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Potency, affinity constants and receptor reserves for noradrenaline and adrenaline on aortae from aged normo- and hypertensive rats

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Abstract

Previously we have determined the potency, affinity constants (K_A values), and α_1-adrenoceptor reserves for noradrenaline and adrenaline on the thoracic aortae of 20-week-old Wistar Kyoto normotensive (WKY) and spontaneously hypertensive rats (SHRs). This study has investigated whether these parameters were altered on the thoracic aortae by ageing, and in hypertension/heart failure. The effects of phenoxybenzamine on the contractile responses of the aortae of 20-month-old WKYs and SHRs were determined. The pD_2 values for noradrenaline and adrenaline were 7.1 and 7.0, respectively, on the aortae of 20-month-old WKYs, and similar values were obtained on age-matched SHRs. On the aortae of 20-month-old WKYs, the K_A values for noradrenaline and adrenaline were 1.85 and 1.95×10^{-6} M, and the receptor occupancies required for 50% maximum responses were 16 and 24%, respectively. There were lower affinities, by approximately twofold, but similar receptor reserves for noradrenaline and adrenaline on the aortae of age-matched SHRs. In comparison with the aortae of 20-week-old WKYs and SHRs, there was a 5-fold loss of sensitivity to noradrenaline and adrenaline between 20 weeks and 20 months. Between 20 weeks and 20 months there was a 50-fold loss of affinity with ageing and a further twofold loss with hypertension/heart failure, and an increase in α_1-adrenoceptor reserves for noradrenaline and adrenaline between 20 weeks and 20 months. There were no differences in the sensitivity and affinity, and minor changes in the α_1-adrenoceptor reserves for noradrenaline and adrenaline between the aortae of 20-month-old WKYs and SHRs. In contrast there were major changes in these parameters in the ageing of the WKY aorta from 20 weeks to 20 months. There were no additional changes in the sensitivity and α_1-adrenoceptor reserves, but a small additional change in affinity for noradrenaline and adrenaline in hypertension/heart failure on the aortae of 20-month-old SHRs.

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Introduction

Changes in vascular α_1-adrenoceptor mediated responses with age or disease may occur at the level of, or distal to, the receptors. Changes at the level of the receptor may be distinguished by studying the effects of irreversible blockers on the contractile responses of isolated tissues to agonists. Following such studies, the K_A value and the fractional occupancy–response relationships may be determined. The K_A value is an agonist constant with a unique value for each type of pharmacological receptor (Kenakin 1987). Thus if α_1-adrenoceptors change in nature with age or disease, the K_A values for α_1-adrenoceptor agonists such as noradrenaline and adrenaline will alter.

The responses to phenylephrine, an α_1-adrenoceptor agonist, on the rat aorta did not change with ageing between 3 and 29 weeks (de Oliveira et al 1998). The levels of α_{1A}-, $_{1B}$- and $_{1D}$-adrenoceptor mRNA declined in the rat aorta at 12 and 24 compared with 3 months (Xu et al 1997). Changes at the receptor level to stimulating aorta α_1-adrenoceptors with noradrenaline and adrenaline in aged rats have not been fully characterized.

In the early stages of hypertension (20 weeks) in spontaneously hypertensive rats (SHRs) there is no difference in sensitivity of the rat aorta to noradrenaline or adrenaline compared with age-matched Wistar Kyoto normotensive rats (WKYs) (Doggrell 1994). Although there is no change in sensitivity, there is an increase in the K_A values and receptor reserves for noradrenaline at the α_1-adrenoceptors of the SHR aorta at 20 weeks (Doggrell 1994). Aged SHRs are a non-invasive and realistic model of heart failure (Bing et al 1995; Doggrell & Brown 1998). The response to stimulating the aged SHR aorta α_1-adrenoceptors with noradrenaline or adrenaline has not been fully characterized.

