YOHIMBINE

A CONTRIBUTION TO THE STUDY

OF

NARCOTIC AGENTS

with an appendix on

PHOTOACTIVE CHANGES IN YOHIMBINE SOLUTIONS.

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by

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C O N T E N T S.

I. GENERAL FACTS REGARDING NARCOSIS.

A. The Theory of Meyer and Overton.

The pharmacology of narcotic drugs has of late years greatly increased in interest. The reason is that we are now for the first time in a position to unravel the physical or chemical process by which they exert their influence. All narcotics are substances that have a selective action on the nervous system. This action consists in a depression of the excitability of the higher nerve centres, while in greater concentration they interfere with the working of reflexes and even with simple nerve conduction. A revival in the purely pharmacological interest of these substances occurred about ten years ago, owing to the work of Hans Meyer (99, A., B.), (OI) and E. Overton (OI).

It has long been known that the nervous system is characterised by the presence of large amounts of substances such as lecithin and cholesterin, some of which, though chemically not fats, resemble them in physical properties. All these fatty-like substances, many of them of undefined chemical constitution, have received from Overton the name"lipoids". The discovery of Meyer and Overton is that narcotics are all markedly lipoid-soluble, or, what is the same thing, lipoid-solvent. When we compare their solubil-

ity in water with their solubility in lecithin or cholesterin we find that they dissolve more readily in the latter. Furthermore, in proportion as their solubility in lipoid substances oversteps their solubility in water, so are they powerful narcotics.

These lipoid substances have been found to be universally distributed throughout the animal and the vegetable kingdom. They seem to be an essential constituent of all cells, though some cells contain them in greater abundance than others. When a narcotic is carried by the blood of a multicellular animal it no doubt accumulates in the lipoid constituents of the cells to an extent determined by the partition-coefficient of the narcotic in lipoid and in the non-lipoid constituents of the tissues (approximately ~~equal to~~ the blood-plasma). Given a definite quantity of any special narcotic to be distributed throughout the organism, the higher this partition-coefficient (i.e. the more soluble the special narcotic in lipoid as compared, say, with plasma) the greater will be the relative ~~accumulation~~ *accumulation* of the narcotic in the lipoid constituents of the cells. This hypothesis was confirmed by Meyer and Overton by a careful series of experiments in which the partition-coefficient of various narcotic substances in oil and in water ~~respectively~~, was compared with their relative power in inducing narcosis.

As stated before, the nervous system is preeminent among the tissues of the body in containing a large amount of lipoid, and so is explained the selective action of narcotics on the nervous system.

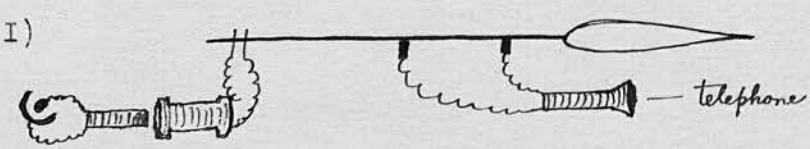
The "theory" elaborated by Meyer and Overton attributes the phenomenon of narcosis entirely to a solubility reaction. It may therefore be said to be a physical or physico-chemical theory. In itself however it does not furnish any final explanation of the phenomenon. It merely indicates in what direction one should look for an ultimately satisfying theory of narcosis. Attempts have been made to utilise this discovery in order to construct a general theory of the phenomenon, but we shall be ~~ina~~ in a better position to appreciate the bearing of these attempts when we have discussed certain work which has lately been carried out in the physiology and pharmacology of nerve fibres.

B. Action of Narcotics on Medullated Nerve.

Medullated nerve, e. g. from the frog, forms an excellent medium on which to study the action of narcotics. In the myelin sheath there is a large amount of lipoid, a fact which no doubt accounts for the prominent action exercised by narcotics on nervous trunks. The nerve itself can be readily isolated and its reactions, which are relatively simple, can be studied with more precision than in the case of any other tissue.

About the time that Meyer and Overton were carrying out their work, Wedensky of St. Petersburg (00), (03) was engaged in a detailed study of the modifications in nerve function produced by narcotic drugs. He devised a new method for studying the electrical reactions of nerve, and the results obtained in the elaboration of this method led to an important discovery.

Each impulse that travels along a nerve is accompanied by an electrical change. It occurred to Wedensky that a number of electrical changes transmitted in succession along a normal nerve might affect a telephone; and indeed he found that when a telephone was connected, as in the accompanying diagram, (Fig. I)



by means of two electrodes applied to the side of the

nerve, stimulation of the tissue, whether by a rapid succession of induction shocks or by a regular series of mechanical taps, produced in the telephone an audible note, which corresponded exactly in pitch with the rate of mechanical or of electrical stimulation. Naturally the greatest rate of stimulation could be obtained by means of electricity, and with the highest rates at his disposal - 200 to 300 per second - a clear musical tone of a corresponding pitch was given out by the telephone so long as it remained in connection with the nerve. It was evident therefore that a nerve may transmit a very rapid succession of individual and distinct impulses.

One advantage of this telephonic method of studying electrical reactions is that the nerve does not require to be cut away from the attached muscle. It suffices simply to place the electrodes some little distance apart <u>on the side of</u> the nerve, whereas in almost all other methods of recording electrical change one of the electrodes must be applied to a freshly cut section.[1] It follows that by the Wedensky method muscle records may be obtained at the same time as the telephone is acting, and the two records may be

[1] This suggested to me an important experiment. The fact that the telephone is worked by the nerve proves that energy may be tapped from the side of the structure during function. Does this withdrawal of electrical energy during function diminish the effectiveness of the transmitted impulse in exciting the muscle? Experiment showed that it does not. In the body there must therefore be a constant leakage of electrical current from active nerves, muscles, glands etc. for the organs in each case lie virtually in a

compared with each other. On the other hand the method has this limitation, that it can be used only with rapid rhythmical stimulation, or with "tetanisation" of the nerve. It is useless for studying the electrical change accompanying one single nervous impulse.

Having devised this beautiful method Wedensky proceeded to test with its help the action of certain drugs on nerve, and he began with members of the ~~anaesthetic~~ narcotic group. Now, a large amount of work had already been done on this subject, and, as is usual where complete knowledge is not available, a somewhat acrimonious controversy had arisen in regard to it. Without going into the details of the controversy I shall state the point at issue.

Schiff (58) had first pointed out that nerve possesses two properties, which, theoretically at least, may be considered distinct. These are excitability and conductivity. By excitability is meant that property in virtue of which a stimulus applied anywhere along the side of the nerve throws the internal living mechanism into action. By conductivity we understand the property whereby an active process once set up in any minute section of the nerve excites a similar process in the next adjoining section and so on. There is no reason to infer a

bath of electrolyte. The experiment does not prove, as I was at first inclined to suppose, that the electrical change in a nerve is a mere bye-product as it were, manufactured during transmission of an impulse, but in itself inessential for causing excitation.

priori from the normal working of nerve in the bodily economy that such a property as excitability is possessed by nerve, for in the living body stimuli reach the nerve only from one or other end i.e. from the attached nerve-cell or sensory end-organ, and never from the side. It is only by direct experiment that we become aware that a nerve is excitable all along its length. Conductivity is the only property of nerve which we recognise to be essential to it. From the present outlook of nerve physiology we are experimentally concerned only with these two properties of the structure.

When the action of anaesthetics on nerve began to be studied - Grűnhagen (72), Szpilman and Luchsinger (8I), Efron (85), Hirschberg (86), Gad (88) (89), Piotrowski (93), Waller (96), Gotch (00) etc. - it was recognised that excitability and conductivity may apparently vary independently of each other, and it was an acutely debated question whether they are distinct. We shall better understand the point at issue if we use an analogy.

Imagine a long wooden trough, V-shaped on section, such as is used for feeding poultry. If a row of tennis-balls all in close contact is placed in the bottom of the trough, we have in such a system a rough model of a nerve. A tap applied to either of the two end balls of the row is transmitted in succession along the series to the opposite end: the system in other words possesses conductivity. Similarly a tap applied from above to any ball in the interior of

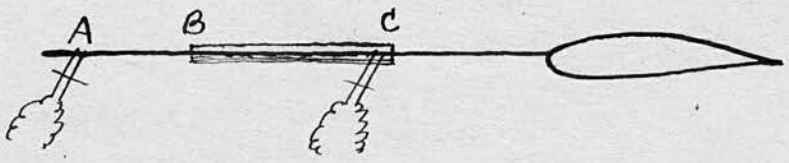


Fig. 2.

The middle portion BC of the nerve is narcotised. Stimulation anywhere over this area requires to be strong in order to excite.

Meantime however a light stimulus applied to AB seems to be readily conducted ~~through~~ the narcotised portion BC.

the row will likewise set the system into action, the impulse being conducted away from the site of "stimulation". Had the whole trough however been covered over with a plank of wood laid longitudinally along the top, it would have been impossible to excite the internal system of balls by tapping the structure anywhere except at the two open ends. The chain of balls is now enclosed in a framework which protects it from mechanical stimulation applied along its length. Its excitability from the side is gone.

Instead of a plank of wood we might have used a layer of some less rigid material to cover the balls. In this case its excitability along its length would be diminished without being absolutely abolished, and it is easy to see that such a material might be applied without interfering in any way with the conductivity from end to end.

Now, experiments with narcotics applied to a localised part of a nerve, say the middle, seemed to show that the conductivity through this portion is not interfered with at a time when its excitability, as tested by induction shocks applied to the narcotised area is considerably depressed. In the accompanying illustration - Fig. 2 - the narcotised portion - BC - would require an induction shock of say 100 mm. distance to excite it, whereas previous to narcosis it had reacted to a shock of 300 mm. distance. At this time however stimulation anywhere AB, the proximal non-narcotised part of the nerve, at a distance of 300 mm. (the original maximal stimulus) would ~~give~~ continue to give

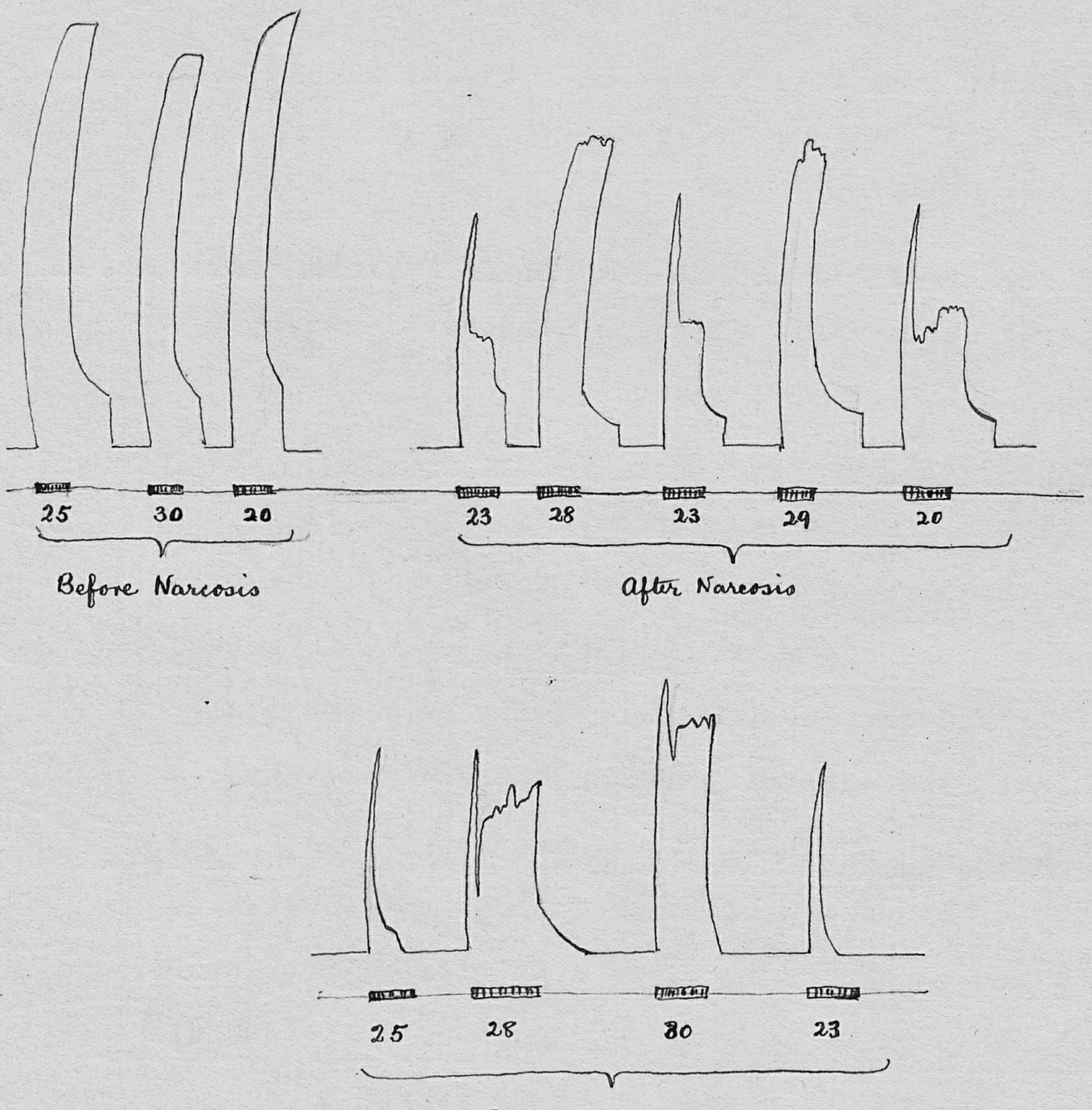
a full maximal contraction of the attached muscle. Apparently conductivity is not depressed at a time when excitability is much diminished, All these experiments were carried out with single induction shocks applied at irregular and varying intervals to the nerve.[1]

We return now to Wedensky's results with the telephone method as applied to narcotised nerve; and we shall confine our attention simply to the observations on conductivity made by him. He painted a portion of the nerve with solutions of cocaine, chloral hydrate or phenol, or subjected it to the action of chloroform or of ether vapour. To detect possible alterations in conductivity he rhythmically stimulated the proximal, non-narcotised portion at a rapid rate and used both the telephone (applied to the distal non-narcotised portion) and the muscle to give a record of the impulses transmitted through the narcotised part. In this way he found that conductivity begins to alter from the very start.

The telephone note, instead of being of uniform pitch and intensity, became weak, dull, and

[1] It should be understood that these and the following results are true only during the preliminary action of the narcotic. Complete narcosis implies total absence of conductivity, but the essential action of narcotics is best studied during the preliminary stage, when function is altered though not abolished.

Fig. 3.



Note that during narcosis the weakest tetani follow upon the strongest stimulations.

In late narcosis strong rhythmical stimulation produces one single twitch of the muscle.

(These curves are copied from Wedensky's paper.)

mixed up with extraneous noises. This change was a progressive one, becoming more and more pronounced as the narcosis went on, and yet the nerve when tested by single induction shocks with the muscle as indicator seemed to have unimpaired conductivity. Turning to the muscle record corresponding to these telephonic sounds, he found that the contractions were not so high ~~or~~ nor so smooth-topped as before narcosis. Strangely enough the tetani elicited by strong rhythmical stimulation were weaker than those elicited by moderate stimulation at the same rate, and further, when the strength of stimulation was kept constant, and only the rate of stimulation varied, the tetanic responses corresponding to rapid stimulation were weaker and more irregular than those corresponding to slow stimulation. It seemed as if the narcotised nerve became more easily thrown out of gear when it had to do individual acts of heavy work, or light work at a rapid rate (Fig. 3).

On further progress of the narcosis he found that with constant rate of excitation, strong tetanisation produced only an initial twitch of the muscle, whilst rhythmical stimulation at moderate intensity caused tetanus. Similarly when the strength of stimulation was kept constant, a single initial twitch was obtained with a rapid rate of stimulation, and a tetanus when the rate was slow. The telephonic record corresponded exactly: with constant rate, strong tetanisation caused either no sound at all or an initial "Knack" followed by silence; moderate tetanisation caused a weak and not quite pure tone, but one

that corresponded unmistakeably to the rate of stimulation used. (See too the illustration facing page 22.)

This remarkable and perfect parallelism between the records of the muscle and the telephone naturally surprised Wedensky. By poisoning the nerve close to the muscle or by cutting it across he made sure that the telephone sound was no backwardly propagated effect from the muscle itself,. and he further controlled his results by other electrical methods - galvanometer. He was consequently led to conclude that a stretch of narcotised nerve begins to alter as regards its conductivity much sooner than was formerly supposed. According to him a"paradoxical modification" of conductivity is established, in that rhythmical tetanisations are not transmitted regularly, strong impulses being transmitted with greater and greater difficulty at a time when weak excitations are handed on without prominent change.

These results of Wedensky are of high importance. They show that in testing the action of narcotics or of other reagents and influences on nerve, it is not sufficient to excite the structure with single induction shocks and use the height of the corresponding maximal response as an index of the state of the conductivity. To test conductivity properly it is necessary to work the nerve at a rapid rate. Incidentally it may be mentioned that Wedensky claimed his results as a proof that conductivity and excitability in nerve are one and the same thing,. for, as he had found, no depression of excitability occurs

without a corresponding depression of conductivity. Lastly, in the case of all the reagents whose action on nerve he tested - and they all happened to belong to the narcotic group - the result was identical. Wedensky as a matter of fact declared that all substances which have any action on nerve affect its function in the same way, by causing in every case a highly characteristic and readily recognisable alteration of function.

[The phenomenon whereby conduction in a partially narcotised nerve ceases when the rhythmical stimulation is strong or rapid, or, better still, both combined, - a certain definite frequency and a certain definite strength of stimulation being necessary at each successive stage of narcosis to produce absence of conductivity - suggests a possible explanation of a fact well-authenticated but otherwise difficult to explain. It is known that local application of cocaine to a mixed nerve first paralyses the sensory and, at a later stage, the motor fibres. Furthermore, different qualities of sensations are paralysed at different times; thus, sensations of pain and of touch are paralysed at an early stage, while sensations of heat and of cold may long persist. If we assume that sensory impulses are of a higher order of frequency than motor impulses, and that of the sensory impulses, those of touch and of pain have the highest frequency, we have in the discovery of Wedensky a feasible explanation of this otherwise obscure phenomenon.]

C. The Work of the Göttingen School on Asphyxia and Narcosis of Nerve.

Meantime an entirely independent series of investigations on nerve had been going on in Verworn's laboratory in Göttingen. These, which were at first directed towards a different object, came eventually to have a close relation to Wedensky's work.

Verworn (00) had begun an investigation into the metabolic processes of nerve-cells, from which he had concluded that abundant O_2-supply is essential to the continued function of these cells. Winterstein (02) then studied the extent to which the two phases of metabolism, assimilation and dissimilation (with regard to oxygen) are affected by narcosis with ether, chloroform, alcohol etc. It had long been known that narcosis interferes with dissimilation generally. Winterstein showed that narcosis hinders assisilation of O_2 to exactly the same extent as it hinders dissimilation.

Verworn's results in regard to the metabolism of oxygen by nerve cells had suggested to him however that one might succeed in asphyxiating a nerve trunk by withdrawal of oxygen,. Under his direction v. Baeyer (02) succeeded in abolishing the conductivity of a nerve by keeping a portion of it subjected for some hours to the influence of an oxygen-free neutral gas (nitrogen, hydrogen). When oxygen gas was once more supplied to the nerve, recovery took place in a

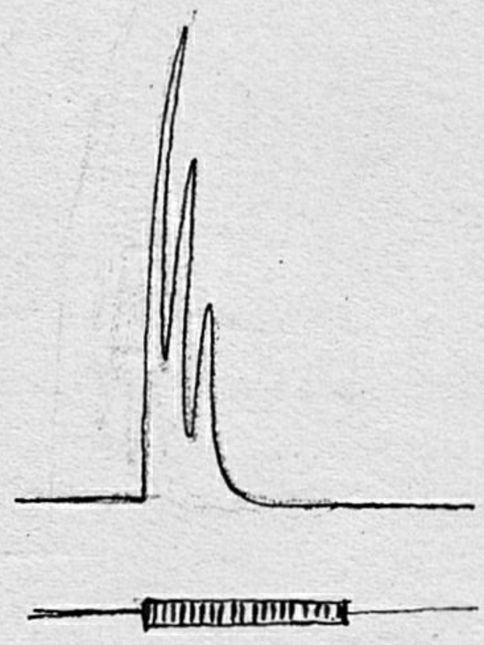
very short time. Von Baeyer was unable however to show that oxygen is consumed by the nerve during activity. Fröhlich (03) then showed that narcosis, while having no influence on the outward diffusion of oxygen from a nerve in an oxygen-free atmosphere, prevents the subsequent assimilation of oxygen once the nerve has become asphyxiated.

