REACTIONS
OF
COOLED NERVE

with especial relation to its

FATIGABILITY.

(An Experimental Research)

Thesis submitted for the Degree of D.Sc. by

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C O N T E N T S.

INTRODUCTORY.

The present thesis contains a record of a number of experiments on the conductivity of the cooled sciatic nerve of the frog. To make clear the general bearing of the work a certain amount of historical introduction is given.

Experimental work on the cooling of frog's nerve falls naturally into two divisions according to the temperature to which the nerve is cooled. In the one case the temperature, while lowered, may still fall short of the freezing point of the nerve. In the other case the nerve may be frozen, and the changes investigated which go on both during the process of freezing and after a subsequent thawing. In the present instance discussion is limited to experiments in which the temperature was not lowered to such an extent as to freeze the nerve.

The attached gastro-cnemius muscle was alone used as an index of the nerve condition: the electrical reactions were not examined. Indications of fatigue were in many cases elicited in the cooled nerve/

nerve. The subject of fatigability of nerve consequently comes up for discussion.

The research of which the thesis forms the subject was undertaken at the suggestion of Professor Verworn in the winter of 1903 - 4 in the physiological laboratory of Göttingen University, and was continued from November 1905 up to the present time in the physiological laboratory of Edinburgh University under the direction of Professor Schäfer.

PART I. HISTORICAL.

Section A:- Grünhagen's Method:

Conductivity v. Excitability.

In the year 1872 Grunhagen[1] by the introduction of a new method into nerve physiology, gave a start to a most important series of investigations. Already in 1858 Schiff[2] from the local variations in excitability along a nerve, and from the behaviour of a nerve when a portion of it was subjected to the action of a constant galvanic current, had argued that nerve possesses two distinct properties, excitability, or the faculty of being excited at any point by an external stimulus, and conductivity, or the faculty of transmitting the excitation so caused from one part of the nerve to another.

These observations were soon backed up from the clinical side by Duchenne de Boulogne, Ziemssen and Weiss, Erb, Eulenberg[3] and others. They showed that in certain peripheral paralyses a nerve supplying definite muscles might be incapable of responding to electrical stimulation, while the patient might still be able to move these same muscles at will. In other words nerves might retain the property of conducting impulses from centre to periphery and yet be incapable of/

of excitation by external stimuli. Ziemssen - Weiss and Erb[3] went further. They carried out experiments on frogs by nipping across certain nerves with forceps and then studying the reactions during the process of repair. Conductivity returned before excitability

Munk[4] carried out a very careful series of investigations on the dying sciatic nerve of the frog, and found that while excitability might be greatly diminished or even lost in peripheral portions, a relatively slight stimulus applied to a more central portion might still cause a full contraction of the muscle.

All these experiments, however lacked definiteness It is to the credit of Grünhagen [1] (see Fig.1)

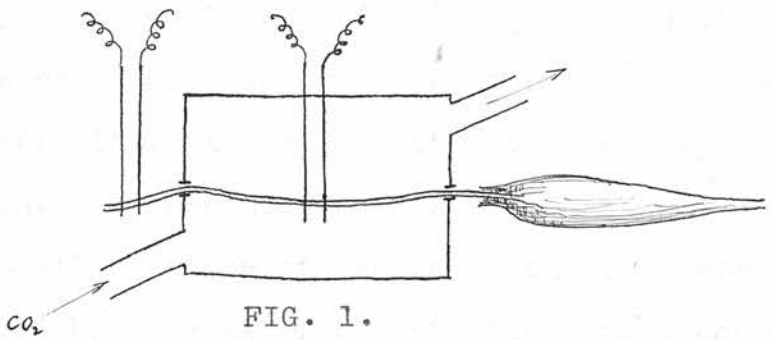


FIG. 1.

that he instituted a method by which the existance of these two separate properties was clearly established and by which the extent of their independence might be with some exactness determined. He enclosed the middle portion of a frog's sciatic nerve in a chamber through which a constant stream of CO_2 gas could be passed./

passed. The middle portion was thus subjected to the influence of the gas while the portions outside the chamber remained unaffected. On stimulating after the lapse of some time by means of electrodes fixed inside the chamber he found that the excitability had undergone a marked diminution, "while on the other hand," to quote his own words, "the excitability of the central portion of the nerve, which was not subjected to this narcotic influence remained constantly at the same height ---- although the excitation started in it had to traverse the peripheral narcotised part. From this, one is justified in concluding that nerve excitation and nerve conduction should be separated as different processes."

With the validity of Grünhagen's deduction from his experiment we are however not immediately concerned; it is the method that claims our attention. It presents these special advantages:- (1) a localised piece of the nerve can be homogeneously acted upon by some external condition (presence of gas) to the exclusion of the rest of the nerve: (2) the influencing agent, being electrically non-conducting, does not prevent the use of electrical stimulation, thus still affording quantitative measurement of the stimulus; (3) the length of the portion influenced by the gas can be varied at will by varying the size of the chamber. Finally its result/

result shews that we have in it a most valuable means of determining the extent to which conductivity and excitability are independent of each other.

Since these experiments a hot dispute has raged round the question of the relative independence of conductivity and excitability, and the multiplication of observations on the subject made the question for a time very complicated.

Szpilman and Luchsinger[5] using the vapours of ether, chloroform, alcohol, and ammonia, distinguished two stages in the process of narcosis. In the first stage the excitability is lowered while the conductivity remains unchanged; in the subsequent stage the excitability is not yet absent, though greatly lowered, while the conductivity is totally gone. They pointed out that Grünhagen by using the weaker agent CO_2 had got only the first stage. The two stages of narcosis which these two observations thus established are of fundamental importance and will often be referred to. These results correspond entirely with those of Efron,[6] who extended his observations to include the influence of various alcohols, different pressures, and different temperatures. Hirschberg[7] repeated Grünhagen's CO_2 experiments while also using different temperatures. He did not get by this means the second stage of Szpilman and Luchsinger.

Gad/

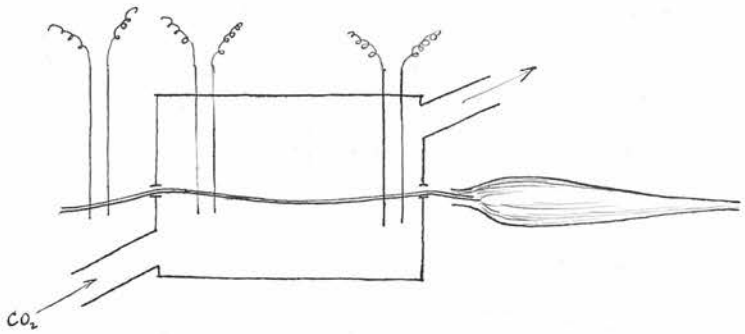


Fig. 2.

Gad, Sawyer and Piotrowski[8] using various methods were able to repeat all the work of Szpilman and Luchsinger, besides directing attention to a new fact, viz. an initial rise in excitability and to a less extent in conductivity on narcosis. They also introduced an important modification into the method., vix. they put twp pairs of electrodes inside the chamber used for anaesthesia, one peripheral and one more central, although the observations they carried out in this way have since been shewn to be erroneous (see pp. 15 - 16)

Gotch carried out a series of observations with ether and chloroform, in which special attention was directed to the recovery of the nerve after anaesthesia. He got the two stages recognised by Szpilman and Luchsinger, while during recovery he found that conductivity returned somewhat rapidly, excitability on the other hand returned more gradually. Some experiments carried out under Gotch's direction shewed that the second stage of nerve anaesthesia could be reached by the use of CO_2 gas, while recovery occurred in the same order as after chloroform or ether.

Waller[9] used isolated nerve and took the electrical response as an indication of the condition of the nerve. His obeervations, however, rather emphasised individual differences in the narcotic agents/

agents than helped forward generalisation on the question of the fundamental properties of nerve.

The question of the relation of conductivity to excitability had now become thoroughly complicated. As an indication of the difficulty of the subject at this stage one need only refer to the account given of it in some of the modern text-books of physiology. The careful and impartial summary of the evidence by Biedermann in his "Elektrophysiologie," 1895, p.p. 493, f.f., shews better than anything the need for some general principle to explain the facts. It tends to leave the mind of the reader rather in bewilderment than in enlightenment. Again the section relating to "the influence of anaesthetics" on nerve in Schäfer's "Textbook of Physiology," Vol.II, 1900, p.p. 491-493 is more a series of observations on individual anaesthetic agents than a generalisation. The fact is, it was not possible to bring order out of the experimental facts at the time when these text-books were written.

As an interesting commentary on the difficulty of coming to definite conclusions on the subject, one may take the following two sentences, which Biedermann in his text-book, as if glad of an opportunity to generalise for once, prints in italics:- "From these (the experiments of Grünhagen, Luchsinger, Gad, Piotrowski, etc.) the very significant fact appeared that/

that with local application of alcohol vapour, ether or chloroform, conductivity was as a rule first and most fundamentally affected before excitability underwent any perceptible diminution. With CO_2 on the contrary, as well as CO, conductivity is quite unaffected, while local excitability is quickly abolished." Not only is the statement in the first sentence a very doubtful deduction from the experiments quoted, but the separation of the action of CO_2 from that of other anaesthetics was soon proved wrong by the experiments just referred to as being carried out under the direction of Gotch. (P. 7.)

The subject was soon to be cleared up in considerable measure. To understand how this was done we must go on to other experiments carried out by slightly different methods.

SECTION B. WEDENSKY'S "PARADOXICAL CONDUCTION."

We have next to consider an investigation which Wedensky [10] carried out on the fundamental properties of nerve. This observer anaesthetised the nerve by painting a portion of it with cocaine, chloral hydrate, or phenol. As indicators of the nerve condition he used simultaneously the telephone and the muscle response. The use of the telephone involved that the stimulation/

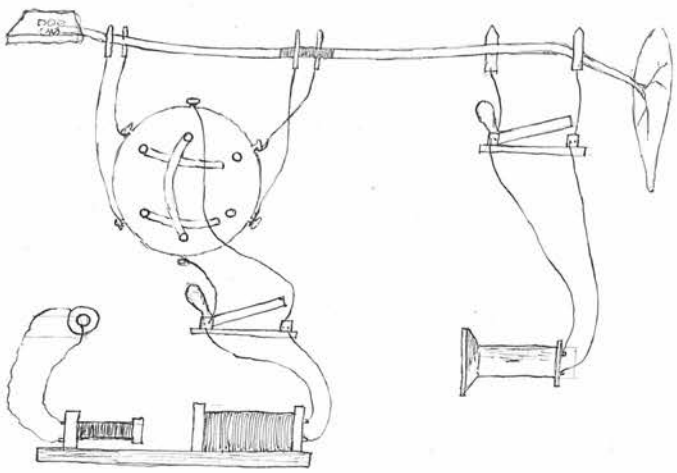


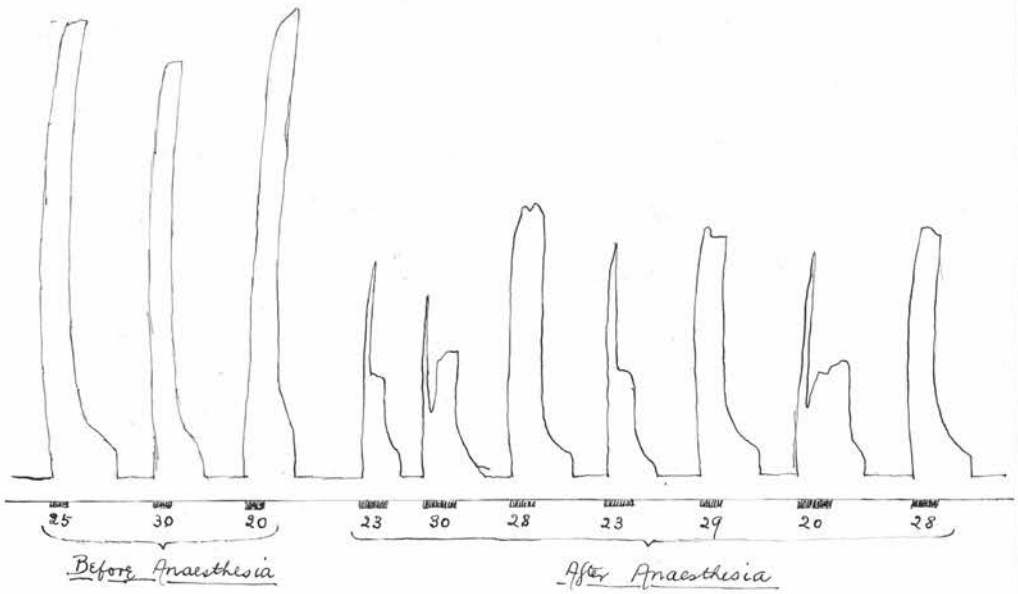
Fig. 3.

the stimulation of the nerve should be rhythmic and rapid, - a tetanus of 100 per second he used as a rule. The responses of the muscle were accordingly tetani.

By means of the muscle record it was easy to confirm the existence of the two stages of Szpilman and Luchsinger. During the first stage the excitability gradually fell away, as shown by the shorter and shorter distance of secondary from primary coil required to elicit minimal tetani in the muscle. The conductivity meantime remained constant. The second stage set in suddenly. All at once the conductivity began to fall rapidly and in a few seconds entirely disappeared. The excitability by this time was low, and it continued thus during further anaesthesia.

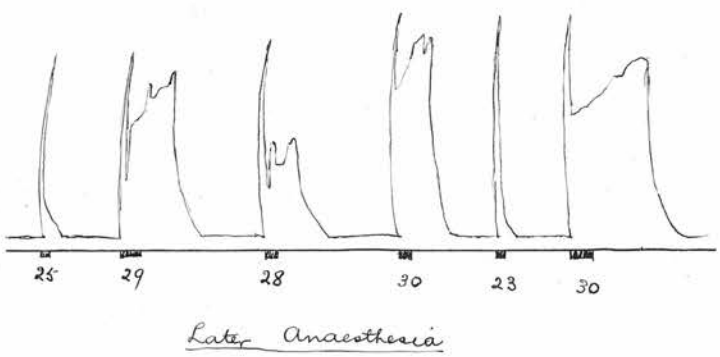
On using the telephone note however as indicator a remarkable phenomenon occurred. So long as he tested the excitability alone, the telephone record and the muscle record ran exactly parallel to each other, i.e. to keep the intensity of the telephone note constant during the progress of the anaesthesia he had to shift the secondary coil nearer to the primary, and at the same rate as he found necessary in order to elicit minimal tetani of the muscle.

When he tested conductivity on the other hand he/



(Curves reproduced from Wedenskyi's Paper)

Fig. 4



(From Wedenskyi's Paper)

Fig. 5.

he noticed that during the first stage, - the stage of apparently "undiminished" conductivity as shewn by the muscle record, - the tone underwent a considerable change. The note became weak, dull and mixed up with extraneous noises, and this was the case for all intensities of stimulation during the first stage. This change was a progressive one, becoming more and more pronounced as anaesthesia went on.

His next observation was that in a certain stage of anaesthesia the tetanic contractions of the muscle (as elicited by stimulating centrally) were not as full and complete as before anaesthesia, and this was the case whether he used strong or moderate tetanising currents. Again, the tetani elicited by strong were weaker than those elicited by moderate stimulation, and more resembled those resulting from very weak tetanisation. The effect was in no way due to diminished conductivity of the peripheral end-apparatus, for peripheral stimulation with a third pair of electrodes caused full and energetic contractions. (See Figs. 4 and 5).

On further progress of the anaesthesia he found that strong tetanisation produced only an initial twitch of the muscle, (with often a final twitch in addition), while stimulation at moderate intensity caused tetanus. The telephonic record corresponded exactly;/

exactly; 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.

This remarkable and perfect parallelism between the records of the muscle and the telephone naturally surprised Wedensky. To make sure that the telephone sound was no backwardly propagated effect from the muscle itself, he put a drop of creosote on the nerve close up to the muscle. Thereupon the contractions of the muscle ceased, while the nerve continued to give the same sounds as before. When he put creosote between the "conductivity" and the "excitability" electrodes the telephone tone disappeared.

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. A "paradoxical modification" of the conductivity is established, in that rhythmical tetanisations are not transmitted regularly; further strong impulses are transmitted with greater and greater difficulty, while weak excitations are handed on without prominent change.

It was interesting to see if the galvanometer would bear out the results of the telephone and of the/

the muscle. The use of the galvanometer involves the interruption of the vital continuity of the nerve. Wedensky consequently brought the anaesthesia to a certain stage, keeping both nerve and muscle intact. After recording the muscle response he quickly killed a portion of the nerve with creosote and led off from the nerve the galvanometer current.

The results confirmed in the most striking fashion his previous observations. During the first stage of anaesthesia, provided strong shocks were used, the galvanometer records of stimulations starting at both central ("conductivity") and peripheral ("excitability") electrodes ran parallel to each other from the start i.e. they shewed a regularly progressing diminution of both conductivity and excitability. When the conductivity had disappeared, as shewn by muscle and telephone, i.e. after the second stage of anaesthesia was established, the galvanometer no longer indicated the presence of any negative variation. When he stimulated peripherally, (excitability) the stronger induction currents still remained for a considerable time effective, but gradually fell off in amount as anaesthesia further progressed.

These results of Wedensky are of high importance. As we/

As we see, they were reached by employing rapid tetanisations of the nerve instead of single excitations, and by directing attention not to responses of the muscle elicited merely by minimal stimuli but to the result of stimuli of all dgrees of strength; while the muscle records were contrclled at every point by means of the telephone and of the galvanometer.

They were confirmed by Boruttau(11) who likewise investigated the condition by means of the capillary electrometer. He writes; "I have been able to confirm out and out the discoveries in a series of analogous experiments, and have also found by registration of the movements in the capillary electrometer that at the commencement of ether as well as of cocaine narcosis of a complete nerve, the steepness of the curve and the backward drop ("definitive Ablenkung") of the meniscus is considerably diminished, i.e. when the nerve is strongly stimulated with very frequent currents (acoustic interruptor); while the curve of the current of action with its single peaks show scarcely any change on stimulation with less frequent and weak induction shocks." Boruttau likewise reports that he cannot appraise too highly the telephone as a means of investigating changes in nerve condition.

Wedensky/

Wedensky further found that when he tested the excitability of a narcotised piece of nerve by means of electrodes applied to the anaesthetised portion, the phenomena altered with alteration in the direction of the exciting current. When the effective current had an ascending direction, so that the point from which the stimulus started lay more centralwards, the phenomena more resembled those got from the "conductivity" electrodes; when on the other hand he used descending currents to excite the nerve he got a quite regular and gradual fall in excitability. He concluded that the latter method of stimulation gives a record of pure excitability, the former method gives a mixture of excitability and of conductivity. Gad and Sawyer[8] by using two pairs of electrodes within the anaesthetising chamber had found no difference in effect according as they stimulated more centrally or more peripherally.

Werigo and Rajmist[12] in a very exact piece of work had previously reached the same conclusions in this regard as Wedensky, and had pointed out the mistake in Gad and Sawyer's experiments, while Tiberg,[13] working under Wedensky's directions, had in 1895 shewn that in the later stages of narcosis each 1 mm. added to the narcotised nerve causes an increased/

increased hindrance to the progress of the excitation. Curiously enough Gotch and Macdonald[14] apparently from purely a priori considerations, had pointed out the fallacy that might arise from testing for excitability at any arbitrary point along the influenced portion of a nerve subjected to local depressing influence. The importance of this precaution has since been confirmed by many investigators.

[The present occasion may be used to discuss a question which, though not directly bearing on the subject in hand, is yet of considerable theoretical importance, viz. the relation which the electrical change bears to the actual excitatory process in a stimulated nerve. The intimate association of the one with the other has again and again been clearly proved - the experiments of Wedensky on "paradoxical conduction" are only a further illustration of the fact. While there is distinct evidence that the electrical may exist without the excitatory process, e.g. the dissected out nerves of warm-blooded animals may be incapable of causing contraction of the attached muscle when stimulated, and yet give the electrical response (Hermann[15], Fredericq[16], Gotch and Horsley[17]-); still, until quite recently (Gotch/

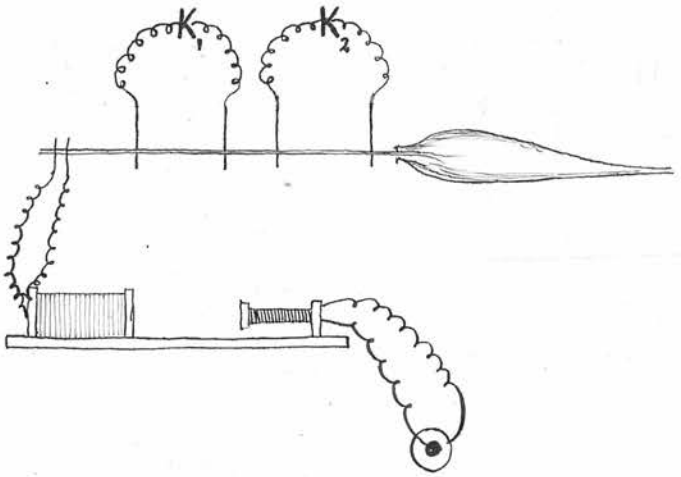


Fig. 6.

(Gotch and Burch[18]), there has been no evidence of the existence of the excitatory alteration without the electrical.

These experiments of Wedensky suggest an interesting question. A portion at least of the electrical energy can be tapped on the progress of the excitatory wave along the nerve; it is this energy that causes the vibrations of the plate inside the telephone. Does this draining away of electrical energy detract from the ability of the excitatory process to stimulate the muscle? In other words is the electrical change causally related to the contractions of the muscle, or is it a mere bye-product in the generation of the excitation along the nerve?

To investigate this question I carried out a few experiments on submaximal stimulation of muscle (1) without, and (2) with, two electric circuits applied to the side of the nerve as shewn in fig.6.

To give a steady current an accumulator was used in the primary circuit. The "break" was affected by means of a Hermann falling key. The resistance of the two applied circuits was low (about 2 ohms.) The leading off electrodes were of platinum and were arranged at varying distances apart (5 mm. to 2 cm.) One or two submaximal break constrictions were first/

were first elicited with the applied currents open, the time interval between the individual stimuli being half a minute.

The keys K_1 and K_2 were then closed and similar records taken after an interval of one minute. The process was repeated a number of times. No regular difference in the height of contraction was observed according as the applied circuits were open or closed. Charts 1 and 2 shew typical examples of the kind of result.

These experiments are naturally rough and inconclusive. Although it was assumed on general physical grounds that energy must be used up in heating the applied circuits, no means were taken to demonstrate that a current actually traversed the extra circuits. A repetition of these experiments with galvanometer and telephone, careful account being taken of the relative quantities of electricity involved and of the length of the electrical wave, might lead to a more definite conclusion.]

SECTION C. QUESTION OF OXYGEN SUPPLY FOR NERVE CELLS AND NERVE FIBRES.

Meantime a new series of investigations had been/

been going on, which though at first directed towards the elucidation of processes within nerve cells, ultimately came to bear on the fundamental properties of nerve fibres. These investigations were started by Professor Verworn, and continued under his direction by his pupils in Göttingen. From our present point of view their first important result was the discovery by von Baeyer that in the presence of an oxygen-free neutral gas a nerve after some time loses both its excitability and its conductivity; on restoration of Oxygen these properties return.

(1) Nerve Cells.

Verworn[19] in an investigation into the metabolic processes in the nerve cells of the frog's cord, had used strychnine to increase the dissimilation relatively to the assimilation. The ultimate paralysis which follows upon greatly increased activity of the cells after injection of strychnine he found to depend in high degree on the fact that the cells had used up their available supply of oxygen. When exhaustion had set in he perfused the aorta with normal saline, (the only possible food naterials being here H_2O and NaCl)- No recovery occurred. When O_2 was present in solution in the normal saline the tetanic convulsions began again and within limits the more/

the more O_2 that was supplied the longer did the renewed convulsions last. The capacity of the nerve centres to undergo excitation he thus directly proved to be a function of the amount of available oxygen.

Winterstein[20] then studied the extent to which the two phases of metabolism, assimilation and dissimilation are affected by narcosis (ether, chloroform, alcohol, CO_2). It had long been known that narcosis interferes with dissimilation. By using Verworn's method of perfusion with solutions of O_2 he shewed that narcosis hinders assimilation to exactly the same extent as it hinders dissimilation.

