Effects of Hypoxia and Metabolic Inhibitors on the Intrinsic Heart Rate and Myocardial Contractility in Dogs

By Anthony D. Jose, M.A., and Frank Stitt, M.B.

ABSTRACT

Previous studies have shown that after autonomic blockade by propranolol and atropine, the intrinsic rate and contractile function of the heart were related in patients with myocardial failure. In this study the intrinsic rate and ventricular contractile force were measured in dogs during acute heart failure produced by agents which impair myocardial energy synthesis, to see whether these conditions reproduced the relationship found in naturally occurring myocardial disease in man. Anesthetized dogs, after propranolol and atropine, were either ventilated with 6% oxygen or given sodium cyanide, parachloromercuribenzoate, or dinitrophenol by intravenous infusion. Hypoxia and cyanide initially increased and then depressed both rate and contraction. Parachloromercuribenzoate depressed both. Dinitrophenol depressed only contraction. During hypoxia, cyanide, and parachloromercuribenzoate the percent changes in contractile force and intrinsic rate were linearly related ($r = 0.87$) with slopes of 3.2, 2.7, and 3.3, respectively. These results together with previous data during pentobarbital and aminophylline were consistent with a single linear relationship between rate and contractility ($r = 0.89$, slope 3.2). This resembled closely the relationship found previously in man. It is suggested that in both situations, this relationship may be determined by a dependence of rate and contractility on similar limited sources of energy.

ADDITIONAL KEY WORDS
denervated heart myocardial failure propranolol atropine myocardial energy synthesis

In a previous study in man, we used propranolol and atropine to study the intrinsic functional properties of the myocardium at different stages of myocardial disease (1, 1a). It was found that after autonomic blockade by these drugs, the heart rate became fixed to most stimuli, at a value which varied in different individuals inversely with the severity of the myocardial disease. This value, which we named the intrinsic heart rate, was directly related to each available index of left ventricular function. The measurement can be made simply and safely in normal subjects and in most cardiac patients, and we have suggested that it may prove useful for the assessment of myocardial function in clinical practice (1).

A similar relationship between spontaneous rate and contractility has been described in isolated rabbit atria during exposure to metabolic inhibiting agents (2, 3) and was evident in published studies of the failing dog heart-lung preparation (4). We found subsequently in dogs that after propranolol and atropine the intrinsic heart rate was linearly related to myocardial contractile force over a wide range of changes in myocardial function produced by pentobarbital and aminophylline (5). Pentobarbital is thought to depress oxidative metabolism in the myocardium (6), and aminophylline also probably affects myocardial metabolism through its inhibitory
action on the enzyme phosphodiesterase (7). Quantitatively, this relationship between rate and contraction present under certain conditions in animals resembled that found previously in patients with myocardial disease (5). Recent studies of the possible role of defective energy synthesis in the development of myocardial failure have continued to give apparently conflicting results (8-11). It was therefore of interest to study the effect of agents known to depress myocardial energy synthesis on the intrinsic rate and contractility of the dog heart after propranolol and atropine.

**Methods**

Details of the experimental preparation and methods were described in a previous report (5). Healthy mongrel dogs were anesthetized with a mixture of pentobarbital and Dial-urethane and ventilated at a constant volume with 100% oxygen. The chest was opened, and a Walton-Brodie strain-gauge arch sewn to the right ventricle to record myocardial contractile force. The electrocardiogram was monitored continuously and used to trigger a tachometer for the measurement of heart rate. Arterial pressure was measured from a catheter in the aortic arch and left ventricular pressure from a short rigid cannula inserted directly through the ventricular apex; the catheter and cannula were connected to Statham P-23g strain-gauge transducers. Blood flow in the ascending aorta was measured by a gated sine-wave electromagnetic flowmeter (Medicon K2000A), calibrated in vitro using normal saline flow through the excised aorta. Ventricular pressure and aortic flow were measured only in the control studies and those of hypoxia.

Satisfactory function of the strain-gauge arch was ensured at the start of each experiment, as described previously (5). Measurements of heart rate were made only in sinus rhythm. Arterial blood was collected periodically for analysis of 
P_{O2}, P_{CO2}, pH, and base excess; intermittent hyperventilation was used to minimize atelectasis, and in some dogs up to 20 ml of 5% sodium bicarbonate solution was given intravenously in the control period to counter early metabolic acidosis.

Body temperature was monitored from a thermocouple probe in the esophagus adjacent to the left atrium and was maintained between 36° and 38°C in all experiments unless otherwise noted.

