Effect of amphetamine, 3,4-methylenedioxyamphetamine, *p*-methoxyamphetamine and related amphetamines on uptake of metaraminol and efflux of noradrenaline in adrenergic nerves of rabbit atria

Noradrenaline and related amines, e.g. metaraminol, are transported in adrenergic nerves by a carrier-mediated process that is inhibited by a number of phenethylamine derivatives including amphetamine (Iversen, 1967). The efflux of noradrenaline from the cytosol of adrenergic nerves also appears to occur by a cocaine-sensitive, carrier mediated process (Paton, 1973a). Some phenethylamine and tryptamine derivatives accelerated the efflux of [^{3}H]noradrenaline, the most potent compounds studied β-phenethylamine and amphetamine (Paton, 1973b, c; Paton & Pasternak, 1974).

Both 3,4-methylenedioxyamphetamine (MDA) and *p*-methoxyamphetamine (PMA) are potent drugs of abuse that have been responsible for a number of deaths (Thiessen & Cook, 1973; Cimbura, 1974). The aim of the present study was to determine the effects of these agents and of related compounds on the uptake and efflux of catecholamines in peripheral adrenergic nerves.

Pieces of rabbit atria were prepared as described previously (Paton, 1973a) and preincubated at 37° for 30 min in a physiological salt solution. [^{3}H]Metaraminol ($2{\cdot}5 \times 10^{-8}$ M) was then added to the media and the incubation continued for a further 30 min. At the end of this period, the total [^{3}H] content of tissues was determined by liquid scintillation spectrometry as described previously (Paton, 1973a). The net accumulation of [^{3}H]metaraminol was expressed as pmol g^{-1} wet weight. The medium used had the following composition (mM): NaCl, 140; KCl, 5; $CaCl_2$, 1·5; Mg SO_4, 1·2; tris HCl (pH 7·4), 10; Na_2 EDTA, 0·03; Na ascorbate, 0·1. The medium was equilibrated with 100% O_2 and contained 2·5 mM D-glucose.

As described previously (Paton, 1973a), atria, from reserpine-pretreated rabbits, were exposed to pargyline (5×10^{-4} M for 30 min) and tropolone (1×10^{-4} M throughout), and thereafter to $5{\cdot}8 \times 10^{-7}$ M [^{3}H](−)-noradrenaline for 60 min. Tissues were then blotted, placed on fine metal hooks and transferred to fresh media at 37°. Drugs (10^{-5} M) were added between 60–90 min of efflux because, during this period, efflux occurs predominantly from adrenergic nerves (Paton, 1973a). At 90 min tissues were removed, blotted and their [^{3}H] content determined as described above. The amount of [^{3}H](−)-noradrenaline remaining in the tissue (in pmol g^{-1}) was then determined.

It can be seen (Table 1) that, at 10^{-5} M, both (+)- and (−)-amphetamine were potent inhibitors of the uptake of [^{3}H] (±)-metaraminol. MDA and PMA also produced very significant inhibition of uptake, MDA being slightly more potent. By contrast, the dimethoxyamphetamine derivatives studied were much weaker inhibitors of uptake, only the 2,3-, 2,4- and 3,5-derivatives producing significant inhibition of uptake but all were less potent than amphetamine, MDA and PMA.

At 10^{-5} M, (+)- and (−)-amphetamine and MDA all caused considerable acceleration of the efflux of [^{3}H]noradrenaline from rabbit atria and were approximately equipotent. PMA proved to be less potent. The dimethoxyamphetamine derivatives did not alter the efflux of [^{3}H](−)-noradrenaline significantly (Table 1).

Previous studies had demonstrated that phenolic methoxy substitutions had an inhibitory effect on the ability of amines to accelerate [^{3}H]noradrenaline efflux and this was true of both tryptamine (Paton, 1973c) and phenethylamine derivatives (Paton & Pasternak, 1974). Substitution of two phenolic methoxy groups was markedly inhibitory for both amphetamine (present study) and phenethylamine

Table 1. *Effect of methoxylated amphetamine derivatives on the accumulation of [^{3}H]metaraminol, and the residual content of [^{3}H]noradrenaline after efflux.* All drugs were present at 10^{-5} M for 30 min during [^{3}H]metaraminol accumulation and from 60–90 min of efflux of [^{3}H]noradrenaline. Values are the mean ± s.e. of 5–12 observations. * $P<0.05$.

Compound	[^{3}H]metaraminol accumulation (pmol g^{-1} wet wt)	Residual [^{3}H]noradrenaline content (pmol g^{-1} wet wt)
	239±12	2783±245
(+)-Amphetamine	82±17*	1197±173*
(—)-Amphetamine	89±17*	1231±157*
PMA	113±14*	1734±206*
MDA	91±19*	1238±143*
2,3-DMA	195±11*	2068±303
2,4-DMA	159±11*	2951±245
2,5-DMA	221±27	3172±679
2,6-DMA	193±28	2766±350
3,4-DMA	204±20	3012±431
3,5-DMA	167±9	2743±378

derivatives (Paton & Pasternak, 1974). However, a single methoxy substitution at the *para* position resulted in potent phenethylamine (Paton & Pasternak, 1974) and amphetamine derivatives (present study). It is also noteworthy how potent MDA proved to be both with respect to inhibition of metaraminol uptake and acceleration of noradrenaline efflux.

These studies have thus demonstrated that both MDA and PMA inhibit the uptake process for noradrenaline in, and accelerate its efflux from, peripheral adrenergic neurons. These findings are in keeping with our recent demonstration that both agents act as indirectly acting sympathomimetic amines (Bell, Yee & others, 1974), and with certain of the clinical features of overdosage which appear to result from catecholamine release (Thiessen & Cook, 1973; Cimbura, 1974). It remains to be established whether MDA and PMA are themselves transported into adrenergic neurons via the membrane carrier system for noradrenaline. Certainly evidence for this in relation to amphetamine is far from conclusive (Paton, 1974).

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