

Return of function was evidenced by the appearance of an occasional flexion of the tail and either normal spreading of the caudal fin or its active motion (side to side fanning or its minute undulating motion constantly present in the normal fish). At first these signs were present only for short periods, following stimulation or when appearing spontaneously. These activities gradually became more constant, more forceful and better coördinated. In a few cases, the behavior ultimately closely approached that of the normal fish. The interval between operation and the first evidence of returning function varies greatly. In 3 fish such evidence appeared on the third day after operation; in 31 from the fourth to the sixth day; in 12 between the seventh and eleventh day. In addition, 13 others, observed for 7 days or less and which were among those sacrificed at intervals after operation for morphological study, showed no evidence of returning function before terminal anesthesia and fixation.

The evidence afforded by these observations indicates that the adult female of the rainbow-fish, *Lebistes reticulatus*, exhibits a high capacity for functional restoration following section of the spinal cord.

### 11136 P

#### Changes of Choline Esterase at End Plates of Voluntary Muscle Following Section of Sciatic Nerve.

R. COUTEAUX AND D. NACHMANSOHN.\* (Introduced by J. F. Fulton.)

*From the Lab. d'Anatomie comparée et de Physiologie générale de la Sorbonne, Paris.*

Choline esterase, present throughout the body, exists in especially high concentrations at the end plates of striated muscle, in the synapses both of the central nervous system and of the sympathetic ganglia.<sup>1, 2</sup> The concentration in these 3 foci is sufficiently high to satisfy the requirements of the theory that acetylcholine (ACh) may be involved in the transmission of nerve impulses from neurone to neurone or from neurone to striated muscle fibre. The alterations in enzymatic concentration which occur after the nerve endings disappear have been investigated.

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\* Now at the Laboratory of Physiology, Medical School, Yale University.

<sup>1</sup> Marnay, A., and Nachmansohn, D., *J. Physiol.*, 1938, **92**, 37.

<sup>2</sup> Nachmansohn, D., *Bull. Soc. Chim. Biol.*, 1939, **21**, 761.

It has been found in guinea pigs that section of the sciatic nerve is followed by a rise in *concentration* of choline esterase in the gastrocnemius muscle.<sup>3</sup> The Q.Ch.E. (mg ACh hydrolyzed per hour by 100 mg of fresh tissue) depends to a large extent on the number of end plates per unit of weight because of the high concentration of enzyme at these points. It was thought that the increase in choline esterase might be due to the decrease in volume of muscle fibers, leading to an increase in the number of fibers per unit of weight and consequently in number of end plates or more precisely "sole-plates" which persist after the nerve endings have disappeared. If the volume of fibers decreased and the Q.Ch.E. increased at the same time to the same extent it would imply that the enzyme concentration at the end plates remained practically the same.

Determinations of the volume of fibers, carried out 1-3 weeks after denervation show that the decrease fully accounts for the apparent increase of enzyme concentration. The rise of concentration actually is not quite as great as the corresponding diminution of volume.

The *total amount* of enzyme in the muscle decreases, the loss of weight being more important than the increase in concentration. Subtraction of the amount of enzyme present in muscle fibers from the total amount permits an approximate estimation of the amount of enzyme present at the end plates after section of the sciatic. During the first 2 weeks at the end of which nerve endings have disappeared, no measurable decrease of the enzyme concentration at the end plates occurs. Since during this time the enzyme concentration in the peripheral part of the cut sciatic falls by 50%, it can be assumed that with the disappearance of nerve endings some enzyme disappears, but that the amount is small compared with the large total amount of enzyme present at the end plates *outside* the nerve endings. After 3-4 weeks the enzyme at the "sole-plates" has decreased by about 30-40%. But a high concentration still persists and remains there for months.

The persistence of the enzyme at the end plates of denervated muscle can be demonstrated by a direct method based on a special arrangement of the nerve in the interior section of the gastrocnemius: The nerve spreads through nearly the whole muscle at one level only, situated in the midst of the muscle except for its entrance and its termination. In the middle third of this muscle all nerve fibers as well as the endings are situated in the middle zone. If this third is cut in slices with a freezing microtome the slices near the

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<sup>3</sup> Marnay, A., and Nachmansohn, D., *C. R. Soc. Biol. Paris*, 1937, **126**, 785.

