Paradoxical thermoregulatory behaviour in rats induced by (+)-amphetamine: blockade by α-adrenoceptor or dopamine receptor blocking agents

The administration of (+)-amphetamine (5–15 mg kg^{-1}, i.p.) causes marked hypothermia among rats kept at ambient temperatures of 5–15° (Yehuda & Wurtman, 1972a). That this effect is mediated by the release of dopamine in its limbic projections is suggested by the similar effects produced by drugs known to stimulate dopamine receptors (e.g. apomorphine and ET-495), the blockade of the (+)-amphetamine effect by pretreatment with drugs that block dopamine receptors (e.g. pimozide and haloperidol) and the absence of the effect in rats previously subjected to bilateral lesions of the olfactory tubercules (Yehuda & Wurtman, 1972b).

Several types of experimentally induced behaviours that follow (+)-amphetamine administration have also been shown to be mediated by central dopaminergic neurons. These include the stereotypy induced by large doses of the drug (Randrup & Munkvad, 1970), and the rotational behaviour observed in rats with unilateral lesions of the nigro-striatal tract (Ungerstedt, 1971).

We have previously observed that (+)-amphetamine also interferes with normal behavioural thermoregulation: when (+)-amphetamine-treated rats are placed in a temperature-gradient apparatus at an environmental temperature of 4°, they elect to locate themselves far away from the end exposed to the rays emitted by a heat lamp (despite their hypothermia). Similarly, when the environmental temperature is raised to 30°, the animals paradoxically place themselves under or near to the beam of heat (despite their hyperthermia). Stimulants of dopamine receptors mimic, and dopamine receptor blocking agents block, this paradoxical thermoregulatory behaviour.

The administration to rats of propranolol or phenoxybenzamine, two drugs that block peripheral adrenoceptors, causes hypothermia among rats kept at 4°, and enhances the hypothermia that followed (+)-amphetamine administration (Yehuda & Wurtman, 1972b). Hence, we examined the effects of these drugs on (+)-amphetamine-induced paradoxical thermoregulatory behaviour. To our surprise, the α-adrenoceptor blocking agent, phenoxybenzamine, like haloperidol or pimozide, was found to block the amphetamine-induced change in thermoregulatory behaviour (Table 1).

Table 1. *Response of rats to various drugs, with and without amphetamine, to a heat source.* The results are expressed as the score the animals registered on treatment as described in the text. Results are means of 6 animals ± s.d.

	Drug alone Heat lamp		Drug+(+)-amphetamine Heat lamp	
Treatment (mg kg^{-1}, i.p.)	Off	On	Off	On
Control	176 ± 18	109 ± 50*	—	—
(+)-Amphetamine (15)	208 ± 23	286 ± 32†	—	—
Phenoxybenzamine (20)	90 ± 8	92 ± 11	210 ± 43	109 ± 35*
Hydergine (15)	258 ± 49	95 ± 29*	244 ± 55	107 ± 33*
Phentolamine (2)	275 ± 45	58 ± 12*	174 ± 60	114 ± 75*
Tolazoline (4)	320 ± 110	201 ± 95*	189 ± 83	144 ± 50*
BE-2254 (2·5)	220 ± 40	90 ± 22*	173 ± 60	45 ± 12*

* $P < 0.01$ statistically-significant movement of animals towards heat lamp.
† $P < 0.01$ statistically-significant movement of animals away from heat lamp.

The effect of this drug, and of the others utilized in the present study, were examined as follows:

Immediately after injection with test drug, each rat of a group of 6 for each drug was placed on a long track (80 cm long, divided into sixteen 5 cm sections) inside an environmental chamber that had been pre-set at 4° and 45% relative humidity. After 20 min, a heat lamp located over one end of the track was turned on. For 10 min during this period, observations were made at 20 s intervals on the location of the rat on the track. Similar observations were also made during the 8 min immediately before the time that the heat lamp had been turned on. The positional score for each animal was calculated as the sum of the section numbers of the 24 observations made during the first or second test period. A low score indicated that the rat had elected to locate itself near the heat lamp. Other rats injected with test drug were kept in a small cage at 4° for 30 min and then injected with 15 mg kg^{-1} (+)-amphetamine; subsequently they were placed onto the track and the positional score determined as described above.

