612.817

THE SHERRINGTON PHENOMENON.

By EDITH BÜLBRING and J. H. BURN.

(*From the Pharmacological Laboratory, the College of the Pharmaceutical Society, London.*)

(*Received August* 9, 1935.)

The observation of contractures in denervated muscle after nerve stimulation was first made in 1863 by Philippeaux and Vulpian; they found that when the hypoglossal nerve was divided and allowed to degenerate, stimulation of the lingual nerve, which normally has no motor effect, caused contraction of the tongue. In 1894 Sherrington observed a similar phenomenon in the muscles of the leg of the cat; he divided the 6th, 7th, 8th and 9th post-thoracic ventral and dorsal roots central to the ganglia; thereupon the motor fibres to the muscles of the hindleg degenerated, but the sensory fibres, being still connected with their ganglia, remained. After an interval of 2–6 weeks, the application of very strong stimuli to the sciatic nerve caused contracture of the muscles of the hindleg. Since Sherrington was unable to obtain this contracture after cutting the roots distal to the ganglia, so that sensory as well as motor fibres degenerated, he concluded that the fibres producing the contracture were sensory.

Much work has since been done on this phenomenon, of which a full discussion is given in the review by Gasser [1930]. It was discovered that the denervated muscle becomes sensitive to various chemical substances which produce no contracture when the normal motor innervation is present, the most powerful of these being acetylcholine. Dale and Gasser [1926] showed that the common property of these substances was a nicotine-like action. The suggestion was made by Hinsey and Gasser [1928] and Dale and Dudley [1929] that the effect of the stimulation of the sensory nerves might be due to the liberation of a chemical substance at the vaso-dilator terminations, which would cause a contracture in the muscle sensitized by degeneration of the

motor fibres. Dale and Dudley indicated acetylcholine as the most likely substance. The experiments of Hinsey and Gasser [1930] gave strong support to the view that the contracture depended on the vaso-dilator fibres in the posterior roots. They found that the appearance of dilatation in the cat's leg when the roots were stimulated coincided with the appearance of a particular wave in the record of the action potential. They repeated the experiment on the Sherrington phenomenon and found that here too the appearance of contracture was exactly coincident with that of the same wave in the action potential. Dale and Gaddum [1930] obtained further evidence in favour of acetylcholine as the humoral transmitter and concluded that "the vaso-dilator effects of...sensory fibres stimulated antidromically and the contractures of denervated muscles accompanying these actions are due to the peripheral liberation of acetylcholine".

There remained, however, some doubt whether antidromic impulses passing down sensory fibres were responsible for the phenomenon. After section of the roots central to the ganglia, there are in the sciatic not only sensory but also sympathetic fibres in a normal condition. It is true that Sherrington [1894] failed to observe the phenomenon after excision of the posterior root ganglia, and it is also true that van Rijnberk [1918] said that he was still able to observe it after removal of the sympathetic chain. Nevertheless, Hinsey and Cutting [1933] found that direct stimulation of the posterior roots failed to cause a contracture; they found also that they were able to obtain the contracture by stimulation of the sciatic after degeneration of sensory as well as of motor fibres. It remained for them to obtain the phenomenon by stimulation of the sympathetic chain; this, however, they were unable to do, though in one cat they observed it on stimulation of the grey rami.

We have recently published evidence [1935] that the sympathetic supply of the blood vessels in the muscles of the dog contains many cholinergic vaso-dilator fibres; in the cat we could not be certain that such fibres were present, though in occasional experiments we obtained results which suggested that they might be. Now, Hinsey and Cutting, like most previous workers, used the cat, and it seemed to us that the dog would be more likely to give a clear result if the Sherrington contracture was actually due to the liberation of acetylcholine from vaso-dilator fibres in the sympathetic. We have, therefore, investigated the phenomenon first in the dog and also in the cat.

METHODS.

