Effect of Early Diastolic Loading on Myocardial Relaxation in the Intact Canine Left Ventricle

Srdjan Nikolic, Edward L. Yellin, Koichi Tamura, Takako Tamura, and Robert W.M. Frater

Early transmitral flow (MiF) patterns depend strongly on the rate of fall of measured left ventricular pressure ($P_m$) determined by both the active decay of pressure ($P_a$) due to myocardial relaxation and the increase in pressure due to stretch of passive elements during filling ($P_p$). This study was designed to uncouple passive forces from deactivation in order to reveal the instantaneous rate and duration of myocardial relaxation. We assumed a parallel combination of passive and active elements: $P_m(t,V)=P_a(t)+P_p(V)$, with no constraints on the form of $P_a(t)$, where $t$ is time and $V$ is ventricular volume. $P_p(V)$ was determined by a retrospective analysis of data obtained in 11 anesthetized dogs instrumented for volume clamping with a remote-controlled mitral valve, with left atrial and left ventricular micromanometers, and with an electromagnetic probe to measure MiF. The passive pressure-volume relation (both positive and negative portions) was determined by clamping at end-systolic volume or after various filling volumes, and fit to logarithmic functions. $P_a(t)$ was then calculated from $P_p(V)$ and $V(t)$ (integral of MiF). Time to end relaxation ($T_a$) was defined as time when $P_a=0$. During isovolumic relaxation, when $dP_a/dt=0$, $dP_m/dt$ is equal to the relaxation rate, $dP_p/dt$. In completely isovolumic relaxations, asymptote $P_a= -7\pm6$ mm Hg (thus, $P_a$ is 7 mm Hg $> P_m$) and $T_a=40\pm15$ msec, compared with 175$\pm53$ msec during normal filling. In high versus low inotropic state, $P_a$ at the beginning of filling was greater (18.1$\pm$6.1 vs. 12.2$\pm$3.9 mm Hg), and $T_a$ was shorter (170$\pm$42 vs. 228$\pm$43 msec). Active pressure $P_a(t)$ during filling is not an exponential function, and at any time, it was always greater after filling than in nonfilling beats, which indicates an increase in the relaxation duration. We conclude that myocardial relaxation is modulated by filling, which slows its rate and increases its duration, and is therefore a function of both ventricular volume and time. Such a mechanism may have an important role in regulating the diastolic pressure–volume relation. (Circulation Research 1990;66:1217–1226)

Early ventricular filling is directly related to the amplitude and rate of development of the atrioventricular pressure gradient. The ventricular component of the gradient is determined by 1) the amount and rate of decay of actively developed force (relaxation); 2) the elastic ventricular properties, that is, the magnitude of the restoring forces (due to storage of potential energy during contraction) and/or the ability of the ventricle to accept blood (compliance of stretched elements); and 3) the amount and rate of filling. For purposes of this study, we define myocardial relaxation as an active decay of force due to intrinsic myocardial processes, and we distinguish it from the measured ventricular pressure decline (chamber relaxation) because the latter is determined by both active force decay and passive elastic forces. Because of complex interconnections between the extracellular matrix and muscle fibers, early diastolic loading is due to elastic forces that are determined by absolute ventricular volume. Thus, diastolic loads may be either in the form of restoring forces in the wall created during the contractions below end-systolic volume or in the form of tensile forces at larger volumes reached during ventricular filling. In addition, at the onset of filling, isovolumic relaxation becomes auxotonic relaxation, and this may influence the process of active force decay.

In a previous study, we used a volume clamping technique to uncouple the active force from the
passive elastic component of chamber relaxation to
determine the positive and negative passive
diastolic pressure-volume relation. We have also shown
that when filling is eliminated, the pressure of a
completely isovolumically relaxing ventricle does not
follow an exponential form.

This study was designed to determine the time
course of myocardial relaxation in the normal filling
ventricle without any a priori assumptions on the
form of active force decay (e.g., exponential) and to
investigate the interaction between myocardial
relaxation and loads in early diastole. We retrospectively
analyzed the data from experiments in which we
clampped the ventricle at end-systolic and at different
end-diastolic volumes to separate effects of filling and
passive elastic forces on active pressure decay. We will
show that in the intact left ventricle, filling decreases
the rate and increases the duration of myocardial
relaxation and that the decay of active pressure is a
function of both time and ventricular volume.

