Layer-specific strain analysis by speckle tracking echocardiography reveals differences in left ventricular function between rats and humans

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Bachner-Hinenzon N, Ertracht O, Leitman M, Vered Z, Shimoni S, Beeri R, Binah O, Adam D. Layer-specific strain analysis by speckle tracking echocardiography reveals differences in left ventricular function between rats and humans. Am J Physiol Heart Circ Physiol 299: H664–H672, 2010. First published July 2, 2010; doi:10.1152/ajpheart.00017.2010.—The rat heart is commonly used as an experimental model of the human heart in both health and disease states, assuming that heart function of rats and humans is alike. When studying a rat model, echocardiography is usually performed on sedated rats, whereas standard echocardiography on adult humans does not require any sedation. Since echocardiography results of sedated rats are usually inferred to alert humans, in the present study, we tested the hypothesis that differences in left ventricular (LV) function may be present between rats sedated by a low dose of ketamine-xylazine and alert humans. Echocardiography was applied to 110 healthy sedated rats and 120 healthy alert humans. Strain parameters were calculated from the scans using a layer-specific speckle tracking echocardiography program. The results showed that layer longitudinal strain is equal in rats and humans, whereas segmental strain is heterogeneous (P < 0.05) in a different way in rats and humans (P < 0.05). Furthermore, layer circumferential strain is larger in humans (P < 0.001), and the segmental results showed different segmental heterogeneity in rats and humans (P < 0.05). Radial strain was found to be homogeneous at the apex and papillary muscle levels in humans and heterogeneous in rats (P < 0.001). Additionally, whereas LV twist was equal in rats and humans, in rats the rotation was larger at the apex (P < 0.01) and smaller at the base (P < 0.001). The torsion-to-shortening ratio parameter, which indicates the transmural distribution of contractile myofibers, was found to be equal in rats and humans. Thus, when evaluating LV function of sedated rats under ketamine-xylazine, it is recommended to measure the global longitudinal strain, LV twist, and torsion-to-shortening ratio, since no scaling is required when converting these parameters and inferring them to humans.

RATS PLAY AN IMPORTANT ROLE in modeling human left ventricular (LV) function, and thus a variety of pathologies, such as myocardial infarction (20, 30) and cardiomyopathy (17, 19), are commonly induced in rats, so that their effect on LV function can be analyzed under controlled conditions. Consequently, the pertinent questions are 1) does the similarity in LV structure between rats and humans (4, 10) result in similar LV function? and 2) is the rat LV an appropriate model for studying human LV function? To evaluate the properties that quantify LV function, a method known as speckle tracking echocardiography (STE) has been recently developed (1, 3, 9, 11, 13, 14, 18, 20, 23). The STE method uses standard two-dimensional (2-D) echocardiography cines and tracks the speckles in the ultrasound B-mode image sequence of the myocardium, and, subsequently, the parameters of local function are calculated (22). STE analysis is independent of insonation angle, and, therefore, the STE method is superior to tissue-Doppler echocardiography. The STE method has several advantages over tagged-MRI, such as lower cost and high availability, which allow routine usage in the clinic. This method is also advantageous in research, since it allows large cohort size to be acquired with a reasonable effort. The STE method was validated by comparing its measures with those assessed by different modalities such as tagged-MRI and sonomicrometry (3, 9, 11, 13, 18, 23). Furthermore, the relation between fibrosis (histology) and segmental strain analysis demonstrated that the STE method can detect areas that undergo the fibrosis process even in small animals, such as rats (20). Recently, the spatial resolution of the STE method was enhanced to perform layer-specific analysis of LV function (LS-STE) (7), and this layer-specific analysis was used for the human LV (1, 14).

