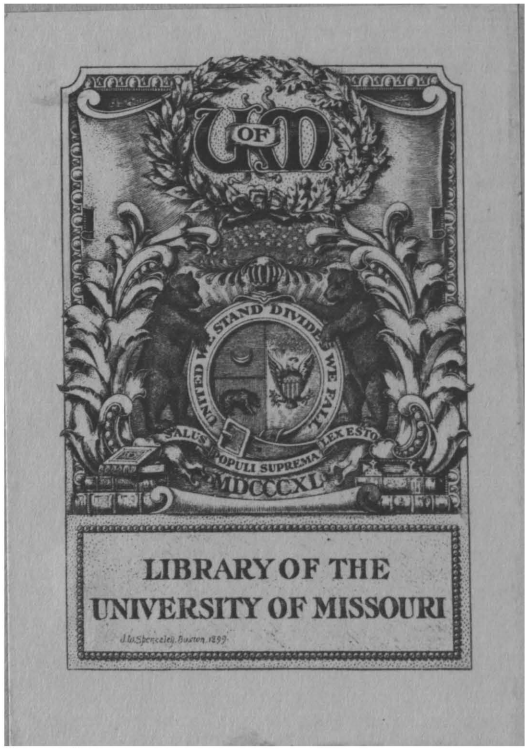

THE EFFECT OF DIGITALIS ON THE CARDIAC PERIPHERAL NEURO-MUSCULAR COMPLEX

by

Lloyd Reuben Boutwell, A. B.

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF ARTS

in the

GRADUATE SCHOOL

of the

UNIVERSITY OF MISSOURI

1914

From the Laboratory of Physiology,

DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY,

University of Missouri.

TABLE OF CONTENTS.

THE EFFECT OF DIGITALIS ON THE CARDIAC PERIPHERAL NEURO-MUSCULAR COMPLEX.

I. Introductory and Historical.

The question of the action on the animal organism of the principles found in the leaves and flowers of Digitalis purpurea has long been one of absorbing interest. A glance at the long list of articles dealing with researches on the subject will indicate to some extent the detail with which this question has been considered. It may not seem obvious that there is any necessity for investigating this subject again. However, as long as there are details of the action of the drug which are not well understood, such investigation is pertinent. It is with the view of making more clear the nature of some of the details that the present paper is presented.

From a historical standpoint, digitalis is one of the oldest known drugs in the pharmacopeia today. The use of the drug dates back long before the middle of the eighteenth century. It was used by the laity as a household remedy sometime before its introduction into

therapeutics. In this connection it was employed chiefly in the treatment of dropsy. The formal introduction of digitalis into therapeutics may be considered to have occurred upon the publication of an article in the year 1875 by W. Withering[55], an English physician. This paper "An Account of the Properties of Foxglove" was based on careful and accurate observations upon the application of digitalis in the treatment of disease. The chief action noticed in the use of the drug clinically was the slowing of the pulse. The advance of the knowledge of the drug was furthered the following year by Schiemann [46], whose observations were made chiefly upon dogs and cats. This work in point of time might be considered as the initiation of a new method as an aid in the solution of the problems of digitalis action. The important observation Schiemann made was that in cats and dogs the pulse was slowed as in man. The first trial to determine the influence of digitalis on the frog occurred at the last of the eighteenth century by an English author, Johnson [26], published in "Medical Essays and Observations" in 1795. Johnson did not, however, observe the effect upon the heart. In 1799 John Ferriar[18], contributed to the literature of digitalis making special reference to the diuretic power of the drug. He also observed the slowing of the pulse and considered that this effect was the primary

action rather than that on the kidney. His observations were all from clinical cases. The knowledge of the subject was extended at the beginning of the eighteenth century successively by Beddoes[5] ; Kinglake [29], and Mossman [41] . Beddoes, 1801, came to the conclusion that the strength of the circulation was increased by a rise in blood pressure. Kinglake, 1801, described the action of digitalis as increasing the activity of the heart. The effect he attributed to an increase in the contraction of the heart both as regards the systole and diastole whereby even though the pulse was slowed more blood was forced into the arteries than under normal conditions. Mossman, 1812, ascribed the entire effect to the slowing of the pulse and the effect on the arteries. Up to the year 1839 the investigations had been with few exceptions, notably that of Schiemann, the results of observations in the clinical experience of practitioners. The first really definite quantitative investigation of the effect of digitalis on the circulation was made by Blake [4] , 1839, who by the aid of a haemodynamometer was able to demonstrate a rise in blood pressure in dogs upon the injection of digitalis infusion. He suggested that the rise in blood pressure was at least in part due to constriction of the capillaries. The next work of note from an experimental standpoint was that of Traube [50] , who first gave the theory of

vagus stimulation for the explanation of the most important clinical observation, the pulse slowing. He came to this conclusion from the results of his experiments on vagotomized dogs. He also observed the rise of blood pressure during the slowing of the heart rate. Briquet [7], and Lenz [34], both working during the year 1853, confirmed the results which Traube had reported. In 1861 Traube added to his former work further experiments in confirmation of his theories. He had previously noted that in large doses digitalis paralyzed the vagus and that the heart thus released from vagus control increased much beyond the previous rate. He suggested that the action was to be explained upon the basis that both the inhibitory and motor nervous mechanism of the heart were first stimulated then paralyzed. Both effects he considered as being manifested first on the inhibitory mechanism followed later by a similar action on the motor apparatus.

From the time of Traube until 1874 the investigation of the influence of digitalis was carried on by a number of men, chief among them being Winogradoff[54], Marme [36], Brunton [8], Ackermann [1], Boehm [6], Gorz [22], and A. B. Meyer [38]. A review of the literature of this period was published by Gorz in 1873. In 1874 Schmiedeberg [43] made extensive investigations into the chemical nature of digitalis, paying particular attention to the isolation of active principles and the determina-

tion of their chemical and pharmacological characteristics. Previous to the time of this work investigations had been carried on with the infusion of the leaves of Digitalis purpurea. Homolle and Quevenne had, however, previously isolated a non-crystallizing substance from digitalis leaves. Also Nativelle had obtained a crystalline product. The work of Schmiedeberg, which has become classic has conclusively shown that the ordinary infusion of digitalis leaves or extracts of the seeds contains a number of substances differing somewhat in their chemical and physical properties and also in their effects upon the animal organism. Schmiedeberg's important constituents are the following:

1. Digitonin, $C_{31}H_{53}O_{17}$ an amorphous substance resembling Saponin.
2. Digitalin, $(C_5H_8O_2)_n$ an amorphous glucoside, insoluble in water.
3. Digitalein, $C_{25}H_{40}O_{25}$ a crystalline glucoside easily soluble in water.
4. Digitoxin, $C_{21}H_{33}O_7$ a crystalline product insoluble in water.

Each of these substances Schmiedeberg was able to break into two or more resinous bodies, some of which were pharmacologically active and some not. Further work of Schmiedeberg was upon the classification of the extracts of plants which by their pharmacological action can be identified with the digitalis group.

In consideration of the experimental work which had been done up to this time on the action of the various substances which are considered as belonging to this group Schmiedeberg reaches the conclusion that the pharmacological action in each case is essentially similar in nature though not always so in intensity.

In 1882 Schmiedeberg published an article in which he reviewed the classification of the substances of the digitalis group together with a rather extensive consideration of the application of digitalis to the treatment of disease.

From the time of Schmeideberg's papers till the beginning of the present century the greater part of the investigation upon the subject has been upon the various members of the group with a view of determining whether or not they conform in their effects with that of digitalis. Near the end of this period there appeared an article by Cushny[13] representing a long series of mammaliam experiments investigating the exact nature of the movement of the heart under the influence of the digitalis substances.

In the last decade the literature on the subject of digitalis has been extensive and varied. Cloetta[11] extended the work of Schmiedeberg in regard to the pure principles of digitalis. Different phases of the subject of the action of digitalis on the circulatory

system have been taken up by various investigators. Gottlieb and Magnus[21], 1902, have been concerned with the effects on the blood-vascular system. Kloptowsky[30], 1903, investigated the histological changes observable in various tissues after digitalis poisoning. Etienne[17], and also Kochmann[32], 1906, have considered the question of the exact effect of the drug upon the vagus inhibitory apparatus of the heart. Edmunds[16], 1907, studied the influence of digitalis upon the rate of the blood stream. Digitalis poisoning, its cumulative action and the question of tolerance have been subjects of researches by v. Lhota[51], 1911-1913.

