Elevated vertebrobasilar artery resistance in neonatal spontaneously hypertensive rats


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Cates MJ, Steed PW, Abdala AP, Langton PD, Paton JF. Elevated vertebrobasilar artery resistance in neonatal spontaneously hypertensive rats. J Appl Physiol 111: 149–156, 2011. First published April 14, 2011; doi:10.1152/japplphysiol.00220.2011.—There is a strong correlation between increased vertebral artery resistance and arterial blood pressure in humans. The reasons for this increased resistance at high systemic pressure remain unknown, but may include raised sympathetic activity. With the recent finding that prehypertensive spontaneously hypertensive (PHSH) rats, which have raised sympathetic nerve activity, but a blood pressure comparable to normotensive rat strains, we hypothesized that its vertebrobasilar vascular resistance would already be raised and, as a consequence, would exhibit a more responsive Cushing response (e.g., brain ischemia evoked sympathoexcitation and a pressor response). We report that PHSH rats exhibited a remodeling of the basilar artery (i.e., increased wall thickness and lower lumen-to-wall thickness ratio) that occurred before the onset of hypertension. In a novel in vitro vascularly isolated, arterially perfused brain stem preparation of PHSH rats of 4–5 wk of age, brain stem vascular resistance was raised by ~35% relative to age- and sex-matched normotensive rats (P < 0.05). In the in situ arterial perfused working heart-brain stem preparation, occlusion of both vertebral arteries in the PHSH rat resulted in a significantly greater increase in sympathetic activity (57 vs. 20%, PHSH vs. control; P < 0.01) that triggered a greater increase in arterial perfusion pressure (8 vs. 3 mmHg, PHSH vs. control; P < 0.01) compared with normotensive rats. These data indicate raised vertebrobasilar artery resistance before the onset of hypertension in the PHSH rat. With the raised responsiveness of the Cushing response in the PHSH rat, we discuss the possibility of brain stem perfusion as a central nervous system determinant of the set point of vasomotor sympathetic tone in the hypertensive condition.

In 1902, Harvey Cushing observed a proportional rise in blood pressure following brain stem ischemia triggered by raising intracranial pressure in conscious dogs (5). He suggested that this response existed to protect the brain against diminished blood supply in the acute setting of raised intracranial pressure. That this mechanism might represent more than just a last-ditch attempt to maintain brain stem blood flow was suggested by Dickinson and Thomson (11). They performed a large postmortem series on 80 patients and discovered that antemortem blood pressure values correlated with a narrowing of the vertebral arteries (VAs); other vessels studied, including the carotid and renal arteries, did not show this correlation to be so strong. The authors commented that their results could be explained in three ways, which were not mutually exclusive: 1) high blood pressure causes preferential narrowing of the VAs; 2) a common factor causes both high blood pressure and restricted VA capacity; and 3) restricted blood-carrying capacity of the VAs causes high blood pressure. They suggested that the first explanation was unlikely, because a number of patients who had hypertension that was secondary to other causes (including renal artery stenosis) showed little or no narrowing of the VAs. In the absence of an obvious common factor causing both findings, the authors concluded the last explanation was the most likely, although the issue of causality has remained both hypothetical and controversial ever since.

We have studied vertebrobasilar artery resistance and sympathetic activity generation in an animal model of essential hypertension, the spontaneously hypertensive (SH) rat. This rat model demonstrates a number of pathological features in keeping with humans with essential hypertension and is the most studied animal model of the condition (39). Moreover, there is good evidence of sympathetic overactivity in both the adult (24, 30), but also in the prehypertensive SH (PHSH), rat (3, 41), despite an absence of hypertension until around 6 wk of age (this study and Ref. 9). Relevant to the present study is that the vertebral and basilar arteries supplying the brain stem in the PHSH rat show morphological differences before the onset of hypertension compared with normotensive rats (19, 38). The functional significance of these morphological changes in relation to blood flow and resistance remains unclear. Based on this, we hypothesized that the vertebrobasilar artery resistance of the PHSH rat would be raised, and that the PHSH rat would exhibit a more responsive Cushing response relative to a normotensive rat. Thus the aim of the present study was to answer the following: 1) when do morphological differences occur in the PHSH rat; 2) do they translate to increased vascular resistance within the brain stem of the PHSH rats; and 3) is the PHSH rat brain stem more responsive to reductions in brain stem blood flow in terms of sympathetic activity generation relative to an age- and sex-matched normotensive rat strain.

