Blood pressure and noradrenaline levels after treatment with α-methyldopa, α-methyldopamine and α-methyl-*m*-tyrosine

SIR,—An important factor in the depletion of tissue monoamines by α-methyldopa is a displacement of these amines by the metabolites α-methyldopamine or α-methylnoradrenaline, or both, which are formed by decarboxylation and, in the latter case, subsequent β-hydroxylation in analogy with the sympathetic transmitter noradrenaline (Carlsson & Lindqvist, 1962; Carlsson, 1964). The α-methylated amines are stored in the sympathetic neurons and released on stimulation of these nerves (Muscholl & Maître, 1963), acting as substitute or "false" transmitters. The activity of α-methylnoradrenaline on the adrenergic receptors is less than that of noradrenaline (Day & Rand, 1964; Brunner, Hedwall & others, 1966, 1967). Day & Rand (1963, 1964) reported that the sympathetic function was impaired after treatment with α-methyldopa and suggested this impairment to be the cause of the antihypertensive action of the drug.

If this is so, one would expect a relatively close time correlation of the effect on the blood pressure and the depletion of noradrenaline after α-methyldopa. A further implication of the false transmitter concept is that a parallellism should exist between the receptor activity of the assumed transmitter and the magnitude of the hypotensive response. The present investigations were made to evaluate these assumptions. Blood pressure and tissue monoamine levels have been examined in parallel following the administration of a single dose of α-methyldopa to rats. The effect of α-methyl-*m*-tyrosine on blood pressure has been compared with that of α-methyldopa; α-methyl-*m*-tyrosine also depletes noradrenaline from the stores through displacement by its decarboxylated amine products (Carlsson & Lindqvist, 1962) and these amines possess much less receptor activity than those arising from α-methyldopa (Brunner & others, 1966, 1967). Finally, one of the proposed false transmitters of α-methyldopa, α-methyldopamine has been tested for its effect on blood pressure and tissue monoamines in the rat.

Sprague-Dawley rats of either sex weighing 180–250 g were used. Renal hypertension was provoked by surgical removal of the left kidney and ligation of a branch of the right renal artery in young animals (age about 6 weeks). This procedure resulted in a slowly developing hypertension with mean arterial blood pressure values ranging from 150–190 mm Hg (mean 170 mm Hg, s.e.m. = 3·1, n = 23) 4–6 weeks after the operation. The mean arterial blood pressure in normal rats of corresponding age was 116 mm Hg (s.e.m. = 1·7, n = 32). Blood pressure was always recorded in unrestrained conscious animals through in-dwelling polyethylene catheters (Popovic & Popovic, 1960) connected to Statham P23Dc pressure transducers and recorded on a Grass Model 5 Polygraph. Basal values were obtained by continuous recording for 20–40 min 3–4 times daily for at least 2 days before drug administration. L-α-Methyldopa (200 or 400 mg/kg) and DL-α-methyl-*m*-tyrosine (400 mg/kg) were dissolved in saline and injected intraperitoneally. (±)-α-Methyldopamine HBr 25 mg/kg (calculated as the salt) was injected subcutaneously in divided doses at about 2 hr intervals. Rectal temperature was checked frequently and the hypothermia, occurring after α-methyldopa but not after α-methyl-*m*-tyrosine (unpublished observations), was prevented by keeping the treated animals at an ambient temperature of about 29°. For determinations of tissue monoamine levels, normotensive animals of the same age as the hypertensive rats were used. Preliminary observations have indicated that no significant differences seem to exist between the two groups in control monoamine levels in tissues.

