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THE EFFECT OF HYPOCAPNIA ON
CORONARY VASCULAR RESISTANCE IN THE DOG

DISSERTATION
Presented in Partial Fulfillment of the Requirements for
the Degree Doctor of Philosophy in the Graduate
School of The Ohio State University

By

Ina Claire Ehrhart, B.S.

* * * * *

The Ohio State University
1972

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## LIST OF TABLES

<table>
<thead>
<tr>
<th>Table</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Raw Data in Untreated Animals</td>
<td>34</td>
</tr>
<tr>
<td>2</td>
<td>Raw Data in Propranolol Treated Animals</td>
<td>36</td>
</tr>
<tr>
<td>3</td>
<td>Response to Hypocapnia in 6-Hydroxydopamine Pretreated Animals</td>
<td>47</td>
</tr>
</tbody>
</table>
# LIST OF FIGURES

<table>
<thead>
<tr>
<th>Figure</th>
<th>Description</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Coronary Occlusion with Hyperemic Response</td>
<td>21</td>
</tr>
<tr>
<td>2</td>
<td>Response to Isoproterenol</td>
<td>24</td>
</tr>
<tr>
<td>3</td>
<td>Response to Isoproterenol after Propranolol</td>
<td>25</td>
</tr>
<tr>
<td>4</td>
<td>Resistance Calculation Curves</td>
<td>27</td>
</tr>
<tr>
<td>5</td>
<td>Response to Hypocapnia in Untreated Animal</td>
<td>31</td>
</tr>
<tr>
<td>6</td>
<td>Response to Hypocapnia after Propranolol</td>
<td>32</td>
</tr>
<tr>
<td>7</td>
<td>Late Diastolic Resistance</td>
<td>38</td>
</tr>
<tr>
<td>8</td>
<td>Late Diastolic Pressure</td>
<td>40</td>
</tr>
<tr>
<td>9</td>
<td>Stroke Coronary Blood Flow</td>
<td>42</td>
</tr>
<tr>
<td>10</td>
<td>Heart Rate</td>
<td>43</td>
</tr>
<tr>
<td>11</td>
<td>Minute Coronary Blood Flow</td>
<td>45</td>
</tr>
<tr>
<td>12</td>
<td>Response to Hypocapnia after 6-Hydroxydopamine Treatment</td>
<td>49</td>
</tr>
<tr>
<td>13</td>
<td>Isoproterenol Response in 6-Hydroxydopamine Treated Dog</td>
<td>50</td>
</tr>
<tr>
<td>14</td>
<td>Calculated Regression Lines for Flow-Pressure Relationships During Induced Pressure Oscillations</td>
<td>52</td>
</tr>
<tr>
<td>15</td>
<td>Calculated Regression Lines for Flow-Pressure Relationships During Induced Pressure Oscillations in 6-Hydroxydopamine Treated Dogs</td>
<td>53</td>
</tr>
</tbody>
</table>
TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACKNOWLEDGEMENTS</td>
<td>ii</td>
</tr>
<tr>
<td>VITA</td>
<td>iii</td>
</tr>
<tr>
<td>LIST OF TABLES</td>
<td>iv</td>
</tr>
<tr>
<td>LIST OF FIGURES</td>
<td>v</td>
</tr>
<tr>
<td>INTRODUCTION</td>
<td>1</td>
</tr>
<tr>
<td>Chapter</td>
<td></td>
</tr>
<tr>
<td>I. METHODS AND MATERIALS</td>
<td>19</td>
</tr>
<tr>
<td>A. Preparation</td>
<td>19</td>
</tr>
<tr>
<td>B. Experimental Design</td>
<td>23</td>
</tr>
<tr>
<td>C. Data Analysis</td>
<td>26</td>
</tr>
<tr>
<td>II. RESULTS</td>
<td>30</td>
</tr>
<tr>
<td>A. Response to Hypocapnia before and after Propranolol Treatment</td>
<td>30</td>
</tr>
<tr>
<td>1. Resistance</td>
<td>33</td>
</tr>
<tr>
<td>2. Pressure</td>
<td>39</td>
</tr>
<tr>
<td>3. Stroke Coronary Blood Flow</td>
<td>41</td>
</tr>
<tr>
<td>4. Heart Rate</td>
<td>41</td>
</tr>
<tr>
<td>5. Minute Coronary Blood Flow</td>
<td>44</td>
</tr>
<tr>
<td>6. Cardiac Output</td>
<td>44</td>
</tr>
<tr>
<td>B. Response to Hypocapnia after Treatment with 6-Hydroxydopamine</td>
<td>44</td>
</tr>
<tr>
<td>III. DISCUSSION</td>
<td>55</td>
</tr>
</tbody>
</table>
### TABLE OF CONTENTS (Continued)

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>IV. SUMMARY AND CONCLUSIONS</td>
<td>69</td>
</tr>
<tr>
<td>REFERENCES</td>
<td>71</td>
</tr>
</tbody>
</table>
Hypocapnia, a decrease in arterial PCO₂ due to over-ventilation, is an area of medical interest during anesthesia or other mechanically assisted respiration. In addition, hypocapnia may occur due to over-breathing during acclimatization to altitude. In spite of the interest in this area, the effects of hypocapnia on the level of coronary blood flow remain controversial. The divergent results observed in animal experiments during hypocapnia may reflect the wide variety of preparations and the multitude of different devices used to measure or estimate coronary blood flow. Individual coronary flow pulses have apparently never been studied in the closed-chest animal during hypocapnia. The flowmeter employed in this work made such a study possible.

The purpose of this paper is to present evidence for a decrease in coronary vascular resistance and an increase in minute coronary blood flow during hypocapnia. Secondly, we will demonstrate that this resistance decrease is mediated through the beta-adrenergic receptors. Finally, a theory is proposed which may account for the decrease in coronary vascular resistance during hypocapnia.

It has been known for many years that CO₂ is a potent vasodilator. Markwalder and Starling (1) demonstrated that increasing the tension of CO₂ in the blood perfusing the heart-lung preparation caused dilation of the coronary vessels which was reversed when the CO₂ was
discontinued. Ventilation of the heart-lung preparation with 7% CO₂ raised the coronary blood flow above the 3% CO₂ level (2). On the other hand, hypercapnia in the intact open-chest dog was found to have no significant effect (3), (4). Some more recent studies demonstrate that myocardial blood flow is increased during hypercapnia (5), (6). Daugherty et al. (7), using extracorporeal circulation and maintaining flow constant, perfused the left common coronary artery in the open-chest dog with blood of varying PO₂ and PCO₂ values. Reduction of the pH of the perfusing blood by raising CO₂ tension produced a fall in coronary resistance to flow. This fall in resistance to flow was concurrent with a decrease in left ventricular contractile force as measured by a 120-ohm strain gauge arch sutured to the surface of the left ventricle. Ledingham (8), in the closed-chest dog, demonstrated that inhalation of CO₂ sufficient to increase the arterial PCO₂ from 40 to approximately 100 mm Hg increased coronary blood flow without any consistent changes in mean arterial blood pressure, heart rate, or cardiac output. During hypercapnia the coronary arteriovenous oxygen content difference, and calculated myocardial oxygen consumption were reduced.

In 1971, Kosche et al. (9) measured coronary blood flow with an electromagnetic flowmeter in open-chest dogs mechanically ventilated with 4% CO₂ during the control period and 8% CO₂ during the experimental period. Heart rate was kept constant by electrical stimulation of the right auricle after elimination of the sinus node. Under these conditions coronary flow did not change with hypercapnia. Systolic
and diastolic aortic pressure, left ventricular pressure, and the maximal rate of pressure rise in the left ventricle remained unchanged. They therefore concluded that alterations of coronary blood flow caused by hypercapnia can be explained by a change of hemodynamic conditions.

Most investigators have reported a decrease or no change in coronary blood flow during hypocapnia. Scheuer (10) mechanically hyperventilated closed-chest dogs with room air and estimated coronary blood flow with $^{131}$I aminopyrine. He found that coronary blood flow decreased significantly from 70.6 to 25.8 ml/100g heart/min. However, mean arterial blood pressure dropped from a control of 128 to 65 mm Hg. McArthur (11) measured coronary blood flow in open-chest dogs by the bubble flowmeter method. He found that coronary blood flow decreased with decreasing arterial PCO$_2$. Concurrently, in each hyperventilation experiment, both aortic pressure and heart rate were also decreased. West et al. (12) visualized and photographed the coronary vessels in the closed-chest anesthetized dog by selective arteriography. Ventilation over a five minute period with 100% oxygen produced a mild constriction of the coronary bed. Sobol et al. (13) estimated coronary blood flow in anesthetized open-chest dogs by measuring coronary sinus outflow. An increase in the oxygen supply to the heart resulted in a decreased coronary blood flow with no change in arterial pH or PCO$_2$. A decrease in coronary blood flow was observed when the arterial blood oxygen content was increased from an average of 18.1 vol.% during air ventilation to 20.9 vol.% by ventilation
with 100% oxygen. A 22% decrease in coronary sinus outflow was associated with an average increase in coronary sinus blood oxygen content from 4.4 vol.% during air ventilation to 5.6 vol.% during 100% oxygen ventilation. There was no change in pH or PCO₂.

Feinberg et al. (14) isolated the right side of the heart in the anesthetized dog and collected blood subsequently flowing into the right atrium on the assumption that it represented total coronary flow. He observed an increase in coronary blood flow relative to cardiac effort during hypercapnia but not during hypocapnia. Cardiac effort was defined as heart rate times mean arterial pressure as these investigators found this product correlated with myocardial oxygen consumption. Rowe et al. (15) determined coronary blood flow by the nitrous-oxide saturation method in closed-chest anesthetized dogs and in unanesthetized human subjects. In dogs, the coronary blood flow tended to remain the same or rose slightly but not significantly with hyperventilation. Coronary blood flow was significantly reduced by voluntary hyperventilation in the human subjects. Daugherty et al. (7) perfused the left common coronary artery in the open-chest dog by extracorporal circulation. The flow was held constant while the PO₂ and PCO₂ of the perfusing blood were varied. Elevation of the PO₂ to greater than 650 mm Hg did not increase perfusion pressure or contractile force as measured by a 120-ohm strain gauge arch sutured to the surface of the left ventricle. Switching from 95% O₂-5% CO₂ to air ventilation of the isolated lung caused a transient fall in perfusion pressure, (decreased resistance), followed by a rise to a
level which by 9 minutes was significantly above that seen during ventilation with the 95% O₂-5% CO₂ mixture. Contractile force immediately rose then leveled off at an intermediate value. Despite this rise in contractile force, aortic pressure fell.