This latter observer then studied (04, A. B.) more carefully than had previously been done the whole question of narcosis and of asphyxia of nerve. He found by Wedensky's method of rapid rhythmical stimulation that asphyxia affects a nerve in exactly the same way as narcosis with ordinary narcotic agents, the only difference being in the time required to produce the various phenomena observed by Wedensky, asphyxia being a much slower process than narcosis in the relation roughly of IO : I. By using the more deliberate asphyxiation method he was enabled to study all the phenomena observed by Wedensky and previous ~~obv~~ investigators more carefully than had before been possible, and he enormously extended the bearing of Wedensky's results. It is unnecessary here to discuss all the work that Fröhlich did in this connection. Much of it is now the common property of physiologists and of pharmacologists. Suffice it to say that he explained Wedensky's results in a very satisfactory manner. According to Fröhlich, narcosis, or asphyxia, as the case may be, slows many of the processes that occur in a nerve. Thus, when an impulse is transmitted along a narcotised or asphyxiated

nerve the rate of propagation is cut down. The slowing of the impulse is confined to the affected portion; on passing through it the impulse once more resumes its normal speed - Fröhlich (04,C), Boruttau (01), Boruttau and Fröhlich (04). Again Gotch and Burch (99) had pointed out that a nerve, like the heart, possesses a refractory period, only the refractory period of nerve is out of all proportion shorter than that of the heart. When two stimuli are applied in rapid succession (say, I - 1000th second interval) to a nerve, the second may or may not be transmitted according to the individual nerve used. When the second stimulus is not transmitted, it is said to fall within the refractory period of the first. Now Fröhlich showed that asphyxia greatly prolongs the refractory period of all nerves. The same fact had been previously demonstrated by Boruttau (00) for narcotised nerve. The refractory period must in part at least be looked upon as a period of recovery from previous katabolism or disintegration, and by stating that the refractory phase is prolonged by asphyxia, Fröhlich meant that the normal reintegration succeeding upon disintegration is slowed.

By means of the prolongation of the refractory period he explained Wedensky's results. In some cases Wedensky had found that the only muscular response to tetanisation of a nerve of which the middle portion is narcotised, is a single initial twitch. Fröhlich pointed out that this twitch is not a summated one, but simply a single maximal twitch. It is a

Fig. 4.



A fatigue tetanus. Note that the muscle ceases to respond before the stimulation of the nerve ends.

The tetanus instead of rising consistently falls off in height.

response merely to the first stimulus of the series, the succeeding stimuli having failed to be transmitted. By control experiments with pairs of stimuli separated by varying intervals, he showed that this peculiar effect is produced only when the interval between the successive stimuli in the rhythmical series is less than the refractory period of the narcotised nerve. Now, as Wedensky had found, this initial twitch is more likely to occur when the serial stimulation is strong than when it is weak. Fröhlich accounted for this on the supposition that strong stimuli give rise to a greater amount of disintegration in the nerve, with consequent necessity for a longer period for restoration.

Furthermore Fröhlich showed that ~~anaesthet-~~ narcotised or asphyxiated nerve is readily fatigued. By rhythmical stimulation at such a rate that the interval between each successive stimulus is just greater than the refractory period (corresponding to that special stage of narcosis or asphyxia), he obtained a tetanus of the form represented in Fig.4. This tetanus consistently falls off in height and finally ceases. It is evident that with each successive stimulation of the series the refractory period gets longer and longer, in other words the nerve functions worse and worse <u>because of</u> continued activity. Interruption of the stimulation but for a fraction of a second restores the nerve to its former degree of conductivity. These results explain why previous observers had failed to fatigue nerve.

Without the help of some method of artificially prolonging the refractory period it is difficult to attain such a rapid rate of stimulation as to wear out the nerve. After each single active process a normal nerve recovers with almost lightning rapidity, and the technical difficulties in obtaining a rate of stimulation comparable with the normal refractory period are great. Nevertheless Thörner (08) has since succeeded in fatiguing normal nerve, and has ~~thus~~ justified Fröhlich's statement with regard to it.

By means of ether narcosis Fröhlich found it possible to prolong the refractory period of nerve to I/I0th second, a prolongation of I00 times the normal refractory period.

All the work which has just been described was carried out on frog's nerve. It was interesting to know if mammalian nerve behaves similarly to nerve from cold-blooded animals. I undertook this investigation in conjunction with Fröhlich. To keep the muscle active it was necessary to operate on the living animal. All our attempts to asphyxiate the nerve by enclosing it in an oxygen-free atmosphere failed however. The failure we found to be due to the presence of a rich recurrent blood-supply derived from the attachment of the nerve to the muscle. When this blood-supply was interrupted the nerve ceased to conduct; when the blood-supply was readmitted conductivity immediately returned. It is probable that the nerves of warm-blooded animals require a <u>constant</u> oxygen supply to maintain function. Although we

failed to asphyxiate the blood-perfused nerve, we were able to narcotise it locally with ether or with chloroform vapour, and so produce typical Wedensky effects in the attached muscle - Frŏhlich and Tait (04).

Frŏhlich's work on narcosis of nerve has a twofold importance. In the first place he subjected to analysis Wedensky's "paradoxical modification of conductivity", and showed that a prolongation of refractory period is chiefly responsible for the peculiar phenomena produced. It is prolongation of the refractory period that makes the nerve incapable of transmitting in normal fashion a series of rapid rhythmical excitations, and fatigue may readily occur in this condition of the nerve. In the second place he showed that narcosis and asphyxia effect the nerve in exactly the same way. The multiform complexity of the Wedensky phenomena, varying in perfectly consistent fashion with (I) the strength of stimulation, (2) the rate of stimulation and (3) the degree of narcosis or asphyxia, makes it a most sensitive test by which to judge of the precise action of any reagent on nerve. Tested by this highly sensitive method, narcosis and asphyxia were found to be the same - a fact in itself most suggestive.

D. Narcosis and Oxygen Supply.

Verworn (09) summing up the results obtained by various workers in his laboratory on the relation of narcosis to oxygen supply, comes to the conclusion that the atomic groups of the cell which are charged with the function of transferring the oxygen, become bound or held by the narcotic, so that they are no longer capable of handing on or transmitting the normal supply from one locality to another. This conclusion is based on an elaborate summary of all the experimental facts relating to the oxygen metabolism of various tissues while under the influence of narcotics.

In an independent publication of the same year by Mansfeld (09), a still more specific theory of narcosis is propounded, in which the question of oxygen supply is definitely correlated with the Meyer-Overton theory. According to Mansfeld, it is the cell-lipoids that handle and transfer from ~~one~~ place to place inside the cell the oxygen required for cell function. He adduces many facts in support of this view. Thus both the central nervous system and the heart are organs that show great sensitiveness to want of oxygen, while on the other hand they possess a high content of lipoid material, which is strictly conserved even during starvation. Again previous experiments by Exner had shown that oxygen has a high partition-coefficient for fat and water, and Vernon (07) had discovered that fatty oils absorb from four to

five times as much oxygen as water. According to Mansfeld these ~~fats~~ facts indicate that the physiological importance of the lipoids consists in facilitating the entrance of oxygen from the tissue-fluid into the interior of the cells.

Now, from experiments in physics, it is known that every solution absorbs less gas than the solvent itself in a state of purity. If a narcotic has penetrated into the cell the oxygen no longer finds a pure solvent but a lipoid-narcotic solution, and this can absorb all the less oxygen in proportion as the solution of narcotic is concentrated; thus the normal solvent activity of the lipoid for oxygen is diminished and narcosis ensues. From this point of view therefore, narcosis is identical with a partial deficiency of oxygen supply.

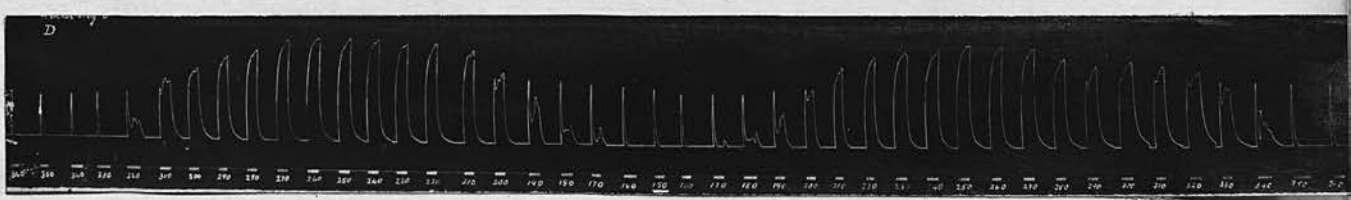
Mansfeld goes on to quote certain experiments of Paul Bert (70), which demonstrated that when a partial pressure of oxygen in the atmosphere surrounding an animal is very gradually reduced, the animal dies in a narcotic condition and does not show the ordinary convulsions which accompany the rapid removal of oxygen. As a further illustration of the same fact I might refer to experiments by Haldane (95), in which it was shown that mere oxygen withdrawal, apart from accumulation of carbonic acid gas, produces unconsciousness without any motor disturbance. Compare too the experiments of ~~Jerusalem~~ Kaya and Starling (09) on "Asphyxia in the Spinal Animal".

Mansfeld then seeks to support his hypothesis

by definite experiments. While giving support to his view, these experiments cannot however be said to prove it. He returns to the subject in a later paper (10) bringing fresh evidence in support of it. Mansfeld's view may at least be accepted as a convenient working hypothesis to investigate the close relation that exists between narcosis and oxygen deficiency.

The same plausible hypothesis had been independently arrived at by W. Cramer of the Physiology Department in Edinburgh University. Cramer had found during certain experiments with lecithin that the substance can occlude or absorb large quantities of oxygen gas. Cramer and I carried out a number of experiments on red blood corpuscles to test Mansfeld's hypothesis relating to narcosis. It is a well-known fact that chloroform or ether is largely carried in the blood by the red blood corpuscles. The high lipoid content of the stroma would account for this phenomenon. Now, supposing the stroma of the deoxygenated red corpuscles to contain a considerable quantity of narcotic in solution, it would transmit oxygen from the plasma to the haemoglobin inside at a slower rate than usual. We extracted the oxygen from equal quantities of blood, of which we subsequently chloroformed one sample. We then exposed the two samples, the one chloroformed, the other not, to a current of oxygen gas, and sought to determine from the rapidity of the colour change, the rate at which the haemoglobin became saturated with oxygen. The experiments however failed to yield any decisive result, because of the difficulty of bubbling the oxygen gas through the two specimens at an equal rate.

Fig. 5.



Photograph of a tracing taken by me to show the Wedensky effect due to cooling of a nerve.

The rate of stimulation was 100 per second. The drum was stopped after each short period of stimulation ceased. Three centimetres of the middle portion of the nerve was cooled to 0°C.

From the left hand side to the middle of the tracing the strength of stimulation measures regularly by 10 mm distance at a time from 360 to 150. From the middle to the right hand end it decreases again by similar steps.

Note that some of the worst developed tetani occur with strong stimulation

Fatigue tetani are seen at the distances 170, 180 and 190.

E. The Action of Low Temperature.

When Gotch and Burch (99) discovered the refractory period of nerve, they also found that it may be greatly prolonged by cooling. Their work was done entirely on the electrical response, but Boycott (99) confirmed their results by means of the muscle record. Now, when Fröhlich had succeeded in explaining the Wedensky phenomena in terms of prolongation of refractory phase, it became of interest to know if the Wedensky phenomena may be produced by local cooling of a nerve. This research I commenced in Göttingen and continued for over a year in Edinburgh. The results, which are as yet only partially published, - Tait (06) (08): compare also my D.Sc. thesis, "Reactions of Cooled Nerve", 1907, in Edinburgh University library - amply showed that low temperature acts on a nerve in precisely the same fashion as asphyxia or narcosis with chloroform, ether, cocaine etc.

By carefully graduating the low temperature to which the nerve was subjected it was found possible to maintain the nerve in one definite stage of depression for a prolonged period of time and thus to study its reactions much more leisurely and exactly than is possible when either narcosis or asphyxia (both of them difficult to control) is used to modify its reactions. An example of the regular type of tracings obtained is shown on the opposite page. (This record incidently serves as an additional illustration of the effects grouped under the heading of "the Wedensky phenomena").

Fatigue of the nerve could likewise be readily studied, and as no chemical treatment of the nerve was employed to facilitate the production of fatigue, it was obvious that Fröhlich's explanation of the ill-success of previous investigators in demonstrating fatigue of nerve, was correct. All that is necessary to fatigue nerve at room temperature is a sufficiently rapid rate of stimulation. In this investigation a considerable knowledge of the technique of experiments on nerve was ~~requored~~. acquired.

As said above the results showed that low temperature acts on nerve in precisely the same manner as narcosis or asphyxia. From the number of different influences that affect nerve in the same way, one might be tempted to conclude, as Wedensky originally did, that all influences which cause depression of nerve conduction act in the same way. Thus the ordinary narcotics, ether, chloroform, chloral hydrate, cocaine etc., all neutral gases which produce asphyxia e.g. nitrogen, hydrogen, carbon monoxide, and simple depression of temperature produce a complex chain of phenomena which are precisely the same in every case. I even found that solution of curara, after some eight or ten hours application produces typical Wedensky effects on the nerve.

There were however on record certain experiments by Waller (99A.,B.)(00) on the action of protoveratrine and of aconitine which might have warned against a ~~rapid-fer~~ hasty generalisation of this nature. I have also from time to time tested the

action of a number of reagents, mostly alkaloids, e.g. quinine, veratrine, strychnine, saponin on nerve. These experiments have shown that various alterations in conductivity differing widely from those described by Wedensky are produced by different reagents.

These results serve only to heighten the interest of the discovery that low temperature, asphyxia and narcosis (with narcotics of the Meyer-Overton type) are identical in their action on nerve. There must be a common process involved in the action of all three. We have already discussed Verworn's conclusions and Mansfeld's hypothesis regarding the relation of oxygen-deficiency to narcosis. A moment's consideration will show that low temperature produces effects on an animal closely similar to narcosis at least.

When a warm-blooded animal hibernates it becomes for the time being cold-blooded, and, instead of maintaining a constant temperature, takes on a temperature approximately the same as that of the surrounding medium i.e. much lower than its ordinary temperature. The animal seems to sleep. Sensation and volition are suspended. At the same time certain reflex movements remain active. Thus Marshall Hall found that a touch applied to the spines of the hedgehog causes it to draw a deep inspiration, a phenomenon that remains present in the narcotised animal. The corneal reflex persists. The frequency of respiration is diminished, and the rhythm is often of the Cheyne-Stokes type, a phenomenon familiar in narcosis with morphia or cocaine - cf. Cushny (10) -

and due in all probability as Haldane and Douglas (09) have shown, to oxygen deficiency in the respiratory centre. The force and frequency of the heart beat likewise becomes reduced, as in narcosis. Apart from the difference in temperature the general symptoms observable in hibernation are closely similar to those seen in narcosis. It might be mentioned too that in both cases the shivering reflex is in abeyance.

Taking all these facts into consideration the statement may be hazarded that both narcosis (with narcotics of the Meyer-Overton group) and low temperature act by restraining the consumption of oxygen in the cells of the body. Now, experiments carried out by directly withholding oxygen from tissues, have repeatedly shown the immediate necessity for oxygen in all changes of katabolic nature in the cells - cf. Verworn (09). Without oxygen there can be no energy output by the living cells of the body, and this holds true when the oxygen has been simply withdrawn by outward diffusion, the cells meantime remaining inactive, and therefore in all other respects charged with energy-producing materials. By simple oxygen-deficiency apart from any consumption of material through activity, it is possible to paralyse the cell completely. On the other hand it has also been shown that ~~oxygen~~ the immediate presence of oxygen is necessary for anabolism after normal activity. If then it is true that narcotics (of the Meyer-Overton group) and low temperature act by restraining the supply or the consumption of oxygen they may both be said to paralyse katabolism and to paralyse anabolism.

II. EXPERIMENTS WITH YOHIMBINE.

A. Nervous System.

Experiments with yohimbine soon showed that it exerts a marked depressant action on the nervous system preceded by symptoms of apparent excitation - Oberwarth (98), Muller (67), Gunn (08, A.,B.). When toxic doses are given to a mammal, the animal after a preliminary period of restlessness, develops paralysis of voluntary movement and finally fails to react to sensory stimulation, although it may remain alive in this condition for a long time. In the frog, as Gunn (08,B.) has shown, there is present at a certain stage abnormal excitation of certain reflex movements, a condition which he could demonstrate to be due to an initial depression of the higher nerve centres. The motor symptoms present in all animals after toxic doses resemble those seen in the disease myasthenia gravis - Gunn (08,C.). When total paralysis of the central nervous system is present, the peripheral nerves still maintain their function to a large extent. It is thus evident that yohimbine has a selective action on the nervous system, producing depression thereof.

Magnani (02),(03) discovered that yohimbine is locally anaesthetic when applied to the cornea, and it is in vogue for this purpose in ophthalmacological

practice. Loewy and Muller (03) discovered that it exercises on isolated nerve an action analogous to that of cocaine, producing complete absence of sensory and of motor conduction. By means of it they were able to abolish conductivity in the vagus nerve, of the inhibitory fibres to the heart and of the afferent fibres to the respiratory centre. In the case of living mammals, the anaesthesia of the nerves was temporary and was followed by complete recovery to the normal state. Applied to the mucous membrane of the tongue, yohimbine produced paralysis of certain taste sensations.

Gunn and I then investigated the action of yohimbine on the motor nerves of the frog. (See No I. of the printed communications appended to this essay). By stimulating the nerve in rhythmical fashion after the method of Wedensky we soon found that, while in certain respects yohimbine resembles in action the ordinary narcotics such as ether, chloroform, cocaine etc., it nevertheless differs from them in important particulars. The various points of resemblance and of dissimilarity we expressed for the most part in reference to and in terms of the phenomena described by Wedensky, for at that time we were unable to analyse in complete fashion the modifications in conductivity produced by the drug. We found that fatigue of the nerve is induced with extreme readiness when it is under the influence of yohimbine, and that the fatigue may last for more than five seconds, while the refractory period, which by Frohlich had been prolonged to a maximum of .I second, we prolonged to .25 second. In a subsequent paper (No.2

of appended publications), I attempted a more precise analysis of the effect of yohimbine on the nerve.

I shall now state in general terms the result of this work on yohimbinised nerve. My own work on cooled nerve had shown that the effects described by Wedensky and by Frohlich as due to narcosis with ordinary narcotics, may be precisely simulated by merely lowering the temperature; in other words the action of ordinary narcotics is equivalent simply to a fall of temperature. Now, low temperature of itself does not necessarily imply any specific alteration in the mode of function of a part. We believe rather that by low temperature each of the individual chemical processes occurring in a tissue are regularly and equably slowed without any of them suffering an abrupt change at any one point. There are good grounds for this belief so long as the change in temperature lies within what may be considered physiological limits.

Experiments in physical chemistry have brought to light the fact that the velocity of a large number of chemical reactions selected at random is increased from two to three times by a rise in temperature of 10^{0} C. The extent to which the velocity of any chemical or physical reaction is altered by a rise of 10^{0} C. in temperature is sometimes spoken of as the "temperature-coefficient" of the reaction; we may therefore convey the meaning of the previous sentence by saying that the temperature-coefficient of chemical reactions in general lies between ~~two~~ 2 and ~~three~~ 3. Of late years a considerable amount of work has been carried out in determining the temperature-coefficients

of certain physiological reactions, and in a great many cases the coefficient has been found to correspond to that of ordinary chemical reactions. Thus, to take an example from our present subject, Snyder(08) found that a rise of 10^{0} C. increases the rate of propagation of an impulse along a nerve from two to three times. Many other similar examples, including the frequency of the heart-beat, might be quoted as showing that the temperature-coefficient of physiological reactions is the same as that of ordinary chemical reactions.

Now, in the activity associated with the propagation of a nervous impulse, we recognise two differently directed phases. There is first of all a katabolic or disintegrative phase, on which immediate function is presumably based, and secondly there is a succeeding anabolic or reintegrative phase, which brings the nerve back to its previous condition so as to be ready for carrying another process when necessary. For both of these acts, as the Göttingen school has shown, oxygen supply is equally necessary; in other words, they are both chemical processes, and the presumption is that if one is slowed by low temperature, the other is slowed to an exactly corresponding degree. As a matter of fact Frohlich's experiments on the rate of propagation of the nervous impulse and on the prolongation of the refractory phase in corresponding stages of narcosis, bear out these statements exactly. Ordinary narcotic agents equally retard the process of disintegration and that of reintegration.