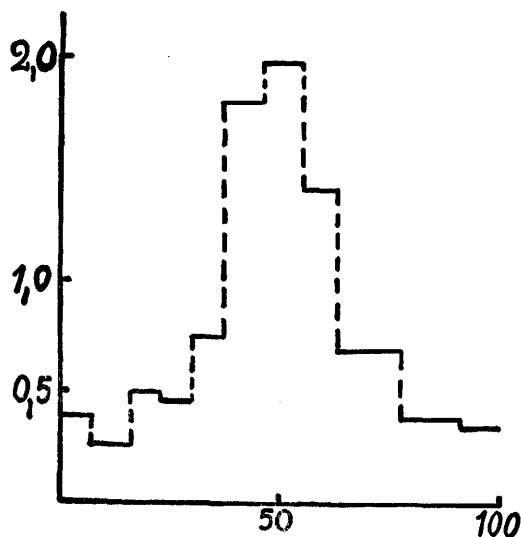


FIG. 1.

Concentration of choline esterase in the middle portion of the interior section of a guinea pig's gastrocnemius cut in 11 slices of similar thickness and weight. Each horizontal line corresponds to one slice and indicates its weight in % of the total weight. Abscissæ: Region from which the tissue was obtained in terms of order of consecutive slices. Point 50 corresponds to the center region where the nerve endings are situated. Ordinates: Q.Ch.E.

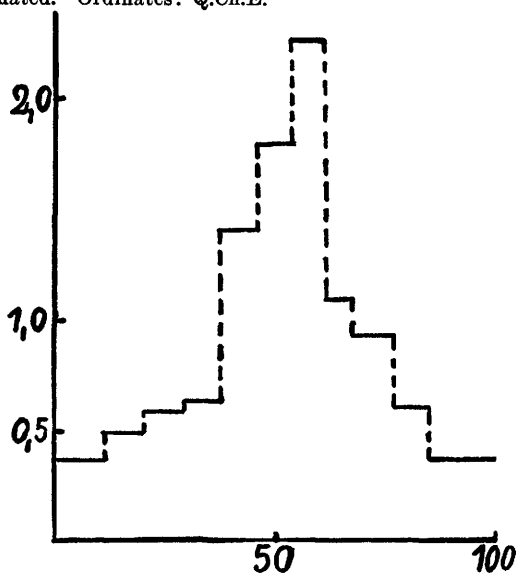


FIG. 2.

Concentration of choline esterase in the middle portion of the interior section of a guinea pig's gastrocnemius cut in 11 slices of similar thickness and weight, 14 days after section of the sciatic. Each horizontal line corresponds to one slice and indicates its weight in % of the total weight. Abscissæ: Region from which the tissue was obtained in terms of order of consecutive slices. Point 50 corresponds to the center region where the nerve endings are situated. Ordinates: Q.Ch.E.

upper and lower surfaces are practically free of nerve endings whereas those of the middle zone contain a large number. Determinations of choline esterase in these slices show a high concentration in the middle zone as compared with low concentration in the 2 nerveless zones. Fig. 1 gives the results obtained from the slices which are arranged in the order in which they were cut. The results confirm previous observations on frog sartorius. After denervation and the disappearance of nerve endings the concentration of the enzyme in the middle zone remains as high as before (Fig. 2).

The results indicate that the main bulk of the enzyme at the end plates is located outside the nerve endings. With their disappearance only a small amount disappears, which cannot be estimated. Later the enzyme concentration decreases to a certain extent but still remains high. Regeneration of the muscle fibers leads to an increase both of the concentration and of the total amount of the enzyme.

