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LOW DOSE INDAPAMIDE PLUS PERINDOPRIL COMBINATION EFFECTS ON CARDIOVASCULAR STRUCTURE AND FUNCTION IN GENETIC HYPERTENSION

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SUMMARY

1. Although the fixed combination preparations of thiazide and angiotensin-converting enzyme inhibitor are gaining wide acceptance in clinical practice, data on the basic pharmacology of the combinations are relatively limited. The long-term structural and functional effects of a fixed low dose (0.24 + 0.76 mg/kg per day) combination of indapamide + perindopril (I + P, S5590) in spontaneously hypertensive rats (SHR) were examined in the present study.

2. Male SHR (10–12 weeks) were treated with I + P or vehicle for 8 weeks. The blood pressure and heart rate were monitored by weekly measurements. At the end of the treatment period, left ventricular, aortic and intramyocardial coronary arteriole structures were assessed. Contractile and relaxant properties of mesenteric arteries were determined by wire-myography.

3. Indapamide + perindopril combination caused a significant lowering of both systolic ($P < 0.001$) and diastolic ($P < 0.001$) blood pressures. Left ventricle plus septum:bodyweight ratio ($P < 0.001$), aortic medial cross-sectional area ($P < 0.05$) and media:lumen ratios ($P < 0.005$) were all significantly reduced by I + P treatment. In contrast, the effect of I + P on intramyocardial coronary vascular structure did not reach statistical significance. There was some improvement in endothelium-independent vasorelaxation of mesenteric vessels but contractile responses to noradrenaline and calcium were unaffected by treatment.

4. In summary, a low dose I + P combination treatment of SHR partly normalizes both systolic and diastolic blood pressures. Cardiac and larger vessel hypertrophy was reversed but intramyocardial coronary arteriole structure was not as readily regressed by the end of the study.

Key words: blood pressure, indapamide + perindopril, intramyocardial coronary arterioles, left ventricular hypertrophy, mesenteric vessels, myography, spontaneously hypertensive rat, vascular hypertrophy.

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INTRODUCTION

Cardiovascular structural changes are commonly associated with a persistent rise in blood pressure[1,2] and are important determinants of tissue perfusion. Over recent years, it has been suggested that blood pressure rises after cessation of antihypertensive therapy because of a failure to normalize left ventricular hypertrophy,[3] although a similar role for vascular structure is more controversial.[4] In the myocardium, persistence of vascular structural abnormalities may contribute to the disappointing reduction in the incidence of coronary events in patients with relatively well-controlled blood pressure.[5] Successful antihypertensive therapy may therefore require not only normalization of blood pressure but also reversal of cardiovascular structural abnormalities.

Indapamide and perindopril are both clinically established first-line antihypertensive drugs. Indapamide is an effective diuretic antihypertensive[6] that causes less marked effects on glucose and uric acid metabolism than the thiazides, although hypokalaemia may still occur.[7,8] Perindopril, a long-acting angiotensin-converting enzyme inhibitor (ACEI) has been shown to lower blood pressure and prevent cardiovascular hypertrophy in models of hypertension[9,10] and in patients.[11] Although both classes of drugs are effective as monotherapy, fixed low dose ACEI + diuretic combinations have been produced recently with a view to reducing the side effects associated with these drugs but moreover, to enhance the antihypertensive efficacy of the individual drugs.[12,13]

Recently, the clinical efficacy of a fixed low dose indapamide + perindopril (I + P, S5590) combination has been reported[14] and although the individual drugs have been extensively studied in both experimental animals and patients,[15,16] data on the basic pharmacology of the I + P combination preparation are relatively limited.[17] In particular, there are no data on the structural and functional effects of the I + P combination on small arteries.

The spontaneously hypertensive rat (SHR), a well-established model of genetic hypertension that develops a raised blood pressure in association with cardiovascular structural and functional abnormalities, was used for this study. Our aim was to determine the long-term effects of a fixed low dose I + P combination treatment in the SHR on blood pressure, vascular reactivity and cardiovascular structure.

