

original method of BARNETT and SELIGMAN^{9,10}. The parathyroid glands were taken from normal and bilaterally nephrectomized rats, and also from toads captured seasonally throughout the year. The glands were fixed overnight in trichlor acetic acid alcohol (1% trichlor acetic acid in 80% ethanol), embedded in paraffin, sectioned at 8 μ and stained.

In the parathyroid glands from bilaterally nephrectomized rats, a noticeable abundance of intensely DDD diazo blue B reactive granules was observed as compared with those from normal animals. These granules were most numerous in the cytoplasm of the parenchymal cells and particularly within its peripheral layer. An example of this abundance is shown in Figure 2.

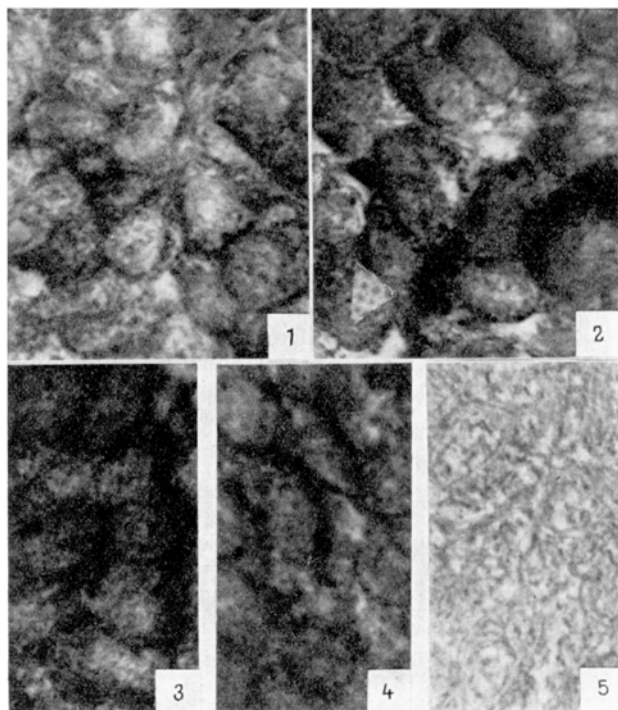


Fig. 1. DDD diazo blue B reactive granules in parathyroid tissue of a normal rat. $\times 1500$.

Fig. 2. An abundance of intensely DDD diazo blue B reactive granules in parathyroid tissue of a rat at 12 h after bilateral nephrectomy. $\times 1500$.

Fig. 3. An abundant amount of intensely DDD diazo blue B reactive granules in parathyroid tissue of a toad captured in July. $\times 1500$.

Fig. 4. A moderate amount of DDD diazo blue B reactive granules in parathyroid tissue of a toad captured in October. $\times 1500$.

Fig. 5. A minimal amount of faintly DDD diazo blue B reactive granules in parathyroid tissue of a toad captured in February. $\times 1500$.

In the parathyroid glands from toads sacrificed in summer, an abundant amount of intensely DDD diazo blue B reactive granules were seen in the cytoplasm of the parenchymal cells, on the outer surface of the plasma membrane and within the connective tissue space (Figure 3)—while in those from animals killed in spring and autumn the amount and stainability of the granules were found to be moderate (Figure 4). In the parathyroid glands from toads in dormant state, the granules were minimal in amount and staining intensity, in fact occasionally absent (Figure 5).

From the physiological point of view, the result is possible to comprehend. The present evidence indicates certainly that the amount and staining intensity of DDD diazo blue B reactive granules are approximately parallel to the extent with which the parathyroid gland is physiologically functioning in both animal species studied. On this basis, it may be concluded that the granules are a reflection of the secretory activity of the glandular cells. It deserves, however, discussion whether the granules represent parathyroid hormone itself or its supporting material. According to RASMUSSEN¹¹, parathyroid hormone consists of a polypeptide which is found on analysis to contain only an exceedingly small amount of cystine. Therefore, although the hormone may be responsible, to a certain extent, for the stainability of the DDD diazo blue B reactive granules, a carrier protein containing significant amounts of sulphhydryl and disulphide groups and existing in association with the active principle appears to constitute the major moiety of the granules. This notion seems significant in view of a biochemical evidence that the biological activity of parathyroid hormone is relevant to its oxidation-reduction properties¹¹.