Bruecke observed a decrease of choline esterase in the superior cervical ganglion of cats after section of the preganglionic fibers and concluded that there was a high concentration inside the nerve endings.<sup>4</sup> These observations were qualitative estimations. Quantitative determinations confirming Bruecke's results, show that during the time when the nerve endings disappear the choline esterase decreases by about 60% (Fig. 3), which apparently indicates a high

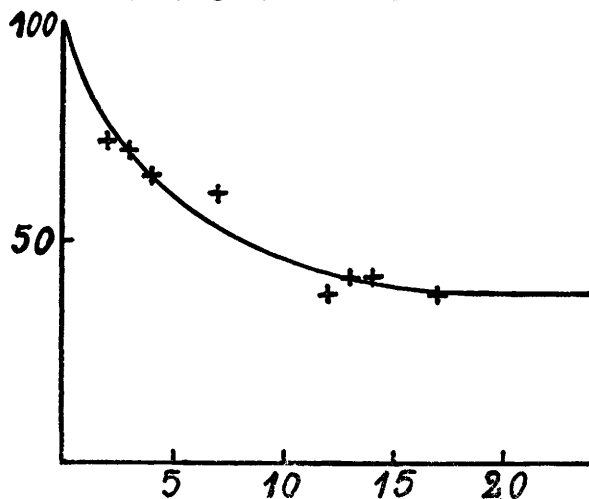


FIG. 3.

Decrease of choline esterase in the superior cervical ganglion of cats, after section of the preganglionic fibres. Abscissæ: Days after section. Ordinates: Amount of enzyme in % of the initial amount.

<sup>4</sup> Bruecke, F. v., *J. Physiol.*, 1937, **89**, 429.

amount localized inside the nerve endings. But then the enzyme power remains constant. After 5 weeks the same value was obtained. The Q.Ch.E. falls from the normal 40-60 to 20-25. This latter value is still very high. It is difficult to explain such a high value by the presence of the remaining fibers and cell bodies. There is reason to assume that considerable fraction of this enzyme is concentrated around the endings of preganglionic fibers and persists there as in the case of striated muscle. Even a fraction of the persisting enzyme if localized at the synapses would be sufficient to split during the refractory period the amount of ACh liberated at this ganglion by stimulation of preganglionic fibers.

The main difference between neuro-muscular junctions and ganglionic synapses seems to be that in the latter a greater fraction of the enzyme is localized inside the nerve endings. The disparity may be related to the powerful end arborization of preganglionic fibers which is not paralleled by that of the motor nerve endings of guinea pigs. The increase of the Q.Ch.E. of preganglionic fibers from 5.0 to a several times higher value at the nerve endings in the ganglion may indicate that the enzyme is localized near surfaces.

### 11137 P

#### Effect of Urine from Gastrectomized and Duodenectomized Dogs on Gastric Secretion.

M. H. F. FRIEDMAN, H. C. SALTZSTEIN AND A. A. FARBMAN.\*  
(Introduced by T. L. Patterson.)

*From the Department of Physiology, Wayne University College of Medicine, and Laboratories of Experimental Surgery, Harper Hospital, Detroit, Michigan.*

Previously we reported that from the urine of normal individuals there can be extracted a substance which inhibits gastric secretion when administered intravenously<sup>1, 2</sup> but not when administered subcutaneously<sup>2</sup> in the same dose. Ivy and his coworkers<sup>3</sup> and Necheles<sup>4</sup>

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\* With the assistance of R. O. Recknagel and H. M. Podolsky.

<sup>1</sup> Friedman, M. H. F., Recknagel, R. O., Sandweiss, D. J., and Patterson, T. L., *PROC. SOC. EXP. BIOL. AND MED.*, 1939, **41**, 509.

<sup>2</sup> Sandweiss, D. J., Saltzstein, H. C., and Farberman, A. A., *Detroit Physiological Soc.*, March 3, 1938; *A.M.A.*, San Francisco Meeting, June 17, 1938; also *Am. J. Digest. Dis.*, 1939, **6**, 6.

<sup>3</sup> Gray, J. S., Wieczorowski, E., and Ivy, A. C., *Science*, 1939, **89**, 489.

<sup>4</sup> Necheles, H., personal communication to Dr. D. J. Sandweiss, June 30, 1938; also Necheles, H., Hanke, M. E., and Fantl, E., *PROC. SOC. EXP. BIOL. AND MED.*, 1939, **42**, 618.