Verapamil Preserves Myocardial Performance and Energy Metabolism in Left Ventricular Hypertrophy Following Ischemia and Reperfusion

Phosphorus 31 Magnetic Resonance Spectroscopy Study

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While calcium entry blockers have a beneficial influence on the postischemic recovery of the nonhypertrophied heart, their influence on the hypertrophied heart has not been determined. The aim of this study was to assess postischemic recovery of myocardial performance and energy metabolites in rat hearts with left ventricular hypertrophy pretreated either chronically or acutely with verapamil. Left ventricular hypertrophy was induced by supraprenal constriction of the abdominal aorta. Hemodynamics and phosphorus 31 magnetic resonance spectra were monitored simultaneously in the isolated hearts during control perfusion, after 30 minutes of global ischemia, and after 30 minutes of reperfusion. All hypertrophied hearts had significantly higher rate-pressure products than normal hearts. Compared with normal hearts, oxygen consumption was significantly lower in all hypertrophied hearts, especially untreated hypertrophied hearts. Also, before ischemia all normal or hypertrophied hearts (treated or untreated) began with comparable phosphorylation potentials (i.e., the supply of energy was not significantly different). Postischemic recovery was not related to energy supply-oxygen demand before onset of ischemia. Furthermore, it was not related to energy levels or intracellular pH during ischemia. For postischemic recovery, the rate-pressure product was 40±5% in the hypertrophied heart, 83±5% in the normal, 100±3% in the hypertrophied heart chronically treated with verapamil, and 82±5% in the hypertrophied heart acutely treated with verapamil. The degree of recovery was related to coronary flow both before and after ischemia. The latter is important for flushing deleterious metabolites and ions from the interstitial space as well as for delivery of oxygen and substrate to the myocardium. (Circulation 1989;80:1837–1845)

Left ventricular hypertrophy caused by systemic hypertension in association with coronary ischemic heart disease has been recognized as a major risk factor for sudden death, postinfarction heart failure, and cardiac rupture.1–4 Myocardial infarct size in a certain area at risk is larger in hypertensive dogs with left ventricular hypertrophy compared with control dogs,5,6 and the ischemically injured, hypertrophied myocardium is more susceptible to rhythm and conduction disturbances.7 Several factors might contribute to this increased susceptibility of hypertrophied myocardium to ischemia including structural and functional abnormalities of the coronary vessels,8–10 which may reduce nutritional support to and metabolic removal from the ischemic myocardium. The increased cell volume to surface area ratio of the hypertrophied myofibers11 and the increased intercapillary distance12 tend to complicate exchange of gases, metabolites, and ions between the intracellular and extracellular spaces.

Verapamil, a slow channel calcium entry blocker, has been shown to reduce infarct size,13,14 hemodynamic deterioration of the ischemically injured myocardium,15 and ST-segment elevation during ischemia15,16 and to increase perfusion of the injured
myocardium in normal dog hearts. Preservation of high-energy phosphate stores and sarcolemmal enzymatic activity by verapamil during ischemia has been documented. Deleterious disturbances of transmembranous electrolyte exchange and intracellular depletion of adenosine triphosphate (ATP) caused by ischemic cell damage and reperfusion might be prevented. Because many infarcts are now being reperfused by thrombolytic and other interventions, it is important to determine whether verapamil protects the hypertrophied myocardium during an episode of ischemia and subsequent reperfusion.

Therefore, the aims of the current study were 1) to compare the posts ischemic recovery of mechanical performance and high energy phosphate stores in chronic pressure overload left ventricular hypertrophy with normal hearts and 2) to determine whether verapamil, administered either acutely or chronically, provides protection of the hypertrophied myocardium during ischemia and reperfusion.

