Determination of cardiac contractility in awake unsedated mice with a fluid-filled catheter

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During recent years, gene-manipulated mice have become the subject of rapidly growing interest in cardiovascular research (1, 2, 17, 20, 21, 23, 27, 35, 36). Quite naturally, this has created a strong need for accurate assessment of cardiovascular variables in mice. Whereas the accurate monitoring of blood pressure and heart rate (HR) in the awake mouse has been established quite rapidly in many laboratories, the determination of cardiac function presents a greater difficulty (34). This is all the more regrettable as several gene-manipulated mouse models of cardiac hypertrophy and cardiac failure have become available (5, 15, 22, 24, 35, 39). So far, cardiac contractility has been measured almost exclusively using the 1.4-Fr Millar catheter (5, 8–11, 15, 17, 39), but because of its stiffness this measuring system can only be used in anesthetized mice. Moreover, because of their fragility and cost, Millar catheters are not used as frequently as they could be in experimental conditions. This is relevant given the well-known effect of anesthesia on cardiac function in large mammals (13, 30, 38) and mice (8, 9, 18, 27).

The present study was undertaken to assess a custom-made, low-cost Pebax 03 (P03) catheter that can be used in awake mice to measure cardiac contractility. The results clearly demonstrate that under anesthesia the P03 catheter yields identical results to those obtained with the 1.4-Fr Millar system but, in contrast to the latter, the P03 catheter enabled us to measure cardiac contractility in awake mice. Our data also confirm that anesthesia profoundly alters the cardiac contractility of mice and show that mineralocorticoids decrease cardiac contractility in conscious mice independently of blood pressure.

METHODS

Characterization of the Pebax Catheter

The Pebax catheter (Figs. 1 and 2) was put together in the following way: 18 mm of Pebax tubing (Medical Extrusion Technology) was joined at the tip to 1 mm of silicone tubing [inner diameter (ID)/outer diameter (OD): 0.30/0.64 mm], which prevents the Pebax tubing from touching the ventricular wall during contraction (Fig. 1A). At the other end, the Pebax catheter was connected to a hydraulic pressure transport line consisting of 12 cm of polyethylene (PE)-50 tubing (SIMS Portex) joined to 7 cm of silicone tubing (ID/OD: 0.51/0.94 mm, Ulrich; St. Gallen, Switzerland). This allows the catheter to be exteriorized and attached to the back of the mouse. The line was connected to a pressure transducer (World Precision Instruments; Sarasota, FL) and a computerized data-acquisition system (Notocord HEM 3.1 software; Croissy sur Seine, France) (Fig. 2). The length of the whole Pebax catheter setup is only 25 cm. It is filled with 0.5 ml of degassed saline containing heparin (300 U/ml). Meticulous care has to be taken to ensure that the system is always free of air bubbles throughout the experimental period.

The natural frequency and damping coefficient of the Pebax catheter pressure transducer system were determined using a transient method (“balloon-pop test”) adapted from Gabe (6). To do so, a Pebax catheter was passed through a stopcock into a 12-cm Plexiglas cylinder (ID: 2.5 cm) filled with degassed saline, and the top of the cylinder was covered and held with a rubber band and membrane obtained from a surgical glove. The balloon inflated pressure was held at 65–75 mmHg. The membrane was then popped with a match, which caused the Pebax catheter pressure transducer system to oscillate at its dampened natural frequency, and these pressure changes in the catheter system were recorded by the Notocord HEM 3.1 software at a sampling rate of 10,000 Hz. Because of the diameter effect on the dynamic characteristics, two sizes of Pebax catheters were tested: a Pebax 02 (P02) catheter (ID/OD of Pebax tubing: 0.20/0.48 mm) and a P03 catheter (ID/OD of Pebax tubing: 0.30/0.48 mm).

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Fifty-seven male C57BL/6J mice (30 g) and ten male N5–6-129Ola/C57BL/6J backcrossed mice (30 g), obtained from the Institute of Pharmacology, University of Lausanne, Switzerland, treated with either DOCA-salt (1% NaCl) or tap water (37) were used in the present study.