The first part of our study compared the contractile responses of the aortae isolated from aged (20–21-month-old) WKYs and SHRs with the endogenous ligands, noradrenaline and adrenaline. We chose to use the endogenous ligands, as these are the agents that will be activating the α_1-adrenoceptors under physiological or pathological conditions. The major aim of the study was to determine whether the sensitivity, affinity and the fractional occupancy–response relationships for noradrenaline and adrenaline were altered by ageing and in hypertension-induced heart failure. We had previously determined these parameters at 20 weeks of age by studying the effects of phenoxybenzamine, an irreversible α_1-adrenoceptor antagonist, on the noradrenaline and adrenaline response curves of aorta from WKYs and SHRs (Doggrell 1994). In this study we used the same method with the aortae of 20–21-month old WKYs and SHRs, as this would allow us to compare our results with those obtained at 20 weeks.

Materials and Methods

Rats

Breeding pairs of WKYs and Okamoto SHRs were purchased from the Animal Resources Centre, Perth, Western Australia. Colonies of these rats were established in the Animal Resources Unit, School of Medicine, The University of Auckland. Adult rats were housed three to a cage with free access to standard rat chow and water.

Measurement of blood pressure

WKYs and SHRs 20–21-months-old were weighed, and then tail-cuff pressure (which approximates systolic blood pressure) was measured using a tail plethysmograph (model 29, Life Sciences Inc). Two similar readings of systolic pressure (mmHg) were obtained for each rat.

General

Rats were stunned and exsanguinated. The thoracic aorta was removed and placed in Krebs solution saturated with 5% carbon dioxide in oxygen. All of the experiments were performed in the presence of a modified Krebs solution (composition in mM: NaCl, 116; KCl, 5.4; $CaCl_2$, 2.5; $MgCl_2$, 1.2; NaH_2PO_4, 1.2; $NaHCO_3$, 22.0; D-glucose, 11.2) that was being bubbled with 5% CO_2 in O_2 at 37°C.

Contractility experiments

Each endothelium-intact thoracic aorta ring of approximately 3-mm length was suspended in a 5-mL organ bath under 1.5 g tension. Contractile responses were measured isometrically with force displacement transducers (Grass model FTO3.C) and displayed on a polygraph (Grass model 79B). Aortae were equilibrated for 60 min whilst 500 mL Krebs superfused the tissues. Each tissue was then cumulatively challenged with noradrenaline or adrenaline. Exposure to each concentration of noradrenaline or adrenaline was continued for a minimum of 3 min or, if a maximum response was not obtained in 3 min, until a maximum response was obtained. Tissues were allowed to recover in a wash of 500 mL Krebs over 60 min. Three of the rings were then treated with differing concentrations of phenoxybenzamine for 30 min while one tissue remained untreated. All rings were then washed with 500 mL drug-free Krebs over 60 min. The tissues were then challenged with noradrenaline or adrenaline for a second time.

At the end of each experiment the aortae were removed from the organ baths and the length was measured. The tissues were blotted, weighed, and the weights calculated as mg mm^{-1}.

Assessment of data

Responses to each concentration of noradrenaline or adrenaline, in the absence or presence of phenoxy-

benzamine, were measured and calculated as a percentage of the maximum obtained during the first challenge. pD_2 values (the negative logarithm of the molar concentration that causes 50% of the maximum response) were determined from regression line analyses over 20–80% of the maximum response in the first challenge.

The affinity constant (K_A) of noradrenaline or adrenaline was determined by the method of Furchgott & Bursztyn (1967). Noradrenaline or adrenaline response curves were obtained from untreated tissues and tissues that had been treated for 30 min with phenoxybenzamine. The following equation describes the relationship that exists between the concentration–response curve of an agonist before and after partial receptor inactivation with the irreversible antagonist phenoxybenzamine:

$$1/[A] = (1-q)/qK_A + 1/q[A'] \quad (1)$$

where [A] and [A′] are corresponding equieffective concentrations of agonist before and after partial irreversible receptor inactivation, respectively, and q is the fraction of active receptors remaining after partial irreversible blockade. K_A values were determined from plots of the reciprocals of noradrenaline or adrenaline concentration before fractional receptor inactivation (1/[A]) against the reciprocals of the corresponding equieffective concentrations of noradrenaline or adrenaline after receptor inactivation (1/[A′]) for individual curves. Furchgott & Bursztyn (1967) demonstrated that more accurate estimates of K_A values could be obtained if only the equieffective concentrations from the linear part of concentration–response curves were used in 'double reciprocal' plots. Consequently, we used the equieffective concentrations from the linear part of the curves and these yielded straight lines in accord with receptor theory. The K_A of noradrenaline or adrenaline was then calculated from the slope and intercept of the resulting 'double reciprocal' plots by the following equation:

$$K_A = (\text{slope} - 1)/\text{intercept} \quad (2)$$

Fractional α_1-adrenoceptor occupancy by noradrenaline or adrenaline was calculated for each bath concentration studied ([A]) using the individual and mean dissociation constant (K_A) values obtained from the interaction of noradrenaline or adrenaline with postjunctional α_1-adrenoceptors, according to the procedure of Ruffolo (1982). Thus the following relationship between agonist concentration ([A]) and dissociation constant was used to calculate α_1-adrenoceptor occupancy by noradrenaline or adrenaline:

$$\%\ \text{Receptor occupancy} = [A]/(K_A + [A]) \times 100 \quad (3)$$

The occupancy–response relationships were constructed by plotting the calculated α_1-adrenoceptor occupancy for noradrenaline or adrenaline against the corresponding response from the normalized concentration–response control curve.

The individual values (percentages, slope, pD_2 values, K_A values) obtained from the same age-group rats were compared by Student's unpaired *t*-test. Comparison between multi-groups involved analysis of variance testing followed by *t*-test. $P < 0.05$ was considered statistically significant. Mean values ± s.e.m. were also determined.

Drugs

Phenoxybenzamine HCl (RBI, Natick, MA) was dissolved at 10^{-1} M in absolute alcohol containing 10^{-2} M HCl. (−)-Noradrenaline bitartrate and (−)-adrenaline bitartrate (Sigma Chemical Co., St Louis, MO) at 10^{-1} M was dissolved in distilled water.

Results

Age and weight

The ages of the WKYs and SHRs were 635 ± 15 days (13) (mean ± s.e.m. (number of rats)) and 600 ± 7 days (17) and the weights were 430 ± 5 g (13) and 404 ± 6 g (17), respectively. The SHR tail-cuff (systolic) pressures were greater than those of the WKY (194 ± 3 (17) and 132 ± 1 mmHg (13), respectively, $P < 0.01$). The SHR aortae rings were heavier than those of the WKY (1.43 ± 0.03 (17) and 1.01 ± 0.03 mg mm^{-1} (13), respectively, $P < 0.01$).

Contractile responses to noradrenaline

Noradrenaline at $10^{-9} - 10^{-4}$ M contracted the aortae of aged WKYs and SHRs (Figure 1). The pD_2 values were obtained from normalized data and were 7.09 ± 0.10 (13) and 7.23 ± 0.11 (17), not significantly different, on the 20–21-month-old WKY and SHR aortae, respectively.

On the aged WKY aortae, 10^{-12} M phenoxybenzamine for 30 min had no effect and at 10^{-11} M abolished the noradrenaline responses (data not shown). Phenoxybenzamine at 2 or 5×10^{-12} M caused nonparallel rightward shifts of the aged WKY aortae noradrenaline responses with a reduction in maximum response (Fig-