Since the echocardiography results of sedated rats are usually inferred to alert humans, in the present study, we tested the hypothesis that differences in LV function may be present between rats sedated by a low dose of ketamine-xylazine and alert humans. To test this hypothesis, the following commonly used properties that quantify LV function were obtained from healthy humans and rats: longitudinal strain, circumferential strain, radial strain, LV rotation, net ventricular twist angle, and LV torsion. Furthermore, the torsion-to-shortening ratio (TSR) parameter was calculated for both rats and humans. TSR indicates the transmural distribution of contractile myofibers and their function (2, 5, 29). For example, it has been found that TSR is different for aortic stenosis patients than for normal subjects due to the change in the transmural distribution of the myocardial fibers resulting from a remodeling process (29). Since the transmural distribution of contractile myofibers is similar in rats and humans (4, 10), this parameter was expected to be equal in these species. In this study, the strain parameters were calculated by a layer-specific STE program at three layers and six segments, and, to our knowledge, this is the first time...
that such a high spatial resolution strain analysis has been performed on rats and humans.

METHODS

Animal experiments were approved by the Animal Ethics Committee and conducted according to institutional animal ethical committee guidelines (ethics number IL-101-10-2007). Approval of human experiments was obtained from the Assaf Harofeh Medical Center Institutional Review Board of Human Studies (NIH number 581944) and the Kaplan Medical Center Institutional Review Board of Human Studies (NIH number IS-0007-08-KMC). Participants gave written, informed consent.

Animal experiments. In this study, 110 adult male Sprague-Dawley rats (weight: 300 ± 28 g; mean ± SD) were investigated as a part of another study. The echocardiograms in this study were baseline ultrasound cines of rats before surgery. The conventional echocardiogram, calculated ejection fraction, and LV dimensions of all rats were normal.

Rat echocardiography protocol. Rats were sedated by an intraperitoneal injection of a subanaesthetic dose of a 29 mg/kg ketamine-4.3 mg/kg xylazine mixture, and the chest was shaved. Rats were placed in left lateral decubitus position and scanned by a commercially available echocamcer (Vivid i ultrasound cardiovascular system, GE Healthcare, Haifa, Israel) using a 10S phased array pediatric transducer and a cardiac application with high temporal and spatial resolutions. The transmit frequency was 10 MHz, the depth was 2.5 cm, and the frame rate was 225 frames/s. The standard 2D echocardiography experiment included scanning of one parasternal long-axis view, which contained the inferior and anterior walls in rats, and three short-axis levels: basal at the mitral valve (MV), midventricle at the papillary muscle tips (PM), and apical (AP) cross sections. Of the 110 rats, 109 AP short-axis cines, 109 PM short-axis cines, and 103 MV short-axis cines were adequately tracked and analyzed, as determined by the tracking quality measurement performed by the LS-STE program. The parasternal long-axis view was acquired in 80 rats, and all 80 cines were trackable by the LS-STE program. Apical long-axis scans were impossible to obtain, since the tilting angle that was required caused a part of the probe to detach from the rats’ chest.

Human experiments. This study included 120 healthy adult human subjects (age: 47.5 ± 18.6 yr; mean ± SD). Subjects had no history of cardiac diseases, valvular disease, or hypertension. All subjects were diagnosed as normal by a cardiologist, who performed a physical examination. Their electrocardiogram and standard 2-D echocardiography examinations were normal.

Human echocardiography protocol. Subjects underwent standard 2-D echocardiography with the Vivid 7 dimension system (101 subjects) or Vivid i system (19 subjects) (GE Healthcare) using a 3S phased array probe and a cardiac application. The transmission frequency was 1.7–3.5 MHz, and the frame rate was 40–70 frames/s. Experiments were performed during rest, with the subjects in the left lateral decubitus position. Three parasternal short-axis scans were acquired: basal (MV), midventricle (PM), and AP cross sections (scanning as closest to the apex as possible while still obtaining an adequate image quality). Moreover, standard long-axis scans were obtained, which consisted of a parasternal long-axis scan and three AP long-axis scans. However, only the two-chamber long-axis scans were included in this study and compared with the parasternal long-axis images of rats. The two-chamber view of humans is anatomically the same as the parasternal long-axis view of rats and contains the anterior and inferior walls. Despite the fact that the spatial resolution in parasternal and AP views is different during acquisition, the longitudinal strains in both views are similar when looking at anatomically identical cross sections (see the Appendix). Subsequently, of the 120 subjects, 96 AP short-axis cines, 108 PM short-axis cines, 99 MV short-axis cines, and 84 two-chamber AP long-axis cines were adequately tracked and analyzed, as determined by the tracking quality measurement of the LS-STE program.

