Pressure-volume relation analysis of mouse ventricular function

Oscar H. Cingolani and David A. Kass

Division of Cardiology, Department of Biomedical Engineering, Johns Hopkins University School of Medicine, Baltimore, Maryland

Submitted 3 August 2011; accepted in final form 12 September 2011

Cingolani OH, Kass DA. Pressure-volume relation analysis of mouse ventricular function. Am J Physiol Heart Circ Physiol 301: H2198–H2206, 2011. First published September 16, 2011; doi:10.1152/ajpheart.00781.2011.—Nearly 40 years ago, the Sagawa laboratory spawned a renaissance in the use of instantaneous ventricular pressure-volume (P-V) relations to assess cardiac function. Since then, this analysis has taken hold as the most comprehensive way to quantify ventricular chamber function and energetics and cardiovascular interactions. First studied in large mammalian hearts and later in humans employing a catheter-based method, P-V analysis was translated to small rodents in the late 1990s by the Kass laboratory. Over the past decade, this approach has become a gold standard for comprehensive examination of in vivo cardiac function in mice, facilitating a new era of molecular cardiac physiology. The catheter-based method remains the most widely used approach in mice. In this brief review, we discuss this instrumentation, the theory behind its use, and how volume signals are calibrated and discuss elements of P-V analysis. The goal is to provide a convenient summary of earlier investigations and insights for users whose primary interests lie in genetic/molecular studies rather than in biomedical engineering.

A comprehensive assessment of mouse cardiac hemodynamics in vivo can be essential to define the physiological significance of a given genetic or pharmacological modification. This is often measured using imaging tools (e.g., Doppler/echocardiography or MRI) and left ventricular (LV) pressure data. However, a more detailed analysis may be desired, and this can be provided by ventricular pressure-volume (P-V) analysis. The P-V approach provides a comprehensive method to assess cardiac systolic and diastolic function in a manner less affected by arterial and venous loading while at the same time quantifying this load. Methods to derive P-V relations in mice, principally using a conductance catheter, were developed in the late 1990s in our laboratory and are now commercially available. The method and analysis require some appreciation of the signals and potential sources of error, as well as the underlying hemodynamics. The original work regarding P-V analysis dates back 40 years, when it was the focus of bioengineering and systems biologists (34). Its emergence in an era of molecular physiology in mice has made it all the more valuable to revisit the principles, assumptions, and limitations of these analytical approaches and the methodologies used to apply them. The contents of this review are not new; however, as the constellation of investigators using P-V analysis has dramatically changed from physiologist/engineers to molecular biologists, we thought it useful to update (remind) researchers of the details and provide contemporary examples of how P-V analysis is providing unique insights into molecular physiology.

A Bit of History

The first studies regarding ventricular P-V relations were reported from frog ventricle and date to the late 19th century with the work of Otto Frank (7). However, it was the seminal work from the Sagawa laboratory in the 1970s and 80s that truly advanced the understanding of these relations and their utility (41, 44–46). These investigators established the concept that ventricular contraction behaves as a time-varying elastance, like a spring with a stiffness constant that changes from diastole to systole and back. They examined the end-systolic P-V relation (ESPVR), the set of P-V points from multiple cardiac cycles generated under different loading conditions, each reflecting maximal elastance for that condition. Over several years, they established the relative (though not absolute) independence of the ESPVR from changes in cardiac filling volume (preload) and aortic impedance (afterload) (44), its modulation by chamber size/geometry (42), nonlinear ESPVR behavior (3, 15), how to couple the ESPVR to the arterial loading system (46, 47), and how to analyze myocardial energetic efficiency (43). All of this work was performed in isolated, excised, blood-perfused canine hearts, and it was not until the mid-1980s that in vivo translation occurred with the arrival of a conductance catheter (2, 20) and sonomicrometer measurements (23) to determine LV volume. Of these, the conductance method was applicable to humans, and by the late 1980s, work by Kass and others (16, 18, 19) revealed the utility of P-V analysis for studies of human heart disease. The conductance (inverse of electrical resistance) catheter was inserted into the LV so it lie along the longitudinal axis. It
provided an alternating constant current field between base and apex and sensed voltage differences between intervening electrode pairs to generate a blood volume signal (volume was inversely proportional to resistance, directly to conductance). P-V relations were obtained by combining a micromanometer sensor with the volume signal.