We come now to the action of yohimbine. This may be expressed in one word by saying that it

slows the process of anabolism or reintegration to an extent out of all proportion great compared with the slowing of katabolism or disintegration produced by it. Gunn and I found that yohimbinised nerve shows the phenomenon of fatigue in far more decided fashion than nerve narcotised by ordinary reagents. In my own experiments I found that a nerve subjected to a spell of rapidly repeated excitations may continue for thirty seconds incapable of its previous degree of functional activity: even the transmission of one single impulse is followed by impairment of function lasting for more than two seconds. When we consider that an ordinary nerve at room temperature is, I/1000 second after the transmission of a first impulse, ready to transmit another, and that Frohlich with great difficulty succeeded by means of chloroform or ether in prolonging this interval of functional depression due to activity to I/10 second, we realise to what an enormous extent yohimbine delays anabolism. On the other hand, the interference with katabolism produced by yohimbine is relatively slight. I carried out a large number of experiments to determine if yohimbine slows the rate of propagation of the impulse in a nerve. Provided the nerve was not fatigued by the passage of an immediately previous impulse, no direct slowing of the impulse due to the drug could be demonstrated.

This specific depression of anabolism after activity is manifested by yohimbine not merely on nerve fibres, but also on the central nervous system. As was pointed out before, the peripheral muscular mechanism of an animal poisoned by injection of the drug

remains capable of almost complete function when reflexes and other central reactions are abolished. This shows that the central action, of whatever nature, is more pronounced than the peripheral. But this central action, though differing in degree, is, as Oberwarth (98), Muller (07) and Gunn (08) have shown, precisely analogous to the action on nerve fibres. The poisoned animal reacts readily to stimulation at first, but each succeeding response gets progressively more feeble, until paralysis ~~int~~ supervenes. After a long period of rest from stimulation, partial recovery occurs and the animal shows the same series of phenomena again. As Gunn has shown, there is a close analogy between yohimbinisation and myasthenia gravis.

In its selective action on the central nervous system and in its subsequent action on peripheral nerves, yohimbine therefore shows a close correspondence with ordinary narcotics. Its mode of action is however different, in that it affects anabolism rather than katabolism.

The next question is: How does it interfere with anabolism? As we have seen, the ordinary narcotic agents probably act by interfering with the supply of oxygen. The action of yohimbine must be different. We perhaps have a clue to the nature of this action in the effect produced by yohimbine on the respiratory centre. Müller first called attention to the fact that yohimbine stimulates respiration, and Gunn showed that this stimulation consists not only in an exaggerated frequency of respiration but in a greater amplitude both of the expiratory and of the inspiratory move-

ments. If one were asked to mention any other substance that produces precisely this effect on the respiratory centre, one would immediately reply, carbon dioxide,. With the solitary exception of protoveratrine, which as we shall see is closely analogous to yohimbine, carbon dioxide is the only other known substance that produces this effect.[1]

When we consider once again the facts relating to the action on nerve fibres of ordinary narcotic substances as contrasted with yohimbine, we see that the fatigue produced with the help of the former is due mainly to an all-round slowing of function, and is, so to speak, a normal phenomenon, artificially brought into prominence by the relatively rapid rate at which the tissue is worked. The fatigue under yohimbinisation is on the other hand a more specific expression of exhaustion depending on previous activity. Now, fatigue or exhaustion is recognised to depend on two principal causes: (I) the using-up of energy-producing materials, e.g. oxygen, and (2) the accumulation of waste products. The first condition may be rendered more imminent by artificially withholding energy materials. Narcosis with ordinary agents probably realises this condition and then, as we have seen, the tissue can neither katabolise nor anabolise at the ordinary rate. The second condition i.e. accumulation of waste products, can normally occur only after activity. Suppose that

[1] The action of acids on the respiratory centre is due to accumulation of carbon dioxide.

yohimbine interferes rather with the removal of these, and we can understand why its action should be exerted so prominently on the reintegration processes occurring after activity. Provisionally therefore, we may assume that yohimbine in some way inhibits the rapid removal of substances like carbon dioxide from the tissues and that part of its action is due to this cause.

This explanation however does not account for the whole of its action nor does it explain why it should exert a specific action on the nervous system. For one thing I found that a yohimbinised nerve, apart from any exercise of activity slowly and steadily loses its ability to function, and if sufficient time is given conductivity may entirely disappear, before any activity has been elicited by direct stimulation. (See No. 2 of appended publications). Whether yohimbine possesses all the properties of ordinary narcotics in addition to the property discussed above, I cannot as yet say. Nor do I know of any experiments devised to determine its partition-coefficient for oil and for water.

B. The Action of Yohimbine on the Heart.

In my experiments on yohimbinised nerve (see No.2 of appended papers) I was able to show that the refractory period of medullated nerve, like that of the heart, consists of two phases, an absolutely refractory phase and a relatively refractory phase. Yohimbine enormously prolongs the relatively refractory phase, and it was the great prolongation of this phase by the drug that first led me to recognise the existence of relatively refractory phase in nerve. This phase obviously corresponds to a process of recovery or of reintegration in the tissue, for during its continuance, function progressively improves.

The correspondence thus established in function between an ordinary nerve and the heart suggested that the action of yohimbine on the heart should be carefully worked out. No. 3 of the appended papers contains a research on this subject, and as the investigation is there set forth in detail, I shall content myself with summarising and commenting upon the results.

For purposes of physiological and pharmacological research, Engelmann had classified the heart properties into four groups: I) power of stimulus-production; 2) excitability; 3) conductivity; 4) contractility. According to him, these functions are all distinct, as evidenced by the varying degree to which they are influenced by the same physiological, and especially pharmacological influences. The fact that these functions are distinct, would imply that each has

its own physical basis or independent mechanism in the heart itself, and that these mechanisms may be for purposes of discussion dealt with as if they were spatially distinct, just as are the corresponding elements in the neuro-muscular mechanism for voluntary movement.

Experiment with yohimbine soon showed that the substance does not directly interfere with the contractility of the heart, nor in the first instance with the property of stimulus-production. What is interfered with is conductivity and excitability. Now Engelmann's separation of these two properties had been made at a time when physiologists believed that they are distinct. Boruttau and Fröhlich (04) however had subsequently shown that in nerve at least they are merely aspects of one and the same fundamental property. The action of yohimbine on the excitability and conductivity of the heart, simultaneous and equal at every stage, suggested to me that in this case too they may be aspects of one common property. I found, on reference to Engelmann's original papers, that his experimental evidence for their separation was inconclusive. In all the investigations on the action of drugs on the heart, where the effect on conductivity and on excitability were separately registered, the two varied together. Hence, until the contrary is proved, one is justified in considering excitation and conductivity in the heart as aspects of one common property. - the property of excito-conductivity.

Having reached this conclusion, it was easy to reconcile two seemingly independent theories that

have been put forward to account for toxic heart-block. According to the more prevalent theory, toxic heart-block is due to a defect in conduction from auricle to ventricle. The second theory, originally propounded by Straub, is that the condition may be due simply to a prolonged refractory period of the ventricle. I had shown that in the case of yohimbine and probably in that of all other substances known to prolong the refractory phase as a whole, the phase of the refractory period which is prolonged is the relatively refractory phase. As this phase is defined purely in terms of excitability, prolongation of it implies depression of excitability. If excitability and conductivity however always vary together, the one essentially depending on the other, then the two above-mentioned theories of heart-block are in reality the same.

I come now to a comparison of yohimbine with ordinary narcotic agents in regard to their action on the heart. Though the ordinary narcotics are not generally treated as having a group action on the heart, one may recognise common features in the action of all of them. As a rule they slow the rhythm of the heart. They all depress contractility. They do not produce heart-block, i.e. they do not interfere with excito-conductivity. From their inability to interfere with excito-conductivity, we might argue that the cardiac conducting mechanism is not similar to medullated nerve with its large content of lipoid, an inference that is confirmed by histological examination.

Now, within the limits of dosage just sufficient to stop the beat, yohimbine affects only the

excito-conductivity of the heart. The excitability to artificial stimulation is greatly reduced. Fatigue changes too are readily demonstrated with yohimbinised heart; these are traceable in every case to fatigue of conductivity.

It is of interest then to discover on further examination that the kind of interference exercised by yohimbine on the excito-conductivity of the heart is exactly similar to that exercised on nerve. It produces an effect of such a nature that the conducting mechanism is now able to undergo anabolism at a very much slower rate than normal. Whether the underlying cause is the same in both cases must be left for further investigation.

APPENDIX.

Photoactive Changes in Yohimbine Solutions.

While engaged in investigating the physiological action of yohimbine I noticed that solutions of the hydrochloride which had been left exposed to direct sunlight became yellow in colour, a change which did not occur in similar solutions kept in the dark. The solutions which underwent this colour change were found to possess no longer the characteristic action of yohimbine either on nerve or on the heart. On the other hand solutions of the substance which had been kept for months in the dark were as active as freshly prepared solutions of the same strength.

The medium used for dissolving the yohimbine hydrochloride consisted of sodium chloride (NaCl) .6gram potassium chloride (KCl) .042 gram, calcium chloride ($CaCl_2$) .024 gram, sodium hydrogen carbonate ($NaHCO_3$) .OIgram and distilled water IOO cc.. Experiments carried out with solutions of yohimbine hydrochloride in distilled water alone and in solutions of the above-mentioned salts by themselves showed that the photoactive change occurred in marked degree only in the presence of potassium chloride. Very slight change in colour occurring only after long exposure to sunlight in the presence both of sodium chloride and of sodium hydrogen carbonate were put down to possible contamination with potassium salt. In the presence of calcium chloride,

commercial samples of which are potassium-free, and in distilled water, no colour change occurred after months of exposure.

It seemed worth while to investigate the action of a more varied selection of both potassium and sodium salts as regards their possible effect in producing photoactive change in solutions of yohimbine hydrochloride. This work I carried out during the last winter with a student in St. Andrews University.

We selected at random a number of potassium and sodium salts and prepared N/10 solutions of these in distilled water. A definite quantity of each solution was then mixed with an equal volume of a I/2500 solution of yohimbine hydrochloride; the solutions were placed in corked test-tubes, and all exposed on Nov. 23rd in a window facing southwards. The amount of sunlight during the time of exposure was not measured. From time to time we made notes in respect to colour change occurring in the various tubes.

On Nov. 28th one of the solutions was faintly yellow, and on Dec. 5th this one was markedly deeper in tint and other four had become tinged, not equally so but in a certain order as regards depth of colour. Reference to our notes showed that the coloured solutions were those of the alkaloid mixed with potassium salts. The order of the potassium salts arranged according to their effectiveness in inducing colour changes was as follows:- iodide, bromide, chloride, chlorate and hydrogen sulphate. None of the tubes containing sodium salts had changed in colour.

On December 14, other three solutions had becone tinged yellow, one being the alkaloid with potassium nitrate, the other two being the alkaloid with sodium sulphate and sodium bromide. The flame test however showed that these two ~~presumably pure~~ sodium salts contained traces of potassium.

On January 30, all the tubes with one exception had a yellow tinge, the least coloured being those containing the sodium salts. With one single exception the sodium and potassium salts used in our experiments were the ordinary "pure" chemicals obtainable in commerce, and the tube which remained without colour was a solution made with carefully prepared potassium-free sodium chloride, for the purity of which we had ourselves been responsible. From these results it was sufficiently clear that ~~the~~ potassium ~~ion~~ is the effective constituent in producing change of colour in yohimbine solutions.

It remains to explain the variations shown by the different potassium salts as regards the rate at which they caused the photoactive change. Reference to tables of electrical conductivity of various potassium salts in solution showed that the order in which we had arranged them from their activity in producing photo-active change corresponded almost exactly with the extent to which they are ionised, the iodide which is most ionised, being the most active. The photo-active change therefore in this alkaloid is due to the presence of the potassium ion alone, and the rate of change is a function of the concentration of the

potassium ion.

We carried out a parallel series of experiments with yohimbine lactate. These however are less available for drawing conclusions, because the lactate tends rapidly to dissociate in watery solution with the formation of needle-shaped crystals of the uncombined alkaloid. Consequently we were able to obtain colour change only with potassium iodide and bromide, the two most ionised salts. In the tubes containing these salts, no needle-shaped crystals appeared. In all the others, which remained colourless throughout, needle-shaped crystals were present. In the case of the first two the photoactive conversion of the yohimbine had evidently occurred before the alkaloid had time to separate out of solution. Once it has so separated, it is evidently removed from the catalytic action of the potassium ion.

These experiments are of practical importance in the conservation of yohimbine solutions in a state of pharmacological activity. It is evident that yohimbine salts may not be exposed to the light in the presence of potassium salts. There are other alkaloidal salts that undergo a colour change on exposure to light (notably physostigmine) and likewise the substance adrenalin. Perhaps admixture with potassium may be the necessary condition for photoactive change in these cases as well.

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THE ACTION OF YOHIMBINE ON MEDULLATED NERVE, WITH SPECIAL REFERENCE TO FATIGABILITY. By JOHN TAIT and JAS. A. GUNN. (From the Physiology Department, University of Edinburgh.)

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THE ACTION OF YOHIMBINE ON MEDULLATED NERVE, WITH SPECIAL REFERENCE TO FATIGABILITY.

By JOHN TAIT and JAS. A. GUNN. (From the Physiology Department, University of Edinburgh.)

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EXPERIMENTS carried out in recent years by Gotch and Burch (1), Boycott (2), Boruttau (3), and F. W. Fröhlich (4), have shown that when medullated nerve is thrown into activity by an external stimulus a certain short period of time must elapse before it can function again. This period of inexcitability, or refractory period, is normally very short—not more than ·002 second for the sciatic nerve of the frog—but can be much prolonged by subjecting the nerve to special conditions. Thus low temperature (1) (2), anæsthesia (3) (4), and asphyxia (4), all greatly prolong the refractory period.

The readiest method of demonstrating the existence of this refractory period is to excite the nerve of a nerve-muscle preparation by two successive maximal stimuli separated by a very short interval of time. By the response of the attached muscle it is then possible to tell whether the nerve has conducted two excitations or only one. If the muscle response is a summated one it is evident that both excitations have been transmitted; if summation does not occur, then the second stimulus must have been in some way ineffective. Absence of summation occurs only when the interval of time between the two maximal stimuli is sufficiently short. In such cases it has been shown by means of the capillary electrometer (1) that the block to the second excitation is seated in the nerve.

The length of the refractory period would seem to be dependent on the intensity of the preceding excitation. Generally speaking, it has been found that the stronger the stimulus applied to the nerve the longer does the nerve take to recover its functional capacity. By combining powerful stimulation with anæsthesia or asphyxia or cooling of the nerve, one would therefore expect to get a maximal refractory period, and indeed Fröhlich was able to prolong it to ·1 second (4).

Besides the method of applying two successive stimuli it is obvious that a series of rapid recurring stimuli might be applied to the nerve; and if a sufficiently high rate of excitation could be attained, one would expect that at least some of the excitations would be ineffective.

Experiments with a rapid rate of stimulation have been carried out on numerous occasions. Neglecting in the meantime those in which the enormously high rates afforded by the discharge of Leyden jars, etc., have been used, where the interval between the individual stimuli is of a different order of magnitude from the refractory period, we shall mention experiments where the rate of stimulation has been over 400 a second and yet not greater than tens of thousands.

Bernstein (5), using a rate of 500 a second, obtained only a single initial contraction of the muscle. Roth (6), with a rate of 1000–5000, obtained tetanus. Langdon and Schenck (7), with a rate of 1800–2000 per second, also got tetanus. Kronecker (8), using a special device whereby he claimed to attain a rate of 20,000 per second, found tetanus, which on repetition became an initial twitch and subsequently failed. From the want of uniformity in these results it was for a time difficult to draw any general conclusion.

Of late, however, thanks to the work of Wedensky (9) and of Fröhlich (4), it has become possible to reach a definite generalisation. Wedensky, who happened to combine the method of rapid stimulation with anæsthesia of the nerve, found that strong excitations at a rate of about 100 per second applied to the proximal end of a nerve whose middle portion is deeply anæsthetised, produces simply a single twitch of the muscle. Fröhlich pointed out that this twitch is of the same height as the twitch evoked by one single maximal excitation, and that the result occurs only when the successive stimuli are separated by an interval less than the corresponding refractory period of the anæsthetised nerve. When, on the other hand, the stimuli succeed each other at an interval greater than the refractory period, then tetanus of the muscle occurs.

The fact that one initial maximal twitch follows upon repeated stimulation of the nerve shows that only the first excitation of the series has taken effect; this excitation prevents the second from being effective, the second prevents the third, and so on; consequently when once the excitations are applied at a sufficiently rapid rate the nerve refuses to conduct any more than the first excitatory process.

Such an effect, though at first sight suggestive of fatigue of nerve, is not necessarily fatigue. It is conceivable that a conducting mechanism built on simple physical principles might give the same result. Nevertheless, by a closely analogous method Fröhlich succeeded in showing that the nerve does actually become fatigued when subjected to rapid stimulation. When during anæsthesia of the nerve he selected a rate of stimulation which just about coincided with the definite refractory period corresponding to the intensity of stimulation used and to the given degree of anæsthesia, he obtained, not a single twitch, but a tetanus of peculiar form. This tetanus, instead of gradually climbing, after the normal fashion of a muscle tetanus, attained its maximum almost immediately, and then rapidly fell off in height until in the space of a second or so the muscle ceased to contract altogether.

Interruption of the rhythmical stimulation but for a second sufficed to restore the conductivity of the nerve to its previous condition.

Now, on the assumption that the degree of anæsthesia does not increase during the short period of observation, this peculiar form of tetanus points to a progressive lengthening of the refractory period due to the continuous activity of the nerve, and any change in the direction of depression of function which is solely due to activity is fatigue. That the gradual prolongation of the refractory period is not due to a temporary and coincident deepening of the anæsthesia is sufficiently clear from the consistent regularity with which the effect occurs even when the anæsthesia is passing off.

Fröhlich's work, while establishing the fact that nerve can be fatigued —a fact of fundamental importance in regard to our views as to the nature of the nerve impulse—serves at the same time to emphasise the high powers of restitution possessed by the structure. Even when anæsthetised almost to the point of complete absence of conductivity, the nerve still required to be stimulated uninterruptedly in order to maintain the fatigued condition. Interruption of the rhythmical stimulation for a fraction of a second left time for an apparently complete recovery.

The anæsthetic agents which were found by Wedensky and Frölich to prolong the refractory period of nerve include most of the common anæsthetics known to medicine (ether, chloroform, cocaine, phenol, etc.). In spite of the chemical differences between these substances, the kind of change produced in nerve by means of all of them seems to be virtually the same. Further, this change corresponds identically with that caused both by asphyxia (4) and by cooling (9), so that one can recognise a common element in the action of all of these things.

The present paper deals with the changes produced in nerve by means of yohimbine. This substance, which is an alkaloid derived from the bark of the Yohimbehe tree (10), was shown by Magnani in 1902 to be a local anæsthetic (11). We have investigated its action on the sciatic nerve of the frog, availing ourselves of the method of Wedensky—i.e. rapid rhythmical stimulation, in order to show changes in conductivity. The response of the attached gastrocnemius muscle was used as an index of the condition of the nerve.

The investigation has shown:—

(1) That fatigue changes in nerve may be demonstrated more readily by the application of this substance than by any method known to us.

(2) Yohimbine seems to differ somewhat in action on nerve from other anæsthetics.

For our experiments we used a 2 per cent. solution of yohimbine lactate in Ringer's fluid, which we applied to the middle portion of the dissected nerve. In order to keep the solution in contact with this part of the nerve, strips of blotting-paper (usually about 3 centimetres long) moistened with the solution were laid under and over the middle portion.

Thus the proximal and distal ends of the nerve were left unaffected by the solution, and to each of these parts a pair of electrodes was applied. To ensure that the solution should not run along the uncovered parts of the nerve and affect either the proximal or distal ends, the middle portion was kept at a slightly lower level than the two ends. The whole nerve-muscle preparation was kept in a moist chamber. The electrodes were connected by means of a Pohl commutator from which the cross wires were removed with a standard Kronecker coil (original pattern), in the primary circuit of which was an accumulator charged to $4\frac{1}{2}$ volts.