Materials and Methods

Conceptual Approach

Because myocardial relaxation is not complete at
mitral valve opening, the instantaneous chamber
pressure reflects the continuously changing state of
dysequilibrium between active and passive forces. In
agreement with models of muscle mechanics, particu-
larly those containing parallel combinations of active
and passive elements, we assume that the
measured pressure is the sum of active and passive
components. Viscoelastic effects due to strain rate
and dynamic effects such as shape changes are
neglected in these definitions. The measured
pressure is a function of time and volume \[ P_m(t,V) \],
the passive component (passive pressure) is due to
elastic properties of the myocardium and is therefore
assumed to be only a function of volume \[ P_p(V) \], and
the active component (active pressure) is assumed to
be only a function of time \[ P_a(t) \]. Thus

\[ P_m(t,V) = P_a(t) + P_p(V) \] (1)

\[ P_a(t) = P_m(t,V) - P_p(V) \] (2)

To solve Equation 2, the passive pressure is
expressed as a function of time as follows: \[ P_p(V) \]
determined by the method of ventricular volume
clamping. \[ V(t) \] during filling is calculated from the
integral of measured mitral flow, and \[ P_p \] is then
expressed as a function of time by eliminating volume
as a parameter. Equation 2 can then be solved
experimentally without any assumptions or con-
straints on the form of \[ P_a(t) \]. In essence, we are
testing the hypothesis that the decay of active
pressure is a function of only time.

The rate and duration of myocardial relaxation are
determined as follows: After volume clamps, there is
a period of isovolumic relaxation so that \[ \frac{dP_a}{dt} = 0 \]
and

\[ \frac{dP_m}{dt} = \frac{dP_a}{dt} \] (3)

Thus, the rate of myocardial relaxation is determined
from the measured pressure during volume clamps.
The duration of myocardial relaxation is defined as
the time when the active pressure, \[ P_a \], becomes zero.
Note that the concept of duration of myocardial
relaxation, based on time to end relaxation assumes
that the active component of pressure can become
zero and that there is no detectable pressure due to
residual activation.

Experimental Approach

Volume clamping. Ventricular diastolic volume was
controlled with a remote-controlled mitral valve (Fig-
ure 1) in experiments described in detail elsewhere;
we will review only the most important aspects. A
modified prosthetic valve was implanted in series
with an electromagnetic flow probe in the mitral
anulus. A controller was triggered by a suitable
physiological signal, and after a programmed delay, it
rapidly closed the valve 1) during systole for a
completely isovolumic diastole, that is, total
occlusion (end-systolic volume clamping); and 2) at any
time after normal filling has started, that is, filling
occlusion.

Animal preparation. Eleven adult mongrel dogs
(25.8 ± 1.2 kg) were premedicated with atropine (0.01
\( \mu \)g/kg i.v.). Anesthesia was induced with thiopental
sodium (15 mg/kg i.v.), followed by intubation and
artificial ventilation at 100% O2 with a pressure-
controlled respirator. Fentanyl (5–10 \( \mu \)g/kg) was
administered every 30 minutes and supplemented
with vecuronium (0.1 mg/kg). After a midline stern-
otomy and left thoracotomy at the fourth intercostal
space, the heart was supported in a pericardial
cradle. Heart rate was controlled by crushing the
sinoatrial node and by pacing to keep the heart rate
at approximately 100 beats/min. During standard left
cardiopulmonary bypass with a bubble oxygenator, the left atrium was opened and the mitral occluder was implanted. Micromanometers (Millar Instruments, Houston, Texas) were placed in the left ventricle and left atrium. The flow probe cable was brought out through the atrial appendage, the atrium was closed, and the dog was weaned from bypass. A flow probe was placed around the cleaned ascending aorta. Flows were measured with a two-channel flowmeter (Carolina Medical Electronics, Burlington, North Carolina). Physiological signals were recorded at high speed (100 mm/sec) on a photographic recorder. Arterial pH, Pco2, and Po2 were measured periodically and maintained normal. Data were recorded with the chest and pericardium open, when the dog was in a stable steady state, and with the respirator turned off.

Great care was taken to accurately measure left ventricular diastolic pressure. The side lumen of the micromanometer was connected to a Statham gauge (Gould, Cleveland, Ohio) positioned at the midventricular level, and the baseline was checked frequently. At the end of the experiment, the heart was arrested in diastole with potassium chloride, vented to the atmosphere, and calibrations and baselines were checked. The left ventricle, including septum, was weighed in eight dogs.

Protocols. A typical run was as follows: 10 control beats, a mitral valve occlusion during systole, 10 normal beats to return to control condition, a mitral valve occlusion after a small amount of filling, 10 normal beats, a mitral occlusion slightly later in time, etc. Each occlusion was held for only one beat. The inotropic state was varied with different rates of dobutamine infusion (maximum, 11 μg/kg/min), and ventricular volume was varied by infusing blood from the oxygenator via the femoral artery cannula with a roller pump.