In 1998, P-V analytic methods were miniaturized by the generation of very thin catheters and custom instrumentation (10). Whereas the use of P-V analysis in humans has remained limited to a few research laboratories worldwide, its use in mice has exploded, and it is now considered the gold standard for ventricular functional analysis. Understanding the principles and use of the method involves two components: the methodology of the volume measurement [see also Pacher et al. (30)] and the analysis of the data obtained. We will deal with both in this order.

From Conductance to Volume

The original P-V catheters designed for larger experimental animals and humans consisted of 8–12 equally spaced electrodes, with a micromanometer mounted between electrodes 3 and 4 so it would lie within the LV. The catheter was placed with its distal tip in the LV apex, and proximal electrode just beyond the aortic valve. A high-frequency (~20 kHz), low-amplitude (~10 μA) alternating current was injected between the most proximal and distal electrodes, generating a current field about the heart, and voltage was measured between pairs of the intervening electrodes. As equal-voltage planes were perpendicular to the current field and thus catheter shaft, this allowed for a reasonable assumption to be made: voltage between two neighboring electrodes would be inversely related to conductivity of the material between the electrodes (blood + heart muscle), and the time-varying component of this voltage largely depended on varying blood volume during the cardiac cycle. One added up the signals from sequential pairs of electrodes, like a summation of disks, to calculate total volume. For the mouse, it was impractical to scale this to a catheter <0.5 mm in diameter, so the electrode count was simplified to four: two outer pairs for stimulation and two inner for voltage determination (Fig. 1). Blood conductivity is approximately three times more than myocardium, and as this source more than myocardial conductivity varies during the cardiac cycle, the signal is thought to primarily reflect blood volume change. This is not absolute volume; there is an offset due to conductance of the wall and surrounding structures and a signal gain that is not unity. Calibration is needed to convert to absolute volume.

The primary equation relating conductance to volume is \( V = \frac{1}{\alpha} (<pL^2>(C - G_p)/2) \), where \( \rho \) is the blood resistivity, \( L \) is the distance between sensing electrodes, \( G \) represents conductance (inverse of voltage with a constant current circuit), \( G_p \) is conductance from muscle wall and surrounding tissues (parallel conductance), and \( \alpha \) is the gain. If the current was injected from parallel plates at base and apex, the field lines would all pass linearly and the assumptions in this equation would be perfectly met. However, we have point sources (ring electrodes) for the current, so the field lines are curved as are the voltage planes perpendicular to them. This introduces a non-linearity that is more and more evident the larger the heart is (further distance away from the point sources). While this impacts large mammalian and human hearts that are substantially dilated, small rodents such as the mouse are at an advantage here, since the short axis is 1 to 2 mm and wall thickness is around 1 mm (relatively hypertrophied even in normal mouse compared with human). Even when the mouse heart is dilated severalfold, one is still dealing with a field distribution that is close to the catheter shaft. This helps stabilize the signal, improves linearity versus gold standard volumes, and also insulates the signal from far field artifacts (10). LV volumes acquired by the conductance catheter correlate quite well with those obtained by other imaging methods, such as cardiac MRI, even in mice with postinfarction cardiomyopathy, where volumes are larger and LV walls present fibrosis (48). In many ways, the small rodent heart has turned out to be the ideal application for this methodology.

A pressure signal can be integrated with other imaging methods to obtain volume such as MRI or echocardiography to convert P-V data to stress/strain (4). However, this has its own limitations requiring careful signal synchronization and is hard to use for transient manipulations, such as load change, used to derive the full set of relations.

Depending on the parameters to be examined, calibration of the conductance signal is important. There are several different methods that have been proposed, some being provided with commercial systems. One is the external cuvette calibration, where a Plexiglas block with cylinder-shaped holes of known volume is filled with heparinized blood. By dipping the catheter into these holes, conductance signals are obtained and plotted against known volume to obtain a correlation. However, this method is rarely accurate, since the in vivo electric field distribution is quite different than in this external system and the offset and gain are thus different. Another method is based on a comparison with another “gold standard” measure of flow,
Doppler ultrasound, thermodilution, or other methods (5, 10, 30). One determines the stroke volume (SV) by this approach and then calibrates the conductance signal to match it. This is done by estimating the average width of the P-V loop and setting this to the independently determined SV. This gives us the “true” SV (Fig. 2A).

