The atrophy was less, but the decrease in cholinesterase more pronounced, than after a big dose of botulinum toxin (Table II). In contrast to botulinum toxin, denervation caused a decreased enzyme activity also when this was expressed on a weight basis (Table I). In the series of rats both denervated and given toxin, the atrophy was greater than after only denervation, while the enzyme activity was comparable to that after denervation (Table II).

Discussion. The results here reported show that two weeks after the injection of botulinum toxin into the anterior tibial muscle there is a decreased cholinesterase activity. The decrease is, however, less than that obtained after denervation of the muscle. The difference can not be explained by loss of enzyme held in the degenerating nerve terminals, since these are known to contain extremely small amounts of enzyme compared with the end-plate (COUTEAUX[3]). On denervation of the muscle in addition to toxin, the decrease was similar to that after only denervation. The toxin therefore does not seem to have any effect in itself on the enzyme, which can explain why botulinum toxin does not have the same effect as section of the nerves. Thus there seems to be some influence of the nerve on the muscle not blocked by botulinum toxin. In this connexion, it is of interest to compare the suggestion put forward that the nerve has some influence on the muscle apart from that caused by release of acetylcholine (BULLER, ECCLES and ECCLES[4]). It may also be recalled that long-continued blocking of the effect of the cholinergic nerves on salivary glands by an atropine-like agent does not cause changes in the cholinesterase activity of the gland (STRÖMBLAD[5]).

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Zusammenfassung

Bei Zufuhr von Botulinum-A-Toxin in den Muskel Tibialis anterior der Ratte war die Cholinesteraseaktivität nach zwei Wochen herabgesetzt, jedoch weniger ausgesprochen als nach zweiwöchiger Denervierung des Muskels.

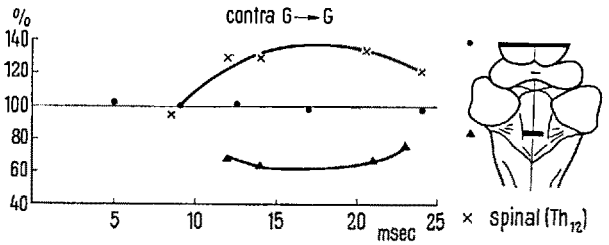
Crossed Reflex Actions Evoked by High Threshold Muscle Afferents

In spinal cats impulses in group II (12–4 μ) and III (4–1 μ) muscle afferents evoke in ipsilateral motoneurones the actions of the general flexion reflex, i.e. widespread excitation to flexor and inhibition to extensor motoneurones. It has, however, been observed that under some conditions these afferents give inhibitory effects to flexors and sometimes also excitatory to extensors[1]. The present investigation is concerned with contralateral effects from these afferents. Unanaesthetized decerebrate cats with both hindlimbs denervated were used. The effect of single conditioning volleys evoked at different stimulus strengths was examined on contralateral monosynaptic test reflexes.

In acute spinal preparations, no group I effects were observed but it has been confirmed that there are usually pronounced effects by volleys in high threshold muscle afferents[2]. Extensors as well as flexors received either inhibition or excitation but in each experiment the same effect was usually exerted on motor nuclei of synergic muscles (extensors or flexors). These effects occurred in combinations which included all four possible variations: (1) generalized excitation or (2) inhibition or (3) excitation to flexors and inhibition to extensors or (4) finally the opposite with inhibition to flexors and excitation to extensors. The last variation was the most frequent one, and would be expected, according to Sherrington's scheme of double reciprocal innervation. In one case a reversal from inhibition to excitation occurred in flexor motor nuclei during the course of the experiment. As group II and III volleys can evoke different crossed effects in different animals under apparently identical conditions, there is, both to contralateral flexor and extensor motoneurones, one inhibitory and one excitatory pathway. Presumably the balance between these channels is labile and easily influenced, whereas on the ipsilateral side the excitatory path to flexor and the inhibitory path to extensor motor nuclei are very predominant in the spinal state.

In the decerebrate cat, the ipsilateral synaptic actions by group II and III muscle afferents may be completely suppressed[3]. This is due to a tonic inhibitory control from the brain stem of interneurones mediating there effects. It has now been found that a similar control is exerted on the interneurones mediating crossed group II and III effects, as is evidenced by the finding that in the decerebrate state stimulation of these fibers never evoked effects in contralateral motoneurones.

The release of supraspinal control of ipsilateral reflex arcs has been investigated after various lesions in the lower reticular formation[4]. After a medial lesion in lower pons, there is a release of the tonic control of the inhibitory path to ipsilateral extensor motoneurones and concomitantly an opening of an inhibitory path to ipsilateral flexor motoneurones. Release of the excitatory pathway to ipsilateral flexors occurs only after a more caudal lesion in the brain stem. Similar investigations have now been made on the supraspinal control of crossed actions. A low pontine lesion gave release of crossed inhibition to extensors. In the Figure a group II + III volley in the nerve to gastrocnemius-soleus had no effect on contralateral extensor motoneurones (gastrocnemius-soleus) in the decerebrate state (•). After a pontine



The effect of a conditioning volley in the nerve to gastrocnemius-soleus on the monosynaptic reflex from the contralateral gastrocnemius-soleus nerve recorded in the ventral root. 100% on the ordinate represents the unconditioned amplitude of the test reflex. Conditioned amplitude, expressed as percentage of control amplitude, is plotted as a function of time interval between the conditioning and testing stimuli. The three series of measurements were obtained after the lesions indicated in the schematic drawing, all at a conditioning stimulus strength supramaximal for group III.