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METHODS

Procedures were carried out according to the United Kingdom Home Office Guidelines on Animals (Scientific Procedures) Act of 1986. The animal research protocols used in this study were also approved by the University of Bristol’s ethics review board. Animals were sourced from either a commercial supplier (Wistar >3 wk old and SH rats; Harlan) or bred within the University of Bristol facility (Wistar rats up to 3 wk old).

Blood Pressure Measurements in Developing Rats

We used radiotelemetry to measure arterial pressure. The advantage of this technique was that we obtained blood pressure readings from unrestrained, stress-free, and conscious animals. For technical reasons, we were limited to measuring arterial pressure in SH rats from 5 wk old with the first reliable postoperative measurement at 6 wk old. Radiotelemetry transmitters were fitted, as previously described by our laboratory (43), to Wistar (n = 6) and SH rats (n = 5). Animals were housed in a temperature, humidity, and light-controlled environment and monitored 24 h per day. Radiofrequency transmitters were calibrated daily before use.

Comparative Morphological Analysis of Basilar Arteries in Wistar and SH Rats

Twenty-four SH and Wistar rats of postnatal age (P) 2, 7, 26, 44, and 92 days were anesthetized with halothane until they failed to respond to noxious pinching of the hind paw and were killed by decapitation. Following a craniotomy, the brain with basilar artery attached was removed carefully. Brains were postfixed in parafomaldehyde (10%) for 4 days when they were transferred to 30% sucrose solution for a further 3–5 days at 4°C. Transcranial perfusion of 2-day-old rats proved difficult technically, so all brains were postfixed to allow comparative analysis. The brain stem was isolated, mounted, and frozen before cutting using a cryostat (coronal sections 10 μm thick) around a level corresponding to the rostral end of area postrema. Sections were mounted onto slides and hematoxylin and eosin stained. Briefly, sections were washed in tap water and submersed in standard Ehrlich’s hematoxylin for 5–10 min and differentiated in acid alcohol (1% hydrochloric acid in 70% alcohol) before washing with water. Sections were exposed to Scott’s bluing solution (3.5 g sodium bicarbonate and 20 g magnesium sulfate per liter of tap water) and rewashed and imaged to ensure that only nuclei had stained. Sections were then stained with eosin (1% aqueous) for 10 s, dehydrated with alcohol, and cleared with Histoclear. Sections were coverslipped with DPX mountant before viewing. To ensure that basilar arteries were measured at corresponding rostrocaudal levels between animals, measurements were made at comparable levels of the inferior olive, as defined by its characteristic shape. Measurements included the external and internal circumferences. Based on these measurements, wall thickness and internal and external diameters and corresponding ratios were computed. Measurements were made using a light microscope at ×40 magnification and Axiovision software (release 4.8) for curve measurement. For each animal, measurements were typically averaged from three sequential sections and normalized against body weight. Data were analyzed using Student’s t-test, and P < 0.05 was taken as significant.

