Abnormal Ion and Water Composition of Veins and Normotensive Arteries in Coarctation Hypertension in Rats

Motilal B. Pamnani, M.D., Ph.D., and Henry W. Overbeck, M.D., Ph.D.

SUMMARY We examined the water, sodium, and potassium composition of the thoracic aorta, abdominal aorta (plus iliac arteries), and veins (vena cava and portal vein) from rats with aortic coarctation. The aortas of 10 rats (group A) were coarcted above the renal arteries to produce hypertension. Control groups consisted of 10 rats sham-coarcted above and 10 rats coarcted below the renal arteries. In group A rats heart weights and carotid artery pressures were elevated over controls (P < 0.01), whereas there were no significant differences in femoral arterial pressures. In group A rats both the hypertensive thoracic aorta and the normotensive abdominal aorta contained about 20% more water per unit of wet weight, and about 35% and 60% more sodium and potassium, respectively, per unit of dry weight than did the corresponding portions of aorta from control rats (P < 0.01). In group A rats water (P < 0.01), sodium (P < 0.02), and potassium (P < 0.05) contents of veins also were increased. There were no significant correlations between level of carotid arterial pressure and magnitude of changes in arterial and venous composition, nor were there significant differences between the magnitude of changes in the normotensive and hypertensive portions of the aorta. These results indicate that in rats abnormalities in vascular wall salt and water content are not necessarily a direct effect of the elevated pressure in hypertension.

Methods

Normotensive male Wistar rats (obtained from the Otago, New Zealand, colony) approximately 2 months old and weighing 150-180 g were randomly divided into three groups. To create coarctation hypertension in 10 rats (group A), we placed a silver clip, 0.813 mm in diameter, around the abdominal aorta and iliac arteries downstream to the renal arteries. In 10 rats (group B) a clip (diameter = 0.927 mm) too large to constrict the aorta was similarly placed. In a final group of 10 rats (group C), in an attempt to create hypertension in the vessels of the hindquarters, we placed a clip 0.800 mm in diameter on the aorta downstream to the origin of the renal arteries. Systolic blood pressure in the hindquarters of all rats was measured weekly by the tail plethysmographic method under light ether anesthesia.

One month after surgery carotid and femoral arterial pressures were measured directly under light ether anesthesia. The rats were then killed by rapid exsanguination and the blood was used for determination of serum sodium and potassium by flame photometry, serum calcium and magnesium by atomic absorption spectrophotometry, and creatinine by the Autoanalyzer. The descending thoracic aorta, abdominal aorta and iliac arteries downstream to the renal arteries (or to the clip in group C), and the inferior vena cava and portal vein were excised rapidly for analysis of vascular wall ion and water composition. Placed on a glass plate, all specimens were rapidly cleaned of adventitia and opened longitudinally. The tissues were blotted once with filter paper to remove blood and surface fluid, and then weighed to the nearest 0.01 mg. The tissue was then oven-dried at
100°C for 24 hours, and, when again cooled to room temperature, reweighed.

The difference between the two weights was considered water content and was expressed as milliliters per kilogram of wet weight. For measurement of ionic content, 2 ml of 0.1 N nitric acid were added to each tissue and the tissue was allowed to digest for 14 days. Digested specimens were again placed in an oven at 100°C for 48 hours to evaporate the nitric acid. Tissue contents of sodium and potassium were then measured by flame photometry (Beckman). Ion contents were expressed as milliequivalents per kilogram of tissue, dry weight. Student's t-test was used to compare pressures and composition of the vessels among the groups. Student's t-test for paired replicates was used within groups to compare composition of thoracic and abdominal aortae. Linear correlation coefficients were calculated to test for associations between mean carotid arterial pressure and vascular composition. The null hypothesis was rejected at P < 0.05.

**Results**

All rats remained healthy and showed no evidence of cardiac or renal insufficiency or malignant hypertension at the time of study. At this time body weights and serum sodium, potassium, calcium, magnesium, and creatinine concentrations did not differ significantly among the groups (Table 1). In contrast, heart weight (Table 1) was significantly increased in the rats with coarctation hypertension (group A). Mean carotid arterial pressure also was significantly increased in group A rats as compared to both control groups (Fig. 1); carotid pressures were similar in the two control groups. Mean femoral arterial pressures in the three groups did not differ significantly. The pressure gradient between carotid and femoral arteries in the groups were expressed as millimeters of mercury and had a significant correlation with body weight. The regression equation was:

\[ \text{Pressure gradient} = -1.2 + 0.04 \times \text{Body weight} \]

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**Table 1 Mean Values ± SEM for Weights and Serum Electrolytes**