Data Analysis

Calculations. The oscillographic records were digitized with a sonic digitizer (model GP-7, Science Accessories, Southport, Connecticut) coupled to an IBM-PC. Changes in volume were calculated from the integrals of aortic and mitral flow. We did not measure absolute ventricular volumes, so all volumes were determined relative to end-systolic volume. Therefore, we assumed that the end-systolic volume remained constant throughout each run, and subsequent volumes were determined from inflow and outflow. Thus, it was important that there should be no change in hemodynamic state during a run. This was tested by visually examining the control beats before each occlusion and accepting only those runs in which there was no detectable change in the control pressures and flows.

Determination of the Passive Diastolic Pressure–Volume Relations and Equilibrium Volume

The pressure-volume coordinates from a complete run (total occlusion and various filling-occlusion beats), shown in Figures 2B and 2C, were used to determine the negative and positive portions of the entire passive diastolic relation. The method has been previously described in detail. Briefly, pressure-volume coordinates for the passive, fully relaxed ventricle were generated from the minimum pressure points reached during occlusions (dots in Figure 2C) and from their corresponding volumes (dots in Figure 2B) to give the entire passive pressure-volume relation (broken line in Figure 2D) relative to the end-systolic volume. Points in the region of negative pressures occur at volumes below the equilibrium volume (V0) and are evidence of elastic recoil and diastolic suction; they also mark the end of relaxation because pressure has stopped falling.

We used a logarithmic approach to characterize the passive pressure-volume relation:

\[ P_p = -S_p \ln \left( \frac{V_m - V}{V_m - V_0} \right) \quad \text{for} \quad V > V_0 \]

\[ P_p = S_p \ln \left( \frac{V - V_0}{V_0 - V_0} \right) \quad \text{for} \quad V < V_0 \]

where \( S_p \) and \( S_r \) are stiffness coefficients in the positive and negative planes, respectively; \( V_0 \) is the equilibrium volume (at zero transmural pressure), read directly from the pressure-volume points (Figure 2D); \( V_m \) is the maximum volume before irreversible yielding of tissue; and \( V_d \) is the minimum attainable volume. We determined \( V_m \) and \( V_d \) from left ventricular weights in eight dogs as described previously. Because the absolute values of \( V_m \), \( V_0 \) and \( V_d \) are not important for the purpose of this study, we calculated \( V_m - V_0 \) and \( V_0 - V_d \) values from the best fit of data points (as described in Reference 3) in three additional dogs and determined relative \( P_p \) for later use in the determination of \( P_s \).

Determination of Active Pressure and Duration of Myocardial Relaxation

To calculate the active pressure from Equation 2, we first used the time variation of volume (Figure 2B) and the passive pressure variation with volume to eliminate volume as a parameter and derive \( P_p(t) \) (broken line in Figure 2E). \( P_p(t) \) was then subtracted from the measured pressure \( P_m(t) \) to give the experimentally determined time variation of myocardial relaxation, \( P_a(t) \). The time to end relaxation, \( T_m \), is then defined as the time when \( P_a = 0 \), that is, when the measured and passive pressures are equal (Figure 2E).

Determination of the Rate of Myocardial Relaxation

We reasoned as follows: if active pressure is a function of only time and is not affected by filling, then regardless of the mode of relaxation (isometric or relengthening) at equal times after a reference time, the rate of relaxation \( (dP_s/dt) \) should be equal. Thus, we compared \( dP_s/dt \) (i.e., \( dP_s/dt \)) of the isovolumic portions of the pressure curves after various amounts of filling at equal times after the onset of normal filling (e.g., lines a and b in Figure 2C). Active force decay may be influenced by absolute length (volume) as well as by the mode of relaxation.
We therefore related dPm/dt at the beginning of the isovolumic relaxation after various amounts of filling (e.g., lines b and c in Figure 2C), with minimum pressures obtained after the relaxation is completed at the corresponding volume (minimum left ventricular pressure [LVPmin] of occlusion, e.g., points 3 and 4 in Figure 2C), as the index of absolute volume (Figure 2D). It is important to note that these pressures represent compressive or tensile elastic forces in the ventricular wall at the volumes below or above equilibrium volume. When the ventricular volume is equal to the equilibrium volume, LVPmin of an occlusion is zero.