Vascular Resistance Study in Vitro

A novel isolated in vitro arterially perfused brain stem preparation was used in which vascular resistance within the basilar artery and downstream arterioles was measured in male 4–5 wk SH (n = 7; weight 91 ± 5 g) and Wistar (n = 9; 97 ± 3 g) rats (see Fig. 2A). Rats were heparinized (1,000 units ip) 1 h before the start of the procedure to reduce the risk of intra-arterial clot formation affecting pressure measurements. Under deep halothane (5%) anesthesia, rats were decapitated at the first cervical level and decerebrated precocollarily. The brain stem with cerebellum attached was carefully removed in cold (4°C) Ringer solution (composition in mmol: 125 NaCl, 24 NaHCO3, 5 KCl, 2.5 CaCl2, 1.25 MgSO4, 1.25 KH2PO4, 10 dextrose) and pinned down in a perspex chamber, ventral side up. The basilar artery was cannulated at its caudal-most end (i.e., at the point of VA convergence), with a double-lumen glass pipette (pulled from theta glass; tip diameter ~150 μm) held in a three-dimensional micromanipulator and tied in place with a fine silk suture (see Fig. 2A). The preparation was perfused through one lumen of the cannula with gas bubbled Ringer (32°C) plus 1.5% polyethylene glycol (osmotic agent; 20,000 molecular weight) at an initial rate of 0.2 ml/min. Perfusate was pumped through a bubble trap and nylon mesh (pore size, 20 μm). To vascularly isolate the medulla oblongata, the basilar artery was then clamped just caudal to its bifurcation into the superior cerebellar arteries to prevent excessive washout of perfusate through cut vessels rostrally. Once warmed for 10 min (32°C, same as the in situ preparation), flow was increased incrementally by 0.2 ml/min every 10 min across five additional flow rates to a maximum of 1.2 ml/min. Pressure was recorded via a transducer attached to the second, fluid-filled lumen of the theta glass cannula. The preparation was superperfused with prewarmed (32°C) Ringer bubbled with 95% O2, 5% CO2.

Working Heart-Brain Stem Preparation In Situ

This study used the in situ perfused brain stem-spinal cord preparation of the juvenile rat (as described previously, Ref. 36) of similar age to those used in the in vitro experiments. In brief, preheparinized male 4- to 5-wk-old Wistar (n = 6; 63 ± 3 g) and SH (n = 6; 71 ± 3 g) rats were anesthetized deeply with 5% halothane until loss of withdrawal reflexes, as described above. Rats were bisected below the diaphragm, skinned, exsanguinated, and cooled in ice-chilled carbogenated artificial cerebrospinal fluid, and the brain was decerebrated precocollarily. Lungs were removed, and the descending aorta isolated before the preparation was transferred to a recording chamber, ventral side up. The aorta was cannulated retrogradely just rostral to the renal arteries with a double-lumen catheter (1.25 mm, DLR-4, Braintree Scientific) and perfused with carbogenated artificial cerebrospinal fluid, and the brain was decerebrated precocollarily. The thoracic aorta was clamped just rostral to its bifurcation into the aortic arch and left descending aorta. The brain stem-spinal cord preparation was superfused with carbogenated artificial cerebrospinal fluid (32°C) plus 1.5% polyethylene glycol (as above) at an initial rate of ~20 ml/min. The second fluid-filled lumen of the catheter was connected to a pressure transducer to record perfusion pressure within the aorta. Neurorvuscular blockade was achieved by adding vecuronium bromide (4 μg/ml to the perfusate). The right phrenic nerve and thoracic sympathetic chain (level T8) were isolated, and simultaneous recordings were made using separate bipolar suction electrodes, each mounted in three-dimensional micromanipulators. Both signals were amplified, band-pass filtered (0.5–5 kHz), and acquired in an A/C converter (CED 1401, Cambridge Electronic Design) to a computer using Spike 2 software. Once the phrenic nerve was isolated, flow rate was increased to 30 ml/min, and 400 μl of vasopressin were added to the perfusate to achieve a target perfusion pressure of 60–70 mmHg; this pressure was chosen as it is comparable with that found in 4- to 5-wk-old in vivo rats (26). At this pressure, an ephaptic pattern of phrenic nerve activity was generated, which was used as an indicator of preparation viability.

