

of the muscle fiber. This observation is important, for it will be recalled that a similar decrease in the resistance of the muscle membrane has been shown in the crab to result from the stimulation of the inhibitory neuron<sup>7</sup>. All of the effects of GAB on the junctional potential were eliminated by washing the muscle free of the amino acid or by perfusing with a solution containing  $10^{-3} M$  picrotoxin and  $2.4 \times 10^{-4} M$  GAB.

In some instances, however, there was merely a slight change in the height of the junctional potential after GAB was added (Figure B), even though the contraction was considerably reduced. This result recalled the idea that the inhibitory transmitter may act at some point between the junctional potential and the contractile machinery, and that changes in the membrane may be merely incidental side effects<sup>8</sup>. The problem then is whether a small change in the height of the junctional potential could possibly lead to a large change in the tension produced by the muscle.

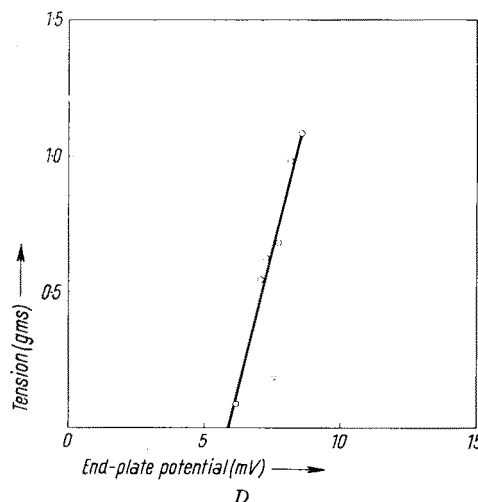
This question was explored by stimulating the motor neuron with pairs of closely spaced stimuli while measuring the junctional potential generated in one fiber and the tension exerted by the entire muscle. Pairs of stimuli separated by more than 2.5 ms produced a double junctional potential (Figure C). As the interval between the first and the second stimuli was increased, the peak depolarization of the muscle membrane was also increased, until a maximum was reached when the interval between stimuli was about 5.3 ms. So by this technique the height of the junctional potential could be varied, and the relation between the height of the junctional potential and the tension produced by the muscle could be determined (the assumption underlying this method is that the behavior of the single junction at which the electrode is lodged is typical of junctions throughout the muscle; recordings with extracellular electrodes appear to support the assumption).



C The double junctional or end plate potential produced by a pair of closely spaced stimuli applied to the 'fast' motor neuron (upper trace) and the tension produced by the closer muscle (lower trace; an increased tension gives a downward deflection)

The results of a typical experiment are plotted as Figure D. Apparently a small change in the peak depolarization of the junctional potential produced a large change in the tension exerted by the muscle. So it seems that even the small decrease in the junctional potential sometimes found after exposure to GAB may be sufficient to account for a pronounced change in the contraction of the muscle. From our results, it appears probable that GAB inhibits

the contraction of the muscle by producing a decrease in the height of the junctional potential.



D The relation between the peak depolarization in the junctional potential and the maximum tension produced by the closer muscle

We conclude that GAB mimics the effects of the inhibitory transmitter on crayfish muscle because this amino acid inhibits the contraction of the muscle and acts on the junctional potential like the inhibitory transmitter, and also because its action is blocked by picrotoxin.

W. G. VAN DER KLOOT and J. ROBBINS

*Department of Pharmacology, New York University College of Medicine and Department of Pharmacology, College of Physicians and Surgeons, Columbia University, New York, July 15, 1958.*

#### Résumé

L'acide  $\gamma$ -amino-butérique et la  $\beta$ -alanine reproduisent l'action du transmetteur inhibiteur du système neuromusculaire de l'écrevisse. Ces deux acides aminés diminuent la contraction musculaire. L'acide  $\gamma$ -amino-butérique réduit la différence de potentiel et augmente sa décomposition. L'action de ces acides aminés est bloquée par la picrotoxine comme l'est celle du transmetteur inhibiteur.

