts; however, we have shown that reproducibility is within acceptable limits.

In the present study, the E and A waves are fused in the mitral inflow signal, and this contrasts with previous reports that show clear E and A wave separation (15). The separation of the E and A waves is heart rate-dependent, and, in the rat, the waves fuse at frequencies above 320–340 bpm (I. Sjaastad, T. Skomedal, J. B. Osnes, and O. M. Sejersted; unpublished data). We sedated the rats with drugs that did not reduce heart rate. Consequently, the heart rates were 320–360 bpm, and the mitral waves were mostly fused. In studies that used cardiodepressant drugs, heart rate was slowed, and the E and A waves could be separated (15).

Transition from MI to CHF. The understanding of CHF development has gradually changed over the years (18), but most investigators still agree that there is a transition from asymptomatic compensated failure to decompensated symptomatic failure. However, there has been some confusing use of the term heart failure in the literature, and some investigators do not distinguish between heart failure and myocardial failure, as defined by Braunwald (5). Acute infarction, in which the remaining myocardium has normal function but is unable to provide sufficient blood supply to the periphery, should also be categorized as heart failure. On the other hand, in myocardial failure, the loss of myocardium is not, initially, substantial enough to reduce the pumping capacity of the left ventricle markedly. However, the function of the viable myocardium gradually deteriorates, which may explain the transition to decompensated failure (10). In human myocardial failure, the myocardium show delayed time to peak force (11, 23) and, usually, slower relaxation velocity (11, 23). Reduced β-adrenergic response (28), altered sarcoplasmic reticulum function, and defective sarcoplasmic reticulum coupling (9) are possible mechanisms that could explain this myocardial dysfunction. Similar phenotypic transformations occur in rats (12); however, it may be difficult to assess the degree of failure in rats with MI. Large transmural infarctions often cause LV dilatation, but our data suggest that dilatation was not always combined with failure, as the left atrium sometimes had normal dimensions and PWSV and LVEDP often remained within the normal range. The left atrium dilates at a later stage than the left ventricle (5). This indicates that dilatation of the left atrium represents one further step towards decompensated failure. In accordance with this, reduced PWSV and high LVEDP were highly correlated with left atrial dilatation.

Criteria of CHF in the rat postinfarction model. There is no general consensus on criteria of CHF in animal models, except for the clinical characteristics of CHF such as pleural effusion, ascites, and tachypnea (2, 25). However, hemodynamic and echocardiographic criteria are also required to settle the diagnosis (2, 25). In our CHF cluster, the animals had the clinical characteristics of CHF, the echocardiographic data indicated CHF, and the animals had LVEDP ≥ 15 mmHg. The Sham cluster did not present any of these criteria. The animals in the MIaf cluster did not, with one exception, have the clinical characteristics of CHF, but only two of the echocardiographic/hemodynamic parameters did not overlap at all between the two MI clusters.

The first parameter, PWSV, was reduced in CHF, in accordance with our hypothesis. We suggest that this parameter may be used as a criterion of postinfarction CHF in small animal models when the basic clinical criteria and echocardiographic criteria of scarring and

Figure 7. Maximal positive first derivative of the left ventricular pressure divided by the instantaneous LV pressure \((\text{LVdP/d}_{max}/\text{IP})\) in the three clusters. A: scatter plot of \(\text{LVdP/d}_{max}/\text{IP}\) in the three clusters. B: relationship between \(\text{LVdP/d}_{max}/\text{IP}\) and PWSV. *\(P < 0.01\).

Table 6. LVEDP in the clusters

<table>
<thead>
<tr>
<th></th>
<th>Sham</th>
<th>MIaf</th>
<th>CHF</th>
</tr>
</thead>
<tbody>
<tr>
<td>LVEDP ≥ 15 mmHg</td>
<td>0</td>
<td>0</td>
<td>16</td>
</tr>
<tr>
<td>LVEDP &lt; 15 mmHg</td>
<td>14</td>
<td>10</td>
<td>0</td>
</tr>
</tbody>
</table>

No. of animals in each cluster grouped by LVEDP values.
LV and left atrial dilatation are fulfilled. This variable can be obtained in chronic studies with repeated measurements.

The second parameter, which could also completely separate the two MI clusters, was LVEDP (Table 6). LVEDP varied between 0 and 10 mmHg in our Sham rats, which is in accordance with reported normal values (12, 15). There are several factors that can explain the variation in LVEDP, e.g., catheter position, anesthesia depth, respiration, and quality of the catheter. A high LVEDP is associated with pathology (5), and elevation of LVEDP ≥ 15 mmHg is regarded as pathologic and indicating CHF. In the present study, we found LVEDP ≥ 15 mmHg in all animals in the CHF cluster and < 15 mmHg in the MI_{nf} cluster. This indicates that LVEDP, in most cases, is a usable criterion for CHF, even when echocardiography is not performed. However, the basic criteria have to be met.

LVEDP/dt_{max}/IP is another hemodynamic parameter that might distinguish MI_{nf} from CHF. However, this parameter directly reflects global LV contractility (19, 30) rather than contractility in the viable myocardium. We found a reduced LVEDP/dt_{max}/IP in CHF, and there was no overlap with Sham. It is not possible, however, to separate MI_{nf} from CHF and Sham using LVEDP/dt_{max}/IP. For this reason, it is not feasible to use this parameter as a main criterion of CHF.

Limitations of the study. The present study was performed under specific experimental conditions, including degree of sedation, age of rats, and time from induction of infarction. The results are valid and reliable, but other investigators should perform echocardiographic studies of their model to set reliable and valid criteria of CHF. However, we suggest that PWSV may be the best parameter to discriminate CHF from MI_{nf}.

In conclusion, highframe rate digital echocardiography allows high quality 2D, M-mode, and Doppler recordings in rats. The time resolution made it feasible to find true systolic and diastolic frames in 2D, and to thereby measure dimensions in both 2D and M-mode. A J-tree cluster analysis separated the rats into one Sham cluster and two MI clusters. One of the MI clusters had clinical characteristics of CHF, a reduced PWSV clearly distinguished this MI cluster from the other clusters, and all animals in this cluster had LVEDP ≥ 15 mmHg. The data indicate that CHF in the rat postinfarction model is associated with myocardial dysfunction. However, MI rats without clinical signs of CHF do not have severely impaired myocardial function, as evaluated by echocardiography. The present study is, to our knowledge, the first to explore the difference in echocardiographic recordings between failing and nonfailing postinfarction rats. The data suggest that it is possible to assess myocardial function in longitudinal rat studies using the echocardiographic variables PWSV and LAD. Further studies are required to describe the development of CHF in the postinfarction period, as evaluated by PWSV, LAD, and other echocardiographic parameters.

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