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ON CARDIAC MONOAMINE OXIDASE ACTIVITY.

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DISSERTATION

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

By

Stanley Grant Harris, B.S., D.V.M., M.Sc.

* * * * * *

The Ohio State University
1970

Approved by

[Signature]

Adviser
Department of Veterinary Physiology and Pharmacology
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VITA

April 2, 1936 ... Born--Wichita, Kansas

1958 .......... B.S. in Science, Kansas State University, Manhattan, Kansas

1960 .......... D.V.M., Kansas State University, Manhattan, Kansas

1960-1962 ...... Private Practice, Little Rock, Arkansas

1962-1964 ...... Chief, Veterinary Services Branch, U.S. Army Chemical Research and Development Laboratories, Edgewood Arsenal, Maryland

1964-1967 ...... Instructor, Department of Surgery and Medicine, Kansas State University, Manhattan, Kansas

1967-1969 ...... Trainee, National Institutes of Health, The Ohio State University, Columbus, Ohio

1969 .......... Research Associate, The Ohio State University, Columbus, Ohio

1970 .......... Assistant Professor, Department of Surgery and Medicine, Kansas State University, Manhattan, Kansas

PUBLICATIONS


FIELDS OF STUDY

Major Field: Comparative Cardiology

Studies in Veterinary Physiology.
Professor C. Roger Smith

Studies in Comparative Cardiology.
Professor Robert L. Hamlin

Studies in Anesthesiology.
Professor Robert Gardier

Professor Jacob E. Mosier
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INTRODUCTION

Catecholamine-induced cardiac arrhythmias during inhalation anesthesia were first shown to occur over 70 years ago when Oliver and Schafer injected adrenal extract intravenously into dogs anesthetized with chloroform. (61) Levy's investigations between 1911 and 1914 demonstrated epinephrine arrhythmia potentiation with chloroform when he produced ventricular tachycardia and ventricular fibrillation with intravenous injections of epinephrine. (41)(42)(43) Following these early studies many investigators have endeavored to show the relationship between catecholamines and inhalation anesthetics in the production of ventricular arrhythmias and to determine the mechanism by which these arrhythmias occur. (2)(10)(25)(26)(36)(64)(65) (69)(78)(80)(82) To date, no one has shown the mechanism; however, numerous reports have unequivocally demonstrated that a catecholamine-anesthetic relationship does exist, whether the catecholamine be exogenous or endogenous.

Arrhythmia is any variation from the normal rhythm of the heart. (11) Those of concern in anesthesia originate from the ventricles. Ectopic excitation or stimulus can arise from two cell types in the ventricle: the His-Purkinje
system or, more rarely, the ventricular myocardial cell. The transmembrane potential of a typical Purkinje or ventricular cell varies from -80 to -100 millivolts. These cells become excited or stimulated by receiving an impulse from another cell or when this transmembrane potential reaches threshold potential, generally -65 millivolts. (74)

When excitation occurs there is a rapid change in transmembrane potential. The cell membrane becomes permeable to sodium ions and an influx of sodium followed by an efflux of potassium ions occurs. This sudden depolarization is designated phase 0. The potassium efflux constitutes repolarization which occurs immediately following depolarization and restoration of ionic and electrical gradients. In ventricular cells, repolarization is characterized by a slight but fast phase (1) followed by a plateau (phase 2) and finally once again a rapid wave of repolarization (phase 3). The resting potential or diastolic period is designated phase 4. His-Purkinje cells have an inherent ability to depolarize spontaneously during the periods of diastole. (70) When threshold potential is reached, providing the cell has not already received an impulse from another cell, a propagated action potential will be initiated. The automaticity can establish pacemaker activity. Phase 0 is slower in the His-Purkinje cells and
the magnitude of potential change is less. Repolarization phases tend to blend into a single wave (Fig. 1). (74)

Velocity of impulse conduction differs in these two cell types. Impulse conduction in His-Purkinje cells occurs at 1.5 - 3 meters/second, whereas in ventricular cells the velocity is 0.9 meters/second. (79)

Excitability changes of the cell occur with changes in resting membrane potential or the level of threshold potential. Increasing the negativity of the resting membrane potential will increase the time required to reach threshold. Similarly, decreasing the negativity of the value of threshold potential will prolong the time required to reach threshold. The converse in each case will decrease the time required to reach the action potential propagating threshold, thereby increasing cellular "excitability."

Increased automaticity or excitability of myocardial tissue will occur with endogenous release or exogenous administration of catecholamines. (36) Receptors in the heart excited by these agents are designated $\beta$ receptors. Stimulation of these receptors will increase myocardial contractile force and/or the rate of diastolic depolarization. Catecholamines have no effect on maximum diastolic potential; however, automaticity may be so enhanced that multiple pacemakers may develop. (31)(44)(50)(78)
Fig. 1.—Transmembrane potential changes in ventricular and His-Purkinje cells during depolarization and repolarization.
Excitability will increase initially by a 10-25% decrease in threshold potential but shortly after this effect threshold potential increases to values 25-50% above normal. (30)

Norepinephrine is formed and stored in nerve endings of sympathetic fibers and its release is responsible for cardiac effects seen with sympathetic nerve stimulation. (12)(37)(60) The precursor of norepinephrine is tyrosine which, acted upon by the enzyme tyrosine hydroxylase, forms dihydropheneylalanine (Dopa). Dopa is rapidly converted to dopamine by an aromatic-amino acid decarboxylase. Dopamine $\beta$-hydroxylase rapidly converts dopamine to norepinephrine. Dopamine $\beta$-hydroxylase is found in storage granules (vessicles) within the nerve ending and these granules appear to be the site of intraneuronal norepinephrine synthesis and binding. (1)(12)

Norepinephrine can be found free in sympathetic nerve endings, in storage vesicles, or extraneuronally near nerve terminals, receptor sites, or free in the circulation. Inactivation occurs primarily by re-uptake in nerve terminals and to a lesser degree by biotransformation by the enzymes catechol-0-methyltransferase or monoamine oxidase. (12) Intraneuronal degradation of catecholamines occurs with monoamine oxidase which is found in high concentration in neuronal mitochondria.
Catechol-O-methyltransferase is believed to inactivate catecholamines extraneuronally and is responsible for inactivation of circulating catecholamines. Liver and kidney cells contain both enzymes.