Factors Influencing Coronary Blood Flow

In a study of coronary blood flow or resistance changes in the coronary vessels, it is necessary to outline the factors that have been demonstrated to participate in the regulation of coronary blood flow. These factors actively or passively determine vessel radius or influence blood viscosity. The chemical influences which actively determine vessel radius include ions, myocardial oxygen tension, and metabolites. Direct neural control via the autonomic nervous system actively determines changes in vessel radius. Passive determinants of vessel radius include myocardial compression of the coronary vascular bed or other changes in extravascular pressure. Those factors which determine blood viscosity also influence the rate of coronary blood flow.

Ions

In addition to changes in hydrogen ion or arterial PCO₂ which were discussed above, potassium ion (K⁺) has been implicated in the control of coronary blood flow. Katz and Lindner (16), in 1938, demonstrated that K⁺ administered into the coronary artery in Langendorff preparations of the dog heart could produce coronary vasodilation. Driscoll and Berne (17) confirmed these findings in
open-chest anesthetized dogs and McKeever et al. (18) observed the local effect of $K^+$ in the perfused coronary vascular tree of the dog. The latter group concluded that the direction of the effect is a function of $K^+$ concentration. Scott et al. (19), in the anesthetized dog, demonstrated that active hyperemia, reactive hyperemia, and the autoregulation of blood flow result, at least in part, from alteration in the chemical environment of the blood vessels but this occurred in the absence of a regular change in the $K^+$ concentration of the blood.

Myocardial Oxygen Tension and Metabolites

Myocardial oxygen tension and metabolites have been cited as primary determinants of active changes in vessel radius. In the heart-lung preparation, Hilton and Eicholtz (2), in 1925, and Gremels and Starling (20), in 1926, clearly showed that a decrease in the oxygen content of the perfusing blood lowers the resistance to flow through the coronary. Shipley and Gregg (21) stimulated either stellate ganglion or its cardiac branches in the anesthetized open-chest dog and observed a large and sustained increase in coronary flow without changes in heart rate or mean blood pressure. They were unable to obtain an increased coronary flow response without evidence of increased cardiac work or metabolism, e.g. increased cardiac input and increased oxygen utilization. The lack of conclusive evidence that the cardiac nerves have a direct vasomotor influence and the observation that cardiac metabolism is considerably increased during sympathetic nerve stimulation supported their thesis that augmented cardiac metabolism was responsible for the coronary vasodilation by elaborating
metabolites and/or producing cardiac hypoxia as a result of the increase in oxygen uptake. Eckenhoff et al. (22) obtained a good correlation of coronary blood flow and myocardial oxygen consumption over a range of arterial blood pressure and cardiac output in anesthetized dogs. Foltz et al. (23) in intact anesthetized dogs found a linear relationship between coronary blood flow and cardiac oxygen consumption with high coefficients of correlation. This relationship was equally well demonstrated for different anesthetic agents as well as for repeated studies on the same animals. Katz and Feinberg (24) concluded that the cardiac performance requiring oxygen determines coronary flow while the arteriovenous oxygen difference across the bed remains constant. However, the level of the arteriovenous oxygen difference is altered by local conditions of oxygen and carbon dioxide concentrations, and the presence of catecholamines. Hence, the level of coronary flow for a given amount of cardiac oxygen consumption is altered by these circumstances. Although it is clear that local hypoxemia can reduce the resistance to blood flow through most systemic vascular beds, there is evidence that this action in the coronary vascular bed is manifest only after the oxygen tension reaches a certain critical level. For example, Daugherty et al. (7) perfused the left common coronary artery of the dog with blood of reduced oxygen tension and content over the range of normal arteriovenous difference and found little change in coronary vascular resistance.

The fall in coronary vascular resistance associated with decreased myocardial oxygen tension might result in part from passive
vasodilation due to a rise in vascular transmural pressure since there is an associated fall in left ventricular contractile force. Ng et al. (25) subjected paced, isovolumetric, canine left ventricle preparations to hypoxia under conditions of constant coronary perfusion pressure or constant coronary blood flow. There was a significant increase in left ventricular contractile strength with moderate hypoxia (oxygen saturation less than normal but greater than 50%) and a depression with severe hypoxia (oxygen saturation less than 50%). Daugherty et al. (7) found that reduction of perfusate PO₂ to 21 mm Hg (7 vol.%) in the coronary vascular bed produced a significant fall in contractile force as well as decreased vascular resistance. These effects were even greater at PO₂ of 9 mm Hg (1.3 vol.%). The latter level could not be maintained for more than a few minutes without complete failure of heart pumping ability.

Berne et al. (26), in 1957, in experiments in open-chest dogs and on fibrillating heart preparations, demonstrated that reduction of oxygen content of arterial blood produces increases in coronary blood flow only when coronary sinus oxygen levels fall below 5.5 vol.%. High perfusion pressures were employed in order to increase coronary flow to the extent that coronary sinus blood became relatively rich in oxygen. Under these conditions it was possible to demonstrate that a moderate lowering of arterial oxygen content does not decrease coronary resistance by a direct action on the vessel walls. Coronary vasodilation in hypoxemia appears to be related to myocardial hypoxia.

Adenosine and the adenine nucleotides, adenosine monophosphate (AMP),
adenosine diphosphate (ADP), and adenosine triphosphate (ATP) are potent vasodilators in most vascular beds and have been implicated in the regulation of coronary vascular resistance. Drury and Szent-Györgyi (27) demonstrated a coronary vasodilation when adenylic acid or adenosine was injected intravenously into the dog. Drury and Wedd (28) confirmed these observations in the perfused rabbit heart where, for a given perfusion pressure, injection of 0.1 µg of adenosine produced an increase in flow which ranged from 100 to 250% of control. Maximal flow was brought about by doses which had no influence upon heart rate. Wolf and Berne (29) demonstrated in the coronary vascular bed of the dog that whereas other nucleotides and nucleotide derivatives, with the exception of uridine triphosphate (UTP), are inactive; adenosine, AMP, ADP, and ATP are potent vasodilators. An increase in myocardial oxygen consumption was observed during infusion of ATP and UTP with an elevation in coronary blood flow greater than that necessary to meet the increased oxygen demand. They therefore concluded that the action of these compounds is primarily on the vessels and not secondary to an increased metabolic rate.

Berne (30) demonstrated in open-chest dogs that cardiac hypoxia resulted in a decrease in coronary vascular resistance and a release of significant amounts of inosine and hypoxanthine, degradation products of adenosine, from the myocardium. He proposed an hypothesis for the metabolic regulation of coronary blood flow based on the assumption that with hypoxia the nucleotide derivatives leave the myocardial cell as adenosine. Fundamental to this hypothesis is not only the demonstration that significant intracellular adenosine formation occurs,
but that rapid extracellular diffusion of the nucleoside is possible. Richman and Uyborny (31) recovered adenosine in the venous effluent of the rabbit heart during perfusion with anoxic Krebs-Henseleit solution during inhibition of adenosine deaminase activity with 8-azaguanine. Washout studies demonstrated the rapid extracellular movement of adenosine and the total content of adenosine was uniformly greater in perfusates than in tissues. Scott et al. (19) demonstrated, in 1965, that, in the intact blood-perfused heart, transient occlusion of the left common coronary artery produced a change in the vaso-activity of coronary sinus blood of sufficient magnitude to cause resistance responses on perfusion into other organs and that these resistance responses are similar to those produced by intra-arterial injection of adenosine and AMP.

More recently, Rubio and Berne (32) demonstrated by chemical analysis that adenosine appears in coronary sinus blood during the hyperemic phase of reactive hyperemia in the blood perfused dog heart. Berne et al. (33) also demonstrated that adenosine is present in normal well-oxygenated myocardium, and the concentration increases several-fold with impairment of the myocardial oxygen supply. ATP, ADP, and AMP were not found in venous blood of ischemic cardiac or skeletal muscle.

**Neural Control**

Until recently, no unifying concept could be derived from the available literature concerning the precise role of the vagal and
cardiac sympathetic nerves in the control of coronary circulation. Much of the controversy appeared to be related to differences in preparations and methods and failure to control critical variables.

Several studies published between 1960 and 1965 demonstrated various responses to sympathomimetic amines dependent upon stimulation of alpha or beta adrenergic receptors as defined by Ahlquist (34) in 1948. Ahlquist's classification was based on the premise that two types of adrenergic receptors could be demonstrated by differing rank orders of potency within a single series of closely related sympathomimetic amines on a variety of different adrenergically controlled functions. "Alpha" was applied to those adrenergic receptors most responsive to norepinephrine and least responsive to isoproterenol and "beta" was applied to those receptors most responsive to isoproterenol and least responsive to norepinephrine. In the isolated dog heart perfused by a donor animal, Hashimoto et al. (35) obtained a coronary vasoconstrictor response with epinephrine and norepinephrine when the metabolic effect of these compounds was blocked by dichloroisoproterenol (DCI), a beta-adrenergic blocking agent, and an enhanced vasodilator response when the vasoconstrictor effect was blocked by phenoxybenzamine, an alpha-adrenergic blocker. With isoproterenol they obtained proportionate increases in coronary blood flow and myocardial oxygen consumption whereas with epinephrine and norepinephrine myocardial oxygen consumption increased to a greater extent than did coronary blood flow. Phenoxybenzamine converted this disproportionate relationship of oxygen consumption and coronary blood flow observed
with epinephrine and norepinephrine to that seen with isoproterenol. Zuberbuhler and Bohr (36) studied the responses to catecholamines in isolated helical muscle strips of large and small coronary vessels from the dog. Epinephrine and norepinephrine in concentrations well below those normally present in the blood uniformly caused relaxation of small coronary vessels. The relaxation was reversibly blocked by the beta-adrenergic blocker, pronethalol. During this blockade, catecholamines either were inactive or produced a slight contraction. On the other hand strips taken from large coronary vessels, were in some cases contracted by catecholamines; and in others, after a transient contraction, they were relaxed. Contraction was blocked by the alpha-adrenergic blocker, phenoxybenzamine. Klocke et al. (37) in open-chest anesthetized dogs observed the response to intravenous isoproterenol (0.1 to 0.3 µg/kg/min). Coronary flow and coronary sinus PO₂ always increased in spite of tachycardia and reduced or unchanged arterial pressure. To rule out vasodilation consequent to increased myocardial activity or an extracardiac factor, additional studies were performed in an isolated heart arrested with K⁺ and perfused with whole blood at constant rates of flow. Isoproterenol was given by single injections and constant infusions and always produced a decrease of perfusion pressure indicating a decrease in resistance. These decreases could be blocked by pronethalol and, as indicated by myocardial oxygen uptake and coronary venous PO₂, did not depend upon increased myocardial metabolism or decreased myocardial oxygenation. Pitt and Gregg (38) demonstrated that direct coronary artery injection of isoproterenol in unanesthetized animals resulted in a very prompt coronary
vasodilation which was attributed to stimulation of beta-receptors.

