Protective Role of Increased Myocardial Glycogen Stores in Cardiac Anoxia in the Rat

By James Scheuer, M.D. and S. William Stetoski

ABSTRACT

To determine whether increased glycogen stores might protect the heart against anoxia, experiments were performed in the isolated perfused rat heart. Marked differences in cardiac glycogen were produced by comparing hearts from rats previously treated with reserpine with hearts from control rats. Lesser differences in cardiac glycogen were produced in hearts by perfusing them for 15 minutes without glucose (0 mM glucose) or with 20 mM glucose. Both groups were then studied during a 5-minute anoxic cycle with 5 mM glucose as the exogenous substrate. Hearts from the reserpine-treated rats had higher left ventricular pressures, maximal rate of left ventricular pressure rise, and lactate output after 2 minutes of anoxia than the hearts from control rats. Similar but less marked mechanical differences were observed between 0 mM glucose and 20 mM glucose hearts. The mechanical differences during anoxia between the two groups were not abolished by simultaneous L-norepinephrine administration. Hearts with greater initial glycogen stores had higher glycogenolytic rates, and proportionately more lactate was produced from glycogen than from glucose. Thus, anaerobic ATP production per mole of hexose was greater in hearts with higher glycogen stores. Calculated ATP production was also greater in hearts from the reserpine-treated rats than in those from control animals.

These studies demonstrate that both marked and minor elevations in cardiac glycogen are associated with greater glycolytic reserve and improved mechanical resistance to anoxia. This appears to be mainly due to enhanced glycogenolysis and anaerobic ATP production.

ADDITIONAL KEY WORDS myocardial metabolism glycogen
glycogenolysis lactate glycolysis anoxia reserpine
catecholamines myocardial energetics high-energy phosphates

Increased glycolysis is an important compensatory metabolic mechanism in cardiac hypoxia (1). Those vertebrate subgroups with the highest potential cardiac glycolytic rates and animals with high cardiac glycogen stores appear to have the greatest resistance to anoxia (2-5). The observation that glycogenolysis accompanies cardiac hypoxia (6, 7) suggests that glycogen stores might be important in the cardiac response to oxygen deprivation. Increased glycogen stores have been shown to have a protective effect during anoxia in the fetal rabbit heart (8), and glycogen has been shown to be a major source of energy in the anoxic turtle heart (8).

Glycogen levels in the heart have a diurnal variation (10) and are affected by a variety of hormonal (11-15), dietary (16) and exercise factors (13, 17), and by the prevention of catecholamine-induced glycogenolysis (18-21). These changes in glycogen stores might be accompanied by altered mechanical responses to oxygen deprivation.

The purpose of the present study was to determine whether alterations in glycogen levels are associated with differences in the mechanical performance of the rat heart during anoxia. The relation of such changes to
glycogenolysis, glycolysis, and high energy phosphate levels were also studied. The results indicate that hearts with increased glycogen stores have improved mechanical performance during anoxia. They demonstrate greater glycogenolysis and higher rates of glycolytic ATP generation. Thus, by increasing glycogen levels in the heart, advantage can be taken of the increased efficiency of glycolytic ATP production from glycogen as compared with glucose.

Methods
The hearts of male albino rats, weighing 200 to 280 g, were studied during aerobic and anoxic periods. One series was designed to study the effects of marked differences in myocardial glycogen levels. In these experiments hearts from control rats were compared with hearts of rats which had been injected intraperitoneally with reserpine, 5 mg/kg, 24 hours earlier. This technique of reserpinization reduces myocardial norepinephrine stores over 80% (22) and raises norepinephrine stores over 80% (23). Another series was designed to study moderate differences in cardiac glycogen. In these experiments, hearts were perfused aerobically for 15 minutes with 20 mM glucose or without glucose (0 mM glucose), Amssorhobic perfusion containing 5 mM glucose was then abruptly introduced, and the mechanical function, glycogen levels, and lactate outputs were studied.