Norepinephrine must first combine with specific sites on the cell membrane of the nerve terminals in order for re-uptake to occur. These have been termed "transfer," "T," or "carrier" sites. Upon entering the neuron norepinephrine is either incorporated into storage vesicles where it is protected from enzyme degradation, metabolized by intraneuronal monoamine oxidase, or remains free in the nerve terminals. Nerve stimulation will release norepinephrine from the synaptic vesicles directly into the extraneuronal spaces.

From this schema of events, several changes could be suggested as possible factors for genesis of ectopic rhythms during inhalation anesthesia. Cellular effects may be a decrease (less negative) in transmembrane potential, increased rate of diastolic depolarization, decrease in threshold potential, or membrane permeability changes. Alterations in synthesis, retention, storage, release, re-uptake, or metabolism of norepinephrine could occur. Receptor sites may respond in an abnormal manner to normal sympathetic stimulation. Finally, increased sympathetic activity may occur from central or ganglionic sites. Any one of these factors could initiate ectopic
ventricular activity. Precisely what factor(s) are involved is not known.

Early indirect evidence suggested increased central sympathetic activity may be involved. Katz studied ventricular arrhythmias in cats breathing halopropane, halothane and carbon dioxide, and halopropane plus exogenous epinephrine. His work demonstrated the complexity of central involvement in anesthetic-induced arrhythmias. Suprapontine structures were necessary for the halopropane arrhythmia, while halopropane - carbon dioxide arrhythmias required only an intact pons and medulla. The halopropane-epinephrine arrhythmia did not require any central nervous structure. (35) Stovstead found increased sympathetic nervous activity with high concentrations of ether, 8% fluoroxene and cyclopropane. Increased sympathetic activity was not affected by decerebration or baroreceptor-denervation; therefore he postulated that these agents caused sympathetic excitations by depressing medullary "depressor" neurons. (76)(77) Price, in earlier studies, had shown that, in dogs with isolated cranial circulation, halothane limited to the cranial circulation still produced cardiovascular depressant changes shown in the intact animal. (63) His conclusion at that time was that para-sympathetic activity was increased, whereas sympathetic activity was inhibited. This may be true in very deep
halothane anesthesia. Sympathetic activity is increased in light halothane anesthesia. Gardier reported that arrhythmias do occur with greater frequency in light than in deep halothane anesthesia, whereas in other inhalation anesthetics the converse is true. (17) In more recent work, Price has shown that the cardiovascular effects seen with halothane result from \( \beta \)-adrenergic stimulation. (64)

Sympathetic stimulation can occur by reflex mechanisms responding to hypoxia or hypercarbia. Incidence of arrhythmias under these conditions has been shown to increase markedly. (34)(47)(64) Incidence of anesthetic arrhythmias will occur with increased frequency by increased heart rate (55), blood pressure (5), and stretch of myocardial fibers, (29)(36) but none of these factors are absolutely essential. (36)(57)

Release of potassium from the liver has been suggested as a cause of epinephrine or cyclopropane-epinephrine arrhythmias. Dresel, however, found that by excluding the liver from the circulation and eliminating the hyperkalemic response, arrhythmias with cyclopropane and epinephrine will still occur. He concluded that releasing potassium from the liver may facilitate production of arrhythmias but it is not essential. (9) By measuring intracellular potassium levels Gutgeseld found
that epinephrine produced a biphasic response. Initially there was a loss of myocardial potassium, which was followed by an uptake of the ion. Similar effects could be accomplished by increasing heart rate. Cyclopropane appeared to inhibit the net uptake of potassium. (24) High levels of serum potassium have been shown to produce cardiac arrhythmias but levels encountered are much higher than seen with anesthesia or anesthesia and epinephrine.

Results on effects of inhalation anesthetics on ganglionic transmission have varied. Larrabee, by perfusing stellate ganglia with various inhalation anesthetics, found ganglionic transmission to be blocked. (40) Garfield studied the effects of pre- and post-stellate ganglionic sympathetic nerve stimulation on heart rate in cats while under ether, halothane, cyclopropane, and nitrous oxide anesthesia. With ether anesthesia, right ganglionic sympathetic nerve stimulation produced no change in heart rate response, whereas pre-ganglionic stimulation produced depressed results. The same depression to pre-ganglionic stimulation occurred with cyclopropane and halothane; however, with these agents, post-ganglionic responses to nerve stimulation were potentiated. (19) Li measured post-ganglionic potentials resulting from pre-ganglionic nerve stimulation and found that the magnitude of the post-ganglionic potential was the same or possibly even increased
with halothane anesthesia. (45) Conclusions, therefore, are that inhalation anesthetics have negligible effect on stellate ganglion transmission, certainly not sufficient to excite or depress ventricular arrhythmias.

Electrophysiology of the myocardial cell and catecholamine activity have been investigated in order to determine their role in arrhythmia production with inhalation anesthetics. Han was able to show that sympathomimetic amines decrease the action potential propagation threshold briefly, then increase the threshold in the cardiac cells. (28) Sympathetic nerve stimulation decreased the threshold potential. Both increase automaticity in autonomic cells. Katy reported that threshold potentials or maximum diastolic potentials were not changed by sympathomimetic amines, although they did increase rate of spontaneous diastolic depolarization in autonomic cells. (36) Levy determined that cyclopropane had no effect on the resting membrane potential of ventricular cells or on the action potential amplitude. (44) Smith reported that neither cyclopropane nor halothane had any effect on diastolic threshold, conduction time or refractory period. (78) Davis reported that cyclopropane had no (effect on) transmembrane potentials but significantly increased the rate of repolarization during phase 2 of the ventricular muscle cell while delaying repolarization
during phase 3. (6) The over-all effect was to shorten the time to reach a transmembrane potential of -60 while the total duration to reach maximum resting potential was prolonged. Trautwein felt that the ectopic beats seen with catecholamines probably resulted from increased automaticity in distal parts of the conduction system. (79) A favorable condition would exist by inhibition of sinoatrial nodal activity, such as seen with increased vagal activity resulting from increased systemic arterial pressure. The probability for the occurrence of ectopic beats will be larger if greater time is allowed for their development.

