SHORT COMMUNICATION

Inhibition of adrenaline-forming enzyme in the brain prevents one-kidney, one-clip hypertension and deoxycorticosterone acetate–salt hypertension in the rabbit

C. ROSENDORFF, J. R. MELAMED, M. L. HURWITZ, A. COULL AND A. JARVIS

MRC–University Circulation Research Unit, University of the Witwatersrand Medical School, Johannesburg, South Africa

(Received 15 June 1982; accepted 20 July 1982)

Summary

1. Phenylethanolamine N-methyltransferase (PNMT) converts noradrenaline into adrenaline and brain PNMT is elevated in spontaneously hypertensive and deoxycorticosterone acetate (DOCA)–salt hypertensive rats. In view of the evidence for the involvement of central adrenergic neurons in renal hypertension, we measured the blood pressure response in one-clip, one-kidney Goldblatt hypertensive and DOCA–salt hypertensive rabbits to the PNMT inhibitor SK&F 64139, injected into the lateral cerebral ventricles.

2. Intracerebroventricular injection of SK&F 64139 (10 µg/kg) significantly attenuated the mean arterial blood pressure rise in one-clip, one-kidney and DOCA–salt rabbits, at 4 and 8 weeks.

3. These findings support the idea that hypertension in this animal model requires an intact adrenaline biosynthetic process, and that central catecholaminergic neurons may be involved in the pathogenesis of low-renin volume dependent forms of hypertension.

Key words: adrenaline, blood pressure, catecholamines, hypertension, noradrenaline.

Abbreviations: DOCA, deoxycorticosterone; PNMT, phenylethanolamine N-methyltransferase

Introduction

Phenylethanolamine N-methyltransferase (PNMT) is the enzyme which N-methylates noradrenaline to form adrenaline [1, 2] in adrenal chromaffin cells [1, 3] and in the brain stem [4]. SK&F 64139 (7,8-dichloro-1,2,3,4-tetrahydroisoquinoline HCl) is a PNMT inhibitor which can cross the blood–brain barrier [5]. Black et al. [6] showed recently that SK&F 64139 reduced blood pressure to normal levels in mineralocorticoid–salt hypertensive rats, a model of hypertension which is volume-dependent and characterized by a low plasma renin activity [7, 8]. By contrast, SK&F 64139 had no effect on blood pressure in high-renin Goldblatt two-kidney, one-clip hypertension in rats [6]. What has not been established is whether PNMT inhibition is effective in other forms of low-renin hypertension, whether it works in animals other than the rat, and whether the action is central or peripheral. For these reasons we have investigated the effects on blood pressure of the PNMT inhibitor SK&F 64139 injected into the cerebral ventricles of rabbits with Goldblatt one-clip, one-kidney hypertension and of rabbits treated with DOCA and a high salt diet.

Methods

All experiments were performed on male New Zealand White rabbits, weighing 2.6–3.1 kg. The conscious rabbits were restrained in stocks, and basal blood pressures were measured in the central ear artery via a 19 gauge needle connected to a Statham P23 AA pressure transducer and a dynograph coupler (Beckman Recorder, R411). Phasic and mean (electronically damped) arterial blood pressures were recorded.

The rabbits were then anaesthetized with
alphaxalone (4 mg intravenously) as a bolus injection with additional doses of 1 mg intravenously as required, and guide headplates were attached to the skull by a modification of the technique described by Monnier & Gangloff. These headplates allow subsequent stereotaxic access to any part of the brain in the conscious restrained rabbit. At the same operative session unilateral nephrectomy was performed via a retroperitoneal approach and the renal artery supplying the contralateral kidney was ligated with size 4 silk, the ligature being tightened until we could just see slight constriction of the artery. The operation wounds were then closed in layers and the animals allowed to recover. At weeks 2, 4, 6 and 8 the conscious rabbits were placed in restraining stocks, a needle was inserted into the central ear artery for the recording of blood pressure, and in ten animals (group B) SK&F 64139, 10 µg/kg body weight, dissolved in 10 µl of sodium chloride solution (154 mmol/l: saline) (pH 5.2) was injected via a cannula into either the left or right lateral cerebral ventricle at co-ordinate aB-10 mm [9]. Group A rabbits (n = 4) were given 10 µl of saline (pH 5.4) alone into the cerebral ventricles. Group C rabbits (n = 4) were given a sham operation (no nephrectomy and no renal artery ligation), and received SK&F 64139 in the same dose as group B.

In the second series of experiments, six rabbits (group B) were given DOCA (25 mg/kg per week) by subcutaneous injection and NaCl solution (0.45 g/dl) as drinking water. At weeks 2, 4, 6, and 8 these rabbits were also given SK&F 64139, (10 µg/kg body weight, by injection into the lateral cerebral ventricle). Group A (n = 6) received the identical DOCA-salt therapy, but had saline injected into the cerebral ventricles, and group C (n = 8) were on a normal rabbit diet and tap water, and received no SK&F 64139.

Statistical analysis of the data was by unpaired Student's t-test.

