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Heart Rate Reflex Responses during Gestation in Normotensive and Spontaneously Hypertensive Rats following Angiotensin II and Vasopressin (42616)

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Abstract. Gestation in the human and in rats is accompanied by a decrease in blood pressure and a reduction of the pressor response to vasoconstrictor agents. In humans, the decreased vascular reactivity to angiotensin II (AII) may occur simultaneously with a state of increased baroreceptor sensitivity. We have consequently evaluated the heart rate response to elevation of blood pressure following administration of either AII or arginine^8-vasopressin (AVP) in conscious unrestrained, nonpregnant, or term-pregnant normotensive rats (Sprague–Dawley, SDR; Wistar–Kyoto, WKR) and in spontaneously hypertensive rats (SHR). The decrease in heart rate in response to increase in blood pressure by AII in nonpregnant animals was similar in SDR and SHR, but much greater in WKR. The heart rate response to increase in blood pressure by AVP was similar in all three strains of cycling rats. Gestation (20th day) did not change the heart rate response to increase in blood pressure by AII in normotensive animals, but increased slightly the reflex responses in SHR, as shown by a significant increase of the slope of the relationship of the decrement in heart rate versus the increment of blood pressure. The heart rate response to increase in blood pressure by AVP was greater during gestation in normotensive SDR and WKR, but not in SHR. These results show that the heart rate responses to an increase in blood pressure by vasoconstrictor peptides is dependent on the strain of animals used and suggest that the baroreceptor reflexes play a minor role in the blunted effect of vasconstrictor agens at the end of gestation in normotensive and spontaneously hypertensive rats.

Hypertension is associated with an increase of the total peripheral resistance, while cardiac output and heart rate are usually within the normal range. The persistence of a normal heart rate in hypertension has been explained by a resetting of baroreceptor function to a higher level of pressure (1–3) proportional to the degree of the disease (4). The same phenomenon appears to be true at the end of gestation in the rat, since despite a substantial decrease in blood pressure in normotensive and spontaneously hypertensive term-pregnant rats, the heart rate is not changed (5, 6). Gestation in humans (7) and rats (5, 8) is associated with a decrease in blood pressure and of the vascular sensitivity to vasoconstrictor agents such as angiotensin II (AII), arginine^8-vasopressin (AVP), and norepinephrine (NE) (6, 9, 10). This decrease of vascular reactivity during pregnancy was associated with an increase of the baroreceptor reflex activity in human (11), while Humphreys and Joels (12) reported that the baroreceptor function does not influence the decreased vascular resistance of the skinned hind-limb vascular bed in pregnant rabbit. There is little information on the role of the baroreceptor reflex in the attenuation of vascular reactivity to pressor agents in pregnant rats.

AVP and AII may have different effects on the baroreceptor. AVP facilitates baroreflexes by local and central mechanisms, while AII attenuates baroreflexes by a central mechanism (13). In term-pregnant rats, either normotensive or hypertensive, the vascular responses to AII and AVP, are similarly decreased (twofold increase of the ED_{50}) by comparison to cyclic rats (6). In light of these results (6, 13), we decided to look at the heart rate responses to intravenous injection of AII and AVP in normotensive and spontaneously hypertensive cyclic and term-pregnant rats, in order to investigate the role of baroreflexes in the blunted effects of *in vivo* vasoconstrictor responsiveness at the end of gestation in the rat. Our results indicate that the

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changes in heart rate response to increases in blood pressure by AII and AVP are not involved in the decrease of the *in vivo* sensitivity to vasoconstrictor agents during gestation. The blood pressure responses to both agents in these rats were reported previously (6).

Materials and Methods. Female Wistar–Kyoto (WKR), Sprague–Dawley (SDR), and spontaneously hypertensive rats (SHR) of 10 weeks of age were obtained from Taconic Farms (Germantown, NY) and were bred with males of the same age. The morning in which vaginal smears were found to contain spermatozoa was labeled Day 1 of gestation. The experiments were performed on the 20th day of gestation, i.e., when the animals reached 13–14 weeks old, in parallel with unmated females of the same age as controls.