Archangelsky [21] in 1873 had shewn that when frogs are warmed up to a temperature of 29° - 34° C. clonic and tetanic convulsions appear, which are soon succeeded by complete paralysis. Examination of the peripheral organs shewed that the cause of this paralysis lies in the centres themselves. Winterstein[22] examined this condition and found that, as in strychnine paralysis so in the paralysis of over-heating, the condition of the nerve cells as regards oxygen is a factor of the first importance. With increase of temperature the amount of oxygen used up by the nerve cells increases rapidly in proportion to the amount taken in, so that after a period of greatly heightened activity the cells become exhausted of oxygen./

oxygen. Recovery does not occur with mere cooling; a fresh supply of oxygen must be available.

Next, von Baeyer[23] investigated the effect of low temperature on the assimilation of oxygen in the ganglion cells of the frog. He shewed that in the cold the assimilation of oxygen outweighs its dissimilation, so that in the cold an actual storing up of oxygen takes place.

Curiously enough Regnault and Reiset[24] in 1849 had directed attention to the same fact. They were investigating the condition of hibernation and found that in one case a marmot actually gained 5-9 grms. in weight in five days during profound torpidity. This increase in weight could only be derived from some constituent of the atmosphere. During the hibernation of marmots generally they found that only about two-fifths of the O_2 taken in respiration appeared in the CO_2 discharged.

The results of Winterstein and v. Baeyer were all confirmed and extended by Bondy[25] who further investigated the effect of narcosis, heat, and cold, on the metabolism of the cells of the frog's cord.

2. Nerve Fibres.

It next occurred to v. Baeyer[26] to try if a nerve/

nerve could possibly be paralysed by want of oxygen. He adopted Grünhagen's method, and passed neutral gases (N_2 and H_2), carefully freed from the last trace of oxygen, over the nerve. After a period varying from 3-5 hours both the conductivity and the excitability became abolished. By once more supplying pure oxygen gas to the nerve recovery occurred in 3-10 minutes. These experiments were done at room temperature. With increase of temperature the process of asphyxiation occurred more rapidly. Not satisfied with using indifferent gases alone, v. Baeyer used chemical reducing means. He packed the nerve with iron filings which had been made red hot in an atmosphere of hydrogen, and found that by this means also the conductivity became abolished, to reappear when oxygen gas was once more supplied to the nerve.

The importance of these experiments need hardly be insisted upon. Their interest lies in their suggestiveness. Although they were taken to shew the existence of metabolism inside the nerve yet in the absence of proof of the existence of end-products, such as CO_2 this deduction is not absolutely justified.

In his experiments v. Baeyer (27) had observed phenomena that bore a strong likeness to fatigue. When/

When he stimulated by means of repeated short tetanisations a nerve that had been asphyxiated and again exposed for a minute or two to the air he obtained "first of all only a single response. Thirty seconds later (the stimulation being meantime suspended) recovery was so far advanced that the muscle responded to three stimulations, and a short time later the nerve had again laid up such a quantity of O_2 that even on continuous stimulation no fatigue was to be seen."

v. Baeyer next endeavoured to determine if a nerve becomes asphyxiated more rapidly when it is kept in constant activity. He placed two nerves from the same frog inside the same chamber and exposed them together to the influence of N_2 gas. The one he stimulated continuously at the rate of 800 per second, interposing a block by means of ether narcosis between the asphyxiated portion and the muscle; the other nerve he allowed to remain unexcited. Numerous experiments shewed that in one case the stimulated, in another the non-stimulated nerve became first asphyxiated. Hence this method does not adapt itself readily to shew fatigue in nerve.

To sum up, these experiments lend countenance to the idea, tenable on general biological grounds, that a nerve while it is alive, "respires" in the wide sense in which we talk of respiration in a unicellular/

unicellular organism. That this propable respiration is increased by activity there is a slight amount of evidence to shew.

Now that it was known that a nerve could be asphyxiated Fröhlich[28] examined the effect of narcosis (ether) on asphyxiated nerve, and found that it prevented the assimilation of O_2 - just as had been found by Winterstein to hold for nerve cells. He further found that the condition of narcosis is no bar to the passing of O_2 out from a nerve when surrounded by an oxygen-free atmosphere; whence he concluded that the O_2 stored in the nerve is dependent on the partial pressure of O_2 gas in the vicinity. His experiments, too, shewed in the most marked manner that nerve, once exhausted of O_2, is in a condition of extreme oxygen-hunger, and takes up with the greatest eagerness O_2 offered to it.

This observer next endeavoured to determine if the presence of a reserve of O_2 in a nerve can be definitely proved. He asphyxiated nerves[29] and offered different amounts of O_2 to them, and found that a certain definite amount is necessary to restore completely the excitability and the conductivity. What falls under this amount causes an incomplete recovery. What is over this amount, while causing no further rise in excitability, yet serves to protect the nerve for a longer period against a second asphyxiation/

asphyxiation. This goes far to prove the existence of a reserve store of O_2.

The time that is required to asphyxiate different nerves at room temperature varies within wide limits. v. Baeyer had given a period of 3-5 hours. Fröhlich found a difference of 1-15 hours in different frogs and established the fact (32) that the nerves of animals kept long on the cold take a much greater time to asphyxiate than the nerves of animals that have been kept continuously exposed to a higher temperature. Further, he determined that the faculty of recovery from asphyxia depends on the state of nutrition of the animals. The nerves of badly fed frogs do not shew complete recovery from asphyxia; if the nutrition of the frogs is good the nerves recover perfectly.

I then endeavoured to carry out asphyxiation experiments with warm-blooded nerve. It is well known that the nerve of a living warm blooded animal can be freed for a considerable distance from the surrounding tissues and yet retain its properties for hours together. On the cessation of the blood supply to the muscle of a warm blooded animal inability to contract very soon sets in. For my experiments the peroneal nerve-muscle group of a live rabbit was used. The spinal cord of the animal was divided in the lower dorsal region. The peroneal nerve was separated/

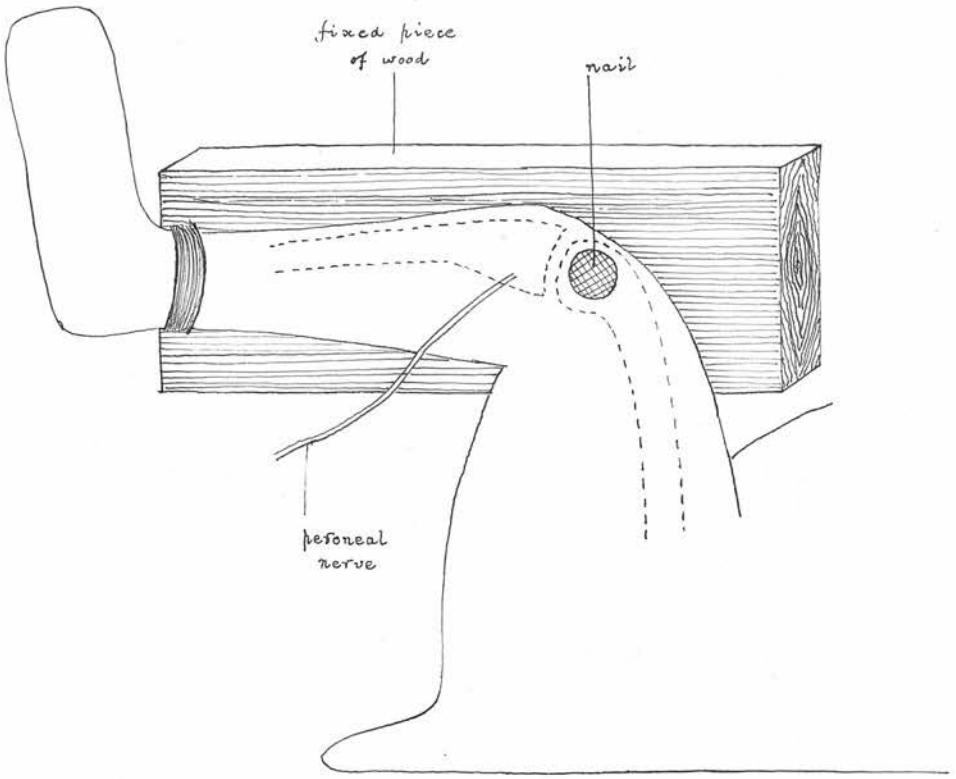


Fig. 7.

separated out from the point where it enters the muscles in the upper part of the leg, right up to its exit from the pelvis, and in this latter situation it was cut across. The separation was easy because the peroneal and tibial nerves in the rabbit are not bound together in one common sheath as is the case in man.

The animal was laid on its back and the lower end of the femur fixed to a rigid piece of wood by means of a flat-headed nail passing through the bone just at the condyles, while one or two turns of tape round the leg just at the ankle served to keep the leg steady. Thus one got a rigidly fixed nerve-muscle preparation, stimulation of which caused flexion of the foot at the ankle. To prevent the possibility of any reflex interference with the responses the tibial nerve going to the calf muscles was likewise divided. The peroneal nerve itself was supported horizontally on electrodes, and lay in a direction perpendicular to the plane of the flexed leg and thigh of the animal (See Fig. 7).

On now surrounding the nerve by a chamber through which N_2 gas was passed, no diminution in either excitability or conductibility was found to occur even though the stream of N_2 was kept going for 15 hours./

15 hours. It then struck me that the persistence of the blood supply might prevent asphyxia. If O_2 were being continuously supplied by means of the blood-vessels the effort to exhaust the nerve from the outside would naturally be fruitless. Control experiments shewed that this conjecture was right, and further I found that a continuous blood-supply is a sine qua non for the functioning of warm-blooded nerve.

Contrary to accepted ideas the nerves of warm-blooded animals possess a richly anastomosing blood supply. The combined peroneal and tibial nerves of the rabbit are in the thigh supplied by at least three visible arteries that enter at different points of their course. By much the largest of these is an artery that enters about the middle of the thigh and sends branches both upwards and downwards along the nerves. On the division of this vessel the central half of the peroneal nerve usually underwent a sudden drop in excitability, often the excitability became quite lost. The half which lay peripheral to the point of entrance of this large vessel remained as a rule fully excitable all along its length. The reason for this was found to be that recurrent vessels from the muscles enter the nerve below, the blood is pumped along these as far as the cut in the afore-mentioned large artery, and there it escapes, (it used)

used to form as an ever-increasing drop which hung pendent on the middle of the nerve). From loss of pressure the blood fails to be pumped further backwards, whence the loss of excitability in the upper half of the nerve. This was beautifully shewn by cutting the nerve across with sharp scissors at different parts of its length. In the upper half - the half which was unexcitable - no blood oozed from the fresh cut section. In the lower excitable half a section at any place shewed bright red blood oozing out from the lower end. Once or twice, too, after the drop of blood forming on the middle of the nerve had clotted, the upper half of the nerve became once more excitable. A cut with the scissors then shewed that blood was again flowing along this part. Obviously by the clotting of the blood in the mouth of the large artery of supply the pressure had become sufficiently raised to drive the blood into the central part once more. It was then found that if this vessel was ligatured before division the excitability of the central half of the nerve remained as a rule quite good.

In the course of a long continued experiment I sometimes omitted to keep the nerve moistened with Ringer's solution. In these cases the central part would/

would become brown, dry and shrivelled, while the more peripheral portion would remain moist and white, and perfectly capable of functioning all the time. The latter condition was obviously due to persistent blood supply. Again, all effects by means of very free dissection to prevent the blood from flowing back into the nerve from the side of the muscles proved fruitless. The blood supply of the muscle and the attached nerve are indissolubly bound up with each other. An injection with methylene blue shewed the presence of fine vessels visible to the naked eye running longitudinally along the nerve into the muscles, and generally speaking it is the easiest thing in a youngish animal during life to demonstrate the presence of relatively large vessels running just inside the sheath of every moderate sized nerve in the body. A low power of the microscope shews them up very plainly.

These experiments throw into strong light not only the absolute dependence of the nerve on a blood-supply, but also the very complete nature of the anastomosis in mammalian nerve; further, that a warm blooded nerve may become temporarily inexcitable and after the lapse of some considerable time regain its excitability on the restoration of the circulation. The clearly suggest that mammalian nerve has a metabolism of a pronounced kind.

An attempt was then made be perfusion to replace the/

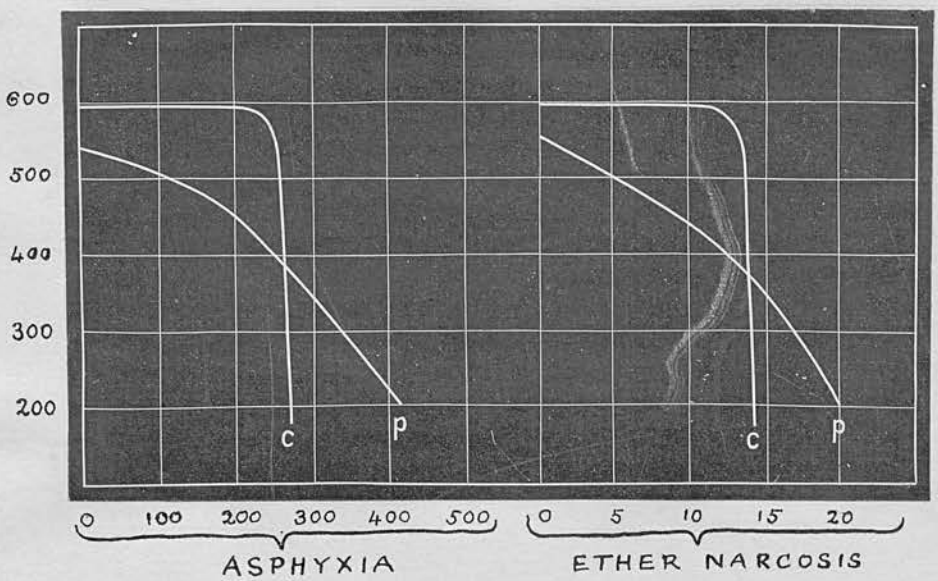
the blood in the combined tibial and peroneal nerves by means of Ringer's solution, with the ultimate object of determining if a supply of O_2 is as necessary for warm-blooded as it is for cold-blooded nerve. All effects however to perfuse the fine vessels of supply failed owing to the smallness of the calibre. Such experiments might succeed in a larger animal than the rabbit.

As was just stated mammalian nerve may undergo temporary suspension of function owing to deficiency of circulation. The nerve is not dead, it resumes its capacity to function normally when blood comes back to it. This might possible be correlated with the fact, previously mentioned (see p.16) that the electrical reactions may be elicited from mammalian nerve at a time when the muscle response cannot be got.

SECTION D. THE RELATIONSHIP BETWEEN EXCITABILITY AND CONDUCTIVITY FURTHER DISCUSSED.

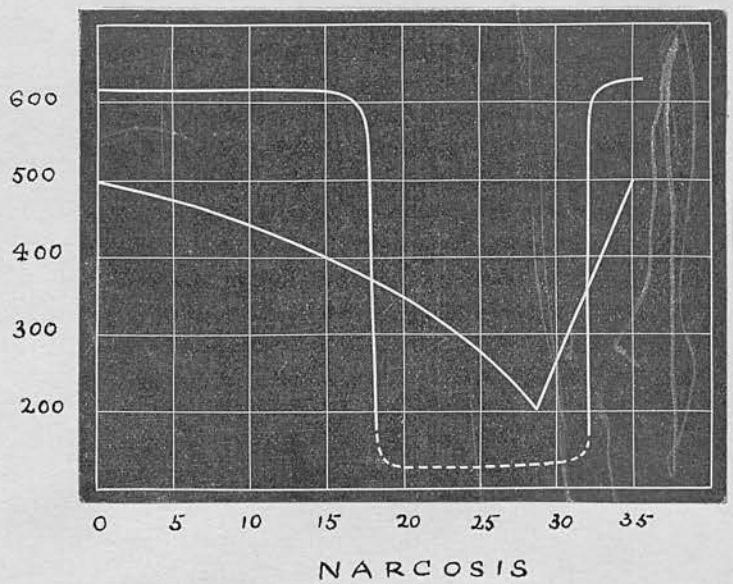
In the course of his investigations Fröhlich had soon convinced himself that the interference with conductivity and excitability caused by asphyxia corresponds exactly with the interference with these properties/

Fig. 7.



Abscissa shews Time in Minutes
Ordinate " Distance of Sec. from Prim. Coil in mm.

Fig. 8.



Line commencing at 610 shews Conductivity.
Line " " 500 " Excitability.

properties by narcosis. The only difference is a difference in the time required to produce the various stages, asphyxia being a much slower process than narcosis, in the relation roughly of 10:1. By using the more deliberate asphyxiation method he was enabled (30) to study the two "stages" of Szpilman and Luchsinger much more carefully than had before been possible, and could easily trace the passage of the one stage into the other. The method he used was the single twitch method with minimal induction shocks, the muscle being employed as the indicator.

On commencing the asphyxiation the excitability was first influenced, while the conductivity shewed no change. When the excitability had fallen to a definite point the conductivity began to fall very quickly, soon to disappear entirely. Meantime stimulation applied inside the chamber not far from the point of exit of the nerve towards the muscle, remained still effective. If the asphyxiation was further pushed the excitability sank more and more until the stimulation began to pass by "Stromschleifen" to portions of the nerve lying outside the chamber. Asphyxiation was never much pushed beyond this stage for the nerve was apt to die.

If O_2 was supplied to the asphyxiated nerve the first signs of recovery occurred after a period of

$\frac{1}{2}$ - 2 minutes/

½ - 2 minutes, and at the peripheral point of excitation situated inside the chamber. At this spot a rise in the excitability took place. Only when the excitability inside the chamber had risen to a definite point did stimuli coming from the central electrodes become effective. The re-installation of the conductivity went on then so quickly that the steps could scarcely be followed. In a few seconds the original conductivity was back again while the excitability inside the chamber continued gradually coming back to its primitive height. Exactly the same results were got with narcosis by means of ether.

By using chambers of varying diameter so as to asphyxiate different lengths of nerve he was able to shew that the conductivity more quickly disappeared when a long stretch of nerve was acted upon than when a short piece was asphyxiated. As stated before (p. 15) Werigo and Rajmist[12] had previously got the same result, and, even before Wedensky made his direct observations on a diminution in the power of conductivity in the early stages of anaesthesia, they had reasoned from the time required to abolish the conductivity when different lengths of nerve were exposed to the anaesthetic that the conductivity undergoes a gradual change from the first moment of anaesthesia.

Fröhlich, however made a further observation[30] He found on asphyxiating different lengths of nerve that there was not/

that there was not merely a difference in the time but also a difference in the degree to which the excitability must sink before conductivity is abolished. The longer the stretch of nerve affected by narcosis or asphyxia the less a drop in the excitability suffices to abolish the conductivity; the shorter on the other hand is the anaesthetised portion the more must the excitability be lowered before the conductivity disappears.

These results clearly prove that instead of being two distinct properties, as Grünhagen had imagined, the conductivity is a function of the excitability.

It is unfortunate that owing to the use of induction coils the graduations of which were arbitrary the exact relation that holds between length of nerve anaesthetised and degree of depreciation of excitability necessary to cause disappearance of conductivity, was not in this minimal twitch method determined. The presumption is that if the anaesthetised portion were homogeneously acted upon all along its length, the nature of the relationship would turn out to be very simple. A series of quantitative investigations with a standard Kronecker coil, care being taken to make as precise

delimitations of the anaesthetised portion as possible, would be interesting.

Dendrinos[31] working in Hering's laboratory with ether narcosis, had also emphasised the importance of the length of nerve anaesthetised on the result of narcosis. This observer, acting on the hint given by Wedensky, had not confined his observations to minimal twitches but had used single over-maximal stimulations as well. Direct observation by this method shewed that conductivity undergoes a decrease at a time, when, as tested by the minimal twitch method, it is apparently unchanged.

Noll[32] who worked in Biedermann's laboratory, repeated Dendrinos work with single over-maximal stimuli and confirmed these results. He also directed his attention to the question whether different narcotic agents have specific effects on nerve, and concluded from experiments ~~on~~ with ether, chloroform, acetone, ethyl-bromide, ammonia, acetic acid and carbon dioxide, that they all act in the same way exactly, the only difference being in the rapidity with which they produce their effect.

In this regard it must be pointed out that the idea, once entertained, that some substances exalt the conductivity of a nerve relatively to the excitability, and vice versa, is now being abandoned. The tendency of recent work has almost invariably been to establish the fact that so-called anaesthetics have/

have nothing peculiar in their action. A whole group of substances varying enormously in chemical constitution all have the same action on nerve. They are depressants and simply abolish the function of the nerve with greater or less rapidity. Fröhlich shewed that asphyxia acts in the same way. Any difference in the action of these things is not manifested by a difference in the behaviour of conductivity to excitability, but rather in the relative toxicity of the chemical substances, recovery from their action being taken as the test. To this point Waller first directed his attention. After him Wedensky emphasised the question. The only serious attempt in quite recent times to establish a specific action for any so-called nerve, anaesthetic was that of Herzen[33] and Radzikowski[34] who claimed that chloralose (or glyco-chloral) could entirely abolish the excitability without affecting the conductivity of a nerve. Their experiments were severely criticised by Wedensky.[10]

In this connection it is interesting to know if CO gas acts in the same was as N_2 or H_2. As we know, CO forms a firmer compound with haemoglobin that O_2 does. It is just possible that after CO asphyxia some compound may be formed in a nerve which might prevent recovery on the admission of O_2.

I/

I performed some experiments in Göttingen to settle this point. The CO gas, prepared by the action of concentrated H_2SO_4 on oxalic acid, and freed from CO_2 by washing with KOH, was allowed to run into the asphyxiating chamber. For some hours the excitability gradually decreased, the conductivity as tested by minimal stimuli, remaining constant. Finally the conductivity disappeared. On readmission of O_2 both conductivity and excitability soon came back again to the full extent, and the nerve shewed a complete recovery. CO acts on nerve like any other neutral gas.

To fix our ideas we shall sum up what we know at this juncture of the relation of conductivity to excitability. The two stages of anaesthesia (Szpilman and Luchsinger) got by the minimal twitch method is only a first approximation to the truth. Just as Kepler's laws, not absolutely true themselves formed the basis of Newton's generalisation, so the recognition of these two apparent stages was the starting point for more exact knowledge. There are in reality no two distinct stages of anaesthesia. Conductivity begins to be influenced from the start. By the minimal twitch method it is not possible to verify this by <u>direct</u> observation, one can only by reasoning/

reasoning on the results of anaesthetising different lengths of nerve reach this conclusion. By the use of the method of over-maximal stimulation one can directly shew that the conductivity begins to disappear at a time when it is still present for minimalstimuli. These facts are sufficient to prove that conductivity and excitability are not two distinct properties as Grünhagen held, but that conductivity is a function of excitability. When excitability is depressed conductivity tends likewise to be depressed.

Wedensky's method of using rapidly recurring stimuli with the "paradoxical conduction" that it disclosed, seems to indicate that the question is a much more complex one that had been supposed, but as we shall see even this paradoxical conduction is capable of analysis and of explanation.

SECTION E. FATIGUE OF NERVE.

Ever since Bernstein's[35] classical experiments on "the fatigue and recovery of nerve" the question of the fatigability of nerve has been a much discussed subject. The majority of the experiments that have been carried out on the subject have been done on motor medullated nerve. The method used has generally been to stimulate the nerve for a number/

number of hours, and meantime exclude fatigue of the end-apparatus by preventing the impulses from reaching the peripheral organs. On removal of the block by which this was attained the activity of the peripheral organ was seen to be present in undiminished strength. Various kinds of block have been used. Bernstein [35] and Wedensky [36] used the constant galvanic current, Bowditch [37] and Durig [38] used curara, Maschek [39] employed ether, and in the case of secretory fibres Lambert [40] made use of atropin.

Waller [41] took the electrical changes in medullated nerve as an index of the activity and photographed the galvanometric variation. The nerve was submitted to a short series of rapid excitations of similar intensity, which recurred at regular intervals of one minute, and the resulting effects shewed no evidence of diminution though fatigue. Some few observers have used non-medullated nerve. In experiments on the cervical sympathetic fibres Eve[42] found the vaso-constrictor apparatus of the vessels of the ear still active after 12 hours stimulation. Again Howell, Budgett and Leonard[43] state that vaso-constrictor and cardio-inhibitory fibres show no functional fatigue after an hour's stimulation.

