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Hemodynamic responses to acute aortic coarctation in conscious sinoaortic denervated rats

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Abstract

The hemodynamic responses to acute (45 min) partial aortic constriction were studied in conscious intact (N = 7) or sinoaortic denervated (SAD) adult male Wistar rats (280-350 g, N = 7) implanted with carotid and femoral arterial catheters, a pneumatic cuff around the abdominal aorta and a pulsed Doppler flow probe to measure changes in aortic resistance. In addition, the hypertensive response and the reflex bradycardia elicited by total (N = 8) *vs* partial (N = 7) aortic constriction (monitored by maintenance of the pressure distal to the cuff at 50 mmHg) were compared in two other groups of intact rats. Intact rats presented a smaller hypertensive response (26 to 40% above basal level) to partial aortic constriction than SAD rats (38 to 58%). The calculated change in aortic resistance imposed by constriction of the aorta increased progressively only in intact rats, but was significantly smaller (193 to 306%) than that observed (501 to 591%) in SAD rats. Intact rats showed a significant bradycardia (23 to 26% change in basal heart rate) throughout coarctation, whereas the SAD rats did not (1 to 3%). Partial or total occlusion of the aorta induced similar hypertensive responses (37-38% *vs* 24-30% for total constriction) as well as reflex bradycardia (-15 to -17% *vs* -22 to -33%) despite a greater gradient in pressure (97-98 *vs* 129-140 mmHg) caused by total constriction. The present data indicate that the integrity of the baroreflex in intact rats can cause the hypertensive response to level off at a lower value than in SAD rats despite a progressive increase in aortic resistance. In addition, they also indicate that the degree of partial aortic constriction by maintenance of the pressure distal to the cuff at 50 mmHg already elicits a maximal stimulation of the arterial baroreflex.

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- Aortic resistance
- Reflex bradycardia
- Sinoaortic denervation

Introduction

The model of acute aortic coarctation provides remarkable hemodynamic changes such as an increase in carotid pressure, which has been attributed to the sudden increase in the resistance to aortic flow, associated with neurohumoral mechanisms triggered by territories below the constriction, especially the kidneys (1-4). In contrast, it is well known that sinoaortic baroreceptors counteract any prompt rise in pressure, as is the case for acute aortic coarctation hypertension (5). Previous observations from our laboratory (6) have demonstrated that conscious rats with intact arterial baroreceptors still exhibit

a gradual increase in aortic resistance during acute (45 min) aortic constriction as a consequence of the overall increase in the impedance to blood flow caused by the release of neurohumoral factors (e.g., angiotensin II and vasopressin) triggered by the kidneys (2). In the present study we investigated the hemodynamic responses (changes in carotid pressure, heart rate and aortic resistance to blood flow) of conscious intact and sinoaortic denervated (SAD) rats during the acute (45 min) hypertensive response to partial aortic constriction (pressure distal to the cuff maintained at 50 mmHg). In addition, we compared both the extent of the hypertensive response and the reflex bradycardia obtained with partial *vs* total occlusion of the aorta in two other groups of intact rats.

Material and Methods

The experiments were performed on conscious freely moving male Wistar rats (280-350 g) equipped with intra-arterial catheters (carotid and femoral), a pneumatic cuff around the abdominal aorta above the renal arteries, and a miniaturized Doppler flow probe. Two other groups used to compare the hemodynamic responses to partial (N = 7) or total (N = 8) aortic constriction were equipped with vascular catheters and the pneumatic cuff only. Carotid and femoral arterial pressures were recorded continuously with a Statham pressure transducer (Model P23-Db, Hato Rey, PR) attached to a Hewlett-Packard recorder (Model 7848, Palo Alto, CA). The wire leads from the flow probe were connected to an ultrasonic pulsed Doppler flowmeter (University of Iowa Bioengineering Department, Iowa City, IA) which allows measurement of changes in blood velocity recorded as Doppler shift in kHz. Aortic resistance was calculated as the ratio of mean carotid pressure (MCP) and aortic blood flow Doppler shift and then normalized in terms of percentage (7).