VA Occlusion In Situ

To isolate the VAs, a modified version of the surgical approach described previously (45) was followed. Using a surgical microscope, the subcutaneous connective tissues and muscles on the ventral surface of the neck were gently retracted to expose the trachea and thyroid. The trachea was separated from surrounding tissue and carefully removed to allow better visualization. The two common carotid arteries were identified, and a fine silk suture placed around both so that they could be visualized at all times. The anterior tubercle of the atlas was identified, and the cervical vertebral bodies exposed.
The VAs were visualized between the second and third transverse processes. Using microvascular clips (Fine Science Tools, product no. 18055-04), the left VA was clamped for 2 min, released for 5 min, and then the right VA was clamped for 2 min, released for 5 min, and then both VAs were clamped simultaneously for up to 1 min. Perfusion pressure, thoracic sympathetic nerve activity (tSNA), and phrenic nerve activity were recorded throughout the protocol. The common carotid arteries were left untouched and remained patent throughout the experiment. Note, perfusion flow rate, composition, pH, arterial PCO2, and O2 were made identical between rat strains; this was a reason for using the working heart-brain stem preparation (WHBP). Another was that it is unanesthetized (and decerebrated), since anesthe sia depresses the Cushing response (5).

Data Analysis of In Vivo and In Situ Studies

Vascular resistance study. All perfusion pressure data were recorded during the experiment using Spike2 software and then analyzed offline. Perfusion pressure readings were taken at the end of the first 10-min warming stage and then at the end of every 10-min phase of increased flow thereafter. Perfusion pressures at each flow rate were compared with a two-tailed Student’s t-test, and differences were considered significant if \( P < 0.05 \).

Working heart-brain stem preparation. All pressure data and sympathetic nerve activity were recorded during the experiment using Spike2 software and then analyzed offline. Perfusion pressure immediately before VA clamping was compared with peak pressure within the 2 min following clamping. Absolute increases in perfusion pressure were compared using a correction factor based on the difference in body weight between the rat strains (see Table 1) further increased the statistical difference in basilar wall thickness at all ages from P7 to P92 (\( P < 0.01 \)).

Functional Studies

In all in vitro and in situ functional studies, there was no significant difference in body weight between PHSH and Wistar rats (\( P > 0.1 \)), as animals used were of different ages (P28–P35).

Vascular resistance study in vitro. Average recorded pressures within the basilar arterial tree were significantly higher (\( P < 0.05 \)) across all flow rates in the PHSH (\( n = 7 \)) compared with Wistar (\( n = 9 \)) rats (Fig. 2B). At the highest flow rate (1.2 ml/min), perfusion pressures were 173 ± 8 vs. 140 ± 6 mmHg for PHSH and Wistar rats (\( P = 0.002 \)). At the lowest flow rate (0.2 ml/min), pressures were 52 ± 6 vs. 38 ± 2 mmHg (Fig. 2B), and this corresponded to a 35% increase in vascular resistance in the PHSH rat basilar arterial tree compared with Wistar rats (i.e., 234 vs. 172 mmHg·ml\(^{-1}\)·min; \( P < 0.05 \)).

SH rats were hypertensive at 6 wk of life (systolic blood pressure: 142 ± 3 mmHg). At this age, Wistar rats had a systolic blood pressure of 104 ± 3 mmHg. Arterial pressure in SH rats plateaued between 12 and 14 wk of age (systolic blood pressure: 176 ± 4 mmHg). At this age, Wistar rats had a systolic blood pressure of 115 ± 4 mmHg.

Comparative Morphological Analysis of Basilar Arteries in Wistar and SH Rats

Wistar and SH rats of identical postnatal age were used. Our main findings were both an increase in basilar artery wall thickness from P7 to P92 (\( P < 0.05 \)) and a lower vessel lumen to wall thickness ratio from P26 (\( P < 0.05 \)) in SH rats relative to age-matched Wistar rats (Fig. 1). Although a trend was apparent (Fig. 1), there was no statistical difference in lumen diameter between the two rat strains. As described previously (9), the hypertensive rat strain was smaller and weighed less in the following age groups: P7 (\( P = 0.08 \)), P26 (\( P < 0.001 \)), and P44 (\( P < 0.001 \)), whereas P2 and P92 groups were similar (\( P > 0.1 \); see Table 1). Consequently, correcting for this difference using a correction factor based on the difference in body weight between the rat strains (see Table 1) further increased the statistical difference in basilar wall thickness at all ages from P7 to P92 (\( P < 0.01 \)).