Methods

Animals

Sprague-Dawley rats, weighing 300–350 g, were used. Normal healthy rats served as controls (n=12). Left ventricular hypertrophy was caused by constriction of the suprarenal abdominal aorta. During pentobarbital anesthesia, the suprarenal abdominal aorta was banded, resulting in a lumen constriction of approximately 50%, which caused chronic pressure overload of the left ventricle. Animals were randomly assigned to three groups. One group of the aortic-constricted rats received no verapamil treatment (n=18); a second group received 1.2 g/l verapamil in drinking water (n=12) starting after surgery until day killed; and a third group received 1.2 g/l verapamil in drinking water for 3 days before being killed (n=11). Not all experiments were carried out on the total numbers because of technical problems with magnetic resonance spectroscopic (MRS) signal acquisition. Four rats from each group were used for freeze-clamping to determine ATP and creatinine (Cr) concentrations. All rats were kept in the same room and given food and water ad libitum. The rats were killed 3 weeks after aortic banding. After successful surgery resulting in uniform left ventricular hypertrophy (35–45% increase in dry heart wt/body wt), no animals were rejected from the study. All work was done in accordance with guidelines from the animal research committee of the University of California San Francisco.

Perfused Heart Model

The rats were anesthetized with ether, and the hearts were excised rapidly through a midline sternotomy and perfused within 30–60 seconds. The isolated heart, paced by a Medtronic model 5320 (Quincy, Massachusetts) pulse generator at a constant rate of 220 beats/min, was perfused by a modified Langendorff method. Because coronary flow in left ventricular hypertrophy associated with spontaneously hypertensive rats has been shown to be constant at perfusion pressures between 140 and 190 cm H2O, all hearts were perfused with a perfusion pressure of 140 cm H2O during a 15–20-minute period of steady-state control. The preparation was stable for 2 hours with this system. Then, the cannula perfusing the aorta was clamped for 30 minutes, producing global ischemia. Subsequently, the heart was perfused under the same conditions as during the control period. Pressure measurements were obtained from a cannula inserted through the left atrial appendage into the left ventricular cavity and connected to a Statham P23Db (Gould, Cleveland, Ohio) pressure transducer.

Perfusion Conditions

The hearts were perfused with a modified Krebs-Henseleit solution containing 117 mM NaCl, 4.3 mM KCl, 2.0 mM CaCl2, 1.2 mM MgCl2, 0.1 mM K2HPO4, 25 mM NaHCO3, 0.5 mM NaEDTA, 15 mM glucose, and 10 units/l insulin. The medium was bubbled with 95% O2-5% CO2 at 35°C; the temperature was raised subsequent to flushing with gases to dissolve maximum oxygen in the perfusate. The oxygen tension of the perfusate was maintained between 650 and 700 mm Hg.

Physiologic Measurements

Oxygen consumption was determined as follows: Arterial samples were aspirated from the aortic chamber, and venous samples were drawn from a catheter introduced into the right ventricular outflow tract for oxygen measurements (Corning model 165/2 gas analyzer, Oberlin, Ohio). PaO2 was measured and the oxygen content calculated as the product of coronary flow and coronary oxygen extraction. Coronary flow was determined by collecting the effluent of the right ventricle for 1 minute. Coronary resistance was calculated as perfusion pressure minus left ventricular end-diastolic pressure divided by coronary flow. Such a measurement is only an approximation of pressure across the coronary circulation because left ventricular end-diastolic pressure rather than sinus pressure was used for this calculation. Coronary dilator reserve and minimal coronary resistance were determined during treatment with 10−4 M adenosine and 10−5 M nitroglycerin.

Phosphorus 31 Magnetic Resonance Spectroscopy

Phosphorus 31 MRS of the beating isolated perfused heart was obtained on a 5.6-T vertical 76-mm bore magnet. The 31P-MRS spectra were obtained without proton decoupling at 97.3 MHz, using an 1180 Nicolet computer, a pulse programmer, and a high resolution 20-mm broad band probe. Pulse angle was 60°, recycle time 1.25 seconds, and spectral width 4,000 Hz. The 236 transients were accumulated during a 5-minute period. The signal-to-noise ratio was approximately 30:1. To correct for partial saturation, fully relaxed spectra were
TABLE 1. Dry Heart Weight, Dry Heart Weight-to-Body Weight Ratio, Coronary Resistance, and Myocardial Oxygen Consumption in Normal Hearts and Left Ventricular Hypertrophy With and Without Verapamil Treatment