Moore used microelectrode techniques and recorded from atrial muscle cells, atrioventricular nodal cells, His bundle cells, and ventricular cells. He found that cardiac arrhythmias resulting from catecholamines occurred below the His bundle electrode and theorized that the ectopic beats were due to increased pacemaker activity in the ventricular conduction system. (56)

Smith and Wallace felt that slow conduction velocity coupled with unidirectional block in ventricular cells under the influence of catecholamines and inhalation anesthetics allowed re-entry of ventricular impulses from areas of slow conduction and block, thereby exciting the arrhythmia. (78)(83)

Ngai, Naito and Gillis, and Greene have shown that cyclopropane and halothane had no effect on synthesis,
re-uptake, or release of catecholamines from the myocardial cell. (22)(23)(58)(59)(60)

Naito and Gillis, utilizing isolated cat atrial strips, found no change in norepinephrine rebinding after sympathetic nerve stimulation. (58)

Gardier et al. demonstrated increase in total plasma catecholamine levels during cyclopropane anesthesia; however, levels of norepinephrine were increased. (18) Li found that myocardial tissue levels of epinephrine decreased with ether, cyclopropane, and halothane anesthesia but myocardial tissue levels of norepinephrine increased. (46)

Work to date suggests that ventricular arrhythmias with inhalation anesthesia occur at the myocardial level and are influenced by an increased concentration of normal adrenergic mediator (Norepinephrine). It is likely that nonuniform distribution of nerve fibers exists throughout the myocardium which undoubtedly could account for uneven distribution of norepinephrine and, through increased concentrations of norepinephrine in susceptible areas, this mediator could account for increased excitability or automaticity.

Gardier and Hamelberg have shown that high levels of cyclopropane in vitro will inhibit catecholamine biotransformation by catecholmethyltransferase. (16) Gardier further suggests that increased titers of plasma
norepinephrine during cyclopropane anesthesia might possibly be a function of decreased amine metabolism. (18) Neither catechol methyltransferase or monoamine oxidase has been studied for its effects in vivo with clinical levels of concentration.

Purposes of this study were to determine normal response to cardiac sympathetic nerve stimulation, find a laboratory model in which increased arrhythmia production can be seen with inhalation anesthesia, and to determine the effect of common inhalation anesthetics on myocardial monoamine oxidase activity.
CHAPTER I.

CARDIOVASCULAR EFFECTS OF RIGHT AND LEFT PRE- AND
POST-GANGLIONIC SYMPATHETIC NERVE
STIMULATION IN DOGS AND CATS

Introduction

Cardiac acceleration and augmentation are known to be mediated through $\beta$-adrenergic receptors in the heart. (74) Exogenous or endogenous catecholamines can combine with these receptors to elicit response. Increased sympathetic activity will increase heart rate (acceleration), cardiac inotropy (augmentation) through neuronally released norepinephrine at the sympathetic nerve terminals.

Augmentor and accelerator fibers are intermingled throughout their course and cannot be functionally separated at any point in their anatomical pathways. One mediator, norepinephrine, is released at the sympathetic nerve terminals. The specific response is apparently dependent upon the receptor site activated.

McKibben and Getty were able to follow the course of sympathetic fibers in the dog and cat. They found
that left cardiac sympathetic nerves passed primarily to the caudal and right surfaces of the left cardiac chambers, and that right cardiac nerves ramified on the right chambers and left portion of the left chambers. Left cardiac nerves were more prominent around the coronary sinus and atrioventricular node, while the right ones contributed more to the area of the sino-atrial node. (48)(49)

Mizeres, Furnival, and Randall and McNally have shown in dogs that stimulation of right cardiac sympathetic nerves has marked positive chronotropic response and minimal positive inotropic response, whereas stimulation of the left nerves has a greater positive inotropic and little if any positive chronotropic response. (15)(54)(66)(67)(68)

Katz found that halopropane plus 10% carbon dioxide produced ventricular arrhythmias in 99% of the animals studied and that removal of the pons and midbrain blocked the arrhythmic response. Drugs and other procedures to interrupt efferent sympathetic pathways also prevented the arrhythmia. However, catecholamine anesthetic arrhythmia can be produced in decerebrate, spinal, and reserpinized animals and in animals with denervated hearts and in heart lung preparations. (35) Therefore, more than efferent sympathetic activity must contribute to arrhythmia production.
Presynaptic and postsynaptic sympathetic activity has been shown to increase in animals under cyclopropane and ether anesthesia. (51)(52)(53)

The purpose of the study is to show cardiac response to right and left pre- and post-ganglionic cardiac sympathetic nerve stimulation in the dog and cat and the effect of a myocardial "sensitizing" inhalant anesthetic on cardiac rhythm during sympathetic nerve stimulation in these species.

Materials and methods

Four healthy dogs and 16 healthy cats were used in the study. Each animal served as its own control. Anesthesia was achieved by intravenous injection of sodium pentobarbital until loss of palpebral reflex. Intubation was accomplished by tying an endotracheal tube securely in the tracheal lumen. The tracheal tube was attached to a Rochester Model Heibrink inhalation anesthesia machine. Ventilation was maintained with a Mark 2 Bird\(^1\) respirator.

The femoral artery was cannulated and attached to a Statham P 23 AA transducer for pressure recording. The left common carotid artery was cannulated and the catheter passed retrograde into the left ventricle by direct fluoroscopic examination or pressure monitoring.

\(^1\)Bird Corporation, Palm Springs, California.
Left ventricular pressure was recorded through a Statham\textsuperscript{1} P 23 AA transducer and the first derivative ($dp/dt$ max.) of the pressure curve recorded on a separate channel. Lead II of the electrocardiogram was monitored with the above parameters on a Brush\textsuperscript{2} or Grass\textsuperscript{3} direct writing oscillograph (Fig. 2).

Ventral midline thoracotomy was performed and the internal thoracic arteries were doubly ligated and cut. Right and left stellate ganglia were located. Preganglionic sympathetic nerves were isolated one centimeter proximal and transected two centimeters proximal to the stellate ganglia. Cranial and caudal trunks of the ansa subclavia to the stellate ganglia were isolated (Fig. 3).

Right and left pre- and post-ganglionic sympathetic nerves were stimulated with a square wave impulse from a Grass Model S4 stimulator through an isolation amplifier. Impulse voltage was varied from 1-10 volts, duration from 2-10 milliseconds and frequency from 2-30 Hertz (Hz).

Following stimulation of cardiac sympathetic nerves the animals were allowed to breathe a gaseous mixture of

\textsuperscript{1}Statham Transducer, Inc., Hato Rey, Puerto Rico.

\textsuperscript{2}Brush Instrument Division, Clevite Corporation, Cleveland, Ohio.