In Waller's experiments on isolated nerve he used various anaesthetic agents, including CO_2 gas.

Large/

Large doses of CO_2 temporarily abolish the electrical response. Very small doses on the contrary increase its activity, and the swing of the galvanometer needle is increased when the nerve is thrown into action. Again, in a non-narcotised nerve mere excitation causes the electrical responses to improve at first, and in a manner exactly similar to the improvement from small doses of CO_2 gas; whence Waller concluded that activity is associated with the discharge of carbon dioxide.

Now to account for the fact that fatigue does not still occur in nerve Waller put forward the following tentative explanation. He says (44) "I wonder does this carbonic acid become altogether dissipated; may it not perhaps be reinvolved in some storage combination, as the nerve-fat, perhaps, that is so prominent a constituent of fully evolved nerve? Such nerve consists of proteid axis and fatty sheath; the axis - which is the off-shoot of a nerve cell - is the specially conductile part, the sheath is a developmental appendix, not directly connected with any nerve cell. Yet, cut the nerve, andsheath as well as axis undergo Wallerian degeneration, which is evident proof of a functional commerce between sheath and axis. All these things to my mind reconcile themselves with the notion that the active grey axis both lays down and uses up its own fatty sheath, and that it is inexhaustible not because there/

there is little or no expenditure but because there is an ample re-supply."

In order to test this hypothesis, Miss Sowton[45] undertook at Waller's suggestion an investigation on the non-medullated olfactory nerve of the pike, and found that the galvanometer responses of the nerve became feebler on repeated stimulation.

Brodie and Halliburton[46] not quite satisfied with the proof of fatigue in these experiments, sought to settle the question in another way. To avoid the use of the electrical response as the test of activity, they selected the non-medullated nerves that go to the spleen of a mammal. They blocked the impulses by means of low temperature (0° - 2°C), and found that even after 6 hours continuous stimulation the nerve conducted impulses just as well as at the start. They likewise made corresponding observations on the vaso-motor nerves contained in the sciatic nerve of the dog, the volume of the leg being recorded with a plethysmograph, and repeated Eve's experiments on the cervical sympathetic running to the ear of the rabbit. In no case were they able to demonstrate any functional fatigue.

These experiments of Brodie and Halliburton however, labour under one serious objection, and the same objection applies to Bowditch's to Eve's to Lambert's and to Howell, Budgett and Leonard's experiments./

experiments. They were done on warm-blooded animals in which, as was pointed out on page 29 the nerves while capable of functioning retain their blood supply more or less intact. So far as we yet know, it is quite possible that the nerves of warm-blooded animals may be easily made to shew fatigue.

But to return to Waller's hypothesis, apart from the function ascribed to the medullary sheath, the suggestion that fatigue is difficult to prove because the restitution processes are so perfect has much to recommend it. Thus Fröhlich's experiments brought into prominence the great rapidity with which O_2 is taken up by an asphyxiated nerve. While it took 3 - 4 hours to remove the conductivity of a nerve in an atmosphere of N_2 exposure to O_2 for $\frac{1}{2}$-1 minute was sufficient to restore conductivity again for a long time. The nerve took up O_2 at a much faster rate than this element could be withdrawn from it.

Garten[47] took up Miss Sowton's work on the non-medullated olfactory nerve of the pike, using the capillary electrometer instead of the galvanometer, and got the same results as she did. On rhythmical stimulation with induction shocks in a constant direction he found that the excursions of the electrometer soon declined in amount. After a rest they/

they returned in full amount again. In order to meet the objection that the result was merely an effect of injury at the point of stimulation, he stimulated at two places; the one point of stimulation he used for long continued tetanisation, the other (nearer to the capillary electrometer electrodes) he kept for short control stimulations. Thus, assuming that the electrical change gives a measure of the amount of the excitation, the fact seems to be proved that non-medullated nerve is capable of being fatigued.

In 1900 Carvallo[48] reported that he had been able to shew fatigue of frog's nerve by cooling. He kept the muscle at room temperature and cooled the nerve to 0°C. By stimulating with single shocks every five seconds he found that after a period of half an hour the excitability had disappeared; and proved to his own satisfaction that both low temperature and stimulation of the nerve were necessary to cause this fatigue. I repeated Carvallo's experiments while in Göttingen, and found that what he took to be fatigue was merely a lowering of the excitability of the nerve by low temperature. He had fixed upon a stimulus that was just maximal at 20°C, and had used it all along. Naturally, when the temperature of the nerve fell this stimulus became no longer effective. In his proof that both low temperature and stimulation were necessary to cause fatigue/

fatigue he had not accurately measured his temperature; and generally speaking his whole conclusions were inaccurate. For one thing the conductivity of the "fatigued" nerve remained unchanged all along!

As we have seen (pp 22,23), v. Baeyer observed phenomena in a nerve recovering from asphyxia, which he took to imply fatigue.

While Fröhlich was examining the curves published in Wedensky's paper[10] it occurred to him that the phenomena might be due to fatigue of the nerve. At a certain stage of the anaesthesia shortly before the disappearance of the conductivity, strong and rapid tetanisation of the nerve calls forth only a single twitch of the muscle, weak and less rapid tetanisation causes tetanus. Fröhlich had found that the same thing occurs in asphyxia just before the disappearance of the conductivity. He consequently set himself to examine the condition more closely.[49]

He kept the nerve narcotised with a dilute stream of ether, and examined it when it had reached the stage at which the conductivity is on the point of disappearing. By varying the rate of stimulation (from 150 to 3 or 4 per second), meantime keeping the intensity constant, he found that high frequency caused only an initial twitch, a slightly less frequency caused also an initial twitch which was slightly higher than the first. Stimulation with

still/

still lower frequency caused tetanus, which increased in height with further decrease in the frequency - Optimum of Frequency - and finally decreased again in height with continued decreasing frequency of stimulation. Eventually he got incomplete tetanus. A frequency of 3 - 4 per second produced twitches quite separate from each other, but of the same height at first as the initial twitches which were got with the high rates.

The same results were got by altering the intensity, when the frequency was kept constant. Tetanisation with strong intensity caused a single twitch, a slightly less intensity caused a single twitch just higher than the first. Weaker intensities caused tetani, which at first increased with decrease in intensity - Optimum of Intensity - and then fell off in height to finally disappear. Single shocks here, too, caused twitches of the same height as those got by tetanisation with strong shocks.

These results corresponded exactly with effects that Wedensky(50) already in 1886 had found in the nerve-muscle preparation of a narcotised frog and which Hoffmann(51) who repeated the experiments using had ascribed to an action on the end-plates of the nerve in the muscle.

That these effects in Fröhlich's experiments were not due to an altered condition of the end-plates or/

or of the muscle was shewn by control experiments with a pair of electrodes applied quite peripherally on the nerve, i.e. near the muscle. Full tetani were then always produced. Besides, the telephone and galvanometer controls of Wedensky's later experiments,(10) and the capillary electrometer results of Boruttau (see pp. 12-14) entirely negative any such objection.

The fact that in a certain stage of anaesthesia strong tetanising stimuli call forth only one twitch which in height corresponds to the effect of one single stimulus, indicates that in this condition of the nerve strong and rapid stimuli are not summated, which must be due to a prolongation of the refractory period of the nerve. Experiments with two stimuli separated by a very short interval of time, shewed that in reality the refractory period of the nerve was greatly prolonged by anaesthesia. This had already been pointed out by Boruttau.(52)

Nay more, there was a definite relation between the refractory period as determined by two successive twitches, and the frequency of stimulation that in any given stage of the anaesthesia just caused an initial twitch. Supposing he found that with a definite intensity of stimulation tetanising shocks of 100 per second just caused an initial twitch, while similar/

similar shocks at the rate of 80 per second caused muscle tetanus, then two stimuli of the same intensity as had been used for tetanisation following each other at an interval of 1/100th. of a second caused no summation, while two similar stimuli, but separated by an interval of 1/80th. of a second caused summation.

This prolongation of the refractory period by means of anaesthesia may sometimes be considerable. Fröhlich found a refractory period in one case of as much as 1/10th. of a second.

The refractory period is a function not merely of the degree of the anaesthesia but also of the strength of the shock used to stimulate the nerve. Strong shocks shew a longer refractory period than weak shocks.

All this suggests that the reason why no one has succeeded in fatiguing nerve is that the refractory period under normal conditions is so short that in the intervals between the individual stimuli the nerve has sufficient time for complete recovery, and fatigue cannot set in. In the condition of narcosis and of asphyxia on the other hand, the refractory period is so much prolonged that on stimulation with repeated shocks that follow each other in an interval of time less than the refractory period, only the first stimulus is effective, while the second stimulus occurs in the refractory period

Fig 9.

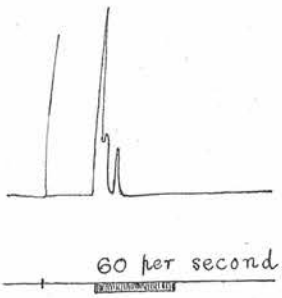
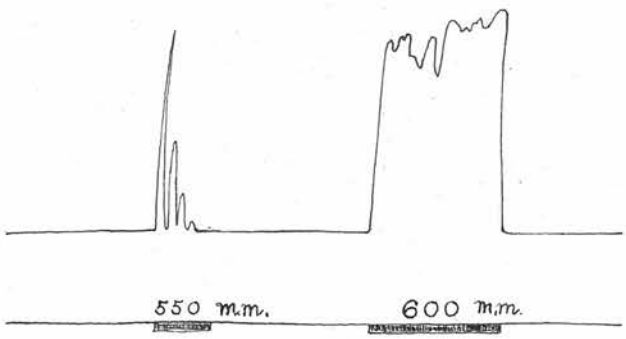


Fig. 10.



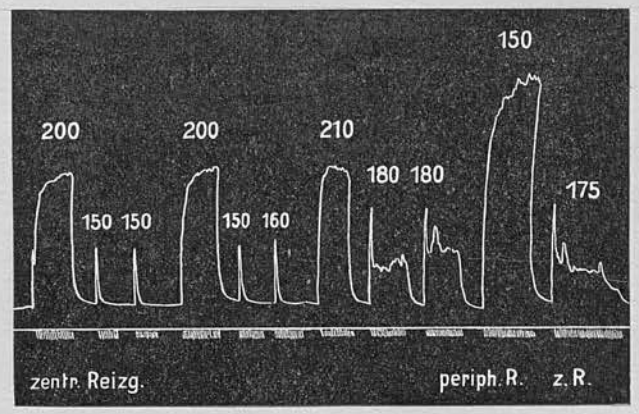
Curves reproduced from Fröhlich's paper.

of the first and is therefore ineffective.

Now, if instead of considering simply the single twitches that are got with rapid tetanisation and the complete tetani that follow much slower stimulation, we direct our attention to the muscle contraction that follows tetanic stimulation of the nerve at an intermediate rate, we find that it consists of an initial twitch followed by a series of twitches of decreasing height and finally by complete rest of the muscle. In figure 9 we see two responses: the first is a single twitch recorded with stationary drum, the second is the result of tetanising at 60 per second at the same intensity Had the tetanising shocks been more rapid we should have got a single twitch, had they been slower we should have got full tetanus. As it is we see that the twitches fall away and are succeeded by complete rest. This is a curve shewing fatigue of the nerve.

This proof of fatigue is ingenious and beautiful. It just misses being absolutely conclusive, because Fröhlich did not prove that the refractory period is prolonged by previous activity of the nerve. A possible objection might be that his anaesthesia was becoming progressively deeper, and that consequently the refractory period was with each/

Fig. 11.


150
200
200
210
180 180
175
150 150
150 160
zentr. Reizg.
periph. R.
z. R.

each small interval of time becoming longer. Until it is proved that the refractory period is actually prolonged by activity one cannot take this as unconditional fatigue. Nevertheless it is by far the best proof of fatigue of nerve that has ever yet been given.

As in my attempts to asphyxiate the nerves of warm-blooded animals I had worked out the operative technique for warm-blooded nerve it occurred to Frŏhlich that by an adaption of the method we might be able to narcotise warm-blooded nerve sufficiently to shew Wedensky"s "paradoxical conduction." This we found (53) quite possible in the peroneal nerve of youngish rabbits by means of ether and chloroform, and we obtained typical "fatigue" curves. (See Fig 11) For some reason we were unable to sufficiently narcotise the corresponding nerves of full-grown animals; whether it was that the connective tissue was more dense or the blood supply better, or for what other reason it is difficult to say.

SECTION F. ARE CONDUCTIVITY AND EXCITABILITY REALLY SEPARATE?

Before concluding this part of our historic sketch it will be of advantage to once more return to/

to these two properties of nerve, conductivity and excitability, so as to finally adjust our ideas on the subject.

Boruttau had found that the rate of transmission of the electrical change in nerve is delayed by narcosis. Fröhlich investigated this discovery by means of the muscle, and established the fact (54) that both asphyxia and narcosis produce a distinct delay in the transmission of the excitation. This delay is greater the greater the length of nerve anaesthetised or asphyxiated, as the case may be, and is most marked just at the stage when the conductivity (as tested by minimal induction shocks) is about to disappear. The slowing of the impulse is confined to the affected portion; on passing through it the impulse once more resumes its normal speed.

As mentioned in last section (p. 46.), Fröhlich determined that the refractory period of the nerve is prolonged by narcosis and asphyxia, a fact to which Boruttau had already drawn attention.(52)

Hand in hand with the prolongation of the refractory period there goes an increase in the actual duration of the electrical wave.(52) This takes the form of a great prolongation of the descending portion of the curve of negative variation, as shewn by the capillary electrometer. The prolongation of the wave is obviously most intimately bound/

bound up with the increased duration of the refractory period. It is limited to the narcotised portion of the nerve, while peripheral to this the normal wave duration reasserts itself.

Thus we have three definite changes occurring in a narcotised stretch of nerve, two at least limited to the affected portion:-

(1) delay in rate of transmission,

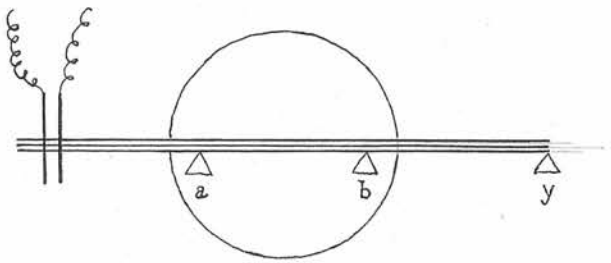
(2) prolongation of refractory period,

(3) prolongation of electrical wave.

There is still a fourth change, this time not localised to the narcotised part. This takes the form of a diminution in the extent or amplitude of the excitation as measured by electrical means. Boruttau first established this change in nerve which had been locally cooled.(55) A similar decrement in the excitatory wave occurs in a narcotised or asphyxiated portion.(56) On leaving the affected stretch of nerve the wave does not return to its original height, it remains permanently diminished, notwithstanding that its original short duration and its original rate are recovered.

This decrement is a function of the intensity of the stimulation. We see this from the experiments of Wedensky and of Noll. Boruttau and Fröhlich(56) set themselves to determine the exact relation between the/

Fig. 12.



the two. They used two electrodes for registering electrical change on the longitudinal part of the nerve, the one, a, nearer to, the other, b, more distant from, the point of stimulation. (See Fig. 12). By means of a third electrode, y, placed in contact with the cross section of the nerve, they could lead off alternately from a and y, or b and y as the case might be.

In any given stage of narcosis then, although the demarcation current was exactly the same in each case, the negative variation with one and the same intensity of stimulation was less in b. than in a. When they varied the intensity, the decrease in the negative variation from a. to b. with strong stimulation was much more considerable than with weak stimulation. The further the narcosis had progressed the more did this hold true. Thus, by tetanising at stated intervals, first with secondary coil set at a distance of 450 m.m. and then at a distance of 200 m.m., the negative variation in the first case decreased in the space of 20 minutes from 4 to 1 scale-divisions, in the second case from 75 to 3.

The decrement in the strong stimulation is great, that in the weak is proportionally slight, and in fact "as one can easily convince oneself, the size of the decrement is directly proportional to the size of the/

the wave of excitation."

This explains how it is that the conductivity disappears at the same moment for both weak and strong stimuli, for the negative variation in each case becomes vanishingly small at the same point. It likewise explains how, at the commencement of narcosis or asphyxia, the conductivity appears unchanged for both strong and weak stimulations. For weak stimulations the decrement is extremely small, and the conductivity disappears only when the negative variation has sunk below the limit necessary to induce a minimal twitch of the muscle. For strong stimulations the decrement is greater, but the maximal twitch only decreases when the negative variation sinks under the limit necessary to call forth a maximal twitch. Once this occurs, the height of the muscle twitch decreases with decreasing extent of the negative variation, to disappear simultaneously with the effectiveness of weak stimulation.

The conductivity for weak and strong stimulations during narcosis is therefore only apparently unchanged, because the various alterations that the excitation undergoes, prolongation of refractory period, increased duration of the electrical wave, and delay in rate of transmission, do not have an opportunity of evincing themselves in the commonly used method of observation of the muscle twitches following single stimuli.

The/

The only other question that it remains to answer is this. Why does the apparently unchanged conductivity disappear before the excitability?

As we saw on page 32 the time at which the conductivity disappears depends on the length of nerve narcotised. The shorter the affected portion of nerve, the longer the time and the greater the drop in excitability required to abolish the conductivity. Pursue this to the limit, i.e. narcotise a vanishingly small portion of nerve, and the excitability and conductivity will disappear together. The "excitability" merely expresses the change at one given point owing to progressive alteration in the nerve through narcosis, the "conductivity" is an expression for the change in a whole stretch of nerve, for the wave of excitation undergoes steady diminution all along its course from one portion of the narcotised nerve to another. In other words there is no separation possible between excitability and conductivity. They are merely different expressions for the same thing.

SECTION G. THE QUESTION OF COOLING.

It had long been held that the excitability of nerve is lowered by cooling, and raised by warming. Efron[6] and Hirschberg,[7] who conducted experiments/

experiments to determine the relation between conductivity and excitability, found that the excitability apparently fell with fall of temperature. Howell, Budgett and Leonard[43] investigated the influence of temperature on nerve in a special research, and came to the same conclusion. Not till Gotch and Macdonald[14] carried out their experiments, however, was the question settled beyond cavil; they found, taking special precautions against manifold sources of error, that the excitability of nerve to break induction shocks is definitely lowered with decrease in the temperature.

The rate of transmission of the nervous impulse is likewise lowered by cooling of the nerve. This was determined by Helmholtz[57] for frog's nerve, by Helmholtz and Baxt[58] and by Oehl[59] for the sensory nerves of man, and by Boruttau[55] for the motor nerves of numerous warm-blooded animals.

Again, Borrutau had found[52] by electrical observation that the amplitude of the wave of excitation undergoes a diminution with cold. The fact that the muscular response is altered in extent by cooling of the nerve is thus referred to by Gotch "On cooling a portion of a nerve, not only is the rate lessened, but the excitatory state diminishes in amount as it traverses the cooled region, and on emerging from this it continues with the same diminished/

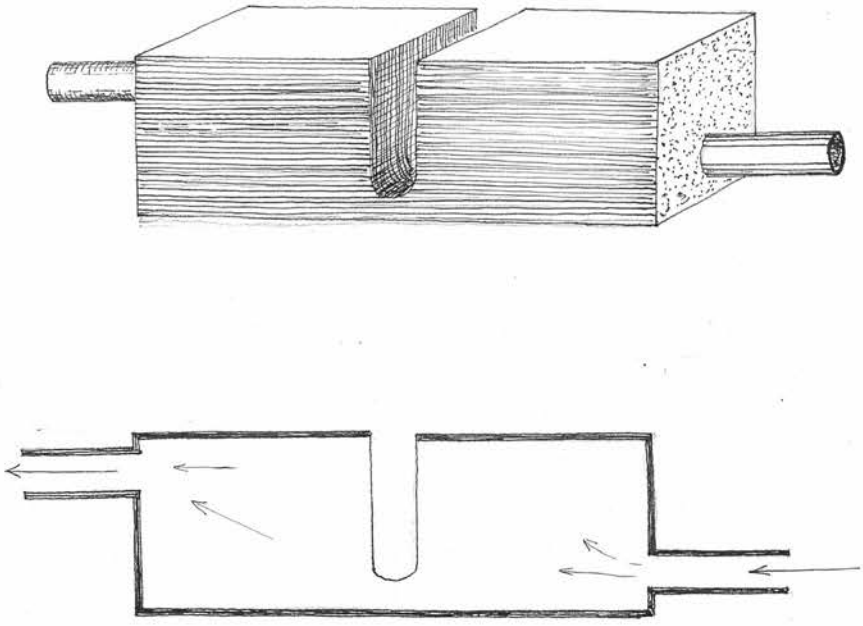
diminished intensity, so that it evokes a smaller muscular response than it would otherwise do."

Boycott,[60] working under Gotch's direction, found that the refractory period of nerve is prolonged by cooling. Boycott used the muscle response; Gotch and Burch[18] determined the same thing on isolated nerve by means of the capillary electrometer.

The descending part of the curve of negative variation is also prolonged by cold. This was first made out by Hermann[61] and confirmed by Verwej [62] and by Boruttau[52]

Putting these facts together, one sees that cold has apparently much the same influence upon nerve as narcosis or asphyxia ~~does~~. Seeing that the muscular responses associated with Wedensky's "paradoxical conduction of nerve" were got with both the latter influencing factors, it was interesting to see if they could be got by cooling. At the instigation of Professor Verworn I undertook this investigation.

Fig. 13.



PART II. COOLING OF NERVE.

SECTION A. APPARATUS.

(1) Cooling Apparatus.

In conducting local cooling experiments on the nerve of a nerve-muscle preparation, many precautions are necessary if one is to get reliable results. Care must be taken that neither the muscle nor the central end of the nerve is cooled. The cooled portion must be as nearly as possible at an equal temperature throughout its length. In working with very low temperatures, too, the nerve is apt to freeze if it touches any part of the cooling apparatus, hence it is necessary to cool the nerve by radiation rather than by actual conduction. Again, for experiments such as I had to carry out, it was found necessary not only to be able to alter the temperature at will, but to keep any one temperature constant for a more or less prolonged period.

The form of apparatus on which I fixed, a form suggested to me by Professor Boruttau, was found well adapted to fulfil these requirements. It consisted of a small metal box (see figure 13.) made out of $\frac{3}{4}$ inch square-sectioned brass tubing. A groove, $\frac{1}{4}$ inch wide by $\frac{1}{2}$ inch deep, was let into the upper side; this was lined with very thin copper foil and served to contain the nerve, which was stretched

along/

Fig. 14.