Fig. 1. A: representative coronal sections of the basilar artery at corresponding brain stem levels of Wistar (i) and spontaneously hypertensive rats (SH); ii) at 2, 7, 26, 44, and 92 days of age [postnatal (P)]. B: age-related changes in basilar artery wall thickness (i) and the lumen-to-basilar wall thickness ratio (ii). According to our studies and those of Refs. 1, 4, 9, and 33, arterial pressure in the spontaneously hypertensive (SH) rat starts to diverge at 28 days and become hypertensive by 6 wk, times when morphological changes in the basilar artery have already occurred. Values are means ± SE. *\( P < 0.05 \), **\( P < 0.01 \).
Table 1. Numbers of animals per age group and their corresponding body weights

<table>
<thead>
<tr>
<th>Age, days</th>
<th>Wistar Body weight, g</th>
<th>SH Body weight, g</th>
<th>P Value</th>
<th>Correction Factor</th>
</tr>
</thead>
<tbody>
<tr>
<td>P2</td>
<td>4.87 ± 0.3</td>
<td>4.20 ± 0.4</td>
<td>0.16</td>
<td></td>
</tr>
<tr>
<td>P7</td>
<td>12.1 ± 0.8</td>
<td>9.6 ± 0.2</td>
<td>0.08</td>
<td>1.26</td>
</tr>
<tr>
<td>P26</td>
<td>65.3 ± 1.0</td>
<td>39.2 ± 1.6</td>
<td>&lt;0.001</td>
<td>1.67</td>
</tr>
<tr>
<td>P44</td>
<td>183.7 ± 5.1</td>
<td>138.5 ± 5.8</td>
<td>&lt;0.001</td>
<td>1.33</td>
</tr>
<tr>
<td>P92</td>
<td>262.9 ± 0.6</td>
<td>266.5 ± 7.4</td>
<td>&gt;0.7</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE; n, no. of animals. P, postnatal age in days; SH, spontaneously hypertensive. To permit a comparison based on similar body weight between rat strains, a correction factor was applied to all basilar artery measurements based on a ratio calculated from the difference in body weight (e.g., Wistar weight/SH rat weight).

**WHBP in situ.** Baseline parameters were similar between groups, with no significant difference in phrenic burst amplitude, duration, or frequency between rat strains (Wistar n = 6; PHSH rats n = 6; P > 0.1). This is consistent with that reported previously for similarly aged SH and normotensive rats (41). There was a clear trend toward an increased basal perfusion pressure in the PHSH compared with Wistar rats (71 ± 4 vs. 64 ± 1 mmHg) at equivalent flow rates (30 ml/min), as reported previously (41), although this difference did not reach significance (P = 0.13).

**tSNA and arterial perfusion pressure changes following VA clamping in situ.** In all Wistar rat preparations (n = 6) and all but one PHSH rat preparation (n = 6), occlusion of either the right or left VAs individually had no significant effect on either tSNA or perfusion pressure. In one PHSH rat preparation, there was a 50% increase in tSNA and a 4-mmHg rise in perfusion pressure following occlusion of the left VA alone. In the same preparation, there was no effect following occlusion of the right VA alone. The same preparation showed a 75% increase in tSNA and a 7-mmHg rise in blood pressure following bilateral VA occlusion. In two other preparations, there were small (~1 mmHg) rises in perfusion pressure immediately after clamping individual VAs. We assumed this was a direct effect of clamping, given the instantaneous nature of the rise in pressure, and that there was no associated rise in sympathetic activity within this time frame. For all subsequent analyses, only data following bilateral VA occlusion were analyzed and presented here.