(*a*) *Operative procedure.*

Dogs have been prepared by section of the 5th, 6th and 7th lumbar and 1st and 2nd sacral roots on the left side. In the first dog simple section of the roots central to the ganglia was performed; in the remaining dogs the posterior root ganglia were also excised. In cats the same roots were cut, but the ganglia were not removed; the removal of the ganglia causes much more bleeding than in the dog, and since we found that the phenomenon occurred after removal of the ganglia in the dog, we did not attempt to remove them in the cat. After the injection of atropine the animal was anæsthetized with a mixture of chloroform and ether, and "Pernocton" (sodium butyl-β-bromallylbarbiturate 10 p.c.) was then given intravenously. The usual dose for a dog weighing 10–12 kg. was 2 c.c. given slowly to ensure that the respiration was not arrested. The dose for a cat weighing 3 kg. was about 0·8 c.c. In dogs the injection was made into the ear vein and in cats into the saphena vein at the ankle. Thereafter very little ether was necessary to maintain anæsthesia. We wish to draw attention to the value of Pernocton for operations. The blood-pressure remains low so that bleeding is minimized; after the operation the animal sleeps for a period varying from 8 to 12 hours so that the wound is not disturbed. During the operations we used cotton-wool swabs sterilized by boiling in saline containing 1 in 3000 acriflavine, and before closing the wound it was washed with 1 in 1000 acriflavine. In the cats all the wounds healed without further attention; in the dogs there was a collection of fluid under the skin for the first few days, but this was usually serous and non-purulent.

The animals were kept for periods varying from 2 to 3 weeks before the final experiment was performed.

(*b*) *Final experiment.*

The observations were again made under Pernocton anæsthesia; in one experiment ether was used at first, but since the tensions observed were poorer than usual, Pernocton was injected and ether was discontinued; we then obtained better results. The animal was eviscerated and the vessels in the peritoneum covering the posterior wall of the abdomen were carefully divided between ligatures, to permit easy access to the sympathetic chain. Usually the left kidney was also removed. The chain was not dissected at this stage. The left gastrocnemius muscle

was then prepared so that changes of tension occurring in it could be recorded on a drum, the details of the preparation being in all respects similar to those described by Dale and Gasser [1926]. We began our observations with the use of a spring tension lever of the pattern described by Hill and Hartree [1920], but came to the conclusion that the tensions developed were too small to be properly recorded by it. We changed to a tension lever of the same type, but fitted with a weaker spring, and used it throughout the experiments on dogs. Even this was too strong for the experiments on cats; for these we used an ordinary lever, the pull of the muscle being exerted against a spiral spring. The levers certainly allowed some shortening, but they enabled us to observe changes of tension which otherwise would have been missed.

The sympathetic chain was stimulated by induced break shocks obtained by the use of Lewis's rotating contact breaker, with a frequency of 32 per second. The chain was divided opposite the lower pole of the kidney, and a portion of the lower end was freed so that stimuli could be applied to the part opposite the 3rd and 4th lumbar vertebræ.

Experimental results.

(*a*) *Observations in the dog.*

The effect of stimulating the sympathetic chain has been to produce a contracture of the gastrocnemius muscle in all the experiments on dogs. The record in Fig. 1 was obtained in a dog in which the roots were cut

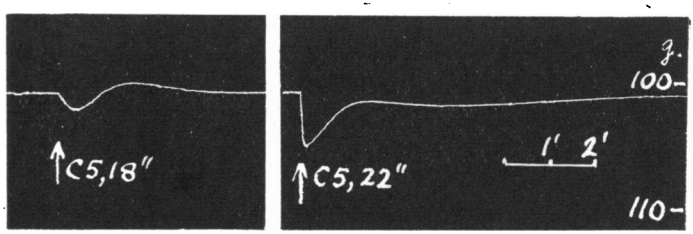

Fig. 1. Record of tension in denervated gastrocnemius of dog, showing the contracture produced by stimulation of the lumbar sympathetic chain (*a*) before and (*b*) after the injection of eserine. Both the motor and sensory fibres to the muscle were degenerated.

and ganglia were excised 18 days previously. Before stimulation the muscle tension was 101 g., and this increased during stimulation to about 103 g. Eserine was then injected in a dose of 1·0 mg., and 10 min. later the same stimulus was repeated, though it was applied for a slightly

longer time. The contraction recorded on the drum was then greater, indicating an increase in tension of about 5 g. The rise of tension before

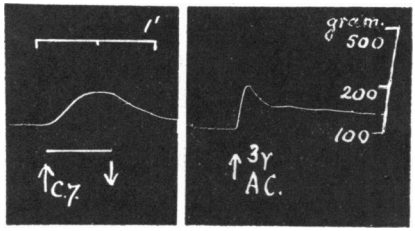

Fig. 2. Stimulation of the sympathetic chain, applied after the injection of eserine, caused a rise of tension of 70 g. Roots cut 22 days previously but ganglia not excised.