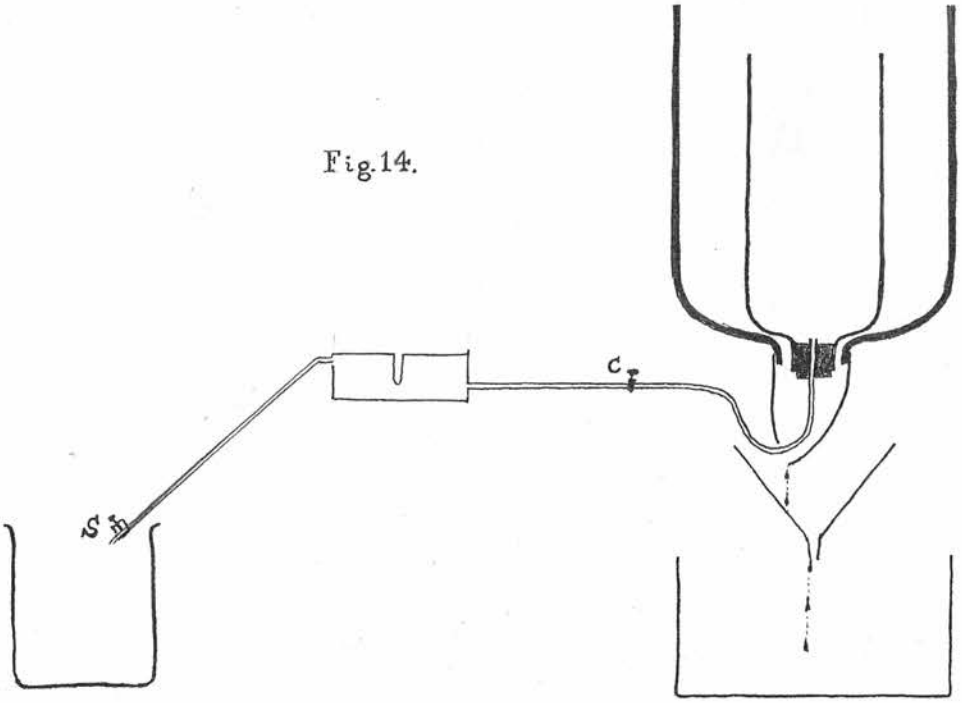
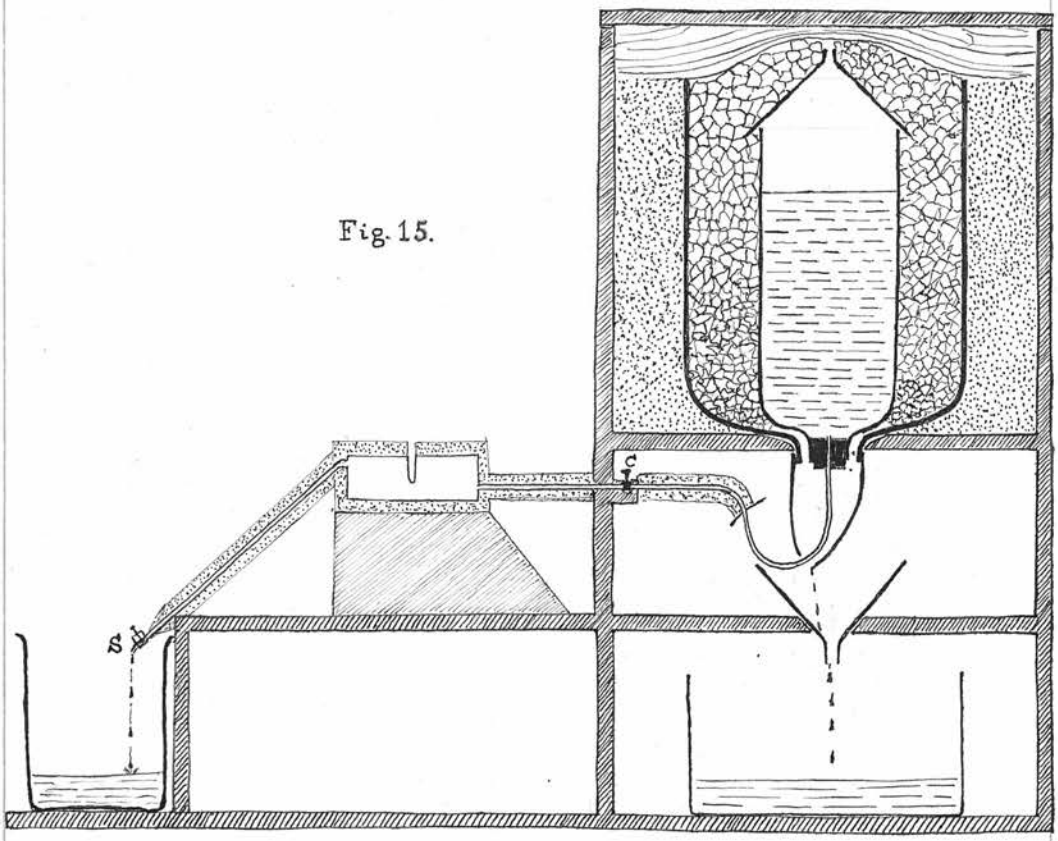


Fig. 15.



along it, near to, but not actually touching the bottom. On each end of the box was a brass pipe which served to conduct cold fluid through it. As the fluid passed along and cooled the walls of the box, the radiation from the thin copper foil cooled the nerve.

The cold fluid (a mixture of alcohol and water to prevent freezing) was got from a special ice and salt cooling apparatus consisting of two glass vessels, one inside the other (see figure 14). The inner vessel (thin walled), containing about a litre and a half, was connected by a pipe running from the bottom directly to the brass box. Between the inner vessel and the outer one a mixture of ice and salt was put. The drippings from this escaped into a funnel placed underneath, and thence into a large glass container.

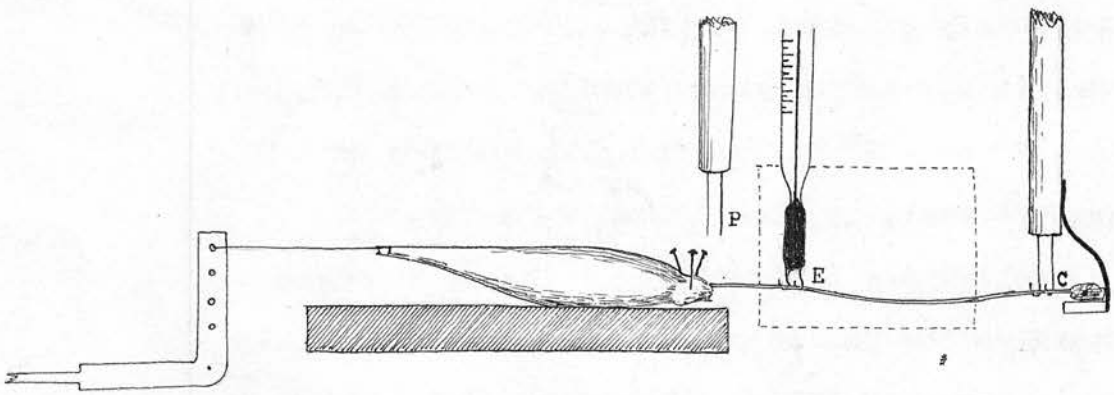
The diagram will explain better than any description how the apparatus was arranged. The upper figure shows the essential parts. The lower shews how insulation was affected, for all the parts designed to hold or convoy cold fluid were thoroughly insulated with felt, while the brass box had an outside jacket of cork, to prevent radiation to the muscle. One thing is not shewn, viz., an additional vessel for holding fluid at room temperature. This was connected by means of a three-way cock, C. to the pipe leading to the brass box. Thus one could run either cold or warm fluid through the box. S. is/

is a screw clamp such as is used on rubber gaspipe for fine graduation of the flow. It was attached to a piece of rubber on the extreme end of the outlet pipe, and served to accurately regulate the flow.

By means of this apparatus one could with a little practice keep the temperature of the brass box absolutely constant for from 20 minutes to half an hour, at any temperature varying from, say, +3° to - 7° or - 8° C. As far as constancy of temperature went, it functioned almost perfectly.

A preliminary series of experiments was done to determine the degree of the heat insulation near the brass box, and it was found that just outside the groove at a distance of ¼" from the box, i.e. at the point where the muscle was fixed with pins, or where, in the case of a short nerve, the central electrodes were applied, the temperature was never more than 6°C below room temperature (12°), when the temperature in the groove was - 5°C. The average temperature of the muscle, as very careful experiments shewed, was certainly never more than 4° below room temperature. When it was necessary that the muscle should be kept absolutely at room temperature, it was fixed in a small cork bath containing Ringer's solution, which was frequently renewed from a bottle kept standing in the room. As regards heat insulation the apparatus was probably as efficient as many similar pieces/

Fig. 16.



To shew arrangement of electrodes in relation to nerve.

pieces of apparatus that are assumed to be perfect.

(2) Nerve-Muscle Preparation.

The muscle was fixed on a small horizontal cork plate, the nerve and it lying in one straight line. Three pins, one through the knee-joint, and one each through the connective-tissue attached to the femur and tibia, were used to fix the proximal end of the muscle. This insured that the nerve should not be pulled upon. The muscle lever gave a ten=fold magnification.

Two pairs of platinum electrodes, bent at the end in the form of a hook, served to support the nerve. The one pair (E. electrodes) ran alongside the bulb of a small vertical thermometer specially made for these experiments; by means of these the nerve was hung just underneath, and almost in contact with, the thermometer bulb. The other pair, fixed in a vertical vulcanite rod, supported the central end, so that the nerve hung like a telegraph wire between these two points of support. As the nerve was removed from the animal with a small piece of the spinal column attached, a support for this end was attached to the vulcanite carrying the C. electrodes. Provision was made for applying a third pair of electrodes (P) to the nerve just as it entered the muscle. These were not always in use, however.

The electrodes, with thermometer etc., were all fixed relatively to the cork plate, and the whole system could be raised or lowered in one piece, so that once the muscle was fixed on the plate and the nerve laid in position, no alteration in the point of stimulation could occur. This relative immobility of the system was found of the greatest importance in freezing experiments, where the slightest mechanical injury to the nerve irretrievably destroys it. The supporting apparatus was raised or lowered by means of a screw arrangement such as is used in the rough adjustment of a microscope. The position of the nerve in the bottom of the groove could therefore be accurately adjusted.

Special screws and sliding arrangements ~~was~~ were also provided for altering at will the position of any member of the system relatively to any other, so as to make the apparatus adaptable to brass chambers of different thickness and to nerves of different length.

The thermometer carrying the E. electrodes served to measure approximately the temperature of the nerve. It was always arranged at the peripheral edge of the cooled piece of nerve (conf. p. 15). A series of preliminary experiments was carried out to determine how the indicated temperature varied with the position of the thermometer bulb in the groove. These/

These shewed that if due care was taken to prevent the bulb from actually touching the sides of the groove, the variation, within the limits of position in the actual experiment, was never 1°C. If the bulb touched the side the reading was about 2° lower than it otherwise was.

The greatest difficulty was to know to what extent the indicated temperature corresponded to the actual temperature of the nerve, and this difficulty with this apparatus seems insurmountable. Experiments done by taking simultaneously the temperature of the fluid, first in the containing glass vessel, and then as it issued from the extreme outlet pipe, proved useless to decide the question. The indicated temperature of the nerve can only be taken as a rough approximation to the real temperature. The error, however, is probably a more or less constant one.

In cooling experiments, fortunately, no provision requires to be ~~taken~~ made to keep the cooled portion of the nerve moistened with saline solution. Moisture from the air condenses on the nerve, so that there is no fear of it drying. The projecting central part of the nerve was kept moist by means of a deep gutter lined with wet cotton wool, which was brought up under the nerve without touching it at any part, and served to keep the atmosphere in the immediate vicinity saturated/

saturated with water vapour. The peripheral part of the nerve, as well as the muscle were carefully brushed from time to time with Ringer's solution.

(3) The Electrical Apparatus.

To stimulate the nerve a standard Kronecker coil was used. The current in the primary circuit was obtained from a large accumulator which was kept at the same potential by regular charging. The difference in potential was $2\frac{1}{4}$ volts. For rapid tetanisation a Bernstein's acoustic interruptor was interposed in the primary circuit, for slower rates the springs supplied with the Kronecker coil were used. In the one case the mercury contact surface was kept covered with alcohol, in the other it was continuously washed with a stream of tap water.

Considerable difficulty was found in determining whether both "break" and "make" shocks were to be considered effective in stimulating the nerve. The Helmholtz side-wire does not adapt itself to rapid contact interruptions. With ~~this~~ the form of interruptor used it was hard to say whether one alone or both stimuli are effective at any given stage of an experiment. Experiments with single make and break shocks do not decide the question, for with increasing rapidity of stimulation the distance of secondary from primary coil requires to diminish in order to produce effective tetanisation of the muscle; so

that/

that the effects of rapid and of more leisurely interruption are not comparable. The figures that are here given represent the number of complete vibrations per second of the interrupting spring.

Until recently I was acquainted with the principle of placing a suitable condenser in the primary circuit of the induction coil, so as to increase the disproportion in intensity between "make" and "break" shocks. It is unfortunate that such a condenser was not used in my experiments.

The electrodes were so arranged that on the break of the primary coil a current in the descending direction went through the nerve. The electrode wires were kept as nearly as possible all parallel with each other, and were not twisted. This was found to be a vital precaution. By a combination of two Pohl commutators without the cross-wires, it was possible to send a stimulus through any one of the three pairs of electrodes at will. There was a simple key in the primary circuit, and a short circuiting key in the secondary circuit.

The above description applies to the apparatus as used by me in Edinburgh. The apparatus I had in Göttingen for example did not include a Kronecker coil; and many minor points tending to greater accuracy were not taken into consideration in Germany.

Fig 17.

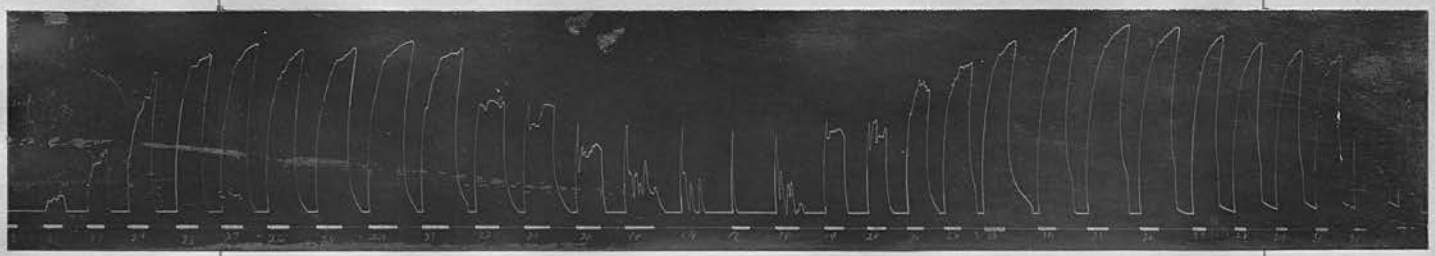
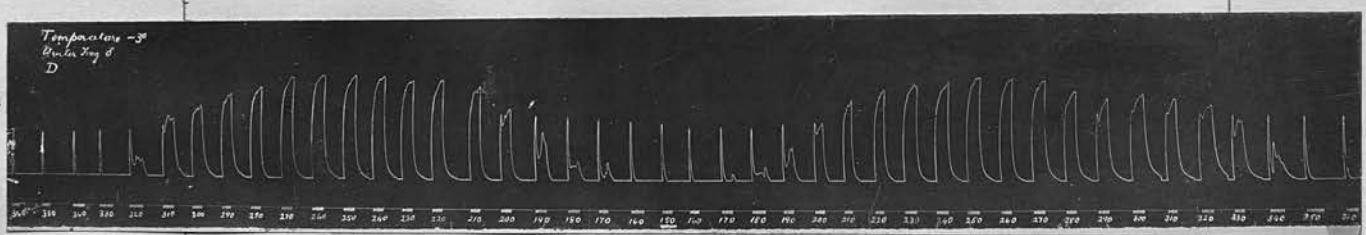


Fig 18.



SECTION B. WEDENSKY'S EFFECT BY COOLING.

On now cooling a portion of the middle of the nerve by lowering it into the cold chamber, it was found that, when the temperature sank below 0°C, tetanisation of the central end at a rate of about 100 per second gave the phenomena which Wedensky had described under the title of "paradoxical conduction of nerve," and which Fröhlich had argued was due to fatigue. When the temperature had become constant, and stood at, say - 2°C, the secondary coil was placed at just such a distance from the primary that no contraction of the muscle followed on tetanisation On moving it 1 cm. nearer to the primary a weak tetanus followed stimulation, on moving it another 1 cm. nearer the tetanus was stronger; and thus it went on until the tetani attained a maximum. They then began to fall off in height as the secondary coil was brought still nearer, curves like Fröhlich's "Fatigue" curves (see illustration facing p. 46) began to appear, and finally with strong tetanisation single twitches were got. (See Fig. 17.)

That this condition was not due to fatigue of the muscle was shewn by the simple device of reversing the direction in which the secondary coil was moved. Instead of bringing it nearer to, it was now moved farther away from the primary coil, and the same muscular effects were repeated in the reverse ~~direction~~ order.

The/

The tetani began gradually to climb up as the intensity of the stimulation decreased, until a maximum was reached; they then fell away in height, and ultimately with weak tetanisation disappeared. (See Fig.18). At the same time as this phenomenon was present peripheral stimulation of the nerve with a third pair of electrodes quite close to the muscle shewed full tetani with all moderate and strong intensities.

The correspondence with the condition described by Wedensky as resulting from anaesthesia, and the similar condition described by Fröhlich as resulting from asphyxia, was seen to be more complete when one varied the rate of stimulation keeping the temperature still constant. On stimulating at a slow rate full tetanus occurred even with the strongest intensity; stimulation with a very high rate produced the "paradoxical condition" even more distinctly, in that single twitches were got with tetanisation at even moderate intensities. These single twitches, or as Hoffmann has named them[51] "initial tetani" (Anfangstetanus), corresponded in height to single maximal twitches of the muscle evoked by one break shock. Finally if the temperature were ~~mechanically~~ more or less quickly raised, say, to - 1°C, the condition passed away for the highest rate of stimulation at my disposal. If the temperature were still further lowered the "paradoxical condition" was more easily elicitable.

From/

From all this one concluded that:-

(1). So far as the response to induced currents is concerned, cold has apparently the same effect on nerve as asphyxia or narcosis. A low temperature corresponds to a deep degree of narcosis or asphyxia as the case may be; a higher temperature corresponds to a lighter asphyxia or narcosis.

(2). For examining the condition that Wedensky called "paradoxical" conduction," cooling of the nerve is a more satisfactory method than either narcosis or asphyxia, in that the curves obtained are more regular. This is probably due to the fact that the physical condition of the nerve during an experiment can more readily be kept constant.

It was now possible to investigate in detail the influence of the various factors that contribute to the production of the Wedensky effect. The relation of the effect to disappearance of the conductivity could likewise be more precisely determined. I at first hoped to be able to establish certain quantitative relations among the factors involved, but it was found that the influence of temperature is more complicated than I had at first imagined. It will however conduce to clearness if we first of all take up in detail the various features of the Wedensky effect and deal afterwards with complicating circumstances.

(1)/

(1). The Height of Muscle Contraction as Influenced by the Temperature of the Nerve.

As we saw (p. 11), the first observation of Wedensky on the reaction of the muscle to anaesthesia of the nerve, was that the tetanic contractions were not so full and complete as before anaesthesia. In his experiments he had to exclude the possibility of this being a mere fatigue effect in the muscle itself. In my experiments there was the additional possibility that the temperature of the muscle might vary along with the variation in the temperature of the nerve. To avoid this contingency the muscle was fixed in a small bath of Ringer's solution at room temperature, and the solution frequently replenished with fresh fluid at a constant temperature. A few observations with a thermometer placed in the bath sufficed to shew that with these precautions the muscle did not vary in temperature more than $1^{\circ}C$. Such a variation is negligible, and besides, the alterations in the temperature of the muscle did not correspond with those in the temperature of the nerve.

On now cooling a portion of the nerve it was found that the muscle responses to central tetanisation of the nerve were less pronounced than before, in that the tetanus curves were lower. At first this change was not so marked, but when the temperature of the cooled piece of nerve fell to a definite point/

Fig. 19.

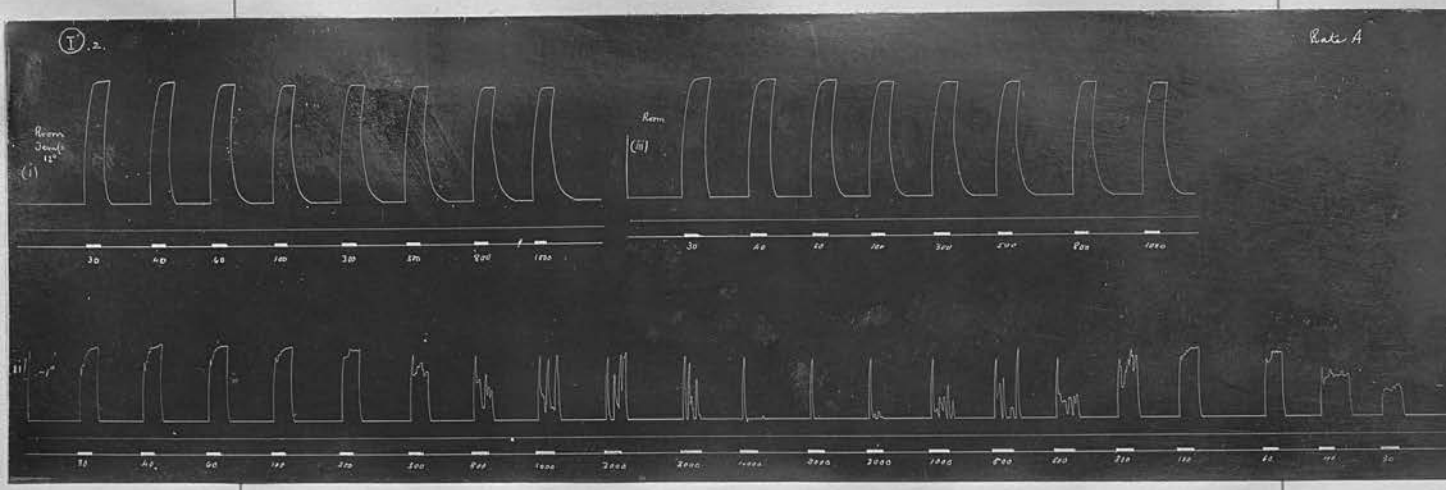
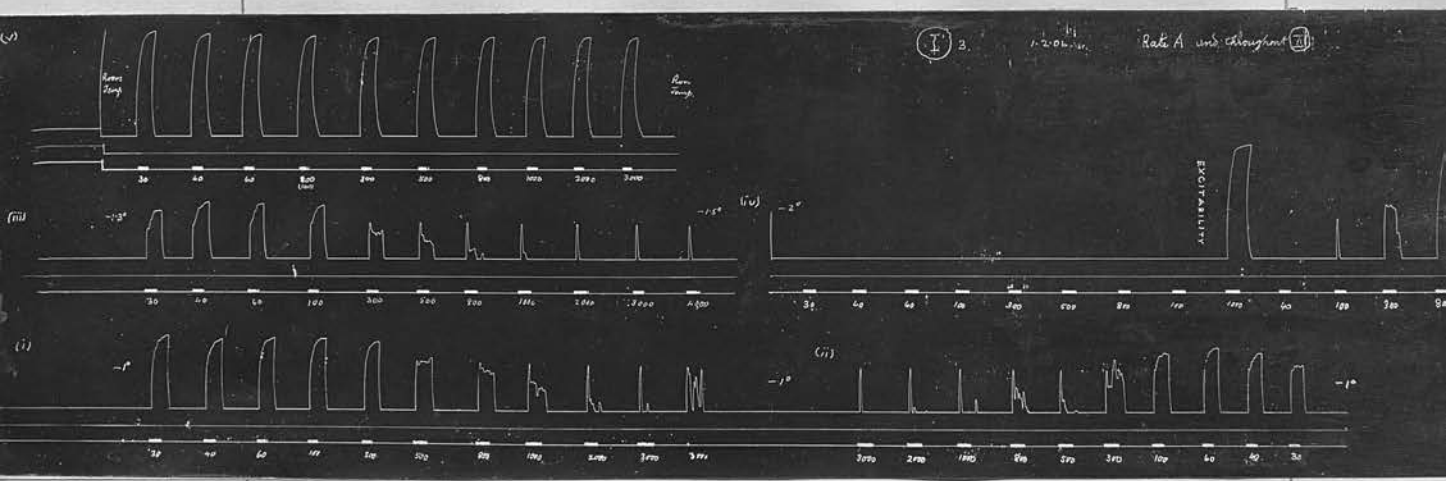

I. 2.
Rate A
Room Temp 12°
(i)
Rem.
(iii)
(ii)

Fig. 20.


I. 3.
Rate A and throughout
Room Temp
(v)
(iii)
(iv)
EXCITABILITY
(i)
(ii)

point, which varied for different preparations, each succeeding change of 1^{o} in the temperature of the nerve was denoted by a distinct falling off in the maximum height of the muscle tetanus; and, generally speaking, the lower the temperature of the nerve the more marked was the drop in the height of the muscle contraction corresponding to a drop of 1^{o}C. in the temperature of the nerve.