**FIGURE 2.** Graphs showing the passive pressure-volume relation and the active pressure in diastole. Panel A: Control mitral flow [MiF(t)]. Panel B: Ventricular volume [V(t)]. Panel C: Pressure [Pm(t)]. Traces of MiF, V, and P are drawn as solid lines. Numbers 1–6 in panel B mark the times of mitral valve occlusions. Number 7 is the end-diastolic point. Integration of the flow curve to the time of occlusion gives the change in ventricular volume relative to control end-systolic volume (VES). Broken lines in panel C denote the pressure traces associated with each volume clamp, and numbered points correspond to the minimum pressure during each occlusion. These minimum pressures are assumed to be the values at end relaxation. Pc is the value of minimum pressure for an end-systolic volume clamp. Thin lines (a,b,c) represent dPm/dt during isovolumic relaxation at the same time (a,b) and at different times (a,c). Panel D: The pressure-volume relation determined by eliminating time as a parameter from panels B and C. Dashed line is a passive pressure-volume relation above and below the equilibrium volume V0. Panel E: Combined pressure-volume diagram of panel D and the volume-time diagram of panel B to obtain the measured and passive pressures as functions of time (solid and broken lines, respectively). From their difference at each instant (Equation 1), we determined active pressure during filling (dotted line). Tc in panel E is the time of the end of myocardial relaxation.

**Determination of the Time Constant of Isovolumic Relaxation**

The time constant, T, of isovolumic ventricular chamber relaxation was calculated as described previously5 on the assumption that the measured pressure in the completely isovolumic ventricle is reasonably close to an exponential:

\[
P_m = (P_0 - P_s) \exp(-t/T) + P_s \tag{6}
\]

where P0 is the pressure at the onset of isovolumic relaxation (taken at dPm/dt<sub>max</sub>), and Ps is the pressure asymptote, determined experimentally from the completely isovolumic relaxation (Figure 2C).
Active Component of Pressure at the Onset of Filling

When low and high inotropic runs were analyzed separately (Figure 3, lower panel), there was no difference between them: for the low inotropic state $P_{MVO} = -0.72V_{es} + 8.2$ ($r=0.75, n=20$), and for the high inotropic state $P_{MVO} = -0.70V_{es} + 10.4$ ($r=0.90, n=23$). The inverse relation between pressure at the onset of filling and end-systolic volume ($V_{es}$) was stronger in the low inotropic state ($A_1=-0.79, A_2=8.2$) than in the high inotropic state ($A_1=-0.70, A_2=8.2$).

Table 1 summarizes the hemodynamic conditions during 68 runs in 11 dogs. In addition, Table 1 also shows the data obtained during low and high inotropic states created by the concentration of infused dobutamine and separated by a difference in $dP_m/dt_{max}$ greater than 30%.

In the normal filling beats the pressure at the onset of flow (mitral valve opening) was $P_{MVO}=9.1±5.2$ mm Hg. The minimum pressure in the completely isovolumic beats, that is, with total occlusions, was $P_T=P_m=-6.8±6.3$ mm Hg. By using Equation 2, the active pressure at the beginning of filling was thus calculated to be $P_{MVO}=16.1±6.4$ mm Hg. In the runs with a different inotropic state, $P_{MVO}$ was not different ($8.7±4.5$ vs. $8.3±5.3$ mm Hg), but due to the difference in $P_a$ ($-3.6±3.7$ vs. $-9.7±8.3$ mm Hg), the active pressure at the beginning of filling was larger in the high inotropic state: $12.2±3.9$ mm Hg in low versus $18.1±6.1$ mm Hg in high.

To test if the active pressure at the onset of filling is a function of the end-systolic volume, we correlated $P_{MVO}$ and relative end-systolic volume, $V_{es}^*$ (where $V_{es}^*=V_{es}-V_0$). As seen in Figure 3, upper panel, the correlation is strong, $P_{MVO}=-0.79V_{es}^*+8.2$ ($r=0.80, n=68$), and the active pressure at the beginning of filling increases as ventricular volume decreases. When low and high inotropic runs were analyzed separately (Figure 3, lower panel), there was no difference between them: for the low inotropic state $P_{MVO}=-0.72V_{es}^*+8.2$ ($r=0.75, n=20$), and for the high inotropic state $P_{MVO}=-0.70V_{es}^*+10.4$ ($r=0.90, n=23$). The inverse relation between pressure at the onset of filling and end-systolic volume ($V_{es}^*$) was stronger in the low inotropic state ($A_1=-0.79, A_2=8.2$) than in the high inotropic state ($A_1=-0.70, A_2=8.2$).