The perfusion apparatus has been described and diagrammed previously (24). Hearts are perfused retrograde through the aorta in a gravity-flow system. The perfusion medium contained 143 mM sodium, 126 mM chloride, 25 mM bicarbonate, 6 mM potassium, 1.2 mM magnesium sulfate, 1.2 mM phosphate, 1.2 mM calcium, and 0.4 mM sodium EDTA. Glucose, 5 mM, was included at all times except during aerobic periods. During anoxic periods, the perfusion medium was then abruptly switched into the perfusion line and continued for 5 minutes. The perfusion apparatus was arranged with the gravity-reservoir 100 cm above heart level. There were two parallel perfusion systems, either of which could be alternately switched into the perfusion line by a stopcock just above the aortic perfusion cannula. Thus, oxygenated and unoxgenated medium could be changed abruptly. Flow rates were monitored by flowmeters of the purge type inserted between the gravity reservoir and the heart. Left ventricular pressures were monitored through a 17-cm polyethylene-60 catheter inserted through the mitral valve and attached to a Statham P23Db strain gauge. The system had a frequency response of 21 cps. The rate of left ventricular pressure rise (LV dp/dt) was recorded on a differentiating channel on an Electronics for Medicine photographic recorder. The time constant for the differentiating circuit was 0.5 msec, and the response was linear within 5% from 1 to 57 cps. Recordings were made at a paper speed of 100 mm/sec. Peak systolic pressures were monitored on a full scale of 0 to 100 mm Hg. End-diastolic pressures were monitored on a full scale 0 to 25 mm Hg.

To avoid bradycardia during anoxia, hearts were paced through a platinum wire attached to a Grass S6X stimulator. The active lead was placed on the left ventricle and the ground lead on the aortic cannula. The pacing rate was constant in any one heart. Rates used in different hearts were between 330 and 350 beats/min. The stimulus had an amplitude of 4 v and a duration of 4 msec. Hearts were perfused for 15 minutes aerobically. Pacing was begun after 10 minutes of control perfusion. After 15 minutes, aerobic perfusion was continued for 5 additional minutes in some hearts. In other experiments, N2-equilibrated solution was abruptly switched into the perfusion line and continued for 5 minutes. During the 15-minute control period, the medium was recirculated. During the 5-minute experimental period, the effluent medium was not recirculated. In a series of experiments on hearts from the reserpine-treated and the control rats, photographs were taken of the heart at the end of the control and anoxic periods to estimate any changes in heart size. For these experiments, rats were paired by weight before reserpinization so that paired experimental and control hearts would be approximately the same size. The photographs were taken with a Nikon F camera using a 50-mm lens and extension rings at a fixed focal length of 31.8 mm. A consistent geometry between different hearts was achieved by always placing the pulmonary outflow tract parallel to the base. The negatives were enlarged 0.75 times with a photographic enlarger. The cardiac length was defined as the distance between the origins of the aorta and the apex of the heart. The diameter of the heart was measured to the midpoint of the

Conclusion

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long axis was used as the width. An 0.25-second exposure duration and an F 16 lens opening and a filter were used. A white reflector was placed behind the heart chamber and light was reflected from both sides on to the heart. The long exposure time in relation to the cardiac cycle and the excellent contrast of the pictures enabled estimation of maximum heart size.

In anoxia experiments on three hearts from the reserpine-treated and three from the control rats, 5 mM glucose-U-14C was used during anoxia to study the relative contribution of glucose to lactate formation. The final specific activity of the glucose was 19,700 dpm/μmole. The effluent was collected and analyzed for total lactate and for lactate-14C.

In one group of experiments on hearts from reserpine-treated rats, the effect of exogenous catecholamines on the mechanical response in anoxic experimental and control hearts was tested. L-norepinephrine was introduced into the aortic cannula during the 5-minute aerobic or anoxic experimental period. Infusion was at a constant rate of 4 X 10^-10 moles/min in 0.1 ml of solution.

When high-energy phosphate compounds were studied at zero time, the rats were anesthetized with sodium pentobarbital (10 mg/kg, ip) and respired with 100% oxygen through a tracheotomy cannula controlled by a Harvard animal respirator. The thorax was opened and the apex of the heart was clamped in aluminum-alloy clamps previously cooled to the temperature of liquid nitrogen. When these compounds were to be measured in perfused hearts, the experiments were terminated by clamping the bearing heart. The frozen hearts were placed in liquid nitrogen. Perchloric acid extracts were prepared as described previously (25). The dry weight of the precipitated protein was used as the dry heart weight (25).

When hearts were analyzed for glycogen, the hearts were removed from the animal or from the perfusion apparatus, placed in buffer at 3°C, blotted dry, and a portion removed for the dry to wet weight ratio. The remainder of the heart was immediately introduced into a preweighed tube containing 30% KOH.

Adenosine triphosphate (ATP) and creatine phosphate (CP) were determined by the methods of Lamprecht, Stein, and Trautschold (26, 27). Lactate and pyruvate were measured by the methods of Wallass and Wallass (30), and the resultant glucose was quantified enzymatically (31). All metabolic results are calculated per gram dry heart weight. P values were determined with analysis of variance using interaction when comparing paired means (32).

