Effect of Angiotensin II Synthesis Blockade on the Hypertensive Response to Chronic Reductions in Uterine Perfusion Pressure in Pregnant Rats

Barbara T. Alexander, Kathy Cockrell, Farrah D. Cline, Maria T. Llinas, Mona Sedeek, Joey P. Granger

Abstract—The purpose of this study was to examine the role of the renin-angiotensin system in mediating the hypertension in response to chronic reductions in uterine perfusion pressure (RUPP) in conscious chronically instrumented pregnant rats. Mean arterial pressure was significantly higher in pregnant rats with chronic RUPP (125±3.0 mm Hg, P<0.01, n=12) than in pregnant rats (100±2.3 mm Hg, n=17). Plasma renin activity in pregnant rats with chronic RUPP was 17.1±2.5 nmol angiotensin I · L⁻¹ · h⁻¹ compared with 21.9±3.5 nmol angiotensin I · L⁻¹ · h⁻¹ in pregnant rats. Chronic oral administration of a converting-enzyme inhibitor (enalapril, 250 mg/L for 6 days) decreased mean arterial pressure to a similar extent in pregnant rats with chronic RUPP (109±4.2 mm Hg, P<0.01, n=9) and in normal pregnant (81±1.8 mm Hg, P<0.01, n=9) rats. Blockade of the renin-angiotensin system, however, had no significant effect on the blood pressure response to chronic RUPP as differences were similar in control (Δ25 mm Hg) and converting-enzyme inhibitor–treated (Δ27 mm Hg) groups. These findings suggest that the renin-angiotensin system does not play a major role in mediating the hypertension produced by chronic RUPP in pregnant rats. (Hypertension. 2001;38[part 2]:742-745.)

Key Words: hypertension, pregnancy ■ preeclampsia ■ renin-angiotensin system ■ enalapril ■ blood pressure

A leading theory concerning the pathophysiology of preeclampsia suggests that a pathway that starts with inadequate trophoblast invasion of maternal spiral arteries leads to decreased placental perfusion. The ensuing placental ischemia results in placental release of factors, subsequent maternal endothelial dysfunction, and systemic vasoconstriction.¹,² A variety of animal models have shown that a reduction in uterine perfusion pressure (RUPP) leads to systemic hypertension in pregnant animals³–⁶; however, the mechanisms involved in mediating this hypertensive response to RUPP are unknown.

The renin-angiotensin system (RAS) plays an important role in the long-term regulation of renal function and arterial pressure during a variety of both physiological and pathophysiologica...
(MAP) and in reductions in kidney function. As significant alterations in vascular sensitivity to Ang II occur in women with preeclampsia, the goal of the present study was to determine the role of the RAS in mediating the hypertension produced in response to chronic RUPP. Specific aims were to determine if blockade of the RAS by use of the ACE inhibitor enalapril would attenuate the hypertension produced by chronic RUPP in pregnant rats.

**Methods**

All studies were performed in timed-pregnant Sprague Dawley rats purchased from Harlan Sprague Dawley Inc (Indianapolis, Ind). Animals were housed one per cage in a temperature-controlled room (23°C) with a 12-hour/12-hour light/dark cycle. All experimental procedures executed in this study were in accordance with Guide for the Care and Use of Laboratory Animals (NIH publication No. 93-23, revised 1985), and the Animal Care and Use Committee at the University of Mississippi Medical Center approved all protocols.

**Experimental Design**

Animals were divided into 4 groups: pregnant (n = 17), pregnant plus enalapril (n = 9), pregnant with chronic RUPP (n = 12), and RUPP plus enalapril (n = 9). The ACE inhibitor enalapril, (250 mg/L, Sigma-Aldrich Co) was administered in the drinking water starting at day 14 of gestation. Enalapril was used as a tool in this study by employing a dose that has been used extensively by our laboratory and others to result in an effective blockade of the Ang II response as determined by a decrease in arterial pressure. Rats that would be in the RUPP group were clipped at day 14; normal pregnant rats were subjected to a sham operation. Rats were instrumented with arterial catheters on day 18. Arterial pressure was measured, and animal and pups weights were recorded on day 20. Water intake did not differ between pregnant and RUPP, treated or untreated, and averaged 45 to 50 mL per animal per day.

**RUPP in Pregnant Rats**

Chronic reductions in uteroplacental perfusion in rats were achieved by a method previously reported by our laboratory. Briefly, all rats undergoing surgical procedures were anesthetized with 2% isoflurane (W.A. Butler Co) delivered by an anesthesia apparatus (Vaporizer for Forane Anesthetic, Ohio Medical Products). Following a midline incision, the lower abdominal aorta was isolated, and a silver clip (0.230-mm ID) was placed around the aorta above the iliac bifurcation. Because compensation of blood flow to the placenta occurs through an adaptive increase in ovarian blood flow, we also clipped branches of both the right and left ovarian arteries that supply the uterus using a silver clip (0.100-mm ID).