Blood pressure and heart rate were measured in conscious rats as previously described (6, 14). In brief, under light ether anesthesia, the left carotid artery and jugular vein were cannulated respectively with PE-50 and PE-90 polyethylene tubing (Clay-Adams). The catheters were passed under the skin and brought out at the scruff of the neck and secured with silk threads. The carotid catheter was connected via a Gould-Statham pressure transducer (P23ID) to a Grass Polygraph No. 7. A digital heart rate monitor was connected to the J6 output of the driver amplifier of the polygraph.

After surgery, rats were put in metal wire cages for the rest of the experiment, where they were allowed to recover from anesthesia (approximately 60–90 min). For the first 20 to 60 min of this period, blood pressure and heart rate increased progressively and then stabilized. Increasing doses of AVP and AII were subsequently injected intravenously in the same rat over a period of 5 to 6 hr. The dosage of both AVP and AII used ranged from 1 to 4000 ng/kg for each peptide and was given in a 50-μl bolus injection followed by 50 μl saline. Sufficient time was allowed between each dose to ensure that blood pressure and heart rate returned to baseline levels.

The maximum decrement in heart rate and increment in blood pressure were measured after the administration of each dose of the peptides. The regression line of the mean decrease in heart rate and the mean increase in blood pressure for each dose of the peptide was evaluated by factorial analysis of covariance (6, 14, 15) to determine the intercept and the slope of the regression line, which respectively estimate the threshold and sensitivity of the baroreflex.

TABLE I. CONTROL VALUES OF MEAN BLOOD PRESSURE (MAP) AND HEART RATE (HR) IN CYCLIC (C) AND PREGNANT (P) SPRAGUE–DAWLEY (SDR), WISTAR–KYOTO (WKR), AND SPONTANEOUSLY HYPERTENSIVE (SHR) RATS

Group		N	MAP (mm Hg)	HR (beats/min)
SDR	C	9	95 ± 3	355 ± 14
	P	9	94 ± 4	408 ± 8*
WKR	C	9	90 ± 5	370 ± 15
	P	9	81 ± 4	372 ± 17
SHR	C	9	135 ± 10	384 ± 15
	P	9	107 ± 4*	391 ± 10

* $P < 0.05$ vs cyclic animal of the same strain by Student's t test.

Pharmacological agents used in these experiments were arginine[8]-vasopressin (AVP) and aspartic[1], isoleucine[5]-angiotension II (AII), both obtained from Peninsula (Belmont, CA).

Results. Control values of mean blood pressure and heart rate for the six different groups of rats are shown in Table I. The only statistically significant differences between pregnant and cyclic animals of a given strain were found in SHR for blood pressure and SDR for heart rate. The blood pressure of WKR was also decreased in the pregnant ones, but the values were at the limit of significance ($P < 0.10$).

Table II summarizes the results of the regression lines for the decrement in heart rate versus the increment in blood pressure produced by AII and AVP in the six groups of rats, while the data are depicted in Figs. 1 and 2. When AII was used as the vasopressor agent, the relationship between the decrease in heart rate and increase in blood pressure was markedly different in the three strains of rats, both cyclic and pregnant (Fig. 1). The slope of the regression line was significantly higher ($P < 0.05$) in WKR than in SDR and SHR (Fig. 1, Table II), both in the cyclic and

TABLE II. SLOPE (m), ORDINATE INTERCEPT (y), AND CORELATION COEFFICIENT (r) OF MEAN REGRESSION LINES OF THE HEART RATE RESPONSE FOLLOWING BLOOD PRESSURE ELEVATION BY AII AND AVP IN CYCLIC AND PREGNANT SDR, WKR, AND SHR

Group	N	Angiotensin II			Vasopressin		
		(m) beats/min / mm Hg	(y) beats / min	(r)	(m) beats/min / mm Hg	(y) beats / min	(r)
SDR C	9	0.90	10	0.98	1.65	0.50	0.95
P	9	0.66	19	0.98	2.51*	−7.00	0.99
WKR C	9	2.27	11	0.99	1.98	−0.50	0.97
P	9	2.39	4	0.96	2.87*	−1.00	1.00
SHR C	9	0.42	26	0.73	2.94	−52	0.95
P	9	1.53*	−17	0.97	2.56	−57	0.97