Berne et al. (39) published observations of transient decreases in coronary blood flow following stellate ganglion stimulation in anesthetized animals. Feigl (40) studied sympathetic control of the left circumflex coronary blood flow in anesthetized dogs. Coronary vasoconstriction was observed during stellate ganglion stimulation after beta-receptors were blocked with propranolol or INPEA. He was unable to demonstrate sympathetic cholinergic vasodilation in the coronary under experimental conditions which elicited sympathetic cholinergic vasodilation in skeletal muscle. Feigl (41) also demonstrated in open-chest anesthetized dogs that direct parasympathetic coronary vasodilation results from vagal stimulation and is independent of vagal chronotropic and inotropic effects.

In summary, there is substantial evidence that the coronary vascular resistance is influenced directly by both sympathetic and parasympathetic discharge. The predominant response to sympathetic impulses is dependant upon the receptor stimulated. Vasoconstriction is the result of alpha-receptor stimulation and vasodilation of beta-receptor stimulation. The response of the coronary vasculature to parasympathetic stimulation is vasodilation.

Myogenic Control

A myogenic response, independent of the central nervous system, of vessels to a stretching force was first postulated by Bayliss (42) in 1902. He proposed that the muscular coat of arteries reacts to increased arterial pressure by contraction and to decreased pressure
by relaxation; according to this concept arterial smooth muscle reacted
to stretch in a manner similar to that of smooth muscle elsewhere.
Bayliss ascribed the dilator response to the elimination of the con-
tinuous mechanical stimulation of the vascular wall by the blood
pressure. Anrep (43) later criticized the myogenic response described
by Bayliss and presented experimental evidence implicating other
factors in the vascular response to variations in perfusion pressure.

The myogenic theory, according to Berne (44), has been repeatedly
criticized because for flow to remain constant in the face of an in-
crease in perfusion pressure the vessel lumen must become smaller and
consequently the vascular smooth muscle cells must shorten. If stretch
is the stimulus which evokes the smooth muscle contraction then once
the vessel contracts to its original size, following the initial
stretch, the stimulus for further contraction is gone.

In 1949, Folkow (45) revived the myogenic response theory and
concluded from his studies that changes in intravascular tension,
rather than mechanical stretch, constitute stimuli instrumental in
maintaining and modifying vascular tone. The revived theory required
some vascular element be sensitive to tension rather than stretch.
According to La Place's relationship, (P=T/R, where P=distending
pressure, T=wall tension, and R=vessel radius) during an increase in
distending pressure, total vessel wall tension can be restored to
control levels only by a reduction in the initial radius to a value
less than that existing prior to the pressure increase. Bulbring (46)
demonstrated in the isolated smooth muscle preparation of the guinea
pig that changes in membrane potential and spike frequency induced by muscle stretch were primarily a function of the tension produced rather than the muscle length. This observation supported the thesis that vascular smooth muscle responds to changes in tension rather than length. In 1960, Folkow (47) suggested that resistance vessels respond to stretch with an increase in frequency of contractions and thus each vessel would spend a greater proportion of time in the contracted state. This hypothesis was supported by the observation of increased rate of rhythmic "vasomotion" in the vessels of the bat's wings as a response to steady distention of the vessels (48).

**Myocardial Compression**

A passive determinant of vessel radius is myocardial compression of the coronary vascular bed. In 1926, Hammouda and Kinosita (49) observed in isolated, perfused rabbit hearts that ventricular fibrillation or abolition of coordinated cardiac contractions often resulted in increased coronary blood flow. More recently, Sabiston and Gregg (50) reinvestigated the restrictive influence of cardiac contraction on left coronary inflow in the dog heart. Asystole, produced by vagal stimulation or surgically induced atrioventricular block, and ventricular fibrillation resulted in immediate flow increases of 59 and 26% respectively. These increases in inflow were associated with slightly delayed increases in coronary sinus outflow. They concluded that contraction of the heart muscle, by compression of the myocardial vascular bed, behaves as a throttling mechanism and impedes coronary blood flow. Kirk and Honig (51) estimated myocardial tissue pressure
on the basis of changes in flow through an analog of a small coronary vessel. A gradient of tissue pressure from epicardium to endocardium was observed with peak tissue pressure two times peak ventricular pressure recorded in the inner half of the wall. The theoretical gradient agreed well with the experimentally determined gradient.

Heart Rate

Heart rate has also been found to influence the rate of coronary blood flow but the mechanism by which this occurs remains in question. Since the greatest proportion of coronary blood flow occurs during diastole, during tachycardia or shortening of the diastolic period one might expect a decrease in flow. However, there is substantial evidence that coronary blood flow increases with increased heart rate. Laurent et al. (52) have demonstrated increases in coronary blood flow and myocardial oxygen consumption in association with increases in heart rate even at constant blood pressure and cardiac output. Wegrzyna et al. (53) demonstrated that atrial and ventricular tachycardia were associated with increases in coronary blood flow even when aortic pressure and cardiac output were at or below control values. In open-chest dogs, Berglund et al. (54) obtained a decrease in intravascular resistance with increased heart rates without any change in the relationship of coronary blood flow and myocardial oxygen consumption. They noted that even at very high heart rates when ventricular work was constant or decreasing, myocardial oxygen consumption and left coronary flow were both elevated and there was a marked decrease in coronary resistance. Lewis et al. (55), using the technique of estimating
extravascular resistance from the flow difference in the beating and asystolic heart at constant perfusion pressure, found that extravascular resistance increased with tachycardia (more time in systole). But this increase was more than compensated by a decrease in intravascular resistance, the net effect being an increase in coronary blood flow. Pitt and Gregg (56) studied the effects of heart rate changes in unanesthetized dogs in which electromagnetic flowmeters had previously been surgically implanted. Their results suggest that stroke systolic coronary flow is relatively well maintained over the entire range of ventricular rates studied. Although stroke diastolic coronary flow progressively decreases with increasing ventricular rate, coronary blood flow per minute increases.

Blood Viscosity

Resistance to blood flow may be due to changes in vessel radius or to changes in blood viscosity, therefore blood viscosity is an important variable influencing the rate of blood flow. Blood viscosity is largely determined by changes in red blood cell concentration, size, shape, or aggregation (57).

The coronary vascular bed actively responds to changes in its chemical environment and to nervous influences with changes in vessel radii. Changes in resistance are also brought about by hemodynamic alterations which vary the degree of myocardial compression or extravascular pressure. The variety of experimental results in studies on the effect of changes in the pH or PCO₂ on the coronary vascular resistance may be attributed to the inability to adequately control
or measure the hemodynamic, neural, and chemical influences on the vasculature in the various preparations. Even opening the chest may drastically alter the normal response of the coronary circulation. Rushmer writes: "Investigators must constantly keep in mind that when they first expose a heart of a mammal, like a dog, the heart may have already been rendered so abnormal that its function may be outside the normal range before any experimental procedure is undertaken." (58).

The methods employed in this study made it feasible to examine phasic coronary blood flow in the closed-chest animal. Monitoring of phasic coronary blood flow made it possible to calculate coronary vascular resistance late in diastole when the effects of changes in myocardial compression are unimportant. Maintaining a closed-chest animal averted those changes secondary to opening the chest, i.e. damage to the nerve supply of the heart and alterations in hemodynamic conditions affecting the heart. During these studies, the factors known to alter coronary vascular resistance or coronary blood flow were either accounted for or controlled. It is our belief that measurements of pulsatile coronary blood flow and the hemodynamic factors affecting it in the closed-chest animal during hypocapnia will shed some light on the influence of high pH and low PCO₂ on the coronary vasculature.
Preparation

Male mongrel dogs weighing 22-30 kg were anesthetized with urethane (0.50 g/kg) and chloralose (0.075 g/kg) intravenously following pre-medication with 30 mg morphine sulfate. The animals were intubated and ventilated at a fixed rate and depth with a Harvard Respiration Pump. The control gas was 95% O₂-5% CO₂ with adequate ventilation to maintain end-expiratory CO₂ at approximately 40 to 45 mm Hg. Oxygen, 100%, was used to hyperventilate the animals during the hypocapnic periods. Rapid change from the control gas to 100% O₂ was accomplished by suitably arranged solenoid valves. The rate and depth of ventilation were maintained constant throughout the control and hypocapnic periods.

In one series of experiments, a sinusoidal piston pump (59) was attached to the abdominal aorta by way of a polyethylene catheter inserted into one femoral artery. The pump was operated with a 4-5 second period for a total of 5 cycles each data collection interval. Injection and withdrawal of 40 ml. of blood with each cycle of the pump caused oscillations of the arterial volume and therefore the arterial pressure. It has been demonstrated by several investigators that operation of the pump at a frequency of 1 cycle per 4 to 5 seconds is rapid enough to prevent compensatory heart rate and blood pressure responses mediated through the carotid sinus reflexes (59,60). This
was confirmed in our laboratory.

Cardiac output was determined by the indicator-dilution method in a few experiments. Indocyanine green was injected by an automatic device into the pulmonary artery or right ventricle via the right jugular vein. The injection was always at the same time in the respiratory cycle. Blood was sampled by a constant-speed pump from the abdominal aorta via a catheter in the femoral artery. This blood passed through a densitometer quantitating the concentration of the indicator before being returned to the peripheral end of the cut artery so that the leg was continuously perfused. A cardiac output computer (61) was employed to integrate the dye concentration curves and calculate the cardiac output for each curve.