<table>
<thead>
<tr>
<th></th>
<th>DHW (g)</th>
<th>DHW/BW (g/kg)</th>
<th>CR (mm Hg/ml/min)</th>
<th>MVo₂ (µM/g DHW/min)</th>
<th>CF (ml/min)</th>
<th>SP (mm Hg)</th>
<th>HR (beats/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>0.17±0.01</td>
<td>0.52±0.02</td>
<td>5.4±0.1</td>
<td>46.7±1.4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>p&lt;0.001</td>
<td>p&lt;0.002</td>
<td>p&lt;0.001</td>
<td>p&lt;0.001</td>
<td>p&lt;0.001</td>
<td>NS</td>
<td></td>
</tr>
<tr>
<td>Hypertrophy</td>
<td>0.28±0.01</td>
<td>0.77±0.05</td>
<td>8.6±0.3</td>
<td>18.4±0.9</td>
<td>12.9±0.4</td>
<td>241.9±6.7</td>
<td>228.6±6.6</td>
</tr>
<tr>
<td></td>
<td>p&lt;0.02</td>
<td>NS</td>
<td>p&lt;0.001</td>
<td>p&lt;0.05</td>
<td>p&lt;0.001</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Hyp+Chr Ver</td>
<td>0.25±0.01</td>
<td>0.70±0.03</td>
<td>4.1±0.2</td>
<td>35.2±4.4</td>
<td>26.9±1.6</td>
<td>244.3±16</td>
<td>234.6±9.1</td>
</tr>
<tr>
<td></td>
<td>p&lt;0.02</td>
<td>NS</td>
<td>p&lt;0.001</td>
<td>p&lt;0.05</td>
<td>p&lt;0.001</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Hyp+Ac Ver</td>
<td>0.23±0.01</td>
<td>0.73±0.04</td>
<td>6.3±0.65</td>
<td>28.5±3.7</td>
<td>18.3±1.6</td>
<td>254.3±9.3</td>
<td>225.0±15</td>
</tr>
</tbody>
</table>

All values are given as mean±SEM; NS, not significant. DHW, dry heart weight; BW, body weight; CR, coronary resistance; MVo₂, myocardial oxygen consumption; CF, coronary flow; SP, pressure; HR, heart rate; Hyp+Chr Ver, chronic verapamil; Hyp+Ac Ver, acute verapamil; Hyp, hypertrophy.

obtained at a 15-second recycle time, and correction factors for phosphocreatine (PCr) and inorganic phosphate (Pi) were determined (3% and 5%, respectively). Chemical shifts were referred to the resonance position of PCr. The peaks characteristic of Pi, PCr, and phosphate groups of ATP were identified. Each spectrum and the corresponding areas were numerically integrated after defining the baseline. Phosphatase peaks were quantitated using manual electronic planimetry and estimated for whole heart detection by comparison to a capillary tube of standard methylene diphosphonic acid fixed inside the nuclear magnetic resonance (NMR) tube. High-energy phosphate values determined by 31P-MRS were standardized by high-pressure liquid chromatography of freeze-clamped tissue.30,31 Extracellular space was estimated by perfusing with K(CoEDTA).30 The cobalt was analyzed by Atomic Absorption.30 All MRS and freeze-clamped data were corrected for intracellular and extracellular water.30

Free ADP was calculated as follows:

\[
ADP_{\text{free}} = (\text{Cr})(\text{ATP})/(K_{\text{eq}})(H^+)(\text{PCr})
\]

The \(K_{eq}\) was 2.36 \times 10^5 at a pH of 7.0. This value was normalized for changes in pH.32 The pH was determined from the chemical shift in relation to the resonance of PCr.33 Intracellular pH was standardized as follows: A standard solution at physiologic ionic strength (150 mM KCl, 8 mM ATP, 10 mM PCr, 5 mM Pi, and 9 mM MgSO₄) was used at a temperature of 37°C to obtain the chemical shift titration curve of pH-dependent Pi to PCr peak difference. This curve was fitted to the Henderson-Hasselbalch equation. Creatine was obtained by analyzing the difference in total creatine (high-pressure liquid chromatography analyses of freeze-clamped tissue) and PCr obtained by 31P-NMR. The nucleosides, adenosine, inosine, and hypoxanthine were analyzed in the coronary effluent by high-pressure liquid chromatography as described earlier.30