\textsuperscript{3}Grass, Instruments, Quincy, Massachusetts.
Fig. 2.—Direct writing oscillograph used for recording electrocardiograms, arterial pressure and myocardial contractility.
Fig. 3.—Completed preparation of cat for sympathetic nerve stimulation with bipolar stimulating electrodes placed around left pre-ganglionic sympathetic fibers.
0\textsubscript{2} and 2% halothane. Various magnitude and frequency square wave impulses were again delivered to the isolated nerves and cardiac response recorded.

**Results**

Results of left and right pre- and post-ganglionic nerve stimulation in dogs and cats are shown in Tables 1 and 2. Variations in impulse duration had little effect on response when kept in the range from 4-10 milliseconds. Maximal response was seen at 16 Hz., and frequencies greater than 16 Hz. had little if any effect in increasing cardiac rate, inotropy or arrhythmia production. Increasing the impulse to 4 volts increased the intensity of response. Exceeding 4 volts increased the magnitude of sympathetic response although the slope of increase diminished rapidly.

Stimulation of right cardiac sympathetic nerves increased heart rate (positive chronotropy) with minimal increase in inotropy, whereas stimulation of left cardiac sympathetic nerves had a greater positive inotropic response with minimal positive chronotropic response (Fig. 4).

Addition of 2% halothane to the inhalation gas failed to cause arrhythmia during sympathetic nerve stimulation in dogs, whereas 13 of 16 cats stimulated during halothane administration demonstrated ventricular arrhythmias (Table 3). Arrhythmias produced in cats originated from the ventricle ipsilateral to nerve stimulated.
TABLE 1. Dogs demonstrating greater than 10% increase in heart rate (HR), left ventricular contractility (dp/dt max), and aortic pressure (A\(\sigma\)) during cardiac sympathetic nerve stimulation. Four dogs were tested.

<table>
<thead>
<tr>
<th>Sympathetic Nerve</th>
<th>HR*</th>
<th>dp/dt max</th>
<th>A(\sigma)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Left pre-ganglionic</td>
<td>2</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Left post-ganglionic</td>
<td>2</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Right pre-ganglionic</td>
<td>4</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Right post-ganglionic</td>
<td>4</td>
<td>2</td>
<td>2</td>
</tr>
</tbody>
</table>

*One dog had negative chronotropic response to left sympathetic nerve stimulation.

TABLE 2. Cats demonstrating greater than 10% increase in heart rate (HR), left ventricular contractility (dp/dt max), and aortic pressure (A\(\sigma\)) during cardiac sympathetic nerve stimulation. Sixteen cats were tested.

<table>
<thead>
<tr>
<th>Sympathetic Nerve</th>
<th>HR</th>
<th>dp/dt max</th>
<th>A(\sigma)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Left pre-ganglionic</td>
<td>6</td>
<td>14</td>
<td>13</td>
</tr>
<tr>
<td>Left post-ganglionic</td>
<td>7</td>
<td>15</td>
<td>13</td>
</tr>
<tr>
<td>Right pre-ganglionic</td>
<td>15</td>
<td>9</td>
<td>10</td>
</tr>
<tr>
<td>Right post-ganglionic</td>
<td>15</td>
<td>8</td>
<td>8</td>
</tr>
</tbody>
</table>
Fig. 4.—Results of right and left cardiac sympathetic nerve stimulation in a dog. Heart rate was 90 beats per minute during the control period, 120 beats per minute during right sympathetic nerve stimulation, and 92 beats per minute during left sympathetic nerve stimulation.
TABLE 3. Arrhythmias produced by sympathetic nerve stimulation in dogs and cats before and after halothane in the inhaled gas.

<table>
<thead>
<tr>
<th></th>
<th>Number of Trials</th>
<th>Arrhythmias Before Halothane</th>
<th>Arrhythmias With Halothane</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dogs</td>
<td>4</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Cats</td>
<td>16</td>
<td>0</td>
<td>13</td>
</tr>
</tbody>
</table>

Addition of halothane to the inspired air decreased aortic pressure and left ventricular contractility in every trial. Stimulation of sympathetic nerves had positive inotropic and chronotropic responses; however, the response in most trials reached only control levels (Fig. 5).

Discussion

Randall and McNally, in a limited number of surgical patients, found no difference in response to right and left nerve stimulation in man. (67) The present study confirms observations of Furnival et al. and Mizeres in dogs and shows additionally that cats respond with a greater positive inotropic response to left cardiac sympathetic nerve stimulation. Right cardiac nerve stimulation produces a greater positive chronotropic response. (13)(14)(54) Right sympathetic nerves to the heart, therefore, contain more fibers distributed to the SA node, whereas left sympathetic nerves contain more fibers distributed to ventricular muscle.
Fig. 5.—Arterial pressure, left ventricular pressure, electrocardiogram, and $dp/dt$ max (ventricular contractility) resulting from left cardiac nerve stimulation before and after addition of 3% methoxyflurane to the gaseous inhalation mixture.
Control  Stimulation  Control + 3% Methoxyflurane  Stimulation + 3% Methoxyflurane

Arterial Pressure

Left Ventricular Pressure

Lead I

Lead II

dp/dt
Addition of halothane to inspired air did not result in cardiac arrhythmias during sympathetic nerve stimulation in dogs. Thirteen of 16 cats tested did demonstrate ventricular arrhythmias under the influence of this halogenated gas. Arrhythmias in cats resulting from inhalation anesthesia cannot be attributed to simple sympathetic nerve stimulation as suggested by Black et al. Sympathetic nerve stimulation was supramaximal in each case, yet no arrhythmias were shown before halothane was administered. The fact that dogs failed to elicit ventricular arrhythmias augments the hypothesis that myocardial tissue in this species is not "sensitized" to the effect of halogenated hydrocarbons to the degree seen in cats.

In cats arrhythmias resulting from sympathetic nerve stimulation were limited to the ipsilateral ventricle. Right sympathetic fibers, therefore, are not limited to the area of the sinoatrial node and atrial musculature but must have augmentor elements in the right ventricle. Greater augmentor response to left sympathetic nerve stimulation and elicitation of left ventricular arrhythmias serve to confirm a functional course of these fibers on the left ventricular myocardium.

Conclusions

Stimulation of the right cardiac sympathetic nerves in dogs and cats result in a greater positive
chronotropic response with minimal positive inotropy, whereas stimulation of the left cardiac sympathetic nerves in these species results in greater positive inotropy.