Figures 19 and 20, representing curves from the same preparation, shew this change, in its final stage at least, very plainly. In Fig. 19. we see, (1) the general height of the tetani got with varying intensities of stimulation at room temperature; (2) the drop in the height of the tetani when the nerve was cooled to -1^{o}; (3) the return to the ~~primitive~~ original height when the nerve was quickly raised out of the cold chamber again, thus proving that the effect is not one of fatigue of the muscle.

Fig. 20 (tracings 1 and 3) shews the relatively large drop in height of tetanus corresponding to a drop of half a degree in the temperature of the nerve, when the latter is already much cooled. A drop of another half degree suffices to abolish the muscle response entirely (tracing (IV) while the height returns more or less to normal on simply raising the nerve out of the chamber and waiting for a minute or two till it shall have attained approximately the temperature of the room. (tracing V.)

(2) Incomplete Tetani with Strong Stimulation.

After Wedensky had noticed the diminution in height of the tetani his next observation, if we remember, was that when the anaesthesia had advanced to a certain stage the muscle contractions got with strong tetanisation were less complete than those got with weaker, ~~stimulation~~ and more resembled these obtained with submaximal tetanisation. A reference to Fig.17 will shew the justice of this remark. In the first series of tracings shewn there, the gradual rise of the tetani to a maximum and their subsequent falling off in height as the strength of stimulation is increased, is well shewn. A further fact, not mentioned by Wedensky, is also indicated, viz, that the tendency of the tetani to climb is more or less abolished, and a change has rather set it in the opposite direction. The tendency of the curves is now to decline rather than to rise in height, and this decline may be pretty regular, as is indicated in the first curve marked 19 in the Chart.

From such curves alone it might with considerable shew of reason be argued, as it was argued by Fröhlich, that the nerve is being fatigued, for the outside conditions are practically unchanged. The muscle is presumably as able to respond to stimulation as ever; the temperature of the nerve keeps constant: the/

the stimuli that come from the electrical apparatus do not vary. Yet the successive excitations in the nerve in traversing the cooled region undergo a change in such a way as to be less and less effective. Assuming then that the internal conducting mechanism of the non-stimulated nerve at this low temperature is stable, the falling off in effectiveness must be the result of the continued activity of the nerve.

It must be granted, however, that all the incomplete tetani got with strong stimulation do not shew this regular decline, and it is interesting in this regard to refer to some of Wedensky's own curves, which do not so plainly conform to the declining type. Nevertheless the very irregularity of the tetani got in this manner is in itself a striking phenomenon, and, as we shall see, it occurs again and again when nerve is cooled.

(3) The "Initial Tetanus" (Hoffmann).

These incomplete tetani from strong stimulation gave way, as Wedensky found, on further anaesthesia to single twitches. It was reserved for Fröhlich to point out - and I have confirmed the fact again and again - that the height of these single twitches corresponds to the height of a single maximal twitch of the muscle; as if only the first member of a series of excitations had succeeded in emerging from the/

Fig. 21.

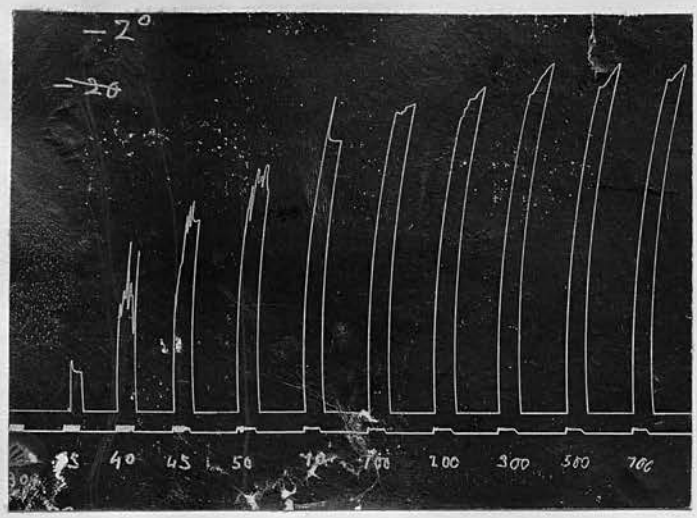


Fig. 22.

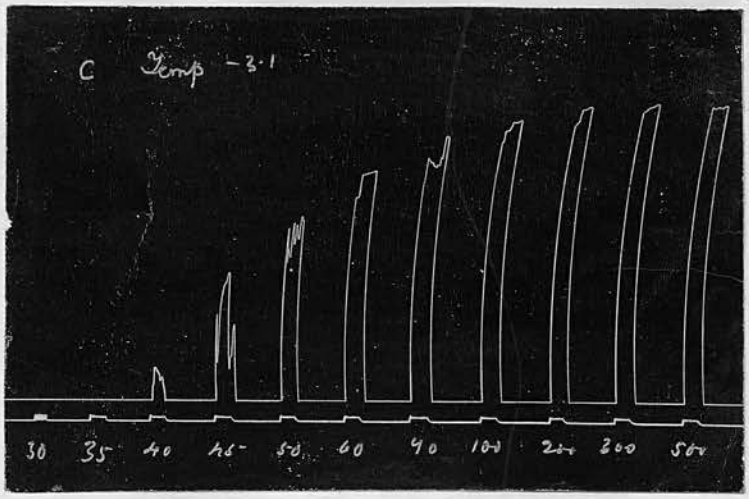
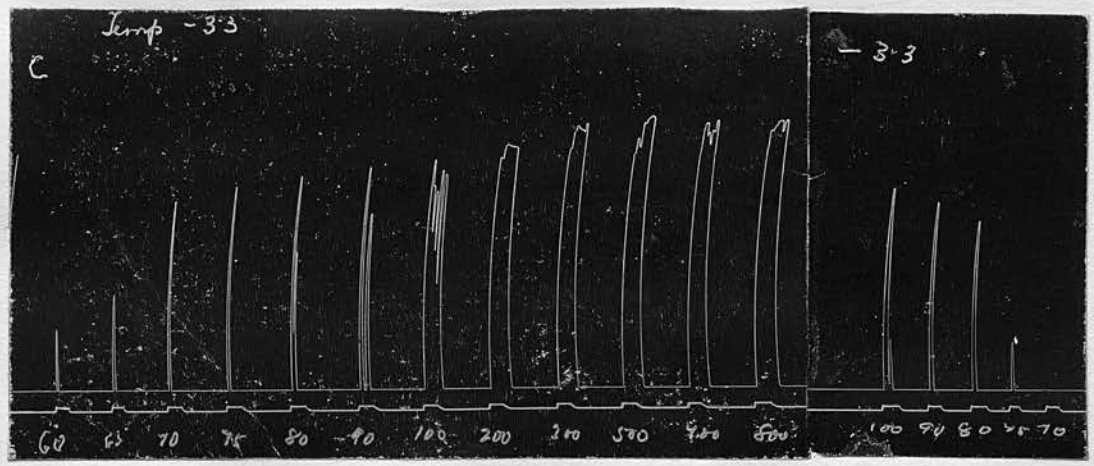


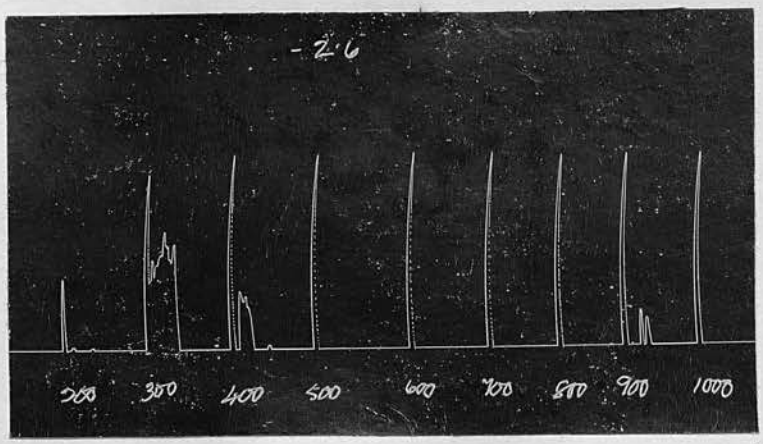
Fig. 23.



the affected part of the nerve with a strength sufficient to be maximal, while the succeeding ones all drop below minimal strength or do not get through at all.

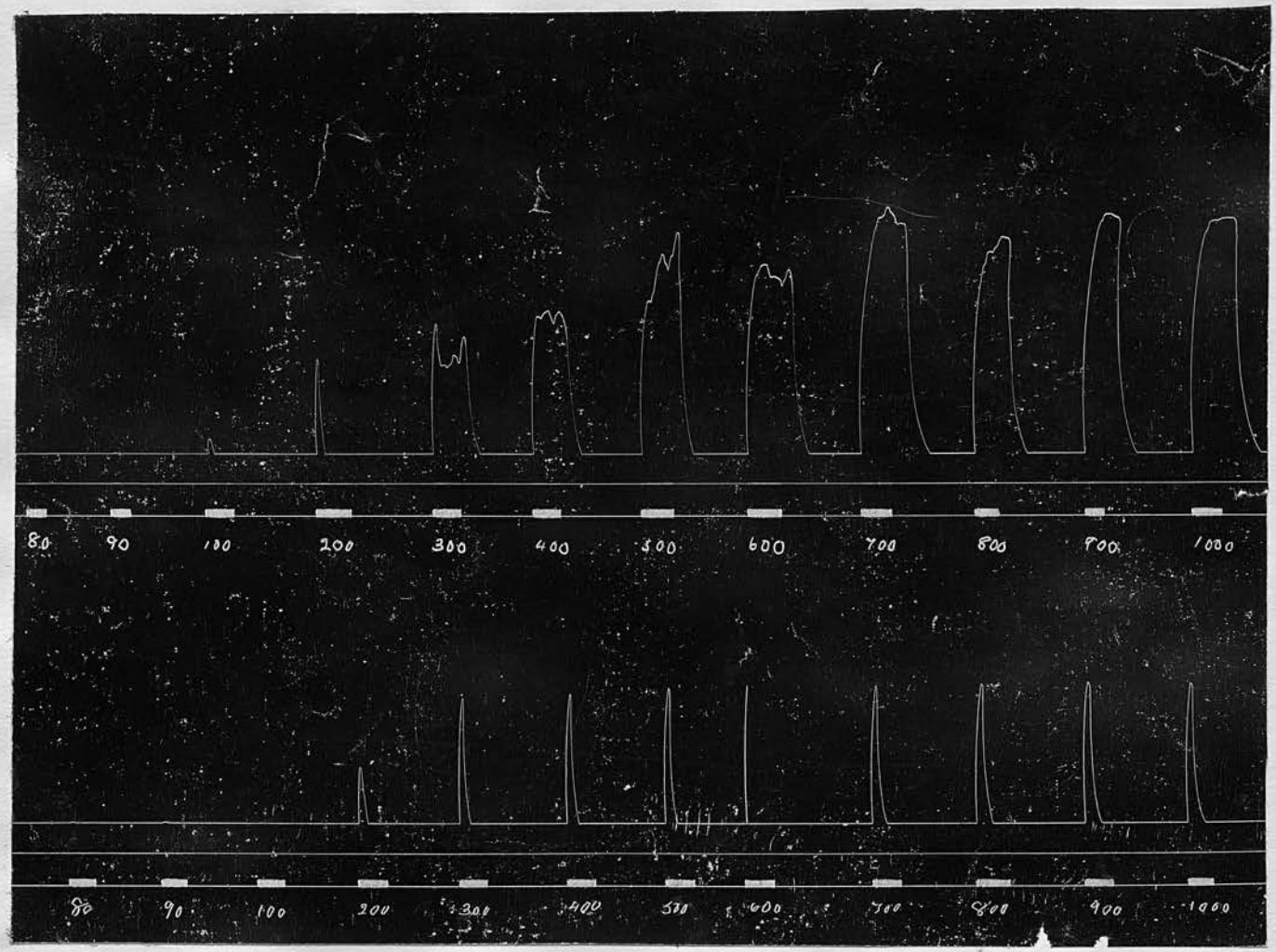
Such initial tetani are to be seen in any of the tracings in figures 17 to 20 In Fig. 18 their presence is complicated by the appearance of another phenomenon, the initial twitch, or "Anfangszuckung" of Bernstein.(63) These initial twitches occasionally occur on weak tetanisation, and the apparently arbitrary way in which they do occur led to a lively dispute between Kronecker and Stirling,(64) on the one hand, and Bernstein (65) on the other. The latter had found them present when the rate of stimulation began to be as high as 250 per second, the former two physiologists had been unable to observe them and denied their existence apart from physical errors in the apparatus. Their presence is indicated in a number of the charts in my possession but in none so plainly as in Fig. 18., where their unvarying height suggests some mechanical cause for their production in this special instance. Generally, however, these initial twitches vary in height with varying distance of secondary from primary coil, as shewn in figure 23 on the opposite page, where for some reason or other they have suddenly begun to appear, whereas in Figs 21 and 22, representing records taken a/

Fig. 24.



Rate 256 per second. Temperature −2·6°

Fig. 25.



Upper Line — Rate 214 per second. Temperature 1° − 2° C.

Lower Line — " 214 " " " −2° C.

a few minutes previously from the same preparation, they do not occur.

To return however to the initial tetani, their existence may be taken as an indication that the Wedensky effect under the given conditions is fully established, and, generally speaking, the better the Wedensky effect is established, the weaker the tetanisation, or, what amounts to the same thing, the lower the rate of stimulation required to produce an initial tetanus.

Thus in Chart 3 (lower line) we see a series of tetani inscribed at different temperatures as the nerve is gradually cooled from room temperature to - 1.3°C. The rate of stimulation throughout is 128 In the seventh series we see an indication of the near approach of the Wedensky effect, in that the tetani with strong stimulation are lower than those got with weak stimulation. In the eighth series we see tetani at 1000 and 2000 which are almost but not quite initial tetani. In the ninth and tenth series we see unmistakeable initial tetani, in each case corresponding to a lower intensity of stimulation as the temperature falls. The third tracing in the upper line shews the more or less complete restoration of the conductivity when the nerve is once more warmed to room temperature.

The same kind of thing is shewn in Chart 4 series(24) —— (31) inclusive.

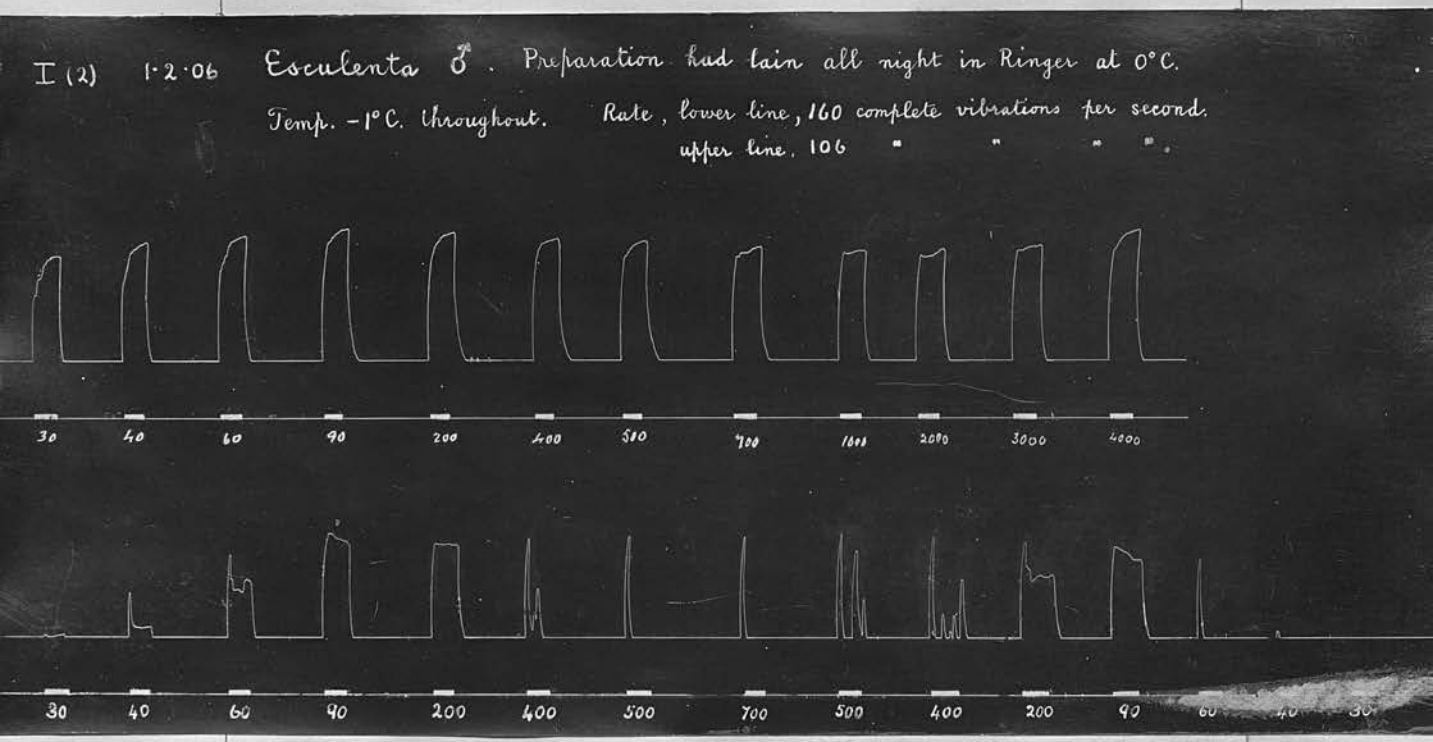
The presence of an initial tetanus, then, shews that for the given conditions the Wedensky effect is thoroughly established, and its position in a series of curves, taken with regularly increasing intensities, is an index to the degree in which the effect is present.

We might express the same fact in another way by saying that, as the conductivity of a nerve becomes more and more depressed with low temperature, the tendency is for the initial tetani in a Wedensky effect to creep in from the end of high intensity of stimulation and replace the full tetani got with lower intensities.

This leads us to ask if it is possible to have a Wedensky effect which shews nothing but initial tetani for all intensities of stimulation. The three tracings on the opposite page supply us with an answer. In the topmost tracing we see a series which is just on the point of becoming entirely initial tetani. Again in the lowermost tracing, got from a different preparation, we see a complete series of initial tetani, when the temperature is - 2°C; while in the middle tracing, taken a few minutes later at a temperature of +1° to +2°C, the normal kind of tetanus has returned again. Similar instances we shall see in many of the tracings to be subsequently described.

It/

Fig. 26.


I (2) 1·2·06 Esculenta ♂. Preparation had lain all night in Ringer at 0°C.
Temp. -1°C. throughout. Rate, lower line, 160 complete vibrations per second.
upper line, 106 " " " "
30 40 60 90 200 400 500 700 1000 2000 3000 4000
30 40 60 90 200 400 500 700 500 400 200 90 60 40 30

It is therefore evident that under special circumstances it may be impossible to obtain anything but single muscle twitches as the result of rapid rhythmical stimulation of the nerve.

This suggests a further question. What happens to such a series of initial tetani when, say, the temperature is still further lowered, or the rate of stimulation increased, or, in short, the conditions modified in such a direction as we already know to facilitate the appearance of the Wedensky effect? The answer to this question we shall give when we come to discuss the method of disappearance of the conductivity of a nerve.

(4) Rate of Stimulation.

It will already have become clear that the rate of stimulation is an important factor in the establishment of the Wedensky effect. The effect is invariably better obtained when the rate is high than when it is low.

Fig. 26 represents two series of tracings taken from one and the same preparation, the temperature being kept constant (1°C) throughout. The lower series was first inscribed with a rate of stimulation of 106 per second, the upper series was obtained after an interval of three or four minutes the rate having been meantime changed to 160 per second. The lower series shews a typical Wedensky effect./

effect. The upper series gives almost no indication of Wedensky effect.

[In this upper tracing we see evidence of a phenomenon which is apt to occur when one uses high intensities of stimulation. When a preparation shews signs of the proximity of the Wedensky effect, in that the tetani following stimulation at high intensities, (800 - 2000 in my experiments), begin to fall off slightly from the height of the preceding tetani, stimulation at a still higher intensity brings the tetani back to their full height. In the present case the tetanus at 4000 is higher than that at 2000. This effect is due to "escape" of the current and is of physical not of physiological origin. To avoid such a source of fallacy I rarely used stimuli of more than 2000 units.]

In chart 3 we have another record of the result of varying the rate of stimulation. All the tracings in the lower line were inscribed with a rate of 128 per second. In the last of the series, taken at a temperature of about $-1.2^{\circ}C$, the Wedensky effect is very pronounced. The first series in the upper line was inscribed when the rate had been changed to 30 ~~per second~~, the second when the rate was 80 per second. The temperature had meantime not risen.

In/

In experiments with an interruptor that consists of a vibrating steel spring, it is naturally difficult to obtain a graduated series of curves showing the effect of different rates of vibration, such as is possible in the case of different intensities of stimulation, the rate being meantime kept constant. The charts, shewn, however, are sufficient to indicate the general effect of varying the rate of stimulation, and the result is exactly the same as can be produced by varying the intensity. High rate corresponds to high intensity in its influence on the tetani of a Wedensky effect.

(5) The Length of Nerve Cooled.

A still greater difficulty is present when we try to vary the length of the cooled part of the nerve. In the absence of a special piece of apparatus with more than one compartment for fluid at different temperatures, the only method is to first of all observe the condition of the Wedensky effect in one definite preparation, with given low temperature, and given rate of stimulation, and then at once repeat the observations when a chamber of different size has been adapted to the pipe through which the cold fluid comes. I had three chambers of different width, 1 inch, $\frac{3}{4}$ inch and $\frac{1}{4}$ inch respectively. I often observed/

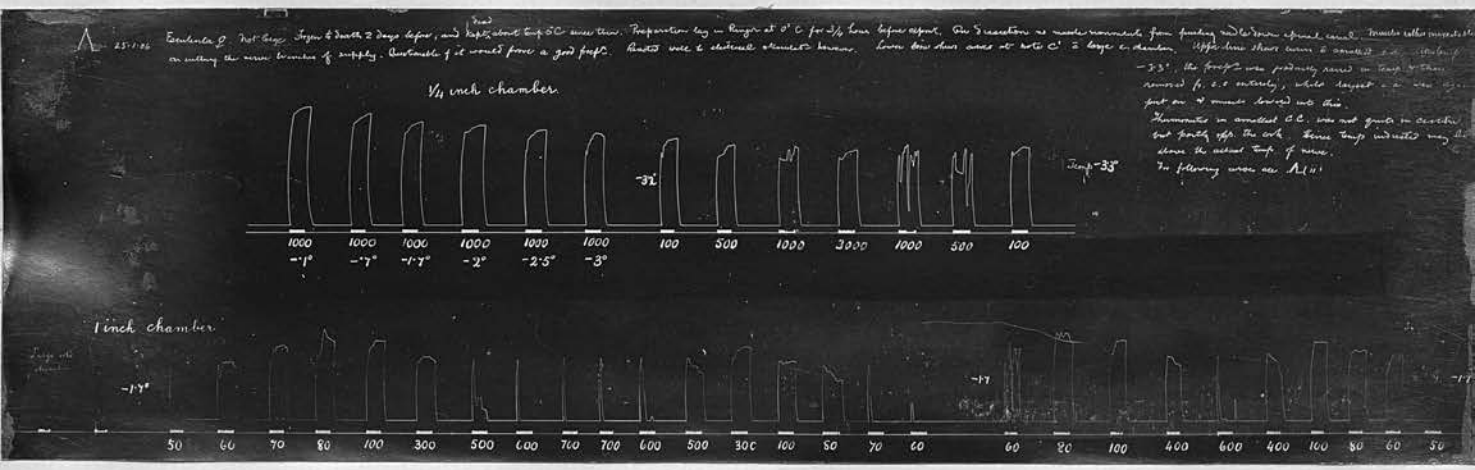

1/11 inch chamber
1000 1000 1000 1000 1000 1000 100 500 1000 3000 1000 500 100
-1° -1·7° -1·7° -2° -2·5° -3°
1 inch chamber
50 60 70 80 100 300 500 600 700 700 600 500 300 100 50 70 60 60 80 100 400 600 400 100 80 60 50

Fig. 27.

observed, ~~that~~ when the Wedensky effect was only with difficulty elicitable by the use of the $\frac{3}{4}$ inch chamber, that on using the largest sized chamber it became fully present.