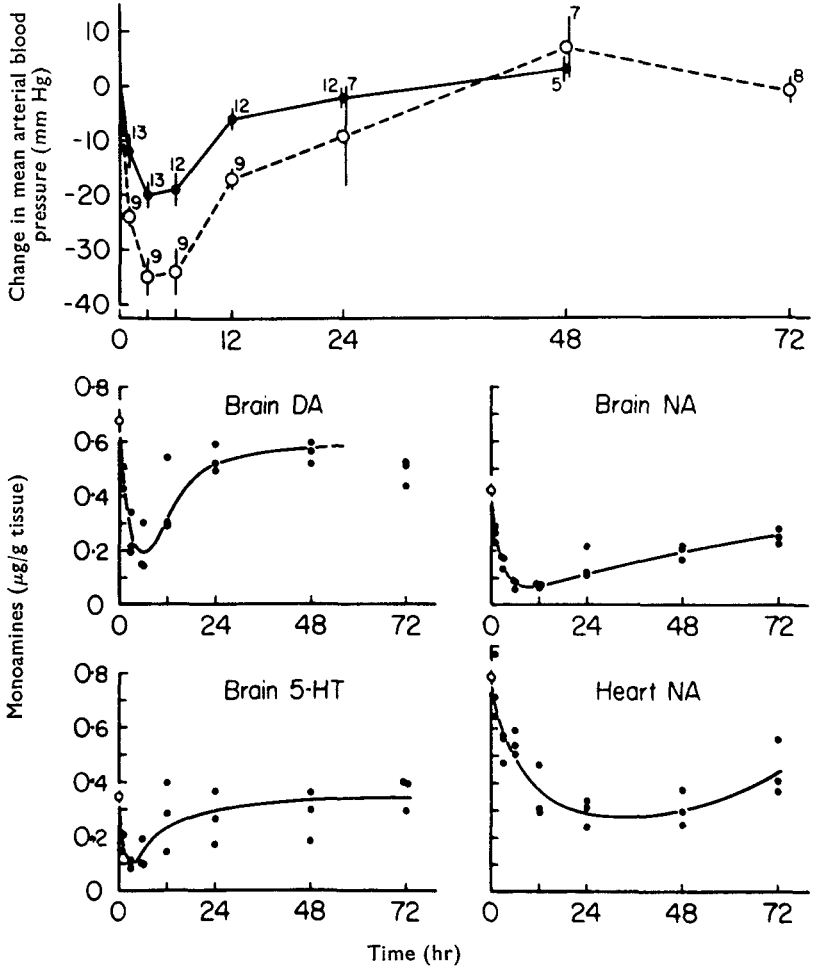

FIG. 1. Decrease in mean arterial blood pressure of conscious normotensive (●—●) and renal hypertensive (○- - - -○) rats (means ± s.e.m., number of experiments indicated by the small figures) and the levels of heart noradrenaline (NA) and brain noradrenaline, dopamine (DA) and 5-hydroxytryptamine (5-HT) following a single dose of L-α-methyldopa 400 mg/kg i.p. Normal blood pressure level of normotensive rats was 116 mm Hg (s.e.m. = 2·1, n = 14), and of hypertensive rats 168 mm Hg (s.e.m. = 5·3, n = 9). Normal amine concentrations: heart noradrenaline 0·79 μg/g (s.e.m. = 0·031, n = 7), brain noradrenaline 0·42 μg/g (s.e.m. = 0·020, n = 7), brain dopamine 0·68 μg/g (s.e.m. = 0·045, n = 6), brain 5-HT 0·34 μg/g (s.e.m. = 0·019, n = 7).

Noradrenaline was determined as described by Bertler, Carlsson & Rosengren (1958), dopamine and 5-hydroxytryptamine (5-HT) by the methods described by Carlsson & Lindqvist (1962) and Andén & Magnusson (1967), respectively.

As shown in Fig. 1, administration of α-methyldopa, 400 mg/kg, lowered the blood pressure of normotensive as well as hypertensive rats. The hypotensive effect appeared to be more pronounced in the latter group, although,