**Phosphorylation Potential**

Corrected peak ratios obtained by 31P-NMR (e.g., PCR/PCR+Pi+β-ATP) were standardized according to values for the same hearts or hearts from parallel experiments analyzed by high-pressure liquid chromatography analyses of the freeze-clamped tissue. Data obtained from freeze-clamped tissue were expressed in micromoles per gram dry heart weight.24 Dry heart weight was obtained after 72 hours at 110°C. The micromoles per gram dry heart weight values were then converted to millimoles after correcting for intracellular and extracellular space.24 The phosphorylation potential was calculated from standardized NMR values and expressed as ln[(ATP)/(ADP)(Pi)]. The phosphorylation potential is expressed in the present study as the natural log and in molar values.

**Statistical Analysis**

Values are given as mean±SEM unless otherwise stated. Schéffe's test for multiple contrasts was applied to detect significant difference by the analysis of variance. The null hypothesis was rejected at the 95% level, considering a \( p \) value less than 0.05 as significant.

**Results**

**Severity of Left Ventricular Hypertrophy**

Left ventricular hypertrophy is characterized in Table 1. Compared with normal, the dry heart weight to body weight increased 48% in untreated hypertrophy, 55% in hypertrophy plus chronic verapamil, and 40% in hypertrophy plus acute verap-
amyl. The wet heart weight was 1.35±0.05 for control hearts, 2.01±0.07 for untreated hypertrophied hearts, 1.83±0.06 for chronically verapamil-treated hypertrophied hearts, and 1.86±0.05 for acutely verapamil-treated hypertrophied hearts. Minimal coronary resistance was significantly higher (p<0.001) in untreated hypertrophy compared with either normal, hypertrophy plus chronic verapamil, or hypertrophy plus acute verapamil.

Mechanical Performance During Ischemia and Reperfusion

Inducing global ischemia caused a rapid decrease of left ventricular developed pressure to zero within a few minutes in all groups. During early reperfusion mechanical performance, as reflected by developed pressure, improved promptly in normal, hypertrophy plus chronic verapamil, and hypertrophy plus acute verapamil but was attenuated in hypertrophy with no treatment. Only the latter was accompanied by a marked instability of the heart rhythm. After 30 minutes of reperfusion, left ventricular developed pressure in normal hearts recovered 86±4% of the baseline value; hypertrophy 44±4% (normal vs. hypertrophied hearts, p<0.001); hypertrophy plus chronic verapamil 106±2% (hypertrophy plus chronic verapamil vs. normal, p=0.005); and hypertrophy plus acute verapamil 88±5% (hypertrophy plus acute verapamil vs. normal, p=0.005) of the baseline value (Figure 1). Left ventricular end-diastolic pressure remained unchanged at 0 mm Hg in hypertrophy plus verapamil (both acute and chronic treatment) but increased in normal hearts from 2±1 to 34±5 mm Hg and in untreated hypertrophied hearts from 3±1 mm Hg to 34±8 mm Hg.

High-Energy Phosphate Metabolism During Ischemia and Reperfusion

The total loss of purine nucleosides in the coronary effluent for 5 minutes of reperfusion was significantly less in hypertrophy plus chronic verapamil compared with hypertrophy with no treatment (3.7±0.3 vs. 8.7±0.6 μmol/g dry heart wt, p<0.001)

![Graph](http://circ.ahajournals.org/)

**Figure 1.** Plots of left ventricular developed pressure during control perfusion (CTR), ischemia (ISCH), and reperfusion (REP) in normal hearts (NORMAL, n=12), left ventricular hypertrophy (HYPERTROPHY, n=12), and left ventricular hypertrophy pretreated with chronic (HYP+CHR VER, n=6) and acute verapamil (HYP+AC VER, n=6) administration.