There was a measurable increase in tSNA and blood pressure following bilateral VA occlusion in all PHSH and Wistar rat preparations (see Fig. 3 for example tracings). The increase in tSNA was significantly higher in PHSH compared with Wistar rats (Fig. 4; 57 ± 13 vs. 20 ± 6%; P < 0.01). This resulted in a significantly higher rise in perfusion pressure in PHSH rats (8 ± 1 vs. 3 ± 1 mmHg; Figs. 3 and 4; P < 0.01), the time course of which was consistent with it being mediated by the rise in sympathetic nerve activity (41). The relatively small rise in perfusion pressure is a result of the open circulatory system in the in situ preparation. This rise in perfusion pressure computed to significantly greater increase in vascular resistance in the PHSH (0.26 mmHg·ml⁻¹·min) relative to the Wistar rat (0.1 mmHg·ml⁻¹·min; Fig. 4; P < 0.05).

Although individual rats showed differences, there was no consistent change in either the frequency or amplitude of the activity in the phrenic nerve during the 2 min following bilateral VA occlusion between the rat strains (P > 0.1). However, if both VAs remained clamped beyond 2 min, all preparations showed a gradual decline in both phrenic nerve frequency and amplitude, and tSNA until PNA activity ceased completely. If one or both VA clamps were released, phrenic nerve activity resumed spontaneously, and tSNA returned to basal (preclamp) levels.

**DISCUSSION**

The present study has demonstrated that there is a vascular remodeling of the basilar artery in the PHSH rat brain stem, and this translates functionally to an increase in vascular resistance of the basilar arterial tree when pump perfused. Our finding of decreased basilar artery lumen to wall thickness in PHSH rats indicates a disproportionate size of the arterial wall, independent of hypertension, which we found developed at 6 wk of life; the latter is consistent with previous studies (1, 4, 9, 33). Furthermore, we found that brain stem ischemia, triggered by bilateral VA clamping, caused a greater increase in tSNA.
and associated rise in peripheral vascular resistance in the PHSH compared with the age-matched Wistar rat, as measured in the in situ preparation. In terms of the sympathoexcitatory response, this suggests that the PHSH brain stem is more responsive to a reduction in arterial perfusion than that in normotensive rats.

Study Limitations

We acknowledge that a caveat of our morphological study was that the basilar arteries were not fixed at pressure that corresponded to the physiological level at each postnatal age. There were two technical issues here: 1) it was not possible to perform intracardiac fixation perfusion even through the left ventricle in the youngest animals; and 2) we were unable to measure arterial pressure in animals up to 5 wk of age. Hence, even if perfusion fixation were possible, it was unknown at what arterial pressure to perfuse-fix the immature groups rats. Because of these limitations, we postfixed the tissue, such that all animals were treated identically. Therefore, we believe our comparative analysis of the differences between rat strains is valid. Additionally, both the isolated brain stem and WHBP have limitations, as both were studied at 32°C, a temperature that does not support cerebral autoregulation (42), as ratified in the present study. Notwithstanding a rightward shift in the autoregulatory pressure range between normotensive and hypertensive rats (34), if autoregulation were intact, one might expect to see lower increments in pressure at lower flow rates. However, even if the preparation was warmed to 37°C, it is...
unlikely that a truly physiological autoregulatory response would be seen due to a lack of various factors present in blood, including erythrocytes (13). Thus partial or aberrant autoregulation in vitro could be seen as a potential confounding factor, and our results may, therefore, give a better indication of fixed or “structural” resistance. That said, there is a distinct difference in this resistance in the PHSH rat.

An important difference between the WHBP and events in vivo relates to the issue of collateral blood flow. One of the biggest advantages of the WHBP is that, once setup, no anesthesia is required, because the preparation is decorticated. Various anesthetics have been shown to affect sympathetic activity in the rat (31), and their use would demand extra caution when interpreting results. However, decortication may affect collateral blood supply to the brain stem via the circle of Willis, given that all rostrally projecting cerebral arteries will be severed. We tried to minimize this by decortication rather than precollricular decerebration, a procedure also used with the WHBP. However, assuming that decortication would have a similar action on brain stem flow in both rat strains, any effect should be the same.