MATERIALS AND METHODS

Blood pressure

Male spontaneously hypertensive rats (SHR) were obtained from Charles River (Kent, UK) and maintained in a controlled environment in the animal

facilities of our institution for at least 2 weeks prior to the performance of any procedure. The rats were housed in a light- and temperature-controlled room and fed on regular rat chow (Labsure, Cambridgeshire, UK) and tap water *ad libitum*. Spontaneously hypertensive rats at 10–12 weeks of age were randomly allocated to the control ($n = 8$) or the experimental groups ($n = 8$). The control group was treated with 1% suspension of hydroxyethylcellulose (vehicle) and the experimental group with the I + P combination (0.24 (I) + and 0.76 (P) mg/kg per day, respectively). Drug or vehicle was administered by daily gavage at 13.00 h for a total of 8 weeks. All treatments were in accordance with statutory regulations for the use of animals in scientific procedures at our institution.

The rats were familiarized with the tail-cuff procedure (IITC Apollo device; Linton Ltd, Norfolk, UK) for conscious blood pressure measurement for a few days prior to recording of basal blood pressures (week 0). Blood pressures were recorded weekly between 10.00 and 12.00 h, prior to dosing and recording of bodyweights. Blood pressure measurements were made in triplicate and the median value recorded. Fluid intake and urine output over a 24 h period were also measured weekly. After 8 weeks of drug treatment, rats were killed with a pentobarbitone overdose and the heart, thoracic aorta and mesenteric vascular tissues were removed. The right ventricle was dissected free from the left ventricle plus septum (LVS) and the wet weight of the latter was recorded. The LVS:bodyweight ratios were thus calculated. The wet weight and lengths of the thoracic aortas were also recorded. The LVS and thoracic aortas were then fixed in phosphate-buffered 2.5% glutaraldehyde and embedded in paraffin wax, prior to histological sectioning.

Morphometric analysis

Cross-sections of aortas (2 µm thick) were cut, stained with Harris' haematoxylin and eosin and examined by light microscopy. Two cross-sections per aorta were analysed with the aid of a video camera attached to the binocular lens of a microscope. The video images were projected onto a computer screen and morphometric analysis was undertaken by a software package (Seescan Solitaire Plus; Seescan, Cambridge, UK). The aortic outer and inner medial circumferences were traced by an individual blinded to the knowledge of whether the tissue was from the control or experimental group. The media, luminal cross-sectional areas and media:lumen ratios were thus calculated. The mean value for each of these vessel parameters was recorded for each animal.

The LVS were randomly orientated prior to embedding in paraffin wax. Two cross-sections (5 µm thick) were obtained from each LVS and stained with Miller's elastic and van Giesons' stains. These stains highlight the elastic laminae (internal and external) of arteries and arterioles and thus enable identification of the target vessels. Discrimination between arterioles and arteries was made on the basis that the external elastic lamina is absent in the former. All the available intramyocardial coronary arterioles ($n = 2$–6) on a LVS cross-section were analysed with the video analyser system described above, by an individual blinded to whether the tissue was from the control or experimental group. Coronary arteriolar outer and inner medial circumferences were traced and the media, luminal cross-sectional areas and media:lumen ratios were calculated. The median value for each of these vessel parameters was recorded for each animal.