The approach to the clinical side of the question of digitalis action has been made very inportant through the careful observations and researches of Mackenzie[35], on clinical cases, 1911.

II. The Effects of Digitalis upon Various Tissues and Systems of the Animal Organism as Determined by Experiments to Date.

Looked at from the experimental standpoint the effects of digitalis can be considered as being limited almost exclusively to the circulatory system. The effects in other regions are for the most part insignificant by comparison and may be considered as in more or less close relation with the effects on the circulatory apparatus. However, mention of the effect of this drug upon some of the other tissues may well be included here.

The continued administration of digitalis in man is attended usually by nausea and vomiting. This effect according to Bastedo[2] is to be ascribed to its effect upon the vomiting center, for it occurs not only upon administration by mouth but when the drug is given hypodermically or by the rectum. Upon the respiratory movements digitalis is usually observed to have no effect though after toxic doses in animals the respiratory movements are increased.

The early use of digitalis was confined chiefly to the removal of accumulations of fluids from the body.

The question has long been in dispute whether this effect is confined only to disease conditions or could be produced in the normal animal. At present it is considered that digitalis diuresis is produced on the normal animal, although this as compared to the effects produced in cases of dropsy are small. As to whether the effect is one of specific kidney action or is merely secondary to the improvement in the circulation which the administration brings about is still a matter of question. There is some discussion as to whether the effect of digitalis on the kidney vessels is one of constriction on dilatation. Marshall[37], and Gottlieb and Magnus[20], reported a constriction of the kidney under the influence of digitalis. It has been conceived that the production of diuresis depends wholly upon the increase in the blood flow through the kidney due to the increase in arterial tension. Jonescu and Loewi[27], working with Digalen and Strophanthin have found that in cases where there is no rise in arterial pressure diuresis occurs. They obtained in their experiments with these drugs a dilation of the kidney. This they considered to be due to direct action of these drugs on the vessel walls since such dilatation occurred when the nervous connection of the kidney was completely cut off. The question then remains open.

The most important effect manifested by digitalis is that upon the circulatory system.

In the frog digitalis produces in small doses a slowing of the heart rate with an increase both toward systole and diastole. Finally the heart stops in systole as first demonstrated by Schmiedeberg. Williams[53], showed that this increased excursion of the heart resulted in a greater outflow at each beat, and that this produces an increase in blood pressure has been shown by the same author using a system of rigid tubes. In mammals in the laboratory dosage digitalis usually produces a rather sudden rise in blood pressure accompanied by slowing of the pulse. This, according to Cushny[13], is to be considered as the therapeutic state of the action of the drug. The second stage he designates as beginning with acceleration in the ventricle and is characterized by this and the periodic changes in the strength of contractions and irregularity of rate. When the irregularity in the strength of contraction becomes sufficiently marked the inefficiency of the heart allows a fall in blood pressure. The mammalian heart according to Meyer and Gottlieb[39], is usually found at post-mortem in a state of diastole a result which they ascribe to the inability of the heart to maintain its activity in the face of a failing coronary circulation a sufficient time for the drug to have the marked systolic effect on the cardiac muscle.

The stages of digitalis action seen in experiments on the mammalian circulation, Cushny interprets as dependent upon two factors. In the first stage the stimulating effect on the vagus center is most in evidence though stimulation of the muscle is shown in the increase of systolic contraction. The beginning of acceleration marks the beginning of the domination of the stimulating effect on the muscle. That the vagus still has an influence in determining the action of the heart can be shown in the diastolic relaxation produced by its stimulation and by the diastolic pauses which occur at this stage. It will be seen, however, that any and all variations must be referable to two factors which control the normal condition of the circulation, the blood vessels on the one hand controlling the size of the bed through which the blood stream flows and the heart on the other governing the amount of fluid introduced into the vessel system and the pressure under which it is introduced. It is upon these two factors that digitalis of necessity acts in producing its effects on the circulatory system.

Beyond the slowing of the pulse which was first noted by Withering and later by Schiemann, no other action of digitalis on the circulatory system was observed until Blake in 1839 reported a rise in arterial tension upon the injection of an infusion into the cir-

culation. This observation was confirmed by Lenz and Briquet and later in 1861 by Traube. Since that time rise in blood pressure has been accepted as the usual effect of digitalis. The problem before investigators has since been to determine the cause of this rise in blood pressure, and assign to each factor its role in the production of this phenomenon.

Blake suggested that the rise in pressure was at least in part due to constriction of the vessels. This factor was overlooked by Lenz, Briquet and Traube, but was later revived by Marme and supported by the work of Brunton,[9] Brunton and Meyer[9], Ackermann, Brunton and Tunnicliffe,[10] Gottlieb and Magnus, Marshall and others. Although there has been considerable opposition to this view by Schmiedeberg, Boehm, Gorz, Williams and Koppe[33], the mass of accumulated data shows that at least in the laboratory dosage digitalis produces for the most part constriction of the peripheral blood vessels. Investigations as to the regions most affected in this stimulation have been carried out by Gottlieb and Magnus using the plethysmographic method of recording the changes in the various organs and regions. They concluded that in small doses the result was a general constriction but upon administration of large doses the greater power of contraction of the splanchnic vessels may result in a dilation of the limbs to accommo-

date the blood excluded from the splanchnic area. With the question of the vasomotor effect of digitalis practically settled, the problem naturally arises as to what part of the vasomotor mechanism is responsible for this change. Traube and Boehm were the first to consider this matter. Their first observation was that the rise in pressure was eliminated by cutting the cord but later Boehm observed that this was not true. He found that there was little variation to be noted upon section of the cord. The work of most of the early investigators as Ackermann, Marshall, Kobert, Gottlieb and Magnus support this view. The weight of evidence seems to be in favor of very little if any influence of the vasoconstrictor center in producing the narrowing of the arterial bed.

The two other factors of the vasomotor complex upon which digitalis may act in producing vasoconstriction are the endings of the vasomotor nerves in the vessel walls and the smooth muscle which makes up the greater part of the wall. Just to what extent each of these factors are affected, experimental work up to the present time does not fully indicate. Kobert[31], and Thomson[49], working on isolated organs were unable to demonstrate a decrease in the outflow from the vein when digitalis was perfused. Kobert reasoned from the fact that the organs used were decomposing the nerve elements, had degenerated and the

effect therefore was only upon the muscle. Donaldson and Stevens[15], working on coldblooded animals reached the probable conclusion that the effect of arteriole constriction was due to the influence on the muscular walls. Bastedo stated that the peripheral effect can be analyzed by the use of apocodeine and ergotoxine which paralyze the vasoconstrictor nerve endings. Vasoconstriction is still obtained after the nerve endings are paralyzed showing that the effect is at least in part upon the muscle. He also says that since digitalis produces vasoconstriction in the pulmonary and coronary arteries we may consider the effect purely on the muscle since vasomotor nerves to these structures have not been domonstrated. It seems then definitely proven that the point of greatest effect of digitalis in producing vasoconstriction is arterial muscle. It may still be regarded as an open question whether any of the effect can be ascribed to the endings of the vasomotor nerves.