Morphological characteristics and global function. LV dimensions, which included outer diameter, length, and wall thickness, were calculated at the end-diastolic phase. The outer diameter and wall thickness were calculated at the PM level from 2-D derived M-mode echocardiograms as recommended (16, 25). LV radii were calculated simply from the diameter. LV wall thickness was calculated at the posterior wall. LV length was calculated from the long-axis B-mode image by the caliper tool. Thereafter, the wall thickness-to-radius ratio and radius-to-length ratio were calculated. The global function estimation included fractional shortening calculation at the PM level by the Teichholz formula (16) and ejection fraction calculation at the parasternal long-axis view for the rats and at the two-chamber AP long-axis view for humans using a single-plane Simpson equation. Heart rate was calculated from the R-R interval of the ECG for both rats and humans.

Speckle tracking echocardiography. The ultrasound cines were postprocessed by the LS-STE program as previously described (7). The program uses a commercial STE program called “2D-strain” (EchoPAC Dimension 98, GE Healthcare). The 2-D strain program tracks the movement of strong reflectors, which are observed in the B-mode images frame by frame. The user marks the endocardium and chooses the width of the myocardium. The program imposes a grid of points on the assigned region, tracks the points, and evaluates their velocities at each frame (22). The velocities, instead of being processed by the built-in smoothing of the commercial program, are denoised by a three-dimensional (3-D) wavelet representation (MATLAB software, MathWorks). Thereby, the spatial resolution of the calculated functional measurements is increased, and, instead of averaging across the myocardium, the values are calculated for three myocardial layers (endocardium, midwall, and epicardium), as shown in Fig. 1. Subsequently, the longitudinal strain, circumferential strain, LV rotation, and net ventricular twist were evaluated at three myocardial layers and six myocardial segments, and the radial strain was evaluated at six myocardial segments. The parameters were evaluated from the locations and velocities of each point on the grid. Longitudinal strain was calculated from the longitudinal velocities and locations of the points on the grid of the long-axis cross section as described by Rappaport et al. (22), and circumferential strain was calculated by the same method from the circumferential velocities and locations of the points on the grid of the short-axis cross section. Radial strain was calculated from the radial myocardial velocities and locations of the points on the grid of the short-axis cross section. LV rotation was evaluated for each layer in degrees from the diastolic state, relative to the center of mass of the short-axis cross section, as described by Notomi et al. (18). The net ventricular twist was defined and measured as the relative rotation between the base and apex. LV torsion was calculated as follows:

\[
T = \frac{\alpha_{\text{twist}}}{L} \times R_{\text{PM}}
\]

where \(L\) is LV torsion, \(\alpha_{\text{twist}}\) is the net ventricular twist (in °), \(L\) is LV length, and \(R_{\text{PM}}\) is the outer radius of the LV in the short axis at the PM level.

TSR was calculated as follows:

\[
\text{TSR} = \frac{\Delta T}{S_{\text{endo}}}
\]

where \(\Delta T\) is the torsion relatively to the diastolic state (in radians) and \(S_{\text{endo}}\) is the endocardial circumferential strain (average over 3 short-axis levels). End diastole was defined by the ECG as the time immediately before the QRS complex, coinciding with the largest diastolic LV diameter.

Statistical analysis. All values are presented as means ± SE. To compare the strain parameters at the different segments/layers of the two groups, two-way ANOVA for unequal variance was used. To determine whether a parameter was heterogeneous or homogeneous
among the different segments, one-way ANOVA for unequal variance was used. To compare the global strain parameters, Student’s \( t \)-test with two distribution tails was used. \( P \) values of \(< 0.05\) were considered statistically significant.