Combined beta-receptor and postganglionic parasympathetic blockade was produced by an initial injection of propranolol, 0.2 mg/kg iv, and atropine, 1.0 mg/kg iv, over 2 minutes, and was maintained by a constant infusion of propranolol, 0.004 mg/kg/min, and atropine, 0.02 mg/kg/min. It has been shown previously that after this dose of propranolol, only small increases in myocardial contractile force and heart rate occur in response to either direct stimulation of the left stellate ganglion (2-msec pulses at supramaximal voltage and at 20/sec) or to isoproterenol injected intravenously (slug injection of 0.4 µg/kg) (1a). Experimental procedures were commenced 10 minutes after the initial injection, by which time the circulation was stable.

Thirty-three dogs were studied. Nine were used in the control studies. Six were subjected to hypoxia by changing the inspired gas to 6% oxygen in nitrogen without altering ventilation and continuing this until death of the animal. In the remainder, metabolic inhibiting agents were given by constant intravenous infusion at rates found previously to cause significant cardiac depression over 20 to 40 minutes. In five, sodium cyanide was infused at rates of 0.05 or 0.1 mg/kg/min. In nine, 2,4-dinitrophenol was given at rates of 0.8, 0.5, or 0.2 mg/kg/min. In three, sodium parachloromercuribenzoate was given at 0.5 to 0.8 mg/kg/min, and in one, 0.01 and 0.1 mg/kg/min. All drug solutions were warmed to 38°C before administration.

Measurements of cardiac function were recorded continuously in each experiment until either the death of the animal or the appearance of an ectopic arrhythmia.

For statistical analysis, readings of heart rate and myocardial contractile force were made from the original records at 2-minute intervals until the contractile force had fallen to 40% of its control level. Changes in both measurements were expressed as percents of their control values. The results in each experiment were inspected (e.g., Fig. 2), and no consistent departure from linearity was found. The individual regression relationships were therefore calculated assuming that they were linear. In each set of experiments, after homogeneity of the regression slopes was confirmed by an analysis of covariance, the data were pooled in the form of regression equations through mean values of rate and contractile force. Mean changes and regression coefficients were compared using Student's t-test. The confidence limits of correlation coefficients were estimated using Fisher's z-transformation, assuming this to be normally distributed.


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Results

Effects of Propranolol and Atropine.—Measurements made immediately before and 10 minutes after the initial injection of propranolol and atropine showed mean changes in rate of —20%, in contractile force of —27%, in mean aortic flow of —8%, and in mean aortic pressure of —9%. These values are similar to those found and discussed in a previous study (5).

The state of the circulation after autonomic blockade is described by the measurements in Table 1, which represent the mean baseline data for these experiments. They closely resemble equivalent data found previously (5), both in their absolute values and in the relatively small scatter of variables of myocardial function in different animals.

Body Temperature Changes.—Unless special precautions were taken to warm the animal, body temperature fell consistently and progressively after the injection of propranolol and atropine. Falls of up to 4°C over 30 minutes were noted in early studies, despite environmental temperatures maintained between 21° and 27°C.

To assess its effect after autonomic blockade, body temperature was varied over the range from 34° to 40°C by external warming and cooling in three dogs. The intrinsic heart rate changed directly with temperature in an approximately linear fashion, with a mean slope of 6.4 beats/min/°C. The changes in myocardial contractile force, however, were inconsistent. From 36° to 38°C, there were no changes greater than 10%; below 36°C there was a 30% rise in contractile force in one dog but no significant change in two; above 38°C there was no change in two dogs but a fall in one. The time from onset to peak of ventricular contraction (measured in milliseconds from recordings of contractile force at a paper speed of 100 mm/sec), shortened consistently as temperature rose, by approximately 9%/°C; simultaneously, but more variably, the rate of tension development increased with temperature; the resultant peak tension was therefore relatively little changed.

In subsequent studies, changes in body temperature during the experiments were limited to ± 1°C by use of an electric warming blanket, partial closure of the chest incision, and by warming all injectates to 38°C.

Control Studies.—Six dogs were observed for 45 minutes after autonomic blockade without any other intervention. The maximum observed changes over this period were: in intrinsic heart rate from +4% to —4%; in contractile force from +2% to —8%; in mean aortic flow from +4% to —12%; and in mean aortic pressure from +3% to —11%. Almost all of the changes in intrinsic heart rate could be accounted for by small changes in body temperature, although this was maintained between 36° and 38°C. Arterial blood pH ranged between 7.4 and 7.25 in different animals, and Pco2 between 30 and 60 mm Hg. Neither mild acidosis in two dogs (dilute hydrochloric acid infused intravenously to an arterial pH of 7.1) nor mild alkalosis in two dogs (5% sodium bicarbonate infused intravenously to a pH of 7.5) caused significant changes in intrinsic heart rate or contractile force.