### Hypothalamic Nerve Fibres in the pars tuberalis and Pia-arachnoid Tissue of the Cat and their Degeneration Pattern after a Lesion in the Hypothalamus

Much experimental data of the last ten years have shown that the function of the anterior pituitary gland is influenced by the hypothalamus. However, the structures and the precise mechanisms of the «connecting link» between the hypothalamus and the adenohypophysis remains still unsettled. It is widely accepted that the secretion of the anterior pituitary is regulated by humoral substances from nerve endings in the median eminence, which are transmitted by hypophyseal portal vessels to the cells of the pars distalis, but unequivocal data which support or establish this thesis as the sole or major mechanism of regulation are absent (SAYERS, REDGATE and ROYCE<sup>1</sup>). Therefore, also other structures and mecha-

<sup>7</sup> P. FATT and B. KATZ, *J. Physiol.* 121, 374 (1953).

<sup>8</sup> C. A. G. WIERSMA, *Recent Advances in Invertebrate Physiology* (University of Oregon Publications, Eugene 1957). – G. HOYLE, *The Nervous Control of Muscular Contraction* (Cambridge University Press, Cambridge 1957).

<sup>1</sup> G. SAYERS, E. S. REDGATE, and P. C. ROYCE, *Ann. Rev. Physiol.* 20, 243 (1958).

nisms of the nervous regulation of the adeno-hypophysis, such as a direct innervation of the gland cells of the adeno-hypophysis by hypothalamic nerve fibres and by nervous structures of the peripheral autonomic nervous system must be taken into consideration (HAIR<sup>2</sup>, URASOV<sup>3</sup>, METUZALS<sup>4,5</sup>).

The problem of a direct innervation of the adeno-hypophysis by hypothalamic nerve fibres was investigated in normal cats and in cats with a lesion in the hypothalamus. The cats were perfused through the heart with Ringer's solution warmed to body temperature, and then with 13% formalin solution. Frozen sections of 35  $\mu$  thickness of the pituitary were cut with the diencephalon attached. The sections were impregnated according to the Bielschowsky-Gros method (ROMEIS<sup>6</sup>).

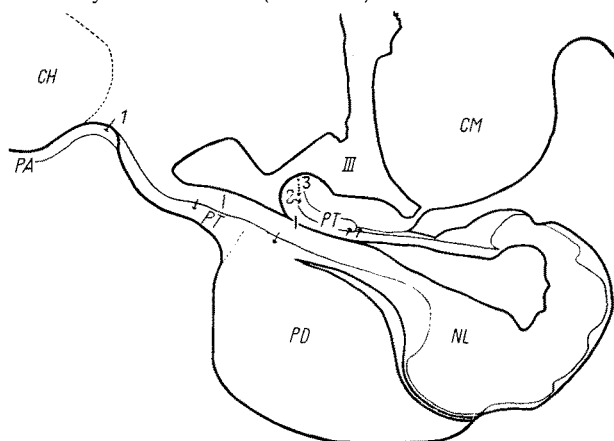


Fig. 1.—Diagram of a midline sagittal section through the hypothalamus and pituitary gland of the cat. PD – pars distalis, PT – pars tuberalis adeno-hypophyseos, NL – neural lobe, I – Infundibulum\*, CH – optic chiasma, CM – corpus mamillare, III – third ventricle, PA – pia-arachnoid,  $\downarrow$  – indicates schematically the places where nerve fibres of the infundibulum pass into the pars tuberalis and pia-arachnoid tissue,  $\downarrow$  – degenerating nerve fibres, 2  $\rightarrow$  – correspond to Figure 2, 3  $\downarrow$  – correspond to Figure 3.  $\times 12$ .

Nerve fibres and nerve fibre strands enter from the infundibulum into the pars tuberalis adeno-hypophyseos. In Figure 1 the places where hypothalamic nerve fibres pass over into the pars tuberalis are indicated schematically ( $\downarrow$ ). Small bundles of nerve fibres were observed to enter from the most cranial part of the infundibulum into the tissue layer of the pia-arachnoid (PA, Fig. 1) cranial to the pars tuberalis (1  $\downarrow$ , Fig. 1). These hypothalamic fibres join to nervous strands of different sizes extending in the pia-arachnoid tissue.