Fig. 27 represents an experiment carried out to shew the dependence of the Wedensky effect on the length of nerve cooled. In the lower line of the figure we see that with one inch of the nerve cooled to -1.7° an initial tetanus is first got at an intensity of 600. For accuracy this observation was repeated twice. In the upper line a series of tetani at a constant intensity of 1000 ~~are~~ is inscribed at varying temperatures. Although the thermometer in this case, from insufficient adaption to the coldest part of the groove, probably registered a higher temperature than the actual temperature of the coldest part of the nerve, yet we see that at a temperature of -3°C, almost complete tetanus follows stimulation at 1000. Only when the temperature falls to -3.2° are there indications that the Wedensky effect is on the point of appearing. After the inscription of this tracing the nerve was once more put back in the larger chamber, and the Wedensky effect was found to return to about its primitive extent at a temperature of -2.7°C.

(6)/

(6) The Relation of the Initial Tetanus to the Refractory Period.

We have already discussed how Fröhlich brought the initial tetanus into relation with the refractory period. I carried out some experiments with the object of determining if an exact relation exists between the two.

The method used to determine the refractory period was that used by Boycott[60], depending on the muscular response to two successive stimuli. Two induction coils, each with an accumulator charged to 2¼ volts in the primary circuit, were placed at a distance from and at right angles to each other. The two secondary circuits were connected by a cross-wire, and the remaining terminals connected with the exciting electrodes. The primary circuits were broken by the knocking over of two keys in a heavy pendulum myograph similar to that used by Sewall ( ) in his experiments.

In one of the primary circuits a vibrating spring interruptor, adjusted to give, say, 100 breaks per second, could be interposed at will instead of the kick-over key of the pendulum myograph. The time interval between the kicking over of the two keys was then adjusted to .01 second. The nerve was cooled/

cooled to a definite low temperature and the position of the one secondary coil sought out which just gave an initial tetanus at the given rate of stimulation. The other secondary coil was arranged at a corresponding distance, and records taken ~~by means~~ of the effect of two successive breaks.

The results, as tending to establish a precise relation between refractory period and initial tetanus, proved very unsatisfactory. All that one could say was that the refractory period was much prolonged by cooling; the actual time measurements obtained in the two cases did not at all correspond, nor was the discrepancy got over by assuming that both make and break of the vibrating spring were effective. Recognising that these were important sources of error in the experimental method employed I gave up these experiments at the time and have not since had an opportunity of repeating them by unobjectionable methods.

The sources of error in the arrangement I used are:- (1) The want of relation between the graduations of two induction coils taken at random. A distance of say, 40 cm. in the one does not necessarily correspond, as regards exciting effect on the nerve, to a distance of 40 cm. on the other. It is known that the refractory period depends on the strength of the stimulus, (Gotch and Burch (18)), not only so, but I have found that the refractory/

refractory period for two stimuli of unequal magnitude may possibly be a very different thing from the refractory period for two equal stimuli (see p.) This difficulty could be got over by using two standard Kronecker coils.

(2) The different rate of break given by the kick-over key as compared with the vibrating spring interruptor. This source of error is present in any experiments where a comparison is made between the shocks given by two different forms of break apparatus. The difficulty might be got over, as suggested to me by Dr. Harry Rainy, by using a falling weight to make contact for any required length of time and thus picking two or three or more excitations at will out of a series of regularly recurring stimuli.

(3) Self-induction and magnetic hysteresis affecting the intensity of the current in the Secondary coil. This trouble would not occur if one used the method suggested in last paragraph.

(4) The inequality between the "makes" and the "breaks" of the vibrating spring. This can be avoided by using proper condensers, or better still when one requires rapid rates of stimulation, by using alternating currents instead of induction shocks to give the Wedensky effect. I have found that the Wedensky effect can be quite well elicited by means of alternating currents.

At present I am making arrangements to carry out a series of experiments with alternating currents and a falling weight to pick out any required number of

stimulations.

In the meatime all we can say is that Fröhlich's claimed relation is extremely likely on general grounds. It can hardly be said to have been experimentally proved, for much the same objections apply to his experiments as to mine.

SECTION C. FACTORS THAT COMPLICATE THE WEDENSKY EFFECT.

Hitherto we have spoken of the Wedensky effect as if it depended merely on certain definite physical quantities, viz., (1) the length of nerve cooled, (2) the degree to which the portion of the nerve is cooled, (3) the rate, and (4) the intensity of stimulation. Given these four quantities it might be imagined that the degree of the effect would be determined. This however is not the case. As stated in the beginning of last section certain complicating factors come in.

First of all, there are physiological differences in nerves, whereby they react in varying degree to similar experimental conditions.

Again, the mere keeping of a dissected preparation affects its responses when the Wedensky effect is tested for.

Further, a nerve is not normally in a stable condition when cooled: low temperature of itself causes a slow and progressive fall in conducting power.

Finally/

Finally, the question of fatigue by repeated stimulation seems to influence the result.

The last factor from its special interest we shall reserve for a separate section. The other influencing conditions we shall now discuss.

(1) Physiological Differences in Nerve.

When I first carried out experiments to determine if the Wedensky effect could be elicited by cooling I had no difficulty in shewing the condition. The experiments were carried out in Göttingen in January and February 1904. Figures 17 and 18 are specimens of the tracings then obtained. On resuming the work in Edinburgh in November 1905, I failed entirely to get the Wedensky effect. No matter how far I reduced the temperature the tetani for all intensities of stimulation kept complete and of good height.

At first I thought the anomaly might be due to some change in the physical apparatus employed. A number of alterations were consequently carried out in the apparatus, all in the direction of making the effect more readily elicitable. Thus, the rate of stimulation was made high, while to keep the interruptions regular the interruptor was removed from the table on which the apparatus stood and placed on the stone ledge of the window where it would be less affected by accidental vibrations. A very broad brass chamber

was/

was made so as to cool as great a length of nerve as possible, and larger frogs were used. The iron core was removed from an inductorium so as to abolish magnetic hysteresis and make the growth and decline of the current in both primary and secondary coils more abrupt, while to compensate for the loss of the core a current of much higher amperage was used. Finally, the whole apparatus was taken down and built up afresh. All was of no avail.

Remembering that Fröhlich had got the effect in the most pronounced form when the conductivity, as tested by single maximal twitches, was just on the point of disappearing, I cooled the nerve gradually, testing excitability and conductivity at each successive stage. Excitability gradually fell and conductivity kept constant, but before conductivity disappeared the nerve always froze and put an end to the experiment. Such experiments I carried out for over a month without once observing either Wedensky effect or abolition of conductivity with low temperature.

It then seemed that the most probable cause of the anomaly must lie in the condition of the nerve itself. In Germany I had got the result with both Rana temporaria and Rana Esculenta, and in male and female specimens of either variety, so that it could not/

not depend on a specific or on a sexual difference in the frogs. Seeing that I was looking for what was possibly a fatigue effect, and knowing as I did that the nerves of summer frogs are less tolerant of exposure etc. than the nerves of winter frogs (conf. too, p.p. 20, 21)- while Fröhlich had found (see p.25) that cooled nerve takes a much longer time to asphyxiate than summer nerve, I imagined that fatigue might be more readily got in warmed than in cooled frogs. A batch of frogs was accordingly kept in the room for some days at a temperature of about 12°C, some of them were even warmed up till tetanic convulsions appeared. Their nerves however gave the same negative result.

It was then suggested by Professor Schäfer that as I had got the result in Germany in February, i.e. during the cold time of the year, I should try the effect of cooling the frogs on ice before using their nerves. On adopting this plan I got the effect at once, and during the rest of the winter I did not fail to elicit the desired effect in one single case where the frogs had been previously cooled. Nay more, it seemed to be the case that the longer a frog was cooled beforehand the more readily was the effect elicited. I did some controls with frogs taken directly/

directly from the ranarium. In two cases I got nothing but a mere indication of the effect: in other cases the effect was elicited, but the weather at that time happened to be cold. From these experiments I at first concluded that the winter condition of a preparation was an essential for the ready obtaining of the Wedensky effect by a subsequent cooling of the nerve.

From January to March 1906, I used cooled frogs and got the result every time. During the months of April and May I carried out no experiments. On continuing my observations in June I found that in spite of previous cooling of the animals the effect was not elicitable. For two weeks during that month I did not get the effect once in spite of prolonged previous cooling. Instead of the normal Wedensky curves a series of very irregular and broken tetani was got with all intensities of stimulation but in no case was an initial tetanus seen. From these experiments it was clear that previous cooling is at least not the only factor involved.

The frogs I used at that time belonged to the same batch as some from which I had got the effect without difficulty in the early part of the year. As the animals had not been fed for some months it was a natural idea that the change might be due to starvation. Some of them were accordingly "stuffed" with frog's flesh in the manner adopted by Fröhlich (29)

(see p.25). They were kept for a few days under normal conditions so that digestive and absorptive processes might go on, after ~~that~~ which they were placed on ice for some time and their preparations examined.

The result of these experiments, important enough as they possibly were, was however overlooked by me at the time owing to my attention being simultaneously directed to a number of other interesting and perplexing facts. Thus, I discovered that a preparation might give the Wedensky effect at a low temperature without any previous cooling of the animal. Nay more, all the presumably starved frogs which I still had now gave the Wedensky effect whether they had been cooled or not.

It is only on looking back over my charts that I have discovered that these feeding experiments are suggestive. I have records of only two animals which were fully fed. They were both kept on ice for some time after feeding. Charts 5, 6 and 7 shew the result of experiments with these. In Chart 5, (tracing 2) we see that conductivity is totally abolished at + 4°C. Chart 6, a record from the opposite leg of the same frog, shews (tracing 3) that a Wedensky effect occurs at + 8°C, while between +8° and +6° nothing but initial tetani are got. Again, Chart 7, shews that the/

the conductivity is abolished at+1.6° (tracing 1), or+2°C (tracing 5). Now, the disappearance of the conductivity at a temperature above 1°C has been observed by me in no other instance, and it would seem that feeding had possibly some effect in bringing about this condition. The experiments, are, however, merely suggestive. It would be dangerous to argue from two single cases. Against the evidence given by them we have the fact mentioned in last paragraph that seemingly starved preparations give the Wedensky effect. Unfortunately since the month of July I have had no frogs that did not give the Wedensky effect, and I have carried out no further feeding experiments.

To return however to the question of previous cooling of the animals, I have found contrary to my original opinion that the "cooled" or "winter" condition of a frog is a factor of practically no importance as tending towards the eliciting of the Wedensky effect by a subsequent cooling of the nerve. A number of frogs was taken from a definite batch and cooled for some days on ice. Their preparations were then examined against the corresponding preparations of their fellows which had been kept under normal conditions of temperature. No difference in result was obtained according as the preparations came from "cooled" or from "non-cooled" animals.

Consequently/

Consequently I never now cool the animals beforehand when testing for the Wedensky effect.

I have investigated the effect of one other possible factor, viz., the influence of altering the amount of water in the tissues of the animal by subjecting it to continuous evaporation. (66) Durig found that frogs may lose in 2-3 days by evaporation from 20 - 30% of their weight; in one case he succeeded in diminishing the weight of a frog by 39% without killing it. In the process of drying a great concentration occurs in the body fluids, the concentration being least marked in the brain and heart of the animals. On the assumption that the nerves participate in this drying up process I took a number of frogs from a batch which were giving the Wedensky effect well. These were weighed and placed in an open wire cage outside the laboratory window. The experiments were carried out in winter when the outside temperature was but a few degrees above zero. In 1 - 3 days they had lost from 10 - 28% of their weight. Specimens were examined in various conditions of dryness. In no case was the slightest departure from the normal degree of Wedensky effect observed, although care was taken not to moisten the dissected nerve in any way.

Charts/

Charts 8 and 9 shew typical examples of the kind of curves got in dried preparations. In Chart 8, when the frog had lost 18% of its weight by drying, we see (tracings 3 and 5) a Wedensky effect setting in when the temperature has reached the normal low point at which a Wedensky effect generally occurs. In tracing 6 initial tetani are seen at all intensities of stimulation. In Chart 9, (the frog having in this case lost 28.6% of its weight), we see a Wedensky effect at - 1° and at -2° followed by loss of conductivity at lower temperatures. A comparison of these two charts with chart 10, which is a record taken from an animal of the same batch, but not dried, shews that the Wedensky effect in each is in substance the same.

[In the experiments of which these charts are records the rate of stimulation was 144 per second. This rate was constantly used by me in a large number of experiments, and hereafter in the description of charts when the rate of stimulation is not specifically mentioned it will be understood that 144 was the rate used.]

Before concluding this question of physiological variations in nerves it might be well shortly to sum up the facts.

It has been found that the conductivity of a nerve/

nerve is influenced by low temperature to very varying degree according to the condition of the nerve. While the conductivity in special cases may be abolished at as high a temperature as + 4°C, and with a rate of stimulation of between 100 and 200 per second the Wedensky effect elicited at + 8°C, in other cases again, no lowering of the temperature short of freezing can either produce the Wedensky effect with ~~the same~~ this rate of stimulation or abolish the conductivity.

Previous subjection of the living animals to definite temperature conditions, within the limits of 0°C and + 16°C, seems to have no effect on the behaviour of their dissected nerves towards cooling.

Different degrees of concentration of the body fluids likewise seem to be without effect on the reactions of nerve to low temperature.

No definite seasonal variation apart from nutrition has been made out, and no definite relation to the reproductive period has been established.

The variations are neither specific nor sexual.

The only influencing factor for which there is any slight experimental evidence is the state of nutrition of the nerve. There is some meagre evidence to indicate that a well nourished nerve may be more affected by low temperature than a badly nourished nerve.

Fig. 28.

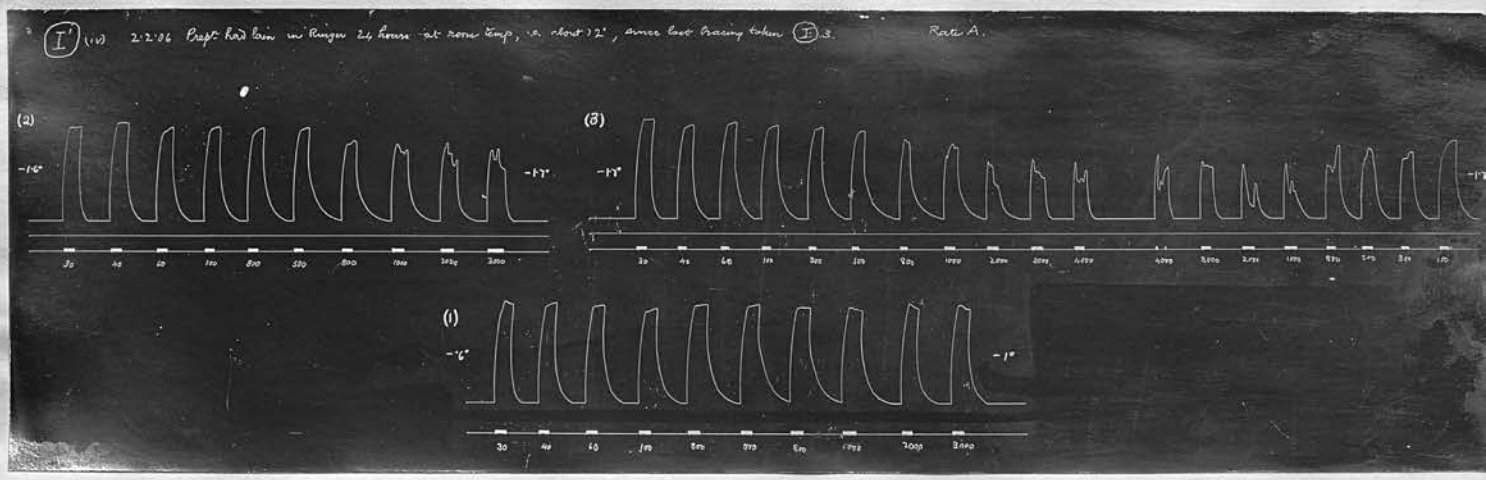
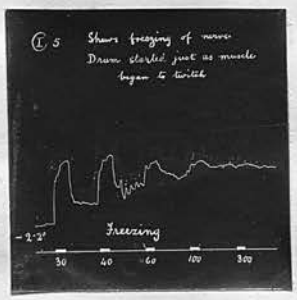

(I') (10) 2.2.06 Brept: had been in Ringer 24 hours at room temp, i.e. about 12°, since last bracing taken (I) 3.
Rate A.
(2)
-1.6°
-1.7°
(3)
-1.8°
(1)
-1.6°
-1°

Fig. 29.


(I) 5 Shows freezing of nerve
Drum started just as muscle
began to twitch
-2.2°
Freezing

(2) Change Undergone by Nerve on Keeping.

I had not experimented long on the Wedensky condition till I began to notice that the preparation from one side of a cooled frog would give the effect quite well in the morning, while the sister-preparation, which had meantime been kept lying in Ringer's solution in a beaker in the room, would fail to give the effect in the afternoon. The fact was so marked that it was difficult to believe that the preparation tested in the afternoon was originally in a different physiological condition from that used in the morning.

I accordingly tested the effect of "keeping" on one and the same preparation.

Figure 28 shews the result of such an experiment. The same preparation which gave the tracings seen in figures 19 and 20, after one night's soaking in Ringer's solution, gives, at corresponding temperatures and rate of stimulation, the result now seen. The nerve was even cooled to the freezing point (Fig.29) and yet the tetani kept complete and full. Previously, as we saw (Fig. 20, tracing 4), the conductivity became abolished at - 2°C. This example is typical of what happens after keeping in Ringer's solution. Though the Wedensky effect may be elicitable at a moderately high temperature, and the conductivity of the nerve spontaneously disappear before the freezing point is reached, yet after soaking in Ringer's solution for some time these effects disappear.

One/

One naturally asks if this result is due to the action of the Physiological solution or whether it is simply an incident in the gradual death of the nerve. As yet I have carried out only two experiments to decide the point. They are not convincing, but would seem to indicate that the change is due to the action of the solution in which the nerve is kept. Charts 11 and 12 illustrate an experiment carried out with a view to decide this point. In Chart 11 we see a record taken from a fresh preparation. The Wedensky effect is apparent at about 0°C. Conductivity disappears at - 1°C. The sister preparation was meantime kept undissected, i.e. with the nerve *in situ* among the muscles, in a beaker of Ringer for 26 hours. The nerve was thus partially protected from the action of the solution during this time. When it was cooled it gave a marked Wedensky effect at temperatures ranging from 0°C to -1°C., with ultimate disappearance of conductivity at a lower temperature. My experience with preparations exposed directly to the action of Ringer's solution had taught me not to expect such a prolonged continuance of the Wedensky effect as this.

That in some cases the effect of keeping in Ringer may be very pronounced is proved by Chart 13, which is a record, after keeping for 24 hours, of one of the "fed" preparations which we saw in Charts 5 and

6/

6 to give disappearance of conductivity at relatively high temperatures. As we see, a real Wedensky effect is not elicitable, though the conductivity (tracing 8) is evidently brought almost to the verge of disappearance.

The broken character of the tetani in this chart is typical of another change often brought about by keeping, viz., the nerve frequently seems to conduct rhythmical excitations in a more erratic fashion than before. Again, after keeping, high intensities of stimulation do not necessarily tend to bring about well marked initial tetani in the stage immediately preceding disappearance of conductivity. The first 7 tracings of Chart 14 illustrates the effect of $3\frac{1}{2}$ hours keeping on a preparation which gave a normal Wedensky effect at $-.3^{\circ}$ followed by complete disappearance of the conductivity (See Chart 15). Tracing 7 shews that the muscle responses, even when the effective conductivity is practically gone, shew traces of being tetanic contractions rather than single twitches. The two tracings on the second part of the same chart shew the broken character of the tetani due to keeping. This record was taken from the sister preparation, which had been kept for 5 hours in Ringer's solution. When it was fresh it would have presumably given a normal Wedensky effect.

An/

Fig. 30.

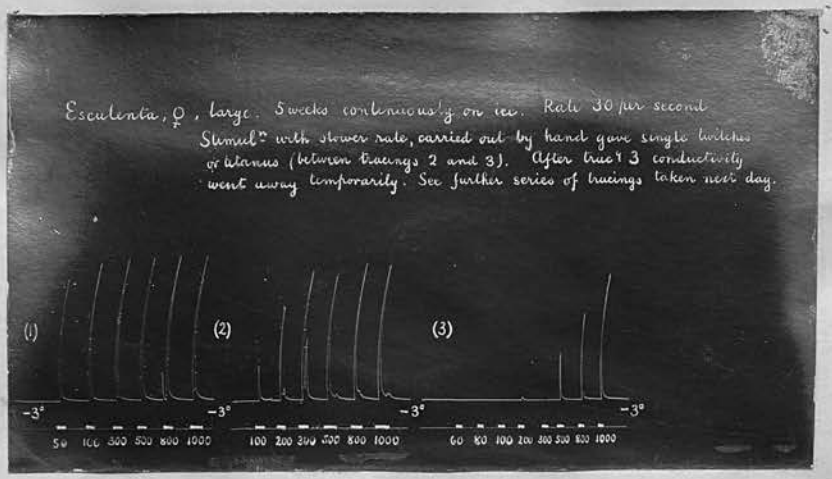


Fig 31.

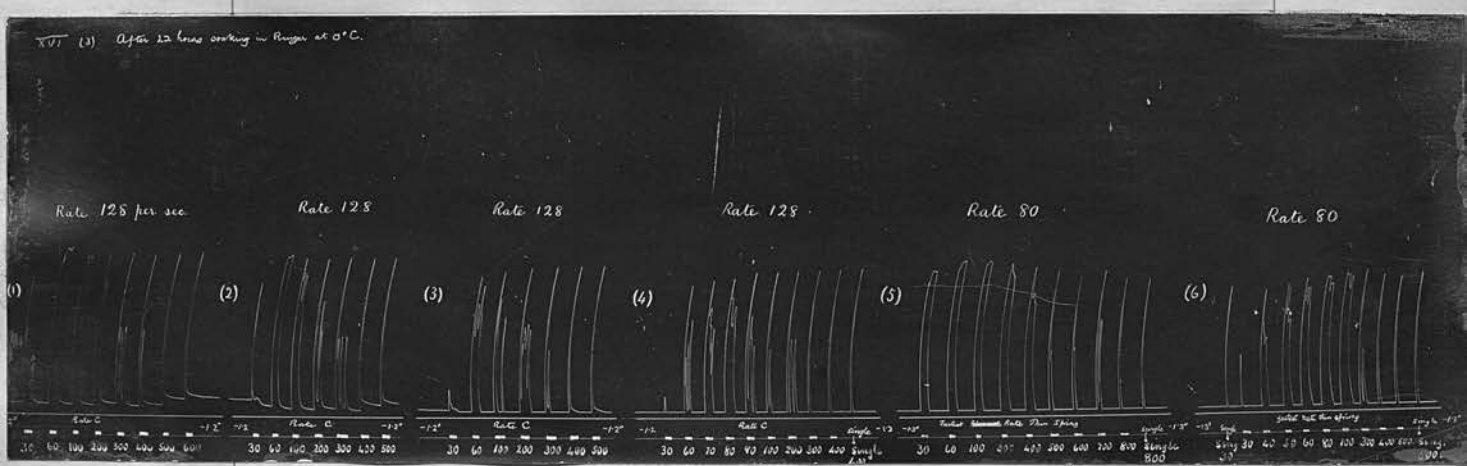


Fig. 32.