More detailed studies on the morphological features of DDD diazo blue B reactive granules in the parathyroid tissues will be published elsewhere.

Zusammenfassung. Granula in Parenchymzellen von Ratten und Kröten-Epithelkörperchen treten durch 2, 2'-Dihydroxy-6, 6'-dinaphthyl-disulfid (DDD) Diazo-Blau-B gefärbt, deutlich hervor. Ihre Menge, Färbbarkeit und Verteilung unter verschiedenen Bedingungen scheinen für die zelluläre Sekretionstätigkeit charakteristisch zu sein.

K. YAMADA

Department of Anatomy, School of Medicine, University of Nagoya (Japan), April 13, 1963.

⁹ R. J. BARNETT and A. M. SELIGMAN, *Science* **116**, 323 (1952).

¹⁰ R. J. BARNETT and A. M. SELIGMAN, *J. Nat. Cancer Inst.* **14**, 769 (1954).

¹¹ H. RASMUSSEN, *The Parathyroids* (Charles C. Thomas Publisher, Springfield, Illinois 1961).

Plantar Reflexes in Cat

SHERRINGTON¹ described that broad innocuous pressure of the plantar cushion in the spinal dog evoked a bilateral brief extension of the whole limb—the extensor thrust. Reflex responses, in man, to painful stimulation of different parts of the foot have been elucidated². The present investigation on acute spinal cats deals with some plantar reflex actions to muscles performing toe and ankle

movements. Reflex actions from different skin areas to the short toe muscles flexor digitorum brevis (FDB) and extensor digitorum brevis (EDB) have been studied in a few cases³ but in general reflex connections to the short muscles of the foot are unknown.

¹ C. S. SHERRINGTON, *Proc. Roy. Soc.* **76**, 161, 269 (1905).

² E. KUGELBERG, K. EKLUND, and L. GRIMBY, *Brain* **83**, 394 (1960).

³ K.-E. HAGBARTH, *Acta physiol. scand.* **26**, Suppl. 94 (1952).

Plantar flexion of toes can be evoked on gentle pressure of the central pad. There are in particular strong excitatory connections to FDB and an electromyographic discharge in this muscle is observed when gentle pressure is applied to the central pad of the hind-foot (Figure 1). Some of the deeper muscles in the foot take part in this reflex from the pad, but these effects have not yet been investigated in detail. Monosynaptic testing has revealed excitatory actions by the same stimulus also to flexor digitorum longus (FDL) and to plantaris, the tendon of which is connected in series with FDB in the cat. All these reflex actions are elicited by gentle pressure, but increase with stronger squeezing of the pad and will also appear on pinching of the plantar skin (Figure 2). No facilitation to the motor nuclei of knee extensors and of the ankle extensor gastrocnemius-soleus was seen from the pad or the

planta but with sharper mechanical stimulation there was the expected inhibitory action of the flexor reflex. The time course of the excitatory action to the motor nuclei of the toe extensors (= plantar flexors) has been studied by conditioning the monosynaptic reflexes with weak single shocks applied through small needles inserted 2 mm into the pad (Figure 2).

Based on their participation in the general flexor reflex and in stepping, hindlimb muscles have been classified as extensors or flexors, thus plantaris (Pl) and FDL are extensor muscles, inhibited in the flexor reflex⁴. A strong pinching of the toes inhibits the Pl and FDL motor nuclei but facilitates in most cases the monosynaptic reflex to FDB, or even causes a weak discharge in this muscle. There is some further evidence of excitatory actions from flexor reflex afferents to FDB, e.g. from skin afferents in the long branch of the sural nerve, and from high threshold knee joint afferents, but other skin nerves have generally given inhibition. The activation of FDB in the extension phase of the step is similar to the activity in hip, knee and ankle extensors in walking.

The antagonist muscle EDB does not seem to be reciprocally inhibited by the moderate stimuli to the pad—it is, however, facilitated by stronger pressure or pinching or by somewhat stronger electrical stimulation of either the central or the peripheral toe pads (to about the same degree as other flexors of the leg). The other antagonist in question, extensor digitorum longus (EDL), has a somewhat varying pattern with excitatory or inhibitory actions dominating from different areas of the foot. It often receives both excitatory and inhibitory actions from the central pad on stronger stimulation. Both EDB and EDL motor nuclei receive facilitation from the flexor reflex afferents but the effects to EDL were often mixed excitatory and inhibitory.