Strands of fine nerve fibres pass from the infundibulum particularly into the dorsal parts of the pars tuberalis. They enter the connective tissue and capillary layer between the infundibulum and the pars tuberalis and extend further among the gland cell cords and the blood vessels. Figure 2 illustrates two such strands (the position of these strands is indicated in Fig. 1, 2  $\rightarrow$ ). They are composed of fine nerve fibres showing fusiform enlargements. Because the strands were cut off in the slide, the manner of the extension of the strands further into the pars tuberalis

could not be observed. The portions of the strands distal from the infundibulum are less heavily impregnated than the portions nearer the infundibulum. These nervous strands in the cat may be comparable with those already described in the horse<sup>4</sup>.

In a male cat, a lesion was made in the hypothalamus by means of the method of HESS<sup>7</sup>. The lesion was caused by a high-frequent current between a pair of electrodes 0.25 mm in diameter. The distance between the electrodes was 1.5 mm. Survival time of the cat was 4 days. The direct lesion extended unilaterally over the caudal part of the nucl. periventricularis arcuatus<sup>8</sup>, the posterior hypothalamic area and the anterior part of the mamillary body. Also some indirect damage of glandular tissue of the pars tuberalis near the hypothalamus was caused by the lesion.

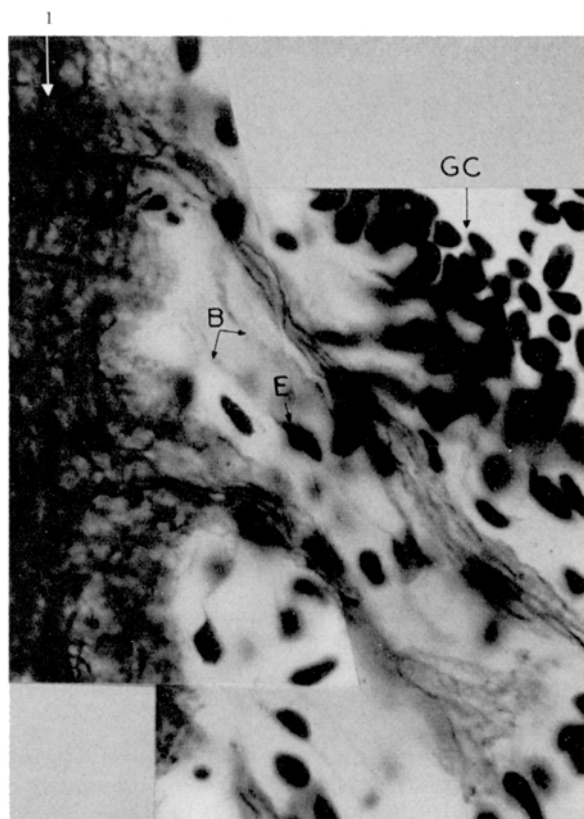


Fig. 2.—Two nerve fibre strands from the infundibulum pass into the dorsolateral part of the pars tuberalis. The position of the strands is indicated in Figure 1 by 2  $\rightarrow$ . I – infundibulum, GC – nuclei of the gland cells of the pars tuberalis, A – strand oriented parallel to a capillary, B – boundaries of the capillary slightly blackened by silver, E – nucleus of an endothelial cell. The connective tissue layer between the nervous and glandular tissues is not impregnated. Cat. Bielschowsky-Gros.  $\times 800$ .

In the dorso-lateral part of the pars tuberalis of the experimental animal nerve fibre strands, partly corresponding to those illustrated in Figure 2, were broken up into conspicuous beaded fragments and granular masses. Figure 3 illustrates two strands showing such typical degenerative appearances. The position of these degenerating strands in relation to the different parts of the pituitary is indicated in Figure 1 (3  $\downarrow$ ). A part of the nerve

<sup>2</sup> G. W. HAIR, *Anat. Rec.* 71, 141 (1938).