Vascular reactivity

At the time of killing, the entire mesenteric vasculature was excised and third-order arteries (230–330 µm internal diameter) were used for assessment of vascular reactivity in a Mulvany-Halpern myograph (J. P. Trading, Aarhus, Denmark).[18] Briefly, vessels were dissected free from the mesenteric arcade and two arteries per rat were mounted as ring segments in an isometric myograph. The myograph contained 15 mL physiological saline solution (NaCl 118; KCl 4.7; $CaCl_2.6H_2O$ 2.5; $MgSO_4.7H_2O$ 1.17; $NaHCO_3$ 25.0; $NaH_2PO4.2H_2O$ 1.0; Na_2EDTA 0.03 and glucose 5.5 mmol/L) maintained at 37°C and aerated with 95% oxygen/5% carbon dioxide. The vessels were allowed to equilibrate for 1 h and then set at normalized internal circumference (L_{100}), estimated to be 0.9-fold the circumference they would maintain if relaxed and exposed to 100 mmHg transmural pressure. This was calculated for each vessel on the basis of the passive length-tension characteristics of the artery and the Laplace relationship. This procedure optimized active force generation by these vessels and the internal diameters referred to above are those derived from this calculation.

Vessel viability was always assessed prior to experimentation by exposing arteries to a depolarizing potassium solution (KPSS, same constitution as physiological saline solution with 118 mmol/L sodium replaced with 118 mmol/L potassium), followed by washout and exposure to noradrenaline (10 µmol/L) in the same solution. Vessels that failed to produce more than 100 mmHg effective pressure (as calculated from the Laplace formula) in response to an equimolar amount of potassium were excluded as they were considered to be non-viable. Cumulative dose–response relationships were obtained to noradrenaline (1 nmol/L–30µmol/L) and to calcium under depolarizing conditions (1 nmol/L–3 mmol/L). Depolarizing conditions were achieved by exchanging the physiological saline solution in the myograph with calcium-free potassium physiological solution.

Active wall tension (increase in wall force upon activation/twice segment length) was determined in response to noradrenaline and calcium for each vessel. The vasorelaxant effects of sodium nitroprusside (1 nmol/L–10 µmol/L) and acetylcholine (1 nmol/L–10 µmol/L) were assessed in vessels preconstricted with noradrenaline (10 µmol/L). Between each response curve, the vessels were contracted with KPSS to ensure continuing viability and then allowed to recover for 20–30 min prior to performing the subsequent response curve. The vascular functional data for each animal were recorded for analysis from the vessel giving the greatest active wall tension response to a depolarizing potassium solution.

Drugs

Indapamide/perindopril (S5590) combination was a gift from Laboratoires Servier, Courbevoie, France. All other drugs were purchased from Sigma, Poole, Dorset, UK.

Statistical analysis

All data are presented as mean±SEM (of *n* observations). Blood pressure, bodyweight, fluid intake and urine output data were compared using repeated measures analysis of variance. If a significant difference between the control and treated groups was detected, the Bonferroni multiple range test was used for *post hoc* analysis of data to determine statistical differences between groups at specific time points (SPSS statistics package, Chicago, IL, USA). Morphometric data were analysed by the two-tailed Student's *t*-test for unpaired data (Excel, Microsoft®, Reading, UK). Individual concentration–response data were fitted to a four parameter logistic function by non-linear regression: $Y = E_{max}/1 + (10^{c}/10^{x})^{D}$ where E_{max} is the maximal effect of the drug, C is the midpoint of the curve (log EC_{50}), X is the logarithm of the drug concentration and D is the Hill coefficient or slope factor. From this analysis the concentration of drug producing a half-maximal response, EC_{50}, was calculated. Non-linear regression was carried out on an IBM-compatible computer using a macro (written by ADH) in Microsoft Excel. Negative log EC_{50} values and maximal tensions or relaxations were compared by the two-tailed Student's *t*-test for unpaired data. $P < 0.05$ was considered significant for all tests.