Two days before the experiment, under sodium pentobarbital (40 mg/kg, *ip*) anesthesia, both pneumatic cuff and flow probe were placed around the aorta, immediately below the diaphragm, by means of ample laparotomy. This procedure was the same as that used in previous studies (6). Briefly, a 4-6 mm length of abdominal aorta was carefully isolated below the diaphragm. A pneumatic cuff, prepared according to a technique developed in our laboratory (8), was placed around the abdominal aorta above the renal arteries, and the miniaturized flow probe secured with 6-0 cotton sutures was placed above the cuff immediately below the diaphragm. The tubing connected to the pneumatic cuff and the wires of the Doppler flow probe were tunneled and exteriorized through the animal's back. Twenty-four hours before the experiment, under ether anesthesia, the animals were submitted to sinoaortic deafferentation (SAD rats, N = 7), according to Krieger's technique (9), or sham operation (intact rats, N = 8) and the catheters were introduced into the femoral and carotid arteries. In animals submitted to partial aortic coarctation the degree of constriction was monitored by reduction of the pressure distal to the occlusion to 50 mmHg. Total occlusion of the aorta was monitored by maintenance of a maximal gradient of pressure between carotid and femoral arterial pressure. The animals did not exhibit any sign of distress with this maneuver. On the day of the experiment the animals had basal MCP and heart rate (HR) measured over a period of 10-15 min followed by acute (45 min) partial or total aortic coarctation. HR was obtained by counting pulses at higher recorder speed.

Statistical analysis of the hypertensive response and reflex bradycardia to aortic constriction was performed by multivariate analysis of variance for repeated measures (MANOVA). Baseline values were also compared by the Student *t*-test. Since differences in baseline values were found, a covariance analysis (MANCOVA) was applied to the

curves. When the time course of the curves was different, individual comparisons were performed during each period by the Student *t*-test. Differences were considered significant if P<0.05.

All animals received humane care in compliance with the Principles of Laboratory Animal Care formulated by the National Research Council (NCR) in the Guide for the Care and Use of Laboratory Animals prepared by the National Academy of Sciences and published by the National Institutes of Health (Publication No. (NIH) 85-23, revised 1985).

Results

Total *vs* partial occlusion of the aorta

MCP and HR values before and during maintained (45 min) partial and total aortic coarctation induced by the pneumatic cuff are shown in Figure 1. The animals submitted to partial occlusion of the aorta presented a basal MCP of 107 ± 2 mmHg. Five min after coarctation, MCP had already reached a plateau (37% above control) and remained close (37 to 38%) to this level throughout the experiment. The animals submitted to total occlusion of the aorta presented a basal MCP of 117 ± 2 mmHg, and five min after the constriction the MCP had also reached a plateau (30% above control) and remained close (27 to 28%) to this level throughout the experiment. During partial constriction of the aorta, the pressure distal to the pneumatic cuff was maintained precisely at 50 mmHg, whereas during total coarctation the distal pressure (data not shown) oscillated between 12 and 17 mmHg. The basal HR of the rats submitted to partial occlusion was 369 ± 6 bpm and five min after coarctation the reflex bradycardia had already leveled off (305 ± 2 to 314 ± 3 bpm). The animals submitted to total occlusion presented a basal HR of 421 ± 7 bpm which declined progressively during total constriction; this reflex bradycardia reached 329 ± 13 and 280 ± 10 bpm 5 and 45 min after the constriction, respectively.

Intact *vs* SAD rats

MCP and HR values before and during the maintained (45 min) aortic coarctation in intact and SAD rats are shown in Figure 2. The intact group presented a basal MCP of 108 ± 4 mmHg. Fifteen minutes after coarctation, MCP reached a plateau (36% above control) and remained close (36 to 40%) to

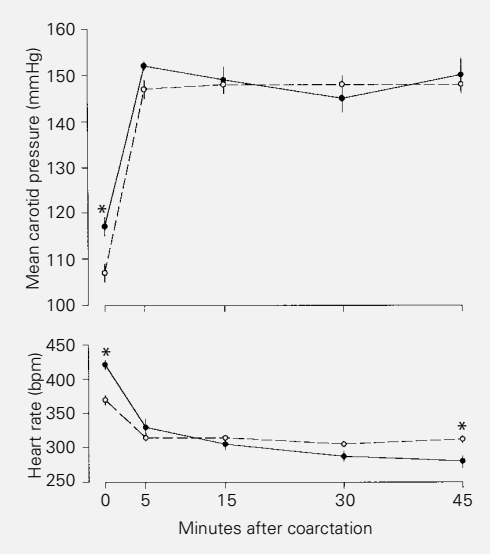

Figure 1 - Mean carotid pressure (upper panel) and heart rate (lower panel) responses to acute aortic coarctation in conscious unrestrained intact rats submitted to total (continuous line) or partial (broken line) aortic constriction. Data are reported as means ± SEM. *P<0.05 compared to partial constriction (Student *t*-test).