Coronary blood flow, aortic pressure, central venous pressure, electrocardiogram, end-expiratory CO₂, and arterial pH and PCO₂ were monitored during the experiments. Coronary blood flow was determined with a velocity sensitive catheter-tip flowmeter (62) inserted into the left common coronary artery or into one of its branches through the right carotid artery. The position of the flowmeter was determined by the characteristics of the flow pulse and by the hyperemic response following brief occlusion of blood flow to the coronary vessel with a sliding sheath on the flowmeter (Fig. 1). The flowmeter position was confirmed on autopsy. Zero flow was determined by occlusion with the sheath at the beginning of the experiment and, in many cases, at the end to ascertain any baseline shifts. The flowmeter was calibrated on the termination of each experiment by pushing fresh dog blood through it with the sinusoidal piston pump.
CORONARY OCCLUSION WITH HYPEREMIC RESPONSE

Fig. 1
Aortic pressure was measured with a miniaturized catheter-tip manometer (63) inserted deep into the left brachial artery. Central venous pressure was monitored in the chest with a Statham transducer. The electrocardiogram was recorded from the standard limb leads. End-expiratory CO$_2$ was monitored with an infrared gas analyzer (Beckman Model LB-1). A glass electrode was used to determine pH and a Stow-Severinghaus electrode was used to determine PCO$_2$. Both electrodes were thermostatically controlled at 37°C. Blood samples were analyzed immediately after they were withdrawn from the right brachial artery. The data were simultaneously stored on electromagnetic tape (Ampex tape recorder) and recorded on line (Grass Model 7 Polygraph).

A few animals were chemically sympathectomized with 6-hydroxydopamine (6-OH-DA; 2,4,5-trihydroxyphenethylamine) in order to determine whether the coronary blood flow response to hypocapnia is present after destruction of the adrenergic nerve terminals. Stone et al. (64) demonstrated that pretreatment of dogs with 6-OH-DA 0.62 to 5.0 mg/kg intravenously 16 to 24 hours earlier caused a reduction or blockade of the pressor effects resulting from stimulation of the central ends of the vagus nerves and those induced by injection of amphetamine. These delayed actions were associated with an ability to deplete the catecholamines from the heart. The catecholamine content of the adrenal glands is reportedly unaffected by 6-OH-DA treatment (65). We used a similar pretreatment schedule with 6-hydroxydopamine hydrobromide which was diluted immediately before injection with a 0.9% sodium chloride-0.1% sodium metabisulfite solution. The sodium metabisulfite was used to minimize oxidation.
Experimental Design

Two series of hypocapnia experiments were performed. In both series, the rate and depth of hyperventilation were constant throughout each control and experimental period. Also, arterial samples for PCO₂ and pH determinations were drawn just before gas changes and after the animals had stabilized on the 100% O₂. In four experiments, cardiac outputs were determined during several minutes before and after gas changes.

In the first series of experiments, the animals were subjected to the control gas mixture, 95% O₂-5% CO₂, for approximately 20 minutes before being switched to 100% O₂ for about 8 minutes. They were then returned to the control gas mixture for about 20 minutes. During this control period, isoproterenol (Isuprel) 0.5 µg/kg was given intravenously to determine the animal's response to this beta-active catecholamine. Figure 2 demonstrates a typical response to this dose of isoproterenol as seen in our experiments. After the animal stabilized, beta-blockade was instituted by propranolol (Inderal) 1 mg/kg intravenously. When the animal became stable following the propranolol, the same dose of isoproterenol was repeated to confirm the beta-blockade. Figure 3 demonstrates a typical absence of response to isoproterenol following beta-blockade with propranolol.

The animals in the second series of experiments were pretreated with 6-OH-DA (5 mg/kg) intravenously in 2 divided doses, one dose each of the 2 days immediately preceding the day of the experiment. In one animal, experiment 72-23, the first dose of 6-OH-DA was preceded
RESPONSE TO ISOPROTERENOL

AORTIC PRESSURE
(mm Hg)

CORONARY BLOOD FLOW
(ml/min)

CENTRAL VENOUS PRESSURE
(mm Hg)

TIME

ISOPROTERENOL INJECTION

Fig. 2
RESPONSE TO ISOPROTERENOL AFTER PROPRANOLOL

Fig. 3
by 30 minutes to 1 hour with chlorpromazine 15 mg intramuscularly. The sinusoidal piston pump was in operation for 5 cycles once during both the control and the experimental period. After the 100% O₂ period, the animals were returned to the 95% O₂-5% CO₂ gas mixture and dextro-amphetamine sulfate 0.5 mg/kg was given intravenously to test the completeness of the chemical sympathectomy; heart rate and blood pressure responses were the criteria used.

**Data Analysis**

Analog records on rectilinear paper were read at the end of the 95% O₂-5% CO₂ control period just a few seconds before the animal was switched to 100% O₂. Readings were also taken at the time of the transient increase in coronary blood flow or an average of 30 seconds into the hypocapnic period. This was followed by readings at 3 minutes, 5 minutes, and 8 minutes into the hypocapnic period and 30 seconds after the return to 95% O₂-5% CO₂. As shown in Figure 4, at each reading, a straight line was drawn through the R wave of the EKG, the aortic pressure curve, and the coronary flow curve on 4 successive pulses. Since the R wave consistently corresponded to the same points on the pressure and flow curves throughout each experiment in spite of changes in heart rate, the R wave was chosen as a convenient reference for a point late in diastole just prior to ventricular isometric contraction. Thus simultaneous flow and pressure measurements were made in late diastole for each of the four beats. The four pressure and flow measurements were averaged for calculation of late diastolic resistance by dividing pressure by flow. The pressure gradient was represented
RESISTANCE CALCULATION CURVES

AORTIC PRESSURE

CORONARY BLOOD FLOW

EKG

Fig. 4
by aortic pressure as changes in central venous pressure were considered negligible in comparison to aortic pressure. This averaging over four pulses covered the range of the oscillating changes in pressure and flow which were probably related to respiration. Averaging also increased the signal-to-noise ratio of the data and decreased the likelihood that random variability would become significant. Late diastolic resistance rather than mean diastolic resistance was utilized as an index of the caliber of the coronary vessels as the former resistance measurement minimizes the effect of changes in systolic myocardial compression. Mean diastolic resistance would include the resistance exhibited during the period of ventricular isometric relaxation when the effect of changes in myocardial compression may still be important.

The areas of the same four pulses were determined by planimetry and average minute coronary blood flow was estimated from flow calibration curves. The stroke coronary blood flow was determined by dividing the minute coronary blood flow by the heart rate at each data collection time. The heart rate was counted on the EKG tracing. The flowmeter was positioned in either the left common coronary artery or in the posterior circumflex branch. Since the blood flow in these two sites differs greatly in magnitude, it was necessary to convert all of the pressure, flow, and resistance data to percentages of control for comparison between experiments. In order to compare the changes in the heart rates of the propranolol and 6-OH-DA treated animals to the changes in the much greater heart rates of the untreated animals, heart rates were also converted to percentages of control. The mean
and standard error of the mean were calculated and plotted for these normalized data. At each data collection interval, the t-test was used to determine the significance of the difference between the mean response of the untreated and the propranolol treated animals.

Data points for late diastolic pressure and flow relationships during pump induced pressure oscillations were determined during the 95% O₂-5% CO₂ control and the 100% O₂ period. Approximately 40 pairs of pressure and flow values for each 5 cycles of the pump during each ventilation period were obtained by drawing a straight line through the R wave of the EKG, the aortic pressure curve, and the coronary flow curve. A linear regression line and r value were calculated and plotted for each set of data points.

The significance of a change in the slope was determined by the t-test when the regression line plotted for the flow-pressure relationship during 100% O₂ ventilation was compared to the line obtained during control gas ventilation.
CHAPTER II

RESULTS

The pH began to increase in all experiments within 15 to 20 seconds after switching from the control gas mixture of 95% O₂-5% CO₂ to 100% O₂. Arterial pH increased approximately 0.2 pH unit from a control average of 7.28 within the first 3 minutes of hypocapnia. The arterial PCO₂ fell from a control of 45 to 20 mm Hg or below within the same time period. The pH continued to rise to about 7.55 or above in most experiments while PCO₂ dropped to 10-18 mm Hg after 8 minutes. The end-expiratory CO₂ stabilized at approximately 2 to 3% during hypocapnia. On return to 95% O₂-5% CO₂, the pH began to drop within 15 to 20 seconds. The pH returned to 7.4 within the first minute and to 7.3 within the first 3 minutes. Within 5 minutes of the switch to 95% O₂-5% CO₂, the pH approximated the value of the previous control period.

Response to Hypocapnia before and after Propranolol Treatment

Figure 5 illustrates the response to hypocapnia in a typical untreated animal. The increases in coronary blood flow and central venous pressure were consistently observed in response to hypocapnia or ventilation with 100% O₂. The decrease in aortic pressure was also quite consistent. Figure 6 presents for comparison the response to hypocapnia in the same animal following beta-blockade with propranolol. Beta-blockade consistently eliminated or
RESPONSE TO HYPOCAPNIA IN UNTREATED ANIMAL

Fig. 5
RESPONSE TO HYPOCAPNIA AFTER PROPRANOLOL

Fig. 6
markedly reduced the increases in coronary blood flow and central
venous pressure and the decrease in aortic pressure observed during
hypocapnia in the untreated animal.

Pulse by pulse analysis of the analog data typified by Figures 5
and 6 is presented in the following sections. Raw data are presented
for the untreated animals in Table 1 and for the propranolol treated
animals in Table 2. These data were converted to percent of control
for the purpose of comparison. Results are expressed in this text as
percentage changes or mean differences from 100% of control. Means
were calculated by totaling the observed values expressed as percent
of control at each time period and dividing by the number of observa-
tions.

Resistance

The change in coronary vascular resistance in response to hypocapnia
in 8 untreated animals was compared to the resistance change during
hypocapnia in 5 of these animals after beta-blockade with propranolol.
The possibility of apparent changes in coronary vascular resistance
secondary to changes in myocardial systolic compression during hypocapnia
was ruled out by calculation of resistance to flow in late diastole.