* $P < 0.05$ (vs C of same strain, by covariance analysis).

pregnant rats. Gestation did not change the characteristics of the regression of heart rate versus blood pressure in normotensive animals (SDR and WKR). In fact, neither slope nor intercept was statistically altered, but the slope was markedly increased in pregnant SHR (Table II). Despite the marked increment in control heart rate in pregnant SDR (Table I), the heart rate–pressure relationship was not statistically modified in these rats by comparison to cyclic ones. These data suggest that when AII was used as a pressor agent, the reflex response of the heart rate of WKR was more sensitive than that of the two other strains. Gestation induced a change of this relationship only in SHR, which had a more sensitive baroreflex-mediated bradycardia.

Figure 2 and Table II describe the results obtained when AVP was used as the pressor agent. There was no statistical difference in the regression lines of the decrease in heart rate in response to the increase in blood pressure obtained from the three groups of cyclic rats. In pregnant rats, the slopes of the

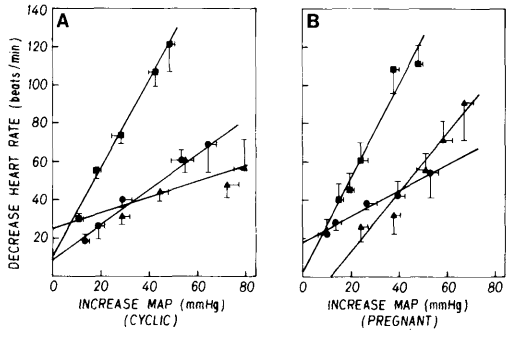

FIG. 1. Decrease in heart rate in response to increase in blood pressure induced by AII in cyclic and term-pregnant rats. Circles, SDR; squares, WKR; triangles, SHR. Ordinate: mean decrease in heart rate for each mean increase in blood pressure for each dose of AII. Bars indicate the standard error of the mean. The slopes of the regression for WKR were significantly steeper than for SDR and SHR in both cyclic and pregnant rats ($P < 0.05$) by covariance analysis.

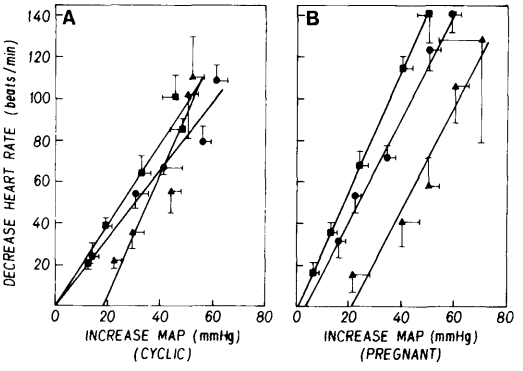

FIG. 2. Decrease in heart rate in response to increase in blood pressure induced by AVP in cyclic and term-pregnant rats. Circles, SDR; squares, WKR; triangles, SHR. Ordinate: mean decrease in heart rate for each mean increase in blood pressure for each dose of AVP. Bars indicate the standard error of the mean. The regressions for pregnant SDR and WKR (B) were significantly ($P < 0.05$) displaced to the left vs pregnant SHR and the three cyclic strains of rats (A) by covariance analysis.

regression lines were almost identical but the intercepts were markedly different, especially for SHR. The threshold dose of AVP for a bradycardic effect in SHR was higher than in the two normotensive strains (Fig. 2, Table II). There was a markedly significant increase in the slope of the regression line for the two pregnant normotensive groups (increase in sensitivity of the baroreflex) when compared to their nonpregnant controls. In pregnant SHR, the slope and the intercept of the regression lines were almost identical to the one of cyclic SHR. Thus, there was no difference in the sensitivity of the baroreceptor between the three strains of rats when AVP was used as pressor agent, but that there was a significant increase in the baroreceptor sensitivity in pregnant normotensive rats by comparison to cyclic ones.