Remodeling of Cerebral Vasculature and High Blood Pressure

A previous study noted vascular remodeling of the aorta and carotid arteries in the PHSH rat, including both an increased wall thickness and lumen-to-wall thickness ratio, indicating a disproportionate size of the arterial wall relative to the luminal space (12). This is consistent with our observations of the basilar arteries in this rat strain. There are reports of hypertrophy in cerebral arteries, but in the adult (hypertensive) stroke-prone SH rat (18, 21). Indeed, Harrap and colleagues (19, 20) found that much of the arterial remodeling could be reduced by treating PHSH rats with an angiotensin-converting enzyme inhibitor, and this included the cerebral vessels. In the latter studies, it was shown that the reversal of the remodeling was most marked in the renal, splanchnic, and cerebral circulations. This may imply that the remodeling is vascular bed specific. It is of interest that these three vascular beds play a major role in determining vascular resistance, either directly (splanchnic) or indirectly via sensory perception of blood flow and blood content (cerebral and renal) to reflexly increase sympathetic vasomotor tone. In a previous pilot study, our laboratory reported a narrower basilar artery lumen in PHSH rats (38). Although a trend was apparent (Fig. 1), we have not confirmed this finding here and suggest it reflects differences in methodology in tissue preparation, such as intracardiac perfusion under pressure vs. postfixation. Nevertheless, a consistent finding between this previous study and the present one is the increased basilar artery wall thickness. This would reduce vascular compliance and explains the increased resistance that we have found through arterial perfusion of the isolated brain stem in vitro.

Sympathetic Nervous System and Remodeling of Brain Stem Vasculature

Sympathetic overdrive may initiate and/or maintain hypertension both in humans (16) and in the SH rat (37, 41). The sympathetic overdrive in the adult SH rat (24, 30) appears to be independent of changes in baroreflex (24) or peripheral chemoreceptor reflex function (22). Sympathetic nerve activity is elevated in juvenile SH rat (41), and neonatal sympathectomy attenuates the development of hypertension in later life (29). As well as the morphological vascular changes described in the PHSH rat brain stem (this study), there is increased vessel wall mass in the aorta (12), carotid (12), and mesenteric vessels (27) of the neonatal SH rat. Neonatal sympathectomy has been shown to prevent hyperplastic changes in small mesenteric arteries and resistance vessels (29). Treatment of PHSH rats with an angiotensin-converting enzyme inhibitor produces a life-long ameliorating effect on their hypertension, and this is associated with a reduction in the hypertrophy of arterial vessels, particularly those feeding renal, splanchnic, and cerebral vascular beds (19, 20, 23). The contribution of the sympathetic nervous system in driving the change seen in the PHSH rat brain stem vessels remains unknown. There is evidence of increased sympathetic innervation of vertebral and basilar arteries of adult SH compared with Wistar-Kyoto rats (28, 32), which some believe to be responsible for the hyper-

Fig. 4. Percentage increase in tSNA absolute PP and absolute vascular resistance following bilateral VA occlusion in SHR and Wistar rats. Values are means ± SE. *P < 0.05, **P < 0.01.
trophy of the main cerebral vessels in the mature SH rat (32). Moreover, prenatal and postnatal treatment of SH rats with hydralazine did not prevent arterial hypertrophy, at least in the renal bed, suggesting that this is not secondary to the hypertension (32). Thus the SH rat appears to have a genetic predisposition to widespread narrowing of the peripheral vasculature, which includes the cerebral arteries, and this is at least partly driven by increased sympathetic activity (29, 46) that is independent of the hypertension (41). Thus remodeling of cerebral vessels before onset of hypertension is not specific to the cerebral circulation. However, what is important to establish is the degree of media thickening (and luminal narrowing) between vascular beds, as this may be vascular bed specific. This remains unknown in the rat, but the human studies suggest it was greatest in the cerebral circulation (11).