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[3] R. M. ECCLES and A. LUNDBERG, J. Physiol. *147*, 565 (1959).
[4] B. HOLMQVIST and A. LUNDBERG, J. Physiol. *148*, 70 P (1959).

lesion, there was inhibition (▲), which changed into excitation after a low spinal section (×). In addition the pontine lesion gave a release of inhibition to contralateral flexor motor nuclei, which sometimes remained, sometimes changed to excitation after a lesion at obex. Thus the effect of a low pontine lesion is a generalized release of inhibitory group II and III pathways to ipsilateral and contralateral extensor and flexor motoneurones with no sign of a reciprocal or a double reciprocal organization.

There are similarities in the supraspinal control of ipsi- and contralateral effects by high threshold muscle afferents which suggest a functional linkage. However, the experiments with supraspinal control have also given further evidence of the existence of two sets of pathways mediating actions of opposite modality from these afferents to contralateral extensor as well as flexor motoneurones. It is possible (as has been suggested for the ipsilateral effect[4] that the opposite actions on a given contralateral motor nucleus are evoked by one and the same afferent system and that supraspinal centres can select either channel or close both.

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Résumé

Chez le chat spinal des impulsions provoquées dans les fibres afférentes à seuil élevé d'origine musculaire chez le chat spinal facilitent ou inhibent des motoneurones contra-latéraux fléchisseurs et extenseurs. Chez l'animal décérébré, ces actions croisées sont supprimées, tandis qu'après une lésion pontine, les actions inhibitrices se manifestent.

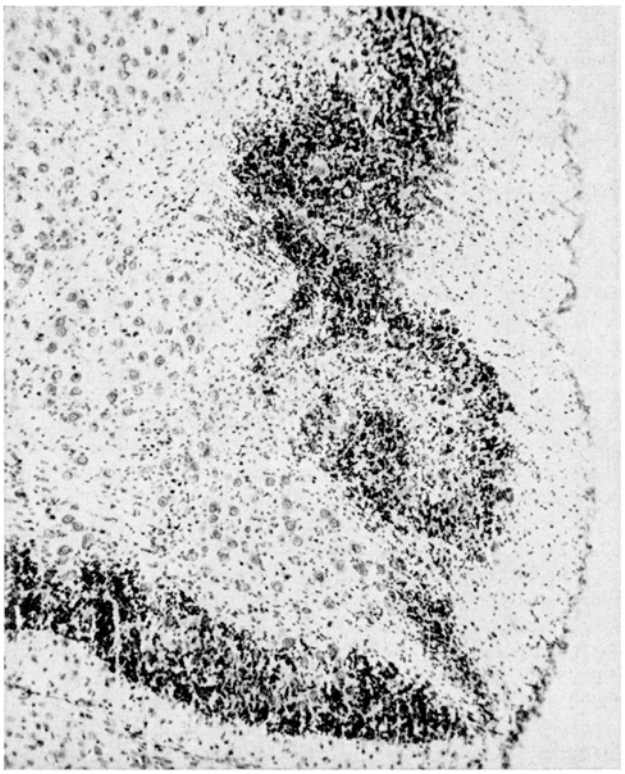

Fig. 1. Sagittal section of cerebellum of a mouse homozygous for 'reeler' gene. Age 18 days. Note: Invasion of molecular layer by various cells, reduction of granular layer and disturbance of the arrangement of Purkinje cells. × 80.

Observations on the Neuropathology of 'Reeler', a Neurological Mutation in Mice[1]

The 'reeler' syndrome is caused by a single recessive gene mutation. The main features of the derangement caused by the 'reeler' gene are: lack of muscular coordination, balancing difficulties and tremors. Mice homozygous for the 'reeler' gene show the disturbance after the 12th day of postnatal age. They rarely survive beyond the 21st day, and we have never been able to raise any of them to breeding age. Mice heterozygous for the 'reeler' gene behave normally.

In a previous communication[2] we reported a 100% increase of cerebellar cholinesterase in mice homozygous for the 'reeler' gene over the values obtained for cerebellar enzyme concentration of normal littermates, who carry the 'reeler' gene either in heterozygous condition or not at all.

A study of the neuropathology of this mutation was begun with a histological analysis of brains of 'reeler' animals.

'Reeler' mice and their normally behaving littermates were sacrificed between the ages 18–21 days postnatally. Their brains were fixed in 10% formalin. The material was embedded in paraffin, cut 15 μ thick in sagittal or parasagittal plane. The sections were stained with cresyl violet.

The histological analysis revealed that the organization of the cerebellum is markedly altered in afflicted 'reeler' animals. The typical appearance of the folia is missing. The arrangement of Purkinje cells which normally surround the granular layer is severely disturbed. The granular layer is much reduced and the area of white matter contains large numbers of cells, which resemble Purkinje cells (Fig. 1 and 2).

The meaning of our findings in terms of neurophysiology is still obscure. The nature of the 'reeler' syndrome, the most dramatic symptoms of which consist of fine intentional tremors, and failure to maintain locomotor balance in homozygous animals certainly suggests cerebellar involvement.

In considering these findings it is of interest that the histological analysis of brains of the 'reeler' mutants revealed a severe disturbance of the cytoarchitectonics of the cerebellum. In a number of neurological mutants in mice, which have been investigated, no gross histological changes within the central nervous system have been detected[3–5]. There are a few exceptions. In the 'agitans' mutation, which resembles the 'reeler' syndrome in some respects, degenerative changes limited to Purkinje cells and to a lesser degree to mossy fibers of the cerebellum of afflicted animals have been reported by MARTINEZ and SIRLIN[6].

[1] Supported by Grant No. B-1716 from USPHS National Institute of Neurological Diseases and Blindness.

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