Late diastolic resistance decreased immediately and remarkably in
the untreated animals on exposure to hyperventilation with 100% O₂.
Figure 7 shows that late diastolic resistance in the untreated animals
decreased 30% at 30 seconds into the hypocapnic period and remained at
least 23% below control throughout the 8 minutes on 100% O₂. This de-
crease in resistance was reversed by return to the control gas mixture.
<table>
<thead>
<tr>
<th>Exp. Variable</th>
<th>95% O₂ - 5% CO₂ Transient</th>
<th>Hypocapnia 3 min.</th>
<th>Hypocapnia 5 min.</th>
<th>Hypocapnia 8 min.</th>
<th>95% O₂ - 5% CO₂</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resistance (mm Hg/ml)</td>
<td>0.53</td>
<td>0.36</td>
<td>0.68</td>
<td>0.50</td>
<td>0.46</td>
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<tr>
<td>Stroke CBF (ml/pulse)</td>
<td>1.16</td>
<td>1.70</td>
<td>0.70</td>
<td>0.92</td>
<td>0.97</td>
</tr>
<tr>
<td>72-10 Min. CBF (ml/min)</td>
<td>153.</td>
<td>254.</td>
<td>110.</td>
<td>143.</td>
<td>140.</td>
</tr>
<tr>
<td>Heart Rate (beats/min)</td>
<td>132.</td>
<td>144.</td>
<td>156.</td>
<td>156.</td>
<td>144.</td>
</tr>
<tr>
<td>Late Diastolic Pressure (mm Hg)</td>
<td>116.</td>
<td>116.</td>
<td>102.</td>
<td>99.</td>
<td>98.</td>
</tr>
<tr>
<td>Resistance</td>
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<td>1.86</td>
<td>1.99</td>
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<tr>
<td>Stroke CBF</td>
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<td>0.713</td>
<td>0.318</td>
<td>0.386</td>
<td>0.402</td>
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<td>72-9 Min. CBF</td>
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<td>77.</td>
<td>42.</td>
<td>51.</td>
<td>53.</td>
</tr>
<tr>
<td>H.R.</td>
<td>108.</td>
<td>108.</td>
<td>132.</td>
<td>132.</td>
<td>132.</td>
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<tr>
<td>Late Diastolic Pressure</td>
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<td>130.</td>
<td>141.</td>
<td>140.</td>
<td>140.</td>
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<td>2.00</td>
<td>2.15</td>
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<tr>
<td>Stroke CBF</td>
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<td>0.147</td>
<td>0.145</td>
<td>0.153</td>
<td>0.153</td>
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<tr>
<td>72-15 Min. CBF</td>
<td>26.</td>
<td>27.</td>
<td>33.</td>
<td>33.</td>
<td>33.</td>
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<tr>
<td>(A) H.R.</td>
<td>180.</td>
<td>180.</td>
<td>228.</td>
<td>216.</td>
<td>216.</td>
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<tr>
<td>Late Diastolic Pressure</td>
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<td>99.</td>
<td>95.</td>
<td>93.</td>
<td>100.</td>
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<td>Resistance</td>
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<td>1.63</td>
<td>1.81</td>
<td>1.84</td>
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<tr>
<td>Stroke CBF</td>
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<td>0.191</td>
<td>0.194</td>
<td>0.154</td>
<td>0.140</td>
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<td>72-15 (B) Min. CBF</td>
<td>37.</td>
<td>39.</td>
<td>42.</td>
<td>35.</td>
<td>32.</td>
</tr>
<tr>
<td>H.R.</td>
<td>204.</td>
<td>204.</td>
<td>216.</td>
<td>228.</td>
<td>228.</td>
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<td>94.</td>
<td>84.</td>
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<td>73.</td>
<td>72.</td>
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<table>
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<tr>
<th>Exp.</th>
<th>Variable</th>
<th>95% O₂</th>
<th>5% CO₂</th>
<th>Transient</th>
<th>3 min.</th>
<th>5 min.</th>
<th>8 min.</th>
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<tr>
<td></td>
<td></td>
<td>Hypocapnia</td>
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<td></td>
<td>95% O₂</td>
<td>5% CO₂</td>
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<th></th>
<th>Resistance</th>
<th>Stroke CBF</th>
<th>95% O₂</th>
<th>5% CO₂</th>
<th>Transient</th>
<th>Hypocapnia</th>
<th>95% O₂</th>
<th>5% CO₂</th>
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<td>72-20</td>
<td>Min. CBF</td>
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<td>H.R.</td>
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<td>156.</td>
<td>144.</td>
<td>144.</td>
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<td>192.</td>
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<td>122.</td>
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<td>.32</td>
<td>.33</td>
<td>.72</td>
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<td>Stroke CBF</td>
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<td>.648</td>
<td>.694</td>
<td>.741</td>
<td>.588</td>
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<tr>
<td>72-8</td>
<td>Min. CBF</td>
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<td>110.</td>
<td>140.</td>
<td>150.</td>
<td>160.</td>
<td>120.</td>
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<td>216.</td>
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<td>204.</td>
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<td>Late Diastolic Pressure</td>
<td>106.</td>
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<td>96.</td>
<td>102.</td>
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<td>Min. CBF</td>
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<td>.253</td>
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<td>72-7</td>
<td>Min. CBF</td>
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<td>171.</td>
<td>150.</td>
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<td>Late Diastolic Pressure</td>
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<td>128.</td>
<td>98.</td>
<td>87.</td>
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### TABLE 2

RAW DATA IN PROPRANOLOL TREATED ANIMALS

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<thead>
<tr>
<th>Exp. Variable</th>
<th>95% O₂-5% CO₂ after Propranolol</th>
<th>Hypocapnia after Propranolol 5% CO₂</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Transient 3 min.</td>
<td>5 min.</td>
</tr>
<tr>
<td>Resistance (mm Hg/ml)</td>
<td>1.96</td>
<td>1.98</td>
</tr>
<tr>
<td>Stroke CBF (ml/pulse)</td>
<td>.325</td>
<td>.295</td>
</tr>
<tr>
<td>72-10 Min. CBF (ml/min)</td>
<td>39.</td>
<td>39.</td>
</tr>
<tr>
<td>Heart Rate (beats/min)</td>
<td>120.</td>
<td>132.</td>
</tr>
<tr>
<td>Late Diastolic Pressure (mm Hg)</td>
<td>112.</td>
<td>105.</td>
</tr>
<tr>
<td>Resistance</td>
<td>2.76</td>
<td>2.54</td>
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<tr>
<td>Stroke CBF</td>
<td>.556</td>
<td>.536</td>
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<tr>
<td>72-9 Min. CBF</td>
<td>40.</td>
<td>45.</td>
</tr>
<tr>
<td>H.R.</td>
<td>72.</td>
<td>84.</td>
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<td>Late Diastolic Pressure</td>
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<td>126.</td>
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<td>Resistance</td>
<td>1.82</td>
<td>1.72</td>
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<td>Stroke CBF</td>
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<td>.409</td>
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<tr>
<td>72-8 Min. CBF</td>
<td>51.</td>
<td>54.</td>
</tr>
<tr>
<td>H.R.</td>
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<td>132.</td>
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<tr>
<td>Late Diastolic Pressure</td>
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<td>Resistance</td>
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<td>Stroke CBF</td>
<td>.206</td>
<td>.200</td>
</tr>
<tr>
<td>72-3 Min. CBF</td>
<td>33.</td>
<td>32.</td>
</tr>
<tr>
<td>H.R.</td>
<td>160.</td>
<td>160.</td>
</tr>
<tr>
<td>Late Diastolic Pressure</td>
<td>241.</td>
<td>245.</td>
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</tbody>
</table>
TABLE 2 - Continued

<table>
<thead>
<tr>
<th>Exp.</th>
<th>Variable</th>
<th>95% O₂-5% CO₂ after Propranolol</th>
<th>Hypocapnia after Propranolol</th>
<th>95% O₂-5% CO₂ after Propranolol</th>
</tr>
</thead>
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<tr>
<td></td>
<td>Resistance</td>
<td>6.56</td>
<td>7.28</td>
<td>9.51</td>
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<tr>
<td></td>
<td>Stroke CBF</td>
<td>0.141</td>
<td>0.120</td>
<td>0.083</td>
</tr>
<tr>
<td>72-7</td>
<td>Min. CBF</td>
<td>13.</td>
<td>11.</td>
<td>8.</td>
</tr>
<tr>
<td></td>
<td>H.R.</td>
<td>92.</td>
<td>92.</td>
<td>96.</td>
</tr>
<tr>
<td></td>
<td>Late Diastolic Pressure</td>
<td>122.</td>
<td>120.</td>
<td>121.</td>
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</tbody>
</table>
LATE DIASTOLIC RESISTANCE

Fig. 7. Late diastolic resistance responses of the groups of dogs were significantly different from each other with P<0.05 at 30 seconds, 3, 5, and 8 minutes of 100% O₂ ventilation.
Thirty seconds after the return to 95% $O_2$-$CO_2$, the resistance had increased to a few percent above control.

In contrast to the decrease in coronary vascular resistance observed in response to hypocapnia before propranolol, there was an increase in resistance during hypocapnia after propranolol treatment. Following beta-blockade, the resistance was unchanged at 30 seconds after the switch to 100% $O_2$ and increased to 30% above control after 5 minutes of hypocapnia. At 8 minutes the resistance averaged 17% of control and, at 30 seconds after the switch to 95% $O_2$-5% $CO_2$, was 10% above control.

Pressure

Late diastolic pressure decreased during hypocapnia before propranolol treatment and increased slightly in response to hypocapnia after propranolol (Fig. 8). At 30 seconds of hypocapnia, late diastolic pressure was slightly decreased both before and after propranolol. Late diastolic pressure was 5% below control in the untreated animals after 30 seconds of 100% $O_2$ hyperventilation and 3% below control after propranolol. In the untreated animals the pressure continued to fall during hypocapnia, reaching a low of 12% below control at 5 minutes. This trend was reversed in the propranolol treated animals as diastolic pressure rose to 6% above control at 5 minutes of hypocapnia but returned to control at 8 minutes. At 30 seconds after the return to 95% $O_2$-5% $CO_2$, pressure had risen in both the untreated and the propranolol treated animals. In the former, diastolic pressure increased to just 6% below control, and in the latter, pressure increased to 10% above control.
Late diastolic pressure responses of the 2 groups of dogs were significantly different from each other with P<0.05 at 3 and 5 minutes of 100% O₂ ventilation.
Stroke Coronary Blood Flow

The presence of a transient increase in coronary blood flow in response to hypocapnia is well illustrated by the 20% increase in stroke coronary blood flow in the untreated animals at 30 seconds of hypocapnia (Fig. 9). This is in marked contrast to a 5% decrease in stroke coronary blood flow at this time after propranolol. Stroke coronary blood flow was decreased both before and after propranolol at 3, 5, and 8 minutes of hypocapnia. However, the decrease at 5 and 8 minutes in the propranolol treated animals was 25% greater than the flow decrease in the untreated animals. On return to 95% O₂-5% CO₂, the stroke coronary blood flow showed a further decrease in the untreated animals and an increase in the propranolol treated animals.