Addition of halothane to the inhalant gas has an arrhythmogenic effect during sympathetic nerve stimulation in cats but not in dogs.
Fig. 6.—Ectopic ventricular beat or ventricular tachycardia resulting from right and left cardiac sympathetic nerve stimulation.
Ectopic Ventricular Beats

Ventricular Tachycardia

Stimulation
Right Cardiac Sympathetic Nerve

1 sec.

Stimulation
Left Cardiac Sympathetic Nerve

1 sec.
Fig. 7.—Systemic arterial pressure drop resulting from addition of 3% methoxyflurane to the gaseous inhalation mixture.
CHAPTER II

NON-UNIFORMITY OF POSITIVE BATHMOTROPIC EFFECTS OF RIGHT AND LEFT SYMPATHETIC NERVE STIMULATION

Introduction

During the past 10 years different investigations have shown that, in dogs, stimulation of the right cardiac sympathetic nerves produced a positive chronotropic response with minimal positive inotropy. Stimulation of left cardiac sympathetic nerves produced a greater positive inotropic response and little positive chronotropy. (54)(68) Pace et al. demonstrated that right ventricular contractility increased more with left cardiac sympathetic nerve stimulation than right cardiac nerve stimulation in dogs. (62)

McKibben and Getty followed cardiac nerves in feline and canine species and found that left cardiac sympathetic nerves passed primarily to caudal and right surfaces of the left cardiac chambers, while right cardiac sympathetic nerves ramified on the right chambers and left portion of the left chambers.

The purpose was to investigate the effects of pre- and post-ganglionic right and left sympathetic nerve
stimulation during the administration of the anesthetic with myocardial "sensitizing" properties. Indirectly, the results could contribute to an understanding concerning the way the right and left nerves are distributed to the cardiac tissues.

**Materials and methods**

Ten cats were anesthetized with sodium pentobarbital. A midline skin incision from the xyphoid cartilage to the larynx was made. The trachea was transsected and a tracheal cannula tied securely in place. The tracheal cannula was attached to a Rochester Model Helbrink\(^1\) inhalation anesthesia machine and the cats allowed to breathe a gaseous mixture of \(O_2\) and 2% halothane through a Flutec\(^2\) Vaporizer. Ventilation was maintained by a Bird\(^3\) positive pressure ventilator.

Lead II electrocardiogram, aortic pressure and left intraventricular pressure with its first derivative were monitored on a direct writing oscillograph.

A midline thorocotomy was made. Left and right internal thoracic arteries were located, double ligated and cut between the ligatures.

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\(^1\)Ohio Chemical Company.

\(^2\)Fraser Sweatman Inc., Hatfield, Pennsylvania.

\(^3\)Bird Corporation, Palm Springs, California.
Left and right stellate ganglia were located and pre-ganglionic fibers were isolated a distance one centimeter proximal to the ganglia. Ansa subclavia from right and left stellate ganglia were isolated a distance 1 centimeter from the ganglia. The nerve for stimulation was placed over a bipolar electrode and stimulated with a square wave impulse from a Grass\textsuperscript{1} model S4 stimulator through an isolation unit. Stimulation voltage, duration, and frequency were carried from 2-10 volts, 3-10 milliseconds and 2-16 Hz at each stimulation site. At the termination of the study ectopic ventricular beats were produced by pricking the ventricular myocardium at various sites with a sharp instrument.

Results

Ectopic beats of ventricular origin were demonstrated in 7 of 10 cats studied. Stimulation of right pre-ganglionic and post-ganglionic nerves produced multiple ectopic ventricular beats originating from the right ventricle (Fig. 8). A supramaximal stimulus applied to the fibers produced right ventricular tachycardia (Fig. 9). Stimulation of left pre-ganglionic and post-ganglionic cardiac sympathetic nerves produced multiple

\textsuperscript{1}Grass Instrument Company, Quincy, Massachusetts.
Fig. 8.—Right ventricular ectopic beats resulting from stimulation of right pre-ganglionic sympathetic nerves. Four volts at 8 Hertz.
Fig. 9. — Right ventricular tachycardia resulting from stimulation of right post-ganglionic sympathetic nerves. Eight volts at 16 Hertz.
left ventricular contractions (Fig. 10). Supramaximal stimulation produced left ventricular ectopic beats followed by left ventricular tachycardia (Fig. 11).

Mechanical stimulation of right and left ventricles resulted in ectopic beats with electrocardiographic complexes that appeared on electrocardiogram to be identical to those created by ipsilateral cardiac sympathetic nerve stimulation (Figs. 12 and 13).

Lead II electrocardiograms had an RS pattern in 7 cats whereas the complexes appeared qR in 3.

Left ventricular dp/dt max increased upon stimulation of both right and left sympathetic nerves but the increase was greater with left nerve stimulation. Positive chronotropy appeared with both right and left sympathetic nerve stimulation; however, the response was greater with stimulation of the right nerve.

Single or multiple ectopic beats were elicited and were dependent upon the degree of nerve stimulation. Supramaximal stimulation excited all sympathetic fibers and produced ipsilateral ventricular tachycardia, whereas submaximal stimulation elicited only ectopic beats. Greater positive inotropy from left sympathetic nerve stimulation suggests the possibility that more of these fibers terminate in the left ventricle.
Fig. 10.—Left ventricular ectopic beats resulting from stimulation of left pre-ganglionic sympathetic nerves. Eight volts at 8 Hertz.
Fig. 11.—Left ventricular ectopic beats and tachycardia resulting from stimulation of left post-ganglionic sympathetic nerve stimulation. Eight volts at 16 Hertz.
Fig. 12.—(a) Ectopic ventricular beats and ventricular tachycardia resulting from ipsilateral sympathetic nerve stimulation. (b) Mechanically induced ectopic beats produced by pricking the left and right ventricular apex and mid-ventricular myocardium.
Fig. 13.—(a) Stimulus and mechanical ectopic ventricular beats resulting from right cardiac sympathetic nerve stimulation or pricking the right ventricular myocardium. (b) Stimulus and mechanical ectopic ventricular beats resulting from left cardiac sympathetic nerve stimulation.
Mechanically stimulated ectopic beats had almost identical configuration to those of sympathetic nerve stimulation, confirming the area in which ectopic beats were provoked during nerve stimulation.

