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CENTRAL INHIBITORY EFFECT OF α-METHYLDOPA ON BLOOD PRESSURE, HEART RATE AND BODY TEMPERATURE OF RENAL HYPERTENSIVE RATS

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The central inhibitory effect of α-methyldopa on blood pressure, heart rate and body temperature was studied in conscious renal hypertensive rats. Systemic administration of α-methyldopa decreased mean arterial blood pressure and body temperature and caused a short lasting increase in heart rate followed by a long lasting decrease. Inhibition of central decarboxylase activity prevented the decrease in blood pressure, heart rate and body temperature but not the initial increase in heart rate. Inhibition of peripheral decarboxylase activity blocked the increase in heart rate and partially reduced the decrease in heart rate and body temperature but did not affect the decrease in blood pressure. α-Methyldopa also decreased blood pressure at an ambient temperature of 30° C, but the decrease of body temperature was absent and the heart rate remained elevated for 7 hr. Similar results were obtained in normotensive rats. The decrease in heart rate was correlated with the decrease in body temperature in normotensive and renal hypertensive rats.

These findings suggest that in the renal hypertensive rat the decrease in blood pressure and in body temperature depends on a central action of α-methyldopa metabolites.

Dopa decarboxylase inhibition | α-Methyldopa | Body temperature
Central inhibitory effects | Heart rate | Blood pressure

1. Introduction

α-Methyldopa has a marked hypotensive action in renal hypertensive rats (Henning, 1967; Baum et al., 1972), which may be caused, at least in part, by a central action of α-methyldopa (Henning and Van Zwieten, 1968; Ingenito et al., 1970; Heise and Kroneberg, 1972). The effect of α-methyldopa on heart rate in the rat is less well documented. A decrease in heart rate has been reported after α-methyldopa administration in man and in anesthetized dogs (Dollery and Harington, 1962; Antonaccio et al., 1974). In the anesthetized rat, however, a short lasting increase with a maximum after 1 hr has been reported (Ayitey-Smith and Varma, 1970). This observation was confirmed in conscious rats, but in addition we observed that the initial tachycardia was followed by a long lasting bradycardia which was associated with a fall in body temperature. The present investigation deals with the relationships between the α-methyldopa-induced changes in blood pressure, heart rate and body temperature and with the influence of inhibition of central and peripheral decarboxylase activity.

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2. Materials and methods

Experiments were performed at a room temperature of 20° C (19—21° C), except in

one case when an ambient temperature of 30°C (29—31°C) was used. Animals were housed in a temperature controlled room at 23 ± 1°C. The rats were transported from the animal room to the room kept at 20 or 30°C at least 20 min before starting the experiment.

Renal hypertension was induced by application of a solid silver clip (internal diameter 0.20 mm) on the left renal artery of male Wistar rats (outbred stock; Cpb, WU, TNO, Zeist, The Netherlands) weighing 130—160 g (Leenen and De Jong, 1971). This procedure results in a regular development of hypertension, reaching a maximum 3—4 weeks after operation. The hypertensive rats were used 4—6 weeks after application of the clip. Body weight range at that time was 200—240 g. In one experiment male normotensive rats, weighing 220—260 g, were used.

A permanent indwelling iliac cannula was used to enable continuous recording of blood pressure and heart rate in unanesthetized rats. The cannula was modified to the model of Weeks and Jones (1960). A U-shaped piece of PE-50 (Clay Adams) (1.5 cm length) was inserted into the left iliac artery with the tip just lying into the abdominal aorta. The PE-50 cannula was connected to a piece of PE-100 (13 cm length) that passed s.c. to the neck. Via a short stainless steel needle (2 cm length, o.d. 1.2 mm) the cannula was connected with a piece of vinyl tubing (Portex, i.d. 0.91 mm, o.d. 1.5 mm). The transition of PE-100 to the needle was protected by PE-240 (2 cm length). When blood pressure was not being recorded the cannula was filled with heparinized 0.9% NaCl (78 I.U./ml) and stoppered with a short steel nail. During the period of measurement the cannula was connected via a Statham transducer (Model P23AC) with PP-100 (Portex) to a Grass Polygraph. Complete pressure curves were registered except at the times indicated under Results when mean arterial blood pressure was recorded during approximately 5 min. Heart rate was calculated from the blood pressure recording at these times. The iliac cannula was implanted under ether anesthesia at least 24 hr prior to the experiment.