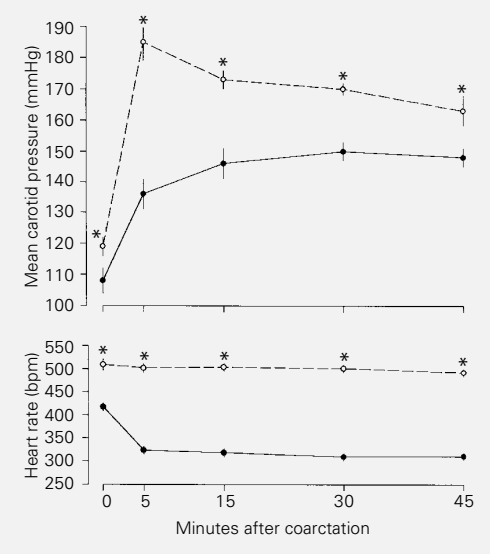

Figure 2 - Mean carotid pressure (upper panel) and heart rate (lower panel) responses to acute aortic coarctation in conscious unrestrained intact (continuous line) and sinoaortic denervated (SAD) (broken line) rats. Data are reported as means ± SEM. *P<0.05 compared to the corresponding point in the intact group (Student *t*-test).

this level throughout the experiment. The SAD group presented a basal MCP of 119 ± 2 mmHg. Five min after constriction, the SAD group presented an increase in MCP almost twice (58%) that observed in intact rats, and remained at a higher level (38-46%) throughout the experiment. The basal HR of the intact group was 417 ± 4 bpm. Five minutes after coarctation the reflex bradycardia leveled off (323 ± 5 to 309 ± 4 bpm) throughout the experiment. In contrast, the SAD group presented a remarkable tachycardia (509 ± 8 bpm) before coarctation and the HR did not change (492 ± 4 to 502 ± 6 bpm) during aortic constriction. Figure 3 shows the calculated changes in aortic resistance (%) of the intact and SAD groups during coarctation. The change in aortic resistance of the intact group increased progressively from 5 min (193 ± 28%) to 30 min (314 ± 41%) after coarctation when it attained a plateau (306 ± 40%). The change in aortic resistance of SAD rats during aortic constriction was significantly higher than that of the intact group. Nevertheless, the change in aortic resistance of the SAD group had already reached a plateau (501 ± 67%) 5 min after constriction, remaining within a relatively narrow range (548 ± 72 to 591 ± 63%) throughout the experiment.

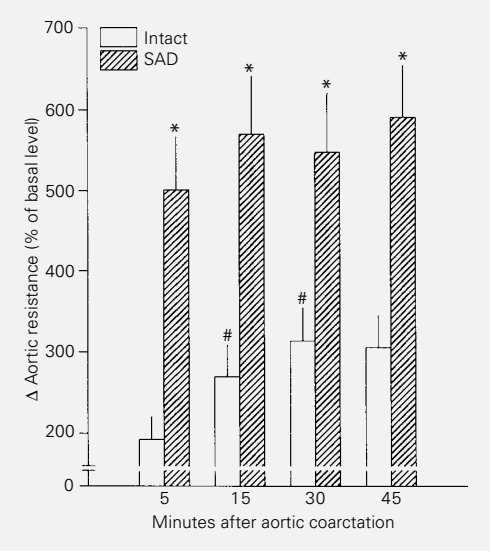

Figure 3 - Bar graph showing change as percentage of basal level (Δ%) in calculated aortic resistance due to aortic coarctation in conscious unrestrained intact and sinoaortic denervated (SAD) rats. Data are reported as means ± SEM. *P<0.05 compared to the corresponding point in the intact group (Student *t*-test); #P<0.05 compared to the anterior value in the same group (Student *t*-test).

Discussion

Partial aortic occlusion with the maintenance of the pressure distal to the cuff precisely at 50 mmHg (8) keeps the renal perfusion pressure well below the lower limit of renal blood flow autoregulation (10) providing a powerful stimulus for renin (1,4) and vasopressin release (2,3,6,11,12). In addition, this maneuver induces a marked mechanical increase in aortic resistance which represents a substantial challenge for circulatory homeostasis (6). In the present study we found that, despite a greater gradient in pressure (129-133 mmHg) through the cuff caused by total aortic occlusion, the hypertensive response did not differ from that of the animals submitted to partial occlusion, but the time course of the reflex bradycardia differed between groups particularly 45 min after constriction.