The result of soaking the nerve for a number of hours (the time varied from two to three hours in our experiments) is to abolish conductivity in

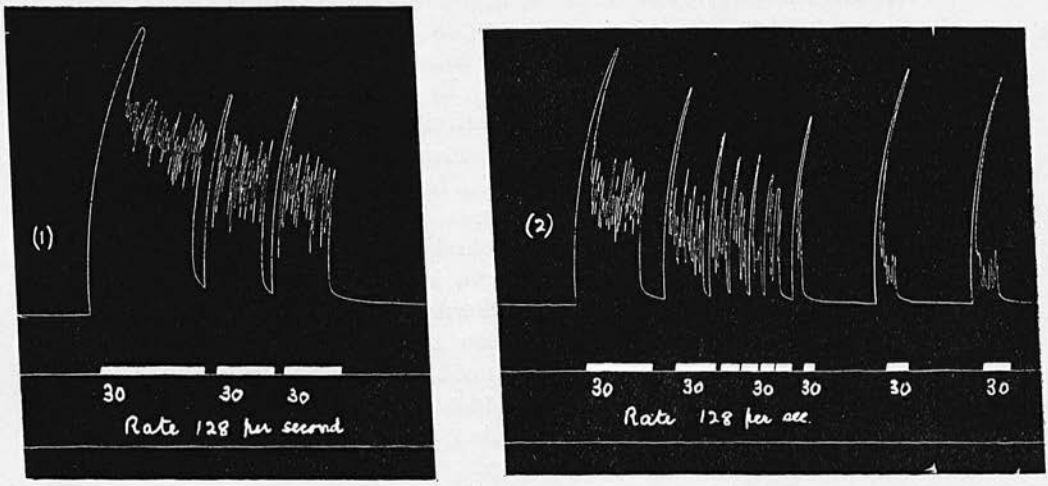

FIG. 1 (reduced to two-thirds).—Yohimbine lactate, 2 per cent., applied for 1 hour to 3 cm. of nerve. Proximal stimulation, rate 128 per sec. Intensity, 30 Kronecker units. Rate of drum, 1 mm. per sec. Tracing (2) taken 40 secs. after tracing (1).

Tracing (1) shows (i.) irregularity of tetanic responses; (ii.) diminution in height of successive tetanic responses.
Tracing (2) shows (i.) diminution in extent of successive tetanic responses with diminishing intervals between stimulations; (ii.) subsequent improved responses with increased intervals of rest between stimulations.

the part affected by the yohimbine solution. As is the case when other anæsthetics are applied to nerve, the abolition of conductivity does not occur abruptly but comes on gradually, so that long before the nerve has actually lost the power of conduction, changes can be detected which indicate a depression of function.

Thus when the proximal end of the nerve is stimulated at some fixed rate lying between 100 and 200 excitations per second, the tetanic responses of the muscle begin to undergo a change; instead of being smooth-topped, they become irregular in form, and the muscle, instead of remaining in continuous contraction, ultimately twitches more or less spasmodically (see fig. 1). On the other hand, the muscle response to distal stimulation is a smooth and regular tetanus, showing that the irregularity

of the muscle tetanus in the former case is not due to fatigue of the muscle or of the nerve ends in the muscle. In this respect Yohimbine resembles in its action other anæsthetics.

At a somewhat later stage the abnormality in the muscle response is clearly seen to be of a definite type. To any given series of continuously applied rhythmical excitations the immediate response of the muscle is a summated tetanus which quickly begins to decline in height, and finally becomes feeble and irregular, or ceases altogether. Thus the general form of the tetanus approaches that of the "fatigue tetanus" described by Fröhlich. (See fig. 1, tracing (1), and fig. 2.)

In the case of other anæsthetic agents applied to nerve it was found by Wedensky that the muscle response is largely dependent on the intensity of stimulation used. Wedensky showed (9) that at any given stage of anæsthesia, provided the rate of stimulation is kept constant, there is one definite intensity of stimulation (optimum of intensity) which produces a maximal height of tetanus; intensities either above or below this optimum cause a less height of tetanic response. In other words, when the nerve is anæsthetised, say by ether or cocaine, weak rhythmical stimulation produces a tetanus of submaximal height, stimulation at some moderate intensity causes maximal height of tetanus, while strong stimulation produces again submaximal tetanus. It is found, too, that tetani of the form which Fröhlich calls "fatigue tetani" are more readily obtained with strong stimulation. Furthermore, with deep anæsthesia and strong rhythmical stimulation, the muscle response, as already mentioned, is a single twitch of the same height as the twitch evoked by one single maximal excitation (4). The same is the case when nerve is asphyxiated (4). A similar effect has been shown by one of us (Tait) to occur when nerve is cooled. All these facts indicate that under these conditions the refractory period of the nerve corresponding to strong stimulation is longer than that corresponding to weak.

Yohimbinised nerve does not conform in this regard to nerve subjected to these other influences. At almost all stages of yohimbine anæsthesia in which alterations of the muscle response to rhythmical stimulation can be detected, this response takes the form of a "fatigue tetanus"—i.e. the last part of the tetanus is at least markedly lower than the first. Furthermore, the highest and best sustained muscle response is not produced by stimulation at moderate intensity, but in every case strong stimulation is more effective than any moderate stimulation as regards both the height and duration of the corresponding tetanus. (See fig. 2.) Thus it is evident that the refractory period of yohimbinised nerve does not increase with the intensity of the stimulation. If anything, the contrary would seem to be the case.

An examination of the tracings in figs. 1 and 2 shows that when series of rhythmical stimulations are applied in closely succeeding sets or groups to the proximal end of yohimbinised nerve, the successive muscular

responses tend to fall off in height with repetition of the successive series of stimulations. Thus in fig. 1, tracing (2), it is very clear that the responses to the first six series of stimulations become progressively lower and lower. This effect may be due either to rapidly deepening anæsthesia of the nerve or to fatigue. That it is due to some fatigue condition and not to progressive and rapid anæsthesia, is indicated by the fact that

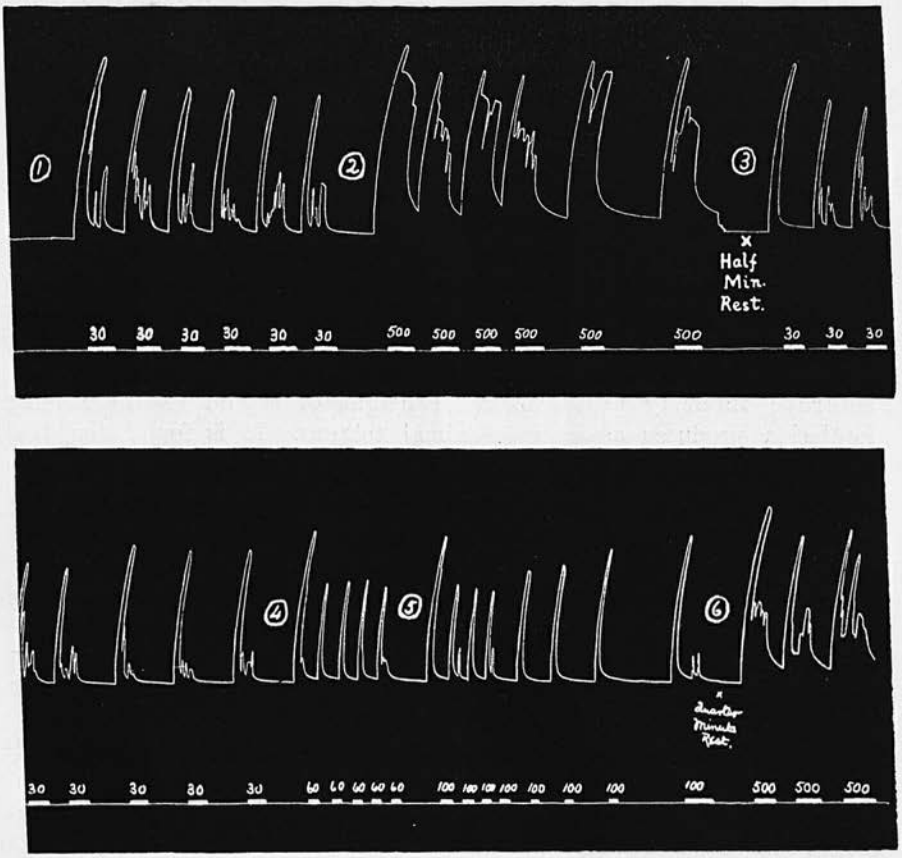

FIG. 2 (reduced to about one-half).—Length of nerve anæsthetised, 3 cm. Duration of application of yohimbine, 40 minutes. Rate of stimulation, 144 per sec. Rate of drum, 1·5 mm. per sec. Six series of responses are shown, corresponding to intensities varying from 30 to 500 Kronecker units.

Note (i.) the tetani are all of the "fatigue" form; (ii.) the responses corresponding to strong stimulation are more marked than those corresponding to weak; (iii.) in any given series with constant intensity of stimulation the height of the responses varies as the duration of the period of rest between stimulations.

if longer intervals of rest are allowed between the successive sets of excitations, the effect does not occur. Further, the effect is most marked when the intervals of rest between successive series of stimulations are made progressively less and less, as is the case in the first six responses of fig. 1, tracing (2), or in the middle series of responses in fig. 3.

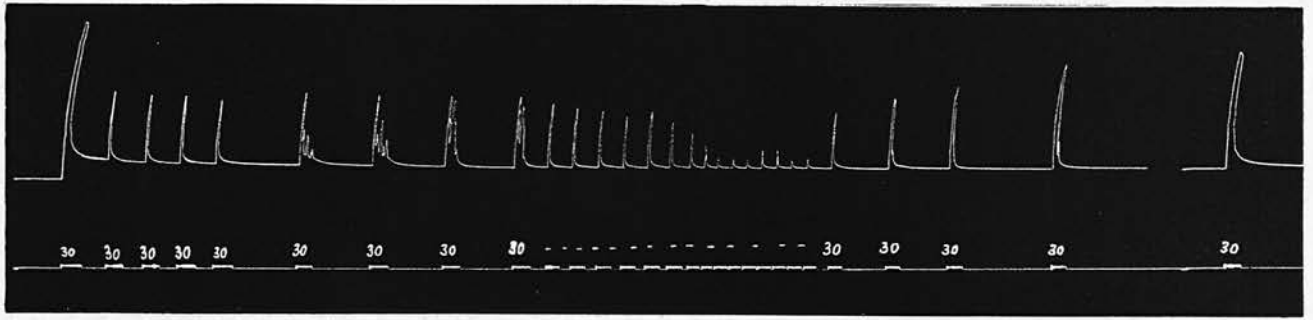

FIG. 3 (reduced to one-half).—Length of nerve anæsthetised, 1 cm. Duration of application, 1½ hours. Rate of stimulation, 144 per sec. Intensity, 30 Kronecker units. Rate of drum, 1 mm. per sec.

The tracing shows that the general extent of the muscle responses varies as the duration of the interval of rest between stimulations.

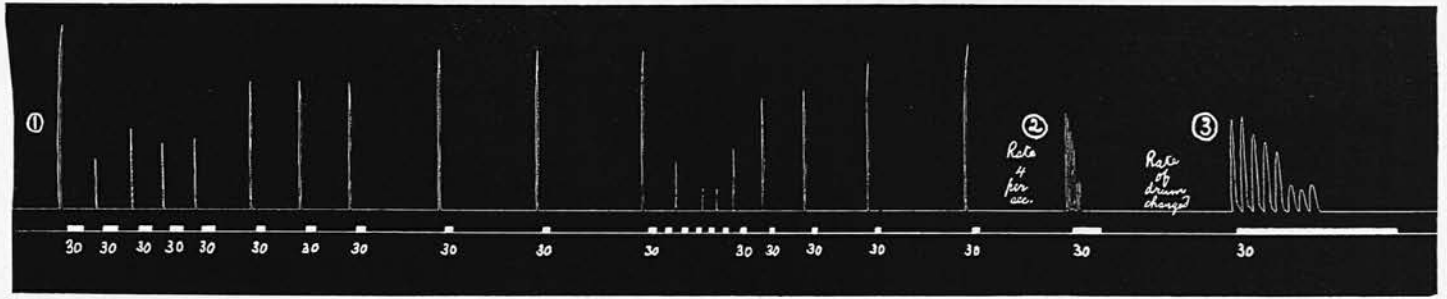

FIG. 4 (reduced to about two-thirds).—Length of nerve anæsthetised, 2·7 cm. Duration of application, 1¾ hours. Rate of stimulation in tracing (1), 128 per sec. ; in tracings (2) and (3), 4 per sec. Rate of drum in the case of tracings (1) and (2), 1 mm. per sec. ; in the case of tracing (3), 10 mm. per sec.

Tracing (1 shows summated muscle twitches varying in height with duration of interval of rest between stimulations.
Tracings (2) and (3) show rapid diminution in height of successive muscle twitches on repetition of stimulations, and final abolition of conductivity of nerve.

On the other hand, when the successive muscle responses have become less and less marked as a result of a steady diminution in the intervals of time between the sets of stimulations, they gradually increase in height again when the intervals of rest between sets of stimulations are made longer and longer. (See the last three responses in fig. 1, tracing (2), or the last five responses in fig. 3.) Provided the same short interval of time elapses in each case between the successive sets of stimulations, the second, third and fourth, etc., responses of the muscle may be all of about the same magnitude, whereas the first response of any series—beginning after an adequate interval of rest—is more marked. (See generally the tracings in figs. 2 and 3.) At any given stage of anæsthesia, therefore, the efficiency of the muscle response induced by stimulation of the proximal end of the nerve is directly proportional to the interval of time during which the preparation has rested from activity. This is clearly a fatigue phenomenon.

During the later stages of anæsthesia with yohimbine the muscle responses to rapid rhythmical stimulation tend to resemble simple muscle twitches rather than tetani (see fig. 4), and this is the case whether strong or weak stimulation is used. These seeming simple twitches are, however, in reality summated muscle responses. This is readily seen when one compares the height of the muscle contractions evoked on the one hand by rhythmical stimulation, and on the other by single maximal break shocks applied to the proximal end of the anæsthetised nerve (care being taken in each case to examine the preparation after an adequate interval of rest). In every instance the effect of rhythmical stimulation is to produce a much higher muscle response than that produced by a single maximal excitation. In this respect the action of yohimbine is once again different from that of other anæsthetics, for in the later stages of anæsthesia with, say, ether or cocaine, rapid stimulation, especially when strong, produces indeed a muscle twitch, but this twitch is of the same height as the response to one single maximal excitation of the nerve.

If we come now to the interpretation of this phenomenon we must conclude that when a series of excitatory processes are made to travel in rapid succession from a normal portion of nerve into a portion deeply anæsthetised with yohimbine, probably the first few excitatory processes succeed in traversing the anæsthetised part, but the passage of these unfits the affected portion of nerve for the immediate transmission of further excitatory processes. Only after an adequate interval of rest is the nerve able to function again, and an examination of the tracings in fig. 4 will show that this interval must be spread over many seconds to restore the conducting mechanism to exactly the same degree of functional capacity as before. From the fact that even with a relatively rapid rate of stimulation (between 100 and 200 per second) the deeply anæsthetised nerve is at the start able to transmit more than the first excitatory process, while after the passage of a few excitations it temporarily ceases to function, we infer that the refractory period of yohimbinised nerve is dependent, not so much

on mere degree of anæsthesia by itself as on the extent to which the anæsthetised nerve is within any given short period of time thrown into activity. It is activity during the anæsthesia rather than the anæsthesia itself which causes the prolongation of the refractory period.

The progressive impairment of function of the nerve with activity is equally well shown if during this stage of deep anæsthesia the nerve is stimulated at a slow rate (4 per second), with break shocks of a strength that is just maximal. (See fig. 4, last two tracings.) Then the individual twitches of the muscle consistently decline in height with each repetition of the excitation and finally, after a certain small number of responses have occurred, die away entirely. A rest of a considerable number of seconds (not more than thirty) suffices to restore the nerve to its previous condition, when the same process can be repeated again by rhythmical stimulation at the same slow rate. On the other hand, if the interval of rest be not sufficiently long (say only two to five seconds), the process of recovery is not so complete, and the next set of muscle responses are fewer in number and of less height. Meanwhile, if the nerve is stimulated at a part distal to the anæsthetised portion, the muscle responds by a continuous series of maximal twitches. (See fig. 5.)

Such experiments demonstrate in striking fashion not only the existence of fatigue in yohimbinised nerve, but also the gradual nature of the recovery from fatigue. In every case after a period of continuous activity the nerve becomes exhausted and requires a rest of a considerable number of seconds before it has regained its previous state of functional efficiency. Nevertheless, by stimulating the nerve after a shorter period of rest it can be shown that the recovery process, though incomplete, has still gone on to a certain extent. Further, the fact that in every case recovery does occur after fatigue indicates that nerve is characterised not so much by non-fatigability as by the possession of an extremely efficient mechanism for repair after fatigue.

How far the refractory period of nerve may be prolonged when the nerve is under the influence of yohimbine anæsthesia we have not determined exactly. Much depends on the signification in which the term "refractory period" is used. If we take the term to mean that period of time which elapses before one maximal stimulus following upon another of equal intensity can be fully effective, then the refractory period has been prolonged to at least ·25 second. If, however, we extend the definition to include the period of time necessary for complete recovery of the nerve after the application of a series of stimuli applied in rapid succession, then the refractory period has been prolonged to a number of seconds (more than five).

In the later stages of yohimbine anæsthesia when the proximal end of the nerve is stimulated by isolated maximal shocks at long intervals, the corresponding muscle responses are definitely lower in height than those produced by similar stimulation of the distal end. This points to a diminution

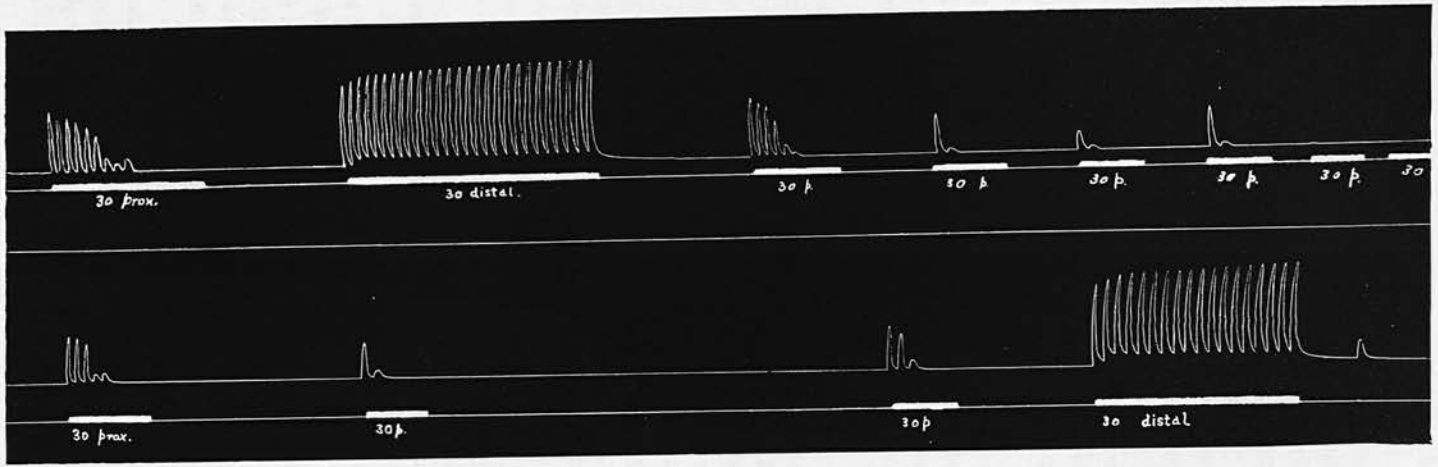

FIG. 5 (reduced to one-half).--From same preparation as in fig. 4. Rate of stimulation, 4 per sec. Intensity, 30 Kronecker units (break shocks alone effective). Rate of drum, 10 mm. per sec.

Tracing in upper line, taken during the same stage of anæsthesia as in fig. 4, shows—
(1) Result of proximal stimulation of nerve.
(2) ,, ,, distal ,, ,, ,,
(3) ,, ,, subsequent repeated proximal stimulation of nerve.
Tracing in lower line, taken at a somewhat later stage, shows—
(1) The difference in result according as distal or proximal stimulation is used.
(2) Partial recovery of the distally stimulated nerve with increased interval of rest.

in amplitude of the excitatory processes as they traverse the yohimbinised area. When the conductivity by deepening of the anæsthesia is just about to disappear, these responses become minimal. In this respect the anæsthetic action of yohimbine corresponds with the action of cooling, of asphyxia, and of anæsthesia produced by means of the more commonly used anæsthetics.