**RESULTS**

**Morphological characteristics and global function.** The morphological characteristics and global function of rats and humans are shown in Table 1. As expected, the dimensions (length, diameter, and wall thickness) of the human LV were larger than those of rats \((P < 0.001)\). The wall thickness-to-radius ratio was equal in rats and humans \((0.30 \pm 0.003)\); however, the radius-to-length ratio was larger for rats \((humans: 0.39 \pm 0.004 and rats: 0.43 \pm 0.003, P < 0.001)\). The global function results showed that the heart rate was higher in rats than in humans \((P < 0.001)\), whereas ejection fraction and fractional shortening were similar in rats and humans at the PM level.

**Longitudinal strain.** The longitudinal strain parameter at the endocardium and midwall was found to be the same for rats and humans \((Fig. 2A)\), and the epicardial longitudinal strain parameter was found to be larger in rats \((P < 0.05)\). Longitudinal strain values decreased from the endocardium toward the epicardium in both rats and humans and by the same extent (rats:

<table>
<thead>
<tr>
<th></th>
<th>Outer Diameter, mm</th>
<th>Length, mm</th>
<th>Posterior Wall Thickness, mm</th>
<th>Wall Thickness-to-Radius Ratio</th>
<th>Radius-to-Length Ratio</th>
<th>Ejection Fraction, %</th>
<th>Fractional Shortening, %</th>
<th>Heart Rate, beats/min</th>
</tr>
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<tbody>
<tr>
<td>Rats</td>
<td>10.3 ± 0.06</td>
<td>12.2 ± 0.1</td>
<td>1.5 ± 0.02</td>
<td>0.30 ± 0.003</td>
<td>0.43 ± 0.003</td>
<td>59 ± 0.6</td>
<td>41 ± 0.5</td>
<td>274 ± 2.7</td>
</tr>
<tr>
<td>Humans</td>
<td>64.3 ± 0.45</td>
<td>83.1 ± 0.68</td>
<td>9.8 ± 0.09</td>
<td>0.31 ± 0.003</td>
<td>0.39 ± 0.004</td>
<td>61 ± 0.7</td>
<td>40 ± 0.5</td>
<td>69 ± 1.1</td>
</tr>
<tr>
<td>( P ) value</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>NS</td>
<td>&lt;0.001</td>
<td>NS</td>
<td>NS</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Values are means ± SE; \( n = 110 \) rats and 120 humans. LV, left ventricular; NS, not significant.
The horizontal lines indicate nonsignificant cardiac segments [basal (baso), mid, and apical inferior segments and apical, mid, and basal anterior segments]. The interspecies differences found to be significant in all myocardial layers [endocardium (Endo), midwall (Mid), and epicardium (Epi)]. The interspecies differences found to be significant in all segments (P < 0.05). The horizontal lines indicate nonsignificant differences between the indicated segments (P > 0.05). The interspecies difference was found to be significant in all segments (P < 0.05) except for the basal inferior segment (indicated by the horizontal double-headed arrow).

endocardium: -21.8 ± 0.7%, midwall 15.5 ± 0.3%, and epicardium: -11.8 ± 0.6%; and humans: endocardium: -21.8 ± 0.8%, midwall: -15.7 ± 0.5%, and epicardium: -10.2 ± 0.6%). On the other hand, the segmental results showed a significant difference between the longitudinal strains of rats and humans (P < 0.05). Longitudinal strain was homogeneous for both rats and humans (P < 0.05), whereas it was distributed differently between the segments, as shown in Fig. 2B, except for the basoinferior wall, which showed the same longitudinal strain in rats and humans.

Circumferential strain. Global circumferential strain (averaged over the 3 cross-section levels) was found to be larger in humans than in rats [humans (n = 82): -22.0 ± 0.4% and rats (n = 103): -20.0 ± 0.3%, P < 0.001]. Circumferential strain values decreased from the endocardial layer toward the epicardial layer (P < 0.001) in both rats and humans at all LV levels (Fig. 3A).