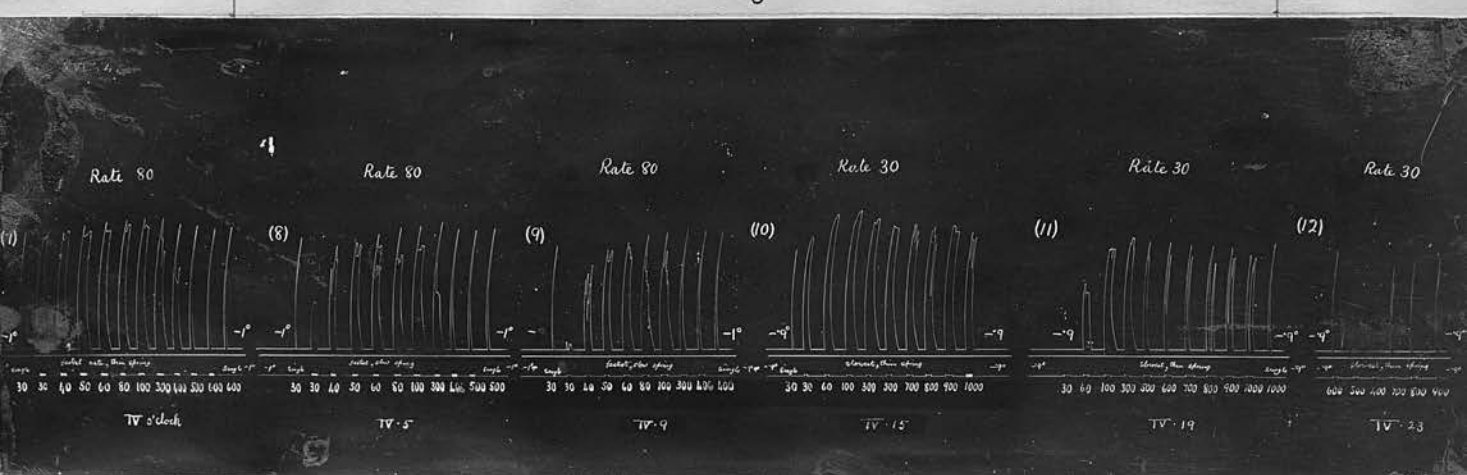


Fig. 33

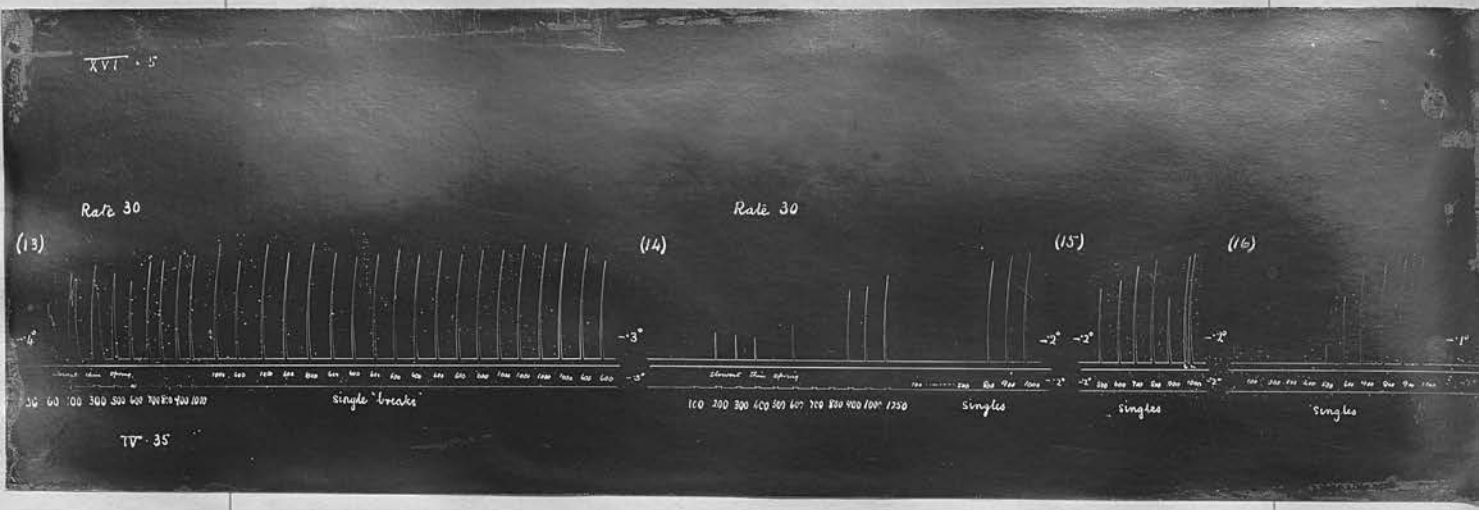


Fig. 34

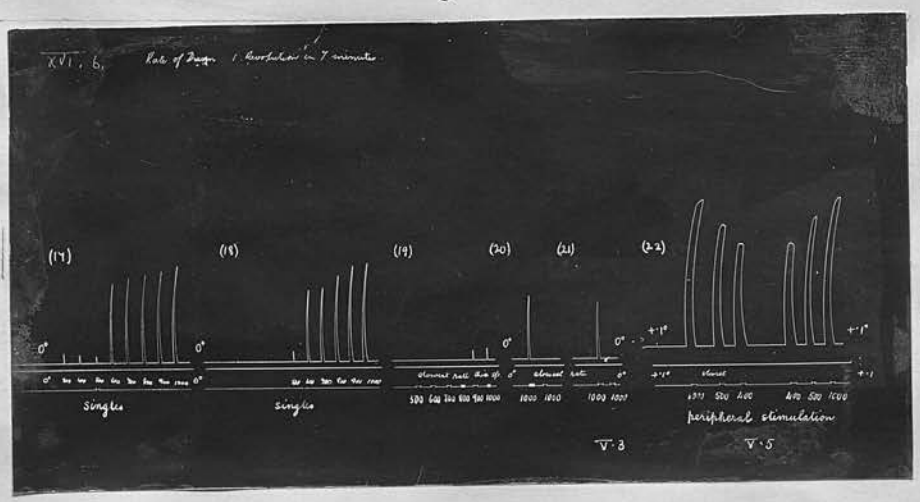
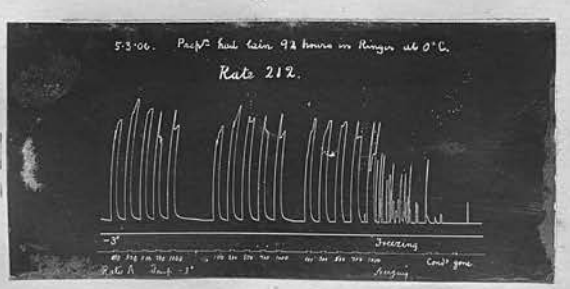


Fig. 35.



An attempt has been made to determine whether the rate of the change that occurs with keeping can be influenced by temperature. A considerable number of experiments has been carried out to settle this question. From the amount of variation in different preparations as regards viability etc. the result is not absolutely definite, but the evidence on the whole tends to shew that temperature has little if any influence on the rate of change. Certainly the change goes steadily ahead in spite of a temperature as low as 0°C. A comparison of Fig. 30 with Fig. 35 shews this well. In Fig. 30 we see an exceptionally well marked Wedensky effect, in that nothing but initial tetani are got with a rate of stimulation of 30 per second. In Fig. 35 again, we have a tracing taken from the same preparation after 92 hours soaking in Ringer's solution at 0°C. The rate of stimulation is now 212, and yet the Wedensky effect is hardly present when the temperature has been brought right down to the freezing point. Figs. 31, 32, 33, and 34 which represent records taken from the same preparation after 22 hours keeping at 0°C, shew that the Wedensky effect is still present even for low rates of stimulation.

A preparation kept at room temperature (about 12° C) shews the same progressive change, whereby the conductivity is less affected by low temperature, but no difference in the rate of this change depending on the temperature at which the living animals have been kept

Fig. 36.

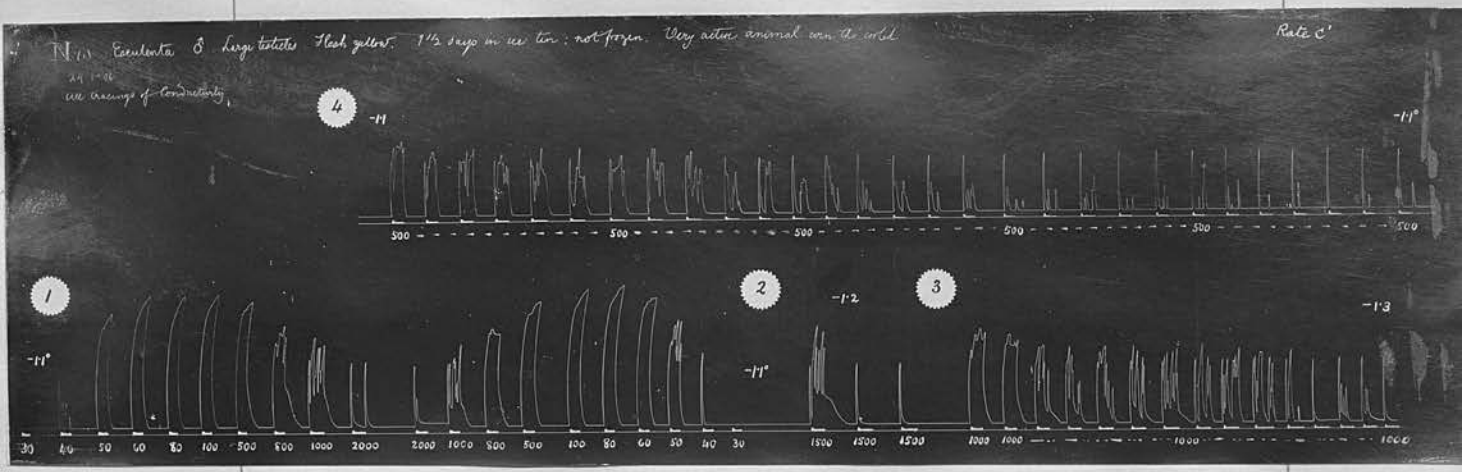
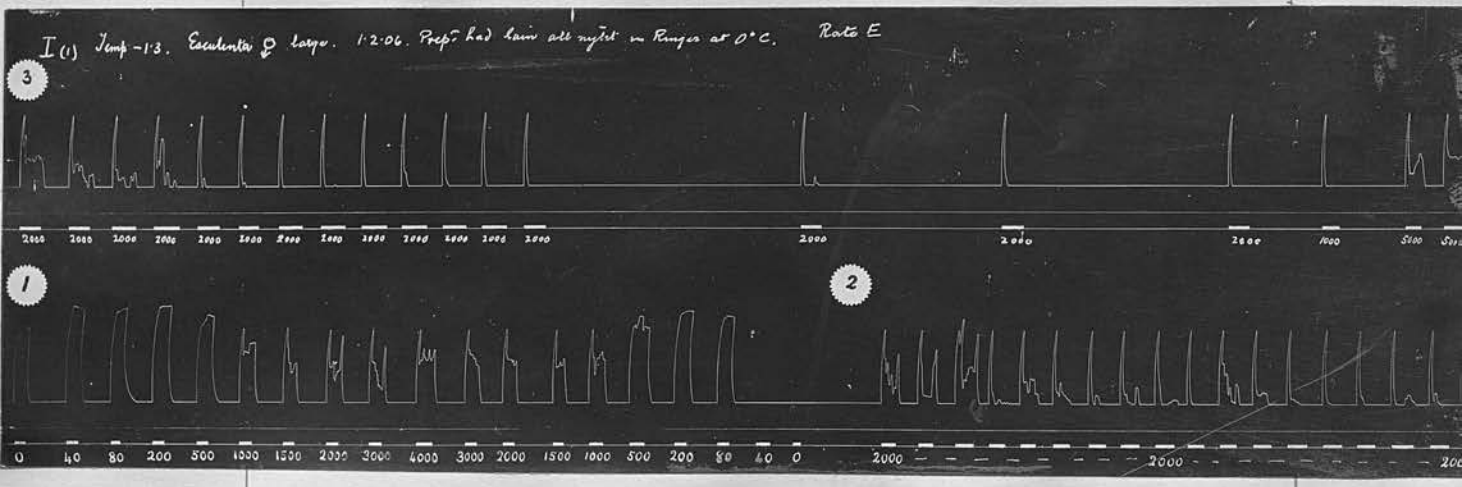

Rate C'
-1·1°
-1·2
-1·3
-1·1°

Fig. 37.


I (1) Temp -1·3. Esculenta ♀ large. 1·2·06. Prep. had been all night in Ringer at 0° C. Rate E
3
1
2
0 40 80 200 500 1000 1500 2000 3000 4000 3000 2000 1500 1000 500 200 80 40 0
2000

(3) Progressive Diminution of Conductivity with Constant Low Temperature.

In most work done by previous investigators on cooled nerve it has been assumed that the conducting power is a function simply of the temperature. If the temperature of a definite length of nerve be kept constant it has been taken for granted that the conducting power of that portion does not vary. The conductivity of nerve, when the temperature is so low that the Wedensky effect is readily elicited, seems however, to be a function of time as well. At a constant low temperature the conductivity tends slowly to disappear.

This gradual effect of constant low temperature may be demonstrated in more than one way. Thus, when a Wedensky effect has been elicited at a definite low temperature, one may, while keeping the temperature steady, repeat at intervals a series of rhythmical stimuli of an intensity just sufficient to give a "descending tetanus." The succeeding tetani then gradually assume the form of initial tetani. Fig.36 illustrates such an experiment. In the first tracing we see a typical Wedensky effect. In tracing 4, where the temperature is recorded as constant throughout, we see that the descending tetani with constant intensity of stimulation gradually tend to become initial tetani.

Fig./

Fig. 38.

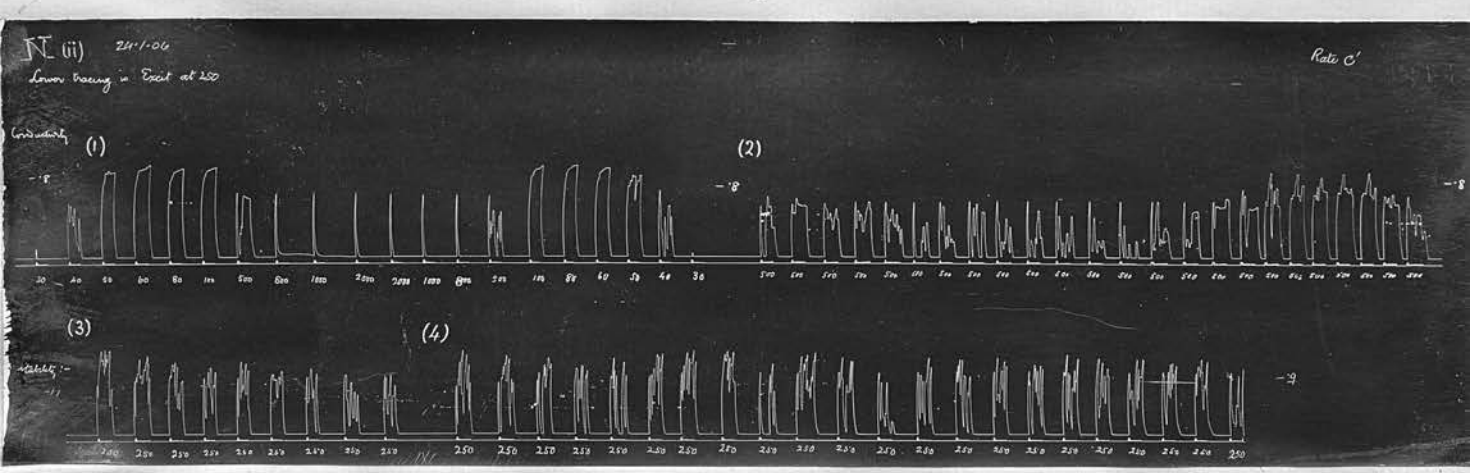


Fig. 37 shews a similar experiment, the temperature being kept constant (- 1.3°C) all the time.

Two possible objections to these experiments might be advanced. First of all, it might be said that the temperature of the nerve is not really constant; there may have been a slight but unobserved fall of temperature all along. Again we do not know but that repeated strong stimulation of a nerve at low temperatures may <u>fatigue</u> it; it may be that when much cooled the nerve recovers less and less perfectly after each successive series of stimuli.

That the first objection is pertinent appears from such a case as is shewn in Fig. 38. Here (tracing 2), in spite of the registered temperature being constant the nerve is affected by a kind of wave of increased conductivity; just as if, say, some accidental current of air had temporarily warmed up the nerve without affecting the thermometer. Whether this be the explanation or no, we cannot in any case say that with the form of apparatus used the nerve is absolutely protected from slight alterations in temperature. All we can say is that in the great majority of cases when the registered temperature remains constant, the result falls out as we have seen in Figs 30 and 31.

But/

But we are not restricted to this method in demonstrating progressive depression of the conductivity of a nerve as the result of it being kept continuously at a low temperature. Possibly the best mode of proving the fact is by experiments carried out in the manner shewn in charts 15 and 16, in which, as soon as a well marked Wedensky effect is elicited, the temperature is allowed slowly to rise, In spite of the fact that the temperature is rising the conductivity may with time disappear and continue absent until a temperature is reached which is much higher than that at which the conductivity originally went away. In the experiment shewn in Chart 15, observations were carried cut every five minutes, the temperature meantime being allowed to rise slowly. A Wedensky effect was first of all elicited when the temperature was falling from - .4° to - .6°C. The temperature continued to descend to - .8° C., after which it was made to rise again by slow stages. At - .3° a Wedensky effect is still present. Five minutes later at 0°C. the conductivity is just present and no more. After another five minutes rest at 0°C the conductivity is absent and continues absent for 35 minutes, until a temperature of + 6°C is reached. Even at + 8° C it is not fully restored. Chart 16 shews the same kind/

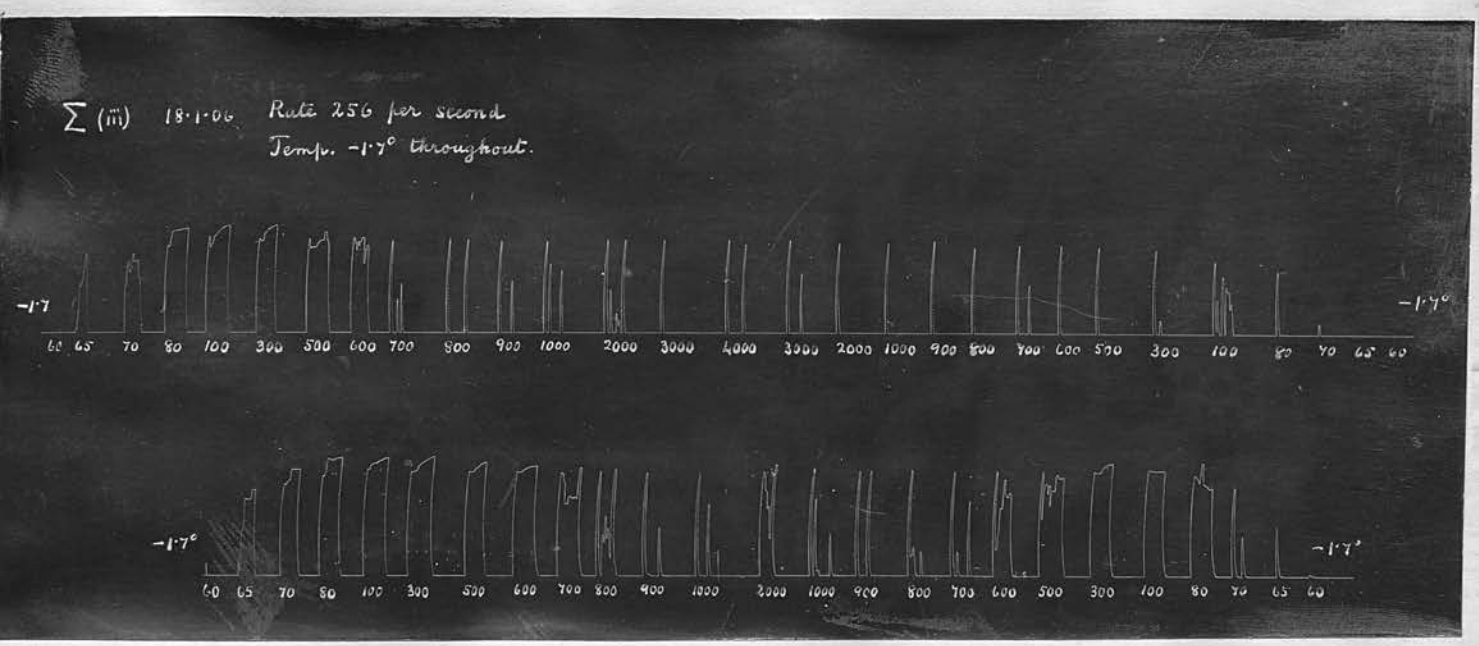
kind of result. These experiments demonstrate in striking fashion that the condition of a nerve at any moment as regards its power of conduction is not a function simply of the temperature of the nerve for the time being. The action of low temperatures is more complex than has been hitherto supposed.

It is possible that the experiments of Gotch and Burch [18] from which they concluded that an excitatory change may take place in a nerve without a corresponding electrical change, may ultimately be explained on the basis of slow changes started in the nerve when once the temperature has been altered.

That the absence of conductivity in Charts 15 and 16 is due, not so much to fatigue, as to the continuance of the low temperature is indicated by the fact that the conductivity seems to progressively disappear with time, and not by jumps, as would be the case if the disappearance were the immediate result of stimulation. A comparison of tracings 2 and 3 in Chart 15, and of tracings 5 and 6 in Chart 16 will shew that during the interval of rest in each case a great diminution of conductivity has occurred. Still, tracings 3 (chart 15) and 6 (chart 16) by themselves would suggest that fatigue may play a part in abolishing the conductivity.

An/

Fig. 39.


Σ (iii) 18·1·06 Rate 256 per second
Temp. -1·7° throughout.
-1·7
-1·7°
60 65 70 80 100 300 500 600 700 800 900 1000 2000 3000 4000 3000 2000 1000 900 800 700 600 500 300 100 80 70 65 60
-1·7°
-1·7°
60 65 70 80 100 300 500 600 700 800 900 1000 2000 1000 900 800 700 600 500 300 100 80 70 65 60

An exactly analogous instance of the gradual action of low temperature on nerve is seen in the series of figures 31 to 34 , which represent consecutive tracings taken on the same day from one definite preparation. In tracing 6, where the temperature is - 1.3°C a Wedensky effect of definite "degree" is elicited with a rate of 80 per second. From this point onwards a gradual fall in conductivity is apparent, although each successive tracing is recorded at higher and higher temperatures. Finally at 0°C (tracing 21) the conductivity disappears. In this case, as contrasted with the cases of Charts 15 and 16, the conductivity at the start was present in considerable amount. By the application of the Wedensky method of examination however it was possible to follow out the gradual change of conducting power, a thing which previous to the introduction of this method would have been very difficult if not impossible.

Still another illustration of the fall in conductivity with constant low temperature is probably provided by experiments carried out after the manner of Fig. 20, tracings 1 and 2, of Figs 26 and 27. or especially of Fig. ~~38~~ 39. In these cases we see that on repetition of a Wedensky series in the reverse direction, i.e. with decreasing instead of increasing intensity/

intensity of stimulation, the second record contains fewer full tetani than the first. The decrease takes place especially at the expense of the right-hand members of the series, i.e. in the responses to weak stimulation. It must at the same time be admitted that this decrease does not occur in all cases where the temperature has been apparently kept constant, (see, e.g. Figs. 17, 18, and 30); and, further, that in the instances in which such a decrease is apparent, very high intensities of stimulation have generally been used.

To sum up, we see that in certain cases at any rate, low temperature brings about a progressive diminution of the conducting power of a nerve. Consequently we cannct assume the conducting mechanism of a nerve, when it has been suddenly lowered in temperature, to reach at once a stable condition.

The diminution of conductivity that goes on in a nerve kept at constant low temperature is a slowly acting process.

Once conductivity has been abolished at a low temperature it does not return for some time provided the nerve be slowly raised in temperature. The ultimate return takes place at a temperature higher than that at which conductivity became abolished.

Section D./

SECTION D. MODE OF DISAPPEARANCE AND OF RETURN OF THE CONDUCTIVITY.

We have already seen (pp. 72-3) that in extreme conditions a Wedensky effect may consist of nothing but initial tetani, and we were led to inquire how the conductivity ultimately disappears. We likewise found (Section C. 3) that when the conductivity does go away at any definite temperature it may remain absent until a temperature is reached which may be much higher than that at which the nerve originally ceased to conduct. It is interesting to know whether in such a case the conductivity reappears in full amount at once or whether it only gradually is restored. The discussion of these two questions we shall now take up.