The finding that the degree of partial occlusion monitored by the maintenance of the mean femoral pressure at 50 mmHg elicited a hypertensive response equivalent to that elicited by total occlusion is not surprising. Previous findings by Gupta and Wiggers (13) demonstrated that 85 to 95% constriction of the aortic lumen is necessary to reduce the distal pressure to 50 mmHg in anesthetized dogs. Therefore, the amount of resistance imposed by the maintenance of the pressure distal to the occlusion at 50 mmHg already elicits a maximal hypertensive response. This finding validates the use in our protocol of partial aortic occlusion to promote maximal stimulation of the arterial baroreflex in conscious intact rats.

The basal MCP of conscious intact and SAD rats differed. Over the last two decades there has been a debate about whether or not sinoaortic denervation causes sustained hypertension (14-16). Currently, there is a general agreement that sinoaortic denervation causes an extreme lability of arterial pressure, not necessarily accompanied by arterial pressure elevation (17), but with the SAD

rats being more reactive to environmental stimuli (18), e.g., noise (19). In the present study we also observed that the SAD rats presented a remarkable tachycardia when compared to their intact counterparts, which might be attributed to sympathetic overactivity (19-22). During acute (45 min) partial aortic coarctation intact rats presented a steady increase in MCP accompanied by a stable reflex bradycardia, indicating a buffering effect of the arterial baroreflex due to parasympathetic activation (23). In contrast, during aortic constriction SAD rats exhibited both a greater hypertensive response and a greater change in aortic resistance to blood flow than the intact animals. The maintenance of the MCP of SAD rats 38 to 58% above the basal level during constriction of the aorta, without a change in HR, indicates complete sinoaortic deafferentation. From a hemodynamic point of view, it is noteworthy that in intact rats both MCP and HR remained stable throughout the period of coarctation, whereas the overall resistance to blood flow increased gradually as indicated by the change in aortic resistance. This finding might be explained by Green's peripheral resistance equation (resistance = pressure/flow) (24) where the fall in cardiac output would compensate for the increase in overall resistance with the maintenance of a steady MCP. Nevertheless, SAD rats exhibited stable MCP and HR, without a gradual change in aortic resistance to blood flow, presumably due to the maintenance of a constant cardiac output and the lack of reflex vasodilatation throughout the period of coarctation.

Studies from our laboratory on conscious rats demonstrated that, in addition to the mechanical effect of aortic constriction, the vasopressor hormones angiotensin II and vasopressin participate in the physiopathogenesis of acute (45 min) aortic coarctation hypertension (1-3,11,12). Moreover, a previous study from our laboratory indicated that the release of these neurohumoral factors increased the overall resistance to blood flow throughout the circulation (6). As demonstrated before (6), in the present study there was a gradual increase in aortic resistance during coarctation of the aorta in intact rats, which may have been due to an increase in overall resistance to blood flow. In contrast, the aortic resistance of SAD rats reached a maximum immediately after constriction of the aorta, a finding that might be interpreted as being due to an already increased sympathetic tone at the beginning of the period of aortic coarctation (20,22). Therefore, the increased sympathetic drive of SAD rats presumably masked the effect of neurohumoral factors on overall resistance to blood flow as seen in intact animals. On the other hand, the smaller and gradual increase in aortic resistance associated with a sharp bradycardia that leveled off 5 min after aortic constriction in the intact rats might suggest the already known rapid resetting of the baroreceptors (25,26) that was demonstrated to be present 30 min after the onset of the hypertensive response (27).

An intriguing aspect of the hemodynamic findings in the intact animals was the stability of reflex bradycardia and hypertensive response while the overall resistance to blood flow increased progressively. These findings suggest an effective role of the baroreflex in counteracting the rise in pressure by slowing the heart but apparently failing to act upon vascular resistance to overcome the effect of vasopressor hormones. However, there is evidence from experiments in anesthetized rabbits (28) and humans with arterial hypertension (29) of a differential control of HR and vascular resistance during activation of the arterial baroreflex. In addition, there is evidence in conscious (30) as well as in anesthetized rats (31) that aortic baroreceptors play a selective role in the regulation of regional blood flow independent of changes in HR. Therefore, the finding in the present study of a stable reflex bradycardia associated with a gradual increase in

overall resistance to blood flow might be explained by a failure of the baroreflex to override the effect of vasopressor hormones and/or by a differential control of HR and vascular resistance during strong activation of the arterial baroreflex.

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