RESULTS

The fixed low dose I + P combination at 0.24 (I) + 0.76 (P) mg/kg per day in 10–12-week-old SHR ($n = 8$) administered for 8 weeks was effective in reducing both systolic and diastolic blood pressures (Fig. 1), as compared with vehicle-treated controls ($n = 8$). Both systolic (191.2±9.2 *vs* 208.1±4.4; $P < 0.005$) and diastolic pressures (157.5±8.9 *vs* 173.1±6.4; $P < 0.02$) were significantly reduced by I + P from the first week of treatment as compared with the controls, with continued declines thereafter. By the end of 8 weeks of treatment, systolic (171.5±5.3 *vs* 223.1±6.5; $P < 0.001$) and diastolic pressures (147.5±3.5 *vs* 185.6±6.4; $P < 0.001$) were significantly

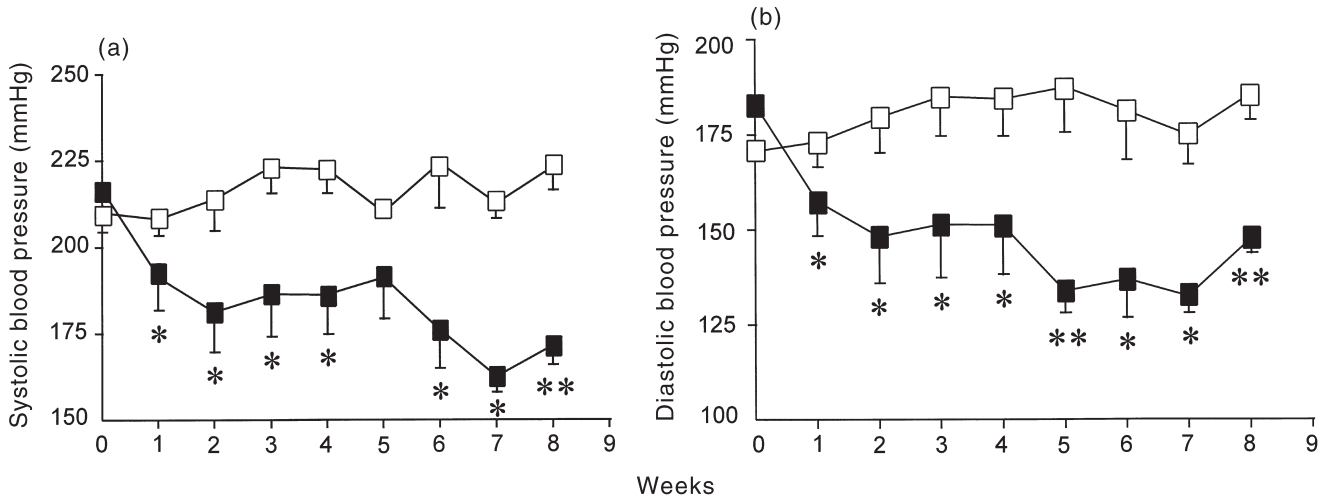

Fig. 1. Effect of a fixed low dose (0.24/0.76 mg/kg per day) combination of indapamide and perindopril (■, I + P, S5590) in spontaneously hypertensive rats on (a) systolic blood pressure and (b) diastolic blood pressure. Both systolic and diastolic blood pressures were significantly reduced ($P < 0.001$ by repeated measures ANOVA) by I + P treatment. Bonferonni comparisons between I + P and vehicle-treated groups at specific time points indicated that blood pressures were significantly reduced from week 1 onwards for the entire study period. One-way error bars are shown for clarity. Values are mean±SEM ($n = 8$ per group). □, vehicle; ■, I + P. *$P < 0.05$ and **$P < 0.01$ between control and I + P-treated groups.

reduced in the I + P-treated group. Heart rate remained unchanged throughout the treatment period (data not shown).

Fluid intake was slightly raised in the I + P-treated group (Table 1) throughout the study period but this effect was statistically insignificant ($P = 0.08$) as compared with the controls. Urine output was significantly ($P < 0.05$) increased by I + P treatment (Table 1), which demonstrates the incidence of diuresis by the second week of treatment. The diuretic effect was not sustained, although a statistically significant difference was achieved again in the last week of treatment. Bodyweights were unaffected by I + P treatment (325.6±8.7 *vs* 333.8±7.5; NS) as compared with the vehicle-treated controls.