**Measurement of Arterial Pressure in Conscious Rats**

At day 18 of gestation under isoflurane anesthesia as described above, a catheter of heat-stretched PE-50 tubing was inserted into the carotid artery. On day 20 of gestation, rats were placed in individual restraining cages. After achieving stabilization, arterial pressure was recorded continuously for 2 hours with a pressure transducer (Cobe III Transducer), followed by collection of an arterial blood sample.

**Measurement of PRA**

PRA was measured by radioimmunoassay. A pool having an average concentration of 7.13 nmole Ang I · L⁻¹ · h⁻¹ exhibited a within-assay variation of 3.0% (n = 5). Two controls having average PRAs of 1.5 and 2.3 nmole Ang I · L⁻¹ · h⁻¹ exhibited respective between-assay variations of 7.0% (n = 16) and 11.4% (n = 16).

**Statistical Analysis**

All data are expressed as mean ± SEM. Comparisons of control pregnant rats with RUPP rats, both treated and untreated, were analyzed using factorial ANOVA followed by Scheffe’s test. A value of P < 0.05 was considered statistically significant.

**Results**

Chronic RUPP in pregnant rats resulted in significant increases in MAP relative to that of pregnant rats. MAP averaged 100 ± 2.3 mm Hg in pregnant rats. Arterial pressure in pregnant rats with chronic RUPP averaged 125 ± 3.0 mm Hg, 25 mm Hg above pregnant rats (P < 0.01 versus pregnant, Figure 1). Chronic oral administration of the ACE inhibitor enalapril (250 mL/L for 6 days) decreased arterial pressure to a similar extent in both RUPP (109 ± 4.2 mm Hg, P < 0.01 versus RUPP, Δ 17 mm Hg) and pregnant rats (81 ± 1.8 mm Hg, P < 0.01 versus pregnant, Δ 19 mm Hg) (Figure 1), representing a 19% and 13% decrease, respectively. Thus, blockade of the RAS had no significant effect on the blood pressure response to chronic RUPP, as differences were similar in control (Δ 25 mm Hg) and ACE inhibitor–treated (Δ 27 mm Hg) groups. No difference in PRA was observed for pregnant rats with chronic RUPP compared with pregnant rats (17.1 ± 2.5 nmole Ang I · L⁻¹ · h⁻¹ versus 21.9 ± 3.5 nmole Ang I · L⁻¹ · h⁻¹, respectively).

Pup weight, although not significant, tended to decrease in the RUPP rats relative to pregnant rats (3.6 ± 0.2 g versus 3.3 ± 0.2 g, respectively). Additionally, chronic oral treatment with the ACE inhibitor tended to decrease pup weight (3.1 ± 0.2 grams) in pregnant-treated rats but resulted in a significant decrease in the RUPP-treated animals (2.2 ± 0.3 g; P < 0.01 versus RUPP, P < 0.01 versus pregnant). In addition, at day 20 of gestation, chronic RUPP in pregnant rats was associated with a reduced litter size relative to that of pregnant rats (8.9 ± 1.2 pups versus 12.2 ± 0.7 pups, respectively; P < 0.05). Chronic oral treatment with ACE inhibitor attenuated the decreased litter size observed in RUPP rats (10.49 ± 1.2 pups) without altering litter size in pregnant rats (12.3 ± 0.3 pups). Chronic RUPP resulted in decreased body weight at day 20 of gestation in RUPP rats compared with pregnant rats (297.7 ± 6.6 g and 336.0 ± 6.4 g, respectively; P < 0.01). Chronic oral treatment with the ACE inhibitor resulted in a further decrease in body weight in both RUPP-untreated (272 ± 6.9 grams, P < 0.05 versus RUPP, P < 0.01 vs RUPP, Figure 1).
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The RAS is activated in human pregnancy and in the pregnant rat. However, activity of the RAS appears to be unclear but with no change in PRA. Chronic administration of ACE inhibitors during pregnancy results in abnormal fetal development and growth. In a study by Valdes et al., treatment with enalapril during the second half of gestation resulted in marked reductions in pwp weight; however, IUGR was also evident in animals receiving enalapril treatment in the first half of gestation. Within this study, ACE inhibitor treatment during the third week of gestation resulted in IUGR within both pregnant-treated and RUPP-treated animals with a marked decrease in pwp weight observed in the RUPP-treated animals. Thus, use of ACE inhibitors, such as enalapril, as a mode of antihypertensive treatment during pregnancy or PIH is contraindicated.

In summary, we found that chronic RUPP in the pregnant rat was associated with significant increases in arterial pressure but with no change in PRA. Chronic administration of the ACE inhibitor enalapril markedly attenuated the increase in MAP observed in the pregnant rats with chronic RUPP; however, ACE inhibition also significantly reduced blood pressure in pregnant rats to a similar extent as observed in the RUPP rats. Therefore, as arterial pressure differences were similar in control and ACE inhibitor–treated animals, these results suggest that RAS does not play a major role in mediating the hypertension produced by chronic RUPP.

**Acknowledgments**

This work was supported by National Institutes of Health (NIH) grants HL38499 and HL51971. The NIH National Research Service Award HL10137-01 supported Dr Alexander.

**References**

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Hypertension, 2001;38:742-745
doi: 10.1161/01.HYP.38.3.742

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2001 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

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