Table 1. Hemodynamic Data and Results (Mean±SD) for 11 Dogs

<table>
<thead>
<tr>
<th></th>
<th>All runs</th>
<th>Low</th>
<th>High</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>HR (beats/min)</td>
<td>107±20</td>
<td>102±20</td>
<td>110±20</td>
<td>NS</td>
</tr>
<tr>
<td>PLVP (mm Hg)</td>
<td>116±25</td>
<td>102±15</td>
<td>130±74</td>
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<tr>
<td>SV (ml)</td>
<td>23.6±7.7</td>
<td>20.1±5.9</td>
<td>27.1±9.5</td>
<td>0.01</td>
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<tr>
<td>PMIF (ml/sec)</td>
<td>138±51</td>
<td>126±33</td>
<td>160±61</td>
<td>0.02</td>
</tr>
<tr>
<td>LVP$_ao$ (mm Hg)</td>
<td>4.6±2.7</td>
<td>5.0±2.3</td>
<td>4.5±2.3</td>
<td>NS</td>
</tr>
<tr>
<td>$P_m$ (mm Hg)</td>
<td>9.1±5.2</td>
<td>8.7±4.5</td>
<td>8.3±5.3</td>
<td>NS</td>
</tr>
<tr>
<td>$dP/dt_{max}$ (mm Hg/sec)</td>
<td>2,560±870</td>
<td>1,750±520</td>
<td>3,160±700</td>
<td>0.001</td>
</tr>
<tr>
<td>$dP/dt_{min}$ (mm Hg/sec)</td>
<td>1,705±570</td>
<td>1,480±380</td>
<td>2,180±490</td>
<td>0.002</td>
</tr>
<tr>
<td>IVRP (msec)</td>
<td>58±25</td>
<td>60±14</td>
<td>47±17</td>
<td>0.02</td>
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<tr>
<td>$P_a$ (mm Hg)</td>
<td>-6.8±6.3</td>
<td>-3.6±3.7</td>
<td>-9.7±8.3</td>
<td>0.004</td>
</tr>
<tr>
<td>T (msec)</td>
<td>30.8±10.5</td>
<td>34.7±14.4</td>
<td>25.1±7.4</td>
<td>0.05</td>
</tr>
<tr>
<td>T-P$_{min}$ (msec)</td>
<td>170±48</td>
<td>182±33</td>
<td>163±51</td>
<td>NS</td>
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<tr>
<td>$T_v$ (msec) (TO)</td>
<td>105±31</td>
<td>125±30</td>
<td>98±28</td>
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<tr>
<td>$T_v$ (msec) (N)</td>
<td>238±67</td>
<td>290±57</td>
<td>216±40</td>
<td>0.05</td>
</tr>
<tr>
<td>n</td>
<td>68</td>
<td>20</td>
<td>23</td>
<td></td>
</tr>
</tbody>
</table>

HR, heart rate; PLVP, peak left ventricular pressure (LVP); SV, stroke volume; PMIF, peak mitral flow; LVP$_ao$, end-diastolic LVP; $P_m$ (mm Hg), measured LVP at mitral valve opening; $dP/dt$, rate of change of measured LVP; IVRP, isovolumic relaxation period; $P_a$, pressure asymptote after mitral valve occlusions; T, time constant of an assumed exponential decay of LVP; T-P$_{min}$, time to LVP minimum in normal filling measured from the time of $dP/dt_{min}$; $T_v$, time to end relaxation measured from time of $dP/dt_{max}$ during total occlusions of the mitral valve (TO), or calculated during normal filling cycles (N).

Results

Active Component of Pressure at the Onset of Filling

Table 1 summarizes the hemodynamic conditions during 68 runs in 11 dogs. In addition, Table 1 also shows the data obtained during low and high inotropic states created by the concentration of infused dobutamine and separated by a difference in $dP_m/dt_{max}$ greater than 30%.

In the normal filling beats the pressure at the onset of flow (mitral valve opening) was $P_{MVO}=9.1±5.2$ mm Hg. The minimum pressure in the completely isovolumic beats, that is, with total occlusions, was $P_T=P_m=-6.8±6.3$ mm Hg. By using Equation 2, the active pressure at the beginning of filling was thus calculated to be $P_{MVO}=16.1±6.4$ mm Hg. In the runs with a different inotropic state, $P_{MVO}$ was not different ($8.7±4.5$ vs. $8.3±5.3$ mm Hg), but due to the difference in $P_a$ ($-3.6±3.7$ vs. $-9.7±8.3$ mm Hg), the active pressure at the beginning of filling was larger in the high inotropic state: $12.2±3.9$ mm Hg in low versus $18.1±6.1$ mm Hg in high.

To test if the active pressure at the onset of filling is a function of the end-systolic volume, we correlated $P_{MVO}$ and relative end-systolic volume, $V_{es}^*$ (where $V_{es}^*=V_{es}-V_0$). As seen in Figure 3, upper panel, the correlation is strong, $P_{MVO}=-0.79V_{es}^*+8.2$ ($r=0.80, n=68$), and the active pressure at the beginning of filling increases as ventricular volume decreases. When low and high inotropic runs were analyzed separately (Figure 3, lower panel), there was no difference between them: for the low inotropic state $P_{MVO}=-0.72V_{es}^*+8.2$ ($r=0.75, n=20$), and for the high inotropic state $P_{MVO}=-0.70V_{es}^*+10.4$ ($r=0.90, n=23$). The inverse relation between pressure at the onset of filling and end-systolic volume ($V_{es}^*$) was stronger in the low inotropic state ($A_1=-0.79, A_2=8.2$) than in the high inotropic state ($A_1=-0.70, A_2=8.2$).