![Spectra](http://circ.ahajournals.org/)

**Figure 2.** 31P-Magnetic resonance spectra of normal hearts (Normal), left ventricular hypertrophy (Hypertrophy), and left ventricular hypertrophy pretreated with chronic (HYP+CHR VER) and acute verapamil (HYP+AC VER) administration during control perfusion, at 30 minutes of global ischemia, and at 30 minutes of reperfusion.
TABLE 2. Inorganic Phosphate, Phosphocreatine, $\beta$-Adenosine Triphosphate, Phosphorylation Potential, and Rate-Pressure Product During Control Perfusion and After 30 Minutes of Postischemic Reperfusion in Normal Hearts and Left Ventricular Hypertrophy With and Without Verapamil Treatment

<table>
<thead>
<tr>
<th></th>
<th>$P_i$ (mM)</th>
<th>$PC_r$ (mM)</th>
<th>$\beta$-ATP (mM)</th>
<th>PP (M)</th>
<th>RPP (mm Hg×beats/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal ($n=6$)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>1.1±0.3</td>
<td>16.3±1.7</td>
<td>8.4±0.3</td>
<td>12.2±0.40</td>
<td>42,482±4,078</td>
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<tr>
<td>Reperfusion</td>
<td>2.4±0.7</td>
<td>14.7±2.0</td>
<td>4.6±0.4</td>
<td>11.2±0.65</td>
<td>33,253±3,921</td>
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<tr>
<td>Hypertrophy ($n=7$)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>3.0±0.3</td>
<td>15.1±1.2</td>
<td>7.3±0.4</td>
<td>11.1±0.39</td>
<td>52,358±3,329</td>
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<tr>
<td>Reperfusion</td>
<td>7.5±1.0</td>
<td>8.5±1.4</td>
<td>2.6±0.5</td>
<td>9.1±0.34</td>
<td>18,030±3,988</td>
</tr>
<tr>
<td>Hyp+Chr Ver ($n=7$)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>1.9±0.3</td>
<td>16.9±0.4</td>
<td>9.0±0.3</td>
<td>11.8±0.17</td>
<td>57,236±4,031</td>
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<tr>
<td>Reperfusion</td>
<td>3.2±0.5</td>
<td>17.9±0.9</td>
<td>6.7±0.3</td>
<td>11.4±0.12</td>
<td>56,497±4,901</td>
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<tr>
<td>Hyp+Ac Ver ($n=6$)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>3.8±1.2</td>
<td>12.1±1.2</td>
<td>8.4±0.1</td>
<td>10.8±0.50</td>
<td>62,340±7,101</td>
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<tr>
<td>Reperfusion</td>
<td>7.7±1.5</td>
<td>12.4±1.7</td>
<td>5.1±0.3</td>
<td>10.0±0.35</td>
<td>50,940±6,694</td>
</tr>
</tbody>
</table>

Values are given as mean±SEM: $P_i$, inorganic phosphate; $PC_r$, phosphocreatine; $\beta$-ATP, $\beta$-adenosine triphosphate; PP, phosphorylation potential; RPP, left ventricular rate-pressure product; Hyp+Chr Ver, chronic verapamil; Hyp+Ac Ver, acute verapamil.

and in hypertrophy plus acute verapamil compared with hypertrophy with no treatment (4.2±0.6 vs. 8.7±0.6 μmol/g dry heart wt, $p<0.001$). Normal hearts gave values of 6.2±0.8 μmol/g dry heart wt. Values given are the total content as measured in the total coronary effluent and normalized for heart weight. Nucleosides accumulated in the interstitial space during ischemia and were released in the first 5 minutes of reperfusion. Verapamil helped prevent the release of nucleotide breakdown.

Representative $^{31}$P-MRS spectra of each group during control perfusion, at 30 minutes of ischemia, and at 30 minutes of reperfusion are shown in Figure 2. The time course of changes in $P_i$, $PC_r$, $\beta$-ATP, and pH during ischemia and reperfusion are shown in Table 2 and Figure 3. At 30 minutes of postischemic reperfusion, $P_i$ was increased 224±36% of the baseline value in normal, 306±51% in hypertrophy untreated, 189±33% in hypertrophy plus chronic verapamil, and 202±15% in hypertrophy plus acute verapamil (Figure 3, Table 2). Phosphocreatine recovered in all except the untreated hypertrophy hearts (Figure 3, Table 2). Because of the loss of the nucleotide pool during ischemia, $\beta$-ATP recovered poorly in all groups but the least in the untreated hypertrophy group (Figure 3, Table 2). The intracellular pH fell severely in all groups but most severely in the hypertrophy group treated chronically with verapamil (Figure 3, Table 2).

