Altered vascular contractility in adult female rats with hypertension programmed by prenatal glucocorticoid exposure

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Abstract

Excessive exposure to glucocorticoids during gestation reduces birth weight and induces permanent hypertension in adulthood. The mechanisms underlying this programmed elevation of blood pressure have not been established. We hypothesised that prenatal glucocorticoid exposure may lead to vascular dysfunction in adulthood. Pregnant rats received dexamethasone (Dex) (100 µg/kg, s.c.) or vehicle (control) daily throughout pregnancy. Blood pressure was elevated (students *t*-test, unpaired; $P < 0.05$) in adult female offspring (aged 12–16 weeks) of Dex-treated mothers (148.0 ± 3.6 mmHg, $n = 10$) compared with the control group (138.0 ± 2.5 mmHg, $n = 8$). Vascular responsiveness in aortae and mesenteric arteries was differentially affected by prenatal Dex: aortae were less responsive to angiotensin II, whereas mesenteric arteries were more responsive to norepinephrine, vasopressin and potassium (mesenteric arteries respond poorly to angiotensin II *in vitro*). Acetylcholine-mediated, endothelium-dependent relaxation was similar in both groups.

Prenatal exposure to Dex had no effect on blood pressure or aldosterone response to acute (15 min, i.v.) infusion of angiotensin II (75 ng/kg per min). In contrast, chronic (2-week, s.c.) infusion of angiotensin II (100 ng/kg per min) produced a greater elevation ($P < 0.05$) of blood pressure in Dex-treated rats (150.0 ± 3.6 mmHg) than in controls (135.3 ± 5.4 mmHg), and aldosterone levels were higher in Dex-treated animals. There was no angiotensin II-induced medial hypertrophy/hyperplasia in mesenteric arteries from Dex-treated rats. These results indicate that vascular function is altered in a region-specific manner in rats with glucocorticoid-programmed hypertension. Despite a striking increase in mesenteric artery contraction in Dex-treated rats, *in vivo* studies suggest that abnormalities of the renin-angiotensin-aldosterone system, rather than enhanced vascular contractility, may be responsible for the elevation of blood pressure in these animals.

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Introduction

Prenatal 'programming' of physiological responses (Barker *et al.* 1993), which could explain the association of low birth weight with increased risk of cardiovascular and metabolic disease in adulthood (Barker *et al.* 1990), has been attributed to several factors, including dietary modification (Langley-Evans *et al.* 1994, Woodall *et al.* 1996) and maternal stress during pregnancy (Lesage *et al.* 2001). Excess exposure of the foetus to glucocorticoids may provide a common mechanism for the programming response (Edwards *et al.* 1993). Indeed, in rats, maternal undernutrition produces a rise in maternal cortisol (Fowden 1995) and impaired glucocorticoid inactivation in the placenta (Langley-Evans *et al.* 1996, Nyirenda & Seckl 1998, Lesage *et al.* 2001). Furthermore, the link between glucocorticoids and adult hypertension is well

established, with prenatal glucocorticoid administration (to rats and sheep) inducing low birth weight, impaired glucose tolerance and elevated blood pressure in adult offspring (Benediktsson *et al.* 1993, Lindsay *et al.* 1996, Dodic *et al.* 1998).

The mechanisms by which prenatal programming induces adult hypertension have not been established. One possibility is that changes in peripheral vascular function contribute to elevation of blood pressure; indeed, enhanced contractility has been demonstrated in femoral arteries isolated from rats exposed to maternal dietary restriction (Ozaki *et al.* 2001). Glucocorticoids could be implicated in programming of vascular function since they alter the function of both endothelial (Johns *et al.* 2001) and vascular smooth muscle cells (Ullian *et al.* 1996, Souness *et al.* 2002). Consequently, infusion of the synthetic glucocorticoid betamethasone elevates blood

pressure and alters vascular contractile and dilator function in preterm lambs (Gao *et al.* 1996, Anwar *et al.* 1999, Docherty *et al.* 2001). The effects of these interactions may extend into later life as prenatal exposure results in permanent changes, such as elevated glucocorticoid receptor expression in adult rats (Nyirenda *et al.* 1998), in glucocorticoid signalling pathways.