To assess the offset or parallel conductance, one can use the hypertonic saline calibration method first proposed by Baan et al. (2) and later applied and validated by our group in mice (9). This is based on the principle that if one only varies the conductivity of blood (introducing a very low volume of hypertonic saline into the venous return) but not actual blood volume, the catheter would interpret this as an increase in volume from one beat to the next (Fig. 2B). This is then used to estimate what the catheter signal would be if there was no blood conductance volume, i.e., signal from non-blood sources (Fig. 2C). This becomes the parallel conductance. Details of this as well as pitfalls have been previously reported (9, 30). Key issues are that LV pressures should be stable during this injection and beats during the initial wash-in phase of rising volume signals are used. Anything else reflects changes in real LV function because of the hypertonic saline typically depressing function.

Most recently, another method to calibrate the conductance signal has been proposed that is based on a real-time assessment of both parallel conductance and gain variability during the cardiac cycle, based on admittance dynamic analysis and Wei’s equation (21, 33). This requires a determination of myocardial admittance (inverse of impedance) but then automates a real-time estimation of any time-varying myocardial admittance contribution to the volume signal and compensates for time-varying gain. Commercial systems with this algorithm are available (ADVantage, Scisense, London, Ontario, Canada), and studies have proposed this as superior to the traditional approach and more forgiving of catheter misalignment in the LV (21). Validation of this claim in various disease conditions has not been reported. In our hands, the traditional approaches work very well and strongly correlate with echocardiographic or cardiac MRI-based volumes. Nonetheless, the flow probe and saline calibration methods are tricky, and a robust automated approach would indeed be valuable.

Surgical Technique and Common Pitfalls

To obtain reproducible, good quality P-V hemodynamics in mice, a skillful operator is crucial. The following steps need to be followed carefully: 1) induction of anesthesia and preparation of the skin for the procedure, 2) intubation and correct placement of the mouse over a heating pad, 3) intravenous access for hydration/volume expansion, 4) surgical approach to the heart.

**Fig. 2.** A: baseline pressure-volume (P-V) loop for the LV, with stroke volume (SV) defined as the average width of the loop (averaging volumes from the 2 “vertical” sides). B: calibration of the parallel conductance. A set of loops are recorded after rapid bolus injection of hypertonic saline (~10 μL). Ventricular pressures are minimally altered if this is performed correctly. C: peak and trough volumes [end-diastolic volume (EDV) and end-systolic volume (ESV)] are plotted, and a regression between them is extrapolated to the line of identity (dashed line) to determine parallel conductance (Vp). Once done, this can be subtracted from the conductance signal to obtain calibrated LV volume.

Review

H2200

ASSESSMENT OF CARDIAC FUNCTION IN MICE

AJP-Heart Circ Physiol • VOL 301 • DECEMBER 2011 • www.ajpheart.org

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ESV = 0.5557 * EDV + 2.03, r² = 0.9973, Vp = 4.6 μL
expose the cardiac apex, and 5) correct introduction and positioning of the catheter into the LV.

Different anesthetics are available (Table 1), though we continue to prefer a combination of etomidate-urethane or etomidate-isoflurane. After the mouse has been placed in a Plexiglas box with isoflurane for several seconds, an injection of etomidate (5–10 mg/kg ip) is given, the chest and neck are shaved (internal jugular vein access is necessary for intravenous fluid administration), and the animal is intubated (or tracheostomized). The mouse is placed on a heating pad to keep body temperature around ~37°C and ventilated (for a 25-g intubated mouse, we use a respiratory rate of 130–150/min and tidal volume of 200–250 μl). Temperature control in anesthetized mice is critical. The mouse heart rate is exquisitely sensitive to changes in temperature, and HR (and contractility) can easily be decreased by half the normal values if heating is inappropriate. A warm pad at 40–45°C will keep body temperature close to 37°C. If possible, monitoring is preferred with a rectal thermometer. It is important to achieve normal baseline contractility parameters, such as HR and peak rate of pressure rise (dP/dt_max) (see Table 1), and maintain them steady throughout the procedure while data are being acquired.

