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VASCULAR REACTIVITY IN SPONTANEOUSLY HYPERTENSIVE NORMOTENSIVE AND HYPOTENSIVE RATS

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1 Three strains of rats spontaneously hypertensive (HRS), normotensive (NS) and hypotensive (HOS), were selected by repeated inbreeding.

2 In order to explain the different blood pressure levels observed, we have studied their vascular reactivity to noradrenaline and angiotensin II. Experiments were performed in anaesthetized ganglion-blocked, vagotomized rats.

3 The reactivity to noradrenaline was significantly higher in HRS than in NS and HOS rats.

4 The reactivity to angiotensin II appeared identical in the three strains used.

Introduction

Vascular reactivity to different pressor agents has been extensively studied in spontaneously hypertensive rats either in whole animals (Okamoto, Tabei, Fukushima, Nosaka, Yamori, Ichijima, Haebara, Matsumoto, Maruyama, Suzuki & Tamegai, 1966; Phelan, 1966; Bing, 1969; Scholtysik & Unda, 1971; Shibayama, Misogami & Sokabe, 1971) or in isolated organs (Spector, 1969; Clineschmidt, Geller, Govier & Sjoerdsma, 1970; Hallblack, Lundgren & Weiss, 1971; Massingham & Shevde, 1971; Fujiwara, Kuchii & Shibata, 1972). According to the strain of rats and the methods utilized, contradictory results have been obtained.

Recently (Dupont, Dupont, Froment, Milon & Vincent, 1973), we have succeeded in isolating by inbreeding three strains of rats differing spontaneously in their blood pressure which are classified as hypertensive (HRS), normotensive (NS) and hypotensive (HOS).

In the present study, the vascular reactivity of these three strains of rats to noradrenaline and angiotensin II was compared in whole animals anaesthetized and pretreated in order to diminish spontaneous cardiovascular regulation.

Methods

The rats used were CFE Albino derived from the Sprague-Dawley strain, supplied by the Oncins breeding centre (Iffa Credo). As previously described (Dupont *et al.*, 1973) by successive inbreeding over a period of three years, three strains of rats differing spontaneously in their blood pressure, have been selected from three pairs of rats of the same age. In this study, females of the sixth generation of the selection have been used. In the hypertensive group 16 animals were used, 10 in the NS and 10 in the HOS group. At the time of the experiment all the animals were old enough (18-25 weeks) to have a stable blood pressure. Their body weight averaged 220 grams. They received water *ad libitum* and a standard diet (containing approximately 0.2% sodium chloride) prepared by Iffa-Rat (Iffa Credo). Starting at the age of five weeks, their blood pressure was taken once a week by the indirect oscillometric method (Racia) without anaesthesia, after slight warming at 38°C for 10 minutes.

For vascular reactivity studies, the animals were anaesthetized with an intraperitoneal injection of urethane (1.4 g/kg). In order to minimize their spontaneous blood pressure regulation they were pretreated with pentolinium (May and Baker; 25 mg/kg i.d.) and atropine sulphate (0.25 mg/kg s.c.). Polyethylene catheters were placed one in the carotid artery for mean blood pressure direct measurement and one in each of the jugular veins for drug injections.

At the same time vagotomy on both sides was performed. Mean blood pressure was measured