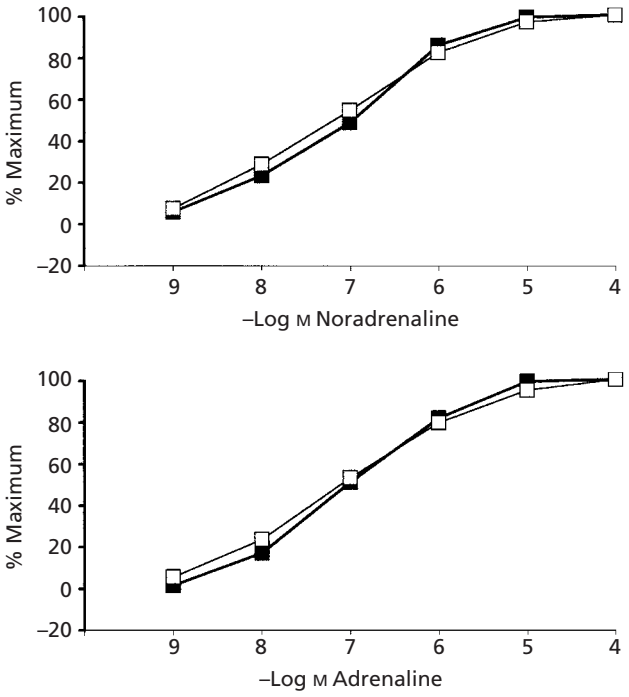

Figure 1 Contractile responses to noradrenaline and adrenaline on the aortae of 20–21-month-old WKYs (■) and SHRs (□). Responses were calculated as % maximum response and plotted against the −log of the molar concentration of noradrenaline or adrenaline. Each value is the mean ± s.e.m. from 13 (WKY) or 17 (SHR) aortae. The error bars are not shown when they are within the symbol size.

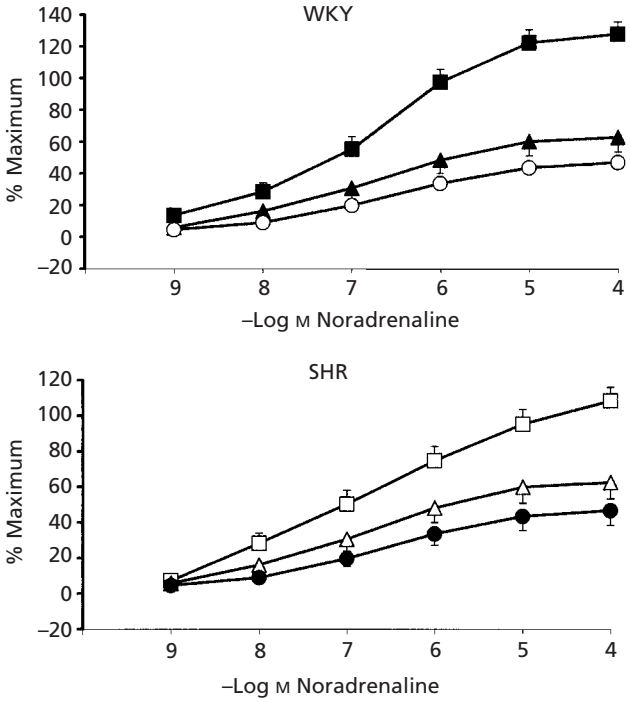

Figure 2 Effect of phenoxybenzamine treatment on the noradrenaline responses of 20–21-month-old WKYs and SHRs. WKY; mean ± s.e.m. from 13 untreated aortae (■), and 14 aortae treated with phenoxybenzamine at 2×10^{-12} M (▲) or 5×10^{-12} M (○). SHR; mean ± s.e.m. from 17 untreated aortae (□), and six or nine aortae treated with phenoxybenzamine at 2×10^{-11} M (△) or 5×10^{-11} M (●). Responses were calculated as % maximum response of the first (untreated) and plotted against the −log of the molar concentration of noradrenaline.

ure 2). The K_A values for noradrenaline were independent of phenoxybenzamine concentration, and were $1.9 \times 10^{-6} \pm 0.8$ M (14) and $1.8 \times 10^{-6} \pm 0.8$ M (14) with phenoxybenzamine at 2 and 5×10^{-12} M, respectively. Noradrenaline produced 20, 50 and 95% maximum responses by occupying 1.9 ± 1.2, 15.5 ± 5.7 and 81.9 ± 5.0% (14), respectively, of the aged WKY aorta α_1-adrenoceptors.