The internal jugular vein should be accessed (e.g., a PE-10 silastic tubing connected to a 30-gauge needle), and a constant infusion of 12.5% albumin in normal saline is given at 5 ml/min, after an initial 50-μl bolus. This is necessary to counteract the peripheral vasodilatation and hypotension induced by anesthesia. A subxyphoid incision is made next, exposing the diaphragm where a window is performed with an electrocautery device (this is crucial since the diaphragm is vascularized and bleeds after sectioning without cautereization). The LV apex is punctured with a 26- to 27-gauge needle, and the catheter is carefully introduced via this puncture. It is important to use the correct needle size, large enough to allow the introduction of the catheter without damaging it, but not too large that the blood will flow back from a hole bigger than the catheter’s diameter. LV catheterization retrograde, with the catheter introduced via the right carotid artery, has also been used by many. Our group prefers the apical approach since there is never a question of placement: the possibility of being trapped along the wall in trabeculae or papillary muscle is minimized, and one knows where the electrodes are.

Once inside the LV, a correct positioning of the catheter is also important. Its shaft needs to be aligned with the LV major (longitudinal) axis, with both outer electrodes right below the aortic valve and adjacent to the apical endocardial border, respectively (Fig. 1). Correct volume and pressure signals should be identified, together with generally squared- to oval-shaped loops (Fig. 2A). Before recording data, a change in loading conditions [either via inferior vena cava (IVC) occlusion or aortic constriction maneuver] should be performed to ensure that the electrodes are not in contact with intracardiac structures (i.e., papillary muscles) during the loading-changing maneuver. The catheter itself has a cross-sectional diameter of 0.625 mm, and with a 7-mm long axis, this occupies a bit more than 2 μl. This is <10% of end-diastolic volume (EDV) but could be a substantial percentage of end-systolic volume, and

### Table 1. Schematic of P-V catheter placement via left ventricular apex in the mouse

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Urethane + Etomidate + Morphine</th>
<th>Isoflurane</th>
<th>Ketamine + Xylazine</th>
<th>Pentobarbital Sodium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight, g</td>
<td>20–30</td>
<td>20–34</td>
<td>20–34</td>
<td>20–34</td>
</tr>
<tr>
<td>Heart rate, beats/min</td>
<td>490–655</td>
<td>470–620</td>
<td>340–510</td>
<td>365–550</td>
</tr>
<tr>
<td>MAP, mmHg</td>
<td>81–105</td>
<td>93–109</td>
<td>72–90</td>
<td></td>
</tr>
<tr>
<td>ESP, mmHg</td>
<td>80–120</td>
<td>92–118</td>
<td>104–125</td>
<td>90–110</td>
</tr>
<tr>
<td>EDP, mmHg</td>
<td>2–3</td>
<td>1–6</td>
<td>1–9</td>
<td>2–8</td>
</tr>
<tr>
<td>ESV, μl</td>
<td>2–12</td>
<td>7–21</td>
<td>9–20</td>
<td>10–29</td>
</tr>
<tr>
<td>EDV, μl</td>
<td>20–33</td>
<td>25–53</td>
<td>25–39</td>
<td>28–54</td>
</tr>
<tr>
<td>Stroke volume, μl</td>
<td>14–26</td>
<td>17–30</td>
<td>14–21</td>
<td>17–25</td>
</tr>
<tr>
<td>CO, ml/min</td>
<td>7–16</td>
<td>8–16</td>
<td>6–10</td>
<td>6–13</td>
</tr>
<tr>
<td>CI, ml·min⁻¹·kg</td>
<td>280–557</td>
<td>350–580</td>
<td>225–400</td>
<td>226–480</td>
</tr>
<tr>
<td>Emax, mmHg/μl</td>
<td>4–6</td>
<td>3–7</td>
<td>5–9</td>
<td>4–6</td>
</tr>
<tr>
<td>TPR, mmHg·ml⁻¹·min</td>
<td>—</td>
<td>6–12</td>
<td>10–19</td>
<td>7–14</td>
</tr>
</tbody>
</table>

