Peripheral vascular smooth muscle relaxation in normotensive and hypertensive rats†

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The adenylcyclase-phosphodiesterase system has been suggested as one of the biochemical mechanisms participating in the regulation of arterial tone and reactivity (Volicer & Hynie 1971; Triner et al 1972). Specific alterations in the vascular cyclic nucleotides system have been advanced as an explanation for increased total peripheral resistance occurring in hypertension (Amer 1973, 1975). Cyclic nucleotides and β-adrenoceptor stimulants effect a decrease in the relaxation of aortic strips of spontaneously hypertensive (SH) rats compared with those of normotensive rats and defects in vascular relaxation have been proposed to contribute to hypertension (Triner et al 1975, Cohen & Berkowitz 1976), while Spector et al (1969) have reported an enhanced relaxation in response to isoprenaline. As the aorta is not the major determinant factor of the total peripheral resistance, it is difficult to extrapolate the results obtained from these studies to small arteries and arterioles which are important in determining the level of blood pressure. In view of these findings, studies were carried out to investigate the vasodilator activities of agents known to increase cyclic(c) AMP content of the tissues in the mesenteric vascular bed of SH, renal hypertensive (RH) and deoxycorticosterone acetate/saline hypertensive (doca/saline) rats.

Male SH rats were direct descendants of the original strain developed by Okamoto & Aoki (1963). For renal hypertension, male Wistar rats were made hypertensive by clamping the left renal artery with silver clip (aperture 0·2 mm), leaving the contralateral kidney intact (Goldblatt et al 1934). Mineralocorticoid hypertension was induced in male Wistar rats by implanting 4 pellets (25 mg each) of doca, subcutaneously after removing the left kidney and substituting 1·0% sodium chloride solution for drinking water (Peterfalvi & Jequier 1960). For the purpose of comparison, age matched normotensive Wistar Kyoto (WKY) rats, and sham operated age matched normotensive Wistar (NW) rats were used as controls respectively (Lais & Brody 1978). The age, weight, blood pressure and ratio of the weight of ventricles to body weight of the animals are summarized in Table 1. Since isoprenaline, papaverine and adenosine are known to cause relaxation of arterial smooth muscle by increasing tissue cAMP content either by stimulating adenylcyclase or by inhibiting phosphodiesterase (Pöch & Kukovetz 1971; Wurm et al 1976), the vasodilator effects of these agents were investigated on the peripheral vascular bed by perfusing the vascularly isolated but neurologically intact mesenteric arteries of hypertensive and normotensive rats.

The mesenteric arteries were auto-perfused at a constant flow, according to Bhattacharya et al (1977). The rats were anaesthetized with a combination of sodium pentobarbitone (20 mg kg^{-1} i.p.) and urethane (500 mg kg^{-1} i.p.). The blood from the carotid artery was forced by a peristaltic pump into the superior mesenteric artery. The pump speed was so adjusted that the perfusion pressure was almost equal to the systemic blood pressure. Intra-arterial injections were given into the tubing leading towards the periphery. Ten animals were used in each group. Results were expressed as a change in perfusion pressure in mm Hg for each rat. Statistical significance between groups was calculated by Student's *t*-test.

Isoprenaline (1 μg), papaverine (50 μg) and adenosine (50 μg), when given intra-arterially elicited a fall in perfusion pressure without an appreciable change in systemic blood pressure. The vasodilator responses in SH rats were found to be significantly greater ($P < 0.001$) compared with normotensive WKY rats (Fig. 1). Similar findings were also reported by Deragon et al (1978), where relaxation of hind limb vessels of SH rats induced by isoprenaline was found to be increased compared with normotensive rats.

Table 1. Characteristics of hypertensive and normotensive rats.

Rats	Mean b.p. (mm Hg)	Age (weeks)	Wt (g)	Ventricle as g kg^{-1} body wt
WKY	118·4 ±5·5	20–24	246 ±7·6	3·2 ±0·1
SH	188·7 ±4·7*	20–24	272 ±6·3	4·52 ±0·14*
NW	121·2 ±5·5	12–14	172 ±6·7	3·41 ±0·15
RH	189·4 ±6·0*	12–14	190 ±9·3	5·34 ±0·24*
DOCA/saline	170·0 ±7·3*	12–14	191 ±5·1	4·1 ±0·09*

Mean blood pressure = Diastolic + 1/3 pulse pressure.
Values are mean ± s.e.m. (12 animals per group).
* Significantly different from appropriate age-matched normotensive rats ($P < 0.001$). Statistical analysis with Student's *t*-test.

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† Dedicated to Prof. R. Sammet, Hoechst A.G., Frankfurt on the occasion of his 60th birthday.