Body temperature was measured with a telethermometer (Yellow Springs Instrument Co., Yellow Springs, Ohio). The probe was inserted 3.5 cm into the rectum and the temperature was recorded at hourly intervals.

α-Methyldopa (l-α-methyl-3,4-dihydroxyphenylalanine hydrochloride; Merck Sharp and Dohme) or vehicle (0.9% NaCl) were injected intraperitoneally (i.p.). Peripheral decarboxylase activity was inhibited by Ro 4-4602 (Seryl-2,3,4-trihydroxybenzyl-hydrazine hydrochloride; Benserazid; Hoffman—La Roche) in a dose of 50 mg/kg i.p. (Bartholini and Pletscher, 1968). This dose was injected 4 times at 90 min intervals, beginning 30 min before administration of α-methyldopa. Central decarboxylase activity was inhibited by intracerebroventricular (i.c.v.) administration of Ro 4-4602 (3 × 0.15 mg/kg, dissolved in 15 μl 0.9% NaCl) via a permanently implanted cannula into the lateral ventricle as described by Hayden et al. (1966). Cannulas were implanted under ether anesthesia 24—48 hr before the experiment. The drug was injected 3 times at 2 hr intervals beginning 30 min before administration of α-methyldopa.

Results are expressed as means ± standard error of the mean (S.E.M.). Significance of the difference between control and α-methyldopa values was determined with the Student's *t*-test.

3. Results

3.1. Effect of α-methyldopa on blood pressure, heart rate and body temperature

The basal values at different ambient temperatures, for blood pressure, heart rate and body temperature of renal hypertensive and normotensive rats are reported in table 1. At 30°C blood pressure of normotensive animals was significantly lower when compared to controls at 20°C. In both groups of rats heart rate was significantly decreased while body

TABLE 1

Basal values of mean blood pressure, heart rate and body temperature in normotensive and renal hypertensive rats at different ambient temperatures.

Values given are means ± S.E.M. of 15—19 animals. Basal values were measured 20 min after the rats were placed at 20 or 30°C.

Temperature	Renal hypertensive rats		Normotensive rats	
	20°C	30°C	20°C	30°C
Blood pressure (mm Hg)	185 ± 7	168 ± 7	119 ± 3	108 ± 3*
Heart rate (b.p.m.)	418 ± 12	372 ± 10*	385 ± 14	334 ± 11*
Body temperature (°C)	37.2 ± 0.1	37.8 ± 0.1*	37.4 ± 0.2	38.0 ± 0.2*

* $p < 0.05$ compared to controls at 20°C.

temperature was significantly elevated at this ambient temperature. α-Methyldopa (400 mg/kg) caused a decrease in blood pressure at 20°C ambient temperature in renal hypertensive rats (fig. 1). The decrease was maximal

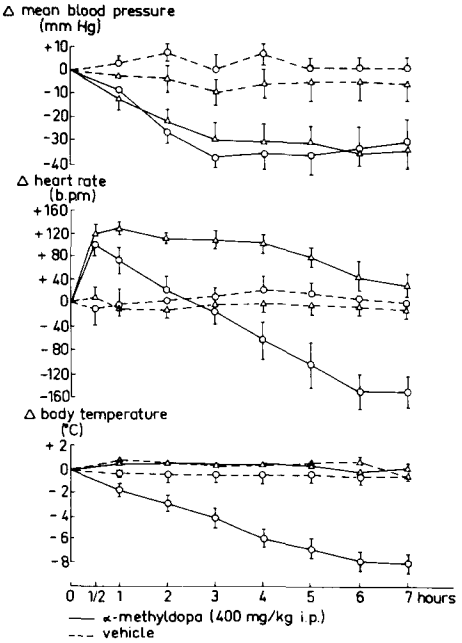

Fig. 1. Effect of α-methyldopa (400 mg/kg i.p.) on mean blood pressure, heart rate and body temperature of renal hypertensive rats. Data are means ± S.E.M. of 7—10 animals. Circles indicate data from rats at an ambient temperature of 20°C and triangles data from rats at 30°C.