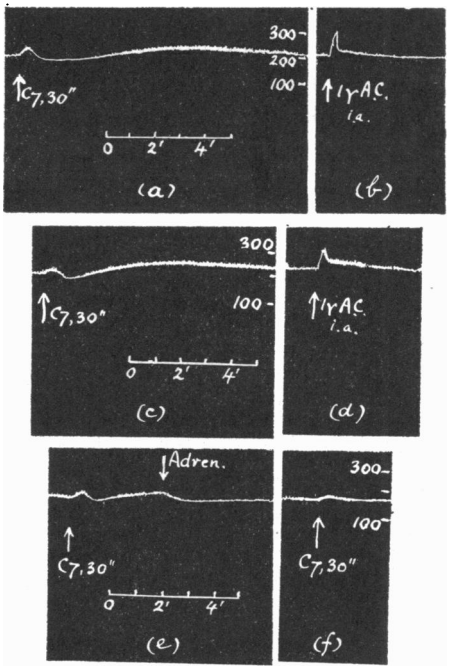

Fig. 3. Showing the double form of the contracture when the chain is stimulated after eserine. In (*a*) and (*c*) there is an initial rise, a fall and a second prolonged rise. Between (*b*) and (*c*) more eserine was injected, and the second rise in (*c*) followed the initial rise sooner than in (*a*). Note that in (*e*) intravenous infusion of adrenaline causes a fall of tension, and while this infusion continued stimulation was ineffective (*f*).

the injection of eserine was followed by a fall below the original level; the rise after eserine was also followed by a fall, but the return to the base line did not occur for about 10 min. A greater effect is shown in Fig. 2

which is a record obtained with a Hill and Hartree lever; in this dog the ganglia were not excised, but the roots were cut 22 days previously. Before eserine was injected the effect of stimulating the sympathetic chain was too small to be reproduced; after the injection of eserine, stimulation produced a rise of tension of about 70 g. as shown in the figure. In the absence of eserine the greatest tension we have recorded has been 40 g. (Fig. 10 (*a*)).

The effect of stimulating the chain was frequently observed to be more than a single rise of tension; there was an initial rise, subsiding in about 1 min., followed by a second slow and prolonged rise which often remained for 10 or 12 min.; this curious double effect is well shown in Fig. 3. For this experiment the roots were cut and the ganglia were excised 16 days previously, but the 2nd sacral ganglion was not removed.

In Fig. 3 (*a*), taken after the injection of 1 mg. eserine, there was an initial rise of tension which subsided in 1 min., and then a second maximum about 7 min. after the stimulation. Further injections of 6 mg. eserine and 5 mg. atropine were made. On repetition of the stimulation the second rise of tension (see Fig. 3 (*c*)) occurred earlier and the maximum was an increase of 50 g. above the initial tension. The stimulation was also applied in Fig. 3 (*e*) and, during the second rise of tension, a slow infusion of adrenaline (1 in 50,000) was begun into the jugular vein. The tension at once relaxed. A fresh stimulation applied during the infusion of adrenaline failed to produce an appreciable effect (Fig. 3 (*f*)).

(*b*) *Observations in the cat.*

In experiments on cats made with a tension lever, we failed to observe an effect when the sympathetic chain was stimulated; when the muscle was arranged to pull against a spiral spring, the initial tension varying from 20 to 100 g., stimulation of the chain produced a contraction represented by an increase in the tension of about 2 g. The greatest effect is recorded in Fig. 4, and this was obtained without previous injection of eserine. Fig. 5 shows a comparison between the effect of stimulating the sympathetic chain and stimulating the sciatic nerve. In (*b*) a stronger stimulus (coil 3, 8 sec.) was applied to the sciatic than to the sympathetic in (*a*) (coil 5, 4 sec.); but thereafter the stimuli were the same. Sciatic stimulation was

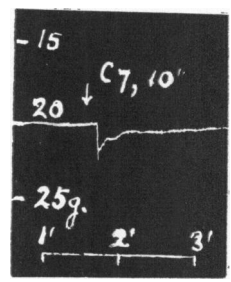

Fig. 4. Contracture of the denervated gastrocnemius of the cat when the sympathetic chain was stimulated. No eserine injected.

throughout more effective than stimulation of the chain, the effect of which in (*c*) and (*e*) was hardly perceptible. Since the posterior root ganglia had not been excised, the stimulation of the sciatic was not only a stimulation of sympathetic, but also of sensory fibres, and the difference in the effect may have been due to this; certainly in one dog

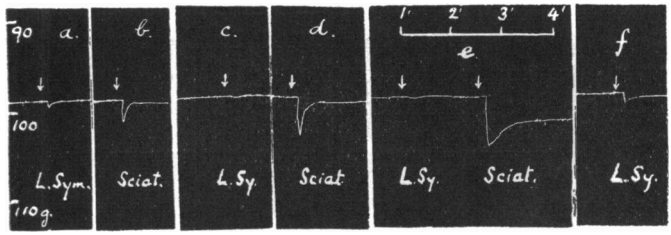

Fig. 5. Showing the greater contracture obtained in the cat when the sciatic is stimulated than when the chain is stimulated. (*a*), (*b*), (*c*) and (*d*) before eserine. (*e*) after 0·3 mg., and (*f*) after 1 mg. eserine. Roots cut 15 days previously.