Results

Fig. 1 (a) summarizes the results of the first experiment. Blood pressure values for the baseline period and at weeks 4 and 8 are shown. There was no significant difference between blood pressure values in the sham-operated animals given SK&F 64139 (group C) and the one-kidney, one-clip rabbits given SK&F 64139 (group B). Also, in both groups, there was no significant increase in blood pressure with time. However, in group A, the one-kidney, one-clip rabbits not given SK&F 64139, there was a significant increase in blood pressure at 4 weeks, a further rise at 8 weeks, and at both 4 and 8 weeks the mean blood pressure was significantly (P < 0.01) greater than in both groups B and C. The conclusion is that the one-clip, one-kidney pro-

---

**Fig. 1.** Mean arterial blood pressure responses in three groups of rabbits. (a) Group A (n = 4): one-kidney, one-clip hypertension; group B (n = 10): one-kidney, one-clip hypertension, with SK&F 64139 (10 µg/kg) injected into the lateral cerebral ventricles at weeks 2, 4, 6 and 8 (arrows); group C (n = 4): sham-operated rabbits with SK&F 64139 injected as described for group B. Blood pressure values are shown as means ± 1 SE. Values for group B are significantly lower than those for group A at both 4 and 8 weeks. (b) Group A (n = 6) rabbits were injected with DOCA (25 mg/kg per week) subcutaneously and given NaCl solution (0.45 g/100 ml) as drinking fluid. Group B (n = 6) rabbits were treated in the same way, but, in addition, were injected with SK&F 64139 (10 µg/kg) into the lateral cerebral ventricles at weeks 2, 4, 6 and 8 (arrows). Group C (n = 8) rabbits were on a normal diet and tap water, and were not given SK&F 64139. Blood pressure values are shown as means ± 1 SE. Values for group B are significantly lower than those for group A at 8 weeks.
intracerebroventricular injection of SK&F treated rabbits (group A) show a significant increase in mean arterial blood pressure over rabbits on a normal diet, who were not given baseline values, at both weeks shown in Fig. There is, as expected, no significant difference between groups A and B, injected with SK&F 64139 (group B), show a smaller rise in blood pressure, significantly different (P < 0.05) from group C only at 8 weeks. There is, as expected, no significant change in blood pressure in group C, the control rabbits on a normal diet, who were not given SK&F 64139. Although the blood pressure difference between groups A and B was significant (P < 0.05) only at 8 weeks, it is clear from Fig. 1(b) that SK&F 64139 attenuated, but did not abolish, the development of hypertension in the DOCA–salt rabbits.

Discussion

It is likely that one-clip, one-kidney hypertension is a low-renin, volume-dependent form of hypertension [7, 8]. It is also possible that, in this form of hypertension, there may be a vasoconstrictor (sympathetic) component [11, 12]. It has been shown [13] that destruction of central nervous system adrenergic neurons by 6-hydroxydopamine prevents the development of renal hypertension in rabbits.

PNMT, the enzyme which catalyses the N-methylation of noradrenaline to form adrenaline, is found in relatively high concentrations in the rat in regions A1 and A2 of the medulla [41], corresponding to regions C1 and C2 of Hökfelt et al. [14]. Of great interest is the finding that brain PNMT is elevated in spontaneously (genetic) hypertensive rats and in DOCA–salt rats [15]. This would suggest that increased adrenaline synthesis in the brain stem may have something to do with the development of hypertension in these experimental animals. In DOCA–salt hypertensive rats plasma adrenaline levels are raised [16].

Although we have not measured PNMT activity directly in this study, there is good evidence that SK&F 64139 is a PNMT inhibitor which can cross the blood–brain barrier [17]. An earlier compound, SK&F 29661, does not cross the blood–brain barrier [18]. Parenteral SK&F 64139 reduces arterial pressure in DOCA–salt hypertensive rats but SK&F 29661 has no effect on DOCA–salt or one-kidney, one-clip hypertension [6]. This suggests that it is a central PNMT inhibition which is important in the anti-hypertensive action of PNMT inhibitors. The present paper has demonstrated that this is so in both one-kidney, one-clip renal hypertension and DOCA–salt hypertension. Also this is not species specific to the rat; these effects are readily demonstrable in the rabbit.

There are still many unanswered questions. One is, how does volume overload, the common factor in one-kidney, one-clip and DOCA–salt hypertension, generate medullary PNMT? Does this have to do with water or sodium retention or both? How specific are the tetrahydroisoquinolines, such as SK&F 64139, as PNMT inhibitors? Recent studies [19, 20] have shown that SK&F 64139 has significant α₁-adrenoceptor antagonist activity. This finding does not, however, explain its anti-hypertensive activity; central antagonists of noradrenaline presynaptic α₂-receptors would not be expected to lower blood pressure. Nor does it negate its function as a PNMT inhibitor; the Kᵢ of SK&F 64139 for PNMT inhibition in the central nervous system (21 nmol/l) is lower than that for α₂-receptor-antagonist (89 nmol/l) [19]. Also, there is some evidence that at least some α₂-receptors in the brain may be adrenaline receptors [21]. If these adrenaline-sensitive α₂-receptors are post-synaptic, and are involved with blood pressure control, then the effects of PNMT inhibition and α₂-receptor blockade on blood pressure would be synergistic.

Acknowledgments

The support of the South African Medical Research Council and of the University of the Witwatersrand is gratefully acknowledged. We also thank Professor D. Mitchell for his help and advice in the early stage of this project, and Smith, Kline and French Laboratories, Philadelphia, for supplies of SK&F 64139.

References


PNMT inhibition and hypertension