As anticipated, control animals placed in an environment of 4° moved towards the heat lamp, while (+)-amphetamine-treated rats moved in the opposite direction (Table 1). Pretreatment of the rats with phenoxybenzamine blocked this movement away from the heat lamp. Phenoxybenzamine may affect other brain receptors besides those for noradrenaline, i.e. 5-hydroxytryptamine (5-HT) (Nickerson & Hollenberg, 1967) and dopamine (York, 1967; McLennan & York, 1967). To determine whether phenoxybenzamine blocked the amphetamine-induced paradoxical thermoregulatory behaviour by actions on noradrenaline, dopamine, or 5-HT synapses, we compared its effects with those of other α-adrenoceptor blocking agents.

The administration of BE-2254 (HEAT, 2-[β(4-hydroxyphenyl)-ethyl-aminomethyl]-tetralone), like phenoxybenzamine, blocked the amphetamine-induced paradoxical behaviour (Table 1). By itself, it did not affect thermoregulatory behaviour, but did produce hypothermia among rats kept at 4°. Since this drug apparently has no effect on 5-HT receptors (Baumgarten, Gothert & others, 1972), we conclude that the blockade of (+)-amphetamine-induced paradoxical thermoregulatory behaviour by α-adrenoceptor blocking agents is not mediated by their effects on 5-HT receptors. All other α-adrenoceptor blocking agents tested also blocked the paradoxical thermoregulated behaviour seen in (+)-amphetamine-treated animals (Table 1).

The possible effects of α-adrenoceptor blockers, such as phenoxybenzamine and BE-2254, on dopaminergic synapses were tested in two experimental behaviour situations, stereotypy and rotational behaviour. Neither drug blocked the stereotyped behaviour induced by either amphetamine or apomorphine. Similarly, the two drugs blocked rotation toward the lesioned side observed after rats with unilateral caudate lesions were given (+)-amphetamine (Ungerstedt, 1971).

These findings indicate that α-blocking agents do not interfere with central dopaminergic synapses; hence their blockade of paradoxical thermoregulatory behaviour (+)-amphetamine-treated rats is not explained by dopamine-receptor blocking actions.

Our findings are best explained by postulating that noradrenergic synapses exist in the pathway which mediates the amphetamine-induced paradoxical behaviour, and that the flow of signals across the synapses is blocked by α-adrenoceptor blocking agents.

These studies were supported in part by a grant from the United States Public Health Service.

The drugs were donated by: Smith, Kline & French ((+)-amphetamine and phenoxybenzamine); Sandoz (Hydergine), CIBA (phentolamine and tolazoline) and by Beiersodorf (BE-2254).

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August 28, 1973

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LETTERS TO THE EDITOR

Do adrenergic fibres have muscarinic inhibitory receptors?

Lindmar, Löffelholz & Muscholl in 1968 put forward the view that adrenergic fibres have receptors with which muscarinic substances combine to inhibit the release of noradrenaline. The original observation which suggested the idea was that of Hoffmann, Hoffmann & others (1945) who found that when a rabbit isolated heart was perfused with fluid containing atropine, then acetylcholine injected into the aortic cannula caused an increase in the rate and force of the heart beat, and liberation of an adrenaline-like substance (later shown to be noradrenaline) in the outflow. Muscholl therefore conceived that the adrenergic fibres might possess receptors which were stimulated by acetylcholine to inhibit the release of noradrenaline and only when these receptors were blocked by atropine was acetylcholine able to release noradrenaline. Since atropine blocked only muscarinic but not nicotinic receptors, he supposed that the inhibitory receptors were muscarinic.

The proposed inhibitory receptors. The use of the terms "muscarinic" and "nicotinic" to distinguish between receptors raises difficulties. Thus Muscholl considers pilocarpine to be a muscarinic substance, but Dale & Laidlaw (1912) showed that pilocarpine, like nicotine, releases adrenaline from the adrenal gland. Moreover, when a 2% solution of pilocarpine nitrate was applied to the surface of the superior cervical ganglion, it caused a brief dilatation of the pupil, and a prolonged contraction of the nictitating membrane, again acting like nicotine. Then came the work of Ambache, Perry & Robertson (1956) which showed that the original conception of a "muscarinic receptor" required modification in view of the finding that muscarine itself had a nicotine action and could stimulate the perfused superior cervical ganglion when injected into the ganglion. This stimulation was effective in doses which in some experiments were as low as 0·1 μg. Muscarine therefore has "nicotinic" as well as "muscarine" properties, though its action on the ganglion is reversibly blocked by atropine. Now acetylcholine also stimulates the perfused ganglion but its action is not blocked by atropine. We have then acetylcholine, pilocarpine and muscarine which have both muscarinic and nicotinic properties. It seems likely that if metha-