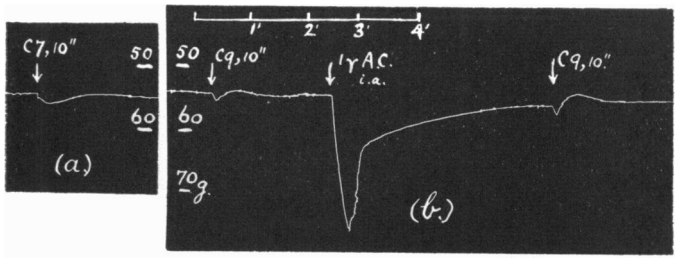

Fig. 6. Contractures in the cat when the chain was stimulated. 0·05 mg. eserine injected before the records were taken. Compare the effect of the second stimulation in (*b*) with that of the first, noting the inhibition of tension after the rise. Roots cut 17 days previously.

in which the ganglia were excised and in which the effects of stimulating the chain and the sciatic were compared, the effects were the same. The effects of stimulating the sciatic in Fig. 5 (*e*) was greater because of the previous injection of 0·3 mg. eserine, and the effect of stimulating the sympathetic in (*f*) was observed after a further dose of 0·7 mg. eserine.

Further examples of the effect in another cat are given in Fig. 6 (*a*) and (*b*); eserine was previously injected. The second stimulation in (*b*) was applied shortly after the intra-arterial injection of 1γ acetylcholine at a time when the tension due to this injection had not completely disappeared; the record shows an increase of tension due to the stimulation,

quickly followed by a fall below the initial level. Since this fall was greater than the fall observed after the previous stimulation, it may indicate a second inhibitory phase of the effect of stimulation upon the tension, which will be discussed later.

(*c*) *Contracture caused by adrenaline.*

In the course of our experiments we have observed that adrenaline produces a contracture in the denervated muscle of cats and dogs. We were anxious to test the effect of adrenaline on the Sherrington phenomenon when produced in the dog by stimulation of the chain after

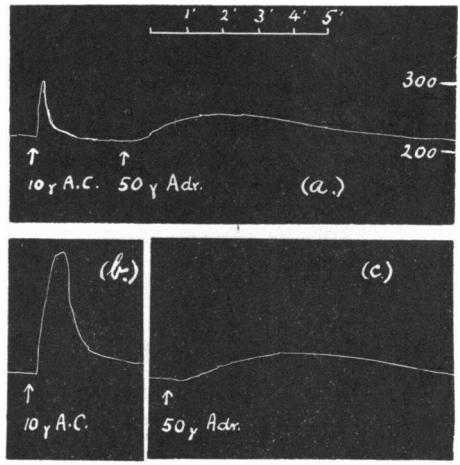

Fig. 7. Contractures in the dog due to acetylcholine and to adrenaline. Ergotoxine previously injected. Between (*a*) and (*b*) eserine was injected; the effect of acetylcholine was augmented (*b*), but that of adrenaline was unaffected (*c*).

a full dose of ergotoxine had been given to exclude the vaso-constrictor action. The injection of adrenaline itself, however, whether intra-arterial or intravenous, was found to produce a contracture. Fig. 7 illustrates the effect in a dog after section of the sciatic nerve 9 days previously. Under Pernocton anæsthesia, 10 mg. of ergotoxine ethanesulphonate was injected, after which doses of 50 and 100γ adrenaline caused a large fall of blood-pressure. The record shows the relative effects of 10γ acetylcholine and 50γ adrenaline, (*a*) before, (*b*) and (*c*) after, the injection of eserine. In contrast to the short contraction produced by acetylcholine, a long contraction, slow in onset, was produced by adrenaline. This contraction was unaffected by 1·6 mg. eserine which greatly magnified the effect of acetylcholine.

