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The effect of oxotremorine on the acetylcholine content of different parts of cat brain

Tremorine (Pepeu, 1963; Holmstedt, Lundgren & Sundwall, 1963) and also its active metabolite oxotremorine (Holmstedt & Lundgren, 1966) cause an increase of brain acetylcholine and tremor in rats. A causal relation between the two effects was proposed (Holmstedt & Lundgren, 1966) but a number of substantial objections have been recently presented (Cox & Potkonjak, 1969a).

We have now investigated whether oxotremorine also increases brain acetylcholine in rats and whether the increase occurs uniformly in all brain regions.

Of eight young cats, each weighing about 1 kg, four received an intraperitoneal injection of saline and four 1·0 mg/kg of oxotremorine. Within a few minutes of giving the drug the cats showed intense tremor, salivation, miosis and behaviour similar to false rage. Fifteen min after the injection the cats were killed under light halothane anaesthesia, the skull opened, the brain removed and placed on ice. From each brain 3 samples were prepared: (I) about 500 mg of cortex were excised from the frontal lobes; (II) the diencephalon and the upper part of the midbrain were dissected following the lateral ventricles, the head of the caudate nucleus and a plane from the posterior colliculi to the rostral border of the pons. The weight of this sample was about 2·0 g; (III) the caudal part of the brain stem including the pons and the medulla oblongata; its weight was about 1·5 g.

Acetylcholine was extracted by the method of Smallman & Fisher (1958) modified by Bartolini & Bedarida-Jarach (1965), and assayed on the dorsal muscle of the leech. Recovery of added acetylcholine was 90%.

The results are reported in Table 1. The values of acetylcholine content found in the control cats are in good agreement with previous observations (Macintosh, 1941; Pepeu, 1966).

Table 1. *The influence of oxotremorine* (1 *mg/kg, i.p.*) *on the acetylcholine content of the cat cortex* (*I*), *diencephalon and upper part of midbrain* (*II*) *and the caudal part of the brain stem* (*III*). (Means ± s.e. of four experiments)

	Acetylcholine (μg/g)			
	Controls	After oxotremorine	% Increase	*P*
I	1·12 ± 0·26	1·43 ± 0·33	27	N.S.
II	2·53 ± 0·28	4·79 ± 0·49	89	<0·01
III ..	3·32 ± 0·61	3·45 ± 0·58	4	N.S.

It appears that oxotremorine increases the content of brain acetylcholine in the cat as it did in rats, but the increase is limited to some parts of the brain. There is an almost two fold rise of concentration in sample II which includes the diencephalon and part of the midbrain, a small but not significant rise in the cortex and no change in the lower brain stem.

The mechanism of action is not yet known. An effect on cholinesterases and on cholinoacetylase seems to be excluded (Holmstedt, Lundgren & others, 1965) and the hypothesis that the drug might act by mobilizing acetylcholine from an otherwise undetected store has been proposed (Holmstedt, 1967). Oxotremorine is known to cause a rise in total acetylcholine also when added to a brain homogenate (Lundgren & Malberg, 1968).

The largest increase in acetylcholine content occurs in the region which includes the caudate nucleus, the substantia nigra and the globus pallidus. Direct injections of tremorine in these nuclei have been shown to produce tremor in the rat (Cox & Potkonjak, 1969b). Before ruling out a causal relation between the effect of oxotremorine on acetylcholine content and the onset of tremor, the possibility of a rapid and strictly localized rise of acetylcholine level in the areas of the extrapyramidal system involved in the control of movement and posture should be excluded.

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