Before concluding, we ought to say that the fatigue effects which we have ascribed to the action of yohimbine can be due only to a change in the nerve itself. In a normal nerve-muscle preparation, strong stimulation, especially of the distal end of the nerve, may so alter the nerve-endings in the muscle that subsequent stimulation of the nerve may produce effects which might be erroneously ascribed to fatigue of the nerve. In all the experiments carried out by us which show fatigue phenomena this fallacy is excluded by the fact (1) that the intensity of the stimulation applied to the nerve was at no time more than just maximal; (2) that the fatigue phenomena in each case appeared before distal stimulation was used.

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1. A two per cent. solution of yohimbine lactate has been applied to the middle portion of the sciatic nerve of a frog's gastrocnemius preparation, and alterations in the conductivity of the nerve observed by means of the muscular response to rhythmical stimulation applied proximally to the alkaloid-affected portion. The rate of stimulation varied between 144 and 4 per second.

2. In its action on nerve, yohimbine resembles in many respects the already known action of other anaesthetics, of low temperature, and of asphyxia. It ultimately abolishes conductivity. The process of abolition of the conductivity is gradual, and is characterised by a progressive diminution in the amplitude of excitatory processes which traverse the affected part of the nerve, and by a prolongation of the refractory period of the nerve. By means of it, too, fatigue changes may be shown to occur in the nerve.

3. On the other hand, the action of yohimbine differs in important respects from that of asphyxia of low temperature, and of anaesthesia with ordinary agents. The tetanic responses of the muscle corresponding to rapid rythmical stimulation of the proximal end of a yohimbinised nerve are always of one type, and resemble the "fatigue tetani" described by F. W. Fröhlich. In stages of deep anaesthesia it is not easy to demonstrate the occurrence of initial non-summated maximal twitches as a result of rapid rhythmical stimulation of the nerve. The duration of the refractory period does not seem to vary directly with the strength of the stimulus applied to the nerve, and is more

clearly dependent on the amount of previous activity than is the case when other agents are used to depress the function of nerve.

4. The anaesthetic action of yohimbine lactate as applied in solution to the outside of a dissected nerve is characterised by great evenness and regularity. Partly for this reason, and partly because of the unusual prolongation of the refractory period due to yohimbine, it has been shown that nerve is a very convenient tissue on which to study the process of fatigue and recovery from fatigue.

5. In spite of the ready fatigability of yohimbinised nerve, complete restoration of function seems in every case to follow the katabolic changes due to activity.

6. The refractory period of nerve has been prolonged to ·25 second; while fatigue changes lasting for more than five seconds have been demonstrated.

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THE REFRACTORY PHASE ASSOCIATED WITH ONE SINGLE EXCITATORY PROCESS IN YOHIMBINISED NERVE. By JOHN TAIT. (From the Physiology Department, University of Edinburgh.) (With two figures in text.)

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THE REFRACTORY PHASE ASSOCIATED WITH ONE SINGLE EXCITATORY PROCESS IN YOHIMBINISED NERVE.

By JOHN TAIT. (From the Physiology Department, University of Edinburgh.) (With two figures in text.)

(*Received for publication* 31*st December* 1908.)

IN a paper previously published in this Journal (1) it was shown by Tait and Gunn that when two stimuli, separated by an interval of ·25 second, are applied to a nerve deeply anæsthetised with yohimbine, the muscle twitch corresponding to the second excitation is lower than that due to the first; whereas, if the interval of rest between the two stimuli is sufficiently long, the second muscle twitch is not diminished in height. (In their experiments they used not two single stimuli but series of rhythmical stimuli at a rate of four per second. What is referred to above applies to the responses to the first two stimuli of such a series.) From this they inferred that in deep yohimbine anæsthesia the refractory period corresponding to one single excitation may be as long as ·25 second.

By refractory period or refractory phase is commonly understood a condition of absolute or relative inexcitability conditioned by the simultaneous or immediately previous occurrence of function. The term inexcitability, or, what is the same thing, the term excitability, is, however, used in two different senses in physiology. On the one hand it may refer to the strength of stimulus which is just sufficient to excite function, no regard being paid to the amount of this function provided only that the organ responds. The German word "Anspruchsfähigkeit" is used to express this meaning. On the other hand, the term excitability is sometimes taken to imply the extent to which the activity of a tissue or organ is called into play by a stimulus of fixed amount. For this the German word "Leistungsfähigkeit" is used. These two conditions, theoretically distinct, are found practically to be closely related to each other, and, so far as the proof of existence of refractory phase goes, it matters not whether we take alteration in Anspruchsfähigkeit or alteration in Leistungsfähigkeit as showing change in excitability. In the present case the proof of prolongation of the refractory period is based on a change in the Leistungsfähigkeit of the nerve.

A point worthy of note in regard to Tait and Gunn's experiments is that the refractory phase would seem to consist of two stages: during one

stage the nerve is absolutely refractory, during the succeeding stage it is only relatively refractory. The stage of relative refractoriness too is much longer than that of absolute refractoriness. Indeed, in no single case in their experiments was the existence of an absolutely refractory phase corresponding to one single excitatory process by itself actually demonstrated, so short is this absolutely refractory stage. Even when the deeply yohimbinised nerve was stimulated at a rate lying between 100 and 200 per second, the corresponding muscular response was a summated twitch, indicating that at least the first two excitatory processes had traversed the nerve, and that at the commencement of the series of rapid stimuli the absolutely refractory period was less than ·01 second.

Still, there is no reason to doubt the existence of an absolutely refractory stage in nerve under the influence of yohimbine. The work of Gotch and Burch (3) and of Boycott (4) on normal nerve, and the obvious presence of an absolutely refractory period after rapidly repeated excitation of yohimbinised nerve (see Tait and Gunn's paper, p. 197, fig. 4, and p. 200, fig. 5), warrant us in assuming an absolutely refractory stage after each excitatory process in yohimbinised nerve.

The recognition of these two stages in the refractory phase of nerve is not new. Gotch and Burch (3), investigating the electrical response to two successive stimuli, found that the absolutely refractory stage of the nerve is followed by an interval in which the second electrical response, though present, is less marked than the first (see illustrations accompanying their paper). In other words, after the cessation of the absolutely refractory stage the Leistungsfähigkeit of the nerve does not recover with a bound. Again, Boycott (4), investigating the Anspruchsfähigkeit during the refractory phase, obtained evidence of a second relatively refractory stage succeeding upon the more prominent absolutely refractory stage. The work with yohimbine, however, emphasises the importance of recognising the relatively refractory stage as a component part of the refractory phase of nerve

As was mentioned before, the experiments of Tait and Gunn indicate that in deep yohimbine anæsthesia the absolutely refractory stage corresponding to one single excitatory process is in duration less than ·01 second. The inference is based on the nature of the muscle response to series of rapidly repeated stimuli. As there may be doubt as to the validity of a conclusion regarding the duration of the absolutely refractory stage drawn from the response to series of stimuli, it seemed advisable to control the results of this method by means of the method of two successive stimuli, the muscle being still used as an index of nerve activity. This method, first employed by Boycott (3) in the investigation of the refractory phase of nerve, depends on the fact that when two successive stimuli, separated by a short interval of time, are applied to the nerve of a nerve-muscle preparation, summation of the corresponding muscle contraction may or may not occur according as the interval

between the two stimuli exceeds or does not exceed the refractory period of the nerve.

The two successive stimuli consisted of break induction shocks of equal magnitude, obtained by means of two Kronecker coils placed at a distance from and at right angles to each other, the secondaries being connected in series through the nerve. In the primary circuit of each was a dry cell (modified Leclanché), these cells being specially picked from a number of new ones as showing the same potential difference between terminals. Each secondary coil was placed at the graduation mark, 15. In this position of the coil no muscle contraction occurred on make, while the break contraction was maximal, a statement which holds good for all the experiments (between thirty and forty nerve-muscle preparations). In the actual conduct of the experiments, however, the induction currents corresponding to make of the primary were not allowed to traverse the nerve, being short-circuited by means of a Du Bois key.

The time interval between the two stimulating shocks was measured by means of a piece of apparatus similar to that used by Boycott. A kick-over key in each primary circuit was opened by the slider of a Fredericq spring myograph. The one key was fixed in position. The other could be slid along a scale graduated in thousandths of a second, and could be clamped in such a position that it might open either simultaneously with the first or at any requisite time interval after it. Considerable care was spent in the calibration of this instrument. It was carefully levelled, and the keys were arranged to tip over on a very slight impulse. Each morning before use the wires on which the slider moved were wiped clean and oiled afresh with clock-oil. Under these conditions it was found to work in a regular and unvarying fashion.

In order to have a means of estimating the degrees of anæsthesia of the nerve at any stage of an experiment, a third Kronecker coil was so arranged that it might be connected at will with the stimulating electrodes. This coil stimulated the nerve with shocks of practically the same intensity as the other two, only in rhythmical fashion. Rates of 100, 50, 25, and 3 per second were used as occasion required. A time-marker in each case measured the duration of stimulation. The battery in the primary circuit of this third coil was an accumulator of about 2 volts potential.

The nerve-muscle preparation, taken from winter specimens of Rana temporaria or Rana esculenta, was rapidly dissected out and placed without the addition of any salt-containing fluid in a moist chamber, where it was fixed horizontally (so as to act on a rectangular lever), the nerve being laid over two pairs of electrodes, the one pair at the proximal, the other at the distal end. When the refractory period of the normal nerve had then been determined by stimulation through the proximal electrodes, 1 cm. of the middle portion of the nerve was moistened with a freshly

prepared saturated[1] solution of yohimbine hydrochloride in Ruysch's fluid. This was done by allowing one or two drops of the solution to fall on two pieces of blotting-paper which had previously been placed in position, one above, the other underneath the nerve, supported on a flat slab of glass 1 cm. in width. A thermometer inside the moist chamber served to indicate the temperature.

As a matter of fact, in nearly all the experiments, not one preparation only, but both preparations from the same frog were enclosed each in a separate moist chamber. They were arranged one above the other so as to write on the same drum by means of levers of identical weight and magnification. The object of using two preparations was at first to let one preparation serve as a control in case changes in the absolutely refractory period of the nerve might occur simply with time, and apart altogether from yohimbine. Thus parallel records were taken, in the one case of a yohimbinised preparation, in the other of a non-yohimbinised preparation. Latterly, however, when it had become sufficiently clear from control experiments that under proper conditions the refractory period of a non-yohimbinised preparation did not vary during the time of an experiment, both preparations were subjected simultaneously to the action of yohimbine and thus a considerable saving of time was effected.

A typical experiment will now be described.

Experiment, 22nd October 1908.—The two nerve-muscle preparations were taken from a large female temporaria. The one to be subsequently yohimbinised was put in the upper moist chamber, the control in the lower. Temperature 15·3° C. First, the height of muscle contraction elicited by one single maximal stimulus was determined in each case. The drum was now moved a slight distance by hand, and a muscle contraction was elicited in each case as the result of two stimuli ·001 second apart. These contractions were of the same height as those corresponding to one single excitation. The drum was again moved a slight distance, and contractions recorded corresponding to the time interval ·002 second These both showed marked summation. The absolutely refractory period consequently lay in each case between ·001″ and ·002″.

A more precise determination of the absolutely refractory periods was now made by reading the scale to ·0005″ interval, with the following result:—

		Upper Preparation.	Lower Preparation.
X. 30	single	max. contraction	max. contraction
	·001″	„ „	„ „
	·0015″	slight summation	„ „
	·002″	marked „	marked summation

At the start, therefore, the absolutely refractory period of the upper nerve

[1] The saturated solution was obtained by adding excess of the hydrochloride to a small quantity of the salt solution. The mixture was then gently heated, being stirred vigorously all the time, and subsequently cooled to room temperature.

lay between ·001″ and ·0015″, that of the lower nerve between ·0015″ and ·002″.

At X. 40 a saturated solution of yohimbine hydrochloride was applied to 1 cm. of the upper nerve, while 1 cm. of the lower nerve was similarly treated with Ruysch's fluid alone.

		Upper Preparation.	Lower Preparation.
X. 42	single	max. contraction	max. contraction
	·0015″	slight summation	,, ,,
	·002″	marked ,,	marked ,,
XI.	single	max. contraction	max. contraction
	·0015″	slight summation	,, ,,
	·002″	marked ,,	marked summation
XI. 15	single	max. contraction	max. contraction
	·0015″	,, ,,	,, ,,
	·002″	slight summation	marked summation
	·0025″	marked ,,	,, ,,

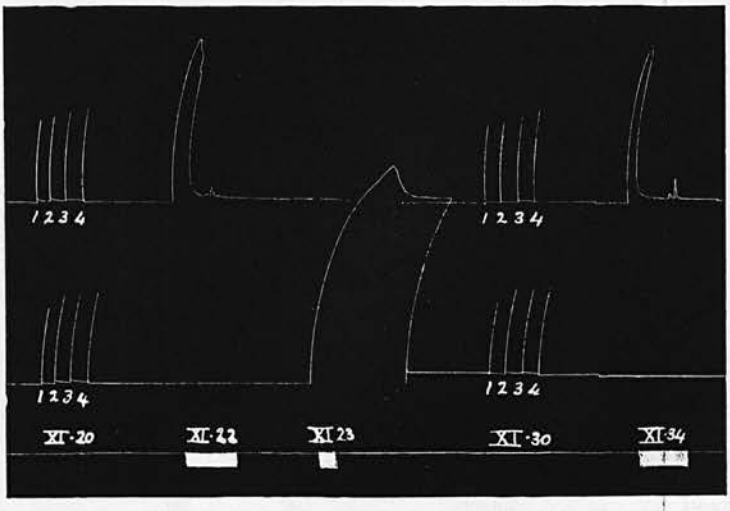

FIG. 1 (reduced to one-half).—Upper tracing from yohimbinised preparation. Lower tracing from control.

XI. 20:—1 = sing. 2 = ·002″ 3 = ·0025″ 4 = ·003″
XI. 30:—1 = sing. 2 = ·002″ 3 = ·003″ 4 = ·004″
XI. 22:—Rhythmical stimulation at 100 per second.
XI. 23:—Rhythmical stimulation at 100 per second. The writing-lever stuck and was drawn down by hand.
XI. 34:—Rhythmical stimulation at 100 per second. The temperature at this point was 15·5° C.

The tracings recorded at XI. 20 and XI. 30 were taken with stationary drum. The rate of drum in the other cases was 6 mm. per second.

Evidently the absolutely refractory period of the upper nerve at XI. 15 was beginning to be prolonged. The further course of the experiment is shown in the accompanying two figures.

The records in the lower line of these show that the absolutely refrac-

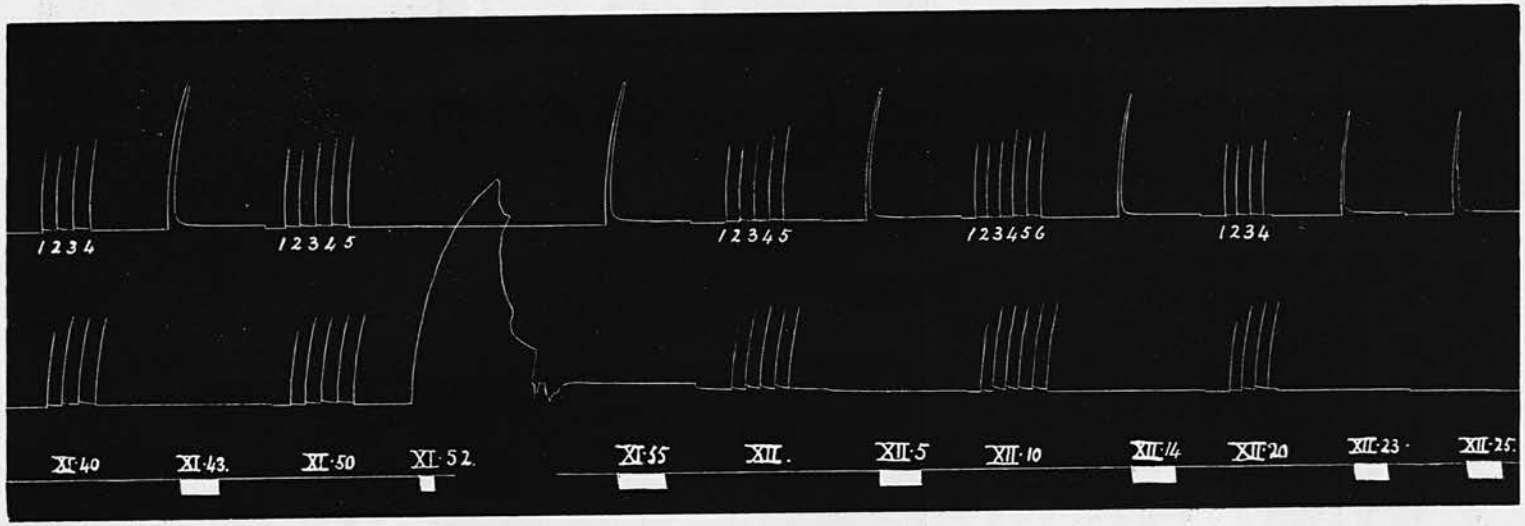

FIG. 2 (reduced to one-half).—Arrangement as in fig. 1.

XI. 40 :—1 = sing.
2 = ·002″
3 = ·003″
4 = ·004″

XI. 50 :—1 = sing.
2 = ·002″
3 = 003″
4 = ·004″
5 = ·005″

XII.:—1 = sing.
2 = ·002″
3 = ·003″
4 = ·004″
5 = ·005″

XII. 10 :—1 = sing.
2 = ·002″
3 = ·004″
4 = ·006″
5 = ·008″
6 = ·010″

XII. 20 :—1 = sing.
2 = ·002″
3 = ·004″
4 = ·005″

The tetanic responses were all taken with same rate of drum and of rhythmical stimulation as in fig. 1, except that the last response recorded, viz., at XII. 25, corresponds to 25 stimulations per second. The lever in the case of the response at XI. 52 was drawn down by hand.

The temperature at XI. 40 was 15·4° C., at XII. 20, 16° C.

tory period of the non-yohimbinised preparation continues less than ·002″ throughout. Rhythmical stimulation at 100 per second, when used, elicits full and sustained tetanus. The control preparation consequently undergoes no perceptible change as regards prolongation of the refractory period within the time occupied by the experiment.

On the other hand, the behaviour of the yohimbinised preparation is different. Certain points call for especial mention.

(1) The absolutely refractory period as measured by the method of two successive stimuli steadily increases with time, but even when last measured is not greater than ·005″.

(2) Meantime a series of many stimuli, each one separated from the preceding by an interval of as much as ·01″, in each case elicits a "fatigue tetanus." These eventually, with increasing anæsthesia of the nerve, approximate to the form of summated twitches.

(3) Such summated twitches resulting from rhythmical stimulation are higher than the summated contractions resulting from two stimuli alone.

(4) In spite of the marked temporary alterations in conductivity produced by spells of rapidly repeated activity, there is no evidence to show that the refractory period corresponding to one single excitatory process suffers thereby any permanent or lasting increase. After each period of "tetanisation" the absolutely refractory period as measured by the method of two successive stimuli is found to have regained almost its previous value. The slow steady increase which does occur might quite well be referred to progressive deepening of the anæsthesia.

This method of experimentation, therefore, amply bears out the conclusion previously deduced from Tait and Gunn's experiments with rhythmical stimulation alone, viz. that the absolutely refractory period corresponding to one single excitatory process by itself is still short, at a time when, by means of repeated stimulation, it is capable of being greatly prolonged. By implication it increases the reliance which can be placed on rapid rhythmical stimulation by itself, as a means of showing changes in the refractory period of a nerve.[1]

The general results obtained from the series of experiments remain to be discussed.

First, with regard to the control preparations. It was found that they might retain a constant refractory period for at least as long as four and a half hours. Such a result was not, however, attained in the first experiments I carried out. These seemed to show that the absolutely refractory period of the control underwent a steady and progressive increase—in one case from ·002″ to as much as ·015″ and even more, for, eventually, rhythmical stimulation at 25 per second produced a very broken and imperfect tetanus response (duration of experiment six and a half hours). On taking great pains to protect the preparation from loss of water during an

[1] Conf. Gotch, Art. "Nerve," Schäfer's Text-book of Physiology, vol. ii. p. 474, and Fröhlich, Zeitschr. f. allgem. Physiologie, Bd. iii. p. 468, 1903-4.

experiment this tendency to alteration on the part of the control preparations disappeared, and thenceforward in every case the refractory period of the control remained constant.

