(Figure) and 40% of those that remained indifferent to peripheral stimulation. The effect of L-glutamate occurred after a latency of 0.2–0.5 sec, and the discharge rate returned to control levels of activity after 0.1–0.3 sec. These findings appear to be in contrast with results obtained in other brain regions, where practically all cells were activated by L-glutamate[3,4]. However, considering that 40% of the cells recorded in the dorsal column nuclei are presumably presynaptic units[5] not responding to L-glutamate administration, the percentage of influenced units is likely to be considerably higher.

Acetylcholine acted only on a few neurones. The effect occurred after a relatively long latency of 15–50 sec, and when the microelectrophoretic current ceased, the activity change persisted for 10–50 sec. Two units localized in the dorsal column nuclei were inhibited by acetylcholine, one of these responded to peripheral stimulation. Three cells that had not been influenced by peripheral stimulation were activated by acetylcholine. These sparing effects suggest that the relay cells of the dorsal column are not themselves activated by cholinergic nerve transmission. The few acetylcholine-sensitive units may be related to other fibre systems which interact with the dorsal column system at the level of the nuclei gracilis and cuneatus (e.g. collaterals of the pyramidal tract, etc.).

Dopamine inhibited a majority of cells (8 of 11) responding to peripheral stimulation and only a minority of cells (3 of 13) that were indifferent to peripheral stimulation. The inhibitory effect occurred after a latency of 0.5–4.0 sec and vanished after the same delay. Dopamine consistently antagonized the facilitatory effect of L-glutamate.

Zusammenfassung. Von 250 Neuronen im Gebiet des Nucleus cuneatus und des Nucleus gracilis der Katze wurden 132 durch periphere physiologische Reizung aktiviert und 6 gehemmt. Mit Hilfe der Mikroelektrophorese wurden die folgenden Substanzen in die unmittelbare Nähe dieser Neurone gebracht: L-Glutaminsäure aktivierte 50% der Neurone. Acetylcholin war nur an wenigen Zellen im Kerngebiet wirksam, von diesen wurden 2 gehemmt und 3 aktiviert. Dopamin hemmte die Mehrzahl der durch periphere Reizung beeinflussbaren Neurone.

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August 2, 1965.

[3] D. R. Curtis, Brit. med. Bull. *21*, 5 (1965).
[4] P. B. Bradley and J. H. Wolstencroft, Brit. med. Bull. *21*, 15 (1965).
[5] D. L. Winter, J. Neurophysiol. *28*, 48 (1965).

Storage Function and Amine Levels of the Adrenal Medullary Granules at Various Intervals after Prenylamine Treatment

Independent work by Hillarp et al.[1] and Kirshner[2,3] has shown that the amine granules of the adrenal medulla are able to take up and concentrate monoamines *in vitro* by a Mg^{++}-ATP dependent storage mechanism. The storage mechanism is blocked by low concentrations of reserpine. In a second paper Hillarp et al.[4] analysed the uptake mechanism in further detail. Apart from reserpine, prenylamine [N-(diphenylpropyl)-amphetamine] proved to be the most potent inhibitor of the uptake mechanism. In higher, but still very low, concentrations it caused complete release of the granule amines. Similar effects were found in adrenergic nerve granules[5,6].

In a previous paper the storage function of the adrenal medullary granules was studied at various intervals after reserpine treatment[7]. In the present work adrenal medullary granules were examined in essentially the same way at different intervals following injection of a single dose of prenylamine to rabbits.

Methods. Rabbits weighing about 1.5 kg were injected with prenylamine (5 mg/kg) intravenously. At different intervals following injection (1–12 h) the rabbits were sacrificed by an injection of air intravenously. The adrenals were immediately removed and chilled with ice. The medulla with some adhering cortical tissue was rapidly dissected and homogenized with a loose-fitting plastic pestle for about 20 sec in 7 ml of 0.3 *M* sucrose. To remove unbroken tissues and cells, but at the same time to prevent loss of amine granules, the homogenate was centrifuged at 800 *g* for 5 min. The supernatant was centrifuged at 20,000 *g* for 20 min. The sediment was suspended in 0.5 ml 0.3 *M* sucrose. The granule suspension was transferred to 1.0 ml of an incubation mixture (at 0°C) containing 0.31 *M* glycyl-glycine (pH 7.3 with NaOH), 0.0025 *M* ATP and $MgCl_2$, 25 μg unlabelled adrenaline, 4.5 μg C^{14}-labelled adrenaline.

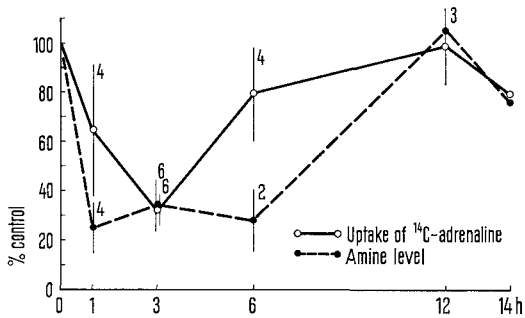
Incubation was performed without shaking at 31°C for 30 min, after which the suspension was chilled to 0°C, diluted 30 times with cold 0.5 *M* sucrose and – after about 1 h at 0°C – centrifuged at 74,000 *g* for 30 min. After thorough rinsing of the tubes with 0.5 *M* sucrose, the granule sediment was extracted with 5.0 ml of 0.01 *N* HCl in 98% ethyl alcohol. The catecholamine content of the extracts was determined spectrophotofluorimetrically[8]. The C^{14}-amine content was determined directly in a liquid scintillation counter. Pure nucleotides from the Pabst laboratories and pure prenylamine-lactate (Segontin) generously supplied by Hoechst, Frankfurt a. M. were used. *dl*-Adrenaline-7-C^{14} was purchased from Commis-