<sup>3</sup> I. G. URASOV, *Vest. Leningr. Univ.* 7, 47 (1955), in Russian; Engl. summary in *Int. Abstr. Biol. Sci.* 7, 4 (1957).

<sup>4</sup> J. METUZALS, *Acta anat.* 20, 258 (1954).

<sup>5</sup> J. METUZALS, in *Pathophysiologia Diencephalica*, Internat. Symposium, Milan, May 1956 (Springer, Wien 1958), p. 148.

<sup>6</sup> B. ROMEIS, *Mikroskopische Technik* (Leibniz, München 1948).

\* According to RIOCH, WISLOCKI, and O'LEARY<sup>8</sup>), the median eminence is the bulbous part of the infundibulum down to the point where the straight infundibular stem clearly commences.

<sup>7</sup> W. R. HESS, *Das Zwischenhirn* (Benno Schwabe & Co., Basel 1949).

<sup>8</sup> D. McK. RIOCH, G. B. WISLOCKI, J. L. O'LEARY, *Res. Publ. Ass. nerv. ment. Dis.* 20, 3 (1940).

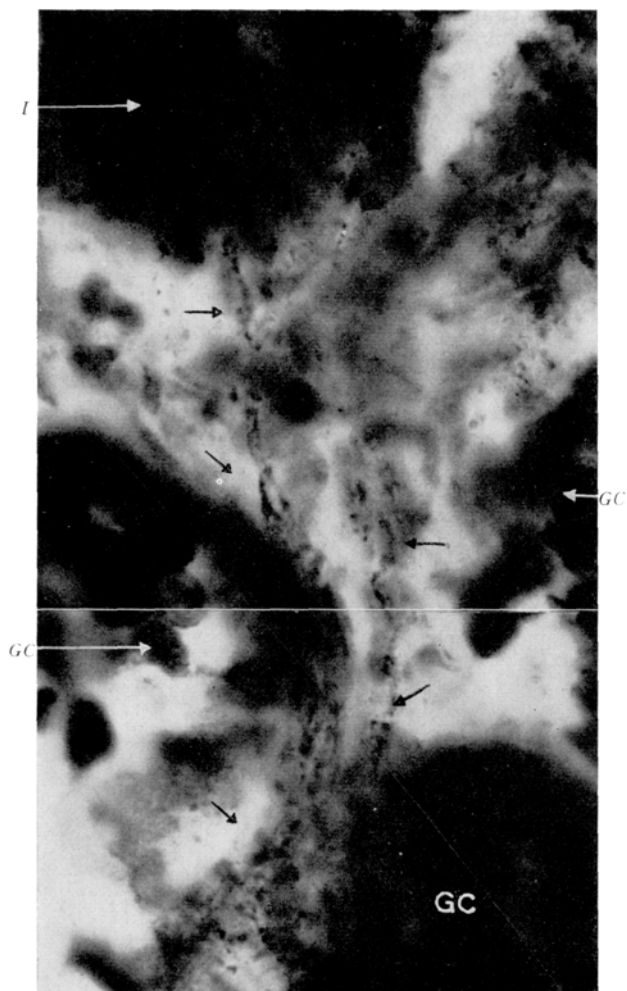


Fig. 3.—Two nerve fibre strands in the pars tuberalis showing typical degenerative appearances — granular masses and beaded fragments ( $\downarrow$ ). The position of the strands is indicated in Figure 1 by 3  $\downarrow$ . GC — gland cell cords of the pars tuberalis, I — infundibulum. Cat. Bielschowsky-Gros.  $\times 1000$ .

fibres of the infundibulum were broken up into loose nerve fibre fragments and granular masses. Such granular masses and beaded fragments ( $\downarrow$ ) can be followed from the infundibulum (I) continuously into the pars tuberalis between the gland cell cords (GC), as is convincingly demonstrated in Figure 3. Some damage of the gland cells caused by the lesion is indicated by the heavy impregnation of the cell cords.