By the end of the 8 week treatment period a significant reduction in LVS:bodyweight (mg/g) ratios (2.7±0.2 *vs* 3.2±0.2; $P < 0.001$) and left ventricular mass (wet weight/g) was observed (Table 2) in the combination-treated group. Aorta wall mass (g wet weight) normalized for vessel segment length/cm was also significantly reduced by I + P treatment (0.009±0.001 *vs* 0.011±0.002; $P < 0.05$). Aortic media:lumen cross-sectional area ratios and medial cross-sectional areas were also significantly reduced by I + P treatment (Table 2).

In the intramyocardial coronary arterioles there was a 12.2±1%, statistically insignificant ($P = 0.43$), reduction of the media:lumen

Table 1. Effect of a fixed low dose (0.24 + 0.76 mg/kg per day) combination of indapamide and perindopril (I + P, S5590) in spontaneously hypertensive rats on 24 h fluid intake and urine output

Week no.	1	2	3	4	5	6	7	8
Fluid intake (mL/24 h)								
Control	29.38±1.75	30.63±2.58	34.38±2.58	35.63±3.05	34.38±3.20	34.38±3.33	34.38±1.99	32.5±1.64
Indapamide/perindopril	29.38±1.13	38.13±2.30	35.63±1.99	35.63±1.99	39.38±2.20	35.63±1.75	36.88±1.62	39.38±2.58
Urine output (mL/24 h)								
Control	6.00±0.53	6.63±1.78	8.94±1.80	7.38±1.44	9.38±2.03	8.50±2.10	8.63±0.70	8.38±0.72
Indapamide/perindopril	4.69±0.70	12.75±2.67*	11.31±1.66	9.81±1.31	10.25±1.14	9.94±0.98	10.94±1.25*	12.19±1.56

Fluid intake and urine output were measured at weekly intervals. Fluid intake did not differ between control and treated groups ($P = 0.08$). Urine output was significantly increased ($P < 0.05$ by repeated measures ANOVA) with I + P treatment. Values are mean±SEM ($n = 8$ per group). *$P < 0.05$ between control and I + P-treated groups.

Table 2. Structural characteristics of the left ventricle plus septum, aorta and intramyocardial coronary arterioles after 8 weeks of treatment with a fixed low dose (0.24 + 0.76 mg/kg per day) combination of indapamide and perindopril (I/P, S5590) in spontaneously hypertensive rats

			Media cross-sectional area (μm^2)		Media:lumen cross-sectional area	
	LVS wet weight (g)	LVS/bodyweight ratio	Aorta	Imca	Aorta	Imca
Control	1.066±0.026	0.0032±0.0002	7646.7±300	79.93±16.5	0.425±0.009	3.61±0.5
Indapamide + perindopril	0.865±0.018	0.0027±0.0002	6676.4±187	78.19±17.2	0.362±0.014	3.17±0.26
Significance	$P < 0.001$	$P < 0.001$	$P < 0.05$	$P = 0.94$	$P < 0.005$	$P = 0.43$

Values are mean±SEM ($n = 8$ per group).

Imca, intramyocardial coronary arterioles; LVS, left ventricle plus septum.

Table 3. Reactivity characteristics of small mesenteric arteries after 8 weeks of treatment with a fixed low dose (0.24 + 0.76 mg/kg per day) combination of indapamide and perindopril (I + P, S5590) in spontaneously hypertensive rats

	Maximal tension (N/m)		Maximal relaxation (%)		$-\log EC_{50}$ (m)	
	Control	Indapamide/perindopril	Control	Indapamide/perindopril	Control	Indapamide/perindopril
Noradrenaline	5.13±0.74	5.85±1.48	–	–	5.86±0.31	5.77±0.14
Calcium	4.56±0.92	4.58±0.78	–	–	3.82±0.56	3.75±0.41
SNP	–	–	44.68±6.43	64.74±9.03	6.62±0.70	7.03±0.09
Acetylcholine	–	–	47.50±5.90	54.50±11.4	8.26±0.66	7.66±0.24