3 hr after administration (—38 ± 4 mm Hg) and slowly returned to the control value within 24 hr. At an ambient temperature of 30°C a similar decrease of blood pressure was observed. Heart rate at 20°C showed an initial increase with a maximum after 30 min (+102 ± 16 b.p.m.), followed by a decrease (fig. 1). The lowest value was reached 7 hr after administration of the drug (—150 ± 23 b.p.m.). At 30°C the initial increase in heart rate was maintained for 6—7 hr, with no subsequent decrease below basal values. The effect of α-methyldopa on body temperature also depended on ambient temperature. At 20°C, body temperature decreased gradually with a maximum after 7 hr (—8.2 ± 0.7°C) (fig. 1). However, at 30°C no decrease in body temperature was observed. After 24 hr body temperature and heart rate were not significantly different from the control values.

The maximal decrease in blood pressure after α-methyldopa (400 mg/kg) administration was less pronounced (—23 ± 9 mm Hg) in normotensive rats (fig. 2) than in the hypertensive rats. The action of α-methyldopa on heart rate and body temperature in normotensive rats (fig. 2) did not differ significantly from the effect observed in the hypertensive animals. Individual values for decrease in heart rate and body temperature, in renal hypertensive and normotensive rats, 6 hr after administration of α-methyldopa (400 mg/kg)

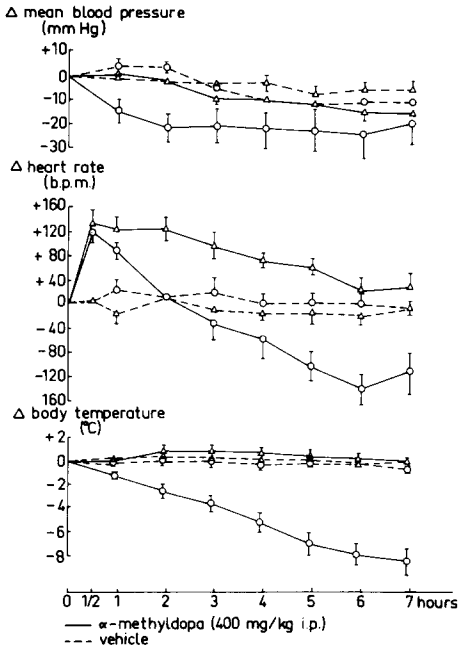

Fig. 2. Effect of α-methyldopa (400 mg/kg i.p.) on mean blood pressure, heart rate and body temperature of normotensive rats. Data are means ± S.E.M. of 7—10 animals. Circles indicate data from rats at an ambient temperature of 20° C and triangles data from rats at 30° C.

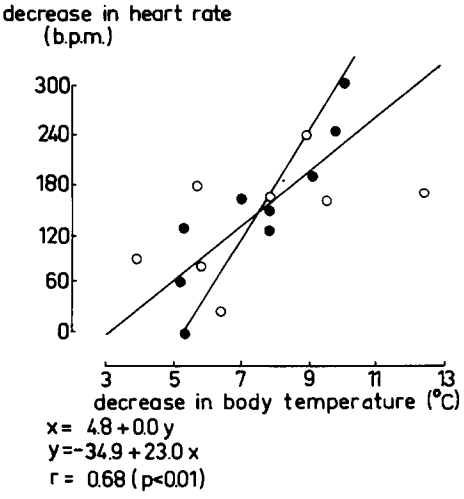

Fig. 3. Correlation between decrease of heart rate and decrease of body temperature 6 hr after administration of α-methyldopa (400 mg/kg i.p.) to renal hypertensive (•) and normotensive (○) rats. Data are from animals in figs. 1 and 2; r = 0.68, $p < 0.01$.

were significantly correlated (r = 0.68, $p < 0.01$; fig. 3).