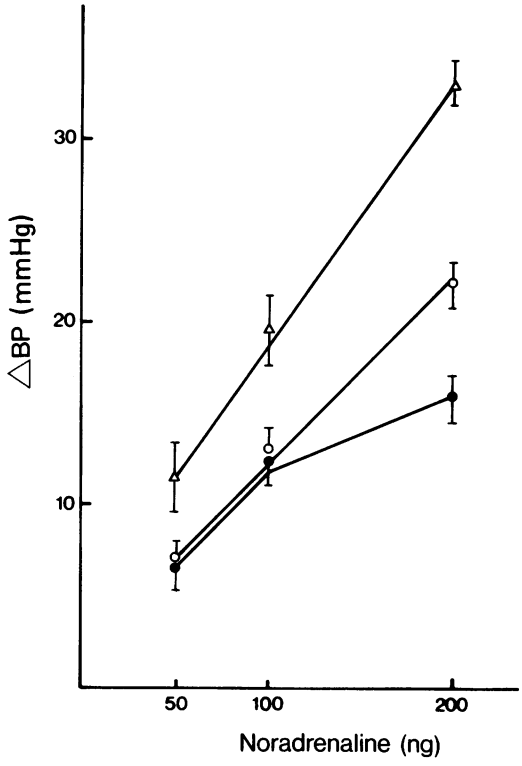

Fig. 1 Vascular reactivity to noradrenaline expressed as variations of blood pressure (△BP) evoked by injection of three doses of noradrenaline. Standard deviations are shown by the vertical bars. Hypotensive rats (○); normotensive rats (●); hypertensive rats (△).

with a Palmer mercury micromanometer.

Angiotensin II was injected twice at doses of 10, 20 and 30 ng (stock solution : 200 ng/ml) and noradrenaline was injected twice at doses of 50, 100 and 200 ng (stock solution : 1 μg/ml).

At the end of the experiment the heart was weighed after careful dissection. The results obtained were analysed statistically by Student's *t* test.

Results

The age and body weight of the animals used is given in Table 1. HRS rats were significantly ($P < 0.05$) heavier than other animals.

For systolic blood pressure measured by the indirect method the mean values are also shown in Table 1. The differences between the three strains were highly significant ($P < 0.001$). When the mean blood pressure was measured directly in the carotid artery in anaesthetized and pretreated rats, there was still a difference between the three groups but, at this time, it was not significant (Table 1).

Vascular reactivity to noradrenaline

Results obtained are illustrated in Fig. 1, which shows that responses of HRS rats were significantly higher ($P < 0.01$) than those of NS and HOS, for the doses of noradrenaline used.

Vascular reactivity to angiotensin II

The values obtained are illustrated in Figure 2. No differences could be found between the three strains.

Heart weight

The heart weight expressed as g/kg body weight (±s.d.) were: 3.49 ± 0.06 g/kg for HRS rats, 3.01 ± 0.05 g/kg for NS rats and 3.20 ± 0.09 g/kg for HOS rats.

The heart weight of the HRS group was significantly ($P < 0.01$) higher than those of the

Table 1 Age, body weight and blood pressure (BP) in spontaneously hypertensive (HRS), normotensive (NS) and hypotensive (HOS) rats

	No. of animals	*Age (weeks)*	*Body weight (g)*	*BP in conscious rats* *Systolic BP (mmHg)*	*BP in anaesthetized rats after pretreatment* (mmHg)*
HRS	16	24 ± 0.2	232.8 ± 5	165 ± 2	65.8 ± 4
NS	10	23 ± 0.9	217.6 ± 4	149 ± 3	59.5 ± 6
HOS	10	23 ± 1.0	215.3 ± 3	138 ± 3	51.0 ± 3

*Animals anaesthetized with urethane (1.4 mg/kg, i.p.) and pretreated with pentolinium (2 mg/kg i.d.) and atropine sulphate (0.25 mg/kg, s.c.). All values are given with s.d.