The other important factor in varying the conditions of the circulation is the heart. Fundamentally there are two elements which must be considered in a discussion of the effect of any drug on the cardiac mechanism, i.e., nerve and muscle. The former of these may be observed from two viewpoints, (1) the nerve centers in the medulla and (2) the nerve elements in the heart itself. The first evidence of stimulation of the vagus center by

digitalis was given by Traube in 1851 who found that the slowing was removed by cutting the vagus in the neck. His observation also included the fact of paralysis of the vagus by large doses and escape of the heart from this control and consequent marked increase in rate. Traube's explanation for the phenomenon of increased blood pressure in connection with the slowing of the pulse was that both the inhibitory and motor mechanism of the heart was first stimulated and then paralyzed and that the inhibitory factor was acted upon first in each case. After the work of Traube the question of the stimulation of the inhibitory nerve center in the medulla was not seriously disputed. The question was raised by Meyer whether this stimulation of the inhibitory center was not due to the increase in pressure which digitalis produces. This explanation has not found favor and has been practically disproved by Kaufmann[28], and Cushny, who have found numbers of cases in which great slowing occurred accompanied by very low blood pressure. The idea of stimulation of the vagus center by the increase of pressure has been recently revived by Kochmann who contends for a part of the stimulation as a slight stimulating effect upon the vagus due to pressure in those cases in which suitable doses produce a rise in pressure. Along with the view of central action of the digitalis substances have been attempts to determine

whether the inhibitory mechanism in the heart plays any part in the production of slowing. There is still considerable doubt about this phase of digitalis action. Traube observed that the slowing of the heart was not entirely removed by section of the vagi. He therefore concludes that the intracardiac inhibitory nerves are stimulated. This observation has been confirmed by others. Ackermann found that the slowing was completely removed by atropine. The discussion has again risen upon the statement of Etienne that the vagus endings of the heart of the dog are not stimulated and that the slowing which is produced by strong hypertoxic doses on the vagotomized dog is not to be ascribed to the effect on the nerve endings but to the effect on the heart itself.

The commonly observed condition of increased rate in the later stages of digitalis was explained by Traube as excitation of the motor apparatus of the heart. Later work by Ackermann showed that this acceleration could not be produced by the paralysis of the vagus alone because such acceleration occurred even when the vagus had been eliminated by atropin. There is no doubt, however, that the vagus has lost control at this stage since it shows no control of the heart upon electrical stimulation. Various attempts have been made to explain this acceleration on the basis of excitation of the accelerator mechanism but experimental proof is not sufficient to support

this view. Cushny from experiments on the non-gangliated apex of the frog's heart which showed an increased excitability in digitalis solution and also from the tendency of digitalis to remove the inhibitory standstill of muscarin is inclined to the view that the important factor of the acceleration of the heart in later stages is the increased irritability of the heart muscle. He stated that the factors of inhibitory action and increasing muscular irritability are in action throughout all stages of the effects of digitalis, but that in the first stage the inhibitory effect is able to keep the other factor in check, and that it is upon the paralysis of the vagus than the muscle factor is made manifest in the alteration of the rate of the heart.

The nature of the action of the digitalis on the heart muscle is not altogether clearly worked out, although the details of the changes which occur in the muscle have been carefully studied by Schmiedeberg. He proposed the theory that the effect upon the heart muscle was primarily one of an increase of elasticity. His work was upon the heart of the frog and his conclusions are based largely upon his observation that the heart of the frog which has come to systolic standstill by the action of digitalis can be made to beat again if the elasticity is overcome by mechanical distention of the ventricle. Roy[42], working primarily on the question of

the elasticity of the heart, came to the conclusion that the effect due to digitalis varied with the strength of the dose. He found that by the use of small doses, at a given pressure the extent to which the ventricle would relax in diastole was increased while with larger dosage the relaxation was much less complete and that a correspondingly high degree of pressure was necessary for producing distension. He concludes then that the elasticity change of the muscle depends somewhat upon the dose or the amount of the drug to which the muscle has been subjected. Cushny, working upon the mammalian heart, gives extensive evidence of the movements which occur in the heart muscle by the action of digitalis. The change that occurs in the early stage of the cycle of the action of the drug is referable to the activity of the inhibitory mechanism opposing the direct effect on the heart muscle. The systolic contraction is always increased, the extent of diastolic relaxation may be increased, remain unchanged or be diminished. During the second stage, the dominant factor is the increased irritability of the heart muscle. This stage is marked by great irregularity both in rate of contraction and extent of contraction. This stage continues until the circulation becomes insufficient. The rather permanent character of the toxic effect of digitalis upon heart muscle when administered in large doses has a suggested explanation in some

experiments by Schliomensum[47], regarding the combining power of heart muscle extracts with digitalis thereby depriving it of its toxic properties. He found that he could obtain such an extract which mixed with digitoxin prevented the latter from readily causing systolic standstill in the frog's heart. Kloptowsky[30], has described the histological changes which occur in various organs in cases of digitalis poisoning. In the heart muscle he found parenchymatous and fatty degeneration and in some cases fragmentation of the fibers.

We see that in the study of a drug which possesses such wide range of activity, the extent to which this action may be limited to any particular field is a difficult matter to determine. The effect of digitalis on the nerve endings as opposed to that on the muscle itself in the peripheral neuromuscular complex of the heart is still one of the open questions. It was with a view of contributing to the knowledge of the relative effect of digitalis on these two factors of the heart i.e., the cardiac nerve endings and the muscle substance, that the present experimental work was undertaken.

III. Materials and Methods.

I. Animal Used.

For the investigation of this problem the turtle's heart was chosen as research material. The specimens were of the mud turtle variety, Emys blandingi, a species common to the region extending from New York to Wisconsin.

2. Solutions and Drugs Used.

The solution used for application in maintaining a normal condition was 0.7% sodium chloride, which is taken as able to keep the tissues of the turtle in a condition fairly normal for a time sufficient for pharmacological tests.

The digitalis preparation used was the Digitalin Pure, guaranteed by the Mallinckrodt Chemical Works. This was the German water-soluble product and contained the soluble active principles of the leaves of Digitalis purpurea. The Digitalin and Digitoxin though insoluble when isolated in pure form are held in suspension in a solution of this preparation by the presence of Digitonin which is readily soluble in water. A stock solution of 0.1% of this drug was made up in

physiological saline and dilutions to the proper per cent were made as desired. Fresh solutions were made frequently to insure against deterioration of physiological activity.

The atropine solution was one of atropine sulphate in Ringer's solution, sodium chloride- .7%, potassium chloride .03%, calcium chloride .026%. The stock solution was .01% in Ringer's and suitable dilutions were made as used.

The apocodeine used was a solution of apocodeine hydrochloride Merck in .7% saline, The stock solution was 1% and dilutions were made therefrom.

3. Methods.

The method used was in principle the one described in Greene's[24], Experimental Pharmacology, p. 66. It was the heart strip method and consisted essentially in suspending in a .7% saline a strip cut from the ventricle of a turtle. One end of the strip was fixed and the other attached to a lever of the first class by which a record of its contractions were obtained. In detail the method was as follows: The turtle's heart was exposed in the ordinary manner and the ventricle entirely removed. The base of the ventricle was grasped with curved forceps and a curved cut made around the apex from one angle of the base to the opposite. In the apex strip thus obtained two cuts were made parallel

to the first. Thereby three strips were obtained; one from the dorsal and one from the ventral surface of the ventricle, and a third from the apex. The cuts were made so that the strips were as nearly alike as possible in width and mass. Silk threads were then tied to the ends of the strips by means of which they were attached, one end to the L-shaped hooks of the ordinary Harvard form of muscle warm chamber support, the other to the short arm of a light straw lever recorder. The three levers of a set were arranged to record one above the other on a smoked cylinder. Five-second intervals were recorded on the cylinder by an electromagnet time marker. A signal marker was used to indicate the time of the application of the drug. The heart strips were immersed in small vials containing about 25 cc of .7 per cent NaCl. The change from one solution to another was accomplished by quickly removing one vial and substituting another containing the desired drug. When the strips had attained a regular rhythm in physiological saline the drug was applied in the manner indicated. No particular uniformity was observed in the length of time of the application of the drug. This was governed somewhat by the effect which was manifested. If after the first application a regular rate and amplitude was restored, a second test was made and so on for consecutive trials as regularity in the after period was maintained.

In obtaining apropinized strips, the heart was exposed in the usual manner. The right vagus nerve was dissected out in the neck and tested by continuous induction from an ordinary induction coil. A strength of stimulation was used which would produce stopping of the heart when applied for a few seconds. A quantity of .01% atropine in Ringer's was injected into the vein of the left side near its junction with the sinus. After a few preliminary trials it was found that 1 cc of .01% atropine was sufficient to produce paralysis of the vagus endings in the heart as indicated by the failure to obtain stopping of the heart by stimulation of the vagus with strength of stimulus much greater than was able to produce inhibition at first. The strips were then cut and mounted as before described.