Hypoxia.—Five minutes after commencing ventilation with 6% oxygen, the arterial Po2 had fallen from control levels of 400 to 600 mm Hg to between 27 and 35 mm Hg, where it remained subsequently.

The first cardiac response to hypoxia in each animal was an increase in all variables of myocardial function; this early phase was then

### Table 1

Control Data Recorded Ten Minutes after the Initial Injection of Propranolol and Atropine in Thirty-Three Open-Chest, Anesthetized Dogs

<table>
<thead>
<tr>
<th></th>
<th>Mean</th>
<th>± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (kg)</td>
<td>16.5</td>
<td>2.4</td>
</tr>
<tr>
<td>Esophageal temperature (°C)</td>
<td>36.9</td>
<td>0.5</td>
</tr>
<tr>
<td>Intrinsic heart rate (beats/min)</td>
<td>136</td>
<td>16</td>
</tr>
<tr>
<td>Mean aortic flow (L/min)*</td>
<td>1.05</td>
<td>0.15</td>
</tr>
<tr>
<td>Left ventricular end-diastolic pressure (mm Hg)*</td>
<td>7</td>
<td>2</td>
</tr>
<tr>
<td>Mean aortic pressure (mm Hg)</td>
<td>98</td>
<td>24</td>
</tr>
</tbody>
</table>

* Measured in 12 dogs only.
Measurements of cardiac function after propranolol and atropine during ventilation with 6% oxygen in dog no. 4 (Table 2). Cardiac arrest occurred in the forty-seventh minute.

Followed by a progressive decline in all functions until death. Typical changes are shown in Figure 1. A similar pattern was found in all studies, though there was considerable variation in its rate of evolution.

The peak of the early positive response was reached after an average of 10 minutes (±6), the contractile force increased by 36.1% (±17), and the intrinsic heart rate by 8.3% (±2.5); at this time there were also
average rises in mean aortic pressure of 13% (SD ± 12), in mean aortic flow of 33% (SD ± 35), in the peak velocity of aortic flow of 31% (SD ± 27), and in left ventricular stroke work of 71% (SD ± 54), while left ventricular end-diastolic pressure was reduced by 2 mm Hg (SD ± 1).

These early changes then became reversed. Contractile force, intrinsic heart rate, and aortic flow reached their control levels approximately together, after an average of 25 minutes (SD ± 15); by this time the mean aortic pressure had already fallen below its control level by 31% (SD ± 20), reflecting considerable peripheral vasodilatation (Fig. 1).

Contractile force reached 50% of its control level after an average of 47 minutes (SD ± 25); at this time the intrinsic heart rate was reduced by 21% (SD ± 10), and both aortic pressure and flow were below 50% of their control values. Shortly after, death occurred by sinus arrest, abruptly in four dogs and preceded by atrioventricular block in two.

Metabolic acidosis was not evident from arterial blood samples taken at 5 and 10 minutes; it was mild in samples at 20 minutes, and thereafter became progressively more severe until death.

Throughout the responses, changes in contractile force occurred predominantly through
TABLE 2

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>Duration of observations (min)</th>
<th>Correlation coefficient (r)</th>
<th>Regression coefficient (%ΔCF/%ΔIHR)</th>
<th>Sample SD of regression</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>28</td>
<td>0.98</td>
<td>2.08</td>
<td>5.2</td>
</tr>
<tr>
<td>2</td>
<td>34</td>
<td>0.86</td>
<td>2.76</td>
<td>17.2</td>
</tr>
<tr>
<td>3</td>
<td>17</td>
<td>0.95</td>
<td>2.67</td>
<td>3.3</td>
</tr>
<tr>
<td>4</td>
<td>24</td>
<td>0.77</td>
<td>4.57</td>
<td>25.7</td>
</tr>
<tr>
<td>5</td>
<td>48</td>
<td>0.98</td>
<td>3.79</td>
<td>5.5</td>
</tr>
<tr>
<td>6</td>
<td>80</td>
<td>0.91</td>
<td>3.87</td>
<td>13.6</td>
</tr>
<tr>
<td>Pooled data</td>
<td></td>
<td>0.87</td>
<td>3.22</td>
<td>14.5</td>
</tr>
</tbody>
</table>

CF = myocardial contractile force. IHR = intrinsic heart rate. * 95% confidence limits.