Mice were anesthetized via inhalation of 1/2% halothane mixed with oxygen. The right carotid artery was exposed for a length of 5 mm. A 1.4-Fr Millar pressure catheter (Millar Instruments; Houston, TX) or a self-made P02 or P03 catheter was advanced into the left ventricle through the right carotid artery. The correct position of the catheter tip in the left ventricle was then confirmed by the waveform of the left ventricular (LV) pressure (LVP) visualized on a Hewlett-Packard monitor. The arterial line was connected to the computerized data-acquisition system to record HR, LVP, maximal rates of LVP rise and fall (LV dP/dt max and LV dP/dt min, respectively), ejection time (ET), and isovolumetric relaxation time constant (τ) at a sampling rate of 1,000 Hz. A venous catheter (PE-10/silicone catheter), made of 4 mm PE-10 tubing (SIMS Oortex) joined to 7 cm of silicone tubing (ID/OD: 0.51/0.94 mm, Ulrich) and filled with saline containing heparin (300 U/ml), was inserted into the left jugular vein for drug administrations. For monitoring of LV hemodynamic signals in the conscious state, the Pebax catheter (silicone tubing) was closed by inserting a copper pin and subcutaneously tunnelled to exit the skin on the back of the mouse, where it was attached with dental cement and a piece of cellophane tape. The mouse was then free to move around in the cage and allowed 3–4 h to recover from the halothane anesthesia. After the catheter was connected to the pressure transducer, and a stabilization period of 30 min, LV hemodynamic signals were simultaneously recorded for 10–15 min and saved for later analysis.

Comparison of Millar and Pebax Catheters

To compare the two methods under identical conditions, either a 1.4-Fr Millar or a P03 catheter was alternatively advanced into left ventricle of the same individual C57BL/6J mouse to record and...
analyze HR, LVP, LV dP/dt_max, LV dP/dt_min, ET, and τ for 5–10 min during an inhalation of 0.5% halothane. Afterward, the mouse was kept in the same position with the same dose of halothane (0.5%). The right carotid artery together with the catheter was held by a self-closing tweezer (Dumont; Montignez, Switzerland), the silk 3-0-made knots on the right carotid artery were carefully released, and the catheter (Millar or P03) was pulled off. Meanwhile, the bleeding was prevented by upstream stretching of a prepared silk 3-0 underneath the right carotid artery. The next different type of catheter was carefully inserted into the right carotid artery via the same hole on the wall. After the right carotid artery was clamped together with the catheter using the self-closing tweezer and the stretched silk 3-0 was released, the catheter was continuously and carefully pushed into the left ventricle with the tweezer. The ventricular parameters were then recorded again for 10–15 min. For this operation, a skilled microsurgical operator was needed. Furthermore, serial bolus injections of isoproterenol [0.2 mg/ml Isuprel (isoprenaline hydrochloridum) per ampoule, Sanofi-Pharma] were administered with doses ranging from 0.75 to 24 pg/g. The injection volume was kept at 1.5 μl/g body wt. The next dose was only given when LV hemodynamic measurements returned to baseline. The injection interval took 5–15 min depending on the dose of isoproterenol.

To investigate the role of anesthesia on cardiac contractility, mice inhaled two different doses (1% and 0.5%) of halothane during the sampling time. To do so, HR, LVP, LV dP/dt_max, LV dP/dt_min, ET, and τ were first simultaneously recorded with either the Millar or P03 catheter for 5–10 min as a baseline during an inhalation of 1% halothane. The dose of halothane was then switched to 0.5% and kept there until the maximum of LV hemodynamic changes appeared. Finally, the halothane concentration was again returned to 1%, and the LV signals were continuously recorded until their values returned to baseline.