Systolic indexes

| Ejection fraction, % | 50–88                          | 55–72     | 49–63               | 44–62                |
| dP/dt_max, mmHg/s   | 9,500–16,000                    | 8,200–14,200 | 7,700–14,480         | 6,900–11,000         |
| SW, mmHg·μl         | 1,200–2,700                     | 1,500–2,600 | 1,600–2,200         | 1,100–2,100          |
| Emax, mmHg/μl       | 6–14                            | 7–14      | —                   | 6–9                  |
| dP/dt_max, EDV, mmHg·s⁻¹·μl⁻¹ | 360–600                  | 180–470  | 160–390             |                      |
| PRSW, mmHg          | 70–130                          | 58–93     | 51–86               |                      |
| Efficiency, %       | 70–85                           | 60–75     | —                   | 55–68                |

Diastolic indexes

| −dP/dt_min, mmHg/s | 6,000–12,000                    | 6,700–10,500 | 6,900–10,400        | 5,400–9,500          |
| τ (W), ms          | —                               | 4.4–7.6     | 4.8–8.5             | 4.9–10              |
| τ (G), ms          | 7–9                             | 7–12       | 8–13                | 7–15                |
| EDPRVR             | 0.04–0.12                       | —          | 0.06–0.2            |                      |

CI, cardiac index; CO, cardiac output; dP/dt_max·EDV, relation of peak rate of pressure rise and end-diastolic volume; −dP/dt_min, peak rate of pressure decline; Emax, arterial elastance (measure of ventricular afterload); EDP, end-diastolic pressure; EDPRVR, pressure-volume (P-V) relation slope; EDV, end-diastolic volume; EDPVR, end-diastolic elastance (slope of the end-diastolic relationship); Efficiency [stroke work (SW)/P-V area]; ESP, end-systolic pressure; ESV, end-systolic volume; MAP, mean arterial pressure; PRSW, preload-recruitable stroke work (slope of stroke work-EDV relationship); τ (G), relaxation time constant calculated by Glantz method (regression of dP/d vs. pressure); τ (W), relaxation time constant calculated by Weiss method [regression of log[pressure]]; TPR, total peripheral resistance, MAP/CO. Values were calculated with correction based on aortic flow measurements and saline calibrations as described from P-V loops obtained with open-chest approach (modified from Ref. 30; used with permission).
during transient IVC occlusion, where these volumes decline further, this can lead to pressure spiking at end systole often because of entrapment. This can be usually fixed with some modest additional fluid repletion. Other than this, having the catheter occupy blood volume space in the chamber does not impact function, and as the signal is calibrated to external standards, this “dead volume” is corrected for.

As mentioned earlier, to change the loading conditions, transient occlusion of the IVC or aorta is usually performed. For the former, this is achieved by either pushing with a cotton tip right before the IVC enters the right atrium or pulling from a suture placed around the vessel. For the aorta, the suture technique is recommended given the higher pressures. For the latter, we personally prefer to perform an incision at the second and third ribs left to the sternum, following the surgical technique used for transverse aortic coarctation (4), and expose the ascending aorta, where a 5-0 to 7-0 suture is passed around the vessel. Several P-V loops are then recorded while changing loading conditions (examples shown in Fig. 3A). P-V analysis is not identical based on these different loading maneuvers, and for a variety of reasons, including analytical and physiological, preload modulation is preferable. This is discussed in more detail below.

Assessment of Ventricular Function

Ventricular P-V data are the in vivo chamber correlate of isolated muscle force-length relations; active and passive force-length relations correspond to the end-diastolic and end-systolic P-V relations. P-V data are affected not only by myocyte properties, however, but also by vascular and extracellular matrix, as well as by chamber geometry. Various parameters derived from these relations can therefore be impacted by nonmyocyte behavior, and this needs to be kept in mind. One can divide the parameters typically obtained into several groupings: 1) resting function, 2) systolic relations, 3) diastolic relations, 4) ventricular-vascular coupling, and 5) energetics.