Changes in the rate of development of tension; the time from onset to peak of ventricular contraction showed no statistically significant change until the terminal phase of myocardial depression, when it shortened abruptly; at the peak of the early positive response, when rate was increased by 8%, the average time from onset to peak of ventricular contraction was prolonged by an average of 9%.

The changes in contractile force and intrinsic heart rate were approximately in phase throughout and maintained an apparently single and linear relationship during both the positive and negative phases of the response (Fig. 2). The linear correlation between these two measurements was highly significant in each experiment (Table 2).

Cyanide.—The infusion of sodium cyanide caused changes closely resembling those of hypoxia. In all experiments there was an initial positive response, maximal after an average of 5.6 minutes (so ± 1.6), with peak rises in contractile force of 20.4% (so ± 7.2) and in intrinsic heart rate of 5.8% (so ± 0.3); the changes in mean aortic pressure were variable and averaged zero at this time.

After its early rise, contractile force fell somewhat more rapidly than the intrinsic heart rate, reaching its control level after 7.8 minutes (so ± 2.0) when the heart rate was still raised by 1.5% (so ± 1.1); the mean aortic pressure by this time was already reduced by 33% (so ± 11).

Contractile force fell to 50% of its control value after 21 minutes (so ± 6), at which time the intrinsic heart rate was reduced by 19.2% (so ± 7.4), and mean aortic pressure by 68%. The experiments were terminated soon after by sinus arrest (in two), atrioventricular block (in two), or an ectopic tachycardia (in one).

Accurate measurements of the time from onset to peak of ventricular contraction and the rate of development of ventricular tension were not made in these studies. There did not appear, however, to be any major changes in the time from onset to peak of ventricular contraction until the terminal stages of depression of contractile force, when there was obvious shortening. There were no systematic changes in body temperature, or in acid-base balance, at least until the terminal stages. Two rates of cyanide infusion were employed (Table 3); their results differed only in the time taken over the phase of myocardial depression.

As with hypoxia, statistical analysis of the changes in contractile force and intrinsic heart rate confirmed a close and approximately linear relationship between them during both phases of the response (Table 3). This did not differ at the two rates of infusion of cyanide.

Dinitrophenol.—Difficulty was encountered at first in preventing rises in body temperature during administration of this drug. In three experiments, rises of 1.8° to 3°C were encoun-
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Regression Relationships between the Percent Changes in Intracellular Heart Rate and Contractile Force in Dogs during Intravenous Infusion of Sodium Cyanide

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>Infusion rate (mg/kg/min)</th>
<th>Duration of observations (min)</th>
<th>Correlation coefficient (r)</th>
<th>Regression coefficient (%ΔCF/%ΔIHR)</th>
<th>Sample SD of regression</th>
</tr>
</thead>
<tbody>
<tr>
<td>13</td>
<td>0.1</td>
<td>20</td>
<td>0.94</td>
<td>3.55</td>
<td>8.7</td>
</tr>
<tr>
<td>14</td>
<td>0.1</td>
<td>22</td>
<td>0.98</td>
<td>2.38</td>
<td>4.7</td>
</tr>
<tr>
<td>15</td>
<td>0.1</td>
<td>23</td>
<td>0.97</td>
<td>2.78</td>
<td>6.6</td>
</tr>
<tr>
<td>16</td>
<td>0.05</td>
<td>30</td>
<td>0.96</td>
<td>2.83</td>
<td>7.8</td>
</tr>
<tr>
<td>17</td>
<td>0.05</td>
<td>21</td>
<td>0.88</td>
<td>1.30</td>
<td>10.6</td>
</tr>
<tr>
<td>Pooled data</td>
<td></td>
<td></td>
<td>0.93</td>
<td>2.72</td>
<td>8.4</td>
</tr>
</tbody>
</table>

CF = myocardial contractile force. IHR = intrinsic heart rate. *95% confidence limits.

Correlation coefficient (r) = (0.89 to 0.96) (اس = ± 0.14)

CF = myocardial contractile force. IHR = intrinsic heart rate.