There was a lower affinity for noradrenaline but a similar α_1-adrenoceptor reserve for noradrenaline on the aged SHR compared with the WKY aortae. On the 20–21-month-old SHR aortae, 10^{-11} M phenoxybenzamine for 30 min had no effect and at 10^{-10} M almost abolished the noradrenaline responses (data not shown). Phenoxybenzamine at 2 or 5×10^{-11} M caused nonparallel rightward shifts of the aged SHR aortae noradrenaline responses with a reduction in maximum response (Figure 2). The K_A values for noradrenaline on the 20–21-month-old SHR were independent of phenoxybenzamine concentration, and were $4.2 \times 10^{-6} \pm 1.3$ M (6) and $4.7 \times 10^{-6} \pm 1.2$ M (9) with phenoxybenzamine at 2 and 5×10^{-11} M, respectively. These K_A values for noradrenaline on the aortae of aged SHRs were approximately twofold and significantly different ($P < 0.02$) from those on age-matched WKYs. Noradrenaline produced 20, 50 and 95% maximum responses by occupying 0.2 ± 0.2, 7.6 ± 3.9 and 86.2 ± 2.9% (9), respectively, of the aged SHR aortae α_1-adrenoceptors, and these values were not significantly different from the values on the age-matched WKYs.

Contractile responses to adrenaline

Adrenaline at 10^{-9}–10^{-4} M contracted the aortae of aged WKYs and SHRs (Figure 1) with similar pD_2 values (7.02 ± 0.10 (13) and 7.13 ± 0.11 (17), respectively).

On the WKY aortae, 2×10^{-10} M phenoxybenzamine had no effect and at 2×10^{-9} M almost abolished the adrenaline responses (data not shown). Phenoxybenzamine at 5×10^{-10} or 1×10^{-9} M caused nonparallel rightward shifts of the 20–21-month-old WKY aortae

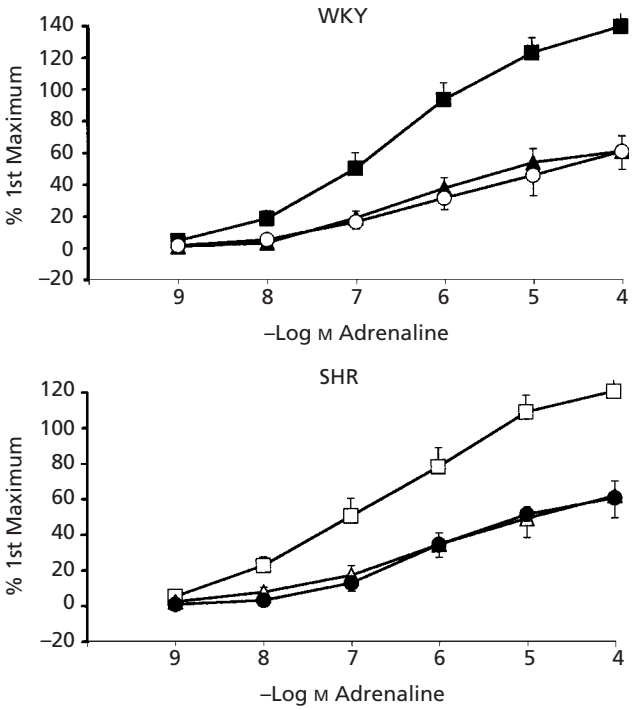

Figure 3 Effect of phenoxybenzamine treatment on the adrenaline responses of 20–21-month-old WKYs and SHRs. WKY; mean ± s.e.m. from 13 untreated aortae (■), and 13 or 12 aortae treated with phenoxybenzamine at 5×10^{-10} M (▲) or 1×10^{-9} M (○), respectively. SHR; mean ± s.e.m. from 17 untreated aortae (□), and six or nine aortae treated with phenoxybenzamine at 1×10^{-9} M (△) or 2×10^{-9} M (●), respectively. Responses were calculated as % maximum response of the first (untreated) and plotted against the −log of the molar concentration of adrenaline.