A similar procedure was followed in the use of apocodeine. Some of the experiments in which this drug was employed were conducted by injection of apocodeine into the vein following the injection of atropine. In others the apocodeine was applied to the unatropinized or atropinized strips in the same manner as the saline and digitalis solutions.

IV Experimental Results.

This series of experiments was performed with the idea of determining the pharmacological effects of digitalis upon the terminal cardiac mechanism, particularly the relative extent of this action upon, 1, the nervous, and 2, the muscular elements in the ventricle.

The strongly developed inhibitory mechanism of the turtle's heart and the probable absence of any accelerator mechanism make it especially adaptable to experimentation which involves a determination of the relative influence of any pharmacological agent upon these two factors. The remarkable control which the inhibitory nervous apparatus exerts upon the heart is a matter of common knowledge. Mills[40] reported an instance in which the contractions of the heart of the terrapin were held in check for more than four hours by continuous stimulation of the right vagus nerve.

That the ventricle muscle of the turtle is directly supplied with inhibitory nerve elements has been shown by Gaskell[19]. He describes a nerve, which he calls the coronary nerve, connecting the sinus directly with the ventricle in the turtle's heart. By severing all connection between the sinus and the auricles, leaving only his coronary nerve intact and allowing time for the development of a spontaneous rhythm in the auriculo-ventricular

portion, Gaskell was able to produce a cessation of the ventricular rhythm by stimulation of the right vagus in the neck. Although these results were not always constant they are very positive and are taken as sufficient evidence of the presence in the tissue of the ventricle of well developed nervous elements the stimulation of which will produce a retardation of the ventricular rhythm. Mills has reported evidence of a cardiac accelerator mechanism in this group of reptiles, work that has lacked confirmation and is now discredited.

A discussion of influence of any agency in producing changes in automatically contracting tissue involves the fundamental question of the causation of rhythm. The more clearly the facts of the phenomenon of automatic contractility are understood the easier will be the interpretation of modifying conditions . Heart tissue is automatic, that is, when placed under suitable conditions it is able to initiate and maintain rhythmic contractions for periods of time the durations of which depends upon the surrounding environment. Whether this property of automaticity and rhythmicity is due primarily to the cyclic changes which occur in the muscle alone, or is brought about by definite nerve elements arranged in a definite manner is a question that has long been in dispute. On purely theoretical grounds it is easily conceivable that either one or the other or both of these factors might be concerned in the initiation of contractions. Neither view can be said to have been conclusivly

demonstrated up to the present time, but the weight of evidence is in favor of the theory that automatic contractions of heart muscle occur independently of a definite nervous mechanism.

Gaskell was the first to show that strips taken from various parts of the heart of cold blooded animals would contract under suitable conditions, a fact which is part of the evidence for the conception that definitly arranged nerve elements are not necessary to the rhythm. This evidence does not exclude the undeniable fact that regulation of the activity of the heart as a whole is maintained chiefly through its connections with the central nervous system. Other influences shown by Greene[23] and by Howell[25] to be of eminent importance in maintaining and controlling cardiac movements is the constitution of the surrounding medium, especially the extent and nature of the inorganic ionic content.

1. <u>The Pharmacological Action of Digitalis Solutions upon the Rate, Amplitude, and Tone of Strips from the Ventricle of the Turtle.</u>

Strips of cardiac tissue, cut in the manner indicated in the chapter on Methods, are physiological complexes containing at least two elements upon which pharmacological agents may act in producing changes in condition or activity. These two factors are the muscle cells and the terminal nerve elements. No definite nerve ganglia have been demonstrated to exist in the ventricle, The

nerve fibres can be left out of consideration hence the only nerve elements which we need to consider are the nerve endings,- the inhibitory nerve terminations, and the questionable accelerator endings. The cardiac strips when immersed in physiological saline begin contracting rhythmically after a latent period of variable length, and if undisturbed continue their rhythm for several hours. This activity of ventricular tissue is essentially physiological. Influences which modify the rate, amplitude, and tone of such preparations may be taken as capable of acting in a similar manner upon this part of the cardiac complex under strictly normal conditions. The terminal effects of digitalis observed in the experiments presented, were determined from such strips. The results obtained were measured in terms of change in rate, in amplitude of contractions, and in change of tone.

(a) <u>The Pharmacological Effects of Digitalis upon the Rate of Ventricular Strips.</u>

In my series of experiments over 86 per cent of the total number showed an increase in rate under the influence of digitalis. The general average percentage increase in rate of the entire group of thirty experiments was 24.26 per cent. It will be noted by referring to Tables I and II that included in this average are all those results from strips which show no changes, and also those which show an actual decrease in rate. The percentage increase in rate, considering only those showing an in-

crease is of course much greater than the general average. Tables I and II show the changes in rate occuring in individual strips. The strengths of digitalis used were .01 and .05 per cent, respectively. In addition to the experiments recorded in these tables, in a series of ten experiments using .005 per cent digitalis but not reported in detail, 9 out of 10 strips gave acceleration. It is evident therefore that digitalis produces a marked acceleration of the contraction rate of ventricular strips.

There are a number of exceptions to the general rule as regards increase in rates shown in Tables I and II. These exceptions occur chiefly in those strips which showed by a condition of irregularity of rate before the application of digitalis that they were in poor condition. Experiments Nos. 3, 7, and 11 are examples of this class. Experiments 19, and 29 though they show a decrease in rate during the application of digitalis are not actual exceptions to the rule since the typical digitalis effect did not occur till later, as shown by the increase in rate in the after period.

(b) The Effect of Digitalis upon the Amplitude of the Contractions.

Reference to Table I and II, or to the Summary of Results given in Table V, will indicate that the usual effect produced by the action of digitalis upon strips of ventricular muscle is a reduction in the amplitude of the contractions. The average percentage of this reduction for the entire series of experiments is 5.4 per cent.

There are two possible explanations for this de-

crease in amplitude. Either there may be a primary effect upon the factors which are immediately concerned in the determination of amplitude, or the change may be secondary to the increased rate. Explanation on the latter basis involves the "all or none" law of contraction and the principle of compensatory pause which was first formulated by Bowditch and now universally recognised. He observed that the heart muscle responded to electrical stimulation, if it responded at all, with a maximal contraction. He also observed the relation of the amplitude of contractions to the rapidity of contractions. He found as the rate was increased that the amplitude was correspondingly decreased. The principle applied to the relation of rate and change in amplitude was observed in the fact described by Gaskell with regard to the heart of the tortoise. He stated that upon stimulation of the vagus nerve a decrease in rate is always accompanied by a corresponding increase in amplitude, and that during the after-acceleration, which he describes, following vagus stimulation, the amplitude was decreased proportionately to the increase in the rate. In other words, although the factors which control the rate and amplitude are probably not identical, they are, at least, closely correlated.

On the basis of the above principle, influences which modify the rate of the heart, either by acceleration or depression, other conditions being equal, should

be held responsible for a corresponding but inverse alteration of amplitude. It is certain that digitalis sets this law into operation when it accelerates the heart rate. The question is, then, to determine whether the extent of this decrease in amplitude is commensurate with the acceleration in rate. If it is, then the amplitude change may be considered wholly secondary. Otherwise one may look for direct effects of digitalis on the amplitude even though masked by the operation of the physiological law. The question is, is there a direct stimulating action of digitalis on the amplitude.

An inspection of the results obtained with normal ventricular strips under the influence of digitalis brings to light certain conclusive facts against the view that digitalis affects the amplitude secondarily only. A comparison of the average percentage increase in rate with the average percentage decrease in amplitude of the series of unatropinized strips in Table VI shows clearly that there is a striking quantitative difference between the two figures. The average percentage of rate increase is 24.26 per cent while the average percentage of amplitude decrease is only 5.6 per cent. This striking lack of correspondence between these percentages throughout the entire series points to a positive stimulating effect upon the factor of amplitude. If we consider the individual groups of experiments the same type of variation is observed. The lack of conformity

between these two factors of percentage increase in rate and percentage decrease in amplitude cannot be explained on the basis of Bowditch's law of maximal contractions. In terms of that principle an increase in the rate produces a corresponding decrease in amplitude of contraction. Consequently, if this principle alone were operative in these experiments one ought to be able to check it out in a comparison of rates and amplitudes in groups of experiments of different strengths of drug. The percentage increase in rate Table VI, Group 6, 25 per cent, was greater than the percentage increase in rate of Group 3, 18.9 per cent. There should be a correspondingly greater percentage decrease in amplitude in Group 6, On the contrary, the percentage decrease in amplitude is less in Group 6, 5.4 per cent, than that in Group 3, 6.4 per cent. The only interpretation, obviously, is that there is a positive tendency for digitalis to produce an increase in amplitude. This conclusion is strengthened by the fact that a solution of .01 per cent digitalis was used in Group 3, and .05 per cent in Group 6. The greater concentration produced a smaller decrease in amplitude. This is interpreted as a direct tendency toward an increase of amplitude. In short, the compensating decrease in amplitude is partially but not wholly overcome by a direct stimulating action of digitalis on the amplitude.