Out of 37 normal preparations examined immediately after dissection at temperatures which varied from 11·8° to 15·5° C., the following values were found for the absolutely refractory period:—

In	6	preparations	between	·001″	and ·0015″
„	13	„	„	·0015″	„ ·002″
„	10	„	„	·002″	„ ·0025″
„	4	„	„	·0025″	„ ·003″
„	2	„	„	·003″	„ ·0035″
„	1	„	„	·004″	„ ·0045″
„	1	„	„	·0045″	„ ·005″

The longest recorded refractory periods do not correspond with the lowest temperatures. Preparations from the same animal as often as not gave slightly unequal values for the absolutely refractory period.

With regard to the amount of prolongation of the absolutely refractory period corresponding to any definite degree of anæsthesia it is impossible to give numerical results. Generally speaking, at the temperature at which the experiments were carried out an interval of 40 to 60 minutes elapses before any change in the refractory period is observed. What is remarkable, however, is that with the first indication of prolongation of absolutely refractory period (i.e. an increase of ·0005″) rhythmical stimulation at a rate of 100 per second shows a well-marked fatigue tetanus, indicating that the nerve is already so much under the influence of yohimbine as to be incapable of sustained activity. From this time onwards a progressive increase in the length of the absolutely refractory period occurs, but at every stage the total increase is slight compared with the marked way in which the conductivity of the nerve succumbs to rhythmical stimulation. Thus, corresponding to a change of absolutely refractory period from ·002″ to ·008″, it was found that rhythmical stimulation at the slow rate of 3 per second gave a series of 7 twitches of successively diminishing height followed by absence of conductivity.

In any one experiment the rate of increase of absolutely refractory period with time is not constant. In the later stages of anæsthesia it is much more rapid than at the start, and becomes increasingly so towards the end. Towards the end, too, a marked diminution in the height of the muscle contractions corresponding to individual excitations sets in. With this rapidly occurring change in the height of the "single" maximal contraction, used as a standard by which to determine the presence or absence of summation, it becomes difficult to fix the duration of the absolutely refractory period. The longest absolutely refractory period directly observed was ·04″, but this is by no means to be taken as the limit. Before such a prolongation occurs, however, the anæsthesia must be very deep. The statement made by Tait and Gunn (loc. cit., p. 198),

that "in every instance the effect of [rapid] rhythmical stimulation is to produce a much higher muscle response than that produced by a single maximal excitation," is not strictly accurate, for it is possible to obtain responses to rhythmical stimulation at a rate of 50 or even of 25 per second which are of the same height as single maximal contractions. In such cases the anæsthesia is extremely deep, however.

From the height of the twitch-like summated responses which in ordinary deep anæsthesia result from rapid rhythmical stimulation Tait and Gunn inferred that "probably the first few excitatory processes succeed in traversing the anæsthetised part." This inference was found to be entirely borne out when the height of the most elevated summated response obtainable by means of two successive stimuli was compared with that of the responses in question. Provided the anæsthesia is not extremely deep, the former is always of less height than the latter.

Before concluding, reference may be made to some experiments carried out on yohimbinised nerve previous to the already described investigation. These had for their object the determination of the maximum total refractory period (corresponding to one single excitation) attainable in extreme yohimbine anæsthesia. While early degrees of anæsthesia are quite suited for determination of the absolutely refractory period, they are obviously not suited for determination of the total refractory period, so long at least as the muscle response is taken as an index of the degree of nerve activity. In later stages of anæsthesia, on the other hand, the muscle record serves as a very convenient means of fixing the duration of the total refractory period.

Two slightly different methods were used. In the one case, by means of two contact pins placed at appropriate intervals in the drum-plate of a Birch revolving drum, the nerve could be periodically excited by means of two stimuli at any required interval, followed by a rest of a longer interval. Thus, when two pins were put into the drum at a distance of ninety degrees on the circumference, and the drum driven at a rate of one revolution in four seconds, the nerve was excited twice with a rest of three seconds before next excitation. The method which was found most convenient, however, was to excite the preparation at appropriate intervals by hand, the drum on which the contractions were recorded meantime revolving at a fixed known rate. A time-marker recorded the moment of application of each stimulus. The interval between the excitations could therefore be read off the drum, either by the time-marker record or by the individual muscle contractions when present. In making determinations, care was of course taken to select for the purpose only such pairs of excitations (with corresponding contractions) as were separated by a sufficient interval of time from other excitations.

In both these sets of experiments the preparation, previous to the attainment of the requisite degree of anæsthesia, was excited as seldom as possible. Although, as will presently be pointed out, the nerve in

every case makes a complete recovery even after the most exhausting activity, this precaution served to keep the muscle fresh. In such experiments one must make sure that any slight falling off in the height of the second muscle-contraction of a pair is not due to progress of the anæsthesia apart from the residual effect of immediately previous activity. This can be settled by observing the height of the next succeeding contraction after a relatively prolonged interval of rest.

By means of the first method I found a total refractory period of 2 seconds, by means of the second of 2·2 seconds duration. At the time, however, I had not realised the importance of simultaneously determining the absolutely refractory period corresponding to these prolonged total refractory periods, otherwise it would have been possible here to give experimental values for the relation that obtains between the two. All that the tracings show is that the absolutely refractory period is at least very much shorter than the total refractory period.

One further point requires mention. It is of great interest to know whether a yohimbinised nerve as a consequence of activity undergoes any, even the slightest, alteration which is not recoverable from with adequate rest. To this question attention was constantly directed throughout. Tait and Gunn found no evidence of such alteration. In each case in their experiments the fatigue tetanus in the adequately rested preparation was virtually as high and as prolonged as it was in the first of a previously elicited series of more rapidly repeated fatigue tetani. This method evidently applies to the nerve a very searching test, for it is calculated to bring out the slightest change (1) in the absolutely refractory period (and possibly the total refractory period), and (2) in the capacity of the nerve to undergo sustained activity.

In my experiments I examined especially for (1) change in absolutely refractory period, and (2) change in the magnitude (i.e. muscle-exciting efficiency) of a single excitatory process, in each case after exhausting rhythmical stimulation. The first process was carried out in moderately deep yohimbine anæsthesia by means of the method of two successive stimuli. The second process was used in very deep anæsthesia, that is, at a time when the nerve is more sensitive than usual to the effects of activity, by comparing the height of a pair of muscle contractions each corresponding to one excitation, one applied before, the other applied at an adequate interval after rhythmical stimulation. In both cases complete recovery could be proved.

Other experiments involving a slightly different process of reasoning lead to a similar conclusion. It might be argued, for example, that the depression of function observed in yohimbinised nerve depends, not on the anæsthesia per se, but on the total extent to which the conducting mechanism has been called into play during the action of the alkaloid. If this were so, we should expect a yohimbinised nerve not to show these special effects until it had undergone a certain minimal amount of activity.

Observation has repeatedly shown that the depression of function associated with yohimbine progresses at a fixed rate quite apart from function. Thus, in not a few cases a preparation was treated with yohimbine and then left alone until such time had elapsed as experience had previously shown to be sufficient for very deep anæsthesia of a repeatedly stimulated preparation to occur. In one or two cases after the lapse of such interval the nerve was found to have lost its conductivity, the seat of loss being localised to the yohimbinised part; in other cases the conductivity when first tested was found to be almost gone, and all the phenomena of very deep anæsthesia to be immediately elicitable. This shows that the slow depression of function which does come on under the action of yohimbine is independent of the question whether the nerve has been made to function during the progress of the anæsthesia.

Summary.

1. In the refractory phase associated with one excitatory process in yohimbinised nerve two distinct stages are present. In the first or absolutely refractory stage conductivity is suspended. In the second relatively refractory stage conductivity, while present, is diminished in amount. Throughout this stage, however, it gradually and progressively increases from zero right up to the extent to which it was present before the excitatory process occurred.

2. Both these stages are present in the recovery of normal nerve after activity.

3. Yohimbine prolongs both the absolutely refractory phase and the relatively refractory phase of nerve. By much the greatest absolute prolongation is in the relatively refractory phase.

4. The total refractory phase of deeply yohimbinised nerve corresponding to one single excitatory process may be as long as 2·2 seconds. The greatest previously recorded prolongation (Tait and Gunn) was ·25 second.

5. Although yohimbine gradually depresses the function of a nerve, and although the immediate effect of activity during yohimbine anæsthesia is to exaggerate this depression of function, yet as regards the repair after activity yohimbine affects merely its rate and not its amount.

The expenses of this research have been defrayed by a grant from the Carnegie Trust. For my supply of yohimbine I am indebted to Drs Hillringhaus and Heilmann, Chemische Fabrik, Güstrow.

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THE ACTION OF YOHIMBINE ON THE HEART, WITH SPECIAL REFERENCE TO TOXIC HEART-BLOCK. By JOHN TAIT. (From the Laboratory of Physiology, Edinburgh University.) (With six figures in text.)

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THE ACTION OF YOHIMBINE ON THE HEART, WITH SPECIAL REFERENCE TO TOXIC HEART-BLOCK.

By JOHN TAIT.
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(*Received for publication* 17*th February* 1910.)

CONTENTS.

THIS communication is meant in the first instance as a general contribution to the question of arhythmia cordis as produced by drugs. The presentation of the subject follows the historical development of the facts.

PART I. GENERAL CONSIDERATIONS.

(a) The Refractory Phase of the Heart.

Since the experiments of Kronecker and Stirling (1), of Marey (2), and of Engelmann (3) on the subject, it has been recognised that the beating ventricle is during a certain phase of its cyclic action inexcitable to even the strongest stimuli. During the immediately succeeding phase it responds indeed to stimulation, but its irritability is low, and the strength of any stimulus adequate to excite it is greater than normal. It has thus an absolutely refractory phase and a relatively refractory phase.

Regarding the precise incidence and duration of the absolutely refractory phase, the experiments of Woodworth (4), and especially of W. H. Schultz (5), have shown that it begins with the commencement of and ends at a definite point just anterior to the end of systole. It thus falls entirely within the period of systole and is of slightly less duration than it, a relation which seems never to vary. Schultz, who tested the effect on the absolutely refractory phase of varying the amount of inorganic

salts supplied to the heart, came to the following conclusion: "The absolute refractory period continues to bear a constant relation to the duration of systole, whether the agents used increase or decrease the irritability of the muscle. In other words, if S equals the duration of systole and R the absolute refractory period, then the ratio R/S is approximately constant."

The investigation of the absolutely refractory phase is simple. It is characterised by zero irritability throughout. When we have, therefore, fixed the points of commencement and of termination in relation to the externally recorded change in volume of the heart, we have, mathematically speaking, determined it completely. It is otherwise with the investigation of the relatively refractory phase. At least two complicating circumstances render the complete determination of it difficult. In the first place, the end-point is not nearly so easy to fix as in the case of the absolutely refractory phase. In the second place, the irritability throughout the relatively refractory phase constantly varies. Nevertheless, the knowledge we possess regarding this phase is of prime importance. The outstanding feature is that in it the irritability, at first infinitely low, steadily increases until it attains a maximum (near which point presumably the next spontaneous heart-beat occurs).

Fröhlich (6), on theoretical grounds, has suggested that the return to normal or maximum irritability during the relatively refractory phase may possibly be represented by a logarithmic curve, in which the gradient is at first steep, and later more gradual. And indeed it is known that the return of irritability does take place rapidly at first and ultimately more slowly. A graph of the irritability during one ventricular cycle might consequently be constructed somewhat after the accompanying diagram (fig. 1). If we take a fixed base-line O X to represent the level of maximum irritability, and measure along it successive intervals of time in the ventricular cycle, then O S might represent the time of systole and S D that of diastole, while O R (just less than O S) would represent the duration of the absolutely refractory phase. If we arrange to represent any condition of diminished irritability by points below this base-line, vanishing irritability being at an infinite distance below, the graph of irritability during the cardiac cycle will take the form represented by the thick line in the diagram.

Constructed according to this convention, the graph illustrates not only the way in which the irritability rises during the relatively refractory phase, but also the fact that a hard and fast distinction exists between the absolutely refractory and the relatively refractory phase. A study of the conditions relating to the relatively refractory phase suggests that it is the expression of a reparative process taking place inside the tissue. On the other hand, it is just as likely that the absolutely refractory phase is conditioned by an oppositely directed change. It falls entirely within the period of systole or output of external energy, and is associated with some kind of change that we are in the habit of calling katabolic. There is

indeed a possibility that it may eventually be regarded as a measure of the duration of some specific (katabolic) process going on inside the tissue. However this may be, it is necessary in every case to make a clear distinction between these two phases in the irritability of the heart during activity.

We have seen that the ratio A/S, where A = the time of the absolutely refractory phase and S = the time of systole, was found to remain constant under varying experimental conditions. Is the ratio R/D, where R = the time of the relatively refractory phase and D = the time of diastole, similarly constant? To this it may be replied at once that the duration of the

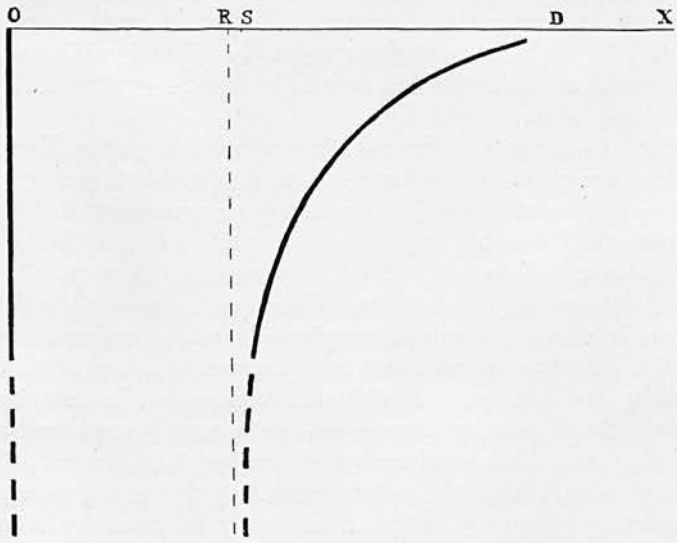

FIG. 1.—Graph of irritability of ventricle during a cardiac cycle.

OS, time of systole; SD, time of diastole; OR (slightly less than OS), duration of absolutely refractory phase.
With commencement of systole the irritability (represented by the thick line) at once becomes infinitely small, and continues thus until the commencement of the relatively refractory phase, when it increases, at first quickly, and later more slowly.

relatively refractory phase bears no fixed relation to the time taken up by the mere diastolic dilatation of the heart.

There are numerous cases in the literature in which change in the duration of the refractory phase as a whole has been recorded. In some cases it has been found to be shortened; in other cases it has been found to be prolonged. A list of such cases is contained in a paper by Rohde (7). Now, in all these cases—and in none of them was any distinction made between absolutely refractory and relatively refractory phase—what was really discovered was in all probability an alteration in the relation R/D. While, therefore, the time taken up in the systolic contraction is an excellent measure of the duration of the absolutely refractory phase, there is no corresponding relation between the time of diastolic expansion and duration of relatively refractory phase. The diastolic relaxation in itself

would seem to be a passive or indifferent process so far as concerns the active reparative changes which bring the heart back to the degree of irritability existing just at the moment of spontaneous explosion.

(b) The Refractory Phase of Nerve and the Action of Yohimbine on it.

Nerve possesses a refractory phase. The first observers to recognise its presence were Gotch and Burch (8). Their work was confirmed and extended by Boycott (9), Boruttau (10), and Fröhlich (11), the latter of whom, by applying the discovery of Gotch and Burch to the explanation of certain curious phenomena described by Wedensky (12), in anæsthetised nerve threw much new light on the experimental bearings of refractory phase in general.

In order to explain the observed phenomena in nerve, Fröhlich (6) assumes the presence at one time of an absolutely refractory phase, at another time of a relatively refractory phase, according as the stimulation used to excite the tissue has been strong or weak. This method of treating the question is apt, however, to lead to confusion. It would be more in accordance with analogy, and probably also with observed facts, to grant the existence in nerve of a refractory phase in which under all conditions there are the same two stages as we have learned to recognise in the heart (13). By slightly modifying Fröhlich's explanation in accordance with this principle, the Wedensky phenomena are as readily explained as before, while at the same time uniformity of treatment is secured. We shall consequently take it that the whole refractory phase of nerve consists of an absolutely refractory stage immediately followed by a relatively refractory stage, the latter at least corresponding to a repair process.[1]

It was shown by Tait and Gunn (15) that the alkaloid yohimbine, which has been used in medicine as a local anæsthetic (16), has the property of enormously prolonging the total refractory phase of nerve. This property it possesses in more marked degree than ordinary anæsthetic substances, e.g., ether, cocaine, etc., which likewise prolong the total refractory phase. Subsequent analysis of its action by Tait (13) showed that yohimbine exercises a selective influence on the relative refractory phase, prolonging it enormously, while any prolongation of the absolutely refractory phase, if present, is slight by comparison. Other anæsthetics do not exercise this exclusive action on the relatively refractory phase; they would seem to have a depressing action as much on katabolism as on anabolism (16). Yohimbine, on the other hand, interferes principally with the repair process that succeeds activity.

[1] The interesting results recently published by Keith Lucas (14) on the "irresponsive period" of muscle also support this idea. If one only assumes that the local excitation due to the application of induction currents to muscle continues in existence for more than $\frac{1}{100}$ second, it seems to me that the phenomena with which Lucas is dealing are phenomena of absolutely and relatively refractory phase.

(c) The Question at Issue in the Present Communication.

We have seen that nerve as well as heart has a refractory phase consisting of two stages, an absolutely refractory stage and a relatively refractory stage. We have further seen that yohimbine exercises a special action on the relatively refractory phase of nerve. The question now arises: Does yohimbine affect the refractory phase of the heart, and, if so, how?

The action of yohimbine on the heart has already been a subject of research, the most complete and detailed report being that of F. Müller (17). Gunn (18) has likewise furnished a good account of the subject, his experiments being in the main confirmatory of Müller's. The work of other investigators is of less importance, and is included in both of the communications just referred to.

Shortly expressed, yohimbine has been found to produce the following results:—An overdose stops the heart in diastole; the ventricle comes to rest first, then the auricle, and finally the sinus. In lesser concentration it slows the heart and produces the phenomenon of "heart-block," the ventricle beating once for every two, three, or more auricular beats. The action of the drug is independent of the extracardial nerves and of the nervous inhibitory mechanism in the heart. These results were obtained by both Müller and Gunn. One interesting point reported by Müller alone, which recalls the work of Tait and Gunn (14) on nerve, is that the heart muscle under certain conditions of yohimbinisation is readily fatigued.

With regard to the question of refractory phase little information is to be obtained from Müller's communication. He states that the "Anspruchsfähigkeit," or irritability of the heart to external stimulus, is heightened in the earlier stages of yohimbinisation, while in the later stages, i.e. with higher concentration of the drug, it is diminished. This he determined by rhythmical excitation of the heart brought to rest by the first Stannius ligature, or of the excised, no longer spontaneously contracting ventricular apex. Observations made by means of slow rhythmical stimulation of the resting heart are, however, of doubtful value in settling a question regarding duration of refractory phase, and Müller rather avoids discussion of the point. On the other hand, Gunn, as the result of one experiment on the Stannius heart, states that the "refractory period" is prolonged, and explains by means of a diagram how prolongation of the refractory period of itself accounts for the phenomenon of heart-block.

The hypothesis thus put forward by Gunn regarding a possible causal relationship between prolonged refractory period and heart-block is not new. A similar hypothesis was advanced in 1901 by Straub (19) to account for the heart-block produced by antiarin, a drug belonging to the digitalis group. Straub's experiments were also carried out on the resting (first Stannius ligature) heart, to which stimuli were applied in rhythmical series. He claimed to have found that antiarin, in addition to

depressing the irritability of the heart, likewise prolongs the refractory phase, and described by means of diagrams how a prolongation of the refractory phase of the ventricle would involve "Halbierung" or heart-block. Without any diagram it is easy to see how this effect would follow from a prolongation of the refractory phase of the ventricle. If we assume that the stimuli which comes in rhythmical succession from the auricle are just sufficient to excite the ventricle at the end of diastole or at some point in post-diastole, i.e. when its irritability is about maximal, any marked delay in the recovery of normal irritability on the part of the ventricle will prevent some of the auricular stimuli from taking effect. The ventricle will beat once, say, for every two auricular beats. If the refractory phase of the ventricle is still further prolonged, the ventricle will beat once for every three auricular beats, and so on.

Straub's results were adversely criticised by Alcock and Meyer (20). They pointed out that what Straub had looked upon as a prolongation of refractory phase was entirely a depression of excitability—a "deepening" of the refractory phase rather than an actual prolongation thereof; and as a matter of fact their objection is relevant, for Straub failed to show in convincing fashion that the irritability, even though depressed by the drug, undergoes a steady increase with time. A similar objection applies to Gunn's experiment on the Stannius heart.