Resting function. Baseline P-V data provide standard measures of volumetric function (e.g., ejection fraction and stroke work) but also peak ventricular power (maximal pressure-flow product), pressures and pressure derivatives, relaxation time constants, and other parameters. Loops are not always perfectly vertical during isovolumic contraction and relaxation, even without any physiological mechanism for deviation. This is most often due to slight signal artifacts which become obvious when pressure is changing rapidly as it does in these two
periods. Rather than use maximum and minimum volumes to determine SV, we prefer averaging multiple volumes from the mid-two sides of the loop, taking advantage of the additional data available. This reduces a bias of selecting one point over another and uses more of the available data. SV is then the mean width of the loop. This calibration impacts other parameters, so it is worth considering. Data are typically derived from multiple signal-averaged beats or can be determined from multiple beats and then averaged.

**Systolic relations.** The primary P-V relation identified by Sagawa and colleagues was the ESPVR. Ideally, this is determined by finding the points of maximal P/(V - V₀) for each beat, using an iterative method to identify V₀, and then fitting the data using perpendicular regression. The latter is preferred over linear regression, since it does not designate a dependent and independent variable but minimizes the perpendicular distance of each point to the regression line. Nonlinear models have also been described, typically a parabolic model (15) or a theoretically based logarithmic model (26). As shown in the two examples in Fig. 3A, the ESPVR may be fit to a linear model, though it is often curvilinear. In the measured data range, the difference may be subtle, but extrapolations clearly differ.

There are a number of alternative systolic relations that have been derived from the same P-V data, all with the goal of deriving a systolic index less susceptible to changes in LV load. Little et al. (22) studied the dP/dt max-EDV relation (22), which is more sensitive to inotropic change from PKA phosphorylation than is the ESPVR. Rankin and colleagues (11) developed preload-recruitable stroke work (PRSW), a linear version of the Sarnoff relation (35), and Sharir and Kass (39) pursued maximal power-EDV relations. Each of these ordinate parameters, dP/dt max, stroke work, and maximal power, vary with preload but less with afterload (unless pushed to extremes). Thus, if a set of P-V data are obtained by transient aortic occlusion, none of these is useful, whereas all can be derived from preload modified data. This is shown in Fig. 3B. Only the preload reduction-derived data are interpretable, whereas the relations from the afterload increase analysis are not. PRSW is particularly useful because its units are force (pressure) and errors in volume calibration are removed. As a result, it is chamber size independent, so mice, rats, dogs, pigs, sheep, and humans all have the same normal rest value of 70–90 mmHg. This is a useful independent check on P-V analysis and on the condition of the hearts being studied. Reports of PRSW values in the 30–40 mmHg range in control animals raise concerns about cardiac depression and experimental conditions and would make corresponding transgenic analysis suspect.

There are several important issues pertinent to comparing ESPVRs. First, the position as well as slope are relevant, a relation that is acutely leftward shifted but with a lower slope does not indicate depressed contractility but more likely nonlinear behavior (15). Acute interventions with inotropes or pacing can impact both slope and extrapolated V₀. Second, a quantitation of shifts of these relations to acute interventions may be misleading when based solely on slope, particularly if the data are nonlinear. As depicted in prior studies (28, 40) and in Fig. 4, differences between nonlinear ESPVRs can easily be assessed by an area defined between them. This P-V area is constrained to lie within the measured range of the data sets and involve physiological pressures. The area is determined from the nonlinear fits using analytical integration, and it represents an altered P-V area that the heart can operate within as a result of an ESPVR change. The third, the ESPVR is relatively but not absolutely load independent. Studies performed in the mid-1980s demonstrated the impact of high afterload on the ESPVR (shifting it leftward) (8), and differences in ESPVRs derived from preload versus afterload modulated contractions (1, 12). The latter more than the former impacts the duration of systole and the extent of shortening, so different loops may not really be derived from identical resting contractile states (17). This is less prominent with preload change.

**Diastolic relations.** One of the common uses of P-V relations is to define the diastolic P-V relation (DPVR), something difficult to do from imaging methods. This ideally requires more than single steady-state beats, as the relations are impacted by relaxation processes, right ventricular load, and pericardial constraint (14). Using a set of loops measured over a range of loading (typically IVC occlusion), one identifies the lower boundary, fitting the data to linear, exponential, logarithmic (14), or harmonic oscillator models (27). The most common is an exponential model: P = α(βV - 1) + P₀, where α is a stiffness and scaling coefficient, and β is chamber stiffness coefficient, and P₀ offset pressure at a volume of 0. Figure 5 shows an example of marked differences in DPVR between mice with desmin-related cardiomyopathy and controls. While Doppler indexes of filling can be used to assess such restrictive type physiology, the direct analysis of P-V relations during diastole is valuable to determine stiffness modulation.