Figure 3. Upper panel: Relation between active pressure at the time of mitral valve opening ($P_{MVO}$) and relative end-systolic volume ($V_{es}^*$) for all 11 dogs combined. $A_0$, y intercept; $A_1$, slope. Lower panel: Same relation as in upper panel for the runs in low and high inotropic states.
onset of filling and volume is a consequence of the increase in elastic recoil at low end-systolic volumes.\textsuperscript{3,5}

**Rate and Duration of Myocardial Relaxation**

To evaluate the influence of filling on myocardial relaxation we calculated the rate and duration of myocardial relaxation in beats with different amounts of filling. In the completely isovolumic beats $\frac{dP_a}{dt}=0$ at $P_a$, and relaxation is complete (line a, Figure 2C). At that same time (line b, Figure 2C), $\frac{dP_a}{dt}$ was always nonzero. As an illustration for this consistent finding, which is quite obvious from Figure 2C, we calculated $\frac{dP_a}{dt}$ during filling beats after the time when $\frac{dP_a}{dt}$ in nonfilling beats had reached zero. With pooled data from all 11 dogs, at $78\pm38$ msec into the filling period, after $4.5\pm3.2$ ml (19\% of the filling volume) had entered the ventricle, $\frac{dP_a}{dt}=96\pm54$ mm Hg/sec. Thus, filling slows the rate of myocardial relaxation. The same conclusion is valid if we compare $\frac{dP_a}{dt}$ after occlusion later in diastole (point 4, Figure 2A; line c, Figure 2C) with early occlusions (points 2 and 3, Figure 2A; pressures after points 2 and 3, Figure 2C). It is obvious from Figure 2C that myocardial relaxation is always completed in early occlusion beats before late occlusions. For example, after $110\pm42$ msec when $8.7\pm5.1$ ml (37\% of the filling volume) had entered the ventricle, $\frac{dP_a}{dt}$ had decreased to $74\pm33$ mm Hg/sec, while $\frac{dP_a}{dt}$ in nonfilling beats was zero.

$\frac{dP_a}{dt}$ at the time of occlusion in the filling-occlusion runs ($\frac{dP_a}{dt}*$) correlated strongly with minimum pressures ($LVP_{min}$) obtained in all 11 dogs (Figure 4, top panel). The same relations for low and high inotropic states are presented in Figure 4, middle and bottom panels. Table 2 shows the linear regression parameters for each dog. Note that all x axis intercepts are positive and have similar values.

The precise time to the end of myocardial relaxation was determined from Equation 2. Active pressure reached zero in the nonfilling beats $40\pm15$ msec after the time when the mitral valve would have opened if it were not occluded (or $105\pm31$ msec after $dP_a/dt_{min}$); myocardial relaxation ended $175\pm53$ msec after the onset of filling in the normal filling beats (or $238\pm67$ msec after $dP_a/dt_{min}$). The time to end relaxation was less in high than in low inotropic states: in nonfilling beats, $49\pm31$ vs. $66\pm23$ msec after the time of what would be the beginning of normal filling (or $98\pm28$ vs. $125\pm30$ msec after $dP_a/dt_{min}$); in normal filling, $170\pm42$ vs. $228\pm43$ msec after the beginning of filling (or $216\pm40$ vs. $290\pm57$ after $dP_a/dt_{min}$).

The active component of pressure ($P_a$) does not decay asymptotically to zero as in an exponential model; therefore $P_a$ cannot be exponential, and the time of the end of myocardial relaxation ($T_{el}$) is a well-defined point (Figure 2E).

**Discussion**

The increasing awareness that diastolic dysfunction is present in many disease states, and frequently precedes systolic dysfunction,\textsuperscript{13} has resulted in a growing emphasis on the study of left ventricular relaxation. This trend has been accelerated by the ability to accurately and noninvasively measure transmural flow velocity. Thus, an understanding of the interaction of early filling and relaxation becomes of great clinical and experimental interest and has been studied in the isolated muscle preparation,\textsuperscript{4,14-20} in isolated and intact mammalian hearts,\textsuperscript{3,5,21-29} and in patients.\textsuperscript{5,7,13,30} Investigations of chamber relaxation have attempted to treat it as a process by formulating mathematical models and to characterize it with an appropriate index. Weiss et al.\textsuperscript{20} described the measured left ventricular pressure curve ($P_m$) during the
TABLE 2. Linear Relations Between $dP_v/dt^e$ and $LVP_{min}$

<table>
<thead>
<tr>
<th>Dog</th>
<th>$A_0$</th>
<th>$A_1$</th>
<th>r</th>
<th>p</th>
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$LVP_{min}$, minimum left ventricular pressure; $A_0$, y intercept; $A_1$, slope; r, regression coefficient.