The human segmental results showed homogeneity of the circumferential strain at the AP level and heterogeneity at the PM level (P < 0.001) and MV level (P < 0.001). Conversely, the rat segmental results showed heterogeneity of the circumferential strain at all three myocardial levels (P < 0.001). Detailed segmental results are shown in Fig. 3B. The AP results showed that the inferior and anterior septum of rats show smaller circumferential strain than the free wall (P < 0.05), whereas in humans the inferior septum shows higher circumferential strain than the anterior, lateral, and posterior walls (P < 0.05), and the circumferential strain of the anterior septum is not different from the circumferential strain at the free wall. Interspecies results at the apex showed similar segmental peak systolic circumferential strain at the posterior and inferior walls and a difference at all other segments (P < 0.05). The PM level results showed that in rats the anterior septum demonstrates smaller circumferential strain than the inferior septum and anterior and inferior walls (P < 0.05); in humans, the anterior wall demonstrates smaller circumferential strain than all other segments at the PM level. Interspecies results at the PM level showed similar segmental peak systolic circumferential strain at the lateral and inferior walls and a difference at all other segments (P < 0.05). Interspecies results at the MV level showed a difference between peak systolic circumferential strains at all segments (P < 0.05).

Radial strain. Radial strain was found to be homogeneous in humans at the AP and PM levels and heterogeneous at the MV level (P < 0.05), whereas rats demonstrated heterogeneity of the radial strain at all LV levels (P < 0.001). Radial strain results are shown in Fig. 4. At the apex, rats showed smaller peak systolic radial strain at the inferior septum than at the anterior and lateral walls. Moreover, rats showed smaller AP peak systolic radial strain at the anterior septum than at the anterior lateral and posterior walls. In humans, the AP peak systolic radial strain was smaller at the anterior septum than at the anterior and inferior walls. Interspecies results at the AP level showed similar segmental peak systolic radial strain at the lateral and posterior walls and a difference at all other segments (P < 0.05). In humans, the posterior wall at the PM level showed larger peak systolic radial strain than all other segments. Interspecies results at the PM level showed similar segmental peak systolic radial strain at the lateral and anterior walls and a difference at all other segments (P < 0.05). At the MV level, rats showed smaller peak systolic radial strain at the inferior septum than at the anterior, lateral, and posterior walls (P < 0.05). Moreover, rats showed smaller values of peak systolic radial strain at the inferior wall than at the anterior and
lateral walls ($P < 0.05$). At this level, rats showed larger values of peak systolic radial strain at the lateral wall than at the posterior, inferior, inferior septum, and anterior septum walls ($P < 0.05$). In humans, the lateral wall showed larger peak systolic radial strain than the inferior and anterior septum at the MV level ($P < 0.05$). Furthermore, the posterior wall in humans showed larger values than the anterior septum, inferior septum, and inferior wall. Interspecies results at the MV level showed a difference between peak systolic radial strains at all segments ($P < 0.05$).

LV rotation, net ventricular twist, torsion, and TSR. Rotation was defined in degrees, as measured from the end-diastolic state. Generally, rotation was significantly larger at the endocardium and decreased toward the epicardium for both LV AP and basal levels in both rats and humans ($P < 0.001$; Fig. 5).
As shown in Fig. 5, the apex rotated counterclockwise in both rats (6.6° ± 0.3°) and humans (5.3° ± 0.3°); however, the global rotation at the apex was smaller in humans than in rats ($P < 0.001$; Fig. 5). Subsequently, the net ventricular twist was equal in humans and rats (9.7° ± 0.5° and 8.7° ± 0.4°, respectively). The PM level served as an equatorial plane in humans (Fig. 5), which almost did not rotate (−0.3° ± 0.3°). However, in rats, the rotation at the PM level was counterclockwise in 82 of 109 rats (average for all rats: 2.3° ± 0.4°). LV torsion was found to be equal in rats and humans (humans: 3.8° ± 0.2° and rats: 3.7° ± 0.2°). Finally, TSR was equal in rats and humans as well (humans: 0.41° ± 0.02° and rats: 0.43° ± 0.02°).