As with ESPVR, one needs to be careful about analyzing DPVRs. The most common fit using an exponential is overparameterized for the data being fit, i.e., that while one can identify parameters by the curve fit algorithm, alternative fits with quite different parameter sets can fit the data virtually as well. Examining the displacement between relations by area integration or other subtraction methods (19), which do not rely solely on the model fits, can be used.

There are other indexes of diastolic function that are derivable from P-V relations. The first is the rate of decline
of ventricular pressure during isovolumetric relaxation, as-
lected from micromanometer data. Here too, assumptions
are often made that may not be appropriate. The most
common model is a monoexponential decay fit to the pres-
sure from peak rate of pressure decline to the estimated
onset of LV filling (end-diastolic pressure + 2–5 mmHg).
The time constant $\tau$ is reported. However, some models
assume pressure decays to zero, and this may not be the case
(more often not in intact humans). Assuming this when not
the case will yield a falsely high $\tau$ value (37, 38). Second,
these models assume that pressure decay follows a mono-
exponential. This is reasonable for normal hearts but is not
the case in depressed ventricles with dilated cardiomyopa-
athy. Here, an alternative logistic model has proven to be the
better choice (25, 37). If one assumes an exponential decay
when the actual pressure decline waveform does not follow
this trajectory, then the calculated $\tau$ will be artificially high
and sensitive to the absolute value of pressure estimated at
end diastole. Lastly, one can determine end-diastolic pres-
sure, which with P-V loops is often defined as the pressure
at the lower right corner. Without volume data, it is often
defined as the pressure at peak QRS or pressure when $dP/dt$
exceeds 10% of maximum. A visual determination from
pressure waves alone is often unreliable.

Fig. 5. Use of P-V analysis to assess the dia-
stolic P-V relation. Data from mice harboring a
R120G mutation in B-crystalin (CryAB$^{R120G}$)
that causes desmin-related cardiomyopathy and littermate controls are shown. The former was
associated with chamber dilation, and marked
diastolic stiffening reflected by the end-diastolic
P-V relation (EDPVR) are shown here fit by a
3-parameter monoeXponential relation.

Fig. 6. A: use of effective arterial elastance ($E_a$)
to assess LV afterload. Three loops at different
levels of aortic occlusion are shown, and $E_a =$
end-systolic pressure/SV is shown for each. B:
normalized time-varying elastance [$E_\alpha(t)$] de-
ferred from mice lacking myosin-binding pro-
tein C (MyBPC$^{-/-}$) and from 4 different ge-
etic models of heart failure. MyBPC deletion
results in a unique temporal shortening of the
time course, abbreviating systole. This was a
primary mechanical defect in this model. My-
card, myocardium; MKK3a, MAPK kinase 3a;
Tnl t/t, troponin I truncation mutation. C: effect
of omecamtiv mecarbil (myosin ATPase activ-
ator) on $E_\alpha(t)$. Unlike dobutamine (left), which
accelerates ventricular stiffening and abbrevi-
ates systole, omecamtiv mecarbil had no im-
 pact on the initial rising phase but markedly
prolonged the duration of systolic stiffening.
Thus the time to reach peak $E_\alpha(t)$ was length-
ened. This is a novel mechanism of action for a
cardiac inotrope, and one that required P-V
analysis to identify. $T_{E_{\max}}$ and $T_{E_{\min}}$, time at
maximal and minimal elastance, respectively.
Ventricular-arterial coupling. While P-V relations can yield characterizations of the heart that are relatively less impacted by the loading constraints imposed on the ventricle, they also yield metrics of this load. EDV, for example, is the most straightforward translation of preload, since it more than pressure relates to the actual stretch of the myocardium before systole. Aortic impedance can also be examined, using the effective arterial elastance proposed by Sunagawa and colleagues (46, 47). This is most commonly derived from the end-systolic pressure-to-SV ratio and is largely dictated by systemic resistance and heart rate (Fig. 6A). The coupling ratio of arterial elastance to end-systolic elastance generally falls between 0.7 and 1.2, a range where heart and vessels are optimally linked to provide the greatest work, power, and efficiency (6). The capacity to selectively manipulate heart versus vascular cells using genetic engineering of mice means that an analysis of the interaction between them can be quite important to the phenotype.