... passive stress from the time variation of left ventricular passive filling pressure.\textsuperscript{6,7} This is in disagreement with the results presented herein, because $P_v$ during filling cannot be modeled as an exponential. Furthermore, the assumption of exponential decay of myocardial relaxation to zero leads to an inherent contradiction: the calculated passive pressure is always zero during isovolumic relaxation when $P_m$ and $P_v$ are assumed equal; thus, regardless of hemodynamic state and myocardial chamber properties, the end-systolic volume is always equal to the equilibrium volume. This approach also makes two additional fallacious assumptions: 1) it assumes the absence of restoring forces, and 2) it neglects the possibility that diastolic loading will affect myocardial relaxation. The technique of volume clamping has allowed us to show conclusively that the normal canine ventricle reaches an end-systolic volume that is below the equilibrium volume and that internal restoring forces lead to the negative phase of the passive pressure-volume relation (Equation 5).\textsuperscript{2} Our data also indicate that ventricular filling modifies the intrinsic process of myocardial relaxation.

Myocardial Relaxation and Diastolic Loads

The process of myocardial relaxation in the intact ventricle is subject to diastolic loading due to elastic forces and to stretch imposed on the myocardial fibers during filling. The change in volume due to ventricular filling introduces time variations in passive elastic forces, that is, in time-varying loads. The control of filling in diastolic volume clamp experiments makes it possible to uncouple the active and passive components of measured diastolic pressure because, at constant volume, passive forces are constant. However, the stretch of still active myocardium introduced by ventricular filling can affect the process of relaxation itself.\textsuperscript{4,8}

We have shown (Figure 2C) that myocardial relaxation after stretch due to filling ends significantly later than the completely isovolumic relaxation in nonfilling diastoles. Myocardial relaxation in the normal filling beat was approximately 130 msec longer than in the completely isovolumic beat (Table 1). Since the average duration of diastole in this study was 310 msec, myocardial relaxation in the beats without filling ended at 17% of diastole, whereas it ended at 59% in the normal beats. In low inotropic states, myocardial relaxation was completed later than in runs at high inotropic states; 21% vs. 15% of diastolic time in nonfilling beats, 75% vs. 54% in normal filling. We may thus conclude that in the intact heart filling slows the rate of myocardial relaxation and increases its duration. In addition, myocardial relaxation in high inotropic states is completed earlier than in low inotropic states but is still present during half the filling period.

It has been concluded that relaxation in the normal filling ventricle is completed at approximately 3.5 time constants after the onset of isovolumic relaxation.\textsuperscript{25} It is more appropriate to state that, in the exponential...
model, relaxation will be 97% complete at 3.5 time constants because an exponential decay never reaches the asymptote. In contrast to the exponential, the experimentally determined active pressure in our study does not approach its final value asymptotically (Figure 2E); it reaches zero at a well-defined point T_{rc}. Therefore, we think that more realistic models of relaxation should be explored.

The model of elastic restoring forces (internal loads) that relates the development of negative pressure in the ventricle to contraction below the equilibrium volume and is dependent on absolute volume of the ventricle^{3,31} explains the diastolic pressure-volume relation, but it is unable to account for the rate and duration of relaxation. In the absence of damping or inertia, the rate at which parallel elasticity exerts its recoil is determined solely by the rate of deactivation of the myocardial sarcomeres. How then can internal loading explain the findings in isolated muscle and in the intact heart that the relengthening rate is a function of the extent of shortening^{1,14,24,28} and minimum end-systolic length?^{20}

We have shown that relengthening rate (filling rate) is a function of both relaxation rate and filling pressure.\(^1\) In the physiologically sequenced isolated muscle the “filling pressure” is constant\(^1,11\); therefore, internal restoring forces must have influenced the rate of deactivation. Similarly, Cailliet and Crozatier\(^28\) demonstrated that internal restoring forces had an independent effect on relengthening. We may thus speculate that increasing the amount of shortening increases the amount of stored energy and that deactivation rate is, in turn, influenced by internal loading conditions.