adrenaline responses with a reduction in maximum response (Figure 3). The K_A values for adrenaline were independent of phenoxybenzamine concentration, and were $2.1 \times 10^{-6} \pm 0.5$ M (13) and $1.8 \times 10^{-6} \pm 0.8$ M (12) with phenoxybenzamine at 5×10^{-10} and 10^{-9} M, respectively. Adrenaline produced 20, 50 and 95% maximum responses by occupying 3.4 ± 2.2, 24.0 ± 6.6 and 93.1 ± 2.0% (13), respectively, of the aged WKY aorta α_1-adrenoceptors.

As with noradrenaline, there was a lower affinity for adrenaline but a similar α_1-adrenoceptor reserve for adrenaline on the aged SHR compared with the WKY aortae. On the 20–21-month-old SHR aortae, 5×10^{-10} M phenoxybenzamine had no effect and at 5×10^{-9} M almost abolished the adrenaline responses (data not shown). Phenoxybenzamine at 1 or 2×10^{-9} M caused nonparallel rightward shifts of the aged SHR aortae adrenaline responses with a reduction in maximum response (Figure 3). The K_A values for adrenaline were independent of phenoxybenzamine concentration, and were $5.2 \times 10^{-6} \pm 1.6$ M (9) and $4.7 \times 10^{-6} \pm 1.2$ M (11) with phenoxybenzamine at 1 and 2×10^{-9} M, respectively. These K_A values for adrenaline on the aortae of aged SHRs were approximately twofold and significantly different ($P < 0.02$) from those on age-matched WKYs. Adrenaline produced 20, 50 and 95% maximum responses by occupying 0.6 ± 0.2, 11.5 ± 3.5 and 77.6% ± 6.5 (11), respectively, of the aged SHR aortae α_1-adrenoceptors, and these values were not significantly different from the values on the age-matched WKYs.

Discussion

Aged SHRs are a non-invasive and realistic model of hypertension-induced heart failure (Doggrell & Brown 1998). Bing et al (1995) tested many markers and found the most consistent marker of the SHR in failure was right ventricular hypertrophy. Bing et al (1995) divided their 18–24-month-old SHRs into two groups, SHR-F (failing) which had right ventricular hypertrophy and SHR-NF (non-failing) which did not have right ventricular hypertrophy. Using right ventricular hypertrophy as a marker of heart failure, all the SHRs in our colony had heart failure at > 18 months (Doggrell et al 1998; Nand & Doggrell 1999). Thus it is likely that the SHR aortae used in this study were from rats with heart failure.

In this study, the 20-month-old SHR aortae were heavier than those of the age-matched WKYs. The 14-week-old and older SHR aortae had a weight gain and this indicated that there was hypertension-associated hypertrophy of the aorta (Doggrell & Liang 1998). By 20 months, the SHR also had heart failure, which is often associated with tissue oedema, which may also contribute to the weight gain in the aortae.

Noradrenaline and adrenaline maybe inactivated by uptake into noradrenergic nerves or extraneuronal uptake into smooth muscle. In most studies with these endogenous agents, it is important to inhibit the neuronal (e.g. with cocaine) and extraneuronal (e.g. with normetanephrine) uptake process to work under equilibrium conditions. The rat thoracic aorta is an unusual blood vessel in that there is no evidence of noradrenergic innervation. The rat thoracic aorta content of noradrenaline is negligible and the responses to noradrenaline are not altered by the neuronal uptake inhibitor cocaine or by reserpine or 6-hydroxydopamine (Maling et al 1971). The thoracic aortae of 20-week and 20-month-old WKYs and SHRs are unresponsive to nerve stimulation (20-week, Doggrell (1995); 20-month,

Doggrell (unpublished observations)). The responses of 3–18-month-old WKY and SHR aortae to noradrenaline are not altered by normetanephrine (Arribas et al 1994). Therefore, on the rat thoracic aorta it is unnecessary to inhibit the neuronal or extraneuronal uptake process when studying the effects of noradrenaline and adrenaline.