Heart Rate

Heart rates increased during hypocapnia both before and after propranolol treatment but the percentage increase was almost twice as great in the untreated animals as in the treated animals (Fig. 10). At 30 seconds into the hypocapnic period, the heart rate was increased 9% in the untreated animals and 5% after propranolol. In the untreated animals, the heart rate peaked at 32% above control at 3 minutes while after propranolol the heart rate peaked at 18% above control at 5 minutes. Thirty seconds after the return to 95% O₂-5% CO₂, the heart rate remained elevated in the untreated animals but had returned toward control after propranolol.

The greater variability in heart rate in the untreated animals as compared to the propranolol treated animals during hypocapnia is
Fig. 9. Stroke coronary blood flow responses were significantly different between the 2 groups of dogs with \( P < 0.05 \) at 30 seconds, 5, and 8 minutes of 100% \( O_2 \) ventilation.
Fig. 10. Heart rate responses were not significantly different between the 2 groups of dogs.
Minute Coronary Blood Flow

Minute coronary blood flow, the product of heart rate and stroke volume, is plotted in Figure 11 to compare left coronary blood flow during hypocapnia before and after propranolol. Figure 11 illustrates that minute coronary blood flow increased during hypocapnia in the untreated animals and decreased during hypocapnia after treatment with propranolol. Minute coronary blood flow increased 30% at 30 seconds of 100% O₂ hyperventilation while there was no change in minute coronary blood flow at this time after propranolol treatment. While minute coronary blood flow remained elevated about 12% in the untreated animals throughout the 8 minutes of hypocapnia, minute coronary blood flow gradually decreased to 25% below control during the same time in the propranolol treated animals. Thirty seconds after the return to 95% O₂-5% CO₂, the direction of the minute coronary blood flow change was reversed in both groups of animals.

Cardiac Output

Cardiac outputs were determined in 4 animals. The average increase in cardiac output during the first 3 minutes of 100% O₂ hyperventilation was 22% in the untreated animals. After propranolol treatment, the average increase in cardiac output during the same time period was 16.5%.

Response to Hypocapnia after Treatment with 6-Hydroxydopamine

A total of 6 dogs were pretreated with 6-hydroxydopamine, (6-OH-DA).
Fig. 11. Minute coronary blood flow responses were significantly different between the 2 groups of dogs with P<0.05 at 30 seconds, 5, and 8 minutes of 100% O₂ ventilation.
Two of these developed severe tachycardia and died within minutes of the first 6-OH-DA dose. One other animal was lost at the beginning of the experiment after developing severe hypotension following the single standard dose of isoproterenol. Our difficulty in maintaining the 6-OH-DA animals indicated that a lengthy series of experiments, beyond the scope of our present study, would be necessary to clearly describe the role of sympathetic innervation in the response to hypocapnia. For this reason, data from only 3 animals are presented (Table 3). Although these results are inconclusive, some trends are indicated.

All three 6-OH-DA treated dogs responded to the period of hypocapnia with an increased minute coronary blood flow and in most cases an increased stroke coronary blood flow. Heart rate also consistently increased during hypocapnia in all cases. In every animal, late diastolic pressure was decreased at least transiently and in one animal, experiment 72-23, the pressure remained below 50% of control until the return to 95% O₂-5% CO₂. Resistance decreased moderately during hypocapnia in experiment 72-16 and markedly in experiment 72-23. In spite of an increase in late diastolic resistance and little change in pressure, there was a marked increase in stroke coronary blood flow in experiment 72-18. The coronary blood flow increase in experiment 72-18 reflected a great increase in systolic rather than diastolic coronary blood flow during hypocapnia.

Two of the 6-OH-DA treated animals responded to a dose of amphetamine with increases in mean aortic pressure that were similar to the aortic pressure responses of two untreated controls. One of
## TABLE 3
RESPONSE TO HYPOCAPNIA IN 6-HYDROXYDOPAMINE PRETREATED ANIMALS

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Variable</th>
<th>95% O₂- 5% CO₂ Transient</th>
<th>HYPOCAPNIA</th>
<th>95% O₂- 5% CO₂</th>
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<tbody>
<tr>
<td></td>
<td>HR</td>
<td>78</td>
<td>84</td>
<td>92</td>
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<tr>
<td></td>
<td>P</td>
<td></td>
<td>54 56 55 55</td>
<td>65 64 63 64</td>
</tr>
<tr>
<td>72-16</td>
<td>F</td>
<td>13 10 13 13</td>
<td>16 15 14 14</td>
<td>16 17 16 17</td>
</tr>
<tr>
<td></td>
<td>R</td>
<td></td>
<td>4.55 3.57 3.88</td>
<td>4.30 3.98</td>
</tr>
<tr>
<td></td>
<td>Stroke Vol.</td>
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<td>0.155</td>
<td>0.158</td>
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<tr>
<td></td>
<td>Min. Vol.</td>
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<td>13</td>
<td>14.5</td>
</tr>
<tr>
<td></td>
<td>HR</td>
<td></td>
<td>72</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>P</td>
<td></td>
<td>85 86 87 87</td>
<td>88 86 85 86</td>
</tr>
<tr>
<td>72-18</td>
<td>F</td>
<td>15 14 15 16</td>
<td>13 6 13 10</td>
<td>15 19 19 19</td>
</tr>
<tr>
<td></td>
<td>R</td>
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<td>4.76 5.39 4.85</td>
<td>5.00</td>
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<td>0.200</td>
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<td>Min. Vol.</td>
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<tr>
<td></td>
<td>HR</td>
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</tr>
<tr>
<td></td>
<td>P</td>
<td></td>
<td>112</td>
<td>106</td>
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<tr>
<td>72-23</td>
<td>F</td>
<td>5 5 5 5 9 9 10 9 6 6 6 5 10 10 8 10 58 52 48 54</td>
<td>22 22 22 20</td>
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<td>22.40 11.48 6.65</td>
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<td>0.027</td>
</tr>
<tr>
<td></td>
<td>Min. Vol.</td>
<td>4</td>
<td>7</td>
<td>4</td>
</tr>
</tbody>
</table>

HR = heart rate in beats/min
P = late diastolic pressure in mm Hg
F = coronary blood flow in ml
R = late diastolic resistance in mm Hg/ml
Stroke Vol. = stroke coronary blood flow in ml
Min. Vol. = minute coronary blood flow in ml/min
these 6-OH-DA treated animals also responded to amphetamine with a small increase in heart rate and the other responded with a decrease. The heart rate in one control showed a marked increase in response to amphetamine while the heart rate decrease in the other control was associated with an amphetamine induced arrhythmia. Arrhythmias are not uncommon in response to amphetamine (66). One of the 6-OH-DA treated animals, experiment 72-23, responded to amphetamine with only a small increase in heart rate and a decrease in pressure. This animal probably represented the most completely chemically sympathectomized of the three 6-OH-DA treated animals. As is illustrated in Figure 12, in spite of a profound drop in late diastolic pressure in response to hypocapnia, this animal, experiment 72-23, was able to increase both minute and stroke coronary blood flow. Late diastolic resistance was tremendously decreased during hypocapnia while heart rate was increased.

Although the 6-OH-DA treated animals in our study probably represent various degrees of chemical sympathectomy, there was a marked difference in the response of the untreated animals to a dose of isoproterenol (Fig. 2) as compared to the response of all of the 6-OH-DA treated animals as typified by Figure 13. Isoproterenol in the untreated animals was followed by a drop in aortic pressure. Aortic pressure began to increase again about 20 seconds after the drop and returned to control within 1 minute. The heart rate increased and remained elevated. The coronary blood flow increased transiently at the time of the aortic pressure drop. There was little, if any, change in end-expiratory CO₂ and control venous pressure was elevated.
RESPONSE TO HYPOCAPNIA AFTER 6-HYDROXYDOPAMINE TREATMENT

Fig. 12
ISOPROTERENOL RESPONSE IN 6-OH-DA TREATED DOG

AORTIC PRESSURE
(mm Hg)

CORONARY BLOOD FLOW (ml/min)

CENTRAL VENOUS PRESSURE
(mm Hg)

ISOPROTERENOL

TIME

Fig. 13
In the 6-OH-DA treated animals, the aortic pressure dropped markedly and did not begin to increase again for 1 to 1 1/2 minutes. The pressure did not return to control levels for 2 to 2 1/2 minutes after the drop. The heart rate increased at the time of the drop, was decreased while the animal was hypotensive, and increased again while the pressure was rising. Coronary blood flow exhibited a biphasic response to the isoproterenol. The first increase was followed by a return toward control while the animal was hypotensive and a second increase occurred when the aortic pressure was increasing. The central venous pressure was elevated. In contrast to the untreated animals, the end-expiratory CO₂ rose at the beginning of the blood pressure drop and again when the pressure returned to control.