The adrenaline contracture is not only seen after the injection of ergotoxine; provided the doses are not too large it may be observed in spite of the vaso-constrictor effect. Fig. 8 (*a*) illustrates the effect in a cat, and (*b*) the effect in a dog in which the respective sciatic nerves were cut 24 and 11 days previously. During the first minute after the intra-arterial injection of 10γ adrenaline the tension remained stationary or fell; it then rose slowly, the rise being interrupted in Fig. 8 (*a*) so that two

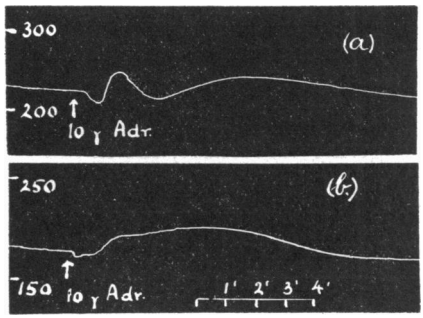

Fig. 8. Contractures in (*a*) the cat, (*b*) the dog due to the injection of adrenaline. No ergotoxine was given.

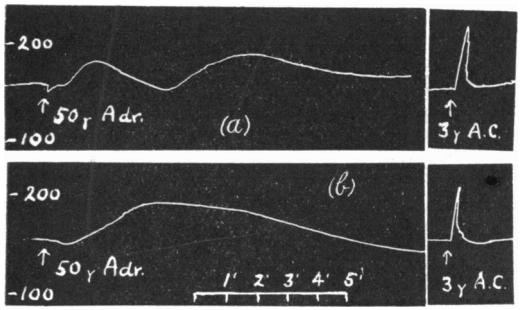

Fig. 9. Contractures in the dog due to adrenaline, (*a*) before ergotoxine, (*b*) after ergotoxine. Note that ergotoxine does not affect the action of acetylcholine.

peaks of tension were recorded. An explanation of the two peaks is obtained when the same dose of adrenaline is injected before and after ergotoxine. Fig. 9 (*a*) records the double rise of tension produced in the gastrocnemius of a dog before the injection of 12 mg. ergotoxine, and (*b*) shows the single rise produced by the same dose afterwards. Evidently the double rise represents the algebraic sum of two opposed effects; the one, an increase in tension seen in its uncomplicated form in Fig. 9 (*b*),

and the other a diminution in tension due to an inhibitor effect which must have reached its maximum in Fig. 9 (*a*) about 4 min. after the injection. The response to 3γ acetylcholine was unaffected by ergotoxine.

The occurrence of the adrenaline contracture depends on the animal and on the dose; an animal which gives a contracture after 10γ may fail to do so after 50γ and, instead, a fall in tension is recorded. In some animals no contracture is observed with any dose before the administration of ergotoxine, and even after ergotoxine the contracture does not always occur at once; in one experiment the injection of 50γ adrenaline, made after giving ergotoxine, caused a rise in the tension of the cat's gastrocnemius of 250 g., but the rise did not occur until 4 min. after the adrenaline was injected. When acetylcholine and adrenaline are injected simultaneously after giving ergotoxine, the initial rise of tension is the same as that produced by the injection of the acetylcholine alone; the tension falls rapidly to the maximum produced by the adrenaline alone, and then follows the course taken by the tension due to the adrenaline.

We have made one experiment to see if adrenaline caused a contracture of the denervated diaphragm of a kitten when suspended in a bath as described by Dale and Gaddum [1930]. The right phrenic nerve was divided and a piece about 3 cm. long was removed. A fortnight later the kitten was killed, and a strip of the diaphragm on the denervated side was excised, and suspended in Ringer's solution. This muscle contracted when acetylcholine was added to the bath, and the contraction produced by a small dose was augmented when adrenaline was previously added to the bath. Adrenaline itself, however, had no contractile effect, either before or after the addition of ergotoxine.

(*d*) *The Sherrington phenomenon after ergotoxine.*

Some experiments were performed to observe the contracture produced by stimulating the sympathetic chain after giving ergotoxine. The record in Fig. 10 is taken from one of these in a dog. At first strong stimulation of the chain gave the response shown in (*a*); a total of 22 mg. ergotoxine was then injected to ensure complete paralysis of the vasoconstrictor effect of adrenaline. As we have previously pointed out [Bülbring and Burn, 1935], larger amounts are often necessary in the dog than the cat, but as may be seen in the remaining parts of the figure, the ergotoxine produced muscular twitching. Fig. 10 (*b*) records the response to the intra-arterial injection of 100γ adrenaline, and (*c*) the repetition of the stimulus applied to the sympathetic chain. Both the injection of adrenaline and the stimulation of the chain produced a fall

of blood-pressure. The stimulation after ergotoxine produced an initial rise of tension similar to that seen before; this rise was followed by a fall below the initial level and then by a second prolonged rise which was