In six subsequent experiments, however, body temperature was controlled to within 1°C. Under these conditions, the changes in contractile force clearly depended on the rate of infusion of dinitrophenol, while there were no changes in the intrinsic heart rate at any infusion rate. When the drug was given at 0.8 mg/kg/min (in two dogs), there were rises in contractile force over the first 10 to 15 minutes, reaching +80% and +110%; these were followed by very rapid falls to zero tension over 2 to 3 minutes. Although the electrocardiogram terminally showed marked S-T segment elevation and numerous ectopic beats, the basic sinus rate remained unchanged for as long as it could be observed. The intermediate infusion rate of 0.5 mg/kg/min (in two dogs) caused transient rises in contractile force to peaks of +20% and +60% in the first 10 minutes, and subsequent falls towards zero tension over 20 to 30 minutes, ending as before in ectopic activity without prior change in the intrinsic heart rate. At the slowest infusion rate of 0.2 mg/kg/min (in two dogs), the only change was a slow fall in contractile force, which reached −40% and −60% after 25 and 30 minutes, and ended abruptly by cardiac arrest in both. In all experiments, the predominant change in mean aortic pressure was a fall, though the control level was maintained for longer during the fast infusions of dinitrophenol. There were no significant changes in the time from onset to peak of ventricular contraction throughout the infusions, except for some shortening terminally.

Parachloromercuribenzoate.—Infusion of this at 0.5 mg/kg/min (in two dogs) and 0.8 mg/kg/min (in one dog) caused simultaneous depression of both contractile force and intrinsic heart rate, without initial stimulation.

Contractile force fell to 50% of its control value after an average of 19 minutes, and at this time the intrinsic heart rate had fallen by an average of 15%. Continued infusion led to the appearance of atrioventricular block in each experiment, after an average of 28 minutes; this was preceded for 2 to 3 minutes by a progressive lengthening of the P-R interval. The time from onset to peak of ventricular contraction remained unchanged until this terminal stage, during which it shortened rapidly by some 30%. The mean aortic pressure generally followed the changes in contractile force, giving no evidence of any major changes in peripheral vessels. There were no significant changes in either body temperature or blood acid-base balance.

There was again a close and apparently linear relationship maintained between the changes in contractile force and intrinsic heart rate throughout each experiment. The calculated relationships are shown in Table 4.

Lest too rapid infusion of parachloromercuribenzoate had masked an early stimulation of the myocardium in these studies, one further dog was given the drug at slower rates: infusion at 0.01 mg/kg/min for 15
TABLE 4
Regression Relationships between the Percent Changes in Intrinsic Heart Rate and Contractile Force in Dogs during Intravenous Infusion of Sodium Parachloromercuribenzoate

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>Infusion rate (mg/kg/min)</th>
<th>Duration of observations (min)</th>
<th>Correlation coefficient (r)</th>
<th>Regression coefficient (%ΔCF/ΔiHR)</th>
<th>Sample SD of regression</th>
</tr>
</thead>
<tbody>
<tr>
<td>27</td>
<td>0.5</td>
<td>34</td>
<td>0.97</td>
<td>4.20</td>
<td>4.3</td>
</tr>
<tr>
<td>28</td>
<td>0.8</td>
<td>25</td>
<td>0.93</td>
<td>2.77</td>
<td>5.5</td>
</tr>
<tr>
<td>29</td>
<td>0.5</td>
<td>30</td>
<td>0.90</td>
<td>3.20</td>
<td>11.2</td>
</tr>
<tr>
<td>Pooled data</td>
<td></td>
<td></td>
<td>0.92</td>
<td>3.32</td>
<td>(0.85 to 0.95)*</td>
</tr>
</tbody>
</table>

CF = myocardial contractile force. IHR = intrinsic heart rate. * 95% confidence limits.

minutes produced no changes in contractile force or intrinsic heart rate; the rate was increased to 0.1 mg/kg/min for a further 15 minutes, and contractile force fell gradually to —12% while the intrinsic heart rate fell to —5%. At no time was there a rise in either measurement.

Comparison of the Relationship between Myocardial Contractile Force and Intrinsic Heart Rate in Different Studies.—In Figure 3 are shown the individual calculated regression relationships between the percent change in contractile force and the percent change in intrinsic heart rate from this study during

![Figure 3](http://circres.ahajournals.org/)