We tested the hypothesis that prenatal exposure to glucocorticoids in rats results in altered vascular function in adult offspring. We used an established model of glucocorticoid-mediated, programmed hypertension (Benediktsson *et al.* 1993) to determine whether exposure to the synthetic glucocorticoid dexamethasone (Dex) *in utero* produced: 1) altered contractile and relaxant function in isolated (conduit and resistance) arteries *in vitro* and 2) altered vasopressor response *in vivo*. We investigated only female offspring, as recent studies (Khan *et al.* 2003, Ortiz *et al.* 2003) and our own experience (O'Regan *et al.* 2004) suggest that females are more likely to develop hypertension in response to prenatal programming.

Materials and Methods

Rats

As in previous studies, female Wistar rats (200–250 g; $n=26$) received daily s.c. injections of Dex (100 µg/kg; $n=14$) or vehicle (0.1 ml 4% ethanol-saline $n=12$) throughout pregnancy (Benediktsson *et al.* 1993). The number of pups per litter (16 ± 0.6 pups from saline-treated mothers) was reduced ($P<0.02$) by prenatal Dex (13 ± 0.9 pups from Dex-treated mothers), but the ratio of males to females was unaffected. This was accompanied by a reduction ($P<0.01$) in pup weight (3.77 ± 0.03 g in the saline group vs 3.36 ± 0.05 g in the Dex group): weight reduction was seen in both male and female pups. No death was observed in either group, litters were culled to eight pups and animals were randomly selected from different litters for subsequent experimentation. Stage of cycle was not taken into account at the time that measurements were made. Experiments conformed to *The Principles of Animal Care* (NIH publication no. 85–23, revised 1985) and were performed under the UK Home Office Animals (Scientific Procedures) Act 1986.

Materials

Salts were obtained from BDH, Poole, Dorset, UK; nor-epinephrine hydrochloride (NE), angiotensin (Ang) II, arginine vasopressin (AVP) and acetylcholine chloride (ACh), from Sigma.

Influence of prenatal Dex on vascular function *in vitro*

Blood pressure was measured in adult female (16 weeks) offspring by tail cuff plethysmography (Evans *et al.* 1994), on four different days, by an operator blinded to treatment.

Rats were killed by decapitation. Functional assessment of aortae and mesenteric arteries was performed by standard organ bath (Martin *et al.* 1986) and small-vessel myograph (Falloon *et al.* 1995) techniques respectively. Isometric force was measured in vessels suspended in physiological salt solutions (PSS) at 37 °C, perfused with carbogen. The arteries were equilibrated at their optimum resting settings (2.0 g (Martin *et al.* 1986) and 0.9 L100 (Falloon *et al.* 1995)) and subjected to a standard start procedure (Martin *et al.* 1986, Falloon *et al.* 1995).

Protocol Cumulative concentration-response curves were obtained with NE (10^{-9} – 3×10^{-6} mol/l for aortae; 10^{-9} – 3×10^{-5} mol/l for mesenteric artery), potassium (KCl; 2.5×10^{-3} – 3.2 mol/l for aortae; high-potassium PSS (KPSS), 2.5×10^{-3} – 0.125 mol/l for mesenteric arteries), Ang II (10^{-11} – 1×10^{-6} mol/l) and AVP (10^{-11} – 1×10^{-6} mol/l). Responses to the endothelium-dependent vasodilator ACh (10^{-9} – 3×10^{-5} mol/l), were obtained after contraction with sufficient NE (3×10^{-8} – 10^{-5} mol/l) to achieve ~80% of the maximum contraction.

Influence of prenatal Dex on the pressor response *in vivo*

Acute Three days prior to infusion, cannulae were inserted, under halothane anaesthesia (Fluothane, Zeneca, UK), into the carotid artery of adult (12 weeks), female pups (Benediktsson *et al.* 1993). Blood pressure was measured directly in conscious, unrestrained rats with the Elcomatic pressure transducer (Glasgow, UK) prior to, and during, venous infusion of Ang II (75 ng/kg per min for 15 min) (Brown *et al.* 1981). Blood samples were taken for measurement of plasma aldosterone.

Chronic Blood pressure was measured in adult (12 weeks), female offspring by tail cuff plethysmography (Evans *et al.* 1994) on three separate days. Minipumps (Model 2002; Alzet, Cupertino, CA, USA) were implanted, under halothane anaesthesia, to infuse Ang II (Hypertensin; Ciba Geigy, Basel, Switzerland), 100 ng/kg per min, or vehicle s.c. for 2 weeks. Rats were killed by decapitation, trunk blood was collected for measurement of hormones, sections of the mesenteric arcade were fixed in 10% neutral buffered formalin for histological analysis, and tissues were retrieved and weighed.