In Figures 14 and 15, calculated regression lines are plotted for the relationship between late diastolic pressure and coronary blood flow obtained with aortic pressure oscillations induced by the pump during 95% O₂-5% CO₂ control and 100% O₂ ventilation. A pair of regression lines is plotted for observations in one untreated animal (Fig. 14). Hyperventilation with 100% O₂ was associated with a decrease in the slope of the regression line or a decrease in resistance. However this change in slope was not significant on the basis of the two-tailed t-test. Two pairs of regression lines are plotted for observations in two of the 6-OH-DA treated animals (Fig. 15). In both experiments, 100% O₂ hyperventilation was associated with a decrease in the slope of the regression line or a decrease in resistance. This decrease was highly significant for experiment 72-23. Regression lines for the
CALCULATED REGRESSION LINES FOR FLOW-PRESSURE
RELATIONSHIPS DURING INDUCED PRESSURE OSCILLATIONS

\[ y = 0.502x + 39.297 \]
\[ r = 0.641 \]

\[ y = 0.400x + 46.840 \]
\[ r = 0.762 \]

Fig. 14
CALCULATED REGRESSION LINES FOR FLOW-PRESSURE RELATIONSHIPS DURING INDUCED PRESSURE OSCILLATIONS IN 6-HYDROXYDOPAMINE TREATED DOGS

\[ y = 11.402x + 45.996 \quad r = 0.749 \]

\[ y = 1.768x + 29.118 \quad r = 0.925 \]

\[ y = 2.161x + 27.772 \quad r = 0.872 \]

\[ y = 2.188x + 17.855 \quad r = 0.809 \]

Fig. 15
third 6-OH-DA treated animal, experiment 72-18, are not plotted as the correlation coefficient during the control gas ventilation was very low with $r=0.389$. These data were obtained at the end of the experiment during a third 95% $O_2$-5% $CO_2$ ventilation period when the preparation was probably deteriorating.
CHAPTER III

DISCUSSION

Our experiments show that the response to hypocapnia in untreated animals is markedly different from the response in propranolol blocked animals. The pronounced decrease in late diastolic resistance in response to hypocapnia in the untreated animals was completely reversed in the beta-blocked animals where late diastolic resistance increased during hypocapnia. In contrast to the decrease in late diastolic pressure observed in the untreated animals during hypocapnia, the pressure remained at control or slightly above in the propranolol treated animals.

Further comparison of the response to hypocapnia before and after propranolol treatment must include coronary blood flow changes. A transient 20% increase in stroke coronary blood flow was consistently observed in the untreated animals about 30 seconds after the initiation of 100% O₂ hyperventilation. In contrast, stroke coronary blood flow was decreased at 30 seconds into the hypocapnic period in the propranolol treated animals. The increase in stroke coronary blood flow at 30 seconds in the untreated animals is probably the result of a marked decrease in resistance, 31%, at this time while the pressure had dropped only 5%. The decreased stroke coronary blood flow in the propranolol treated animals at 30 seconds may be attributed to a slight decrease in late diastolic pressure with no decrease in resistance and a slight increase in heart rate.
There was a decrease in stroke coronary blood flow in both the propranolol treated and untreated animals at 3, 5, and 8 minutes of hypocapnia. But the average stroke flow was always greater in the untreated animals than in the propranolol treated animals during hypocapnia.

In the experiments we have presented, heart rate remained elevated in both the untreated and propranolol treated animals during hypocapnia but the increase was greater in the untreated animals. The difference in the percentage increase in heart rate between the two groups was not significant.

Minute coronary blood flow increased to 30% above control in the untreated animals at 30 seconds after the initiation of 100% O₂ hyperventilation. Minute coronary blood flow as the product of heart rate and stroke coronary blood flow reflects both an increase in heart rate and an increase in stroke flow at 30 seconds. In contrast to the 30% increase in minute coronary blood flow in the untreated animals, minute coronary blood flow remained at control levels 30 seconds into the hypocapnic period in the propranolol treated animals. The absence of any change in minute flow in the propranolol treated animals at 30 seconds may be attributed to the slight decrease in stroke coronary flow which is just offset by a slight increase in heart rate.

Minute coronary blood flow was elevated at least 10% in the untreated animals throughout the remainder of the hypocapnic period. A greater increase in heart rate than decrease in stroke flow was probably responsible for this elevation. The reverse trend was demonstrated in
the propranolol treated animals where minute coronary blood flow was decreased throughout the rest of the hypocapnic period. This decrease may be attributed to the gradually increasing late diastolic resistance. The increasing coronary vascular resistance is responsible for the decrease in stroke flow which is not fully compensated by an increase in heart rate.

In order to examine the change in coronary vascular resistance during hypocapnia from an entirely different perspective and over a range of pressures, pressure-flow relationships were determined during pressure oscillations induced by the sinusoidal piston pump in one untreated animal, experiment 72-20. Regression lines describing the relationship between flow and pressure were plotted. The high correlation coefficients indicate that the data fit a linear regression line. These regression lines show a decrease in slope, ΔP/ΔF or resistance, in this animal during the O\textsubscript{2} ventilation as compared to the 95\% O\textsubscript{2}-5\% CO\textsubscript{2} ventilation. Although the decrease in the slope of the regression line during hypocapnia in this one animal is not significant, this does not invalidate the observed resistance decreases calculated in a number of animals during hypocapnia.

The responses to hypocapnia seen in the untreated animals could not be blocked by chemical sympathectomy with 6-hydroxydopamine (6-OH-DA), and, in fact, were enhanced. In all three of the 6-OH-DA treated animals, both minute and stroke coronary blood flow showed a greater percentage increase during hypocapnia than did these variables in the untreated dogs. Also, the average stroke coronary blood flow for these
three dogs remained elevated throughout the hypocapnic period. This is a reversal of the decrease in stroke flow seen in the untreated animals at 3, 5, and 8 minutes. Decreased diastolic coronary resistance was observed in two of these animals and decreased systolic resistance accounted for the increase in stroke coronary blood flow in the third. Heart rate increased in all 3 animals and late diastolic pressure decreased in 2 of the 3 animals. In one of these 2 animals, the drop in pressure was very severe and prolonged. The regression lines for the 6-OH-DA treated animals showed a decreased slope, that is a decreased resistance, during hypocapnia as compared to the 95% O₂-5% CO₂ control.

The completeness of chemical sympathectomy in our experiments remains uncertain although there is substantial evidence in recent literature that 6-OH-DA treatment is capable of producing an effect similar to that of surgical sympathectomy. In 1968, Tranzer and Thoenen (67) demonstrated by electron microscopic studies that treatment of cats with 6-OH-DA produced a selective and reversible degeneration of postganglionic adrenergic nerve endings. The cholinergic nerve endings were not affected by the treatment. These results were confirmed in rats by histofluorescence studies in 1971 (65). Haeusler et al. (68) administered 6-OH-DA to cats according to a schedule which had previously been found to produce a selective and almost complete destruction of peripheral adrenergic nerve terminals. The norepinephrine content of the heart was reduced below 10% of the control value. At this time the effects of sympathetic nerve stimulation on the heart were strongly reduced. These authors concluded that when a selective destruction of
adrenergic nerve terminals is important, chemical sympathectomy with 6-OH-DA is superior to surgical sympathectomy.

Studies in dogs are not as numerous as those in other species but Stone et al. (64) were able to block the arterial pressure and heart rate increases in response to amphetamine by pretreatment of dogs with the same dose of 6-OH-DA employed in our experiments. However, with the exception of one animal, experiment 72-23, we were unable to demonstrate a marked decrease in the response to amphetamine. Nonetheless, the marked alteration in response to a dose of isoproterenol in the 6-OH-DA treated animals suggests some degree of sympathectomy.

Many factors that have been demonstrated to influence coronary vascular resistance and coronary blood flow have been discussed in the introduction to this study. These factors will now be examined for their possible role in the coronary vascular response to hypocapnia as observed in our experiments.

The rate of coronary blood flow is directly related to the perfusion pressure across the heart. If the perfusion pressure of the coronary vessels is aortic pressure minus right atrial pressure, then a decrease in right atrial pressure might increase the perfusion pressure across the coronary vascular bed in spite of an associated decrease in aortic pressure. In our experiments central venous pressure was monitored as a parallel to right atrial pressure. An increase in central venous pressure was consistently associated with hypocapnia. This eliminates an increase in perfusion pressure due to a decrease in right atrial pressure as being responsible for the increase in coronary blood flow.
Various groups (16,17) have demonstrated that elevation of blood or myocardial $K^+$ concentrations can produce coronary vasodilation. It has also been shown that a myocardial uptake of $K^+$ is associated with hypercapnia (69). Fenn et al. (70) have demonstrated that arterial $K^+$ levels increase during carbon dioxide inhalation but there is little evidence that hypocapnia is associated with $K^+$ increases in the blood or myocardium.

A decrease in myocardial oxygen tension or an increase in myocardial metabolism have been demonstrated to increase coronary blood flow (23,26). It is possible that the myocardial oxygen tension decreases even during ventilation with 100% $O_2$ if the work load of the heart increases. An increase in the cardiac output during the first 8 minutes of hypocapnia indicates that the volume work of the heart has increased and therefore myocardial metabolism might increase. An increase in metabolism would implicate an accumulation of metabolites and a decreased myocardial oxygen tension in the decrease in late diastolic coronary vascular resistance. However, cardiac output is also slightly elevated during hypocapnia following propranolol but is associated with an increased coronary vascular resistance and a decreased minute coronary blood flow. This would indicate that cardiac metabolic factors are probably not responsible for the coronary resistance and flow changes seen during hypocapnia in the untreated animals.

An increase in sympathetic outflow is associated with coronary vasodilation (21). Increased activity in the sympathetic nervous system during hypocapnia is not well supported by our experiments since
coronary vasodilation and increased coronary blood flow were observed in 6-OH-DA sympathectomized animals. Further evidence that the activity of the sympathetic nervous system is not increased in the response to hypocapnia is presented by Moster (71). Moster found that the sympathetic discharge of a post-ganglionic fiber of the celiac ganglion decreased during hypocapnia. However, this alone does not rule out the possibility of an increased sympathetic discharge in other nerves during hypocapnia.

Feigl (41) has presented evidence for coronary vasodilation in response to increased parasympathetic activity. Vasodilation due to increased parasympathetic activity would not conceivably be blocked by propranolol as was the vasodilation observed in our experiments.

The possibility of an elevation of blood catecholamines as a cause of the coronary vasodilation during hypocapnia might be ruled out by the appearance of a marked decrease in coronary vascular resistance as early as 20 seconds after the initiation of 100% O₂ ventilation.

A decrease in resistance brought about by a myogenic response to a decrease in pressure cannot account for the coronary vasodilation during hypocapnia. The decrease in resistance observed during hypocapnia in our experiments preceded the decrease in pressure and was also greater in magnitude than the decrease in pressure.