This positive stimulating effect of digitalis upon amplitude stands out clearly in a number of individual

experiments which have been noted previously. Experiment 25, Table II, in the after period showed an acceleration of rate of over 30 per cent while at the same time an amplitude increase of 50 per cent was observed. Experiment 19 is similar in all respects to Experiment 25, showing in the after period an increase in rate of 4 per cent and an increase of amplitude of more than 10 per cent.

In the light of these facts digitalis undoubtedly has a stimulating effect upon the contraction volume factor in cardiac muscle, tending to produce an increase in amplitude.

(c) The Effect of Digitalis on Cardiac Muscular Tone.

The property of continuing in a more or less permanent condition of contraction is one which is possessed by muscular tissue in general. This is however especially characteristic of the heart muscle, particularly that from the venous end of the organ. It was first noted by Greene that strips of the ventricle of the turtle continually lost tone during an experiment. This gradual fall in the base line was also observed in practically all of the experiments upon which this paper is based. The tone of the heart muscle differs from that of skeletal muscle tone in the fact that it is present even after the connections with extrinsic nerves are severed. It seems probable that this property is one particularly belonging to cardiac muscle.

Table V gives the percentage of the total number of experiments on unatropinized strips which show rise of tone. This is 70 per cent of the total number. Table VI shows by comparing the first three groups of unatropinized strips with the second three that the greater concentration of digitalis produces a correspondingly greater effect upon the tone thus indicating a primary and positive action.

The effect of the application of digitalis in producing tone changes is not limited to the time during which the drug is applied. In fact in the greater number of experiments the maximum rise in the base line, the measure of the state of tone, did not occur during but after the application of the drug. It is also noticeable that the return to the normal state of relaxation is very slow, a fact which illustrates the prolonged action of digitalis. Digitalis then markedly increases the general condition of contraction of ventricular strips as expressed by tone.

Just here, we may make an additional point as regards the influence of digitalis on the amplitude of the contractions of heart strips. The usual measure of the amplitude does not indicate the total state or condition of contraction. One must add to this measure the amount of tonic contraction as usually indicated. Measured by the sum of the change in amplitude and the change in the tone, the effect of digitalis now becomes sharply and unquestionably stimulative. The sum of the two factors

gives an idea of the effect of digitalis on the state of contraction. Take for instance Experiment 13, Table II. The amplitude of contraction before the application of digitalis was 26 mm. During the use of the drug it was decreased to 24 mm. However, considering the fact that at the same time the tone rise was 6 mm., the strip was actually in a state of contraction greater than before the drug.

Still another factor which should be included in any full determination of the total state of contraction is the fact that if left alone the strip would show a gradual and continual loss of tone. Thus, an expression of the total state of contraction must include a consideration of the proportionate rate of loss of tone taking place before the rise of tone occured.

2 The Pharmacological Action of Digitalis upon Strips of the Ventricle of the Turtle cut after the tested Elimination of the Inhibitory Control of the Vagus by Atropin.

The purpose of this group of experiments was to determine the effect of digitalis upon strips cut from the ventricle of the turtle after the elimination of the inhibitory control of the vagus by atropine. In the atropinized strips, we were presumably dealing with only one kind of tissue, i.e., cardiac muscle. By making a comparison of the effects of digitalis upon muscle alone with its effects upon a preparation including both muscle

and inhibitory nerve endings, it was hoped to determine the influence exerted upon each separate factor.

In subjecting ventricular strips to the influence of atropine in order to paralyze the vagus inhibitory endings, the question arose as to whether the muscular tissue was not also affected by atropine. Fortunately the influence of atropine on strips of the turtle's ventricle has been investigated by Stowers[48]. While the greater number of his experiments have shown that in the concentrations used, no effect was produced upon either the rate or the amplitude of heart strips during the application of atropine, the number of variations was about equally divided, showing rather great extremes. The concentration which Stowers used in determining these facts was double that used in my experiments to paralyze the vagus endings.

Additional evidence that atropine did not in my experiments produce any very obvious alteration in the condition of the muscle is in the fact that the average initial rates of the unatropinized and the atropinized strips are in such close correspondence. Table VI shows the average rate before the first application of the drug for the entire series of unatropinized muscles as 11.8 per minute. The corresponding average for the atropinized strips is 11.1 per minute. This comparison and the facts Stowers brought out justify the conclusion that whatever the effect atropine may have, it was in the present percentages confined chiefly to paralyzing the

inhibitory nerve endings. The condition of the muscle after poisoning the inhibitory nerve endings with atropine compares favorably with the normal.

(a) The Influence of Digitalis on the Rate of Atropinized Heart Strips.

The effect of digitalis upon the atropinized strips was the same in nature as that produced upon the unatropinized strips. As has been stated, the average rates before the application of digitalis in both series were nearly identical. Digitalis produced acceleration in 77.7 per cent of the total number of experiments of the atropine series. The percentage increase for the entire series shown in Table VI was 17.1 per cent. The average percentage increase in rate for the different groups was quite variable. For instance Group 3a gave an average increase in rate of 7.1 per cent. The average percentage increase in rate for Group 5a was 35.5 per cent. These variations become of relatively little significance when we consider the wide difference which Stowers found in the rate of strips under the influence of atropine. As regards the influence of digitalis on rate, it is seen that although the average for all the strips atropinized is somewhat less than that of the ones unatropinized nevertheless the effects which digitalis exerts upon cardiac muscle strips is nearly as great when the inhibitory nerve endings have been paralyzed by atropine as when the endings are present.

The effect to be expected in the series of unatro-

pinized strips, if digitalis stimulated exclusively the inhibitory nerve endings in the muscle, is a primary slowing with strong concentrations or even a slowing with weaker concentrations. If the assumption is made that digitalis stimulates these nerves even in part, with the nerve endings eliminated by atropine, the expected result is a faster average rate. This was not found to be true. The average increase in rate in the series of unatropinized strips was in fact higher than in the series of atropinized strips. It might be argued that the hypothetical stimulating effect of digitalis on the inhibitory nerve endings may possibly have been masked by stimulation of the muscle to greater excitability. If such were the case the strips which have the inhibitory factor eliminated should show a greater percentage increase in rate. No such increase in rate was observed in the atropinized strips. In fact, the average percentage increase in rate for the entire series was less than that of the unatropinized group. We are led to the conclusion, therefore, that as far as the factor of rhythmicity is concerned the marked acceleration of rate which digitalis produces is due to its influence upon the muscle, and that the inhibitory nerve endings are not acted upon by this drug.

(b) The Influence of Digitalis on the Amplitude of Atropinized Heart Strips.

In considering the effect of digitalis upon the ampl-

itude of atropinized strips we meet, as in the case of rate, with the fact of wide variations in the individual results. The percentage of the total number showing a decrease in amplitude was 55, indicating the general tendency of the influence of digitalis. The percentage decrease in amplitude for the entire series, shown in Table VI was 1.8 per cent. The variations shown by groups of experiments on strips cut from different regions of the ventricle are considerable. Group 3a, Table VI shows a decrease in the amplitude of 6 per cent, while Group 6a has a positive increase of 6.1 per cent. These rather extreme individual variations are probably due to some action of atropine on the heart muscle, since no other new variant had been introduced.