On many blood vessels noradrenaline and adrenaline act on smooth muscle α_1-adrenoceptors to contract, and on β_2-adrenoceptors and endothelial α_2-adrenoceptors to relax. The β_2-adrenoceptor-mediated relaxation of the aorta is reduced or absent in ageing (O'Donnell & Wanstall 1984) and hypertension (Cohen & Berkowitz 1976; Doggrell 1994). On the endothelium-intact aorta of 5-week-old WKYs and SHRs, the relaxant responses to noradrenaline are inhibited by propranolol but not by yohimbine, and this indicates that β_2-adrenoceptors, but not endothelial α_2-adrenoceptors, are involved in the relaxation (Arribas et al 1994). The relaxant responses to high concentrations of noradrenaline are not apparent on the 3–18-month-old WKY and SHR endothelium-intact aorta (Arribas et al 1994), indicating the lack of α_2- and β_2-adrenoceptor-mediated relaxation at these ages. Thus it seems likely that noradrenaline and adrenaline were acting predominantly at smooth muscle α_1-adrenoceptors to contract the endothelium-intact WKY and SHR aortae in this study.

At present it is not clear which α_1-adrenoceptor is predominant in the rat aorta. The functional responses to phenylephrine of the 29-week-old Wistar rat aorta predominantly involves α_{1B}-adrenoceptors with some input from α_{1A}-adrenoceptors (de Oliveira 1998). The α_1-adrenoceptors of the WKY and SHR aorta seem to be different to those of the Wistar rat aorta. In the presence of propranolol, the responses of 10–12-week-old WKY and SHR aortae to noradrenaline are predominantly mediated by α_{1A}-adrenoceptors (Fujimoto 1994). In contrast, studies of the effects of antagonists on the responses of 6-month-old WKY and SHR aortae to methoxamine indicated that the functional receptor was α_{1D}-adrenoceptors (Villalobos-Molina & Ibarra 1996). Our study was designed to determine changes in responses to the endogenous ligands, noradrenaline and adrenaline, not receptor subtypes. Noradrenaline and adrenaline are equipotent at α_{1B}- and α_{1D}-adrenoceptors and noradrenaline is usually more potent than adrenaline at α_{1A}-adrenoceptors (Alexander & Peters 1999). Noradrenaline and adrenaline are equipotent on the aortae of 20-week-old (Doggrell 1994) and 20-month-old WKY and SHR (this study), suggesting that the functional α_1-adrenoceptor subtypes in these aortae are α_{1B} or α_{1D}. Further studies with α_1-adrenoceptor subtype selective agonists and antagonists are required to clarify which α_1-adrenoceptor subtype/s change with age in aorta.

Comparison of the pD_2 values for noradrenaline and adrenaline between the studies in 20-week-old WKYs (Doggrell 1994) and 20-month-old WKYs (this study) showed that there was a 5-fold loss of sensitivity of the aortae to noradrenaline and adrenaline between these ages. There was no change in sensitivity of the aortae to noradrenaline and adrenaline in early SHR hypertension (Doggrell 1994) or SHR hypertension-induced heart failure (this study).

In this study we used phenoxybenzamine as an irreversible antagonist at α_1-adrenoceptors. Higher concentrations of phenoxybenzamine than those inhibitory at α_1-adrenoceptors are required to inhibit responses mediated by α_2-adrenoceptors and serotonin (5-HT), muscarinic or histamine receptors. Phenoxybenzamine also inhibits the neuronal and extraneuronal uptake process for noradrenaline. As neither of these processes was important in the rat aorta (discussed previously), the inhibition was unlikely to have contributed to the effect of phenoxybenzamine in this study.