**DISCUSSION**

In this study, we tested the hypothesis that differences in LV function may be present between rats sedated by a low dose of...
ketamine-xylazine and alert humans, since the echocardiography results of sedated rats are usually inferred to alert humans. LV function properties were calculated using a novel LS-STE approach, which allowed the high spatial resolution of three myocardial layers and six myocardial segments. The results demonstrate the heterogeneity of contractile function of the LV in alert humans and in rats sedated with a low dose of ketamine-xylazine. Our results show that 1) peak systolic longitudinal strain is equal in sedated rats and alert humans at the endocardium and midventricular layer; however, it is different at the myocardial segments; 2) peak systolic global circumferential strain is larger in alert humans than in sedated rats at the basal and midventricular levels, whereas it is equal at the AP level; 3) segmental peak systolic circumferential strain is different between sedated rats and alert human at most segments; 4) radial strain was found to be homogeneous in humans at the AP and PM levels and heterogeneous at the MV level, whereas rats demonstrated heterogeneity of the radial strain at all LV levels; 5) segmental peak systolic radial strain is different between sedated rats and alert human at most segments; 6) net ventricular twist is the same in rats and humans; however, in rats, the AP rotation is larger and the basal rotation is smaller than in humans; and 7) LV torsion and TSR are equal in rats and humans. Since rats are increasingly becoming a major experimental model of human heart pathologies, it is important to carefully infer LV contractile function of rats to that of humans.

LV dimensions. In addition to the obvious finding that the LV dimensions (length, diameter, and wall thickness) of rats are smaller than those of humans, we found that the wall thickness-to-radius ratio is equal in rats and humans and the radius-to-length ratio is significantly smaller in humans ($P < 0.001$), as shown in Table 1. The radius-to-length ratio results demonstrate that the rat LV is more spherical than that of humans. A previous tagged-MRI study by Liu et al. (15) also reported that the radius-to-length ratio is smaller in humans; however, these authors found that the wall thickness-to-radius ratio is smaller in humans. The difference of the wall thickness-to-radius ratio between the two studies may be explained by the larger wall thickness in humans (9.8 ± 0.1 mm) and smaller wall thickness in rats (1.5 ± 0.02 mm) in this study compared with Liu et al. (15). However, our wall thickness results are in agreement with previously reported values (27, 31).

Longitudinal, circumferential, and radial strains. The results of this study show that global peak systolic longitudinal strain is equal in rats and humans, whereas global peak systolic circumferential strain differs. This result confirms that global longitudinal strain is not affected by heart size, whereas global circumferential strain is affected by its size and morphology (radius-to-length ratio). This interesting result contradicts a previous report (21) showing that for small animals, the contraction along the long axis contributes less to cardiac function. Therefore, it can be concluded that although the longitudinal displacement of the base is scaled allometrically to heart size (21), global longitudinal strain is the same and no scaling is needed.

Circumferential strain is equal in rats and humans at the AP level; however, it becomes different when moving toward the base, as shown in Fig. 3A (PM level: $P < 0.05$ and MV level: $P < 0.001$). These results are in agreement with the tagged-MRI study of Liu et al. (15). The circumferential strain results for the different levels, compared with the tagged-MRI results of Liu et al. (15), are shown in Table 2. The divergence in circumferential strain between rats and humans occurs possibly due to the difference in morphology. The fact that the rat heart is more spherical than that of humans affects circumferential strain at the basal level more than circumferential strain at the midventricular level, whereas the AP level is not affected by it. A possible explanation is that according to Laplace’s law, the stress in the myocardium is larger when the radius is larger. The rat heart is more spherical than the human heart, and thus there is more stress at the basal level.

The segmental results show different distributions of longitudinal, circumferential, and radial strains in rats and humans, as shown in Figs. 2B, 3B, and 4, respectively ($P < 0.05$). Such a segmental comparison of strain parameters between rats and humans has not been previously reported.