Cardiac Contractility in Conscious DOCA-Salt Mice

As reported previously (37), DOCA-salt induces cardiac hypertrophy without hypertension in one-renin gene mice. Whether cardiac contractility in this mouse model of cardiac hypertrophy is also altered is not known. Therefore, 6-wk-old backcrossed Ns.6-129Ola × C57BL/6J mice harboring one renin gene were uninephrectomized and received a DOCA implant containing 35 mg of DOCA (Sigma) with a release rate of 21.3 ± 0.22 μg of DOCA/h (see Ref. 37 for details) and 1% NaCl drinking solution. Control mice of the same strain were uninephrectomized and received tap water as drinking fluid. Three months later, cardiac parameters were recorded for 15 min from conscious mice catheterized with the P03 catheter.

All experiments were carried out following the principles in the care and use of animals and were accepted by the local veterinary services.

Statistical Analysis

Average values of HR, LVP, LV dP/dt_max, LV dP/dt_min, ET, and τ were determined for 120 s from the sampling data. Results are expressed as means ± SE, and comparisons between groups were analyzed by paired and unpaired Student’s t-tests or by one-way ANOVA followed by a Newman-Keuls test. A P value <0.05 was considered statistically significant.

RESULTS

Dynamic Characteristics of the Pebax Catheter

Six P03 and five P02 catheters were characterized with the balloon-pop test (see Fig. 1B for an example). The natural frequency and damping coefficient of the P03 catheter were 202 ± 2 Hz and 0.058 ± 0.004, respectively, and the flat frequency of the P03 catheter was up to 50.5 ± 0.6 Hz, calculated by the natural frequency × the cutoff frequency (0.25; Fig. 1C). In contrast, the natural frequency and damping coefficient of the P02 catheter were 162 ± 2 Hz and 0.134 ± 0.004, respectively, and the flat frequency of the P02 catheter was 40.5 ± 0.5 Hz (P < 0.0001 vs. P03 catheters). Whereas LV systolic pressure (LVSP), HR, and ET were not affected by catheter size, LV dP/dt_max, LV dP/dt_min, and τ were reduced by 40% in conscious mice catheterized with the P02 catheter compared with those with a P03 catheter. This is clearly illustrated by the different wave shapes of LVP obtained with P02 and P03 catheters (Fig. 3A). When the cardiac cycle-matched LVP measurements obtained with P03 and P02 catheters were directly compared point by point, an open loop relationship resulted for the P02 catheter, thus further demonstrating the shortcomings of the P02 catheter (Fig. 3B). Table 1 shows the comparative dynamic characteristics of the Pebax catheters and the Millar catheter.

Comparison of Millar and Pebax Catheters

Direct comparison under anesthesia. Five individual mice anesthetized by inhalation of 0.5% halothane were alternatively catheterized under identical conditions with the 1.4-Fr Millar and P03 catheter, and the individual values are presented in Table 2. As shown, there was no difference between the two catheters. Figure 4A depicts LVP tracings obtained with the two catheters. It is clearly apparent that the two pressure curves are identical and that the detailing of the curve obtained with the P03 catheter is at least as good as that obtained with the 1.4 Fr Millar catheter. Further evidence that the two catheter systems yield identical results is provided in Fig. 4B: when the timed pressure points of different cardiac cycles obtained with the two catheter systems are related, a regression line not different from identity is observed.

In a further experiment, dose-response curves for isoproterenol were established in 12 mice under halothane (0.5%) anesthesia. Figure 5 illustrates the observed changes in cardiac contractility. It is again evident that the two catheters yield identical results when blood pressure and HR are changed acutely.

Effect of anesthesia on LV hemodynamics. The effects of two doses of halothane on cardiac contractility are shown in Table 3. As shown, when the dose of halothane was reduced from 1% to 0.5%, HR and LVSP progressively rose and the absolute values of LV dP/dt_max and LV dP/dt_min were almost doubled, whereas ET and τ were significantly shortened (P < 0.01, 0.5% vs. 1% halothane). These dose-dependent changes in LV hemodynamics induced by anesthesia were reversible when the dose of halothane was returned to 1%. This supports the evidence that LV hemodynamics are profoundly altered by anesthesia. Notwithstanding, the data obtained with the two catheter systems were again identical.