Individual regression slopes calculated from pooled data during hypoxia, cyanide infusion, and parachloromercuribenzoate infusion in this study, and during aminophylline and pentobarbital infusions from a previous study (5). Small intercepts on the ordinate shown by the calculated relationships during hypoxia (positive) and cyanide (negative) were neglected for this comparison. The intercepts of the relationships during aminophylline, pentobarbital, and parachloromercuribenzoate (PCMB) did not differ significantly from zero. From the pooled data of all 23 experiments, the calculated linear regression equation was Y = 3.24 X + 4.2, (r = 0.89).
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hypoxia, cyanide infusion, and parachloromercuribenzoate infusion, together with those during infusion of pentobarbital and aminophylline from a previous study (5). Their close similarity is apparent. Statistical comparison of the regression slopes indicated that the relationship during cyanide differed significantly in slope from those during aminophylline and pentobarbital (0.02 < P < 0.05), but not from those during hypoxia or parachloromercuribenzoate (0.2 > P > 0.1), which themselves did not differ from the other two (0.2 > P > 0.1). We have no basis on which to interpret these small differences in slope. In view of the nature of the experiments, and with the knowledge that the absolute control levels of the intrinsic heart rate and contractile force are probably determined independently by different factors in each animal, it is at least possible that each of the separate regression relationships in Figure 3 is in fact describing one single relationship between the heart rate and contractile force existing throughout these experiments. On this assumption, analysis of the pooled data from all 23 experiments (four with aminophylline, five with pentobarbital, six with hypoxia, five with cyanide, and three with parachloromercuribenzoate) yielded a linear regression coefficient for the relationship between percent change in contractile force and percent change in intrinsic heart rate of ±3.24 (±0.08), and a linear correlation coefficient of 0.89.

Additional Studies.—In some experiments, particularly those with hypoxia and cyanide, there occurred falls in mean aortic pressure of considerable magnitude which, despite the coronary vasodilatation which was presumably present, might have directly influenced the changes in contractile force, at least over the latter part of the experiments. To minimize this possibility, the data used for statistical analysis in each case excluded values of contractile force below 40% of its control level. At this stage in the hypoxia and cyanide experiments, the mean aortic pressure had fallen to between 25% and 55% of its control level in different dogs.

Attempts were made in extra studies with hypoxia to maintain the arterial pressure. In two dogs, rapid blood transfusions failed to do this, apparently because of the very large capacitance of the vascular bed under these conditions. In two dogs, an intravenous infusion of methoxamine for 3 to 5 minutes during the phase of depression of contractile force and intrinsic heart rate raised the mean aortic pressure from 50% and 55% to 80% and 90%, respectively, of its control level; this did not significantly interrupt the downward course of changes in contractile force; it appeared, in fact, that methoxamine under these conditions had a small depressant effect on contractile force; certainly there was no rise in it coincident with partial restoration of the arterial pressure.

In further control studies after propranolol and atropine, falls in mean aortic pressure were produced by procedures thought not to influence myocardial function directly under these conditions. In two dogs, acetylcholine was infused into the ascending aorta at a rate which lowered mean aortic pressure by 50%; this did not significantly change either the intrinsic heart rate or contractile force. In other studies, mean aortic pressure was lowered by hemorrhage (in two dogs), by infusing phenoxybenzamine intravenously (in 3 dogs), and by intra-aortic infusion of histamine (in two dogs); hypotension by these methods had no immediate effects on contractile force; when after sustained hypotension in some dogs there was a fall in contractile force, it was in each case accompanied by a fall in the intrinsic heart rate, in a relationship similar to the one already described.

It was noted in previous studies with aminophylline (5) that the mean aortic pressure fell to low levels although contractile force and intrinsic heart rate were markedly increased. Finally, a similar relationship between rate and contractility has been described in isolated rabbit atria beating spontaneously under isometric conditions (2, 3).

We do not think, therefore, that the falls in mean aortic pressure in the present experi-
ments had any important influence on the relative changes observed in contractile force and intrinsic heart rate. However, studies with cardiac performance and coronary perfusion maintained constant may be required to prove this.

Discussion

The primary aim in these experiments was to study relative changes in rate and contractile performance of the heart after propranolol and atropine during gradual depression of its energy-forming processes, rather than to examine specifically the mode of action of each agent used on the myocardium. In most regards, these specific results were consistent with previous findings in the literature.

Hypoxia typically produces a biphasic change in the performance of mammalian hearts. In intact animals (12, 13) and in man (14), the early rise in cardiac performance is partly attributable to peripheral vasodilatation and partly to increased sympathoadrenal activity. Early rises in contractility during hypoxia have also been found, however, in isolated perfused atria (3), in the dog heart-lung preparation after treatment with reserpine (15), and in dogs after beta-receptor blockade (13). In contrast, no early stimulation was found in isolated perfused rat hearts after treatment with reserpine (16), perhaps because in that study myocardial oxygen deprivation was more rapid and more severe than in the others described. The early myocardial stimulation by hypoxia in this study may have arisen partly from stimuli to beta receptors that were incompletely blocked. However, it has been shown (1a) that the dose of propranolol used here reduces the responses in contractile force to electrical stimulation of the left stellate ganglion from +125% to +11%, and to injected isoproterenol from +110% to +9%. It is, therefore, unlikely that the present rise in contractile force, which averaged +36%, could be explained solely by increased activity of beta receptors. Moreover, the slight prolongation of the time from onset to peak of ventricular contraction (by 9%) at the peak of the contractile force response, at which time the rate was also increased (by 8%), suggests some other mechanism, since stimuli to beta receptors characteristically shorten this interval. These data therefore suggest that early in hypoxia, stimulation of the myocardium may occur through some non-adrenergic mechanism, the nature of which is unknown. Such a mechanism, if it exists, may also be concerned in the residual increase of heart rate during exercise in dogs after combined cardiac denervation and beta-receptor blockade, which appears to be myogenic in origin (17). It may also be concerned in elevations of the measurement of intrinsic heart rate in man which we have observed in some patients with angina pectoris (unpublished data).