Results

When rat hearts are perfused on a retrograde apparatus, they retain fluids (33). Furthermore, when hearts are subjected to ischemia or hypoxia, they also tend to increase their water content (34). Table 1 shows the dry to wet heart weight ratios found in the various experiments we performed. These data demonstrate that the water content increased with perfusion, but the changes were similar in experimental and control hearts. The brief period of anoxia that we imposed was not associated with an increase in water content.

Dynamic performance was similar in experimental and control hearts during the first 15 minutes of aerobic perfusion. Figure 1 shows the dynamics of hearts from control and reserpine-treated hearts perfused aerobically for the experimental period, from 15 to 20 minutes. At 15 minutes, coronary flow was slightly but

<table>
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<th>Table 1</th>
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<td><strong>Dry Heart Weight to Wet Heart Weight Ratios in Perfused Hearts</strong></td>
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<tr>
<td><strong>In vivo (15 min)</strong></td>
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<tr>
<td>CH (5 mM glucose)</td>
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<tr>
<td>RH (10 mM glucose)</td>
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<td>CH (20 mM glucose)</td>
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Numbers in parentheses are number of hearts. Values are mean ± SE for hearts perfused with the various experimental designs. See text for full details.

*Hearts from control rats. fHearts from rats treated with reserpine.
significantly higher in the experimental hearts, but there was no difference in cardiac performance during the 5-minute period. The similarity in performance of isolated hearts from reserpinized and non-reserpinized animals has been noted previously (35). Figure 2 plots the same measurements for hearts from reserpinized and control animals subjected to anoxia. Coronary flow rose with anoxia in both groups. Left ventricular end-diastolic pressure (EDP) rose significantly only in the controls. Peak left ventricular systolic pressure (PLVSP) and maximum LV dp/dt fell in both groups at approximately the same rate for 2 minutes, but then declined more rapidly in the controls. After 5 minutes of anoxia, these variables were significantly higher in the experimental hearts than in the controls.

To determine whether the differences observed between the two groups might be due to release of endogenous catecholamines in the control hearts, studies were performed in which L-norepinephrine $4 \times 10^{-5}$ moles/min was infused during anoxia. This dose increased PLVSP over 100% and maximum LV dp/dt 150% above control levels in aerobic perfusion of hearts from control and reserpinized animals. Figure 3 shows the dynamics in
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Dynamics of perfused hearts made anoxic between the 15th to the 20th minute of perfusion. The zero time signifies the end of 15 minutes of aerobic perfusion and the beginning of anoxic perfusion. Abbreviations same as in Figure 1. Asterisks along the lines indicate \( P < 0.05 \) when 0- and 5-minute paired mean values are compared. Asterisk at the 5-minute points signifies that at that time, \( P < 0.05 \) when means for control and reserpine-treated heart are compared. \( \ast \) indicates that change in EDP is significantly greater in control than in experimental hearts.

Experiments in which this dose of norepinephrine was infused during anoxia. The left ventricular mechanical differences between hearts from the control and reserpine-pretreated animals during anoxia were maintained in the presence of L-norepinephrine. In fact, in the control hearts after 5 minutes of anoxia, PLVSP was 7 mm Hg (15% of aerobic) in the presence of norepinephrine versus 14 mm Hg (26% of aerobic) when norepinephrine was
not infused (P<0.1). In the hearts from reserpinized animals although the mean PLVSP was slightly higher during anoxia in the presence of norepinephrine than in its absence, the difference was not statistically significant. After 5 minutes of anoxia, maximal LV dp/dt was lower in the control hearts in the presence of norepinephrine than in its absence (P<0.05), and higher in the experimental hearts in the presence of norepinephrine than in its absence (P<0.01). Coronary flow was lower in the controls during anoxia when norepinephrine was present than when it was absent.

The results of the photographic analyses of hearts from the control and the reserpinized

*Dynamics of perfused hearts in which L-norepinephrine was infused during the anoxic period. Experimental design and abbreviations same as in Figures 1 and 2.*

**Figure 3**

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Glycogen levels and lactate output of control and reserpine-treated hearts during aerobic and anoxic perfusions. Units are per gram dry heart weight. Glycogen values at time zero were obtained upon opening the chest and immediately processing the heart. Each glycogen point is the mean of six hearts ± 1 se. Numbers on the right of glycogen panel indicate the mean calculated glycogen breakdown during anoxia in the two groups. O₂ signifies aerobic perfusion; N₂ signifies anaerobic perfusion. Each lactate curve is derived from the mean of six experiments.