Coronary Flow

As shown in Figure 4 and Table 1, coronary flow was significantly higher in normal hearts (19.7±0.8), hypertrophied hearts treated acutely with verapamil (18.3±1.6), and hypertrophied hearts treated chronically with verapamil (26.9±1.6) compared with untreated hypertrophied hearts (12.9±12.9). Flow recovered to nearly control levels of coronary flow with 30 minutes of postischemic reperfusion: normal hearts 16.5±1.4, hypertrophied hearts 10.9±1.0, hypertrophied hearts plus chronic verapamil 25.6±1.3, and hypertrophied hearts plus acute verapamil 17.8±1.9. Coronary resistance in normal hearts (5.4±0.1) and hypertrophied hearts treated chronically (4.1±0.2) or acutely (6.3±0.7) with verapamil was significantly different ($p<0.01$) from untreated hypertrophied hearts (8.6±0.3). The pressure used in defining the rate-pressure product is developed pressure.

Oxygen Consumption

As shown in Figure 5 and Table 1, oxygen consumption was significantly lower in hypertrophied hearts compared with normal hearts ($p<0.001$) before ischemia. Hypertrophied hearts treated chronically or acutely with verapamil had a significantly higher oxygen consumption ($p<0.05$) than untreated hypertrophied hearts. After 30 minutes of reperfusion myocardial oxygen consumption was significantly lower ($p<0.05$) in normal (46.7±1.4 vs. 38.3±3.3 μmol/g dry wt/min) but remained unchanged in hypertrophy (18.4±0.9 vs. 15.4±1.6 μmol/g dry wt/min), hypertrophy plus chronic verapamil (35.2±4.4 vs. 33.8±3.5 μmol/g dry wt/min), and hypertrophy plus acute verapamil (28.5±3.7 vs. 28.2±4.4 μmol/g dry wt/min). Oxygen consumption described above was based on dry weight. Control values for oxygen consumption, based on wet weight, were 5.9±0.3 μmol O$_2$/g wet heart wt/min for normal rat hearts, 2.5±0.2 for untreated hypertrophied hearts, 4.8±0.3 for chronically treated hypertrophied hearts, and 3.6±0.02 for acutely treated hypertrophied hearts.

Phosphorylation Potential

Before ischemia, the phosphorylation potential was not significantly different among the four groups (Figure 6, Table 2). The phosphorylation potential remained significantly depressed postischemia in
FIGURE 3. Bar graphs of the course of inorganic phosphate, phosphocreatine, β-adenosine triphosphate (β-ATP), and pH during control perfusion (CTR), 30 minutes of global ischemia (ISCH), and 30 minutes of reperfusion (REP) in normal hearts (NORMAL, n=6), left ventricular hypertrophy (HYPERTROPHY, n=7), and left ventricular hypertrophy pretreated with chronic (HYP+CHR VER, n=6) and acute verapamil (HYP+AC VER, n=6) administration.

Discussion

The aim of the present study was to assess the effects of verapamil treatment on postischemic recovery relative to myocardial mechanics and energetics of rats with left ventricular hypertrophy induced by chronic pressure overload. We compared 1) untreated rats with hypertrophied hearts, 2) hypertrophied hearts from rats treated acutely with verapamil, 3) hypertrophied hearts from rats treated chronically with verapamil, and 4) normal hearts. All hypertrophied hearts, untreated or treated acutely or chronically with verapamil, had significantly higher dry heart weight-to-body weight ratios compared with controls and were not significantly different in dry heart weight-to-body weight ratios among themselves. After 30 minutes of ischemia and reperfusion, hypertrophied rat hearts treated chronically with verapamil-recovered rate-pressure product completely (100±3% of the baseline value). Rate-pressure product recovered 82±5% in hypertrophied hearts treated acutely with verap-

the untreated hypertrophy group (p<0.05), but returned to values not significantly different from control in all other groups (Figure 6, Table 2).