Data show contractile responses to noradrenaline (control, $n = 8$; I + P, $n = 7$) and calcium (control, $n = 8$; I + P, $n = 7$) expressed as a percentage of the maximal response to noradrenaline and calcium, respectively. Maximal relaxant responses to sodium nitroprusside (control, $n = 8$; I + P, $n = 7$) and acetylcholine (control, $n = 7$; I + P, $n = 6$) as a percentage relaxation of noradrenaline-preconstricted vessels are shown. EC_{50} (concentration of drug causing 50% of the maximal contractile or relaxant response as appropriate) are shown. All values are mean±SEM.

cross-sectional area ratios by I + P treatment. There was also a very small 2.2±0.05% ($P = 0.94$), statistically insignificant decrease in the medial cross-sectional areas by I + P treatment (Table 2).

Contractility of mesenteric small arteries to noradrenaline and calcium as assessed by maximal contractile responses and the sensitivity, expressed as the $-\log EC_{50}$, were not affected by I + P treatment (Table 3). Analysis of concentration–response curves of contractile data expressed as a percentage of the maximal response also did not demonstrate a difference between the control and I + P-treated groups (Table 3).

Vessels were also preconstricted with noradrenaline and the effect of the endothelium-independent vasodilator sodium nitroprusside was tested. Relaxant responses expressed as a percentage relaxation of the tone induced by 10 µmol/L noradrenaline indicated that sensitivity to nitroprusside ($-\log EC_{50}$) was increased as a result of I + P treatment but did not reach statistical significance (Table 3). There was also a 44.9±6%, but statistically insignificant ($P = 0.088$) improvement in the maximal relaxation responses of vessels to nitroprusside from treated animals (Fig. 2). The responses of noradrenaline-preconstricted vessels to the endothelium-dependent vasodilator acetylcholine from both control and I + P-treated groups failed to relax completely in response to acetylcholine. The maximal relaxation and the sensitivity to acetylcholine were relatively not affected by I + P treatment (Table 3).

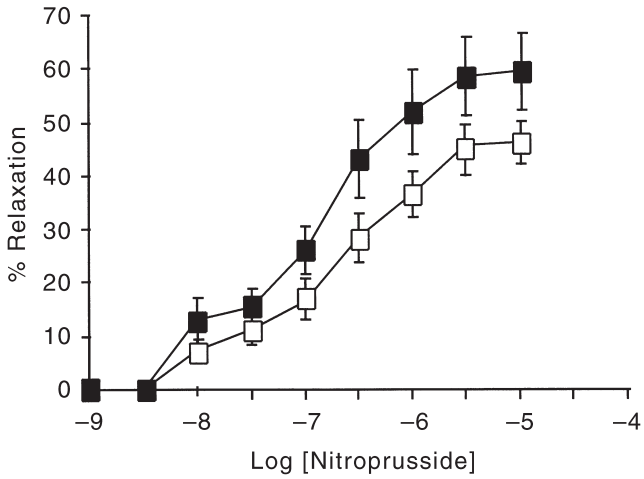

Fig. 2. Effect of a fixed low dose (0.24/0.76 mg/kg per day) combination of indapamide and perindopril (■, I + P, S5590) in spontaneously hypertensive rats on noradrenaline preconstricted mesenteric vessel relaxant responses to sodium nitroprusside. Relaxant responses are expressed as percentage relaxation of the tone induced by 10 µmol/L noradrenaline. Values are mean±SEM (control, $n = 8$; I + P, $n = 7$). □, vehicle; ■, I + P.