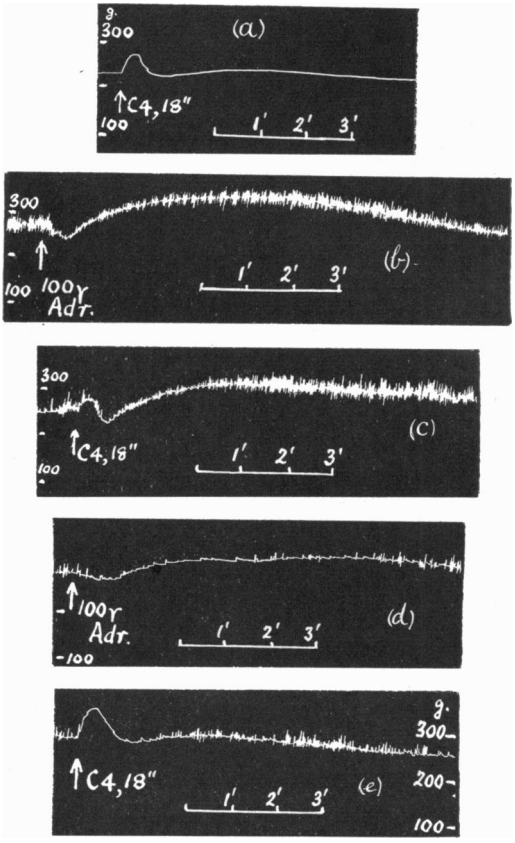

Fig. 10. Showing effect of ergotoxine on the contracture produced by stimulating the sympathetic chain in the dog. Note in (*a*) the initial rise of 40 g. and the small second rise. (*b*) shows the effect of adrenaline after ergotoxine. (*c*) shows the effect of stimulation after ergotoxine; the initial rise seen in (*a*) remains, but is followed by a fall and a second rise resembling that produced by adrenaline in (*b*). (*d*) and (*e*) are records after the injection of eserine which does not increase the adrenaline contraction or the second rise after stimulation. Eserine does increase the first rise after stimulation.

almost absent before the administration of ergotoxine; this second rise was indeed very like that produced by the injection of adrenaline. The records (*d*) and (*e*) were taken after the injection of 1 mg. eserine; the initial effect of stimulation (*e*) was thereby augmented but the adrenaline

contraction and the second prolonged rise after stimulation were less than before. We are of opinion that the lessening of these effects was not a specific effect of eserine, for in other experiments (see Fig. 7) eserine did not diminish the adrenaline response. Since, however, the initial rise of tension was augmented by eserine while the second prolonged rise was not, and because of the parallelism between the second rise and the rise produced by adrenaline, we think that this second rise is more likely to be due to adrenaline liberated by the sympathetic stimulation than to acetylcholine.

Discussion.

The experiments described leave no room for doubt whether the Sherrington phenomenon can be produced by stimulation of the lumbar sympathetic chain; they show that it is readily so produced in dogs, and that small contractures are also obtained in cats. These findings agree very well with the results of our previous experiments [Bülbring and Burn, 1935] which showed that cholinergic dilator fibres are easily demonstrated in the sympathetic chain of the dog, but that proof of the existence of similar fibres in the cat is difficult to obtain. The chain of evidence seems now complete that the Sherrington phenomenon occurs, in part at least, because of the liberation of acetylcholine from sympathetic vaso-dilator fibres; the liberated acetylcholine then diffuses into the muscle substance, which, being denervated, responds to the presence of the acetylcholine by contracture.

Hinsey and Cutting were unable to obtain a contracture by stimulating the chain in cats, though in one cat they were successful when they stimulated the grey rami; the tensions we have observed in cats have been so low as 1–2 g., and it may be that the recording system used by Hinsey and Cutting was insensitive to these tiny effects.

We have been very puzzled by the smallness of the tensions we have observed, since Hinsey and Gasser [1928] record tensions as high as 900 g. in the cat's gastrocnemius after stimulating the sciatic nerve; these were observed after root section without excision of the ganglia. It may be that our experiments furnish a clue to the difference.