Discussion

Stimulating cardiac sympathetic nerves in dogs, Mizeres found that right cardiac sympathetic nerves went primarily to the right atrial wall and musculature and left sympathetic fibers went to the left ventricular myocardium. This produced a greater positive chronotropic response with right cardiac sympathetic nerve stimulation and greater positive ventricular inotropic response with left sympathetic nerve stimulation. (54) Randall and McNally found that these differences did not exist in man. (65)

Pace et al., measuring right ventricular $dp/dt_{max}$, demonstrated in cats that left sympathetic nerve stimulation produced a greater positive inotropic response in the right ventricle than right cardiac sympathetic nerve stimulation. This may reflect only the greater positive inotropic response of the left ventricular contraction and its effect on right ventricular ejection, or it may reflect increased right ventricular contractility. (62)

McKibben and Getty pointed out that the course of right and left sympathetic fibers ended in the main
portion of their respective ventricles. (48)(49)
Norepinephrine has been shown to induce ventricular ectopic beats and is the neurotransmitter at terminal ends of cardiac sympathetic nerves. Concentration of available hormone would be greatest in areas of greatest nerve concentration. The fact that right ventricular premature beats occurred from right sympathetic nerve stimulation indicated that the peripheral course of the right cardiac sympathetic nerves to the ventricles is primarily to right ventricular myocardium in addition to sinoauricular nodal area and right atrium. Left ventricular ectopic beats from left cardiac sympathetic nerve stimulation suggests that the course of these fibers to the ventricles primarily is to the left ventricular musculature as well as atrioventricular nodal areas.

Conclusion

Stimulation of the right and left cardiac sympathetic nerves shows increased excitability (positive bathmotrophy) in the ipsilateral ventricle, supporting the hypothesis that right cardiac sympathetic nerves innervate the right ventricular myocardium and left cardiac sympathetic nerves innervate the left ventricular myocardium.
CHAPTER III

MYOCARDIAL MONOAMINE OXIDASE ACTIVITY DURING INHALATION ANESTHESIA IN CATS

Introduction

Various inhalant anesthetic agents cause a depression in monoamine oxidase (MAO) activity in vitro. (72) Diaz et al. by in vivo methods were unable to show, from measurements of serotonin levels, any change in MAO activity in brain following cyclopropane, halothane or diethyl ether anesthesia. (8) In a series of patients anesthetized with diethyl ether and compared to a series of patients with halothane, Schweizer et al. found significantly higher levels of lactate formation with diethyl ether anesthesia, suggesting a depressant effect on electron transport or possibly a decreased transfer of reducing agents across the mitochondrial membrane. (73) Gardier et al. found that cyclopropane anesthesia in the dog reduced the total concentration of circulating catecholamines, but increased the plasma norepinephrine (NE). (16) Li et al. found a significant increase in NE levels in cardiovascular tissue during diethyl ether and cyclopropane anesthesia, but a significant decrease in
epinephrine following diethyl ether, cyclopropane or halothane. (46) Goldberg and Ullrick demonstrated a direct effect of halothane on the isolated myocardium and postulated that this agent appeared to affect the "active state intensity" (contractility) of the myocardium. (20) Davis and Horita suggested that MAO may be the limiting factor in the termination of the pharmacologic responses to exogenous catecholamines and to indirectly acting sympathomimetic amines. (7)

The effects of various agents on the activity of MAO in the myocardium are described.

Methods

Animal studies.— In preliminary investigations, it was found that the cat, but not the dog, developed cardiac arrhythmias during sympathetic nerve stimulation following inhalation anesthesia. The present study was divided into three parts: (1) a control group which received only oxygen, (2) four groups which received different inhalant anesthetics and (3) two groups which received MAO inhibitors. Drugs in part 3 were administered by a slow intravenous drip so the 10 mg/kg of either JB-516 (Catron)\(^1\) or tranylcypromine\(^2\) was given during the

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\(^1\)Lakeside Laboratories, Milwaukee, Wisconsin.

25-minute anesthesia phase of the experiment. Six animals were in each group. The anesthetics used with oxygen were: halothane (1%), methoxyflurane (1%), diethyl ether (3%) and cyclopropane (30%) initially, followed in 3 minutes by 10% for the remainder of the anesthesia.

Cats were weighed and given Pentobarbital sodium at an approximate dose of 30 mg/kg to the stage of surgical anesthesia. Tracheotomy was performed and a cannula approximating the inside diameter of the trachea tied securely in place. The cannula was attached to the "Y" piece of the breathing tubes on a Rochester model Heibrink gas anesthesia machine and the animal allowed to breathe 100% oxygen. Respirations were maintained at the rate of 9/minute by an Emerson positive pressure ventilator. The left common carotid artery was isolated and a polyethylene cannula (2 mm diameter) was passed down the common carotid artery to the root of the brachiocephalic artery. The cannula was connected to a Statham P23Db pressure transducer.

A midline incision was made through the skin and the sternum split along its midline. Both right and left internal thoracic arteries were double ligated and transected between the ligatures. Pre-ganglionic sympathetic fibers, 1 cm proximal to the left stellate ganglion, were isolated and placed on bipolar stimulating electrodes.
Electrocardiogram was monitored via leads I and II recorded simultaneously. All recording was done on a Grass Model 7 polygraph.

Exactly one hour after the induction of anesthesia with pentobarbital, the left pre-ganglionic nerve was stimulated for 1 minute with a square wave impulse of 4 volts and 3 msec. duration, 4 Hz. Exactly 4 minutes later, the stimulation was repeated at 8 Hz., and similarly at 16 Hz. In earlier studies, no increase in response was noted using greater voltages or higher frequencies, which concurs with the findings of Vassalle et al. (81) Four minutes after the control tests, animals in parts 1 and 3 continued breathing oxygen while either saline (part 1) or MAO inhibitor (part 3) were infused over a 25-minute period. Animals in part 2 received a saline infusion, but were given an inhalant anesthetic over this 25-minute period. The separate groups of animals in part 2 received halothane and diethyl ether through a Dragor vaporizer; methoxyflurane was administered through a calibrated #8 Heibrink ether vaporizer. Cyclopropane was administered directly.

Five minutes after the cessation of anesthesia or drug infusion, the left pre-ganglionic nerve was again stimulated as previously. Four minutes after the final electrical stimulation, the animal was euthitized by removal of the heart.
Approximately 500 mg sections of the atria and the ventricles of the heart were immediately taken, weighed and quickly frozen in liquid nitrogen for determination of MAO activity.