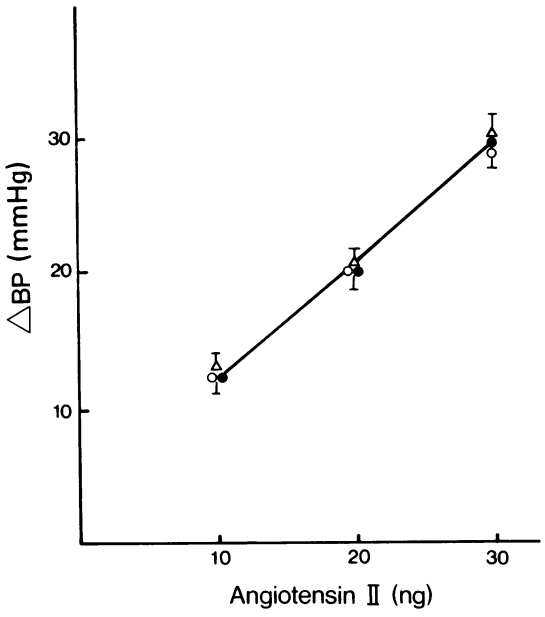

Fig. 2 Vascular reactivity to angiotensin II expressed as variation of blood pressure (△BP) evoked by injection of three doses of angiotensin II. Standard deviations are shown by the vertical bars. Hypotensive rats (○); normotensive rats (●); hypertensive rats (△).

two other strains, while there was no significant difference between NS and HOS rats.

Discussion

In the present study, the three strains used have been selected by successive inbreeding from the same three pairs of Sprague-Dawley rats. As emphasized by other investigators (Clineschmidt *et al.*, 1970; Dejong, Lovemberg & Sjoerdsma, 1972), this procedure is essential for the sake of comparison between the hypertensive animals and their controls.

Concerning the technique of vascular reactivity evaluation, this has been tested with the lowest hormonal concentrations leading to clearcut biological effects. Under these conditions, the maximum effect of the agonists could not be measured, but, according to the classical concept of Ariens (1954) the slope of the dose-response curves allows an evaluation of the affinity of the vascular receptors for the drugs used.

As previously described by Okamoto *et al.* (1966), Phelan (1966) and Folkow, Hallblack, Lundgren & Weiss (1972), the pretreatment showed (in absolute values) more marked hypotensive effects in HRS than in NS and HOS rats. However it must be noted that the blood pressure was reduced to the same extent, approximatively 60% in the three strains. Nevertheless pretreatment allowed reactivity to be studied in animals in which blood pressure was not significantly different at the beginning of the experiments. This is an essential point because, as emphasized by Phelan (1966), the initial blood pressure level can modify vascular responses to pressor agents. Thus a direct and precise evaluation of the vascular reactivity might be expected.

If HOS rats are compared with NS rats, no significant difference was found in the reactivity to noradrenaline and angiotensin II. Since we had previously found an identical cardiac output in these two strains of rats (Dupont, unpublished observations), the physiopathology of the hypotension observed remains unexplained. However, hyporeactivity to noradrenaline can be excluded on the results reported here.

In contrast, the responses to fixed doses of noradrenaline have been found significantly higher in HRS than in NS rats ($P < 0.01$) in spite of the greater body weight of the former strain. Other investigators, using different strains of spontaneously hypertensive rats reported such an increase (Okamoto *et al.*, 1966; McGregor & Smirk, 1968; Folkow, Hallblack, Lundgren & Weiss, 1970; Haeusler & Haefely, 1970) but this is not an invariable finding (Spector, 1969; Scholtysik & Unda, 1971; Fujiwara *et al.*, 1972). Such a discrepancy could probably arise from the nature of the strains used and more probably from widely differing experimental designs.

The vascular reactivity to angiotensin II has been found to be identical in HRS and NS rats. This is in agreement with the work of Scholtysik & Unda (1971) in conscious rats. However, with higher doses of this hormone an increase in its maximal effect (Shibayama *et al.*, 1971) as well as hyper-reactivity (McGregor & Smirk, 1968; Shibayama *et al.*, 1971) or a hyporeactivity (Phelan, 1968; Bing, 1969) have been reported.

In conclusion, in the experimental conditions used, it has been possible to demonstrate a significant hyper-reactivity to noradrenaline, but not to angiotensin II, in HRS when compared to NS rats. The results reported here suggest that an increased sensitivity to catecholamines might be one of the possible causes of the spontaneous hypertension developed in the strain selected.

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