3.2. *Effect of inhibition of central decarboxylase activity*

Basal values of blood pressure, heart rate and body temperature of the renal hypertensive rats at an ambient temperature of 20° C did not differ significantly from those reported above (*3.1.*). Inhibition of the central decarboxylase activity with Ro 4-4602 (3 × 0.15 mg/kg i.c.v.), prevented the decrease in blood pressure, heart rate and body temperature (fig. 4). The initial increase in heart rate by α-methyldopa was not significantly affected by prior administration of Ro 4-4602. Heart rate was still significantly elevated 6 hr after administration of the drug (+46 ± 26 b.p.m.) $p < 0.05$.

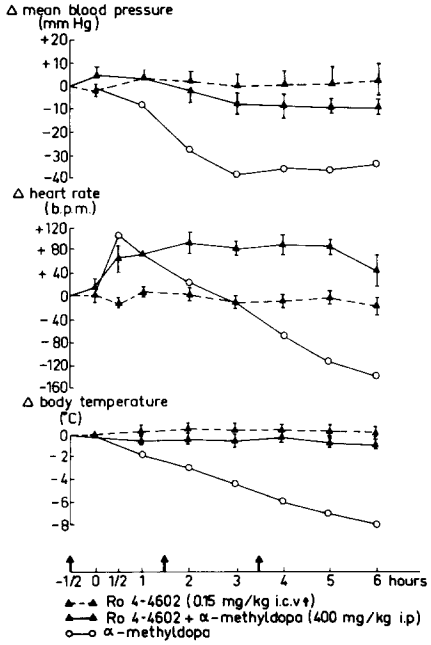

Fig. 4. Effect of α-methyldopa (400 mg/kg i.p.) on mean blood pressure, heart rate and body temperature of renal hypertensive rats after inhibition of central decarboxylase activity with Ro 4-4602 (3 × 0.15 mg/kg i.c.v. ↑). Data are means ± S.E.M. of 7—8 animals. The group of rats treated with α-methyldopa alone is from fig. 1.

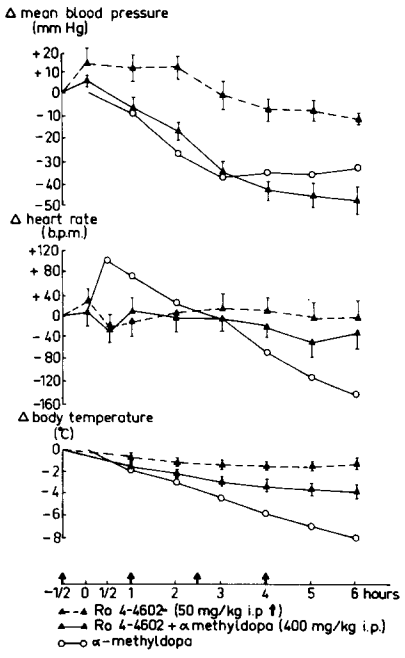

Fig. 5. Effect of α-methyldopa (400 mg/kg i.p.) on mean blood pressure, heart rate and body temperature of renal hypertensive rats after inhibition of peripheral decarboxylase activity with Ro 4-4602 (4 × 50 mg/kg i.p. ↑). Data are means ± S.E.M. of 5—7 animals. The group of rats treated with α-methyldopa alone is from fig. 1.

3.3. Effect of inhibition of peripheral decarboxylase activity

Basal values of blood pressure, heart rate and body temperature of the renal hypertensive rats at an ambient temperature of 20° C did not differ significantly from those reported above (*3.1.*). Inhibition of peripheral decarboxylase activity with Ro 4-4602 (4 × 50 mg/kg i.p.) did not affect the decrease in blood pressure (fig. 5). Although heart rate decreased somewhat, this difference was not statistically significant. The fall in body temperature was also reduced after inhibition of peripheral decarboxylase activity (fig. 5). All differences were statistically significant (at least $p < 0.05$). The decrease was maximal after 6 hr (—3.7 ± 0.6° C). However, body temperature of the controls was also slightly decreased.