A decrease in systolic myocardial compression would be associated with an apparent decrease in coronary vascular resistance (50). In our studies, the effect of changes in systolic myocardial compression on coronary vascular resistance has been eliminated by calculation of resistance from pressure and flow values obtained late in diastole.
Changes in coronary vascular resistance may be directly related to changes in heart rate. Pitt and Gregg (56) have demonstrated, in un-anesthetized dogs in which electromagnetic flowmeters were previously implanted, that late diastolic coronary resistance decreases with increasing heart rate while stroke coronary blood flow also decreases. However, minute coronary blood flow increases since the increase in rate more than compensates for the decrease in stroke flow. Gregg (74) has stated that an increase in stroke coronary blood flow accompanying an increase in heart rate has been demonstrated under only two conditions; i.e. in thyrotoxicosis and with cardiac sympathetic nerve stimulation. The heart rate increase observed during hypocapnia is not responsible for the decreased diastolic resistance and increased minute coronary blood flow observed in the untreated animals at 30 seconds. At this time, although resistance is 30% below control and stroke coronary blood flow 20% above control, the heart rate is increased only 9%. Heart rate cannot be so easily discounted as a factor responsible for the decreased vascular resistance and increased minute coronary blood flow throughout the rest of the hypocapnic period. However, heart rate is also elevated during hypocapnia in the propranolol treated animals but the vascular resistance is increased and minute coronary blood flow is decreased. This suggests that the increase in heart rate is not the major factor involved in the decreased vascular resistance and increased minute coronary blood flow in the untreated animals during the 8 minute hypocapnic period.

A final factor which might be implicated in coronary blood flow
changes observed during hypocapnia is blood viscosity. In a few ex-
periments microhematocrits were done on arterial blood samples drawn
both before and during hypocapnia. The absence of any large change in
hematocrit implies that viscosity changes were most likely not respon-
sible for the coronary blood flow increases observed during hypocapnia.

The experiments described in this paper support the hypothesis that
the decreased coronary vascular resistance observed during hypocapnia is
a result of beta-adrenergic stimulation. In addition, this stimulation
of the beta-receptors in the heart during hypocapnia does not appear to
be the result of increased sympathetic nervous activity. These hypo-
theses were supported by the response to hypocapnia observed after
interruption of the "sympathetic nerve to beta-receptor pathway" at two
sites. With propranolol the pathway was interrupted by blockade of the
beta-receptors. Beta-blockade eliminated the decrease in coronary
vascular resistance in response to hypocapnia. 6-Hydroxydopamine in-
terrupted the pathway by destruction of the adrenergic nerve terminals.
The decrease in coronary vascular resistance in response to hypocapnia
was not eliminated or even diminished in the animal that appeared to be
the most completely sympathectomized by 6-OH-DA. In fact, pretreatment
with 6-OH-DA appeared to enhance the coronary vasodilation observed
during hypocapnia.

Further support for a beta-response to hypocapnia is the observation
that hypocapnia is associated with other known beta-responses of the
heart. These beta-responses are an increased heart rate and an increased
force of contraction as is suggested by the increased cardiac output.
Since coronary vasodilation during hypocapnia is blocked by propranolol and is not blocked by chemical sympathectomy as performed in these experiments, we propose that decreased arterial $H^+$ ion concentration induced by 100% $O_2$ hyperventilation is associated with beta-receptor stimulation. The suggestion that beta-receptors are involved in the response to hypocapnia does not eliminate the possibility of an alpha-adrenergic receptor response also. In fact, the increased coronary vascular resistance observed during hypocapnia after treatment with propranolol may be due to an unmasking of increased alpha-adrenergic activity. The only known alpha-effect in the heart is coronary vasoconstriction (75) which would be masked by activity in the beta-receptors during hypocapnia in the untreated animal. The dominance of a beta-effect in the heart during hypocapnia might be attributed to the preponderance of beta-receptors in the heart (76).

Stimulation of beta-receptors in the heart during hypocapnia could be brought about either by an increase in the level of circulating catecholamines or by an increase in the sensitivity of the beta-receptors to the catecholamines already present in the blood or myocardium. This increase in sensitivity could represent an unblocking effect of alkalosis on the receptors (77) or an alteration in some other factor peculiar to the receptor itself.

Increased levels of circulating catecholamines at 20 to 45 seconds into the hypoxic period might be eliminated as a predisposing factor in the coronary vasodilation observed at this time on the basis of the experiments of Daugherty et al. (7). These investigators observed a
transient decrease in coronary vascular resistance at 1 minute of perfusion of the coronaries in the dog with hypocapnic blood. These results are similar to our observations of a marked transient decrease in coronary vascular resistance at 20 to 45 seconds after the initiation of hypocapnia in the whole animal. However, their experiments would eliminate increased circulating catecholamines as a cause of coronary vasodilation at this time since perfusion would presumably have only local effects and not stimulate the adrenal medulla. It must be pointed out, however, that Daugherty et al. observed an increase in coronary vascular resistance after 1 minute of coronary perfusion with hypocapnic blood. Therefore, the possibility that elevated blood catecholamine levels are responsible for the sustained decrease in coronary vascular resistance seen in our experiments after 1 minute of hypocapnia cannot be excluded.

The period of time required for the onset of coronary vasodilation after the initiation of 100% O₂ hyperventilation in our experiments was as short as 20 seconds. This rapid response may eliminate the possibility that elevated blood catecholamine levels are responsible for the transient increase in stroke coronary blood flow observed at this time. However, more work in this area is necessary to completely eliminate increased blood catecholamine levels as a factor possibly involved in the decrease in coronary resistance during hypocapnia. On the other hand, the rapid response to 100% O₂ hyperventilation is consistent with a direct effect of decreased H⁺ concentration on the myocardial vasculature since arterial pH begins to increase within 20 seconds of the
Many investigators have observed a decrease in the response of the myocardium to catecholamines during acidosis and an increase in the response during alkalosis. Burget and Visscher in 1927 (78), observed an increase in the response to adrenaline in cats as the pH was elevated. The increased response was attributed either to an increased irritability of the sympathetic nervous system because of an increase in the pH of the blood, or to the possibility that as the pH of the blood rises adrenaline is oxidized more rapidly and completely and thus constitutes a stronger stimulus at the receptor site. Harry et al. (79) demonstrated that acidemia reduced the reflex increase in heart rate with stimulation of left atrial receptors and that the extent of the reduction was related to the degree of acidemia. Others have confirmed that metabolic or respiratory acidosis reduces myocardial responsiveness to exogenous catecholamines in reserpinized dogs (80) or in dogs after parasympathetic and sympathetic blockade (81). Nahas et al. (77) observed alterations of the lipolytic and calorigenic effects of norepinephrine during metabolic or respiratory acidosis and alkalosis. Decrease in arterial pH to 7.0 in their experiments inhibited the metabolic effects of norepinephrine while an increase in arterial pH to 7.55 was accompanied by a consistent increase in $V_{O_2}$, free fatty acids, and glycerol concentrations. However, further increases in these factors, when norepinephrine was administered at alkaline pH, were comparable to the increases observed at normal pH. These authors suggest that an increased $H^+$ concentration has an inhibitory effect comparable to that of beta-blocking
agents.

Darby et al. (82) were able to reverse the depression of the response to test injection of levarterenol during acidosis in dogs by correcting the acid-base changes with a H⁺ ion acceptor, THAM (2-amino-2-hydroxymethyl, 1,3-propanediol). Rovere et al. (83), in isolated strips of rat atria, demonstrated that propranolol block of the response to norepinephrine could be overcome by alkalinization of the bathing medium. In other in vitro studies, Tobian et al. (84) have shown that the contractions of isolated muscle strips, spirally cut from the rat aorta, were maximal after exposure to norepinephrine in states of relative alkalosis and minimal in states of acidosis.

On the other hand, some authors (85) have pointed out that an apparent depression in reactivity to norepinephrine during acidosis might be observed if endogenous release of catecholamines by acidosis caused a partial "saturation" of receptor sites before exogenous administration has begun; however, results of experiments in reserpinized dogs would seem to eliminate this objection.

Work in our laboratory has demonstrated that the uptake of K⁺ by the heart during acidosis is a beta-effect (86). We have also demonstrated that K⁺ uptake by the myocardium increases in the immediate post-hypercapnic period when the H⁺ concentration is rapidly decreasing (6). This K⁺ uptake has been interpreted as evidence of an unblocking of the beta-receptors as the pH increases.

The clearly demonstrated effects of beta-blockade on coronary vascular resistance definitely point to an increased beta-adrenergic
activity during hypocapnia induced alkalosis. This increased beta-adrenergic activity might logically be due either to increased sympathetic outflow, to elevated levels of circulating catecholamines, or to increased beta-receptor sensitivity. The experimental work discussed above indicates little evidence for increased sympathetic activity during hypocapnia. The role of circulating catecholamines, although an aspect deserving further study, does not appear to be of primary importance at least at the time of the early transient increase in coronary blood flow. An increase in beta-receptor sensitivity during hypocapnia remains as an attractive explanation for the increased beta-adrenergic activity at this time.
CHAPTER IV

SUMMARY AND CONCLUSIONS

The effects of hypocapnia on coronary vascular resistance were studied in the anesthetized closed-chest dog ventilated with 95% O₂-5% CO₂ during the control period and 100% O₂ during the experimental period. Left coronary blood flow was measured with a velocity sensitive catheter-tip flowmeter. Coronary vascular resistance was calculated from the ratio of aortic pressure to coronary blood flow late in diastole.

The results of these studies indicate that hypocapnia is associated with a marked decrease in late diastolic coronary vascular resistance. This decrease in resistance is reversed in response to hypocapnia after beta-adrenergic blockade with propranolol. Both the transient increase in stroke coronary blood flow approximately 30 seconds after the onset of hypocapnia and the sustained increase in minute coronary blood flow observed during hypocapnia were also reversed in response to hypocapnia after propranolol. The coronary vasodilation seen in response to hypocapnia was enhanced in the dogs chemically sympathectomized with 6-hydroxydopamine. In addition, the coronary vascular resistance determined from the slope of a regression line relating flow and pressure during pump induced arterial pressure oscillations was decreased during hypocapnia as compared to the control ventilation.

These experimental findings indicate that coronary vascular
resistance decreases during hypocapnia and that this decrease in resistance is mediated by beta-adrenergic receptors. Factors which may be involved in the beta-receptor stimulation observed during hypocapnia are increased sympathetic activity, elevated levels of circulating catecholamines, and increased sensitivity of the beta-receptors at a high pH. Although the possibility of increased sympathetic outflow or of an elevated level of circulating catecholamines as a mediator of the increased beta-receptor activity cannot be completely ruled out on the basis of our data, increased sensitivity of the beta-receptors at an alkaline pH deserves serious consideration as a possible explanation.
REFERENCES