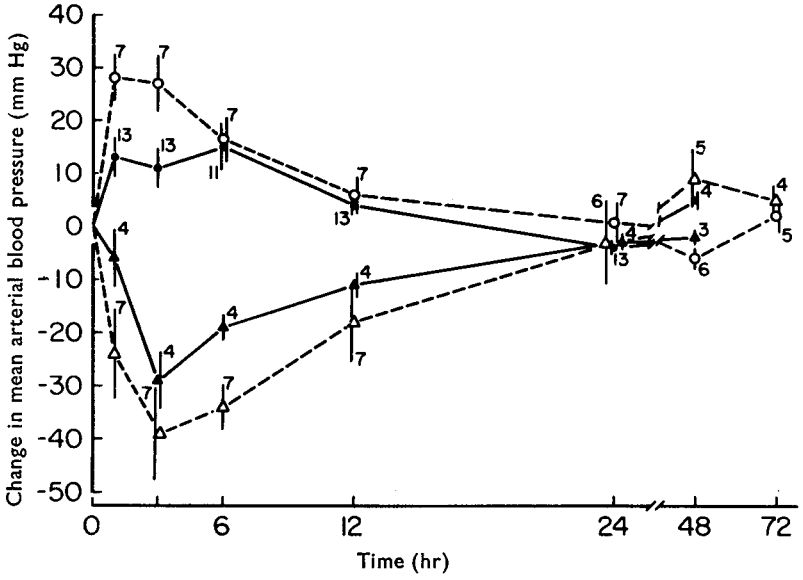

FIG. 2. Change in mean arterial blood pressure of conscious normotensive (solid symbols) and renal hypertensive (open symbols) rats (means ± s.e.m., number of experiments indicated by the small figures) after L-α-methyldopa 200 mg/kg i.p. (triangles) and DL-α-methyl-*m*-tyrosine 400 mg/kg i.p. (circles). Normal blood pressure levels of animals treated with α-methyldopa: normotensive 121 mm Hg (s.e.m. = 3·8, n = 4), hypertensive 171 mm Hg (s.e.m. = 5·9, n = 7); of animals treated with α-methyl-*m*-tyrosine: normotensive 114 mm Hg (s.e.m. = 3·0, n = 14), hypertensive 171 mm Hg (s.e.m. = 5·6, n = 7).

if calculated as % basal values, the response was about the same in the two groups. The effect occurred within 1 hr of the administration and was maximal after 3–6 hr. After 24 hr the blood pressure had returned almost to control values. During the period of blood pressure fall, the animals showed a tendency to develop hypothermia and were sedated.

The levels of monoamines in the brain were lowered by α-methyldopa 400 mg/kg (Fig. 1). Dopamine and 5-HT levels declined to a minimum of about 25% of the normal values after 3–6 hr and then increased to control values after 24 hr. The depletion of brain noradrenaline was slower in onset and longer lasting than that of dopamine and 5-HT. The lowest noradrenaline values were observed after 6–12 hr (about 15% of normal) and control values had not been reached after 72 hr, the depletion still amounting to about 50%. In the heart, maximal depletion, about 70%, was observed after 24–48 hr and the level at 72 hr was about 50% of the normal value.

A comparison between the effect of α-methyldopa on blood pressure and tissue monoamines (Fig. 1) shows that the former effect appeared at a time when there was only a small depletion of brain noradrenaline and an insignificant lowering of heart noradrenaline. The blood pressure had returned to normal values long before the noradrenaline stores were refilled.

Fig. 2 shows the effects of single doses of α-methyldopa (200 mg/kg of the L-form) and of α-methyl-*m*-tyrosine (400 mg/kg of the racemate) on the blood pressure of normotensive and renal hypertensive rats. Since only the L-isomers

of these amino-acids are decarboxylated *in vivo* (for references, see Holtz & Palm, 1966), these doses may be considered roughly equivalent. The hypotensive response of the two groups of rats to α-methyldopa in this dose was in all respects similar to that seen after the higher dose (Fig. 1). α-Methyl-*m*-tyrosine, surprisingly, did not lower the blood pressure in either group of animals for up to 72 hr after the administration; on the contrary, there was a tendency to an initial increase in blood pressure for the first 6–12 hr.

The degree of noradrenaline depletion from the tissues after α-methyldopa 200 mg/kg was nearly as great as that seen after 400 mg/kg (data not presented here). Since α-methyl-*m*-tyrosine seems to be more active than α-methyldopa in depleting catecholamines from the tissues (Porter, Totaro & Leiby, 1961; Udenfriend & Zaltzman-Nirenberg, 1963), it may be inferred that in the present study α-methyl-*m*-tyrosine produced at least the same degree of noradrenaline depletion as α-methyldopa, yet failed to lower the blood pressure.