When the sympathetic chain is stimulated, not only is acetylcholine liberated from whatever cholinergic dilator fibres are present, but adrenaline (or sympathin) is liberated from the constrictor fibres and from adrenergic dilator fibres. Now Hinsey and Gasser [1928] showed that the Sherrington phenomenon was suppressed by circulating adrenaline; our own experiments illustrate this observation (see Fig. 3 (*e*)). In 1930 Dale and Gaddum showed that the contracture produced by acetyl-

choline was abolished when the sympathetic chain was stimulated. These results make it reasonably certain that when a contracture is observed to follow the stimulation of the chain, the initial increase of tension is only the remnant which the adrenaline (or sympathin) liberated is unable to inhibit. There is evidence of the inhibitory effect in different experiments; thus in Fig. 1 (*a*), after the preliminary rise of tension there was a fall below the initial level. This may be interpreted as an inhibition of tension due to adrenaline, or to the natural course of the relaxation; in another experiment, however, the size of the fall was seen to depend on the tension existing in the muscle before the stimulation was applied. Thus, in Fig. 6 (*b*), the same stimulus was applied before and also after the injection of 1γ acetylcholine; the second stimulus, applied when part of the tension due to the acetylcholine was still present, gave a rise like that given by the first, but the following relaxation was much greater. This relaxation, we think, must be regarded as an active inhibition of tension.

Further evidence of the inhibitory effect is provided by the contracture observed in the dog after the injection of eserine. The records in Fig. 3 show that the contracture has two phases, an initial rise which has disappeared in 1 min., and a later prolonged rise. This twofold rise of tension is explained by supposing that after the first rise due to the liberation of acetylcholine, the inhibitory effect of adrenaline is exerted and the tension falls; when the inhibitory effect passes off, the acetylcholine, preserved by the presence of eserine, once more causes a contracture.

Evidence that the contracture following stimulation of the chain is the resultant of the opposed effects of acetylcholine and adrenaline does not, however, explain the greater effect obtained when the sciatic nerve is stimulated. Hinsey and Cutting [1933] failed to obtain any effect by stimulating the chain, but regularly obtained the contracture by sciatic stimulation when both motor and sensory fibres had degenerated. More than one factor may be responsible. Stimulation of the chain at one point may stimulate only a few of the sympathetic fibres leading to the gastrocnemius, while stimulation of the sciatic may affect all of them; if this is so, the amount of acetylcholine liberated in the neighbourhood of the muscle will be greater when the sciatic is stimulated. In the second place stimulation of the chain will cause the release of adrenaline (or sympathin) from vaso-constrictor endings in all arteries peripheral to the bifurcation of the aorta, whereas stimulation of the sciatic will affect far fewer vaso-constrictor endings. We may summarize the difference between stimulation of the sciatic and of the chain by saying that sciatic stimulation will

probably yield more acetylcholine and also less adrenaline than stimulation of the chain; consequently the tension developed will be much greater after sciatic stimulation.

Dale and Gaddum [1930] investigated the inhibitory action of adrenaline on the response of denervated muscle to acetylcholine and concluded that it was not due to vaso-constriction; they suggested that adrenaline reduced the permeability of the capillary walls to acetylcholine. They showed further that adrenaline had an adjuvant effect which followed the initial inhibitor effect, and which could be demonstrated *in vitro* on a strip of denervated diaphragm. We have now found that adrenaline produces a contracture in the denervated gastrocnemius of both cat and dog, similar to that observed by v. Euler and Gaddum [1931] in the denervated facial muscles. When adrenaline is injected after full doses of ergotoxine have been given, the contracture usually appears without a latent period as a slow and prolonged rise of tension; in the absence of ergotoxine the contracture may not appear at all or there may be an alternation of rise and fall of tension. Evidently adrenaline can inhibit its own effect in causing contracture just as it can inhibit that of acetylcholine and, since part of this inhibitor effect is removed by ergotoxine, that part appears to be due to vaso-constriction. Sometimes, however, after ergotoxine, the rise in tension appears after an interval; this latent period may be caused by the reduced permeability of the capillary walls, suggested by Dale and Gaddum.

We may now attempt to analyse the actual form of the Sherrington phenomenon produced in the dog by stimulating the sympathetic chain; hitherto it has been looked upon as a single contraction, but our results indicate that it is more complex. There are three conditions in which the response should be considered: in the absence of drugs, in the presence of eserine and in the presence of ergotoxine; to explain the responses it is to be remembered that both acetylcholine and adrenaline (or sympathin) are liberated by sympathetic stimulation and both substances can cause a contracture.

The response in the absence of drugs is best seen in Fig. 10 (*a*); it consists of an initial rise of tension which quickly passes off, followed by a small but perceptible rise of tension which is slow and prolonged. We consider that the initial rise is due to acetylcholine and that the second small but prolonged rise is due to adrenaline. In the absence of eserine the acetylcholine produced by the stimulation is destroyed too quickly for it to be responsible for this late second rise, the time relations of which agree with those of an adrenaline contracture.