(1) Disappearance of Conductivity.

Figure 30 shews an extreme condition of the Wedensky effect. Not only are the muscle contractions for all intensities of stimulation initial tetani, but the rate of stimulation is only 30 per second. In tracing 1 the contraction at 30 is less than those at higher intensities. In tracing 2 a small contraction is elicited at 100, while only at 800 and 1000 are maximal contractions got. The height of these maximal contractions is less than in tracing 1.

In/

In tracing 3, again, it is very evident that the contractions are higher, the higher the intensity of stimulation. A very short time after this record was taken the conductivity was found to have disappeared. Similar results may be seen in Charts 16 and 17 (upper line in each case).

In these instances the conductivity has been caught just in the act of disappearing, as it were, and we see that it goes away in a definite march or order. It disappears first for low intensities of stimulation, while the conductivity for higher intensities is the last to go.

This is the order of disappearance not only when series of rhythmical stimuli are used to excite the nerve, as in the cases just seen, but also when single excitations of different intensity are used. Figs. 33 and 34 , (which represent an experiment carried out next day on the same preparation which gave the tracings of Fig. 30) , shew the result of single excitations applied to the nerve when the conductivity was on the verge of disappearing. In tracing 13 we see that the single stimuli at 600 are sub-maximal while those at 1000 are higher. Tracing 16 shews the result of a graduated series of single stimuli; at 500 the muscle gives a slight response; the succeeding responses are sub-maximal till at 1000 the/

the highest contraction is reached. Tracings 17 and 18 are further illustrations of the same phenomenon. The conductivity whether for single stimuli or for series of rhythmical stimuli disappears in the same order, namely, first of all for weak and later for strong excitations.

Another method of studying the gradual disappearance of the conductivity is to repeat at regular intervals an excitation (or series of rhythmical excitations) of constant intensity, the temperature being meantime allowed to fall more or less rapidly. Chart 4, tracing is an illustration of this method, a series of rhythmical excitations of intensity, 30 being used to excite the nerve. The muscle responses are seen to gradually change from descending tetani to initial tetani; these diminish regularly in height and finally almost disappear just as freezing of the nerve occurs. [In this case by a coincidence the conductivity has spontaneously disappeared at the freezing point of the nerve.] Corresponding results are obtained when single stimuli are used instead of a series of rapid stimuli.

As the conductivity disappears, therefore, at least two simple changes can be demonstrated as occurring/

occurring simultaneously:-

(1) The height of the muscle contractions is gradually cut down; this speaks for a greater and greater amount of diminution in the excitatory process as it traverses the cooled part of the nerve.

(2) Strong stimuli remain effective longer than weak stimuli. In other words, the effective conductivity does not disappear simultaneously for all strengths of excitation, as one might conclude from the statement of Boruttau and Frohlich[56] (see pp. 50-1); an excitatory process of originally small amplitude gets lost, as it were, in the cooled part at a time when an excitatory process of greater amplitude can still plough its way through.

(2) Return of Conductivity.

As the conductivity returns on raising the temperature it does not immediately come back to its full extent. Provided the change in temperature be brought about slowly it is generally possible to follow out definite stages in the re-establishment of full conduction. Then it is seen that return occurs in the reverse order to that in which conductivity disappeared, viz. the muscle does not respond to weak stimuli applied to the central end of the nerve so soon as to strong stimuli; further, when contractions do begin/

do begin to occur on the application of weak stimuli these are not so pronounced as the contractions following strong stimuli. Such observations may be made by using single break shocks to excite the nerve.

When one uses a series of rhythmical stimuli on the other hand, so as to test for the presence of a Wedensky effect, the result seems to fall out in two different ways, according to circumstances. In the one case typical Wedensky effects may be elicited as the nerve is recovering. Instances of this kind of result we may have already observed in Charts 15 and 16. A further instance is afforded in Chart 18. In other cases, again no Wedensky effect may be elicitable. Charts 19 and 20 are illustrations. In these we see that during the return of conductivity the most complete tetani are got with strong stimulation rather than with weak, in contradistinction to the examples shewn in Charts 15, 16 and 18. To express this result in the language of Fröhlich's interpretation of the Wedensky effect, we should say that in certain cases the refractory period of the nerve during the process of return of conductivity is greater for strong stimuli than for weak; in other cases again it is not so. The question will require further investigation.

That in some cases the re-establishment of full conductivity/

conductivity may take place very rapidly is shewn in Chart 21. Here at 7°C. the nerve does not conduct. Two minutes later at 8.6°C the conductivity has almost fully returned, in that complete tetani are got for all intensities of stimulation with the special rate used (144 per second). A similar speedy return of conductivity almost invariably occurs when the cooled and non-conducting nerve is suddenly warmed by raising it out of the cold chamber. Often after prolonged cooling I have imagined that a nerve was possibly dead: no muscle contractions may have been elicited for a considerable time: on raising the nerve up the immediate change in its condition is most striking: the muscle at once responds with full tetani to rhythmical stimulation of the central end of the nerve.

To the consideration of the state of the nerve during return of conductivity after cooling we shall once more return in next section.

SECTION E. FATIGUE.

In a consideration of fatigue of nerve we must at the outset exactly define what is meant by the expression "fatigue." A mere absence of conductivity in a living nerve is not fatigue. Thus, the conductivity may be temporarily abolished by asphyxia, by narcosis, by freezing, or even by simple low temperature, and yet the nerve is not necessarily fatigued. Before one can definitely say that a tissue is fatigued one must prove that it is either incapable of functioning or thus shews a diminution in functioning capacity, because of previous activity.

(1) Fröhlich's Proof.

The method by which Fröhlich proved fatigue is familiar to us. His argument, as I take it, is as follows. The existence in a Wedensky effect of an "initial tetanus" from strong stimulation is due to a prolongation of the refractory period. As soon as the refractory period for a definite strength of stimulus becomes greater then the interval between the successive stimuli of a rhythmical series of the same definite strength, excitation of the nerve with such a series causes only one twitch. From the fact that this twitch (or intial tetanus" if we care to so call it) corresponds in height to the twitch evoked by one/

one single stimulus, we gather that only the first member of the series of rhythmical stimuli has succeeded in affecting the muscle; the succeeding members of the series have got lost, as it were.

The existence of an "initial tetanus" by itself does not prove fatigue. It is conceivable that a conducting mechanism built on simple physical principles might give the same result. To prove fatigue Fröhlich relies on the "descending tetanus" got with slightly weaker stimulation. In this case, as the result of a series of rhythmical stimuli, the muscle responds at first well, but as the stimulation of the nerve goes on the muscle response becomes less and less pronounced and finally dies away. This would point to a progressive lengthening of the refractory period as the result of activity of the nerve, and consequently to fatigue, were it not that the nerve, even when not excited, can hardly be assumed to be in a stable condition under the action of anaesthesia or of asphyxia as the case may be. For all we know, this prolongation of the refractory period may be due to progressive depression of conductivity arising simply from the anaesthesia or asphyxia.

The fact that on interruption of the rhythmical stimulation even for a short interval the nerve conducts again, does not shew that the previous absence/

absence of conductivity is due to fatigue, unless the second muscle response also takes on the form of a descending tetanus, in other words, unless the refractory period for the same intensity of stimulation can be shewn to have again become shorter during the period of rest. Fröhlich, in his paper, did not give illustrations of such cases and his proof of fatigue is in consequence not absolute. Fatigue is, however, better proved by such a series of curves as was seen in Fig. 30, tracing 4, for here, after each short rest of the nerve the refractory period must have become shorter again. Until the exact relation that Fröhlich claims to hold between initial tetanus and refractory period is rigidly established even this can not be taken as unimpeachable fatigue.

The above criticism may appear superfine. It is not put forward in order to detract in any way from the credit due to Fröhlich for shewing how nerve fatigue may be proved, for by bringing Wedensky's results into relation with the question of fatigue he has thrown a flood of light on the whole subject of nerve conduction. The criticism is made rather to throw into stronger relief the exact bearing of his proof, for in some quarters at least, vague and unspecified objections have been brought against it.

(2) /

(2) Can Fatigue of Nerve be proved in a still more Extended Sense?

When first I began to investigate the Wedensky effect as the result of cooling I was not aware of the slow depression of conductivity that goes on in a cooled nerve apart from activity, and consequently imagined from the tracings obtained that a wide extension might be given to the meaning of fatigue as understood by Fröhlich. It seemed as if a nerve might by successive repetition of a series of rapid excitations be ultimately worn out so that not only the time for recovery at the given low temperature might be very greatly prolonged, but the actual extent of the recovery process might be interfered with, or recovery even entirely prevented; just as a muscle by repeated stimulation may be shewn to undergo a steady deterioration in functioning capacity in spite of intervals of rest between the successive contraction. As far as Fröhlich's presentation of the question goes there is nothing to indicate that recovery is not complete under the given conditions, provided a very short rest be given to the nerve. If one intermits the rhythmical stimulation but for a fraction of a second apparently full recovery occurs.

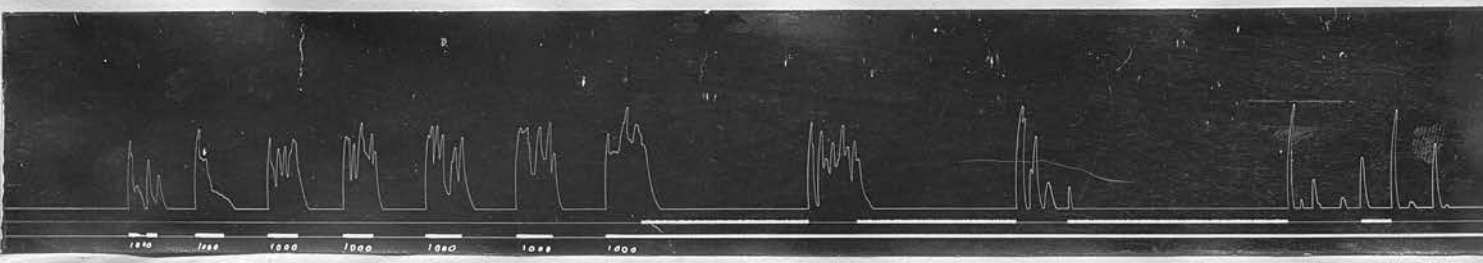
Now on the assumption that at constant low temperature/

temperature the conducting mechanism of the non-stimulated nerve is stable, tracings such as we have seen in Figs. 30 and 31, in Fig 33, or in the series (31-34) could be due to nothing else than a progressive fatigue of the nerve owing to repeated stimulation. But the assumed stability of the nerve at constant low temperature is negatived by experiments in which the conductivity has been found to similarly disappear almost in the absence of stimulation. Evidently therefore, such tracings can not be taken as instances of progressive fatigue.

Another method of investigating the question of fatigue is to continuously stimulate the central end of the cooled nerve, meantime blocking the impulses on their way to the muscle by means of a constant current. If the nerve refuse to conduct on the removal of the block, then it must have been fatigued.

This method was first of all used by Bernstein,(35) and later by Wedensky,(36) and as their results are generally cited as one of the best proofs of the non-fatigability of nerve, it was interesting to see how cooled nerve reacted under the same conditions. I carried out some experiments after the same fashion. Although my object at the time of carrying them out was not to obtain evidence of more or less lasting fatigue but simply to confirm if possible Fröhlich's results, the experiments/

Fig. 40.



experiments are worth recording as tending to support Fröhlich's argument.

For supplying the constant current an accumulator of 4 volts potential was used. By means of a rheocord a current with a potential difference of from 0 to 4 volts could be sent through the nerve at a place peripheral to the cooled area. Non- polarisable electrodes were employed to make contact with the nerve, and the current was arranged in the ascending direction. On testing the efficiency of the block it was found that a potential difference of 2 to 3 volts between the two leading - in electrodes was sufficient to keep the muscle at rest when strong tetanisation of the central end was employed. The effect of a block of this strength was however sufficient to induce polarisation within the nerve of such extent that it did not recover for some seconds afterwards.

On cooling the nerve (the Wedensky condition was established, and when this was present) a strength of stimulation was selected which just gave a descending tetanus. See Fig. 40, (the lower time-marker registers the time of tetanic stimulation, the upper the time of application of the constant current). To prevent any possible fallacy from the slow action of/

of constant temperature the flow of the cold fluid was so arranged that the temperature should slowly ascend during the experiment. Repeated tetanisation with the same strength of stimulus now caused a tetanus which gradually began to increase in height. Previous observation with the constant current had shewn that for these Descending Tetani a considerably lower potential difference than 2 to 3 volts is sufficient to act as a block. Consequently when the tetanus that was being repeated was on the point of becoming a complete tetanus, a block of $1\frac{1}{4}$ volt was put in, and the current kept running for about 8 seconds. On removing the block the tetanus was now found to be lower and more broken. The block was again put in for another 8 seconds: on removing it the tetanus had become a typical descending tetanus. The block was again put in, this time for about 10 seconds, and on removal the muscle reacted only by isolated twitches. (The twitches that occur at the moment of application and of removal of the blocking current are, of course, not due to central stimulation). During the whole of this time the nerve was being continuously stimulated at the central end, and the explanation of the result might be that owing to the uninterrupted excitation the refractory period has become/

become progressively longer and longer. Whether the "fatigue" so induced is more or less lasting I have not as yet investigated by this method.

In such an experiment as this, however, there is always the risk of injurious polarisation of the nerve by means of the constant current, so that we cannot look on the result as incontrovertible proof of fatigue. One thing in the above experiment would indicate that the fall in height of the tetanus is not due to polarisation, namely, the fact that the muscle response after the second application of the block takes the form of a descending tetanus. Had the nerve been polarised recovery of conduction would have at once set in on removal of the block, and the tetanus, as control experiments have shewn, would have begun to take an ascending form. The amount of polarisation depends, apart from the resistance of the nerve, on the potential difference between the leading-in electrodes, and in this case the difference was low. Exactly similar results to that shewn in Fig. 40 have been got in other cases where I used a blocking current with low potential difference.

Wedensky, who in his experiments used a blocking current with a potential difference of 2 volts, improved on Bernstein's method by regularly changing the direction/

direction of the constant current, and thus minimising the effects of polarisation. Although he does not specifically mention the length of time that he allowed the current to run in one definite direction, one gathers that it must have been for some minutes and not merely for a few seconds, otherwise the muscle would soon have become fatigued. The method of blocking by means of a constant current requires to be very carefully used to give reliable results. As yet I have done no experiments with blocking agents other than the constant current.

In order to find evidences of fatigue we are not however limited to the afore-mentioned methods, and in the cases subsequently to be described we shall see that there is considerable evidence of a breaking down process in the nerve which is only tardily recovered from, the process of recovery being spread over many seconds if not minutes.

In Chart 22 we have a record taken from a preparation, in which, after the Wedensky effect had been elicited at a low temperature, the nerve was allowed to slowly rise in temperature. The successive tracings taken with higher and higher temperature shew that conductivity is gradually restored. The method of return in this case is one in which

(to/

(to use Fröhlich's explanation of the phenomena) the refractory period for weak excitations is longer than for strong excitations (see p.p. 45-6). Eventually in tracing 16 an almost complete tetanus is obtained with an intensity of 1000. The nerve, however, as the next tracing shews, has not completely recovered its conductivity, for continucus stimulation at 1000 produces a very marked descending tetanus, which ultimately is resolved into irregular and weak twitches. A rest of a second or so suffices to improve the conductivity slightly but the improvement is only momentary. A slightly longer rest (2 or 3 seconds) causes a better improvement. Increasingly longer rests between the intervals of stimulation cause progressively increasing improvement in conduction. This is a case where we can watch not only the process of fatigue occurring in the nerve, but also the more or less tardy recovery from fatigue. During the whole process the temperature was continually rising.

The same tracing, however, shews a further phenomenon, which may ultimately prove to be one of the best methods of showing fatigue. At a certain moment peripheral stimulation of the nerve at an intensity of 1000 was carried out. Thereafter central/

central stimulation at 1000 is seen to be ineffective. Only after a rest of half a minute is the nerve able to conduct again and then the conductivity is very imperfect. A second peripheral stimulation of the nerve once more abolishes the effective conductivity completely.

To understand this phenomenon it is necessary to recall what was said in the Section dealing with Disappearance of Conductivity. The conductivity of cooled nerve when on the point of disappearance goes away in a definite order. In a certain stage strong excitations can still plough their way through the affected portion of the nerve, while weak excitations are stopped on the way; an excitation of intermediate strength is so cut down on passing through the cooled portion that on emerging at the other end it is submaximal in effectiveness. If we imagine for a moment what happens when a single excitatory process entering a cooled portion of the nerve is only able to emerge from it cut down to a fraction of its original effectiveness, we are practically justified in concluding that the molecular commotion in each minute length of the cooled portion declines and declines in extent the further the excitatory process penetrates towards the peripheral end. Associated with this diminished disturbance/

disturbance of the more peripheral portions of the cooled part is a more complete recovery than in the centrally lying parts of the cooled area. If however, after having rhythmically stimulated the nerve from the central end and partially fatigued it, we can now reverse the direction of stimulation and break it down from the peripheral side as well, the conductivity may be destroyed entirely. This is a probable explanation of what has happened in the above experiment.

When the nerve is in a condition of depressed conductivity strong rhythmical stimulation by means of the peripheral electrodes so affects the peripheral parts of the nerve that subsequent stimulation at the central end is ineffective in bringing about contraction of the muscle. So marked in fact is this action that in conducting experiments on the conductivity of cooled nerve by means of rhythmical stimulation one must never apply peripheral stimulation to test whether the muscle has become fatigued. The result is invariably a more or less prolonged abolition of the conductivity of the nerve to central stimulation and a consequent interruption of the experiment. Numberless tracings might be shewn all illustrating the statement.

To/

To the explanation above given two objections might be advanced. In the one case it might be said that the apparent abolition of conductivity after peripheral stimulation of the nerve is due to a diminished excitability of the muscle through over-much stimulation. Control experiments with stimulation of the muscle at very varying intensities have shewn that this explanation does not account for the phenomenon. A more serious objection is that the result is due to polarisation of the peripheral part of the nerve. As yet I have done no reliable control experiments to exclude this objection, but the length of time that the conductivity remains abolished is an argument against polarisation. It is only in a few cases that I have seen the conductivity again return after strong peripheral stimulation and then when the temperature was rising. If the preparation again be immediately raised out of the cold chamber after the conductivity has been abolished by peripheral stimulation the nerve almost immediately regains its conductivity.

A continuation of the experiment of which Chart 22 is a record is seen in Chart 23 This record was commenced a few minutes after the inscription of tracing 17 in Chart 1, and we see that/

that at a temperature of 8.2°C the conductivity has again returned though in an imperfect fashion. Typical "fatigue" curves are obtained and more readily now with a low intensity of stimulation than with a high. The last tracing on the chart shews the sudden return of more or less complete conductivity on raising the nerve out of the cold chamber.

Still further instances of very tardy recovery after continuous rhythmical stimulation may be seen in other cases. Thus in Fig. 34, tracing 20, we see that after a rest of ½ to 1 minute the nerve is in a better conducting condition than when tracing 19 was inscribed. On a second repetition of the stimulation in tracing 20 no muscle response occurs, while after a short rest of ½ to 1 minute (tracing 21), the nerve can once again conduct. How far the prolonged absence of conductivity in a case such as is shewn in Chart 12 tracings 15 - 28, is due to mere temperature conditions, and how far it is due to possible fatigue owing to recurrent stimulation, is hard to say. Further experiments are necessary to clear up the relation between absence of conductivity due to fatigue and absence of conductivity due to simple temperature conditions.

To/

To sum up the results of the section, we might say that while the fatigue which Fröhlich shewed is in high degree probable, there is reason to believe that fatigue may occur in a more extended sense than he at first imagined. We have seen evidence for believing that the process of recovery may be delayed to a great extent if not entirely prevented.

Section F. Conclusion.

The results of the foregoing research on nerve conduction have been arrived at mainly by taking advantage of two distinct principles. The one is that of applying not simply single maximal stimuli but series of rhythmical stimuli of all degrees of intensity to the nerve. The other is that of altering the conducting power of the nerve by subjecting it to a depressing environment The combined application of these two methods, the systematic introduction of which we owe to Wedensky and Fröhlich, had rendered it possible to make a more complete examination of the processes involved in nerve conduction than had previously been the case.

The present research confirms the results of these two investigators, and establishes the fact that cold has an influence on frog's nerve closely similar to that of anaesthesia and of asphyxia.

The/

The peculiar condition of the nerve associated with the phenomena to which for shortness, I have throughout referred as the "Wedensky effect," has been more or less carefully studied. Wedensky's claim that these phenomena can be relied on as indicating depression of the conductivity long before the actual stage of non-conduction is reached has been consistently verified throughout.

Special attention has been devoted to the exact mode of disappearance of the conductivity. It has been found that under the influence of cold the effective conductivity disappears sooner for weak excitations than for strong. Whether this is a contradiction of the principle enunciated by Boruttau and Fröhlich, whereby the amount of decrement in the excitatory process, when the nerve is acted on by a depressing agent, is proportional to the wave of excitation, cannot be said offhand.

The special action of low temperature on nerve has been more systematically studied than before. Different nerves have been found to vary considerably in the degree to which their properties are affected by any definite low temperature. Taking as a special case the temperature at which the conductivity of a definite length of nerve just becomes abolished, it has been found that this varies throughout a

considerable/

considerable range. Whereas in some cases complete abolition of conductivity has been observed at + 4°C, in other cases again the indications are that the corresponding temperature lies considerably below the freezing point of the nerve. As to the conditions responsible for this variation in different nerves no definite results have been obtained.

Again, any given nerve kept constantly at one definite low temperature cannot be regarded as necessarily in a stable condition as regards conducting power. When a nerve is kept at a fixed low temperature slow changes would appear to go on inside it in such a way as to tend towards abolition of conductivity. Further, when once conductivity is abolished at any definite temperature it does not return at once on slowly warming the nerve. In some cases the temperature may be raised through at least 10 degrees before conductivity reappears.

All this goes to shew that the influence of temperature on nerve is complicated. There has been a tendency to assume that nerve reacts to change of temperature much as a piece of inorganic material, say, a wire, does, and that what might be called the "physiological constants" of the structure are definite/

definite for a definite temperature. We are not entitled to assume that the "constants" of nerve - rate of transmission of impulse, refractory period, etc. - are fixed when once the temperature is fixed. The analogy with a physical structure such as a wire holds in so far as the action of low temperature consistently tends in one direction, and we might even illustrate the variations in nerves from different frogs by saying that they correspond to variations arising from different proportions of alloy, for example in a wire. In the case however of the slow action of constant low temperature on nerve, and the response of a cooled nerve to a subsequent rise of temperature, the simple physical analogy breaks down.

Again, nerve is very susceptible to the influence of physiological saline solution. "Kept" nerve reacts to low temperature in a different way from fresh nerve.

If we might assume, as Fröhlich holds to be established, that the refractory period of nerve can be as well determined by the application of a series of stimuli, as by two successive stimuli, then we might say that generally speaking the duration of the refractory period varies in the same direction as the intensity/

intensity of the excitation, ~~viz.~~ i.e. the refractory period is greater, the greater the stimulus applied to the nerve. In some cases, however, this principle does not seem to hold, for during recovery of conductivity with rise of temperature, the refractory period for excitations of moderate magnitude is often greater than that for strong excitations.

Nerve can apparently be readily fatigued when at a low temperature, and under special conditions the effect of fatigue seems to last for a number of seconds.

If we compare the results of the present investigation, where all the nerve processes were slowed by means of low temperature, with other experiments carried out at room temperature but with very rapid rates of stimulation, there is ground for supposing that more or less lasting fatigue of nerve may possibly be brought about by extremely rapid stimulation. Some experiments which Kronecker carried out using very rapid rates of stimulation are thus referred to by Gotch (66); "The production of a note indicating 20,000 vibrations per second was effectual for the excitation of the nerve, the muscular response being at first a tetanus, but on repetition it became an /

an initial twitch, and subsequently failed." This looks almost as if Kronecker had actually fatigued the nerve. Again, the more recent researches of d'Arsonval, who, ~~by using~~ used rates of interruption of a different order of magnitude from those previously possible, shew that nerve anaesthesia may be brought about simply by using a high enough rate of stimulation. Is this fatigue?

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