Energetics. P-V relations have also been extensively analyzed as a means to extract useful mechanoefficiency information. The total P-V area as first defined by Suga and colleagues (29) includes the external work of a P-V loop and “internal work” bounded by the left side of the loop, ESPVR, and DPVR. P-V area is linearly related to total myocardial oxygen consumption and has been useful for examining mechanisms by which energetics of the heart are altered by disease and molecular processes. Most applications of this analysis have been conducted in isolated hearts, where precise changes in the P-V area and global measures of oxygen consumption are feasible. This has been achieved in isolated mouse hearts as well (13, 32).

Other applications: normalized time-varying elastance. The preceding discussion has reviewed traditional uses of P-V relations, i.e., more comprehensively examined systolic and diastolic function of the heart, identified cardiac-loading interactions, and quantified energetic efficiency status. However, there are other parameters that are not a traditional focus but can be very important. A major example is analysis of the time course of ventricular elastance \( E(t) = \frac{P(t)}{[V(t) - V_e]} \). While the absolute magnitude of \( E(t) \) clearly varies with contractility and heart size (mouse is \( \sim 4,000 \times \) human) and its time course is influenced by heart rate, a binormalized curve (to both amplitude and time to peak) is remarkably conserved. This was first shown in patients encompassing a broad range of clinical heart diseases, heart rates, inotropic status, and other features (36). The human relation is very similar to mouse (10).

Two recent studies have revealed how normalized time-varying elastance \( E_{n(t)} \) can be profoundly altered by either genetic or pharmacological interventions to greatly impact systolic function. In both instances, more traditional measures such as \( \frac{dP}{dt_{max}} \) were little impacted. The first example is a genetic model involving the absence of myosin binding protein C. In these mice, the \( E_{n(t)} \) curve was profoundly abbreviated, peaking just before the onset of ejection and then declining (Fig. 6B). By contrast, \( E_{n(t)} \) was little altered in mice with a broad array of genetic manipulations that altered calcium cycling, signaling kinases, myofilament, and cytoskeletal proteins (31) (Fig. 6B). The impact of myosin-binding protein C on contraction and relaxation kinetics was subsequently explored by P-V relations, in a study that revealed the criticality of this protein to shortening of the diastolic filling period at faster heart rates and load dependence of relaxation (28). The second example pertains to a pharmaceutical study of a drug currently in clinical trials for heart failure. The drug, omecamtiv mecarbil, activates the myosin ATPase (24), enhancing the probability of forming a tightly bound cross bridge. This results in a prolongation of the systolic phase of \( E_{n(t)} \) without accelerating early contraction as would dobutamine (Fig. 6C).

Here too, \( \frac{dP}{dt_{max}} \) was unaltered by the drug, though ejection properties were. The P-V relation analysis proved central to defining the mechanism of action.

Conclusion

P-V analysis remains the most detailed and precise analytic approach to chamber heart function yet developed. Its introduction into mouse research has truly ignited interest from many investigators outside the biomechanics field, though some more sophisticated hemodynamic understanding is needed to employ it correctly. The more one appreciates the pitfalls and strengths, the more likely real insights will be derived. Most of the primary resources for this analysis are old, and to our knowledge there are no recent monograms summarizing the work for nonengineers. Hopefully, this review helped address that gap.

DISCLOSURES

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

AUTHOR CONTRIBUTIONS

Author contributions: O.H.C. and D.A.K. prepared figures; O.H.C. and D.A.K. edited and revised manuscript; O.H.C. and D.A.K. drafted manuscript; O.H.C. and D.A.K. edited and revised manuscript; D.A.K. approved final version of manuscript.

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AJP-Heart Circ Physiol • VOL 301 • DECEMBER 2011 • www.ajpheart.org