To sum up, therefore. The amount of positive knowledge we possess with regard to the action of yohimbine on the refractory phase of the heart is extremely meagre. Further, the hypothesis of Straub with regard to the causation of heart-block, while plausible enough, lacks experimental proof. The object I had in view in undertaking the present research was, in the first place, to compare the action of yohimbine on the refractory phase of the heart with its known action on the refractory phase of nerve. In the event of it being found to prolong the relatively refractory phase of the heart—and from the work of Müller and of Gunn we know that it causes heart-block—it was evident that the experimental proof necessary to establish Straub's hypothesis would then be furnished.

Part II. Experimental: the Yohimbinised Heart.

(a) Technique.

The contractions of the profused frog heart were recorded by means of a Schäfer heart plethysmograph (21). The apparatus was not, however, filled in the usual manner with oil. The lower part of the bottle which receives the heart contained Ringer's fluid; the upper part, including the two horizontal arms, contained liquid paraffin. In this way it was arranged that while the attached base of the heart was surrounded with paraffin, the apical portion dipped into Ringer's fluid. A platinum wire, fused into the bottom of the bottle, projected by one end into the

Ringer's fluid, by the other to the exterior. Consequently it was possible to send an electric current from this wire through the Ringer's fluid and heart to the metal cannula which conveys the perfusion solution, and at the same time to ensure that the whole of this current should pass through the heart-wall. (See fig. 2.) As a precaution against the formation of poisonous electrolytic products, the cannula for conveying the perfusion solution was gilded.

The exciting currents were break induction shocks obtained from a standard Kronecker coil (new pattern), in the primary circuit of which was an accumulator, sometimes of $2\frac{1}{8}$ volts, sometimes of $4\frac{1}{4}$ volts potential. The accumulator was always kept fully charged. Each excitation was signalled on the heart tracing by means of a spark coil, the method used

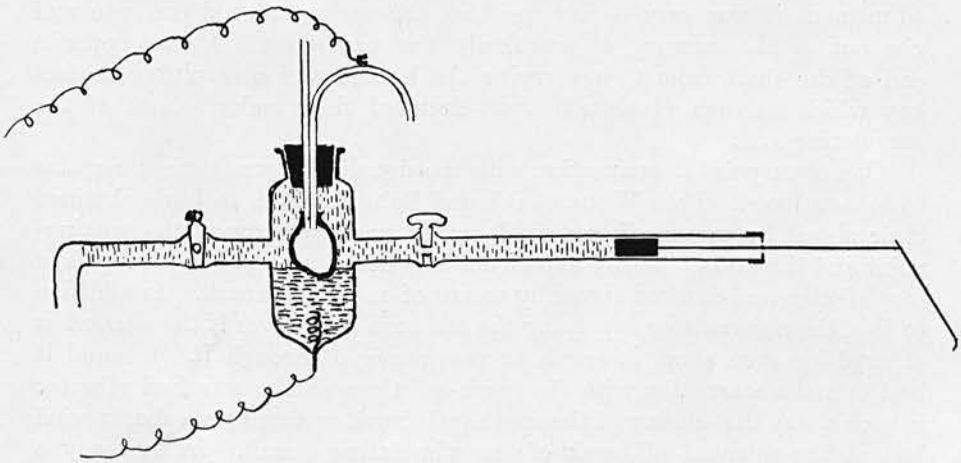

FIG. 2.—Schäfer's heart plethysmograph arranged to allow of electrical stimulation of the heart.

The lower half of the bottle containing the heart is filled with Ringer's fluid (horizontal shading). The upper half and the arms of the apparatus contain liquid paraffin (vertical shading).
A platinum wire fused into the bottom of the bottle projects into the Ringer's fluid. The double cannula used for perfusion is of metal. A current can consequently be sent from the metal cannula through the heart to the Ringer's fluid and platinum wire below.

being practically the same as that described by Schultz (5), which is a modification of a similar method used by Woodworth (4), this in its turn being a modification of a method first introduced into physiology by Schäfer (22). The final modification consists, firstly, in keeping the source of current for the signalling coil distinct from that for the stimulating coil, simultaneous breaking of the two being achieved by means of a specially constructed key; and, secondly, in arranging that only the "break" induction shocks should traverse the heart, the "make" shocks being short-circuited. I was not aware of the existence of Schultz's communications dealing with heart refractory period until the work described in this paper was half finished; and I happened to adopt a somewhat different mechanical device from his.

The special key which I used for the purpose of simultaneously breaking

two contacts consisted of an iron rod or shaft fixed horizontally in bushes, so that it could be rotated about its long axis by means of a crank and handle at the end. Excentrically attached to this shaft, and some little distance from each other, were two circular vulcanite cams. Pressing against the upper surface of each cam was a straight steel spring (the springs actually used were those supplied with Kronecker induction coils for producing a slow rate of interruption), one end of which was clamped in such a way that the spring stood out more or less horizontally above and at right angles to the direction of the underlying shaft. Thus the springs moved once up and down with each revolution of the shaft, their free (platinum-tipped) ends dipping each into a separate pool of mercury, over which in each case a constant stream of water flowed. By proper adjustment it was easy to arrange that the springs should dip into and rise out of the mercury at practically the same time. At the opposite end of the shaft from that carrying the handle was a revolving contact key which on each revolution short-circuited the "make" shock of the stimulating coil.

One other point in connection with the signalling arrangement remains to be mentioned. Both Woodworth and Schultz seem to have obtained their signal by means of one single spark passing between the writing-point and the drum. In my experience it is difficult to get a reliable and consistently well-marked signal by means of one single spark. In addition to this, the accumulator supplying the coil soon runs down if the current is allowed for even short intervals to pass straight through it. I found it best to make connection with the spark-coil through the attached vibrator in such a way that closure of the spark-coil circuit occurred for a short period just at the moment of break of the stimulating circuit. In this way a rapid but short-lasting succession of sparks passed between the writing-point and the drum, and thus one ensured that the moment of stimulation should be well marked, while at the same time the accumulator in the signal circuit remained efficient for a much longer period.

The perfusion solution for the heart consisted either of Ringer's fluid (Rusch's modification thereof), or of a mixture, as originally recommended by Kronecker, of one part of defibrinated ox-blood with two parts of Ringer's solution. As a rule the heart was perfused with either of these fluids for a varying period, from fifteen minutes to two hours or more, before yohimbine was admitted. Perfusion with the ox-blood mixture was found to have a remarkably beneficial and steadying effect on a heart which at the start was inclined to beat feebly or irregularly. As a rule, if such a heart was left for some time under steady perfusion with blood mixture, it settled down to beat strongly with even and regular rhythm. This effect was especially marked in the experiments which were carried out in the breeding season. Usually, too, a marked increase in the rate of the heart became established. This effect, however, I put down as due, at least partially, to rise of temperature, for both frogs

and blood were kept outside, and were taken into the warm laboratory only when an experiment was about to be carried out.

The yohimbine (in the form of the relatively insoluble hydrochloride) solution employed was never of less concentration than 1 in 20,000. For some purposes I used concentrations as high as 1 in 5000—these stop the heart at once—but as a rule I used 1 in 10,000, 1 in 15,000 (most often of all), or 1 in 20,000. The hydrochloride is a more stable salt than the lactate, though not altogether stable. In equal concentration I found it to be more active than the lactate. The only trouble in dealing with the hydrochloride is the slowness with which it dissolves. I used a stock solution of 1 in 5000 parts of Ringer's fluid. Even with repeated vigorous shakings this took over an hour to dissolve.[1]

The admixture of yohimbine with blood does not in any way interfere with the action of the drug, at least in the freshly made mixture, two solutions of the same concentration, one in blood mixture, the other in Ringer, having precisely the same quantitative effect on the heart. Care was nevertheless taken not to interchange the variety of perfusion fluid used during an experiment; thus, if the heart was first perfused with blood mixture, yohimbine was then applied dissolved in a similar medium. The pressure of the Ringer's fluid or of the blood mixture was adjusted at the start, so as to give the best range of excursion, conjoined with a rapid diastole, and care was taken to have the yohimbine solution on first application at this level. Mariotte's bottles were not used.

In the first few experiments carried out, I endeavoured to adapt the ligature with which the heart was fastened to the end of the cannula so closely to the auriculo-ventricular junction that the ventricle should be altogether isolated from the auricle. Owing to the fact, however, that the line of the auriculo-ventricular junction does not lie in one plane, but bends in sinuous fashion, the auricular tissue at one place projecting into a bight in the ventricular, I found this difficult to do. If, again, the ligature was arranged to include a portion of the basal part of the ventricle, the remaining uninjured part of the ventricle did not beat properly. I therefore gave up the attempt completely to isolate the auricle from the ventricle, and contented myself with simply applying the ligature over auricular tissue in the immediate neighbourhood of the ventricle. None of the sinus was left in connection with the preparation.

The heart was attached to the cannula in such a way that its long direction should coincide with the line of the cannula, and not cut it at an angle. In this way it was easy to arrange that the apex of the organ, when placed in the plethysmograph, should dip into the Ringer's fluid not merely during diastole, but also during systole. Owing to the possibility of outward diffusion of yohimbine through the wall of the heart into the surrounding Ringer's fluid, the plethysmograph bottle was after each

[1] I intend shortly to publish experiments regarding the conditions of stability of yohimbine salts in solution.

experiment emptied of Ringer's fluid by suction through a rubber tube washed out two or three times, and refilled with fresh Ringer. The paraffin was not changed except for slight addition from time to time to replenish loss.

The species of frog used for the experiments was Rana temporaria, no distinction being made as to sex. The experiments were carried out during the months of January to March and October to December, no experiments being carried out during the summer months. Some of the frogs used in February and March had been artificially fed with fresh muscle removed from the limbs of dead frogs. Hearts which on exposure were found dilated and full of blood, and into which it was easy to introduce the cannula, were rarely found to beat well. Wherever, owing to extreme irritability and contraction of the ventricle, difficulty was experienced in cutting the interauricular septum, the heart on successful attachment to the cannula continued to beat for hours without sign of impairment.

(b) The Absolutely Refractory Phase.

If the irritability of the normal heart is tested about the time of full systole, an extra contraction can be elicited by fairly strong stimulation applied just before the summit of systole. The finding of Schultz that the ratio A/S (A, duration of absolutely refractory phase ; S, time of systole) is just less than unity, I have been able to confirm in every case. As a matter of fact, however, A is so nearly equal to S that for practical purposes it is sufficient, in disproving prolongation of absolutely refractory phase, to show that a stimulus applied at the summit of systole evokes an extra contraction.

When yohimbine is admitted, it is found that a stimulus which previously was just strong enough when applied at the summit of systole to evoke an extra contraction, now fails to produce any visible effect. Nevertheless, by increasing the strength of the stimulus an extra contraction may be elicited. The degree to which the stimulus must be increased depends on the degree of yohimbinisation, being greater the more the heart is slowed, i.e. the higher the concentration of yohimbine in the perfusion solution. In no case, however, even in extreme yohimbinisation, does one fail to get this extra contraction from a stimulus applied at the summit of systole, provided simply that sufficiently powerful stimulation is used. In other words, the absolutely refractory phase of the heart is not prolonged by yohimbine.

Experiment, March 12, 1909. (See fig. 3.)—Heart perfused with blood mixture beats regularly from the start. Stimulation applied at apex of systole with 800, 900, 1000, and 1250 Kronecker units ($2\frac{1}{2}$ volts in primary circuit) causes no extra systole. Stimulation with 1500 Kronecker units, however, causes extra systole.

Yohimbine hydrochloride (1 in 15,000) then perfused. Almost complete

stoppage of heart at first, from which it gradually recovers, beating with almost regular rhythm at one-fourth of its original rate. Rhythmical stimulation (about 4 per second) with 4000 Kronecker units shows that heart fails to respond immediately on completion of systole. On the other

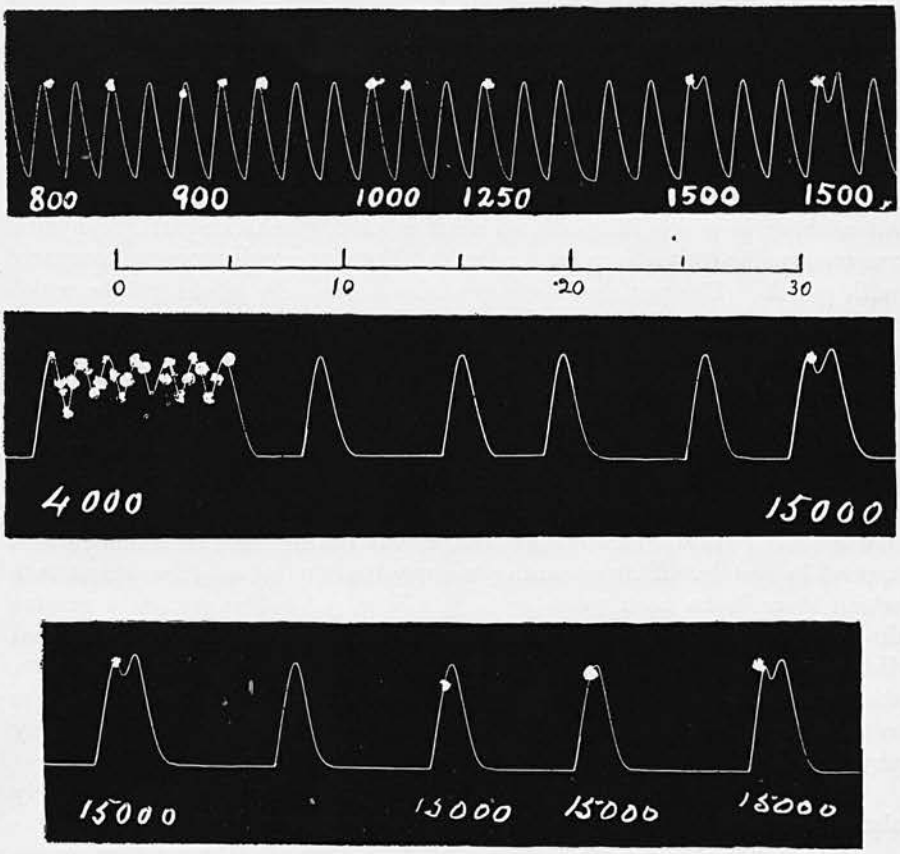

FIG. 3.—To show that the absolutely refractory phase of the heart is not affected by yohimbine.

The upper tracing was taken from the non-yohimbinised heart. Stimulation with 1500 Kronecker units is just sufficient to excite at the summit of systole, stimulation at any less intensity being insufficient.
The middle and lower tracings were taken from the same heart during perfusion with 1 in 15,000 yohimbine hydrochloride. Stimulation with 4000 Kronecker units is now seen to be even less effective than was the case with 1500 units previously. At the same time very strong stimulation—15,000 units—still suffices to excite at the summit of systole.
(The time-scale is marked in intervals of five seconds.)

hand, stimulation applied at the apex of systole with 15,000 Kronecker units causes extra contraction.

(c) The Relatively Refractory Phase and the Latent Period.

From the illustration accompanying the experiment cited in last section, it is evident that, at least in the earliest stages of diastole, the irritability of the yohimbinised heart to induced currents is diminished. Thus, when

1500 Kronecker units stimulates the non-yohimbinised heart in the very earliest stages of diastole, 4000 such units fails to stimulate the yohimbinised heart until diastole is considerably advanced. Now, Müller states that at certain stages of yohimbinisation the irritability to induced shocks is increased. I have not been able to confirm this observation. In my experience yohimbine, in whatever concentration, diminishes the irritability during every phase of activity in which the heart is excitable at all. Numberless tracings might be given in support of this statement.

At the same time, I should be unwilling to lay undue stress on observations carried out by a method which is not capable of directly settling the question. The method of testing irritability used by me, in common with all methods in which the perfused heart is used, gives, for purely physical reasons, under-estimations of the irritability during diastole as compared with systole. The perfused heart contains a quantity of electrolyte which varies in amount with each contraction and expansion. In systole, when the amount of the electrolyte is small, the total current conveyed by the heart tissue is high. On the other hand, in diastole the contained (neutral) electrolyte conveys a considerable proportion of the current, and the current-density in the wall of the heart is less, an effect which is still further enhanced by the greater volume of the whole mass of heart and contained electrolyte combined. Thus a current applied to a heart in diastole has, ceteris paribus, less chance of exciting than the same current applied in systole. This reasoning is supported by actual experiment, into which there is no need to enter here. Now, yohimbine causes a greater diastolic expansion of the heart than usual, and the above-mentioned disturbing factor becomes exaggerated in importance. At the same time, the apparent difference in irritability of an expanded yohimbinised heart is so very great that I am loath to ascribe the whole difference to a purely physical cause.

The irritability is diminished not only to induced currents, but probably also to chemical stimulation, as the following considerations will show.

It is impossible to send strong electric currents through an organ attached to a metal cannula without causing some electrolysis at the site of attachment, and as a matter of fact I obtained unusual effects with strong stimulation. An almost constant abnormality of this nature is shown in fig. 4. This takes the form of a quickly developing contracture, superposed on which are rapid contractions, both the amount of the contracture and the rate of the superposed contractions dying away gradually and simultaneously, while the heart eventually returns to its normal beat. Now this effect is almost certainly due to the formation of electrolytic products (acid or alkali, as the case may be) within the actual heart-wall. The stimulation they cause is intense at first, but grows gradually weaker as the substances become neutralised or washed away. This effect I have never seen in a yohimbinised heart, no matter how strong the electrical stimulation. The natural inference is that yohimbine

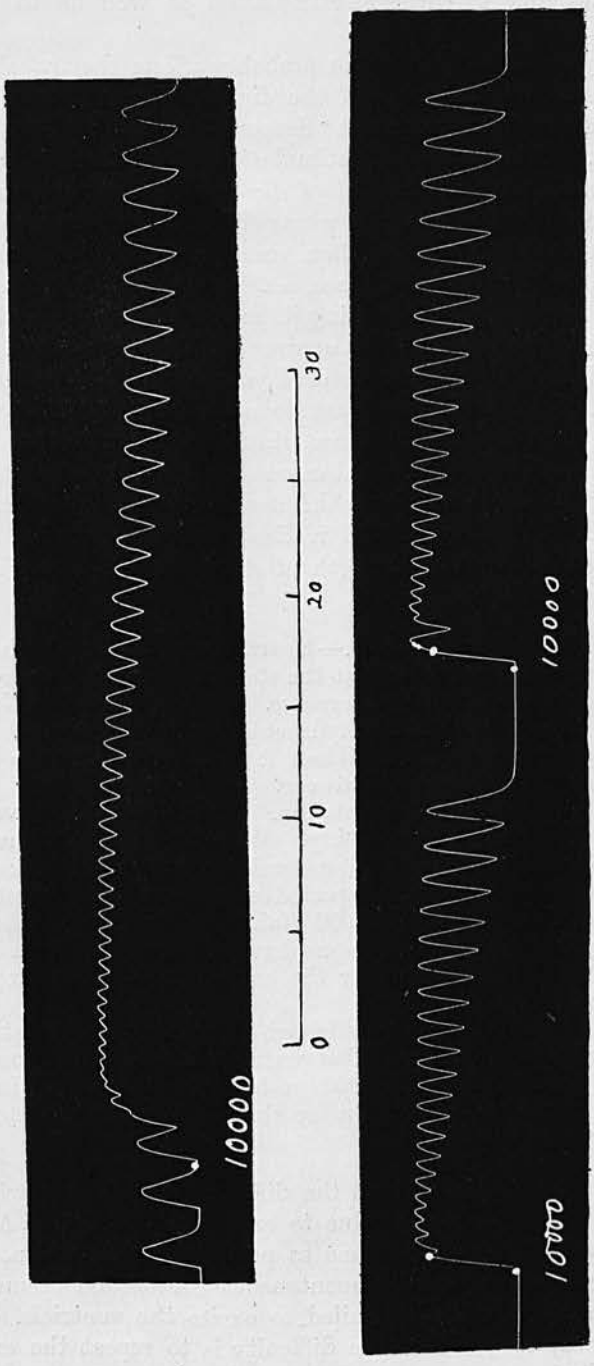

FIG. 4.—To show abnormal effect due to powerful electrical stimulation of the normal perfused heart.