Hormone assays

Plasma renin activity was measured as Ang I generated from plasma incubated at 37 °C for 30 min (Miller *et al.* 1980). Ang I, corticosterone (Kenyon *et al.* 1993) and aldosterone (DPC, Los Angeles, CA, USA) concentrations were measured by RIA.

Histological analysis

Fixed vessels were embedded in paraffin wax and 3 µm sections stained with elastic van Giessen. Image analysis

Table 1 The influence of prenatal Dex on contraction and relaxation of aortae from adult offspring

	E_{max} (mN/mm or % maximum relaxation)		Sensitivity (pD ₂ or -log IC ₅₀)	
	Vehicle	Dex	Vehicle	Dex
Ang II	0.97 ± 0.28 (5)	0.59 ± 0.17 (6)	9.13 ± 0.15 (5)	8.58 ± 0.10 (6)**
NE	3.47 ± 0.72 (6)	2.61 ± 0.39 (8)	7.94 ± 0.05 (6)	7.78 ± 0.06 (8)
AVP	4.57 ± 0.43 (6)	3.99 ± 0.36 (8)	8.41 ± 0.06 (6)	8.51 ± 0.09 (8)
KCl	4.64 ± 0.48 (6)	4.60 ± 0.35 (8)	1.56 ± 0.12 (6)	1.51 ± 0.10 (8)
ACh	142.0 ± 51.9 (5)	138.6 ± 21.5 (8)	7.59 ± 0.17 (5)	7.52 ± 0.12 (8)

Values are mean ± S.E.M. and were compared by Student's two-sample *t*-test; ***P* = 0.01. Sensitivity values are quoted as the -log of the concentration required to produce 50% contraction (pD₂) or 50% inhibition of precontractile tone (-log IC₅₀). Ang II: angiotensin II; NE: norepinephrine; AVP: arginine vasopressin; ACh: acetylcholine.

was performed by an operator blinded to treatment, using MCID-M4 software (Imaging Research Inc, Brock University, St. Catharines, Ontario, Canada).

Statistics

Values are mean ± S.E.M. for *n* pups from different mothers with the exception of *in vivo* data, where *n* refers to number of pups from five vehicle-treated and four Dex-treated mothers. Data were analysed by two-way, one-way or repeated-measures ANOVA with Tukey's multiple-comparisons test or Student's *t*-test, as appropriate. Significance was set at *P* < 0.05.

Results

Influence of prenatal Dex on vascular function in vitro

At 16 weeks of age, female pups exposed to Dex *in utero* had elevated systolic blood pressure (control 138.0 ± 2.5 mmHg, *n* = 8; Dex 148.0 ± 3.6 mmHg, *n* = 10; *P* < 0.05).

Aorta

The sensitivity of aortic rings to Ang II was reduced by prenatal Dex exposure. A similar magnitude reduction in sensitivity to NE did not achieve significance (*P* = 0.07) (Table 1). Maximum contractions and endothelium-dependent relaxation were unaffected by prenatal exposure to Dex (Fig. 1; Table 1).

Mesenteric artery

Mesenteric arteries from control and Dex groups were similar in size (internal diameter 282 ± 14 µm (*n* = 6) and 274 ± 17 µm (*n* = 8) respectively; *P* = 0.74). Contractile responses to NE, KCl and AVP were enhanced in mesenteric arteries from the Dex group (Fig. 1; Table 2). The sensitivity of the mesenteric artery to AVP (Table 2)

was unaffected by prenatal Dex treatment (*P* = 0.06). Ang II produced virtually no contraction in arteries from the Dex group and an unreproducible, spasmodic response in controls (data not shown). ACh-mediated relaxation of mesenteric arteries was unaffected by prenatal exposure to Dex (Table 2).

Influence of prenatal Dex on pressor response in vivo

Acute pressor response The elevation of basal blood pressure measured by carotid cannulation in Dex-exposed pups was again apparent (Table 3). In both groups of rats, Ang II infusion resulted in an acute rise in blood pressure evident a few seconds after the onset of the infusion (Table 3; ANOVA, *P* < 0.01). There was no difference, however, in the incremental rise in blood pressure in response to Ang II (control Δ18.5 ± 6.2 mmHg, *n* = 6, Dex Δ16.5 ± 1.7 mmHg, *n* = 5).