Animals during anoxia are shown in Table 2. All dimensions increased significantly during anoxia. The mean width of hearts from reserpine-treated rats was slightly less at the start of anoxia than that of the controls (P < 0.1), and based upon the calculation for the "volume," experimental hearts were slightly smaller than the control hearts after 15
TABLE 2

<table>
<thead>
<tr>
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<th>Length (mm)</th>
<th>Width (mm)</th>
<th>Volume (mm³ × 10⁻³)</th>
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<tr>
<td></td>
<td>(15 min)</td>
<td>(10 min*)</td>
<td>(15 min)</td>
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<td></td>
<td>(20 min)</td>
<td>(20 min)</td>
<td>(20 min)</td>
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<tr>
<td>CH(10)</td>
<td>130 ± 4</td>
<td>151 ± 4</td>
<td>83 ± 2</td>
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<td></td>
<td>99 ± 4</td>
<td>90 ± 2</td>
<td>502 ± 33</td>
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<tr>
<td>RH(10)</td>
<td>134 ± 4</td>
<td>148 ± 4</td>
<td>76 ± 2</td>
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<tr>
<td></td>
<td>98 ± 2</td>
<td>138 ± 5</td>
<td>453 ± 37</td>
</tr>
<tr>
<td></td>
<td>635 ± 37</td>
<td>629 ± 51</td>
<td><strong>P &lt; 0.05</strong></td>
</tr>
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</table>

Values are mean ± SE. Number in parentheses is number of hearts. All values are in millimeters measured after enlarging the silhouettes with a photographic enlarger. Abbreviations as in Table 1.

* Fifteen minutes is the end of aerobic perfusion and onset of anoxia. Twenty minutes is the end of anoxic perfusion.

† *P < 0.2, comparing CH and RH.

‡ *P < 0.01, comparing the paired 15- and 20-minute values.

§ *P > 0.2, comparing CH and RH.

$ *P < 0.1, comparing CH and RH.

|| *P < 0.05, comparing CH and RH.

Volume was calculated by the equation \( V = \frac{1}{6}(Length)(Width)^2 \).

Conversion of glucose-U-¹⁴C to lactate-¹⁴C in three control and three reserpine-treated hearts during anoxia. Individual points for each heart are shown. First-minute values are shown in an expanded scale in the upper left hand corner.

Figure 4 shows the glycogen levels and lactate output during the experimental procedure. Glycogen was twice as high in the hearts minutes of aerobic perfusion (P < 0.05). However, after 5 minutes of anoxia, no significant differences were found between the two.
ATP values and creatine phosphate (CP) levels in control and reserpine-treated hearts during aerobic and anoxic perfusions. Results are expressed in micromoles per gram dry heart weight. Each point is the mean ± SS of six hearts. Values at time zero were obtained from artificially resired, open-chest rats.

From reserpized animal as in those from the control at the time of sacrifice, and this relationship persisted throughout aerobic perfusion up to 20 minutes. With anoxia, marked glycogen depletion occurred but at the end of 5 minutes, glycogen remained significantly higher in the experimental than the control hearts. When each 20-minute anoxic glycogen value was subtracted from the mean 15-minute aerobic value, experimental hearts had over 3 times as much glycogenolysis as the controls during anoxia (P<0.001).

Lactate output was low during aerobic perfusion. This is the usual finding when hearts are perfused with glucose-containing, lactate-free medium (25). With the onset of anoxia, there was an abrupt increase in lactate production which reached 20 to 40 times the...
control rate at 2 minutes. Thereafter, in the hearts from reserpine-treated animals, lactate production remained constant, whereas in the control hearts, it fell progressively. The calculated glycogen breakdown accounted for close to 100% of lactate production during anoxia in the experimental and 40% in the control hearts. The mean lactate/pyruvate ratios in the cardiac effluent were 6 ± 2 in aerobic perfusions for both groups. These ratios rose to 62 ± 3 in the control and 51 ± 4 in the experimental hearts ($P < 0.05$).