We are here, as in the series of unatropinized strips, confronted with the fact that the influence of digitalis upon contractility as judged by the effect upon the amplitude is confusing, i.e. masked. There is the same lack of percentage correspondence between the increase in rate and the decrease in amplitude. The differences are even more marked in some groups of this series than in the unatropinized groups. Group 5a, Table VI shows a rate increase of 35.5 per cent while the amplitude is decreased only 3.6 per cent. Group 6a of the same series, shows a rate increase of 11 per cent, while, contrary to the general rule there is an actual increase in the amplitude to 6.1 per cent. These marked differences on atropinized strips give further confirmation of the conclusions reached in the

studies of strips from normal hearts. If still more convincing evidence is needed it is furnished by Group 6a in which the stimulation of the amplitude factor more than compensates for the normal reduction due to the accompanying increase in rate.

Another fact giving evidence in support of the argument that digitalis is stimulative to the contraction factor in the heart strip is seen in Table V. With the concentration of .01 per cent solution of digitalis twelve out of eighteen experiments showed a decrease in rate, while .05 per cent solution of digitalis produced a decrease in only eight out of eighteen experiments. Although this result is not of very great statistical importance it illustrates that the tendency toward an increase in amplitude is greater with the greater concentrations of the solution.

The positive tendency toward increase in amplitude shown in the above atropinized preparations, and the fact that this tendency is greater with the stronger solutions of digitalis leads to the conclusion that digitalis stimulates the rate factor in the heart by direct muscular action.

(c) <u>The Effect of Digitalis upon the Tone of Atropinized Ventricular Strips.</u>

The effect which digitalis has upon the tone of atropinized ventricular strips is the same in character as that upon this factor in strips under normal condi-

tions. As in the unatropinized strips the usual result is a well marked rise in the tone. The percentage of the entire number of experiments on atropinized strips which show increase in tone was 72 per cent. The average rise in tone for this series was .58 cm. Although this is somewhat less than the average rise in tone of the unatropinized group as a whole, the results in Group 1a, Table VI, which is an exception to the rule accounts for the greater part of this difference. With this single exception the rise in tone in the atropinized strips is practically as great as in those unatropinized.

As to the factor upon which digitalis acts in producing this rise in tone, it is easily seen that the effect is not modified to any extent by the elimination of the inhibitory nerve endings. Stimulation of the vagus in the intact turtle produces marked relaxation of the ventricle. Assuming that stimulation of the vagus nerve endings produces a similar effect in the unatropinized strip, we would expect a more marked tendency toward relaxation in the unatropinized than in the atropinized strips. On the same grounds, .05 per cent. digitalis ought to produce either an equal or a less marked rise in tone than the weaker concentration, .01 per cent. The facts are that unatropinized strips actually show a greater tone change than atropinized, and, from a similar cause, the effect shown by .05 per cent on the atropinized strips is greater than that

shown by .01 per cent digitalis in the same series. This clearly indicates that digitalis acts only upon the muscle in producing tone changes in the ventricular strips.

It is evident that these experiments support the view that digitalis in its peripheral action influences the heart by a direct muscular change in both rhythmicity and contractility to the exclusion therefore of the terminal inhibitory nerve endings.

3 The Pharmacological Effect of Digitalis Solutions upon Strips of the Ventricle of the Turtle after being Subjected to Apocodeine.

As the purpose of this investigation was to determine if possible the effect of digitalis on all the factors in the peripheral neuro-muscular complex a consideration of possible effects which might be due to the presence of accelerator nerve elements in the cardiac tissue was necessary.

This part of the question was approached on somewhat hypothetical grounds. The presence of a definite accelerator mechanism in the turtle's heart has not been demonstrated or at least not confirmed. Mills[40] has given in detail the innervation of the slider terrapin in which he describes a very definite path for cardiac accelerator fibers. However, his results have not been generally accepted. Besides his investigation was confined to one particular species of terrapin.

An accelerator mechanism certainly has not been demonstrated for the particular species of turtle used in the present work.

But considering the possibility that accelerator nerves might be present in this species it was thought advisable to attempt to eliminate any such factor and determine the subsequent influence of digitalis. Accordingly a drug was selected which has been shown to paralyze the accelerator endings in the mammalian heart. The drug used was the alkaloid apocodeine. This substance has been the subject of investigation by Dixon[14]. He has shown that apocodeine when injected into the circulation of the cat in sufficiently large quantities will produce paralysis of the intra-cardiac ganglia first, then the vagal nerve endings, and finally the accelerator nerves. About twice the amount is required to produce paralysis of the accelerator endings as of the vagus.

A few preliminary perfusion experiments upon the isolated heart of the turtle demonstrated that a prolonged perfusion with .04 per cent apocodeine was required to produce any paralysis of the vagus. Even then the inhibitory control was only partially eliminated. Marked reduction in the rate and amplitude of contraction indicated also a toxic effect upon the muscle. Accordingly some experiments were tried in which the strips were cut after slowly injecting 2 cc.

of 1 per cent apocodeine into the veins, following it by an injection of a sufficient amount of atropine to paralyze the vagus. Other experiments were performed in which the strips cut from the atropinized ventricle were subjected to a solution of .05 per cent apocodeine. The number of experiments of this series is not large. The direct depression of the rhythm and contractility of the muscle produced by apocodeine greatly interfered with the definiteness of the proposed tests.

The results obtained from application of digitalis to strips so treated are given in Table VII. The limited number of experiments and the uncertainty of the muscular influence of apocodein itself, which has not been investigated in this relation, do not justify very definite statements. The experiments at hand indicate that the influence of digitalis after apocodeine and atropine are not far different from the average obtained upon strips that were tested after treatment with atropine alone.

In a word, previous treatment of turtle hearts with apocodeine to eliminate any hypothetical cardiac accelerator nervous mechanism has only added another to the list of negative attempts to prove the presence of this apparatus. However, the experiments give the support of negative evidence to the conclusion that the peripheral influence of digitalis is all on the cardiac muscle.

V Summary.

1. The primary local influence of digitalis on the ventricle is to accelerate the rate, stimulate the amplitude and increase the tone.

2. The action of digitalis is directly stimulative to cardiac muscle.

3. There is no evidence of a stimulating action of digitalis on inhibitory nerve endings, as tested by its action on atropinized and unatropinized ventricular muscle.

4. There is no evidence of stimulation of any hypothetical accelerator nerve endings, as shown by the tests of the influence of digitalis after apocodeine.

5. The stimulating effect of digitalis on amplitude of contraction is more or less masked by the compensatory correlation between rhythm and amplitude.

TABLE I.

Tabulated Results of Experiments Showing the Effect of .01 per cent. Digitalis in physiological saline upon the Rate, Amplitude, and Tone of Strips from the Ventricle of the Turtle.

Exp. No.	Drug, Sec.	Rate per Minute			Amplitude in mm.			Tone change in mm.	
		Before	During	After	Before	During	After	During	After
1	35	5.9	8	8	48	4.7	44	+1	+6
2	60	16	16	15	24	23	23	0	-2
3*	120	12.5	12	12	24.5	24	23.5	+0.5	+1
4	120	10	11	10	34	32	31.5	+2	+3.5
5	120	14	16.5	16.5	20	18	19	+1	+1
6	85	14	17	12	27	25	28	-2	-3
7*	75	16	16	16	9	9	8	_	_
8	120	5.5	7	7	30	29	29.5	+0.5	+0.5
9	120	12.5	18.5	14	8	7	8	+1	+1
10	170	13.5	15	16	21	20	21	0	0
11*	120	11	11	11	16.5	16.2	16	+0.5	+0.5
12	120	10	11.7	11	24	23	24	_	-1

* Strips in Experiments No's. 3, 7, and 11 were irregular in rate before the application of the drug.

TABLE II.

Tabulated Results of Experiments Showing the Effect of .05 per cent. Digitalis in physiological saline upon the Rate, Amplitude, and Tone of Strips from the Ventricle of the Turtle.