DISCUSSION

The main findings of this study are that a low dose I + P combination treatment of adult SHR for 8 weeks reduces both systolic and diastolic blood pressures. Blood pressures were reduced within 1 week of I + P treatment with variable further decline in pressures during the remainder of the study. Since all blood pressure measurements were made approximately 24 h following gavage, this suggests that the combination produces effective blood pressure control for at least 24 h. Indapamide + perindopril treatment also significantly reduced left ventricular and aortic wall hypertrophy but the effect on intramyocardial coronary arteriole structure did not reach statistical significance. Contractile reactivity of mesenteric vessels was not significantly affected by treatment.

Indapamide is a thiazide-like diuretic which our group has previously demonstrated to have acute vasorelaxant effects *in vitro*.[19] The direct relaxant action of indapamide in isolated resistance arteries appears to be mediated in part by a calcium antagonistic-like mechanism[20] and may also contribute to the inhibitory influence on tissue growth such as left ventricular[21] and aortic hypertrophy[22] when given chronically. The direct effect of I + P treatment on structural regression independent of a reduction in blood pressure is not known but merits investigation.

The effects of perindopril treatment in SHR on blood pressure are well known. Thus, a daily dose of 0.8 mg/kg of perindopril from 4 to 24 weeks has been reported to cause a 29% reduction in systolic blood pressure.[9] In another study, a daily dose of 0.76 mg/kg perindopril (equivalent to the perindopril dose in the combination used in this study) in 16-week-old SHR for 6 weeks[17] caused a 23% lowering of blood pressure. In the same report, treatment with an I + P combination at an identical dose to that used in the present study caused 30% reduction in systolic blood pressure. This is somewhat greater than the 14% blood pressure reduction observed in this study and may be related to the higher baseline pressures in our study that may be more resistant to antihypertensive treatment. A daily dose of 0.24 mg/kg indapamide (equivalent to the indapamide dose in the combination used in this study) has been reported to cause only an 8% reduction in systolic blood pressure.[17]

The antihypertensive effects of the I + P combination in relation to the blood pressure lowering effect of equivalent doses of perindopril and indapamide alone have been reported previously to

be relatively greater than the hypotensive effect achieved by either drug as monotherapy.[17] There are several theoretical reasons for an additive/synergistic antihypertensive action of indapamide and perindopril, arising from their activation of divergent mechanistic pathways which have a complementary effect on net efficacy. First, reflex activation of the renin–angiotensin system in response to diuretic-induced sodium loss causes blunting of the decrease in blood pressure and this is counteracted by perindopril, by decreasing angiotensin II formation. Second, diuretics also stimulate the release of catecholamines in response to a reduction in circulating blood volume or the decrease in blood pressure itself and this may antagonize the effects of both the diuretic and ACEI. Perindopril, in common with other ACEI, dampens the reflex activation of the sympathetic nervous system and thus enhances the antihypertensive effect.[13]

Left ventricular hypertrophy (LVH) is a common feature of experimental and clinical hypertension. To some extent, LVH is an adaptive response to the raised afterload but LVH is also recognized as a powerful independent risk factor[23] and its regression is therefore a desirable goal of antihypertensive therapy. In our study, LVS:bodyweight ratio was significantly reduced by I + P treatment to 2.7 mg/g (approximately 16%) in 18–20-week-old rats. This is comparable to the reduction in heart:bodyweight ratio found by others. For example, Campbell *et al*. showed that ventricle (left and right):bodyweight ratio was reduced to approximately 2.7 mg/g by perindopril (3 mg/kg) treatment from 6 to 20 weeks.[10] Similarly, Thybo *et al*. showed that a similar dose of perindopril to the one used in the present study in 4-week-old SHR for 16 weeks resulted in heart:bodyweight ratios of 2.68 mg/g (17% reduction).[9] Our study demonstrates that the same degree of heart:bodyweight ratio reduction can be achieved by I + P treatment of older SHR over a shorter treatment period. Moreover, the heart:bodyweight ratios in our study were reduced more effectively than those reported by Richard *et al*:[17] 3.10 mg/g (approximately 17%) using an equivalent dose of I + P in SHR from 16 to 28 weeks of age. This may be explained by the comparatively younger SHR in the present study although it is noteworthy that the relative reduction in heart:bodyweight ratios was fairly consistent between all of these studies.