Acute infusion of Ang II, which does not allow time for compensatory suppression of renin, increased plasma aldosterone levels 2–3-fold (Table 3; *P* < 0.05 in both groups). Basal aldosterone levels were not different in control and Dex-treated groups. After Ang II infusion, there were no differences between Dex and control groups in either the absolute values of plasma aldosterone concentration (Table 3) or the incremental rise from baseline (control Δ2330 ± 526 pM, *n* = 6 vs Dex Δ1888 ± 431 pM, *n* = 5).

Chronic pressor response Immediately before starting treatment of 12-week-old pups, mean arterial pressure was higher in the Dex group (control 118.9 ± 1.4 mmHg, *n* = 16; Dex 123.4 ± 1.9 mmHg, *n* = 16; *P* = 0.05). After 7-day treatment with Ang II, blood pressure had increased in both groups (*P* < 0.01) (Fig. 2). The Dex/Ang II group had a higher blood pressure than the control/Ang II group (150.0 ± 3.6 mmHg, *n* = 8 and 135.3 ± 5.4 mmHg, *n* = 8; *P* < 0.05). The increment in blood pressure compared with pretreatment values (Fig. 2 inset) was greater in

Figure 1 The influence of prenatal Dex on contractile responses of aortae and mesenteric arteries taken from adult offspring. Prenatal exposure to Dex had no effect on aortic contraction (a–c), but enhanced contractile responses to (d) norepinephrine, (e) arginine vasopressin and (f) KCl in mesenteric arteries ($\blacktriangle$: Dex, $n=8$; $\blacksquare$: control, $n=4$ –6). Each point represents mean $\pm$ S.E.M. Curves were compared by two-way ANOVA.

prenatal Dex-exposed rats, indicating that they were more sensitive to Ang II than the control group ($P<0.04$).

Plasma corticosterone levels were similar in Dex- and vehicle-exposed adult female rats and were unaltered by Ang II infusion (Table 4a). Plasma renin activity was similar in vehicle- and Dex-treated rats (Table 4a). Plasma renin suppression by Ang II was evident in control and Dex-treated groups (Table 4a); Ang II suppressed plasma renin activity (PRA) to similar levels in both groups, reflecting an 81% reduction in activity in Dex-treated animals compared with a 50% reduction in controls.

In contrast to the acute Ang II study, the elevation of basal plasma aldosterone levels detected in Dex-treated rats was significant compared with controls, but chronic Ang II infusion did not produce a significant increase in plasma aldosterone in either group (Table 4a).

Image analysis was performed on 3–13 arteries of similar size (Table 4b) from each rat. The medial areas of mesenteric arteries from rats exposed to Dex *in utero* were smaller than controls (Table 4b), but no difference in medial thickness was observed between Dex and vehicle rats after chronic infusion of Ang II. These values were

Table 2 The influence of prenatal Dex on contraction and relaxation of mesenteric arteries from adult offspring

	E_{max} (mN/mm or % maximum relaxation)		Sensitivity (pD ₂ or -log IC ₅₀)	
	Vehicle	Dex	Vehicle	Dex
NE	3.36 ± 0.71 (6)	4.42 ± 0.42 (8)	5.54 ± 0.12 (6)	5.67 ± 0.08 (8)
AVP	2.87 ± 0.72 (6)	4.16 ± 0.40 (8)	9.86 ± 0.15 (6)	9.16 ± 0.27 (8)
KCl	2.61 ± 0.56 (4)	4.52 ± 0.53 (8)*	1.36 ± 0.03 (4)	1.34 ± 0.03 (8)
ACh	75.3 ± 11.8 (6)	81.1 ± 10.5 (8)	7.25 ± 0.34 (6)	7.27 ± 0.20 (8)

Values are mean ± S.E.M. and were compared by Student's two-sample *t*-test; **P* = 0.03. Sensitivity values are quoted as the -log of the concentration required to produce 50% contraction (pD₂) or 50% inhibition of precontractile tone (-log IC₅₀). NE: norepinephrine; AVP: arginine vasopressin; ACh: acetylcholine.

reinforced by lack of differences in media:lumen ratios in saline- and Ang II-treated groups (Table 4b). Neither Dex treatment *in utero* nor Ang II treatment in adulthood led to changes in organ (adrenal, kidney, liver, heart and thymus) weights (data not shown).

