so long as the acetylcholine administration was continued. On discontinuing the acetylcholine perfusion the duration of the atrial potential gradually rose to its original value.

Consequently we have come to the conclusion that shortening of the duration of the auricular potential during vagal stimulation does not depend upon the rate of the atrial contractions.

We are indebted to the Dutch Organization for Pure Scientific Research for its financial support.

P. A. Biersteker
J. Th. F. Boeles
L. N. Bouman

Department of Physiology,
University of Amsterdam.

[1] Vaughan Williams, E. M., *J. Physiol.*, **147**, 325 (1959).
[2] Burgen, A. S. V., and Terroux, K. G., *J. Physiol.*, **120**, 449 (1953).

Effect of Methylpentynol on Acetylcholine in the Rat's Brain

Recent observations that methylpentynol, a sedative-hypnotic agent, interferes with transmission in certain cholinergic synapses[1], have led to the demonstration by Marley and Paton that this action is due in part to a reduction in the output of acetylcholine, at least from the superior cervical ganglion of the cat[2]. In the course of work in this and other laboratories on the influence of a number of appropriate neuro-pharmacological agents on the acetylcholine content of the rat's brain, it became apparent that the administration of depressants of the central nervous system in general is followed by a rise in brain acetylcholine (unpublished work and ref. 3). It seemed essential, therefore, for us to include methylpentynol in our survey, because this central-depressant might, in the light of the findings of Marley and Paton, depress the output of acetylcholine by the brain, possibly leading to a depression of the total level of acetylcholine. On the other hand, if release of acetylcholine alone, and not its synthesis, were interfered with, one might expect an accumulation and an increase in the amount of acetylcholine in the brain.

Adult male rats (150–250 gm.) were given intraperitoneal injections of methylpentynol in doses varying from 200 to 500 mgm./kgm. At a time shortly after loss of the righting reflex, when the animals were believed to be at the point of greatest central depression, the rats were decapitated, their whole brains (without olfactory lobes and pituitaries) quickly removed and extracted for acetylcholine by the method of Smallman and Fisher[4]. The content of acetylcholine in these extracts was determined by bioassay on the frog's rectus abdominis muscle preparation. Methylpentynol has no direct effect on this preparation in concentrations up to 20 μgm./ml. (approximately seven times the maximal amount expected in the extracts), nor does it sensitize the muscle to acetylcholine. The results are shown in Table 1.

Table 1. Effect of Methylpentynol on Acetylcholine Content of the Rat's Brain

Intraperitoneal dose (mgm./kgm.)	Killed (min. after onset of anæsthesia)	Acetylcholine in brain (μgm./gm. ± S.E.)
None (2 animals)	—	2·77
200 + 100	4	3·03
300	3	3·19
300	2·8	2·80
300	2·5	3·15
		Mean 3·04 ± 0·17
None (6 animals)	—	3·05 ± 0·23
500	12	5·88
500	15	5·08
500	12	5·83
500	15	4·55
500	12	4·60
500	12	5·10
500	12	5·00
500	12	5·66
		Mean 5·20 ± 0·52

The animals given the lower doses were all lightly depressed, and their acetylcholine-levels were only slightly higher than those of the two control animals. On the other hand, those animals given 500 mgm. of methylpentynol per kgm. were deeply depressed and the levels of acetylcholine in the brain were considerably higher than those of the control animals. It appears that, in rats, methylpentynol causes a rise in the level of acetylcholine in the brain, and that the extent of this rise is related to the degree of depression induced by the administration of the agent. If methylpentynol causes a rise in brain acetylcholine by reducing the output of that substance at certain central synapses, these results would indicate that there is no interference with the synthesis of acetylcholine. It has already been established that methylpentynol does not inhibit cholinesterase (unpublished work).

Since preparing this communication, we have conducted experiments to show that there is no difference in the synthesis of acetylcholine by slices of cerebral cortex from brains of untreated rats and by those from brains of rats under anæsthesia induced by methylpentynol (in a dose of 500 mgm./kgm.).

This work was aided by grant *B*–940 from the National Institute of Neurological Diseases and Blindness, U.S. Public Health Service.

Giancarlo Pepeu
Nicholas J. Giarman

Department of Pharmacology,
Yale University School of Medicine,
New Haven, Conn.

[1] Nicholls, J. G., and Quilliam, J. P., *Brit. J. Pharmacol.*, **11**, 151 (1956). Quilliam, J. P., *Med. Press*, **238**, 121 (1957); and *Brit. J. Pharmacol.*, **14**, 277 (1959). Marley, E., *ibid.*, **14**, 284 (1959).
[2] Marley, E., and Paton, W. D. M., *Brit. J. Pharmacol.*, **14**, 303 (1959).
[3] Feldberg, W., in "Metabolism of the Nervous System", 493, edit. by Richter, D. (London and New York, Pergamon Press, 1957).
[4] Smallman, B. N., and Fisher, R. W., *Can. J. Biochem. and Physiol.*, **36**, 575 (1958).

Effect of Severing the Olfactory Bulbs on the Intake and Excretion of Water in the Rat

Ecologists and experimental workers maintain that the sense of smell plays an important part both in the search for food and the regulation of alimentary reactions[1–3]. In recent investigations we studied the significance of olfactory signalization for the intake and excretion of water.

Adult male rats in groups of eight or more were used. The skull was trephined above the olfactory bulbs and the bulbs severed with a scalpel: the experiments were begun after a period of 1–2 months. Control animals were sham operated. During the experiment animals were housed individually in metabolism cages, and the intake and excretion of water together with food intake and body-weight were determined.