FIGURE 4. Plots of relation of coronary flow/g wet heart wt (CF/HWWT) and left ventricular work during control perfusion (CTR) and after 30 minutes of postischemic reperfusion (REP 30', arrowheads) in normal hearts (NORMAL, n=6), left ventricular hypertrophy (HYPERTROPHY, n=7), and left ventricular hypertrophy pretreated with chronic (HYP+CHR VER, n=7) and acute (HYP+AC VER, n=6) verapamil administration. RPP, rate-pressure product.
amil, remained impaired in untreated hypertrophied hearts (40±5%), and returned to 83±5% in normal hearts. The phosphorylation potential recovered to 96.8±2.4% of its baseline value with chronic verapamil-treated hypertrophy, to 93.7±4.6% with acute verapamil treatment, to 82.2±3.7% with untreated hypertrophy, and to 91.2±2.7% in normal hearts. Myocardial oxygen consumption decreased significantly in normal hearts and remained unchanged in the three groups with left ventricular hypertrophy. Recovery of rate-pressure product after ischemia, however, was directly related to coronary flow and inversely related to coronary resistance.

Coronary blood flow and oxygen consumption for untreated hypertrophied hearts was significantly lower than that for normal hearts (Table 1). This observation was true when coronary flow was not corrected for heart weight (Table 1) or calculated as a factor of heart weight (wet) (Figure 4). The diminished coronary flow correlated with the depressed oxygen consumption in the untreated hypertrophied hearts whether oxygen consumption was corrected for dry heart weight (Table 1 and Figure 5) or expressed as a factor of wet heart weight as described in the "Results." Similar findings were obtained for the spontaneously hypertensive rat, another form of hypertension.26 Such a reduced oxygen requirement with hypertension for retaining a high-energy phosphate level during a comparable or greater rate-pressure product output may be due to a shift in metabolism and/or a shift in isomyosins.26 Thus, the depressed coronary flow with hypertension may be specific to rodents and related to the shift in isomyosins and a decrease in oxygen requirements and energy demands.26,34,35 We demonstrated in earlier studies that hypertension and hypertrophy in the spontaneously hypertensive rat was associated with a shift toward isomyosin V3, a decrease in oxygen consumption, and a concurrent increase in rate-pressure product.26,34 This increase in work, a shift toward isomyosin V3, and a corresponding decrease in oxygen consumption were attributed to greater thermodynamic efficiency with such an isomyosin shift.26 A similar shift in isomyosins occurs with hypertension and hypertrophy induced by aortic stenosis in rats.35 The shift toward isomyosin V3 may be assuaged by calcium entry blockers.34

Coronary flow was not significantly different between normal and hypertrophied hearts treated chronically with verapamil. Coronary flow from both was significantly higher than from hypertrophied hearts treated acutely with verapamil or not treated at all, although hypertrophied hearts treated acutely with verapamil had a significantly higher coronary flow compared with untreated hypertrophied hearts. Postischemic recovery of hypertrophied hearts was directly related to coronary flow before ischemia. Verapamil helped improve coronary flow by improving the calcium homeostasis of the vasculature. Similar findings were also obtained with the working heart model and the spontaneously hypertensive rat.26,34 The lower coronary flow with hypertension may be due to an imbalance in the calcium homeostasis of the heart leading to structural and functional abnormalities of the coronary vessels.8–10 A shift in isomyosins and a resultant reduction in oxygen requirements may then occur in rodents to compensate for the depressed coronary flow.

Mortality is increased threefold in patients with electrocardiographic evidence of left ventricular hypertrophy compared with the average general mortality when adjusted for age and sex.1 The risk
for sudden death in patients with coronary artery disease associated with left ventricular hypertrophy is increased fivefold compared with the general population. In the presence of increased heart size and arterial pressure, heart failure and cardiac rupture after infarction are more frequent. In dogs with left ventricular hypertrophy and systemic hypertension, coronary occlusion is associated with a threefold increase in sudden death, a substantial increase in infarct size, and a greater susceptibility for serious arrhythmias than in animals with normal cardiac mass. Thus, the course after a coronary ischemic event tends to be more severe in the presence of left ventricular hypertrophy. In the studies described here, hearts from rats subjected to aortic stenosis but not treated with verapamil recovered poorly after 30 minutes of global ischemia compared with normal hearts or hypertrophied hearts treated acutely or chronically with verapamil.