The time constant of relaxation has been found to depend on the same passive mechanisms that are responsible for the magnitude of the end-diastolic pressure, that is, with the passive ventricular properties that determine the diastolic pressure-volume relation.\(^21,22\) In addition, our results confirm that there may be an interaction of restoring forces with deactivation. First, active pressure at the time of mitral valve opening is a function of absolute volume (Figure 3, upper and lower panels); second, \(dP/dt\) is related to the amplitude of restoring forces (Figure 4, top, middle, and bottom panels); and third, relaxation is completed when the LVP_{min} of occlusion is positive at volumes in the range outside the influence of restoring forces. Note also that the amplitude of restoring forces (LVP_{min} of occlusions) increases as volume decreases (Reference 3, Figure 1). Thus, it seems reasonable to conclude that relaxation in the ventricle reflects the interaction between active and passive elements and that the process of myocardial relaxation is modulated by restoring forces and is therefore volume dependent.

**Indexes of Relaxation: Isolated Muscle Versus Intact Ventricle**

In the physiologically sequenced isolated muscle preparation it was found that load clamps imposed late in the isometric relaxation phase, or during the isotonic phase, resulted in an increase in the rate of muscle relengthening.\(^1,14,17\) That is, that relaxation is load dependent.\(^15\)

The hypothesis of load-dependent relaxation in diastole was advanced in a teleological argument that early filling is facilitated by diastolic loads after the onset of filling\(^16\) and that the load due to filling would produce more filling. This argument was deduced solely on the basis of the results of isolated muscle experiments extrapolated to the normal ventricular diastole. It is now reasonable to ask: can we compare these results with the data from intact ventricles?

Perhaps not. Myocardial relaxation in the intact heart starts when both pressure and length are changing. Pressure may be decreasing in late systole while the ventricle is still ejecting, and late outflow could be produced by blood momentum (leading to chamber shape change) rather than as a result of muscle shortening.\(^32\) There may even be muscle length changes due to internal elastic forces and shape changes. It is possible that there is never a truly isometric relaxation of active elements in the wall due to internal elastic forces. The greatest difference between the isolated muscle and intact ventricle is at the onset of filling. The ventricle always fills while pressure is falling and the myocardium is relaxing; there is never an isotonic phase of filling; and once filling starts, the pressure is determined by a complex interplay of active and passive forces. Furthermore, the rate of filling cannot be used as an independent index of relaxation because it is determined by the time-varying atrioventricular pressure difference, which in turn is profoundly influenced by left atrial pressure at the time of mitral valve opening.\(^1\) Thus, in the intact ventricle, only the isovolumic pressure decay can be related to the process of relaxation; there is no analogy to isotonic relengthening of isolated muscle. Our results indicate that filling of the ventricle prolongs the duration of myocardial relaxation, which might correspond to an augmentation of the isometric force after isotonic reextension in afterloaded isotonic contractions (Figure 4 in Reference 18), that is, delayed relaxation. Because our data on active pressure decay from the intact ventricle, which alone is capable of volume clamping, do not correspond to the data on relengthening from the physiologically sequenced isolated muscle, we urge caution in the data extrapolation obtained in such models to the interpretation of the underlying mechanism of ventricular filling.

**Conclusion**

Late systolic loads have been shown to lead to the premature onset and increased rate of ventricular pressure. But this occurs at a relatively high level of end-systolic activation and/or activation decay, and this effect may not be present at the time of mitral valve opening, when relaxation is associated with low levels of activation and activation decay.\(^19,33\) Our
findings indicate that diastolic loading and relengthening in the intact ventricle actually slows the process of myocardial relaxation. In addition, relaxation apparently decays at very similar volumes, greater than the equilibrium volume (Figures 3 and 4). We can thus conclude that the process of active tension decay in diastole is volume dependent. According to our results, this mechanism will include the dependence of myocardial relaxation on absolute volume, its interaction with internal restoring forces, and its modulation by stretch, that is, by filling. The experimental data lead us to reject the assumption that the active component of pressure decline depends only on time and rewrite Equation 1:

$$P_{m}(t,V) = P_{d}(t,V) + P_{p}(V)$$

(7)

We also want to emphasize that this conclusion is consonant with the length-dependent activation described in isolated cardiac muscle.34

Thus, observations that the normal diastolic pressure-volume relation is unaffected by heart rate and contractility may have to be explained by a volume-dependent regulation of myocardial relaxation in diastole and, perhaps, with a hypothesis of optimal coupling of these ventricular mechanisms with active and passive properties of the atrium.35 Further investigations may be required to determine if these mechanisms are part of normal left ventricular diastolic function.

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References


KEY WORDS • diastole • relaxation • ventricular filling • pressure-volume relation
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S Nikolic, E L Yellin, K Tamura, T Tamura and R W Frater

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