To be valid, the K_A values derived from contractility studies with irreversible antagonists should be independent of concentration of antagonist (Kenakin 1987). In keeping with this requirement, the K_A values for noradrenaline and adrenaline derived in this study were independent of the concentration of phenoxybenzamine used.

The major aim of our study was to determine whether the K_A values for noradrenaline and adrenaline and the fractional occupancy–response relationships were altered on the aorta with ageing or hypertension-induced heart failure. The K_A values for noradrenaline and adrenaline on the 20-week-old WKY aorta were 4.2 and 5.0×10^{-8} M, respectively (Doggrell 1994). Comparing those values with this study, the 20–21-month-old WKY showed that there was a large loss of affinity (50-fold) for noradrenaline and adrenaline on the aorta with ageing. This suggested that there were major changes in the α_1-adrenoceptors with age.

This has been the first study to show an increase in α_1-adrenoceptor reserve for noradrenaline and adrenaline with ageing. Noradrenaline produced 50 and 95% maximum responses by occupying 24 and 95% of the 20-week (Doggrell 1994) and 15 and 82% of the 20-month-old WKY aortae α_1-adrenoceptors (this study), respectively. A similar increase in receptor reserve also existed for adrenaline on the aortae between 20-week (Doggrell 1994) and 20-month-old WKY aortae (this study).

In this study, higher concentrations of phenoxybenzamine were required to inhibit the noradrenaline and adrenaline responses on the aged SHR than WKY aortae, and this may be indicative of a change in affinity of phenoxybenzamine for the α_1-adrenoceptor with hypertension. There was a twofold lower affinity, but similar receptor reserves, for noradrenaline and adrenaline on the aged SHR compared with the WKY aorta. This suggested that there were minor changes in the aortic α_1-adrenoceptors with hypertension-induced heart failure.

There was a 100-fold lower affinity to noradrenaline and adrenaline on the 20-month-old SHR (this study) compared with the 20-week-old WKY aorta (Doggrell 1994). Half of this affinity difference (50-fold) on the aorta during the continuation of SHR hypertension from 20 weeks to 20 months also occurred on WKY during this age period. Thus it seemed likely that age was responsible for most of the loss of affinity to noradrenaline and adrenaline on the SHR aorta between 20 weeks and 20 months. Hypertension was only responsible for twofold lesser affinity to noradrenaline and adrenaline on the 20-month-old SHR compared with aged-matched WKY aorta.

This finding of a loss of affinity to noradrenaline and adrenaline on the SHR aorta cannot be extrapolated to other agonists, age points and/or other SHR blood vessels. Nyborg & Bevan (1988) showed an increase in affinity for noradrenaline on 3-month-old SHR compared with the WKY mesenteric artery. A decreased receptor reserve for phenylephrine has been reported on the SHR mesenteric arteries at 13 weeks (Kojima et al 1989).

To our knowledge, this has been the first study to determine whether α_1-adrenoceptor K_A values and fractional occupancy relationships for noradrenaline and adrenaline were altered in ageing over a long period. We have shown a major loss of affinity accompanied by a loss of sensitivity and increase in α_1-adrenoceptor reserve for noradrenaline and adrenaline in the ageing of the WKY aorta. Only a minor additional loss of affinity to noradrenaline and adrenaline was observed in the SHR aorta.

It is not known whether similar alterations in α_1-adrenoceptors and their functional responses occur in the ageing of human blood vessels. If similar differences were apparent in the ageing of human blood vessels, there would be clinical implications. Firstly, as most pre-clinical pharmacology uses tissues from young animals, the therapeutic potential of selective α_1-subtype adrenoceptor agonists and antagonists may not have been correctly assessed. Secondly, changes in dose of commonly used α_1-adrenoceptor stimulants (e.g. pseudoephedrine in congestion) and antagonists (e.g. terazosin, doxazosin in hypertension and heart failure) with age may be required.

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