It is suggested that the described reflex from the pad, being elicited by stimuli that must be considered far from noxious, is not a functional variety of a protective flexor reflex in the cat, but that it is part of a reflex system assisting in locomotion or related motor performances of the foot.

Zusammenfassung. Spinale Reflexe an Zehenstrecker- und Zehenbeugermuskeln der Katze wurden elektrophysiologisch untersucht, wobei eine effektive Reflexbahn zu den Zehenplantarbeugern, deren Reflex durch leichten Druck auf den Sohlenballen der Hinterpfote ausgelöst wird, gefunden wurde.

I. ENGBERG

Department of Physiology, University of Göteborg (Sweden), 24. April 1963

⁴ C. S. SHERRINGTON, *J. Physiol.* 40, 28 (1910).

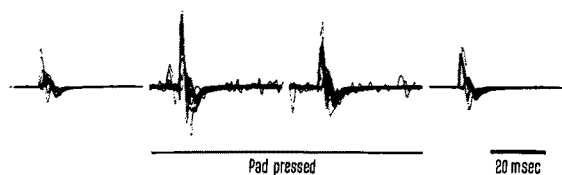


Fig. 1. Electromyography from FDB showing excitatory effects from the central pad of the hind-foot. Several superimposed records with monosynaptic test reflexes (evoked by stimulation of the intact tibial nerve) are taken before, during and after the application of a continuous, very light pressure on the pad. The interval between each set of records is about 5 sec. (A reflex discharge is evoked from the pad besides the facilitation of the test.)

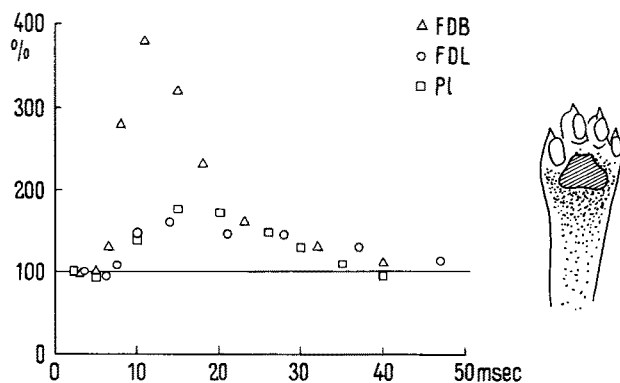


Fig. 2. Facilitation of monosynaptic reflexes to FDB, FDL and Pl obtained by electrical stimulation of the central pad with single shocks (condensor discharges of 12 V, half decay time 25 μ sec) 100% on the ordinate represents the height of the unconditioned test reflexes. The time is measured between the conditioning and testing stimuli. The drawing indicates the areas that give excitatory actions to the same muscles on adequate stimulation. At the central pad only gentle pressure is required; in the dotted area, stronger pinching.

$\text{Cu}(\text{CH}_3\text{CN})_2^+$, ein Mittel zum Studium homogener Reaktionen des einwertigen Kupfers in wässriger Lösung

Im Rahmen unserer Studien zur Koordinationschemie der Flavocoenzyme¹⁻³ suchten wir nach einer einfachen Möglichkeit zum quantitativen Studium von Komplexbildungsreaktionen des einwertigen Kupfers in homo-

gener, wässriger Lösung. Wir fanden, dass der erstmals von MEERWEIN et al.⁴ aus Cu_2O , CH_3CN und BF_3 -Ätherat erhaltene Komplex $\text{Cu}(\text{CH}_3\text{CN})_4\text{BF}_4$, welcher aus CH_3CN in schönen farblosen, kaum luftempfindlichen Nadeln kristallisiert, auch gegen Hydrolyse überraschend beständig ist in Gegenwart von überschüssigem CH_3CN , sofern ein kritisches $\text{pH} = f([\text{CH}_3\text{CN}])$ nicht überschritten wird. Die polarographische Untersuchung zeigt, dass in