LV rotation, net ventricular twist, torsion, and TSR. The contraction of oblique myofibers that are present in the subendocardium and subepicardium causes the apex and base to rotate in counter directions, i.e., net ventricular twist (5). The net ventricular twist is created by different torques in the myocardial layers, and the balance between the torques produces torsion (5, 6). This balance was defined by Arts et al. (5) as homogeneous shortening of all myofibers during the ejection phase. This balance is interrupted and transluminal inhomogeneity of the myofibers’ shortening arises when a dysfunction occurs, e.g., aortic stenosis (29). At present, this inhomogeneity can be quantified by the ratio of LV torsion to endocardial circumferential shortening (TSR) (2, 5, 29). Thus, the functional property quantified by TSR is strongly dependent on LV structure and tissue viability. The fact that the fiber orientation is the same in rats and humans (4, 10) may explain the similarity in TSR between them. It is vital to mention that the definition of LV torsion in this study was net ventricular twist per unit of length multiplied by the LV radius at the PM level.

Table 2. Global circumferential strain and global LV rotation measured by tagged-MRI and LS-STE

<table>
<thead>
<tr>
<th></th>
<th>Circumferential Strain, %</th>
<th>Rotation, °</th>
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<tbody>
<tr>
<td></td>
<td>Apex</td>
<td>Papillary muscle</td>
</tr>
<tr>
<td>Tagged-MRI</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Humans</td>
<td>$-22 \pm 1.2$ ($n = 6$)</td>
<td>$-21 \pm 0.8$ ($n = 6$)</td>
</tr>
<tr>
<td>Rats</td>
<td>$-21 \pm 0.4$ ($n = 6$)</td>
<td>$-19 \pm 0.4$ ($n = 6$)</td>
</tr>
<tr>
<td>LS-STE</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Humans</td>
<td>$-24 \pm 0.5$ ($n = 96$)</td>
<td>$-22 \pm 0.5$ ($n = 108$)</td>
</tr>
<tr>
<td>Rats</td>
<td>$-24 \pm 0.5$ ($n = 109$)</td>
<td>$-21 \pm 0.4$ ($n = 109$)</td>
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</tbody>
</table>

Values are mean ± SE. Tagged-MRI results are from the study of Liu et al. (15). LS-STE, layer-specific analysis of LV function with speckle tracking echocardiography.
as recommended by Arts et al. (5). This LV torsion was found to be equal in rats and humans. However, different studies have provided various definitions of LV torsion, and thus one must be careful when comparing these results with other results. For example, Liu et al. (15) found that torsion is larger in rats than in humans, and Henson et al. (12) found that torsion is equal in mice and humans; however, they defined LV torsion as a net ventricular twist per unit of length without taking into consideration the difference in LV radii between rats and humans. Their definition is correct only if the LV of rats and humans are cylinders with the same radii but with different lengths, but actually the LV radii are different between rats and humans.

The similarity of the net ventricular twist angle between rodents and humans is controversial. Previous MRI studies (15, 32) have shown similarity, whereas one study (12) showed a significant difference in the net ventricular twist angle. Our results demonstrate that the net ventricular twist is similar; however, the AP and basal rotations, each by itself, are significantly different in rats and humans (Fig. 5). In rats, the AP rotation is larger (rats: 6.6 ± 0.3° and humans: 5.3 ± 0.3°) and the basal rotation is smaller than in humans (rats: −2.2 ± 0.3° and humans: −4.4 ± 0.3°). The equatorial plane, which is the short-axis cross-section level that hardly rotates, is different for rats and humans. For humans, the equatorial plane is at the PM level (Fig. 5). For rats, the rotation at the PM level is counterclockwise, which is the same direction of rotation of the apex (Fig. 5). The equatorial plane for rats probably exists between the PM level and basal level. A previous comparative tagged-MRI study by Liu et al. (15) reported the same AP and basal rotations for rats and humans, which are different from the results presented here. A possible explanation for that might be the subjects’ position during the scan. In our study, humans and rats were in the left lateral decubitus position, whereas in the MRI study, they were placed in the prone position. It is important to mention that the cohort size in the present study was much larger than the cohort size in the tagged-MRI study [n = 6 rats and 6 humans in Liu et al. (15) vs. n = 110 rats and 120 humans in this study], and thus the results are probably more reliable in this study. Nonetheless, the rotation values reported by Liu et al. (rats: 9.0 ± 0.9° and humans: 9.7 ± 1.2°) were larger than the values measured by the LS-STE method (rats: 6.6 ± 0.3° and humans: 5.3 ± 0.3°). A possible explanation for these differences is that when using tagged-MRI it is possible to obtain the AP short axis very near to the apex, whereas with ultrasound it is possible to view the apex only at a higher cross section, due to the imaging limitations imposed by the ribs, which cause attenuation of the ultrasound beam (1).