The results of the present investigation indicate that considerable number of hypothalamic nerve fibres pass

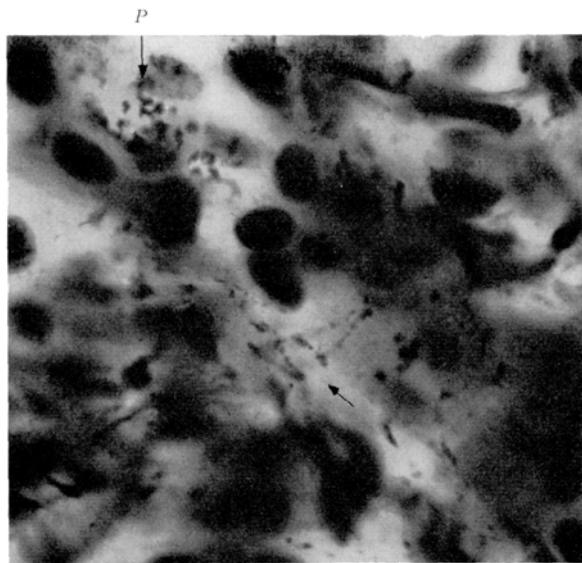


Fig. 4.—Beaded fragments ( $\downarrow$ ) among the gland cells of the pars tuberalis, showing similar arrangement as degenerating nerves. The structures are probably processes of connective tissue cells. P — precipitate of silver on the surface of the slide. Cat. Bielschowsky-Gros.  $\times 1000$ .

from the infundibulum into pia-arachnoid tissue and into different parts of the pars tuberalis, contrary to the general opinion that few or no such fibres exist.

Granular masses could also be found among the gland cells in the dorsal pars tuberalis of the experimental animal, but not in the pars tuberalis of normal animals. At the present time, it is not possible to determine exactly the nature of these structures. Beaded fragments among the gland cells of the pars tuberalis (Figure 4), which could be easily interpreted as degenerating nerve fibres, are probably processes of connective tissue cells. Such connective tissue cells could be found in the dorsal pars tuberalis of the operated animal.

J. METUZALS

Zoologisch Laboratorium der Rijksuniversiteit te Groningen, Haren (Nederland), August 18, 1958.

#### Zusammenfassung

Mit der Bielschowsky-Gros-Methode wurden bei Katzen Nervenfasern untersucht, die vom Infundibulum in die Pars tuberalis ziehen. Die Befunde an normalen Tieren und an einer Katze mit einer Hypothalamus-Läsion zeigen, dass eine beachtliche Zahl hypothalamischer Fasern in verschiedene Teile der Pars tuberalis, sowie in das Gewebe der Pia und der Arachnoidea übergehen.

Bei der operierten Katze wurde das Degenerationsbild der in den dorsalen Teil der Pars tuberalis ziehenden Hypothalamus-Fasern beschrieben.

## Informations - Informationen - Informazioni - Notes

### STUDIORUM PROGRESSUS

#### Sur le dédoublement par entraînement. La notion de sursaturation «rémanente»

Par G. AMIARD<sup>1</sup>

Une variante du dédoublement spontané des racémiques par cristallisation, récemment appelée dédouble-

ment par *entraînement*<sup>2</sup>, consiste à provoquer la cristallisation dans une solution renfermant un excès de l'énantiomorphe désiré<sup>3-7</sup>. Si l'opération est conduite dans certaines conditions, on recueille une quantité de cristaux de

<sup>2</sup> G. AMIARD, Bull. Soc. chim. France 1956, 447.

<sup>3</sup> F. ST. KIPPING et W. J. POPE, J. chem. Soc. 75, 36 (1899).

<sup>4</sup> E. DARMOIS, C. R. Acad. Sci. 237, 124 (1953).

<sup>5</sup> A. WERNER, Ber. deutsch. chem. Ges. 47, 2171 (1914).

<sup>6</sup> R. DUCHINSKY, Festschrift Emil Barends, 375 (1936).

<sup>7</sup> E. CALZAVARA, B. F. 763374 du 23, 1. 33.

<sup>1</sup> Services de Recherches Roussel-Uclaf, Paris, le 29 juillet 1958.