4. Discussion

The present study demonstrates that α-methyldopa decreases arterial blood pressure, heart rate and body temperature both in conscious renal hypertensive and in normotensive rats. These effects apparently depended on centrally formed metabolites of α-methyldopa as they were completely blocked after inhibition of central decarboxylase activity. Henning (1969) has reported similar results for the decrease of blood pressure in unanesthetized renal hypertensive rats. The metabolite which mediates the central effect on blood pressure is thought to be α-methylnoradrenaline, because inhibition of dopamine-β-hydroxylase activity blocks the central hypotensive effect of α-methyldopa (Henning and Rubenson, 1971; Day et al., 1973; Nijkamp and de Jong, 1974). This is also indicated by the greater effectiveness of α-methylnoradrenaline compared to α-methyldopa or α-methyldopamine in decreasing blood pressure after intraventricular administration (Heise and Kroneberg, 1972). The decrease of blood pressure after α-methyldopa administration was associated with a pronounced decrease of body temperature which developed somewhat slower. In fact, the hypothermia may equally well be mediated by α-methylnoradrenaline. Central administration of noradrenaline or α-methylnoradrenaline in the rat has been reported to decrease body temperature (Feldberg and Lotti, 1967; Bruinvels, 1970; Breese et al., 1972) and α-methylnoradrenaline can replace noradrenaline in the storage granules in the brain (Carlsson and Lindqvist, 1962). The hypothermia could thus be explained by release of noradrenaline although a direct action of α-methylnoradrenaline appears more likely because rats which have their brain noradrenaline depleted to ca. 25% by administration of 6-hydroxydopamine (Breese and Traylor, 1970) give an enhanced response to α-methylnoradrenaline (Breese et al., 1972). Although α-methyldopa may decrease brain serotonin and 5-hydroxyindoleacetic acid (Carlsson and Lindqvist, 1962; Pletscher et

al., 1964; Henning, 1967), this effect seems unrelated to the observed hypothermia. In the rat elevated brain serotonin mainly appears to decrease body temperature (Feldberg and Lotti, 1967; Francesconi and Mager, 1974).

α-Methyldopa had a marked dual action on heart rate. This was apparently due to a peripheral tachycardic component, which was blocked by inhibition of peripheral decarboxylase activity, as well as a slower occurring bradycardia which appeared to depend on the central conversion to α-methylnoradrenaline. At 30° C environmental temperature no decrease in body temperature and heart rate was observed, while at 20° C the decrease in both parameters was significantly correlated in individual animals. These findings suggest that the bradycardia is secondary to the fall in body temperature. Such a correlation between the decrease in body temperature and heart rate has also been reported in rats in which body temperature was decreased by forced swimming in water at 20° C (Baker and Horvath, 1964). The reduction of the hypothermia and bradycardia observed after administration of α-methyldopa in rats in which peripheral decarboxylase activity had been inhibited cannot be explained but may be caused by a direct action of the inhibitor on the peripheral factors mediating the fall in body temperature. However, the possibility that the administration of Ro 4-4602 caused a partial inhibition of central decarboxylase activity cannot be excluded.

It is of interest that two other centrally acting hypotensive agents, l-dopa (Henning and Rubenson, 1970) and clonidine (Kobinger and Walland, 1967; Sattler and Van Zwieten, 1967) caused a centrally mediated decrease in heart rate (Schmitt and Schmitt, 1969; Osborne et al., 1971). These drugs have also been shown to cause a centrally mediated decrease in body temperature in unanesthetized rats (Tsoucaris-Kupfer and Schmitt, 1972; Maj and Pawlowski, 1973). This hypothermic action of clonidine as well as that of noradrenaline and α-methylnoradrenaline is inhibited after central administration of α-adrenoceptor blocking agents (Marley and Stephenson, 1970; Burks, 1972; Tsoucaris-Kupfer and Schmitt, 1972). The central hypotensive effect of clonidine, noradrenaline and α-methyldopa is also inhibited after central administration of α-adrenoceptor blocking drugs (Finch and Haeusler, 1973; Schmitt et al., 1973; Sinha and Schmitt, 1974).

These findings may indicate that the central effects of α-methyldopa, l-dopa and clonidine on blood pressure, body temperature and indirectly on heart rate are mediated through a common pathway of action in the brain. Accordingly, both the hypotension and the hypothermia may be caused by central noradrenergic receptor stimulation.

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