FIGURE 7 shows the rate of oxidation of 5 mM glucose-$\text{U}^{14}$C to lactate-$\text{U}^{14}$C during anoxia of hearts prepared by prior cardiac perfusion with 20 mM glucose or 0 mM glucose. Anoxic perfusion was with 5 mM glucose in both groups. DLVSP = developed left ventricular systolic pressure, *($P < 0.1$); **($P < 0.05$). All other designations same as in Figure 5.
anoxia in three hearts from control and three from reserpinized animals. Even during the first half minute, lactate from glucose was greater in the control than the hearts from the reserpinized animals, and this relationship remained true for all points during the 5 minutes of anoxia.

Figure 6 shows the results of high-energy phosphate analyses in hearts from the control and the reserpinized animals. ATP and CP concentrations were comparable in the two groups of animals before perfusion and remained at similar values for the first 15 minutes of aerobic perfusion. There were slight decreases in the concentration of these compounds between 15 and 30 minutes of aerobic perfusion in the controls so that at 20 minutes, their concentrations were slightly higher in the experimental than in the control hearts. With anoxia, marked reductions in both compounds occurred. CP reached very low levels, and was the same in the two groups. ATP was higher after 5 minutes of anoxia in the experimental than in the control hearts.

Figure 7 shows the performance during anoxia of hearts which had been perfused with 20 mM glucose or 0 mM glucose during the 15-minute aerobic period. Coronary flow did not rise as rapidly in hearts prepared with 0 mM glucose, but at the end of 5 minutes of anoxia, coronary flow was the same in the two groups. Left ventricular end-diastolic pressure rose significantly during anoxia in the hearts with 0 mM glucose hearts, and was higher after 5 minutes of anoxia in these experiments than in hearts with 20 mM glucose. Because of large increases in end-diastolic pressure in 0 mM experiments, developed left ventricular systolic pressures (DLVSP) instead of PLVSP are plotted in Figure 7. DLVSP and maximum LV dp/dt declined significantly during anoxia in both groups, but both variables remained significantly higher during anoxia in hearts prepared with 20 mM glucose than in hearts prepared without glucose.

Figure 8 shows the glycogen levels and lactate production rates for hearts prepared with 0 mM glucose or 20 mM glucose during aerobic and anaerobic perfusion. The 20 mM hearts had 30% more glycogen at the onset of anoxia than 0 mM hearts. Almost complete glycogen depletion occurred in both groups. Glycogen utilization, calculated from individual 20-minute anoxic values and 15-minute mean values, was 40% greater in hearts prepared with 20 mM glucose versus 0 mM glucose (P < 0.001).

Lactate output rose to a peak after 2 minutes of anoxia, and then declined in both groups. There was no significant difference between the two groups with regard to lactate production. Calculated glycogen breakdown during anoxia could account for 60% of lactate production in hearts with 20 mM glucose versus 40% in those with 0 mM glucose.
Discussion

In the present experiments, proof that hearts with higher glycogen stores had improved mechanical performance rests upon the ability to measure the dynamics of perfused rat hearts. Ideally, performance might be expressed in terms of tension development, contractility, and fiber shortening or external work. These properties all relate to energy utilization by the myocardium (36). The approximation of tension development necessitates knowledge of the left ventricular pressure and volume.

Rat hearts perfused retrograde through the aorta with standard techniques developed left ventricular systolic pressures that slightly exceed perfusion pressure (37), and systolic ejection probably occurs. In the present experiments, the ionized calcium concentration in the perfusion medium was lowered slightly so that ejection did not occur. Therefore, if no mitral regurgitation was present, the ventricle should have contracted isovolumically, with no net fiber shortening or external work. Although a small amount of mitral regurgitation might have been present, the consistent increases in the end-diastolic pressures during anoxia in hearts with profound deterioration is evidence that severe mitral regurgitation did not occur. Although we did not measure ventricular volumes, our photographic analyses of hearts from control and reserpinized animals were sensitive enough to detect dimensional changes that resulted from anoxia within each group (Table 2). Control hearts were slightly larger than experimental hearts after 15 minutes of aerobic perfusion, but this difference was eliminated after 5 minutes of anoxia. Since LVSP was higher in experimental hearts, calculations of wall tension, based upon the assumption that left ventricular volume would be proportional to the volume of heart tissue, suggest that tension development in experimental hearts was approximately twice that in controls after 5 minutes of anoxia.