Biochemical studies

Frozen heart tissues were minced and crudely homogenized in chilled isotonic KCl by means of a "Duall" homogenizer. The activity of the MAO was determined on these homogenates using a modified Kramel method. (39) The reaction mixture was as follows: 1.0 ml of tissue homogenate in isotonic KCl, 0.5 ml kynuramine (100 µg kynuramine dihydrobromide), 0.5 ml phosphate buffer (0.5 M, pH 7.4) and water to 3.0 ml. Incubation was carried out at 37°C for 30 minutes in unstoppered glass tubes. The addition of perchloric acid (10%) rather than trichloroacetic acid to stop the reaction allowed an increase in sensitivity of approximately five-fold due to the decreased quenching effect of perchloric acid in this fluorometric assay. The reaction product, 4-hydroxyquinoline (4-HOQ), was activated at 315 µm and exhibited an intense fluorescence at 380 µm. The 4-HOQ was determined spectrofluorophotometrically in an Aminco-Bowman spectrofluorophotometer with off-axis elliptical optics. A clear

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1 Kontes Glass Company, Vineland, New Jersey.
plexiglass square rod was used to adjust the instrument for each assay. Reference curves were constructed from data obtained using isolated rat liver mitochondrial outer membrane MAO. (71)

Results

The results of the myocardial MAO activity measurements after treatment with inhalant anesthesia are indicated in Table 4. Student's T-test for paired data was employed for statistical analysis of groups of animals given anesthetics and compared to control hearts. Halothane and methoxyflurane anesthesia caused a significant (p < .01) increase in MAO activity in the atrial tissues. While ventricular tissue demonstrated an increased activity, the results are not as exceptional as the atria (p < .05). Diethyl ether also caused an increased MAO activity in atrial and ventricular tissues (p < .025; p < .10). Cyclopropane anesthesia caused a very significant increase in all samples taken from the heart (p < .0005). Both MAO inhibitors, JB-516 or tranylcypromine, reduced the levels of MAO activity to nearly negligible values from control (p < .0005).

Ventricular arrhythmias occurred in 2 of 42 cats before treatment and 17 of 42 cats following treatment with an anesthetic agent or monoamine oxidase inhibitor. None of the arrhythmias occurred in control cats (part 1). In the two cats demonstrating pre-treatment arrhythmias, the incidence of ectopic beats increased following treatment.
TABLE 4. Monoamine oxidase activity (millimicromoles 4-hydroxyquinoline formed in 30 minutes per gram of tissue) of myocardial tissue homogenates in cats after general inhalation anesthesia

<table>
<thead>
<tr>
<th>Agent</th>
<th>Tissue</th>
<th>X</th>
<th>S.E.M.</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Halothane</td>
<td>RA</td>
<td>68.14</td>
<td>10.96</td>
<td>.01</td>
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<tr>
<td></td>
<td>LV</td>
<td>27.14</td>
<td>1.84</td>
<td>.10</td>
</tr>
<tr>
<td></td>
<td>LA</td>
<td>54.12</td>
<td>3.94</td>
<td>.0025</td>
</tr>
<tr>
<td></td>
<td>LV</td>
<td>32.34</td>
<td>4.26</td>
<td>.0125</td>
</tr>
<tr>
<td>Cyclopropane</td>
<td>RA</td>
<td>93.7</td>
<td>4.83</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>RV</td>
<td>50.73</td>
<td>2.72</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>LA</td>
<td>119.07</td>
<td>5.40</td>
<td>.0005</td>
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<tr>
<td></td>
<td>LV</td>
<td>55.72</td>
<td>2.37</td>
<td>.0005</td>
</tr>
<tr>
<td>Ether</td>
<td>RA</td>
<td>49.3</td>
<td>4.52</td>
<td>.025</td>
</tr>
<tr>
<td></td>
<td>RV</td>
<td>24.37</td>
<td>1.09</td>
<td>N.S.</td>
</tr>
<tr>
<td></td>
<td>LA</td>
<td>48.37</td>
<td>3.72</td>
<td>.005</td>
</tr>
<tr>
<td></td>
<td>LV</td>
<td>22.62</td>
<td>1.66</td>
<td>N.S.</td>
</tr>
<tr>
<td>Methoxyflurane</td>
<td>RA</td>
<td>49.73</td>
<td>5.67</td>
<td>.05</td>
</tr>
<tr>
<td></td>
<td>RV</td>
<td>25.55</td>
<td>2.07</td>
<td>N.S.</td>
</tr>
<tr>
<td></td>
<td>LA</td>
<td>47.33</td>
<td>6.96</td>
<td>.05</td>
</tr>
<tr>
<td></td>
<td>LV</td>
<td>26.12</td>
<td>1.99</td>
<td>.025</td>
</tr>
<tr>
<td>JB-516</td>
<td>RA</td>
<td>5.88</td>
<td>1.08</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>RV</td>
<td>4.53</td>
<td>0.39</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>LA</td>
<td>3.94</td>
<td>0.55</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>LV</td>
<td>5.07</td>
<td>0.65</td>
<td>.0005</td>
</tr>
<tr>
<td>Tranylcypromine</td>
<td>RA</td>
<td>4.85</td>
<td>0.58</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>RV</td>
<td>2.88</td>
<td>0.47</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>LA</td>
<td>4.27</td>
<td>0.74</td>
<td>.0005</td>
</tr>
<tr>
<td></td>
<td>LV</td>
<td>2.61</td>
<td>0.30</td>
<td>.0005</td>
</tr>
<tr>
<td>Controls</td>
<td>RA</td>
<td>34.97</td>
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<td>LA</td>
<td>30.98</td>
<td>3.89</td>
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</tr>
<tr>
<td></td>
<td>LV</td>
<td>19.79</td>
<td>2.05</td>
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</table>
Ventricular arrhythmias occurred during nerve stimulation following treatment as shown in Table 5. Both positive and negative chronotropic responses were observed during nerve stimulation after treatment. Negative chronotropic response occurred in those cats receiving monoamine oxidase inhibitors. Nerve stimulation increased both mean aortic pressure and pulse pressure pre- and post-treatment. Aortic pressure recorded just before either nerve stimulation did not change in part 1 (Fig. 14a). In part 2, the aortic pressure always decreased post-treatment from the pre-treatment values (Fig. 14b). Animals in part 3 showed an increase in aortic pressure before post-treatment nerve stimulation from pre-treatment values (Fig. 14c).