Brain Stem Hypoperfusion and Arterial Pressure

Our data indicating higher brain stem vascular resistance in PSHH suggest that, at an equivalent arterial pressure, the brain stem of this rat strain may be less well perfused relative to that of the Wistar rat. This is most likely to be the case in the PSHH rat when its vertebrobasilar resistance is relatively high, yet its arterial pressure is not different from that of age-matched normotensive rats. In cases in which arterial pressure may fall below the lower autoregulatory limit, such as during sleep (10), we hypothesize that brain stem hypoperfusion might lead to oxygen debt in the SH rat, which, in turn, increases sympathetic activity, causing arterial hypertrophy and raised arterial pressure. The latter response would provide sufficient perfusion pressure to overcome the enhanced vascular resistance within the brain stem, thereby maintaining oxygen levels (Cushing response; Ref. 5). This hypothesis remains to be fully tested. However, it is notable that, in the SH rat, autoregulatory curve is right shifted, such that a small fall in arterial pressure causes a precipitous fall in brain stem oxygenation (J. F. R. Paton, unpublished observations). Also, there appears to be a shift toward nonoxidative metabolism in the SH rat brain stem (M. Toward, S. Kasparov, and J. F. R. Paton, unpublished observations) and poor neuronal vascular coupling (i.e., functional hyperemia) in the SH rat brain stem, indicating an inability to vasodilate when oxygen demand is increased (J. T. Potts and J. F. R. Paton, unpublished observations). Thus the cerebral circulation in SH rat is highly compromised and has marginal reserve to maintain normal oxygenation.

The sensing mechanism(s) and location within the brain that detects reduced blood flow to augment sympathetic drive are unknown. In the rabbit and rat, the sympathoexcitatory response triggered by brain stem ischemia persists following transection at the pontomedullary junction (2, 7). In the rabbit, the response was subsequently shown to be markedly reduced following bilateral lesions to the rostral ventrolateral medulla (8). However, in the rat, a good sympathoexcitatory response persisted after spinalization, suggesting multiple loci are involved throughout the central neural axis (2). While Cushing originally envisaged a mechanism that sensed anoxia within the brain, the speed of response seen in some experiments has led other authors to argue in favor of a “central baroreceptor” (35, 40). Presympathetic rostral ventrolateral medullary neurons are responsive to anoxia (44). Based on current evidence, the transduction mechanism may involve increased potassium (44) or sodium (25) channel conductance or involve a heme oxygenase mechanism (6). These mechanisms may all contribute to the sympathoexcitatory responses to VA occlusion reported herein and may even be sensitized in the SH rat. But the actual stimulus that mediates the elevations in sympathetic activity when vertebrobasilar flow is reduced remains unknown and could include the following: changes in shear force, or activation of an intracranial flow or pressure detector system or hypercapnia. The latter is unlikely, as we have recently found that premotor rostral ventrolateral medulla neurons responsive to hypoxia and cyanide are not excited by hypercapnia (T. Koganezawa and J. F. R. Paton, unpublished observations).

Conclusions

The SH rat has a remodeled basilar artery from the 1st day of life, yet hypertension is not evident until 6 wk of life. This suggests that the structural alterations occur in utero and may be congenic and, as discussed here, driven by high sympathetic drive. We also show that brain stem vascular resistance is raised in immature SH rats before the onset of hypertension. The higher vertebrobasilar vascular resistance in the PSHH rat, together with an equivalent arterial pressure to the normotensive rat, may sensitize the Cushing response. Thus small reductions in brain stem blood flow result in sympahtoactivation and increases in vascular resistance and vascular hypertrophy in the PSHH rat compared with normotensive rats. We hypothesize that such a mechanism could participate in the development and/or maintenance of hypertension in the SH rat.

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GRANTS

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

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