This effect is probably due to chemical stimulation from electrolytic products formed at the base of the ventricle and inside the metal perfusion-cannula. The degree of the effect is proportional to the intensity of the exciting current. This is, however, not shown in the present tracings, of which the upper (single) tracing is from a different heart from that which gave the lower (double) tracing. (The time-scale is marked in intervals of five seconds.)

depresses the irritability to chemical stimulation as well as to induced currents.

We have now seen that yohimbine probably affects the irritability of the heart in the same way as drugs of the digitalis series (Straub). To use Alcock and Meyer's expression, it "deepens" the relatively refractory phase throughout. This means that stimulation of a strength sufficient to excite the normal heart at the end of diastole or in post-diastole fails entirely at any phase of post-diastole whatever to excite the same heart when yohimbinised. The question then comes to be: Does yohimbine prolong the relatively refractory phase, i.e. is the yohimbinised heart slower than the normal heart in regaining its maximum irritability?

It has not been so easy to obtain an answer to this question as I at first imagined it would be. Under any dose of yohimbine sufficient to cause marked heart-block the ventricular beats do not always occur at regular intervals, and it is then difficult to know whether the contractions which do occur from time to time are spontaneous or due to the externally arising stimulus. The only index by which one can judge is the duration of the latent period. It was not until I realised that the duration of this period is itself dependent on the strength of stimulus that I was able to reach a definite conclusion.

Experiment, November 15, 1909.—Heart first perfused with blood mixture. Though beating irregularly at the start, it soon becomes regular, and the rate of the beats gradually increases. At the end of an hour it is perfused with 1 in 5000 yohimbine hydrochloride, upon which it comes to a standstill in extreme diastole. When it is stimulated with single strong induction shocks it contracts, however.

Fig. 5 shows the effect of such stimulation. To 6000 units ($2\frac{1}{8}$ volts in primary circuit) it responds fairly quickly. With 5000 units stimulation the latent period is longer. To the first excitation at 4000 it does not respond, contracting with long latent period only when the excitation is repeated some little time later. To 3000 (fairly late in post-diastole) it responds with long latent period. A considerable interval elapses before it responds to 2000, and that only after the excitation has been repeated thrice—latent period is again long.

A rest of about a minute was now allowed, after which stimulation at 2000 was thrice tried, without effect. The ventricle now, however, contracts of its own accord. Two stimuli of 15,000 units give contractions in each case with short latent period. Shortly after this the heart began to beat at a slow but fairly regular rate.

The experiment just cited illustrates the difficulty of deciding whether a given contraction is spontaneous or due to artificial excitation. At one point stimulation with 2000 units seems to provoke a contraction. Not very long afterwards an apparently spontaneous contraction occurs just after exactly similar stimulation has failed to excite the ventricle. It is clear that the only way to get over the difficulty is to repeat the experiments sufficiently often.

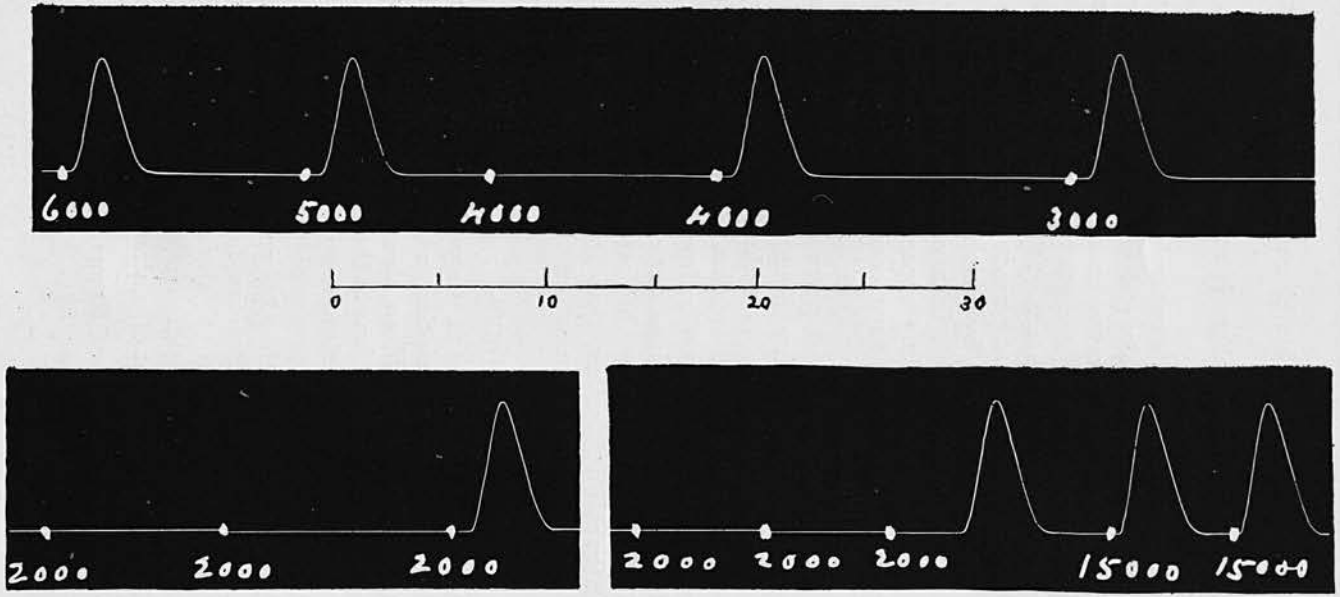

FIG. 5.—To show the differences in latent period corresponding to different strengths of stimulation under the action of yohimbine.

The upper tracing and the first part of the lower tracing are in continuity. Between the first and second part of the lower tracing a rest of about a minute intervened. Concentration of yohimbine solution, 1 in 5000.
The figure also serves to illustrate the fact that the relatively refractory phase is prolonged by yohimbine. (Cf. the effect of the two successive excitations at 4000 units and the first three successive excitations at 2000 units.)
(The time-scale is marked in intervals of five seconds.)

The result of a number of experiments carried out in this way has shown:—

(1) That the latent period of the yohimbinised heart is longer for weak stimulation than for strong.

(2) That the relatively refractory phase is not merely deepened, but is also greatly prolonged by yohimbine.

The first of these two statements is a general one, true of the normal as well as of the yohimbinised heart, of skeletal as well as of heart muscle. Cf. Bornstein (23). Yohimbine, however, exaggerates the differences in latent period corresponding to varying strength of stimulation.

Analysis of the subject by Tigerstedt (24) in the case of skeletal muscle has shown that the shorter latent periods occur when all the fibres are simultaneously thrown into action by spread of the current owing to excessive stimulation; the whole muscle is in consequence excited at the same moment and the attached recording apparatus moves more quickly. The longer latent periods, on the other hand, correspond to contractions in which the portion in the immediate vicinity of the electrodes first contracts, followed in succession by contraction of parts more remote, the impulses being in this case conducted by the living tissue to the more remote parts. The fact that yohimbine exaggerates these time differences, making the latent period corresponding to weak stimulation longer than it usually is, points to a slowing of the normal conduction from fibre to fibre.

We may therefore say that yohimbine depresses the normal irritability of the heart, and delays the restoration of full irritability after contraction; further, that it slows the conduction of impulses from element to element of the heart-wall.

(d) Fatigue.

That fatigue phenomena are present in the yohimbinised heart has been noted by Müller. I quote his description, which contains the essential facts:—"In conclusion, I wish to add an observation which shows that sometimes in the poisoned heart exceptions to the all-or-nothing law occur. The isolated apex was beating in a mixture of frog's blood and 0·7 per cent. saline solution. It was stimulated by means of the polyrheotome and the curves recorded. After poisoning with $\frac{1}{1000}$ yohimbine (lactate) solution in a similar mixture, the height of contraction fell in ten minutes with unaltered strength of stimulation almost to the abscissa. The heart muscle was very readily fatigable, and with varying strength of stimulation in six-second rhythm the following values for height of contraction were obtained:—

1st	contraction	with	coil-distance	of	50	mm.	. . .	4·2	mm.	height.
2nd	„	„	„	„	50	„	. . .	1·2	„	„
3rd	„	„	„	„	50	„	. . .	0·4	„	„
4th	„	„	„	„	30	„	. . .	1·1	„	„
5th	„	„	„	„	0	„	. . .	1·6	„	„

"Thus, in contradistinction to the normal rule, the response to stimulation depends on the strength of the stimulation."

To the above account some further details and discussion may be added. The fatigue is observable in every case where yohimbine is perfused in sufficient concentration. Fig. 6, which shows the general nature of the phenomenon, reminds one of the fatigue effects obtained by Tait and Gunn in a nerve-muscle preparation to the nerve of which yohimbine had been locally applied. (See (15), p. 200.) In both cases—nerve-muscle preparation

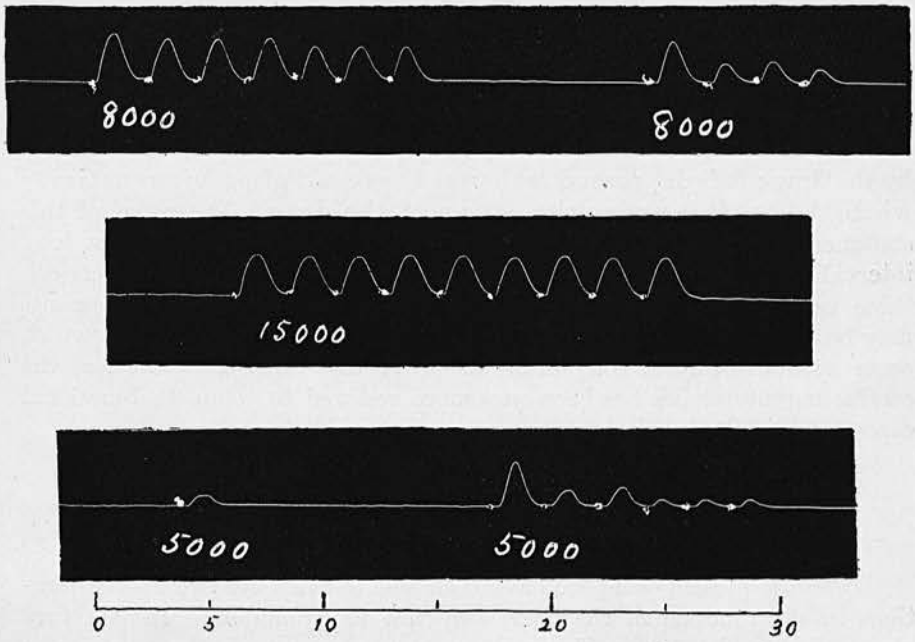

FIG. 6.—To show fatigue effects in the yohimbinised heart.

In the upper tracing one sees marked fatigue with 8000 Kronecker units.
The middle tracing, taken immediately after the upper, shows no apparent sign of fatigue when the strength of stimulation is changed to 15,000 units—this effect is due to direct spread of the current.
In the lower tracing, taken at another stage of the experiment, one sees recovery from fatigue with time.
Concentration of perfused yohimbine, 1 in 5000.
(The time-scale is marked in intervals of five seconds.)

and heart ventricle—repeated excitation causes a progressive diminution in functional capacity, which is recovered from with adequate rest. The similarity is even more close than this, for the present fatigue does not involve the actual contracting mechanism. This is shown by the fact that more powerful stimulation is able to elicit full contractions of the heart. It is fatigue of the conducting mechanism. Inspection of the heart through the glass wall of the plethysmograph shows that in this condition only a localised portion of the heart muscle is involved in each contraction, while the stimulus arising in this portion does not spread all over, as in the normal heart.

The subject has already been discussed by Bornstein (23) with direct reference to this very question of the yohimbinised heart. Similar interference with conductivity is a recognised phenomenon in the action of other heart poisons. Cf. Böhme (25) and Schultz (5). The explanation of the more powerful contractions obtained with strong stimulation is the same as that already adduced to account for the shorter latent period corresponding to strong stimulation. In these full contractions conductivity is really almost abolished. Its absence is masked, however, by the counterbalancing effect of spread of the current. We are thus provided with another proof that the property of conductivity in the heart is interfered with by yohimbine.

At the same time, and this is a point which requires to be emphasised, we are dealing in this case with a definite fatigue phenomenon. It would not account for the condition to say simply that conductivity is depressed by the drug; it is depressed rather as the result of previous activity while the heart is under the action of the drug. The proof of this statement is the fact that the deeply yohimbinised heart executes at long intervals full spontaneous contractions. Tested in the intervals between these contractions, the conductivity between individual muscle-segments may be found to be more or less abolished. The full contractions which occur spontaneously at long intervals prove that with sufficient rest the conducting mechanism has been once more restored to complete functional capacity.

Part III. Excito-Conductivity and Heart-Block

(a) Excito-Conductivity one Single Property.

Up to the present point we have been able to attribute two main alterations in the function of the heart ventricle to yohimbine. In the first place, the conductivity from element to element of the ventricular chamber is modified. This modification is twofold: (i.) conductivity is slowed—this we deduce from the prolonged latent period and likewise from the fact (not hitherto mentioned), established by Müller, that the A_s–V_s interval is increased; (ii.) the conducting mechanism is subject to fatigue —a deduction made from the extent of the ventricular response when tested after varying intervals of rest. In the second place, the relatively refractory phase is prolonged; this implies an alteration in irritability. These alterations are both present at a time when the power of contraction possessed by each cell of the ventricle is undiminished. Indeed, I have never seen a "negative inotropic effect," i.e. a lessened power of contraction of the individual cells, as the result of yohimbinisation. The heart brought absolutely to a standstill by the strongest solution used by me always responded with full contraction to powerful stimulation. My observations in this regard are at variance with those of Müller.

Whether the two properties, irritability and conductivity, are in the

case of the heart separate and distinct things, or whether they are indissolubly interconnected, is a point that has not yet been determined beyond cavil. The view put forward by Engelmann (26) and held by physiologists of his school is that they are distinct. On the other hand, we know, thanks to the work of Werigo and Rajmist (27), of Wedensky (12), of Fröhlich (28), and especially of Boruttau and Fröhlich (29), that in the case of nerve conductivity and irritability, once believed to be separate properties, Schiff (30), Grünhagen (31), Gad, Sayer, and Piotrowski (32), etc., are in reality expressions for one and the same thing.

The fact that in one physiological structure irritability and conductivity, generally held to be distinct properties, have been shown to be merely different aspects of one and the same fundamental property, is presumptive evidence that in other tissues in which we recognise their coexistence they are likewise aspects of a single common property; and it is an interesting fact that in the special case which we have had under review the two things are found to vary together. To tell the truth, the distinction which Engelmann is able to draw between irritability and conductivity is more theoretical than experimental, and is not convincing in the same way as his separation of irritability, say, from contractility, or of conductivity from the power of stimulus-production.

If, then, irritability and conductivity are essentially the same property, it follows that the internal mechanism responsible for their existence is a single thing. We shall call it the excito-conducting mechanism.

This is merely another statement, arrived at by a slightly different process of reasoning, of the view held by Kronecker with regard to the physiological properties of the heart (33). Kronecker contends that cardiac muscular tissue is in itself inexcitable except through the mediation of nerves, and that all conduction from fibre to fibre takes place by means of nerves. Without going so far as to specify the actual structure responsible for the property of excito-conductivity (for in regard to this the present communication furnishes no evidence), it is perfectly justifiable to hold the view that what Engelmann has distinguished as four separate properties of the heart are in reality only three, two of Engelmann's "cardinal properties" being fundamentally identical.

(b) Toxic Heart-Block.

On the above described hypothesis, viz. that the mechanism responsible for the irritability of the heart is the same as that responsible for its conductivity, it follows that the refractory phase (a condition defined in terms of irritability) is a phenomenon which directly involves the conducting mechanism. This statement does not imply dissociation of refractory phase from the phenomena of the contractile mechanism. On the contrary, there is the closest relation between the excito-conducting and

the contractile mechanism. Thus, the only available apparatus in the heart by which we can register change in irritability is the contractile mechanism itself; further, as we have already seen, the phenomena of the refractory phase are experimentally found to be closely related to the events of contraction.

We know that the heart muscle is ready to continue in contraction for any moderate length of time, provided only that it can be appropriately excited—the phenomenon of "heart-tetanus" shows this. It is no unfair assumption that in the case of an ordinary heart-beat the excito-conducting mechanism, having once played its part in producing contraction, is throughout the remaining period of this contraction kept in a state of paralysis or exhaustion, unable to undergo anabolism owing to the demand made upon the available energy materials by the contracting mechanism. Either this or the excito-conducting mechanism functions to the full during practically the whole of contraction. In any case, we have to account for the fact that repair of the excito-conducting mechanism is for the first time possible when contraction is virtually over. When repair of this mechanism does set in, the rate at which it occurs depends on factors which are not the same as those leading to relaxation of the muscle. This we deduce from the fact that the time consumed in muscular relaxation is not an accurate measure of the duration of the relatively refractory phase.

If the refractory phase of the heart is attributable in the first instance to the excito-conducting mechanism, one must recognise two main differences in behaviour between the cardiac excito-conducting mechanism and those analogous mechanisms which, occurring elsewhere in the body, we know as nerves:—(i.) the cardiac excito-conducting mechanism is less independent of the effector organ, in this case the contractile mechanism, being able to undergo repair only when the contractile mechanism has slowed down in activity; (ii.) the repair process, once started, takes place more slowly than in nerves. The second difference is one of degree rather than of kind. The first difference points to a closer functional, and probably anatomical, relationship between the excito-conducting and the contractile mechanism than we find, for example, in a nerve-muscle preparation.

It is still, however, permissible to argue in general terms from the case of a pure excito-conducting mechanism, such as a medullated nerve, to the more complicated form which exists in the heart. Now, Boruttau and Fröhlich (29), summing up the chief modifications in function produced by depressing agents, e.g. anæsthesia, asphyxia, low temperature, on medullated nerve, specify four individual changes which are always found associated:—

(i.) Slowing of rate of conduction;
(ii.) Diminution in intensity of impulse, whether measured electrically or by effect on attached organ;
(iii.) Prolongation of refractory phase;
(iv.) Prolongation of electrical wave.

To this may perhaps be added the fact that fatigue changes are more readily induced in the nerve. As said above, these changes all go together; when one is present the others are present at the same time.

Suppose, now, that the excito-conducting mechanism of the heart is influenced in a similar way by depressing agents, we should have at once an array of conditions, almost any one of which, taken solely by itself, might conceivably be considered a cause of heart-block, and as a matter of fact the modifications of function which have at different times been adduced by different observers to account for toxic heart-block are found among the above specified group. But if the excito-conducting mechanism of the heart is modified by depressing agents in the same way as nerve, all these changes must be present at the same time. Whether they are or not can of course be determined only by experiment. In any case, it is clear that an explanation of heart-block based on prolongation of refractory phase may be—however unlikely at first sight—fundamentally identical with an explanation based simply on deficient conduction.

IV. Summary.

This communication consists of a special and of a general part, of which the latter is the more important. The special part deals with the action of yohimbine on the heart. The general part arises out of the special, and includes a discussion on three of the four cardinal properties of heart tissue recognised by Engelmann, viz. conductivity, irritability, and contractility.

A. Special:—

(1) Yohimbine does not alter the absolutely refractory phase of the heart, or, to be more precise, it does not affect the ratio A/S, where A = duration of absolutely refractory phase and S = time of systole.

(2) In sufficient concentration it greatly prolongs the relatively refractory phase.

(3) It depresses the irritability of the tissue to induced currents and probably to chemical stimulation.

(4) In concentration sufficient to bring the heart to a standstill it has no depressing action on contractility.

(5) Under yohimbinisation the conducting mechanism as distinct from the contracting mechanism of the heart is readily fatigued.

B. General:—

Evidence is adduced in support of the following conclusions:—

(1) Irritability and conductivity in the heart, just as in the case of nerve, are essentially the same property. No change in irritability can occur without a corresponding change in conductivity and vice versa. When Engelmann attempted to separate the two properties he was led astray by a wrong assumption.

(2) It follows from the above that the internal mechanism responsible

for the existence of irritability and conductivity is a single thing. We may for convenience call it the excito-conducting mechanism.

(3) It also follows, provided the distinction initiated by Engelmann between irritability and contractility is upheld, that the heart muscle is capable of excitation only through the excito-conducting mechanism.

(4) The excito-conducting mechanism is in more close functional and possibly structural relationship with the contractile mechanism than are the nerve fibres with the contractile mechanism in a skeletal muscle. After activity the cardiac excito-conducting mechanism undergoes repair at a much slower rate than ordinary nerve fibres.

(5) Toxic heart-block, which has received different explanations at the hands of different observers, is due to a depressing action on the excito-conducting mechanism. The individual explanations advanced to account for the condition are all aspects of one complex change, each feature of which is present at the same time, while no single one can be present without the others.

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