Exp. No.	Drug, Sec.	Rate per Minute			Amplitude in mm.			Tone change in mm.	
		Before	During	After	Before	During	After	During	After
13	50	8.9	16	17	26	24	31	+6	+2
14	65	9.5	13	12	30	27	29	+3	0
15*	60	9	11	7	25	26	28	+1	+1
16	60	10	12	11	45.5	42.5	47	+3	+0.4
17	65	14	18	15	63.5	61	66	+2.5	-1
18	55	11	12	10	53	51	51	+3.5	+2
19**	45	23	22	24	29	29	32	+3	-1
20	70	12.5	14.5	22	21	19	18	+2	+1
21	40	11	18	18	37	35	38	-5	-1
22	65	10.5	12	11.5	16	15	15.5	0	+2
23	60	13.5	20	18.5	49	43	46	+5	+2
24	55	8	12	10	47.5	41	38	+7	+9
25**	55	17	16	22	10	11	15	+1	0
26	80	12	16	23	30	27	20	+3	+2
27	45	11.4	18	13	12	10	13	0	-2
28	80	9.5	12	10.5	20.5	20.5	22	+0.5	+1
29	85	16	18	17	24	22.5	24	+2	-0.5
30	55	12	11	11.5	13.5	15	19	0	-0.5

* Strip in Exp, 15, amplitude gradually increasing before the application of the drug.

** Strips in Exp's. 19 and 25 show unusually rapid initial rate.

TABLE III.

Tabulated Results of Experiments Showing the Effect of ,01 per cent. Digitalis in physiological saline upon the Rate, Amplitude, and Tone of Strips cut from the Ventricle of the Turtle after elimination of the inhibitory control of the Vagus by Atropine.

Exp. No.	Drug, Sec.	Rate per Minute			Amplitude in mm.			Tone change in mm.	
		Before	During	After	Before	During	After	During	After
1a	70	10	13	11.5	38	37	38	0	+1
2a	100	11.5	14	14.5	28	27	28	-2.5	-1
3a	50	12.5	16	17	38	36	33	-1	0
4a	80	8	8.9	9	36	36	37	-1	-2
5a	120	10	12	9	30	28.9	30	+0.2	+0.6
6a	120	4.5	6	6	55.5	53	55	+2.5	0
7a	100	21.5	22	21.5	8	8	8	0	0
8a	135	16	19	17.5	26	22	18	+1	-1
9a	140	5.9	7.9	10	28	28	28	-2	-4
10a	120	16	15.5	16	16	15	14.5	+0.1	0
11a	120	10	10.5	9	13	13.5	13	+0.5	0
12a	120	7.5	9.5	_	24	23	24.5	+4	0
13a	75	17	16	17	25	23	22	+2	+1
14a	55	15	16	14.5	17	15	14	+3	+5
15a	90	5	6	7	29	28	30	0	0
16a	210	11	11.9	12.5	37	34	34	+3	+3
17a	120	14	15	15	8.5	8.5	9	+0.5	0
18a	120	12	13	11	12.2	11.5	12.5	+0.3	+0.7

TABLE IV.

Tabulated Results of Experiments Showing the Effect of .05 per cent. Digitalis in physiological saline upon the Rate, Amplitude, and Tone of Strips cut from the Ventricle of the Turtle after elimination of the inhibitory control of the Vagus by Atropine.

Exp, No.	Drug, Sec.	Rate per Minute			Amplitude in mm.			Tone change in mm.	
		Before	During	After	Before	During	After	During	After
19a	140	9	9	7.9	32	31	33	+2	+5
20a	90	6.9	10	27	36	32	34	+5	+2
21a	55	12	13	14	50	48	48	+2	+1
22a	60	13	12	12	38.5	40	38	+1.5	+1.5
23a	60	11.5	14	15	34	32.5	35	+1.5	+0.5
24a	65	9.5	9.5	8	44	44.5	46	+2	+1
25a	480	9	8.5	8	39	35	20	+1	+2
26a	90	9	12.5	13	13	13	15	-1	-2
27a	70	4	8	7	26.5	24.5	26	+1	-1
28a	55	8	7.5	10	23.5	23.9	22.5	0	+0.5
29a	55	11	16	14	32	29	32	+7.5	+4
30a	55	10	11	14	20	21	21.5	+2	+1.5
31a	170	9	9	7	27	28	30	+2	+4
32a	105	16.5	16	23	25	26	26	+1	+1
33a	55	16	18	16	10	9	10	-0.5	-0.5
34a	60	11	10	11.2	17	20	18.5	+2	+1.5
35a	75	16	18	20	14.5	18.5	22.5	-5	-5.5
36a	60	16	16	15	12.5	12.5	13	+1.5	0

TABLE V.

Classifying Experiments with .01 per cent. and .05 per cent. Digitalis on Unatropinized and Atropinized Strips from the Ventricle of the Turtle to indicate the Percentage of Experiments showing No Change, Increase, or Decrease,- in Rate, Amplitude, and Tone.

Per cent. of Drug	No, of Exp'mts	Number showing								
		No Change			Increase			Decrease		
		Rate	Ampl.	Tone	Rate	Ampl.	Tone	Rate	Ampl.	Tone
Unatropinized Strips										
.01	12	2	2	4	9	0	7	1	10	1
.05	18	0	2	3	17	3	14	1	13	1
Total	30	2	4	7	26	3	21	2	23	2
Average per cent. of total		6.6	13.3	23.3	86.6	10	70	6.6	76.6	6.6
Atropinized Strips										
.01	18	0	4	2	16	2	11	2	12	5
.05	18	3	2	0	12	8	15	3	8	3
Total	36	3	6	2	28	10	26	5	20	8
Average per cent. of total		8.3	16.6	5.5	77.7	27.7	72.2	13.9	55.2	22.2

TABLE VI.

Showing Per Cent. of Change produced by Digitalis in Rate, Amplitude, and Tone of Unatropinized and Atropinized Strips classified according to the Region of the Ventricle from which the Strips were obtained.

Unatropinized Strips

Group	Region	No. of Exp'mts	% of Drug	Average Initial Rate	Average % Rate Increase	Average % Ampl. Change	Av. Tone Change in mm.
1	Apex	4	.01	10.9	16.3	-3.4	+0.87
2	Dors.	4	.01	12.3	22.1	-5.9	-0.1
3	Vent.	4	.01	11.7	18.9	-6.4	+0.18
4	Apex	6	.05	10.4	30.7	-4.7	+3
5	Dors.	6	.05	13	32.6	-7.8	+2
6	Vent.	6	.05	12.9	25	-5.4	+1.06
General average				11.8	24.26	-5.6	+1.16

Atropinized Strips

Group	Region	No. of Exp'mts	% of Drug	Average Initial Rate	Average % Rate Increase	Average % Ampl. Change	Av. Tone Change in mm.
1a	Apex	6	.01	9.4	24	-3.2	-3.3
2a	Dors.	6	.01	12.8	14.2	-1.5	+0.6
3a	Vent.	6	.01	12.3	7.1	-6	+1.3
4a	Apex	6	.05	10.3	11.1	-3	+2.3
5a	Dors.	6	.05	8.5	35.5	-3.6	+2.5
6a	Vent.	6	.05	14	11	+6.1	+0.1
General average				11.1	17.1	-1.8	+0.58

TABLE VII.

Tabulated Results of Experiments Showing the Effect of .05 per cent. Digitalis in physiological saline upon the Rate, Amplitude, and Tone of Strips of the Ventricle of the Turtle cut after elimination of the inhibitory control of the Vagus by Atropine and treatment with Apocodeine.

Exp. No.	Drug Sec.	Rate per Minute			Amplitude in mm.			Tone change in mm.	
		Before	During	After	Before	During	After	During	After
1aa	230	5.5	5.9	6.5	9	9.5	12.5	-0.2	-0.5
2aa	540	11	10	4.5	47.5	40	23	+0.5	+5
3aa	120	5.5	7.5	7.5	24	21.8	24	+3	+3
4aa	65	7	8	10.5	40	38	37	+1.5	+1.5

Average rate at the beginning of experiments	7.2 per min.
Average per cent of increase of Rate	12.2 per cent.
Average per cent of decrease of Amplitude	4.3 per cent.
Average increase of Tone in mm.	1.2 mm.

Literature List

* Read in the original.

1. Ackermann: Über die physiologischen Wirkungen des Digitalins auf den Kreislauf und die Temperatur. Deutsch. Arch. f. klin. Med. XI, 125, 1873.