Contractility can be represented by the relationship of maximum dp/dt to mean systolic tension (38). In our experiments the diameter of hearts was the same in the control and experimental hearts after 5 minutes of anoxia, and therefore relative contractility at that time would be a function of the maximum LV dp/dt divided by mean left ventricular systolic pressure. The ratios of maximum dp/dt to mean left ventricular systolic pressure for the groups after 5 minutes of anoxia are 84 sec⁻¹ for controls and 88 sec⁻¹ for the experimental hearts. These apparent differences in the contractile state are further supported by the observation that during anoxia, hearts with higher initial glycogen stores developed higher left ventricular systolic pressures and maximum dp/dt from lower left ventricular end-diastolic pressures than did hearts with lower initial glycogen stores (Figs. 2, 3, 7).

After 15 minutes of aerobic perfusion, PLVSP and left ventricular maximal dp/dt were similar in hearts from both control and reserpinized animals. This finding is in accord with the several papers recently reviewed by Roberts (23) indicating that it takes several days of reserpine administration before its direct negative inotropic effects are demonstrable.

Analysis of the three factors in energy utilization (36) indicates that during anoxia, experimental hearts consumed more energy than the control hearts. High energy phosphate levels were higher in hearts from reserpinized animals than in control hearts after anoxic perfusion (Fig. 6). Since energy utilization was also greater in experimental hearts during that period, these hearts must have been generating more ATP by anaerobic pathways. This supposition is supported by the finding of higher lactate production rates during the final 3 minutes of anoxic perfusion in hearts from reserpinized animals than in control hearts (Fig. 4). This is the same period of perfusion during which experimental hearts demonstrated enhanced mechanical performance (Fig. 2). The findings indicate that the experimental hearts had higher glycolytic reserves than did the controls.
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14C studies demonstrate that more lactate came from glycogen in hearts that had enhanced performance during anoxia, whereas in hearts that deteriorated more rapidly a higher percent of the lactate was generated from glucose. During glycolysis, 3 moles of ATP are generated for each mole of glycogen used but only 2 moles for each mole of glucose used. Therefore, hearts with a greater fraction of lactate produced from glycogen would have proportionately higher rates of ATP generation.

It has been shown that cardiac function continues in the face of anoxia and cardiac glycogen depletion in the western painted turtle (39). However, in that situation, marked increases in blood glucose occur, perhaps offsetting the reduced endogenous carbohydrate supply.

The present studies demonstrate that in the rat heart, even in the presence of physiologic levels of exogenous glucose, glycogen availability can be a limiting factor in the potential glycolytic rate. The experiments were conducted without insulin being present in the perfusion medium. In rat hearts perfused anaerobically with 100 mg/100 ml of glucose, insulin increases glucose uptake by approximately 30% (40). Therefore, although insulin might have diminished the differences seen between control and experimental hearts, a 30% increase in glucose conversion to lactate would not have abolished the metabolic advantage of the reserpine-treated hearts. Also, very little glucose was used by these hearts, suggesting that glycogen was metabolized preferentially in them. Thus, glucose transport probably reached a maximum only in control experiments. The very low residual glycogen levels after anoxia in all except the group of experiments on reserpine-treated hearts similarly suggests that glycogen was used preferentially.

The possibility had to be considered that the release of endogenous catecholamines from control hearts in some manner contributed to more rapid deterioration of these hearts. If this were true, the infusion of L-norepinephrine into reserpine-treated hearts during anoxia should have caused a similar deterioration. However, when norepinephrine was infused during anoxia, it appeared to accentuate the deterioration in the control hearts but did not affect the experimental hearts adversely (Fig. 2).

Diminished phosphorylase activity may be responsible for glycogen accumulation in hearts of reserpinized rats or rats treated with beta-receptor blocking agents (21, 41). Activation of adenyl cyclase by catecholamines and the resultant conversion of phosphorylase to its active form is one mechanism for the initiation of glycogenolysis in cardiac anoxia (41). In the presence of catecholamine depletion with reserpine, anoxia stimulates phosphorylase activation and glycogenolysis normally (21, 42), perhaps because of the changing balance between ATP, and its breakdown products (43). The present study confirms the observation that normal catecholamine activity need not be present for adequate glycogenolysis to occur during anoxia.

The studies with 0 mM glucose and 20 mM glucose suggest that relatively small differences in glycogen levels can influence the cardiac response to hypoxia (Fig. 7). The lowering of cardiac glycogen in the presence of excess thyroid hormone (12, 13) may partially explain the relative intolerance of patients with angina pectoris to thyroid extract. Cardiac glycogen may also be lowered in adrenocortical insufficiency (15) and elevated in diabetes (11), hypothyroidism (14), hyperadrenalism (15), and physical training (13, 17). Whether such changes in cardiac glycogen are of clinical significance is unknown.

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