In untreated left ventricular hypertrophy, minimal coronary resistance was increased compared with normal hearts. Functional and anatomic alterations of the coronary vasculature in chronic pressure-overload hypertrophy are well known. In spontaneously hypertensive rats treated with a slow channel calcium entry blocker, both hypertension and left ventricular hypertrophy were still present, but myocardial capillary density was comparable to that of normal Wistar-Kyoto rats. Because verapamil exerts a coronary vasodilator effect, thereby increasing coronary blood flow, it may also induce proliferation of myocardial capillaries. In this study, minimal coronary resistance in verapamil-treated hypertrophied hearts was diminished compared with untreated hypertrophied hearts. Alterations of the coronary circulation in untreated pressure-overload left ventricular hypertrophy may partially account for the impaired posts ischemic recovery, causing a delayed supply of nutrients and oxygen and a retarded removal of noxious metabolites during reperfusion. Verapamil treatment may minimize these deleterious changes of the coronary vasculature and, additionally, may protect functional integrity of mitochondrial and cell membranes during ischemia. However, the sensitive protective effect conferred by acute as well as chronic verapamil administration indicates that other mechanisms must also be operative.

Verapamil has been shown to exert a protective effect on the normal myocardium during ischemia and acidosis. In dogs treated with verapamil, coronary occlusion resulted in less infarction volume, reduction of ST-segment elevation, and prevention of hemodynamic deterioration. Preischemic treatment with verapamil protects the normal myocardium against some of the deleterious effects of ischemia, including breakdown and depletion of intracellular ATP, increase of intracellular calcium, loss of mitochondrial ATP generating ability, and depression of the sarcosomal enzymatic activity profile. Inducing global ischemia stops the supply of nutrients and oxygen to the heart as well as the removal of deleterious products within seconds in all heart preparations. The rapid increase in inorganic phosphate, the reduction in muscle stretch after the fall of perfusion pressure, and a decrease of the action potential duration may account for the decline of left ventricular developed pressure to zero within the first few minutes of ischemia. The ATP breakdown was attenuated in verapamil-treated hypertrophied hearts, resulting in a significantly higher ATP level at 30 minutes of ischemia compared with untreated hypertrophied and normal hearts. This protective effect of verapamil on ATP stores has been described previously in normal hearts and might be caused by a reduced ATP use due to lower intracellular calcium levels. During reperfusion, fewer membrane-diffusible nucleosides were washed out, which may reflect quantitatively diminished accumulation of these nucleosides during ischemia. Although pH was lowest at 30 minutes of ischemia in the chronic verapamil-treated group, this group, nevertheless, showed the best posts ischemic recovery. Furthermore, even though Pi was highest in the two verapamil-treated groups at 30 minutes of ischemia, these two groups, again, showed the best posts ischemic recovery.

It might be presumed that normal hearts pretreated with verapamil might show an even better recovery during reperfusion. Normal animals pretreated with verapamil were not investigated in the current study because the salutary influence of verapamil on the normal heart during ischemia has already been established. A previous study has shown a salutary effect of verapamil on high-energy phosphate compounds of the reperfused ischemic myocardium of normal rats.

In summary, the criteria for posts ischemic recovery were retention of rate-pressure product and the phosphorylation potential. Posts ischemic recovery of the hypertrophied heart was directly related to coronary flow before inducing ischemia. Acute or chronic verapamil treatment of the hypertrophied heart may improve posts ischemic recovery by its improvement of coronary flow. The results obtained here by inducing hypertrophy with aortic stenosis, thereby causing hypertension, may not be common to all types of hypertrophy. Furthermore, the isolated perfused heart is a hyperperfused system. The isolated heart, as described here, has excessive coronary flow and oxygen delivery so that the results obtained in this study may not occur in situ.

References


23. Key Words • hypotrophy • ischemia • perfusion • calcium antagonists
Verapamil preserves myocardial performance and energy metabolism in left ventricular hypertrophy following ischemia and reperfusion. Phosphorus 31 magnetic resonance spectroscopy study.

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