Study limitations. In the present study, we compared LV function of alert humans with that of sedated rats. Indeed, the anesthetic mixture of ketamine-xylazine is known to reduce LV heart rate and contractility in anesthetic doses (24, 28); however, the amount of ketamine-xylazine used in this study was subanaesthetic and rather small (29 mg/kg ketamine and 4.3 mg/kg xylazine). The heart rate and fractional shortening of the rats in this study were lower than the values reported in conscious rats (28); however, it is important to mention that conscious rats that undergo echocardiography are in a state of anxiety, and their LV function is probably very different from that of conscious, relaxed rats.

The results of this study were compared with the results of Liu et al. (15), and we found that the heart rate and circumferential strain results were similar in both studies, although Liu et al. used isoflurane as the anesthetic material. This comparison is valid since in rats the effects of ketamine-xylazine and isoflurane are similar on both heart rate and fractional shortening (28).

Another limitation is that in the present study we used 2-D and not 3-D echocardiography. Accordingly, out-of-plane motion may have occurred, and this could have affected the measurements (26). Yet, only images in which the tracking was appropriate were used. Furthermore, due to this limitation, torsion was approximated differently from the MRI study of Aelen et al. (2). Aelen et al. used the real distance between the AP and basal short-axis levels; however, it is not possible to assess this value from 2-D echocardiography, and thus total LV length was used instead. Moreover, Aelen et al. used the average outer radius of the AP and basal cross sections. Since it was difficult to detect the epicardium in the basal short axis of the rats, the outer radius of the LV at the PM short-axis level was used instead of averaging over the radii measured at the AP and basal levels.

Conclusions. In conclusion, the similarity in the orientation of the contractile myofibers between rats and humans is reflected in the TSR parameter. Moreover, global longitudinal strain, net ventricular twist angle, LV torsion, and AP circumferential strain are equal in rats and humans, and thus no scaling is needed while comparing these parameters among rats and humans. It is recommended that these parameters should be measured while evaluating LV function of a rat model of human heart disease, as they appear to accurately reflect human cardiac function.

APPENDIX

In this study, longitudinal strain was measured from different scan views in rats and humans. In rats, longitudinal strain was measured from the parasternal long axis, since AP scans were difficult to obtain. These images were obtained with a good tracking quality, as determined by the “2D-strain” commercial program (EchoPAC Dimension 08, GE Healthcare). The parasternal long axis of rats contains the inferior and anterior walls, which appear in human subjects at the two-chamber view. There is a problem in comparing the longitudinal strains when measured from these two views, as there are differences in the spatial resolution of the images. To test the effect of the different spatial resolution of the parasternal and AP scans, the five-chamber AP views and parasternal long-axis views of humans were analyzed, since anatomically they are alike. In both views, the anterior septum and posterior wall were scanned.

Forty parasternal long-axis scans with a good image quality were chosen from the human data. Longitudinal strain of the anterior septum in this view was compared with the one measured from the five-chamber view. A Bland-Altman test was done to compare the two measurements (9). The mean value of the difference was −0.83%, and the upper and lower limits of agreement were 5.9% and 7.5%, respectively. Agreement between the two longitudinal strain measurements was found in 39 of 40 subjects.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).
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