Discussion

This investigation demonstrated enhanced contractility in mesenteric arteries, but not aortae, from rats with glucocorticoid-programmed hypertension. Functional changes in the mesenteric arteries were not, however, associated with an increase in the acute pressor response to Ang II *in vivo*. This suggests that increased Ang II-dependent vasoconstriction does not contribute to elevated blood pressure in these animals. Similarly, enhanced sensitivity to chronic blood pressure elevation in response to Ang II was probably the result of increased basal renin-angiotensin-aldosterone system (RAAS) activity rather than to changes in the resistance vasculature. We conclude that a non-selective, systemic change in vascular function and structure is unlikely to contribute to hypertension programmed by exposure to glucocorticoids *in utero*.

Glucocorticoid exposure, whether in rats *in utero* (Docherty *et al.* 2001) or to rat arteries *in vitro* (Ullian *et al.*

1996), can influence contraction by selective upregulation of contractile receptors in the vascular smooth muscle and/or by altering the production of endothelium-derived nitric oxide (Anwar *et al.* 1999, Johns *et al.* 2001). Impaired endothelium-dependent relaxation has been reported in arteries from rats after maternal dietary restriction (Lamireau *et al.* 2002, Khan *et al.* 2004, 2005). The present study, however, consistent with another recent report (Ozaki *et al.* 2001), found no impairment of endothelium-dependent relaxation in either aortae or mesenteric arteries from the Dex-treated rats. It is unlikely, therefore, that programmed endothelial cell dysfunction contributes to the elevation of blood pressure in this model.

Glucocorticoid-dependent changes in contractile function are agonist-dependent and involve several distinct mechanisms; increased α -adrenergic contraction appears to be receptor-independent (Smith *et al.* 1987), whereas enhanced Ang II-mediated contraction is secondary to AT₁-receptor upregulation (Ullian *et al.* 1992). Mechanisms for other agonists are less well established (Ullian 1999). In the current study, the comparison of functional results from aortae and mesenteric arteries indicates that, as in prenatal protein restriction in rats (Torrens *et al.* 2003) or prenatal Dex administration in sheep (Docherty *et al.* 2001), functional changes in Dex-programmed hypertension vary in arteries from different anatomical locations.

Table 3 Acute effects of Ang II infusion on blood pressure (mmHg) and plasma aldosterone concentration (pM) in control and Dex rats

	Treatment			
	Control/saline (n=6)	Control/Ang II (n=6)	Dex/saline (n=5)	Dex/Ang II (n=5)
Systolic	129.6 ± 4.7	147.8 ± 5.8**	146.3 ± 6.1	162.0 ± 5.5**
Diastolic	100.8 ± 2.5†	119.5 ± 7.9**	118.9 ± 6.6	135.8 ± 6.4**
Mean	110.4 ± 1.7†	128.9 ± 6.6**	128.0 ± 6.1	144.6 ± 5.7**
Plasma aldosterone	1254 ± 131	3584 ± 571**	1888 ± 494	3775 ± 676**

Values are mean ± S.E.M. ***P* < 0.01 compared with appropriate saline-treated group and †*P* < 0.05 compared with appropriate Dex-treated group.

Figure 2 Serial blood pressures before (dotted lines) and after (solid lines) treatment with Ang II in adult rats exposed to dexamethasone (Dex: ●/■, *n*=8), or vehicle (control: ○/□, *n*=8), *in utero*. Repeated-measures ANOVA indicates that blood pressure was significantly higher in animals treated with Dex *in utero* (*P*<0.05). Inset compares Ang II responses in Dex and control groups. Each point represents mean ± S.E.M.

In the aorta, the failure of prenatal glucocorticoid exposure to enhance contractility (indeed, the tendency was for reduced contraction) suggests that there is no upregulation of contractile receptors in this vessel. Similarly, the enhanced contraction observed in mesenteric arteries from the Dex-treated rats was receptor-independent – the failure of Ang II to contract resistance arteries in isometric systems has been reported previously (Falloon *et al.* 1995). Given that endothelium-dependent relaxation is unaltered in these animals, the enhanced contraction in mesenteric arteries is probably due to changes in the vascular

smooth muscle. Histological analysis indicated that structural remodelling was not a factor, suggesting, therefore, changes in post-receptor signal transduction. This would be consistent with persistence into adult life of glucocorticoid-mediated modulation of G-proteins (Haigh *et al.* 1990), second messengers (e.g. inositol triphosphate (Sato *et al.* 1992)) or sodium–calcium ion exchange (Smith & Smith 1994). Acute and chronic infusions of Ang II *in vivo* were used to assess different mechanisms of blood pressure regulation. The use of Ang II is particularly relevant, as changes