The response in the presence of eserine may be examined in Figs. 1 and 3. There is a larger initial rise, a fall of tension and again a second rise which in Fig. 3 is nearly as great as the first one. The initial rise augmented by eserine must be due to acetylcholine; since eserine does not increase an adrenaline contracture, the second rise must also be due in the main to acetylcholine; we have already suggested that the fall in tension between the first and second rise is caused by the inhibitory effect of adrenaline (or sympathin) and that the second rise represents the continuation of the effect of the persisting acetylcholine as the inhibitory action of adrenaline passes away.

The response in the presence of ergotoxine is shown in Fig. 10 (*c*). There is an initial rise due to acetylcholine no greater than that seen before ergotoxine. The second rise is much greater than before, and we consider that it is almost certainly due to adrenaline, now able to exert a greater effect in causing contracture, when its vaso-constrictor action is paralysed; the second rise closely resembles the effect of an intra-arterial injection of adrenaline, and it is not increased when eserine is subsequently injected, as is the initial rise (see Fig. 10 (*e*)).

In concluding this discussion there is one possibility which should not be overlooked, though we think the evidence of Hinsey and Cutting [1933] makes it very unlikely. They were unable to obtain the Sherrington phenomenon when they stimulated the posterior roots, and from this and other evidence it is clear that the sensory fibres play no principal part in the phenomenon. The sensory fibres may, however, play some part, so that when they are stimulated together with the sympathetic fibres in the sciatic trunk, the effect of the sympathetic stimulation is augmented.

Summary.

1. After degeneration of the motor nerve supply, a contracture of the gastrocnemius muscle of the dog and the cat can be obtained by stimulation of the lumbar sympathetic chain. Thus the Sherrington phenomenon is due to the stimulation of the sympathetic fibres in the sciatic trunk.

2. The contracture which occurs when the chain is stimulated is much more readily seen in dogs than in cats; since we have shown that cholinergic dilator fibres are much more numerous in dogs than in cats, our evidence supports the view that the contracture is due to the liberation of acetylcholine from the endings of the sympathetic vaso-dilator fibres.

3. The denervated muscle of the leg responds to an injection of adrenaline by a slow prolonged contracture, though this is not always seen unless ergotoxine is previously injected.

4. Stimulation of the sympathetic chain liberates adrenaline (or sympathin) as well as acetylcholine. The liberated adrenaline affects the muscle in two ways; it reduces the rise of tension caused by acetylcholine, but it also causes a second late rise of tension. The reduction of tension is well seen after eserine when the prolonged effect of acetylcholine is broken by a period of reduced tension into two phases. The rise of tension caused by the liberated adrenaline is readily seen after ergotoxine, as a second rise following the rise due to acetylcholine.

The expenses of this research have been paid by a grant from the Government Grant Committee of the Royal Society. Our thanks are due to Mr H. W. Ling for his help, and to Messrs J. D. Riedel-E. de Haën, A. G., Berlin, for placing a supply of Pernocton at our disposal through their agents, the Old Strand Chemical and Drug Co., Ltd.

REFERENCES.

Bülbring, E. and Burn, J. H. (1935). *J. Physiol.* **83**, 483.
Dale, H. H. and Dudley, H. W. (1929). *Ibid.* **68**, 97.
Dale, H. H. and Gaddum, J. H. (1930). *Ibid.* **70**, 109.
Dale, H. H. and Gasser, H. S. (1926). *J. Pharmacol.*, Baltimore, **29**, 53.
Euler, U. S. v. and Gaddum, J. H. (1931). *J. Physiol.* **73**, 54.
Gasser, H. S. (1930). *Physiol. Rev.* **10**, 35.
Hill, A. V. and Hartree, W. (1920). *J. Physiol.* **54**, 84.
Hinsey, J. C. and Cutting, C. C. (1933). *Amer. J. Physiol.* **105**, 535.
Hinsey, J. C. and Gasser, H. S. (1928). *Ibid.* **87**, 368.
Hinsey, J. C. and Gasser, H. S. (1930). *Ibid.* **92**, 679.
Philippeaux, J. M. and Vulpian, A. (1863). *C. R. Acad. Sci.*, Paris, **56**, 1009.
Rijnberk, G. van (1918). *Arch. néerland. Physiol.* **1**, 262.
Sherrington, C. S. (1894). *J. Physiol.* **17**, 211.