The effect of α-methyldopamine was studied in normotensive rats receiving 25 mg/kg, s.c., daily for two subsequent days. The blood pressure before the treatment was 110 mm Hg (s.e.m. = 2·8, n = 9) and 16–18 hr after the last injection the blood pressure was 110 mm Hg (s.e.m. = 4·4, n = 9). After 40–42 hr the blood pressure was still within the control range (mean value 113 mm Hg, s.e.m. = 6·0, n = 9). In another series of rats, similarly treated, the heart noradrenaline content was reduced to 0·16 μg/g (s.e.m. = 0·010, n = 6) after 16–18 hr. This reduction was at least as great as that observed after α-methyldopa 400 mg/kg (Fig. 1). The brain noradrenaline was not changed. Apparently, then, depletion of the noradrenaline of peripheral tissues by α-methyldopamine is not accompanied by a reduction of the blood pressure. This may indicate that amine depletion in neurons within the central nervous system is important for the hypotensive effect of α-methyldopa. Also α-methyl-noradrenaline is devoid of blood pressure lowering properties (Henning, unpublished observations, Brunner & others, 1966, 1967).

Thus, after α-methyldopa the time correlation between the noradrenaline depletion and the decrease in blood pressure is poor, the latter effect being much shorter-lasting than the former. This observation is not easily reconciled with the false transmitter concept in its simplest outline. The failure of α-methyl-*m*-tyrosine to lower blood pressure is also difficult to explain assuming a transmitter function of the amine products of this amino-acid.

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Enteral absorption of hyoscine *N*-butylbromide

SIR,—In a recent paper, Herxheimer & Haefeli (1966) investigated the oral absorption of hyoscine *N*-butylbromide (Buscopan) in man using the appearance of atropine-like effects as a criterion of oral efficacy. Oral doses of the drug up to 600 mg (approximately 10 mg/kg) failed to produce the effects seen after parenteral injection and it was concluded that the drug was not absorbed from the gastrointestinal tract. However, the results of experiments comparing the toxicity of hyoscine *N*-butylbromide administered by different routes provides evidence for the enteral absorption of the compound.

Three similar groups of Sprague-Dawley rats (females), which had been fasted for the preceeding 12 hr, underwent laparatomy under ether anaesthesia, after which the drug or physiological saline was injected through a fine needle into the lumen of the stomach and duodenum. The operation wound was then sutured, and each animal injected subcutaneously with physiological saline or drug. One group of rats was given the drug subcutaneously, saline being injected into the stomach and duodenum. A second group was given the drug into the stomach, saline being injected subcutaneously and into the duodenum. In the third group of animals, the drug was given into the duodenum and saline was injected subcutaneously and into the stomach. By this experimental design, operative stress and number of injections per animal were evenly distributed throughout the groups. The animals were observed for the following 24 hr and the LD 50 obtained for each route of administration was calculated using the method of Litchfield & Wilcoxon (1949). The values were as follows: LD50 of hyoscine *N*-butylbromide (1) subcutaneously = 510 (386–673) mg/kg, S = 1·365; (2) instilled into the stomach = 1040 (897–1206) mg/kg, S = 1·130; (3) instilled into the duodenum = 180 (154–211) mg/kg, S = 1·195.

In the group of animals given the drug into the duodenum, convulsions started 1 to 2 min after injection and death occurred within 4 to 10 min. When the drug was given subcutaneously or injected into the stomach, the animals did not die for several hr. The drug given into the duodenum was 2·8 times more toxic than when given subcutaneously, which, together with the quick onset of effects, indicates its rapid and good absorption. The possibility that the lower toxicity of hyoscine *N*-butylbromide given into the stomach is caused by decreased gastric peristalsis and slowed passage of the drug to the site of absorption in the small intestine is now being investigated.

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