Table 4. Influence of prenatal Dex on the response of plasma hormones and mesenteric artery structure to chronic Ang II infusion

	PRA (ng Ang I/ml per h)		Aldosterone (pM)		Corticosterone (nM)	
	Saline-treated	Ang II-treated	Saline-treated	Ang II-treated	Saline-treated	Ang II-treated
(a) Plasma hormones.						
Control (8)	0.54 ± 0.06	0.27 ± 0.09**	990 ± 127	800 ± 101	590 ± 77	496 ± 102
Dex (8)	0.64 ± 0.07	0.12 ± 0.06**	1171 ± 82 [†]	1348 ± 252 [†]	660 ± 71	624 ± 56
(b) Mesenteric artery structure	Artery diameter	Medial area		M:L ratio		
	(mm)	(mm ²)	(% within extended elastic laminae)			
Control (4)	127 ± 9	0.012 ± 0.001	62 ± 3	1.84 ± 0.20		
Control/Ang II (4)	104 ± 6	0.008 ± 0.001	61 ± 7	2.11 ± 0.56		
Dex (4)	112 ± 11	0.007 ± 0.001*	47 ± 7	1.08 ± 0.26		
Dex/Ang II (4)	99 ± 13	0.009 ± 0.002	68 ± 4	1.68 ± 0.21		

Values are mean ± S.E.M., where *n* indicates the number of different pups. Data were compared by ANOVA with Tukey's multiple-comparisons test. **P*<0.05 compared with control; ***P*<0.01 compared with saline-treated group; [†]*P*<0.05 compared with control animals; PRA: plasma renin activity.

in the RAAS are implicated in programmed elevations of blood pressure: for example, AT₁-receptor antagonism in early postnatal life prevents development of hypertension after maternal dietary restriction in the rat (Sherman & Langley-Evans 1998). Acute administration of a pressor dose of the hormone assesses immediate, AT₁-receptor-mediated changes in steroidogenesis and vasoconstriction. Adrenal sensitivity to Ang II was not different, as indicated by similar elevation of aldosterone levels in both groups. Continuous infusion of Ang II below the threshold required for an immediate vasoconstrictor response is a widely used protocol for investigating long-term pressor effects, including hypertrophy and hyperplasia of smooth muscle cells in resistance arteries (Griffin *et al.* 1991, Su *et al.* 1998). Over this time course, elevated aldosterone levels are not sustained, although PRA was markedly suppressed, reflecting adrenal compensation for high Ang II levels, as is well recognised. Importantly, however, aldosterone levels were higher in Dex-treated animals at baseline and after Ang II, and PRA was not suppressed in the face of systemic hypertension. This suggests that there is increased RAAS activity in the basal state in the Dex-programmed animals, either as a result of a primary abnormality of renin secretion or perhaps secondary to sympathetic nervous system activation (Pladys *et al.* 2004). This is consistent with previous results with a different Dex paradigm in the rat, suggesting programming of increased RAAS activity (O'Regan *et al.* 2004). It is surprising, however, that Ang II-mediated suppression of PRA is increased in rats exposed to Dex *in utero*. The mechanism responsible for this alteration cannot be identified in the present study, although it may relate to higher blood pressure response with increased renal perfusion pressure.

The lack of effect of Dex on pressor responses to acute Ang II, consistent with the data from isolated vessels, supports the conclusion that receptor-mediated contraction is not enhanced after prenatal exposure to Dex. Chronic Ang II infusion, in contrast, produced an enhanced blood pressure elevation in the Dex group, indicating a greater sensitivity to Ang II. This could not be attributed to vascular remodelling, as the medial area was unchanged in resistance arteries. It is possible that the increased sensitivity to chronic Ang II is due to glucocorticoid-mediated sensitisation of contractile signal transduction (Hai *et al.* 2002) and/or free radical formation and inhibition of endothelium-derived nitric oxide (Rajagopalan *et al.* 1996), mediated in part by AT₁-receptors (McEwan *et al.* 1998). Alternatively, since there is evidence of activation of the RAAS programmed by glucocorticoids, there may be increased sodium retention during chronic Ang II infusion. These mechanisms will require further studies, but the combination of lack of enhanced acute pressor response to Ang II and lack of vascular remodelling during chronic Ang II infusion does not suggest that altered structure or function of

resistance vessels underlies glucocorticoid-programmed hypertension.

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