When LV hemodynamics were measured again in nine mice with a P03 catheter system in place 3–4 h after anesthesia in the conscious state, HR, LVSP, and absolute values of LV dP/dt_max and LV dP/dt_min were substantially higher and ET and τ were markedly shortened versus the data obtained with the same catheter system under anesthesia. The data of all nine animals are depicted in Table 3.
Cardiac Contractility in Conscious Normotensive Mice With DOCA-Salt-Induced Cardiac Hypertrophy

Recently, it has been demonstrated that N5.6−129Ola/C57BL/6J mice harboring only one renin gene develop cardiac hypertrophy without hypertension when treated with DOCA and salt (37). In the present experiments, the P03 catheter system was thus used in conscious N5.6−129Ola/C57BL/6J mice previously maintained on DOCA (21.3 ± 0.22 μg/h) and saline for 3 mo. The data are summarized in Table 4. The ratio of cardiac weight to body weight, an index of cardiac hypertrophy, was again found to be increased compared with matched controls, whereas blood pressure was not different. Absolute values of LV dP/dt max and LV dP/dt min were significantly reduced in DOCA-salt-treated mice. Similarly, τ was markedly prolonged, whereas ET was not changed. Thus, with the use of this new methodology in awake mice, a clear defect in cardiac contractility could be demonstrated in this normotensive mouse model of cardiac hypertrophy.

### DISCUSSION

With the general availability of transgenic methodology, the mouse has become an important experimental animal also for cardiovascular research. This has made it necessary to adapt the various measuring techniques to this small animal. So far, the measurement of cardiac contractility in the mouse was only possible with the Millar catheter, but the Millar catheter cannot be used in the conscious state and renders anesthesia or at least a sedation mandatory (10). As a result, all data available so far on cardiac contractility in mice were obtained under experimental conditions known to substantially alter most cardiovascular parameters (34). In addition, Millar catheters are quite expensive and cannot be used frequently in physiological studies.

![Fig. 3](image_url)

**Fig. 3.** A: example of tracings of LVP obtained from conscious mice catheterized with either the P03 (ID: 0.3 mm, top) or Pebax 02 (P02; ID: 0.2 mm, bottom) catheter. The average value of each LV hemodynamic parameter was determined for 120 s of recording. B: regressed curve made by point to point from cardiac cycle-matched LVPs determined by the LV end-diastolic pressure (LVEDP). 3rd LVP03 and 3rd LVP02 indicate the third LVP recorded with either the P03 or P02 catheter on the tracing in A.

<table>
<thead>
<tr>
<th>Mode</th>
<th>P03</th>
<th>Millar Micro-Tipped Catheters</th>
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<tbody>
<tr>
<td>Natural frequency, Hz</td>
<td>202 ± 2</td>
<td>1.4-Fr/SPR-671</td>
</tr>
<tr>
<td>Flat frequency, Hz</td>
<td>50.4 ± 0.6</td>
<td>1.8-Fr/SPR-612</td>
</tr>
<tr>
<td>Damping coefficient</td>
<td>0.058 ± 0.004</td>
<td>2-Fr/SPR-407</td>
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</table>

Data are means ± SE. The natural frequency and damping coefficient of the Pebax 03 (P03) catheter were analyzed by the “ballon-pop test” adapted from Gabe (6). Characteristics of the Millar catheters were taken from [http://www.wpi-europe.com/animalphy/MIKRO.html](http://www.wpi-europe.com/animalphy/MIKRO.html) and [http://www.pmsinstruments.co.uk/miller.htm](http://www.pmsinstruments.co.uk/miller.htm).
In the present study, a new, custom-made, low-cost catheter is presented that allows investigators to obtain LV hemodynamic measurements in the awake mouse and to derive key contractility parameters. The data demonstrate that the system exhibits an adequate frequency response and damping coefficient in vitro and under anesthesia yields results that are identical to those obtained with the Millar catheter. However, and maybe not surprisingly, results of further experiments confirm that anesthesia profoundly modifies cardiac contractility of the mice. This technique was then used in additional experiments to investigate LV contractility in mice with cardiac hypertrophy induced by the administration of DOCA and 1% NaCl. The results illustrate a markedly decreased contractility in these animals with hypertrophied hearts. Given all these data, it would seem preferable that, in the future, measurements of cardiac contractility in mice are made in the awake state.