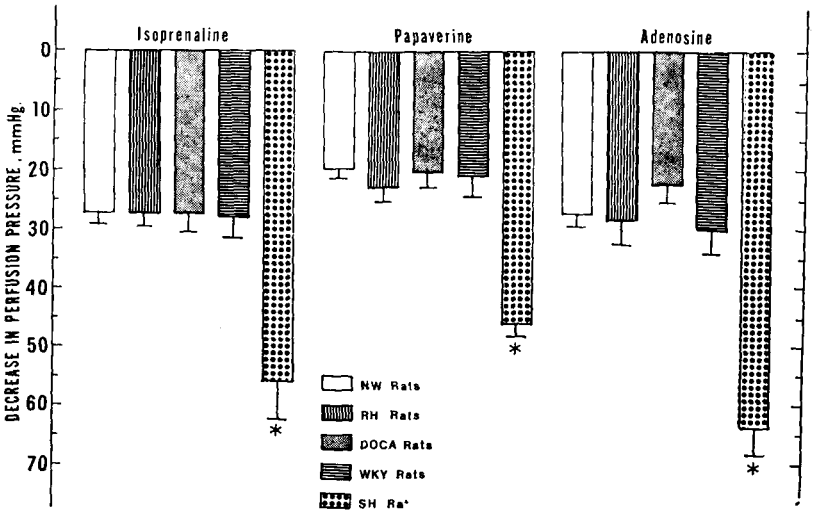

FIG. 1. Decrease in perfusion pressure (ordinate) mm Hg after intra-arterial administration of isoprenaline (1 μg), papaverine (50 μg) and adenosine (50 μg) in the perfused mesenteric artery preparation from normotensive Wistar (NW) rats (open columns), renal hypertensive (RH) rats (Vertically hatched columns), doca/saline hypertensive rats (small stippled columns), normotensive Wistar Kyoto (WKY) rats (horizontally hatched columns) and spontaneously hypertensive (SH) rats (large stippled columns). Each column represents the mean value from 10 animals; vertical lines indicate s.e.m. Value of significance shown is: $*P < 0{\cdot}001$ statistical analysis with Student's t-test.

The increased sensitivity of the blood vessels to the vasodilators in SH rats which is observed in the present study could be due to their ability to increase the intracellular cAMP content. This assumption is based on the findings that increased vascular tone in SH rats is due to decreased concentration of intracellular cAMP (Amer 1973).

These observations are not in agreement with the results obtained from in vitro studies and also do not support the hypothesis which suggests that defects in vascular relaxation are involved in the development of hypertension in SH rats (Triner et al 1975; Cohen & Berkowitz 1976). This discrepancy could be due to the different experimental procedures. In the studies where isolated aortic strips were used, the vascular relaxation was tested against 5-hydroxytryptamine (5-HT)-induced contractile responses. Assessment of results obtained by such an indirect method is difficult because (i) the contractile response to 5-HT is different in various models of hypertensive rats and normotensive rats (Haeusler & Finch 1972) and (ii) the extent of relaxation induced by any compound depends upon the degree of contraction.

In contrast to the results obtained in SH rats, the vasodilator agents did not effect any significant increase in vascular smooth muscle relaxation in RH and doca/saline rats compared with NW rats (Fig. 1). These differential vasodilator responses may be attributed to the difference in pathogenesis of hypertension. This argument is based on the findings that the increased plasma concentration of angiotensin and vasopressin

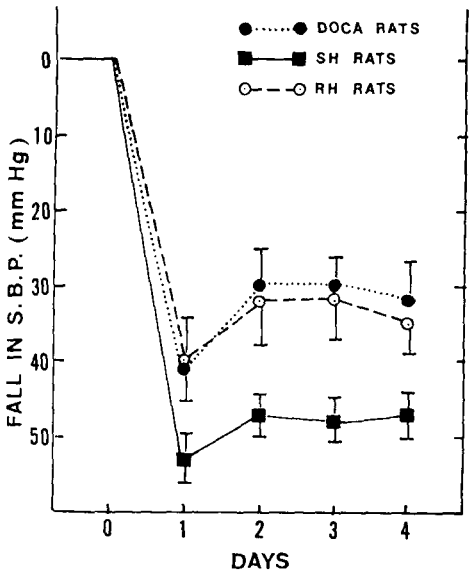

FIG. 2. The effect of prazosin (0·5 mg kg^{-1}, orally) given daily for 4 days, on systolic blood pressure of spontaneously hypertensive (■), renal hypertensive (○) and doca/saline hypertensive (●) rats. Each point represents the mean ± s.e.m. (n = 12 animals per group). Data from different groups were examined using analysis of variance (i.e. area under the curve). Ordinate; Fall in systolic blood pressure (S.B.P.) mm Hg. Abscissa: Time in days.

plays a significant role in the maintenance of elevated blood pressure in RH and doca/saline rats respectively and not in SH rats (Brunner et al 1974; Möhring et al 1977). Furthermore, angiotensin and vasopressin are known to increase synthesis of vascular cGMP (Amer 1975). This imbalance of cyclic nucleotide synthesis in favour of cGMP may cause an increase in vascular tone and peripheral resistance. Thus it appears that increased plasma concentrations of angiotensin and vasopressin may be antagonizing vasodilator responses to isoprenaline, papaverine and adenosine in RH and doca/saline rats either directly or indirectly by modulating cGMP levels in the vasculature.

This possibility was further investigated by studying the effect of the antihypertensive agent prazosin in different models of hypertensive rats. Prazosin, in addition to its selective antagonism at the post synaptic α-adrenorceptor site (Brogden et al 1977), is also a phosphodiesterase inhibitor and its direct relaxant effects on arteriolar smooth muscle is partly mediated through an increased intracellular concentration of cAMP (Oates et al 1976; Genazzani et al 1978).

Prazosin was administered once daily in a dose of 0·5 mg kg^{-1} given by mouth for 4 days. Systolic blood pressure was determined in conscious rats by the tail cuff method using a piezo-electric detector. Prazosin caused a significantly greater fall in the systolic blood pressure of SH rats, than that in RH and doca/saline rats, between which there was no significant difference (Fig. 2). Thus it appears that differences in the pathogenesis of hypertension influence the hypotensive activity of vasodilator agents. Furthermore, the results also support the possibility that the ratio of cGMP/AMP is more important in maintaining an increased arterial tone and reactivity in hypertensive rats than their individual levels.

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