It has been suggested that aortic hypertrophy is associated with a reduced vascular compliance and therefore confers an increased cardiovascular risk.[24] The aorta in clinical hypertension and various experimental models of hypertension is modified with a hypertrophied wall and an increased wall:lumen ratio. Indapamide + perindopril treatment of adult SHR in this study caused a marked regression of both the aortic media:lumen ratio and aortic medial cross-sectional area (CSA). This confirms the findings of an earlier report of aortic hypertrophy regression in SHR using an equivalent I + P dosing regimen.[17]

Although systolic and diastolic blood pressures, left ventricular and aortic hypertrophy were significantly reduced by I + P treatment in this study, the reduction in coronary arteriole media:lumen CSA did not reach statistical significance. Given that the arteriolar media:lumen CSA ratio was reduced by 12.4%, then it is possible that statistical significance was not attained due to the variability in our luminal measurements because the coronary vasculature was not pressure fixed. It has been suggested previously that a relaxed luminal diameter can be estimated from non-pressure-fixed vessels but such a process necessarily assumes inelasticity of the internal elastic lamina and this may not always be valid.[25] Alternatively, the limitations of such data can in some ways be circumvented by focusing on the changes in wall or medial CSA because this parameter has been shown to be unaffected by the physiological (contracted or relaxed) state of the vessel.[26] Thus, it is interesting to note in the present study that medial CSA was regressed in the coronary arterioles by 2.2±0.5% and in the aorta by 12.7±0.4%. The contrast in magnitude of effect between the two types of blood vessels may reflect the more prominent role of eutrophic remodelling in the former which is considered to be more difficult to regress than hypertrophic remodelling. A more detailed morphometric study of pressure-fixed vessels is required to determine whether this is the case.

There is growing evidence indicating conduit and small arterial endothelial dysfunction in hypertension. In the present study, aorta wall mass was significantly reduced by I + P treatment with a possible effect on endothelial function. Therefore, we examined small mesenteric arterial endothelium-dependent and -independent reactivity. We found there was no significant change in relaxation responses in mesenteric vessels from I + P-treated rats, although an insignificant increase in maximum relaxation in response to the endothelium-independent vasodilator sodium nitroprusside was noted. There is some evidence in the literature that ACEI can increase the levels of vascular cyclic guanosine 3′,5′-monophosphate[27] which may have contributed to the enhanced vasorelaxation observed in this study. Our findings, however, are in contrast to a previous study which found that 21 weeks of perindopril treatment in SHR younger than those used in this study, increased endothelium-dependent relaxation.[28] Whether this difference reflects the age of the animals, duration of treatment or the dose of drug combination used is unknown.

In conclusion, a low dose I + P combination is an efficacious antihypertensive agent which partially normalizes systolic and diastolic blood pressures and reverses cardiac and large vessel hypertrophy. Small intramyocardial coronary arteriole structure is not as readily regressed in adult SHR over the 8 week treatment period of this study. Taken together, these findings suggest a temporal dissociation between regression of cardiac and large vessel structure with that of small arterioles and may reflect differential involvement of local vascular factors. Finally, it is noteworthy that the ratio of indapamide to perindopril in the I + P combination used in this study is equivalent to that in a pharmaceutical formulation reported to be effective in clinical hypertension.[11,14,29] The beneficial effects of the low dose I + P combination on cardiovascular structure demonstrated in the present study suggest that use of such a combination may prove an effective antihypertensive therapeutic agent with desirable effects on left ventricular and large vessel hypertrophy.

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