Fluid-filled catheter systems with an inadequate dynamic response will cause phase and amplitude errors resulting in over- or underestimation of LVP. To avoid this problem, it is

| Table 2. Individual LV hemodynamic values obtained from C57BL/6J mice catheterized with either 1.4-Fr Millar or Pebax 03 catheters and anesthetized with 0.5% halothane |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|                | Millar          | P03             | Millar          | P03             | Millar          | P03             | Millar          | P03             | Millar          | P03             |
| Body weight, g  | 30              | 31              | 31              | 29              | 29              | 30 ± 1          |
| HR, beats/min   | 406             | 391             | 446             | 481             | 519             | 425             | 526             | 545             | 522             | 513             | 484 ± 24        | 471 ± 28        | 0.60            |
| LVSP, mmHg      | 112             | 115             | 122             | 122             | 107             | 114             | 96              | 99              | 114             | 111             | 110 ± 4         | 112 ± 4         | 0.29            |
| LVEDP, mmHg     | 9               | 4               | 13              | 14              | 3               | 8               | 2               | 2               | 5               | 2               | 5 ± 3           | 6 ± 2           | 0.84            |
| LV dP/dt max, mmHg/s | 8,434 | 8,577 | 8,347 | 9,201 | 8,364 | 8,098 | 7,526 | 8,124 | 8,785 | 8,393 | 8,291 ± 207 | 8,479 ± 201 | 0.47           |
| LV dP/dt min, mmHg/s | −5,944 | −5,624 | −6,091 | −7,393 | −6,500 | −6,457 | −6,706 | −6,986 | −7,862 | −7,044 | −6,621 ± 339 | −6,701 ± 308 | 0.83           |
| ET, ms          | 62              | 72              | 57              | 60              | 53              | 60              | 60              | 57              | 55              | 57              | 57 ± 2          | 61 ± 3          | 0.16            |
| τ, ms           | 11              | 10              | 10              | 8               | 6               | 9               | 5               | 5               | 7               | 6               | 8 ± 1           | 8 ± 1           | 0.96            |

Each value was averaged by determination of 120 s of LV signals recorded. HR, heart rate; LVSP, left ventricular (LV) systolic pressure; LVEDP, LV end-diastolic pressure; LV dP/dt max and LV dP/dt min, maximal rates of LV pressure rise and fall; ET, ejection time; τ, time constant of isovolumic relaxation. Student’s t-test (paired) was performed between Millar and P03 catheter groups.

In the present study, a new, custom-made, low-cost catheter is presented that allows investigators to obtain LV hemodynamic measurements in the awake mouse and to derive key contractility parameters. The data demonstrate that the system exhibits an adequate frequency response and damping coefficient in vitro and under anesthesia yields results that are identical to those obtained with the Millar catheter. However, and maybe not surprisingly, results of further experiments confirm that anesthesia profoundly modifies cardiac contractility of the mice. This technique was then used in additional experiments to investigate LV contractility in mice with cardiac hypertrophy induced by the administration of DOCA and 1% NaCl. The results illustrate a markedly decreased contractility in these animals with hypertrophied hearts. Given all these data, it would seem preferable that, in the future, measurements of cardiac contractility in mice are made in the awake state.