Cyanide infusion caused functional changes in the myocardium which closely resembled those of hypoxia. This has been noted previously in isolated atria (2) and would be predicted from the primary site of action of cyanide on the electron transport system.

The effects of dinitrophenol were similar to those found previously in the isolated heart, except that an initial stimulatory effect of large doses on contraction has not usually been described in isolated preparations; we are able to offer no explanation for this. Resistance of the heart rate to depression by dinitrophenol has been a uniform finding in all studies of isolated atria (2, 3) and isolated heart preparations (18, 19). Too little is known of the intracellular effects of dinitrophenol to interpret these findings, but in the present context they constitute an unexplained exception to our general finding that related changes in contractile force and intrinsic heart rate occur when energy synthesis in the myocardium is impaired.

Parachloromercuribenzoate was used as an inhibitor of sulfhydryl group-dependent enzymes, after earlier studies with iodoacetate showed its cardiac action to be too slow for the conditions of our experiments. It is unlikely that parachloromercuribenzoate has any single specific site of action on myocardium metabolism; recent studies of its effect on isolated cardiac tissue (20) suggested
several different sites of metabolic inhibition. Its functional effects in this study were similar to those described in isolated atria after prolonged exposure to iodoacetate (2).

A relationship between the intrinsic rate and contractility was sought in these experiments because previous studies in man had suggested its existence in the course of naturally occurring heart disease (1, 1a). A relatively uniform relationship between these two functions was found under the specific conditions of these experiments, during each of the five procedures illustrated in Figure 3. From what is known of the separate determinants of the sinus rate and of myocardial contractility, it appears clear that under many other conditions, there can be no close relationship between them. Some such conditions were implied and others demonstrated in the present study. Variations in autonomic nervous and humoral activity in the myocardium apparently obscure the relationship; presumably this partly explains why no such relationship has been found in previous studies comparable to these but without autonomic blockade. Even in surgically denervated or isolated heart preparations, it seems that locally acting norepinephrine can modify the pattern of myocardial functional changes seen in many experiments enough to obscure the relationship; as discussed elsewhere (5), however, the relationship does become evident in dog heart-lung preparations under conditions associated with acute norepinephrine depletion (4), and has been found regularly in studies of isolated mammalian atria (2, 3).

After autonomic blockade, three conditions were noted in these studies in which changes occurred in either the intrinsic heart rate or contractile force unrelated to changes in the other: during variations of body temperature, after very large doses of aminophylline (5), and during the infusion of dinitrophenol. It is likely that other such conditions exist, though it is difficult to predict these, since there is little or no previous published data describing the simultaneous behavior of spontaneous rate and myocardial contractility in hearts deprived of autonomic activity.

Quantitatively, there was little resemblance between the relationship found here (Fig. 3) and that which underlies the Treppe phenomenon in cardiac muscle; the latter characteristically has a very much lower slope (21) and may not be demonstrable at all by strain-gauge arch measurements of isometric force made in vivo (22). Also, in recent experiments (unpublished), we have confirmed that periods of altered ventricular rate produced by external pacing do not significantly influence the relationship existing between the intrinsic heart rate and contractile force.

Not enough evidence is yet available to explain the relationship found with any confidence. A direct interdependence of the intrinsic heart rate and contractile force appears unlikely. It is more probable that under certain conditions both functions are dependent on some third common factor. Each of the five procedures in Figure 3 is thought or known to influence energy synthesis in the myocardium. It is not suggested that they had single or specific biochemical sites of action. It is even possible in our experiments that some of the myocardial changes were produced indirectly by blood-borne products from elsewhere in the body, though the similarity of our results to those described in isolated hearts suggests that such factors were not important. Together these procedures represent a variety of different influences on the biochemical processes concerned in energy synthesis, and from what is known of their individual specific actions on the myocardium, they would appear to have little else in common. It is therefore possible that the availability of energy was the common factor on which the intrinsic heart rate and contractile force were dependent in our experiments.