Discussion. In this study, we have attempted to look at the possible role played by baroreceptor function in the blunted *in vivo* pressor effect of vasoconstrictor agents at the end of gestation in the rat. In fact, several reports have shown that the blood pressure responses to AII, AVP, and norepinephrine are decreased at the end of gestation and have speculated on the possible mechanisms of this effect (6, 9, 16). The buffering capacity of the baroreceptor reflex could play a role if its sensitivity were to increase in pregnant rats. We therefore looked at the decrease in heart rate produced by increases in blood pressure induced by two vasoconstrictor agents, AII and AVP, to which normotensive (6, 9) and spontaneously hypertensive rats (6) are less sensitive at the end of gestation.

Our data show that the bradycardia produced by increases in blood pressure is different if the pressure is increased by AII or by AVP, which is consistent with previous observations (13). The degree of bradycardia produced relative to the increase in blood pressure was also different from one strain to another, either normotensive or hypertensive, as shown by the greater slope of the regression line in WKR by comparison with the two other strains when AII was used to increase pressure. Furthermore, at the end of gestation, baroreceptor function was not modified consistently by a given agent in the three strains of rats. Different changes were obtained for each of the two vasoconstrictors. In fact, Table II shows that the effect of gestation of the heart rate reflex response to an increment in blood pressure was increased in normotensive rats (SDR and WKR) only when AVP was used as the pressor agent. The baroreceptor reflex was also sensitized in SHR when AII was used to induce the increase in pressure. The absence of changes in heart rate reflex response in pregnant normotensive rats (SDR and WKR) with AII and SHR with AVP suggests that an increase in sensitivity of the baroreflex does not play a role in the blunted effects of vasoconstrictor agents *in vivo* in pregnancy. Recent results obtained from this laboratory (A. Parent, J. St-Louis, E. L. Schiffrin, manuscript in preparation) showed that the sensitivity to two vasodilators (bradykinin and sodium nitroprussiate) is increased in term-pregnant normotensive SDR, without modification of the tachycardic response following blood pressure decreases induced by these agents.

Similar conclusions were reached by Humphreys and Joels (12), who studied the reflex response of the skinned hind-limb vascular bed of the pregnant rabbit to isolated carotid sinus pressure stimulation. These results suggested that the baroreceptor function does not influence the modified resistance of this vascular bed in the pregnant rabbit. These authors indicated that the reduction of the resistance to the blood flow in pregnant rabbits, during sympathetic stimulation, is due mainly to either a different size of the stimulated vascular beds or a different compliance of their vessel walls, rather than to any intrinsic alteration in their response to sympathetic stimulation. *In vitro* studies, that avoid the confounding influence of whole body compensatory or reflex mechanisms, also revealed that the vascular reactivity to vasoconstrictor agents (AII, NE, AVP, and barium chloride) in many vascular tissues (mesenteric and lung vascular bed; aortic, femoral, and portal vessels) are blunted during pregnancy in the rat (10, 17–19). The blunting of vasoconstrictor responses to AII, AVP, and NE in pregnant normotensive and spontaneously hypertensive rats (10) and the present data suggest that the decreased responsiveness to vasoconstrictor agents in pregnant rats *in vivo* is a consequence of a

phenomenon affecting directly the vascular smooth muscle, rather than an increase of the baroreceptor reflex. This is in agreement with previous observations obtained in the pregnant rabbit showing that the role of baroreceptor function in these changes is not important (12). In contrast, Seligman (11) reported that the control of arterial pressure by the baroreceptor reflex was set at a high level of sensitivity in normotensive pregnancy in the human, while it was diminished during pregnancy in chronic hypertensive women and was virtually lost in preeclampsia. In the present study, the heart rate–pressure relationship was more sensitive in pregnant SHR by comparison to normotensive SDR and WKR only when AII was used as the pressor agent, while with AVP the reflex mechanism was identical in pregnant and cyclic animals.

In conclusion, our data shows that the heart rate reflex response to increases in blood pressure by vasoconstrictor agents is different in normotensive and spontaneously hypertensive rats and that pregnancy does not modify this reflex mechanism in a similar manner in the three strains of rats and for the two vasoconstrictors used. Our results indicate that an increase in baroreceptor sensitivity does not appear to be involved in the blunted effects of vasoconstrictor agents and in the decrease of blood pressure at the end of gestation in the rat.

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