A fluid-filled catheter system with an inadequate dynamic response will cause phase and amplitude errors resulting in over- or underestimation of LVP. To avoid this problem, it is
Determination of Cardiac Contractility in Awake Mice

than those obtained with the P03 catheter even though LVSP

generally believed that a fluid-filled catheter system must have a frequency response that is flat up to five times the source frequency to be recorded and in addition must exhibit a low damping (7, 25, 26, 29, 38). The natural frequency of a fluid-filled catheter varies with its radius and stiffness and inversely with its length. The damping coefficient is proportional to its length and inversely related to its radius and stiffness (25, 26, 38). Therefore, to obtain a high-frequency response and a low damping, the catheter length should be short and the internal diameter of the catheter must be as large as possible. With this strategy in mind, the P02 and P03 catheters were produced and characterized in our laboratory.

To obtain the same frequency response and a low damping, the catheter length should be short and the internal diameter of the catheter must be as large as possible. With this strategy in mind, the P02 and P03 catheters were produced and characterized in our laboratory.

As expected, the P03 catheter turned out to have a high natural frequency, a low damping coefficient, and a flat frequency up to 50 Hz, which should be high enough for accurate analysis of LVP with a frequency (HR) of ~10 Hz in conscious mice. In contrast, the pebax 02 catheter had a flat frequency up to 40 Hz. As a consequence, when both Pebax catheters were used in conscious mice, the absolute values of LV dp/dtmax and LV dp/dtmin recorded with the P02 catheter were ~40% lower than those obtained with the P03 catheter even though LVSP was not different between the two groups. However, the wave shape of LVP was substantially different (Fig. 3A), and consequently the regression curve derived from cardiac cycle-matched LVPs monitored with two catheters revealed an open loop (Fig. 3B). Apparently, the lower limit of the internal diameter of the Pebax catheter to exhibit the dynamic characteristics to allow analyses of LV dp/dtmax and LV dp/dtmin in conscious mice must be over 0.2 mm. This is in agreement with a recent report (40) of absolute values of LV dp/dtmax and LV dp/dtmin obtained with a PE-200 stretched fluid-filled catheter with an internal diameter of 0.1~0.24 mm that were 18% lower than those measured with a Millar catheter in mice anesthetized with the same dose of ketamine. Obviously, the OD of the catheter should be as small as possible. Actually, the OD of the P03 catheter tip including the silicone “cap” is 0.64 mm, which is still acceptable for mice with a body weight of 25 g.

The comparison between the Millar and the P03 catheters had to be performed in anesthetized mice because the former can be used only in this condition. Either the 1.4-Fr Millar or P03 catheter was alternatively placed into the left ventricle of the same individual mouse to record HR, LVSP, LV dp/dtmax, LV dp/dtmin, ET, and τ during an inhalation of 0.5% halothane. Neither the wave shapes of LVP nor the values of all recorded parameters differed between the two catheters. These in vivo experiments demonstrate very clearly that under equal anesthetia the 1.4-Fr Millar catheter and the P03 catheter produced identical LV hemodynamic measurements in the same mouse.

Table 3. Effect of halothane dose on LV hemodynamics obtained with Millar or P03 catheters in C57BL/6J mice

<table>
<thead>
<tr>
<th>1.4-Fr Millar Catheter</th>
<th>P03 Catheter</th>
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<tr>
<td><strong>1.0% Halothane</strong></td>
<td><strong>1.0% Halothane</strong></td>
</tr>
<tr>
<td><strong>0.5% Halothane</strong></td>
<td><strong>0.5% Halothane</strong></td>
</tr>
<tr>
<td><strong>n</strong></td>
<td><strong>n</strong></td>
</tr>
<tr>
<td>Body weight, g</td>
<td>29±0.4</td>
</tr>
<tr>
<td>LVSP, mmHg</td>
<td>95±4</td>
</tr>
<tr>
<td>HR, beats/min</td>
<td>377±13</td>
</tr>
<tr>
<td>LVEDP, mmHg</td>
<td>8±2</td>
</tr>
<tr>
<td>LV dp/dtmax, mmHg/s</td>
<td>4,854±244</td>
</tr>
<tr>
<td>LV dp/dtmin, mmHg/s</td>
<td>−3,371±136</td>
</tr>
<tr>
<td>ET, ms</td>
<td>72±3</td>
</tr>
<tr>
<td>τ, ms</td>
<td>14±1.7</td>
</tr>
</tbody>
</table>