Lack of information makes it difficult to discuss this suggestion in terms of the regulation of intrinsic rate and contractility in the myocardium. Little is known of the processes which determine the intrinsic rate in pacemaker tissue, or of the metabolism on which they depend. Recent evidence has
suggested that the contractile force and total high-energy phosphate stores of the myocardium do not always change together (13, 10). However, present understanding of the processes involved in excitation-contraction coupling (23, 24) implies that these processes inherently impose a control over the force of contraction, prior to the utilization of energy by the contractile process itself. Since these processes, like those which determine rate, are probably membrane phenomena, their energy requirements are probably small in relation to the total energy stores of the cell (25). It is conceivable, therefore, that rate and contractile force could be mutually dependent on a limited availability of energy at the membrane, without such limitation being detectable from measurements of the total stores or capacity for energy synthesis in the cell.

To some extent, this loose hypothesis is consistent with the negative results in our study as well as with the positive. Norepinephrine would predictably increase the rate of synthesis and availability of energy and may therefore obscure the relationship by rendering one or both functions no longer dependent on this as a limiting factor. The dissociation of the intrinsic heart rate and contractile force during changes in temperature seems at first to contradict their mutual dependence on metabolic processes; however, temperature changes directly influence the contractile process itself in a complex manner, with results which vary with the extent of shortening of the contractile elements (26), and such changes could mask any simultaneous changes caused by altered metabolism; the effects of temperature change on the intrinsic heart rate were quite consistent with its direct dependence on some metabolic process. As discussed previously (5), high concentrations of aminophylline may interfere with the entry of calcium ions into the cell, which would alter the contractile force at a stage in excitation-contraction coupling beyond that at which we suppose the relationship between contractile force and intrinsic heart rate may be determined. We have no explanation for the findings with dinitrophenol.

Because the relationship found in this study bears a close quantitative resemblance to that found in patients with myocardial disease between the intrinsic heart rate and the contractile performance of the heart (1, 1a), it is likely that both findings describe the same myocardial phenomenon. In the present study, related changes in intrinsic heart rate and contractile force were found in each animal during progressive changes in myocardial function, while in man the relationship between the intrinsic heart rate and contraction was found in different patients, each with a different severity of myocardial disease. To apply our hypothesis from this study to our findings in man, therefore, it is necessary to think in terms of an energy-forming pathway of a given capacity in each individual, which under normal conditions of substrate and oxygen supply and in the absence of norepinephrine, yields energy at a characteristic rate which itself determines the absolute levels of the intrinsic heart rate and contractile strength in that subject. Such a concept appears consistent with the observed behavior of the intrinsic heart rate in man with increasing age (27) and in different forms of heart disease (1).

It remains uncertain at present whether or not there is a necessary impairment of energy synthesis in failing heart muscle (8-11, 13). Some of the apparently conflicting evidence may be related to methodological differences (9, 11); it may also be that myocardial failure, as defined mechanically, can under different circumstances arise from more than one alternative biochemical lesion in the cell. If the analogy we suggest between the present results and our findings in man is valid, and if our hypothesis to explain the present results is correct, two relevant conclusions would follow. First in naturally occurring myocardial disease in man, a depression of intrinsic myocardial contractility may be causally related to a diminished capacity for energy synthesis, not necessarily throughout the myocardium but involving only those supposed pathways concerned in the control of the intrinsic heart rate and contractile force.
INTRINSIC HEART RATE AND CONTRACTILITY

Secondly, such a depression of the intrinsic myocardial contractility could be assessed in man by measurement of the intrinsic heart rate, which we have shown is a simple clinical procedure (1).

To establish the validity of these conclusions will require further experimental and clinical studies of the conditions under which the intrinsic heart rate and contractility of the myocardium are, and are not, related. At present, most of the conditions shown experimentally to dissociate the two either do not occur or can be avoided in clinical circumstances. Clinical measurements of the intrinsic heart rate have meanwhile been made in normal subjects and in many forms of heart disease (1 and unpublished observations). The results suggest that the intrinsic heart rate is consistently and quantitatively related to functional myocardial changes which occur in heart disease, particularly to such tangible measures of the overall functional capacity of the myocardium as exercise tolerance, mortality rates in surgery, and hemodynamic estimates of ventricular function. These may all be clinical counterparts of intrinsic myocardial contractility.

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References


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