37°C. Agonist-antagonist interactions were examined using the method of Arunlakshana & Schild (1959). Control preparations were used in each experiment so that allowances for changes in sensitivity could be made.

In the aortic strip both noradrenaline (NA) and 5HT caused contractions in the same concentration range ($1.0 \times 10^{-8} - 2.5 \times 10^{-5}$ mol/l). In the ear artery NA ($1.0 \times 10^{-11} - 2.0 \times 10^{-9}$ mol) and 5-HT ($1.0 \times 10^{-8} - 2.5 \times 10^{-6}$ mol) produced vasoconstrictor responses at different doses. In the case of 5-HT the response was biphasic at submaximal doses. Methysergide ($1.0 \times 10^{-8} - 1.0 \times 10^{-6}$ mol) also produced biphasic contractile responses in the ear artery and was similar in potency to 5-HT. Both phases of the response to methysergide and 5-HT were still evident in arteries removed from animals treated with reserpine (1 mg/kg 18-24 h previously. The interactions between agonists and antagonists are summarized in Table 1.

The results suggest that in the aorta 5-HT and NA combine with different receptors whereas in the ear artery they appear to combine with the same receptor which can also be activated by methysergide. The profile of action of methysergide in these two preparations once again raises the question of whether its therapeutic action is mediated through 5-HT-receptor activation or antagonism.

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A comparison of the accumulation of noradrenaline and 5-hydroxytryptamine into arterial smooth muscle

P. BUCHAN*, A.J. LEWIS & M.F. SUGRUE

Department of Pharmacology, Organon Scientific Development Group, Newhouse, Lanarkshire ML1 5SH

5-Hydroxytryptamine (5-HT) is capable of being taken up into both central (Shaskin & Snyder, 1970) and peripheral (Thoa, Eccleston & Axelrod, 1969) adrenergic neurones. In addition to its neuronal uptake, noradrenaline (NA) is extraneuronally accumulated (Gillespie, 1973). The objective of this study was to determine, first, if 5-HT like NA, is accumulated by arterial smooth muscle and secondly, if the accumulation characteristics of 5-HT are similar to those of NA.

Isolated segments of rabbit ear artery were incubated with either NA or 5-HT in 5 ml of Krebs-bicarbonate medium containing ascorbic acid (0.2 mg/ml), EDTA (0.05 mg/ml) and pargyline (0.025 mg/ml) at 37°C under an atmosphere of 95% oxygen and 5% carbon dioxide. Incubation was terminated by removing and washing the tissue in ice-cold Krebs-bicarbonate for 20 minutes. The effect of drugs on amine accumulation was studied by pre-incubating the tissue for 30 min with drug prior to the addition of either NA or 5-HT (3×10^{-3} M for 10 minutes). Segments of artery were subjected to the fluorescence procedure of Falck, Hillarp, Thieme & Torp (1962) and the fluorescence in arterial smooth muscle due to amine was measured photometrically in 7 μm transerve sections.

Histochemical examination of segments of rabbit ear artery incubated with 5-HT revealed an intense yellow-green fluorescence in the smooth muscle cells thus indicating an accumulation of 5-HT. The accumulation characteristics of NA and 5-HT were similar. Above a threshold concentration (3×10^{-5} M) accumulation of NA and 5-HT was concentration-dependent up to 10^{-2} M. The amount of amine accumulated rose rapidly over the first 10 min of incubation and reached a maximum after 20 minutes. Amine fluorescence was decreased in arterial smooth muscle when the incubation was continued in amine-free medium at 37°C (washout $t\frac{1}{2}$ for both amines was 8-12 min) but not after incubation in ice-cold Krebs bicarbonate.

Accumulation of both NA and 5-HT into rabbit ear artery smooth muscle was decreased in a concentration-dependent manner by oestradiol., phenoxybenzamine and normetanephrine. The accumulation of 5-HT was less susceptible to inhibition by these drugs than that of NA. 5-HT accumulation was unaltered by methysergide (10^{-4} M). Following a pre-incubation period of

5 min, histamine (3×10^{-4} M to 3×10^{-3} M) produced a concentration-dependent inhibition of both NA and 5-HT accumulation.