*2. Bastedo: Text book of Materia Medica, Pharmacology and Therapeutics. Philadelphia, 1913.

3. Blake: Observations of the Physiological Effects of Various Agents introduced into the Circulation as indicated by the Haemodynamometer. Edinb. Med. and Surg. Journal LI, 330, 1839.

*4. On the Action of Digitalis. Jour. of Physiol. IV, 365, 1883.

5. Beddoes: Observations on the Powers of Digitalis purpurea. London, 1801.

6. Boehm: Untersuchungen über die physiologische Wirkung der Digitalis und des Digitalins. Arch. f. d. ges. Physiol. V, 153, 1872.

7. Briquet: Traité thérapeutique du quinquina et de ses préparations. Paris, 1853.

8. Brunton: On Digitalis. Thesis. Edinburgh, 1866. Journal of Anat. and Physiol. I, 154.

9. Brunton and Meyer: Action of Digitalis on the Bloodvessels. Jour. of Anat. and Physiol. VII, 134, 1873.

*10. Brunton and Tunnicliffe: On the cause of the Rise of Blood Pressure Produced by Digitalis. Jour. of Physiol. XX, 354, 1896.

11. Cloetta: Über die Bestandtheile der Folia digitalis. Arch. f. exper. Pathol. u. Pharmacologie. XLI, 421, 1899.

12. Zur Kentniss der Darstellung und Zusammensetzung der Digitalis glykoside. Arch. f. Pathol. u. Pharmacologie. XLV, 435, 1901.

*13. Cushny: On the Action of Substances of the Digitalis series on the circulation in Mammals. Journ. Exper. Med. II, 233, 1897.

*14. Dixon: The Paralysis of Nerve Cells and Nerve Endings with special reference to the Alkaloid Apocodeine. Journ. of Physiol. XXX, 97, 1903.

*15. Donaldson and Stevens: The Influence of Digitaline on the Work of the Heart and on the Flow through the Blood-vessels. Jour. of Physiol. IV, 165, 1882.

*16. Edmunds: The Influence of Digitalis, Strophanthus and Adrenalin upon the velocity of the Blood Current. Amer. Journ. of Physiol. XVIII, 129, 1907.

*17. Etienne: De l'action de la digitale sur le nerf vagus. Arch. Internation. de Pharmacodynamie et de Therapie. XX, 265, 1911.

18. Ferriar: An essay on the medical properties of Digitalis purpurea. London and Manchester, 1799.

*19. Gaskell: On the Innervation of the Heart with especial reference to the Heart of the Tortoise. Journ. of Physiol. IV, 43, 1882.

*20. Gottlieb and Magnus: Über die Gefässwirkung der

Körper der Digitalisgruppe. Arch. f. exper. Pathol. u. Pharmakol. XLVII, 135, 1901.

*21. Über den Einfluss der Digitalis Körper auf die Hirncirculation. Arch. f. exper. Pathol. u. Pharmakol. XLVIII, 262, 1902.

22. Gorz: I. Untersuchungen über die Nativelle'schen Digitalin Präparate in chemischer und physiologischer Beziehung. II. Ein Beitrag zur physiologischen Wirkung des Digitalins auf den Blutdruck. Dissertation. 1873. (Schmidt's Jahr. CLVIII, 234-238).

*23. Greene: On the relation of the Inorganic Salts of Blood to the automatic activity of a strip of Ventricular Muscle. Amer. Journ. of Physiol. II, 82, 1898.

*24. Experimental Pharmacology. Philadelphia, 1909.

*25. Howell: An Analysis of the Influence of Sodium, Potassium, and Calcium Salts of the Blood on the Automatic Contractions of Heart Muscle. Am. Journ. Physiol. VI, 181, 1901.

26. Johnson: Medical Essays and Observations. Evesham 1785.

*27. Jonescu and Loewi: Über eine spezifische Nierenwirkung der Digitalis Körper. Arch. f. exper. Path. u. Pharmakol. LIX, 71, 1908.

28. Kaufmann: Effets physiologiques de la digitaline amotphe: applications a la therapeutic. Revue de Medicine. IV, 381, 1884.

29. Kinglake: Cases and Observations on the medical efficacy of Digitalis purpurea in phthisis pulmonalis. London, 1801.

30. Kloptowsky: Über die Veränderungen in den Ganglien und der Muskulatur des Herzens, in der Leber und in den Nieren bei Digitalis Vergiftung. Russisch med. Rundshau. X, 1903.

31. Kobert: Über die Beeinflussung der peripheren Gefässe durch pharmakologischen Agentien. Arch. f. exp. Path. u. Pharmak. XXII, 77, 1886.

32. Kochmann: Beitrag zur Wirkung einiger Körper der Digitalis Gruppe auf den Nerv Vagus. Arch. Internat. de Pharcodyn. et de Therap. XVI, 221, 1906.

33. Koppe: Untersuchungen über die pharmakologischen Wirkungen des Digitoxins, Digitalins und Digitaleins. Arch. f. exp. Path. u. Pharmak. III, 274, 1875.

34. Lenz: Experimenta de ratione inter pulsus frequentiam, sanguinis fluentis celeritatem obtinente. Diss. Dorpat, 1853.

*35. Mackenzie: Digitalis. Heart II, 273, 1911.

36. Marme: Die wirksamen Bestandtheile des Helleborus niger, viridis und fostidus. Ztschr. f. rationelle Medicin. XXVI, 1, 1866.

*37. Marshall: On the Antagonistic action of Digitalis and the members of the Nitrite group. Journ. of Physiol. XXII, 1, 1897.

38. Meyer: Zur Lehre den Herzgiften. Untersuch a.

d. physiolog. Laboratorium der Zurich. Hochschule, 1869.

*39. Meyer and Gottlieb: Pharmacology, Clinical and Experimental. Philadelphia, 1914.

*40. Mills: The Innervation of the Heart of the Slider Terrapin. Journ. of Physiology, VI, 246, 1885.

41. Mossman: Essai sur les proprietes medicales de la digitale pourpree. Paris, 1812.

*42. Roy: On the Influences which modify the Work of the Heart. Journ. of Physiol. I, 452, 1878.

43. Schmiedeberg: Über die Digitalinwirkung am Herzmuskel des Frosches. Beiträge zur Anatomie und Physiologie. Festgabe an C. Ludwig. Leipzig 1874.

*44. Untersuchungen über die pharmacologisch wirksamen Bestandtheile der Digitalis purpurea. Arch. f. Pathol. u. Pharmak. III, 16, 1875.

*45. Beiträge zur Kentniss der pharmakologischen Gruppe des Digitalins. Arch. f. Pathol. u. Pharmak. XVI, 149, 1882.

46. Schieman: De Digitali purpurea. Diss. Inaug. Med. Göttingen 1786.

*47. Schliomensum: Über die Bindungsverhältnisse zwischen Herzmuskel und Digitalis. Arch. f. exp. Pathol. LXIII, 295, 1910.

*48. Stowers: The Pharmacological Action of Atropine on Cardiac Muscle. Thesis. University of Missouri, 1911.

49. Thomson: Über die Beeinflussung der peripher-

ischen Gefässe durch pharmacologische Agentien. Petersburg. med. Wochenschr. XII, 1887. Schmidt's Jahrb. CCXV, 235.

50. Traube: Gesammelte Beiträge zur Pathologie und Physiologie, 1871, I, 190, 252, and II 907.

51. v. Lhota: Untersuchungen über die chronische Vergiftung mit Digitoxin und Digitalis. Arch. Internat. de Pharcodyn. et de Therapie. XX, 369, 1911.

52. Versuche über Angewöhnung an Digitoxin und Digitalis. Arch. Internat. de Pharmacodyn. et de Therapie. XX, 451, 1911.

53. Williams: Über die Ursache der Blutdrucksteigerung bei der Digitalinwirkung. Arch. f. exp. Path. u. Pharmak. XIII, 1, 1880.

54. Winogradoff: Über die Einwirkung des Digitalins auf den Stoffwechsel und auf den mittleren Blutdruck in den Arterien. Virchow's Archiv. XXII, 457, 1861.

55. Withering: An Account of the Foxglove and some of its Medical Uses; with Practical Remarks on Dropsy and other Diseases. Birmingham, 1785.