Values are means ± SE; n, no. of mice. *P < 0.05, **P < 0.01, and ***P < 0.001, 1% vs. 0.5% halothane; aP < 0.01 and bP < 0.001, conscious state vs. 0.5% halothane.
To investigate the role of anesthesia on cardiac contractility, mice inhaled different doses (1% and 0.5%) of halothane, and LV hemodynamic parameters were again determined with either the Millar or the P03 catheter system. All the variables were markedly altered by changing the dose of halothane, and these dose-dependent changes were reversible. Again, no difference between the two recording methods was observed. In marked contrast, however, when HR, LVSP, and absolute values of LV dP/dtmax and LV dP/dtmin were again measured in conscious mice, they were substantially higher, and ET and τ were shortened. This may not come as a complete surprise because anesthesia has been shown repeatedly to substantially alter hemodynamic variables (3, 4, 13, 18, 19, 29, 30, 33, 39). Table 5 summarizes the data published in several papers in which cardiac hemodynamics were measured in anesthetized mice. Whereas our LV hemodynamic data obtained under anesthesia with the P03 catheter are in full agreement with previous reports, the parameters obtained with the same catheter in the awake state are clearly different. As a consequence, experiments exploring cardiovascular physiology or pharmacology in intact animals should be carried out in the awake state whenever possible to avoid erroneous results. Unfortunately, so far, this requirement could not be fulfilled in mice with the Millar catheter due to its rigidity and cost. The present results obtained with the P03 catheter in awake mice set a new benchmark for cardiac contractility in mice and underline once more the importance that observations be made in the conscious state.

Similar to previous studies (11, 14, 16, 17), we found that many of the LV hemodynamic parameters, such as LV dP/
d_{fl, max}, LV \frac{dP}{dt_{min}}, and \tau, are related to HR. Furthermore, LVSP correlated with LV \frac{dP}{dt_{max}} and LV \frac{dP}{dt_{min}}, which provides further evidence that anesthesia-induced hypotension and bradycardia distort LV \frac{dP}{dt_{max}} and LV \frac{dP}{dt_{min}}. ET was so closely correlated with HR (r = 0.93, P < 0.0001) that it seems to be almost entirely determined by HR.

Finally, the P03 catheter was used in a one-renin gene mouse model of cardiac hypertrophy without hypertension induced by DOCA and salt (37) to assess cardiac contractility. After 3 mo of DOCA and salt administration, the ratio of heart weight to body weight was increased compared with the control, as reported previously (37). It was now possible to show that absolute values of LV \frac{dP}{dt_{max}} and LV \frac{dP}{dt_{min}} were more than 50% decreased and τ was three times longer compared with control animals. These data clearly demonstrate that these DOCA and salt-treated mice exhibit decreased cardiac contractility even in the absence of hypertension, suggesting that pressure-independent factors such as DOCA, salt, hypokalemia, or cardiac hypertrophy per se may be the cause of this decrease in cardiac contractility, as suggested in other animal models (32).

In conclusion, the present study clearly demonstrates that anesthesia markedly alters cardiac contractility and causes dose-dependent changes in LV \frac{dP}{dt_{max}} and LV \frac{dP}{dt_{min}}. With our new P03 catheter with a natural frequency of \sim 200 Hz, a damping factor of 0.06, and a flat frequency of up to 50 Hz, LV hemodynamics can now be accurately measured in conscious mice. An additional advantage of this new catheter is the fact that it costs a fraction of the Millar probe. In contrast to some Millar catheters, the Pebax catheter does not enable the measurement of pressure-volume relationships. However, even these relationships can be obtained in conscious mice with the actual technologies. It is hoped that in the future, cardiac contractility of mice will be measured in the awake state. This new device can be of great help in the investigation of cardiac function in normal and genetically engineered mice.

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REFERENCES