Gillespie & Rae (1972) have shown that NA accumulation by the guinea-pig superior mesenteric artery is appreciably less than that observed with the rabbit ear artery. As in the case of NA, accumulation of 5-HT was markedly less in the guinea-pig superior mesenteric artery than in the rabbit ear artery.

The results of this study reveal that 5-HT is accumulated by arterial smooth muscle cells and that drugs which decrease NA accumulation also reduce the accumulation of 5-HT. Furthermore, both amines appear to exhibit the same species selectivity. Such findings, together with the observation that histamine blocks the accumulation of both NA and 5-HT, raise the possibility that amine accumulation by arterial smooth muscle may be mediated via a common mechanism.

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Some unexpected pharmacological effects of *p*-chlorophenylalanine methyl ester (PCPA) methyl ester)

E. MARLEY AND JENNIFER E. WHELAN*

Department of Pharmacology, Institute of Psychiatry, De Crespigny Park, London SE5 8AF

Effects *in vitro*: PCPA methyl ester contracted the rat's isolated stomach strip (RSS) and uterus, as did 5-hydroxytryptamine (5-HT), these effects were prevented by methysergide but not by mepyramine, hyoscine, phentolamine and propranolol. Cross-tachyphylaxis occurred between β-phenethylamine, dexamphetamine, substances acting on 5-HT receptors in the RSS (Vane, 1960) and PCPA methyl ester, and 5-HT. Contractions of the RSS to PCPA methyl ester were unaffected by changing bath pH from 7.0 to 2.0, reduced at pH 8.0 and abolished at pH 8.5. Pretreatment of rats with an L-aromatic decarboxylase inhibitor Ro4-4602 (N^1-(DL-seryl)-N^2(2,3,4-trihydroxybenzylhydrazine)), 335 μmol/kg, or amine oxidase inhibitors, did not affect response of the RSS to PCPA methyl ester. Contraction of the guinea-pig ileum to 5-HT was immediate, whereas that to PCPA methyl ester occurred only on wash-out to large doses (0.5 mmol/ml). Such contractions last 20 min, being partly dependent on neuronal integrity since they were diminished by tetrodotoxin (3.0 nmol/ml); wash-out contractions were still obtained after doses of acetylcholine (0.2 nmol/ml), that produce maximal membrane permeability changes (Burgen & Spero, 1968). Additionally, PCPA methyl ester (1.0mmol/ml) immediately abolished twitches and tetanus to coaxial excitation of the guinea-pig ileum and the contractions to acetylcholine, histamine, and 5-HT. PCPA methyl ester (6.0 μmol), like 5-HT (60.0 nmol), caused bronchoconstriction of guinea-pig isolated lungs but unlike 5-HT, the effects were not prevented by methysergide (6.0 μmol), nor by mepyramine, hyoscine, phentolamine and propranolol. Effects *in vivo*: Similar effects were obtained with guinea-pig lungs as *in vitro*. In cats, increases in total and free acidity of gastric secretion evoked by histamine (3.0 nmol kg^{-1} min^{-1} i.v.) were markedly reduced by PCPA methyl ester (0.6 μmol kg^{-1} min^{-1}) or 5-HT (2.6 nmol kg^{-1} min^{-1}) infused into the aorta; volume of gastric secretion was reduced to a lesser degree. 5-HT evokes adrenaline secretion in dogs (Eble, Gowdey & Vane, 1972). In abdominally eviscerated cats, 5-HT (0.06 μmol) or PCPA methyl ester (2.0 μmol) injected retrogradely into the ligated superior mesenteric artery, evoked adrenaline secretion from the adrenal medullae, effects prevented by methysergide (0.215 μmol). In chicks, PCPA methyl ester (10 μmol/100 g) induced behavioural and electrocortical sleep and a 1.0-1.5°C fall in body temperature, effects unaltered by methysergide; equimolar doses of *p*-chlorophenethylamine elicited arousal.

Results with PCPA methyl ester on the RSS, rat uterus and cat adrenal medullae, indicate a