Discussion

These studies provide biochemical evidence for MAO concentration changes in the myocardium during anesthesia with representative agents employed for general anesthesia clinically. By stimulation of the pre-ganglionic sympathetic fibers proximal to the stellate ganglion we attempted to induce arrhythmias in both experimental and control groups of cats. Pilot studies of animals sacrificed for normal values of MAO activity showed no change from control animals receiving pentobarbital.

In all groups from part 2 (inhalant anesthetics), we observed increases in MAO activity. Cyclopropane
TABLE 5. Post-treatment induced ventricular arrhythmias by left cardiac sympathetic nerve stimulation

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. Observed/6 Cat Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Control</td>
<td>0</td>
</tr>
<tr>
<td>2. Volatile Anesthetics</td>
<td></td>
</tr>
<tr>
<td>a. Cyclopropane</td>
<td>4</td>
</tr>
<tr>
<td>b. Ether</td>
<td>1</td>
</tr>
<tr>
<td>c. Halothane</td>
<td>4</td>
</tr>
<tr>
<td>d. Methoxyflurane</td>
<td>2</td>
</tr>
<tr>
<td>3. MAO I</td>
<td></td>
</tr>
<tr>
<td>a. JB-516</td>
<td>4</td>
</tr>
<tr>
<td>b. Tranylcypromine</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>17/42</td>
</tr>
</tbody>
</table>

Anesthesia demonstrated the greatest increase in myocardial MAO activity compared with other drugs tested. Arrhythmias are known to occur with an increased frequency with cyclopropane. (12) Arrhythmias, including ventricular fibrillation, occurred with minimal nerve stimulation. Halothane, methoxyflurane and diethyl ether all increase the myocardial MAO activity to a lesser degree than cyclopropane, and arrhythmia production required a greater nerve stimulation. Aortic pressure and myocardial contractility decreased when
Fig. 14.—(a) Electrocardiogram and aortic pressure change resulting from left pre-ganglionic cardiac sympathetic nerve stimulation before, during, and following saline infusion. (b) Electrocardiogram and aortic pressure change resulting from left pre-ganglionic cardiac sympathetic nerve stimulation before, during, and following addition of 3% halothane to the inspired air. (c) Electrocardiogram and aortic pressure change resulting from left pre-ganglionic nerve stimulation before, during, and following infusion of monoamine oxidase inhibitor.
<table>
<thead>
<tr>
<th>PRE-TREATMENT</th>
<th>TREATMENT</th>
<th>POST-TREATMENT</th>
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<tbody>
<tr>
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<td></td>
<td></td>
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<tr>
<td>A</td>
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<td>B</td>
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</tbody>
</table>
animals were given the inhalant anesthetics, which is consistent with previously described results. (3)(63)(75)

All animals given MAO inhibitors (part 3), administered acutely, had severely depressed myocardial MAO activity (p < .0005). It can be concluded from this information that adequate amounts of the MAO inhibitors were administered.

MAO inhibitors increased systolic and diastolic blood pressure. Slow intravenous infusion of the drug elicited ventricular arrhythmias suggesting catecholamine release or failure of inactivation as described by Goldberg and Shideman (21) and Kopin and Axelrod. (38) Maintenance of high blood pressure and positive inotropy during the course of the experiment gave further evidence of prolonged norepinephrine activity.

Cyclopropane anesthesia demonstrated the greatest increase in myocardial MAO activity over other drugs tested. Arrhythmias are known to occur with an increased frequency with this agent. (81) Arrhythmias, including ventricular fibrillation, occurred with minimal nerve stimulation in cats receiving this cyclopropane.

Halothane, methoxyflurane and diethyl ether all increase myocardial MAO to a lesser degree than cyclopropane, and arrhythmia production required greater nerve stimulation. Aortic pressure and myocardial contractility decreased when animals were given the inhalant anesthetics,
which is consistent with previously described results. (3)

(26)(34)(63)(82)

We have shown a decreased MAO activity with anesthetic agents using isolated rat liver mitochondrial outer membrane MAO in previous in vitro studies. (72) The data in this study indicate that the MAO activity of cardiac tissue in situ will respond in an opposite manner to these previous studies.

Increased levels of NE in myocardial tissue and plasma during anesthesia have been shown. (18) Diaz et al. concluded that cyclopropane, halothane and diethyl ether had no effect on rat brain MAO in vivo, but an increased brain serotonin turnover was observed with diethyl ether. All of these agents caused increases in 5-hydroxyindole acetic acid in the brain, indicating an increased MAO activity. (8) Several authors have reported no observable change in synthesis, release or re-uptake of NE during anesthesia. (12)(59)(60)

The circumstances necessary for arrhythmia production may be dependent on both an enzyme inhibition and an increased activity present on the biologic membranes as a consequence of the anesthetic agent. Finally, the ability of inhalant anesthetic to penetrate certain tissues or microstructures to a variable degree may be responsible for the cardiac effects of these agents.
ADDENDUM
Following completion of this thesis a recent article by Reuter, H. and Beeler, G. W. (Science 163, 399-401, January 24, 1969) was brought to my attention. They show an influx of Ca\(^{++}\) ion into the cell during depolarization following the rapid Na\(^{+}\) influx. This Ca\(^{++}\) influx results in the plateau of phase 2 of the action potential of ventricular cells. Since Ca\(^{++}\) is essential for excitation-contraction coupling perhaps the increased augmentation with catecholamines results in membrane permeability danger to Ca\(^{++}\) with increases in intracellular Ca\(^{++}\) during contraction.
BIBLIOGRAPHY


14. Furnival, C. M., Linden, R. J., and Snow, H. M.:  
"Chronotropic and Inotropic Effects of Isoproterenol and L. Nor-Adrenaline in the Dog."  

15. Furnival, C. M., Linden, R. J., and Snow, H. M.:  
"Response to Stimulation of the Cardiac Sympathetic Nerves."  

16. Gardier, Robert W., Endahl, Gerald L., and Hamelberg, William:  
"Cyclopropane: Effect on Catecholamine Biotransformation."  

17. Gardier, Robert W., and Hamelberg, William:  
"Effect of Anesthetics on Catecholamine Biotransformation."  

18. Gardier, Robert W., Reier, Charles E., Traber, Daniel L., Rowe, Howard M., and Hamelberg, William:  
"Elevated Plasma Norepinephrine during Cyclopropane Anesthesia as a Possible Function of Decreased Amine Metabolism."  