[1] A. Carlsson, N.-Å. Hillarp, and B. Waldeck, Med. exp. *6*, 47 (1962).
[2] N. Kirshner, Science *135*, 107 (1962).
[3] N. Kirshner, J. biol. Chem. *237*, 2311 (1962).
[4] A. Carlsson, N.-Å. Hillarp, and B. Waldeck, Acta physiol. scand. *59*, Suppl. 215 (1963).
[5] U. S. v. Euler, L. Stjärne, and F. Lishajko, Life Sci. *3*, 35 (1964).
[6] U. S. v. Euler, L. Stjärne, and F. Lishajko, Life Sci. *11*, 878 (1963).
[7] P. Lundborg, Exper. *19*, 479 (1963).
[8] Å. Bertler, A. Carlsson, and E. Rosengren, Acta physiol. scand. *44*, 273 (1958).

sariat de l'Energie Atomique, France, and stock solutions in 0.01 *N* HCl were stored at −30°C.

Results. In each experiment one or two prenylamine-treated and one control animal were used. In each experiment the amount of incorporated C^{14}-amines after prenylamine injection is given in % of the control value. The amine levels of the granules are also given in % of the control values.

Prenylamine caused a pronounced blockade of the uptake of adrenaline by the storage granules (Figure). The effect was observed 1 and 3 h after the injection. After 6–12 h the incorporation was restored. Also there was a pronounced reduction of catecholamine levels in the medullary granules. 3 h after prenylamine treatment the amine content was only 25% of normal. After 12 h the amine levels of the granules are restored.



Adrenaline level and uptake of C^{14}-adrenaline by adrenal medullary granules in vitro at various intervals following injection of prenylamine (5 mg/kg) i.v. in rabbits. The bars indicate s.e.m. and the figures the numbers of experiments.

Prenylamine thus resembles reserpine in blocking the storage function of the adrenal medullary granules not only when added in vitro but also after an intravenous injection of the drug to rabbits. The effect is not as long-lasting as after reserpine. After reserpine the storage function was restored within 48 h while the amine levels remained very low for a much longer time. After prenylamine both storage function and amine levels of the granules were rapidly restored. Experiments are in progress to study the effects in further detail[9].

Zusammenfassung. Es wurde der Speichermechanismus der Amingranula im Nebennierenmark von Kaninchen, in verschiedenen Intervallen, nach Injektion einer Dosis Prenylamin (5 mg/kg intravenös) untersucht. Prenylamin zeigt dabei einen reserpin-ähnlichen Effekt auf Nebennierenmarkgranula nicht nur in vitro, sondern auch nach intravenöser Injektion.

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Vestibular Origin of the Rapid Eye Movements During Desynchronized Sleep[1]

The neuronal mechanisms which are responsible for the appearance of the rapid eye movements (REM) during desynchronized sleep have been analysed. The importance of these REM, which have been described both in animals[2] and man[3], is stressed by the fact that in human sleep they are related to the visual content of dreams[3]. Attention was concentrated on the vestibular complex because (a) single vestibular neurones recorded from unrestrained, unanaesthetized cats during desynchronized sleep show bursts of rapid discharge associated with the REM[4], and (b) the second-order vestibular neurones control the oculomotor activity[5].

Methods. The experiments were performed on 15 unrestrained, unanaesthetized cats. Electrodes for recording electroencephalographic activity, the cervical electromyogram, and eye movements (electro-oculogram) were permanently implanted following a technique which has been previously described[6]. Electrolytic lesions of the vestibular nuclei were made using electrodes oriented with the Horsley-Clarke stereotaxic apparatus. The electrodes were inclined 30 degrees from the vertical axis, thus avoiding the bone of the tentorium. Recording sessions were made before and after the vestibular lesions.

Results. During the desynchronized episodes of sleep, two types of ocular movements were observed in the normal animals: (a) the well-known bursts of REM, which were binocularly synchronous, conjugate and grouped in clusters, and (b) slower, non-conjugate movements, which were present sporadically during the periods of quiescence intervening between REM. Occasionally isolated jerks of both eyes occurred. These phenomena were controlled for several days prior to making the lesions.

Bilateral lesions of the vestibular nuclei did not prevent the normal rhythm of sleep and wakefulness, nor did they substantially alter the phases of synchronized and desynchronized sleep.

The most remarkable change was the complete abolition of the bursts of REM typical of desynchronized sleep. Only slow ocular movements could be detected after such lesions. At times rare and isolated jerks of the eyes were observed. The phase of deep sleep in these

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[2] W. DEMENT, EEG clin. Neurophysiol. *10*, 291 (1958).

[3] W. DEMENT and N. KLEITMAN, EEG clin. Neurophysiol. *9*, 673 (1957). – E. ASERINSKY and N. KLEITMAN, Science *118*, 273 (1953); J. appl. Physiol. *8*, 1 (1955).

[4] E. BIZZI, O. POMPEIANO, and I. SOMOGYI, Science *145*, 414 (1964); Arch. ital. Biol. *102*, 308 (1964).

[5] A. BRODAL, O. POMPEIANO, and F. WALBERG, *The Vestibular Nuclei and their Connections. Anatomy and Functional Correlations* (Oliver and Boyd, Edinburgh 1962), p. 193.

[6] O. POMPEIANO and J. E. SWETT, Arch. ital. Biol. *100*, 311 (1962).