<table>
<thead>
<tr>
<th>Measurements</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body wt (g)</td>
<td>262.2 ± 6.6</td>
<td>264.6 ± 7.0</td>
<td>280.8 ± 4.9</td>
</tr>
<tr>
<td>Heart wt (g/100 g body wt)</td>
<td>0.48 ± 0.02</td>
<td>0.34 ± 0.01</td>
<td>0.35 ± 0.01</td>
</tr>
<tr>
<td>Serum [Na⁺] (mEq/liter)</td>
<td>143.6 ± 2.9</td>
<td>144.6 ± 1.2</td>
<td>145.2 ± 1.7</td>
</tr>
<tr>
<td>Serum [K⁺] (mEq/liter)</td>
<td>4.0 ± 0.4</td>
<td>4.5 ± 0.2</td>
<td>4.5 ± 0.3</td>
</tr>
<tr>
<td>Serum [Ca²⁺] (mEq/liter)</td>
<td>4.9 ± 0.1</td>
<td>5.1 ± 0.1</td>
<td>5.0 ± 0.1</td>
</tr>
<tr>
<td>Serum [Mg²⁺] (mEq/liter)</td>
<td>2.23 ± 0.13</td>
<td>2.17 ± 0.07</td>
<td>2.26 ± 0.20</td>
</tr>
<tr>
<td>Serum creatinine (mg/100 ml)</td>
<td>0.8 ± 0.1</td>
<td>0.8 ± 0.1</td>
<td>0.9 ± 0.1</td>
</tr>
</tbody>
</table>

Values were obtained from 10 rats in each group except for serum creatinine, for which n = 8 in group A, 6 in group B, and 8 in group C. *P < 0.01 for comparison with group A rats.
VASULAR WALL IN HYPERTENSION/Pamnani and Overbeck

GROUP: B A C B A C

ABDOMINAL AORTA

THORACIC AORTA

VEINS

GROUP: B A C

ABDOMINAL AORTA

THORACIC AORTA

VEINS

FIGURE 2 Mean (± SEM) values for content of water (top), sodium (middle), and potassium (bottom) in abdominal aorta, thoracic aorta, and venous tissue of rats from groups A, B, and C. The P values are represented for comparison of each control group with the hypertensive group (group A).

introduced by tissue drying during our dissection procedures which were carried out at room temperature. This systematic error does not affect our conclusions, because we used identical dissection techniques for all rats in the study.

Discussion

In this study hypertension was produced only in the rats whose aortas had been constricted above the renal arteries (group A), confirming the findings of Nolla-Panades and supporting his conclusion that renal factors play an essential role in the development of this form of hypertension in rats.

The results of our present study additionally clearly indicate that abnormalities in vascular wall salt and water content are not necessarily the effects of elevated intravascular pressures in coarctation hypertension in rats. The changes we observed in the composition of both the normotensive abdominal aorta downstream to the coarctation and the veins cannot be attributed to the direct effects of increased intravascular pressure. Furthermore, changes in composition of the hypertensive thoracic aortas upstream to the coarctation were no greater than those in the normotensive abdominal aortas of the same rats. Finally, there were no significant correlations between level of carotid arterial pressure and magnitude of changes in vascular composition.

Our results for rats, therefore, do not agree with those of Hollander et al. and Villamil and Matloff, who found, in dogs with a similar degree and duration of aortic coarctation, that changes in aortic sodium and water composition were confined to the hypertensive portions upstream to the coarctation. Other than species differences, there were technical differences between these previous studies and ours: (1) Our coarctation was of the abdominal aorta, whereas the other groups produced midthoracic coarctation; (2) we sampled the entire aorta, whereas the other groups took only representative samples; and (3) control animals in the other series apparently had not undergone sham surgery, whereas ours had. Thus the differences in results may be explained either on the basis of species differences or on the basis of such technical differences.

Regarding species differences, even in the hypertensive thoracic aorta the changes we observed in rats differed qualitatively and quantitatively from those found by the other two groups in studies on dogs. For example, increases in potassium content were the most impressive change we observed in coarcted rats. Similar changes in potassium were reported by Phelan and Wong in two-kidney Goldblatt hypertensive rats of the same strain, and by other investigators also studying rat aorta in one- and two-kidney Goldblatt hypertension. In contrast, in coarcted dogs, Hollander et al. found no significant changes in aortic potassium content and Villamil and Matloff reported significant decreases in potassium content of both hypertensive and normotensive portions of the aorta (which they attributed to tissue injury). It is of interest that in dogs with bilateral perinephritic hypertension, aortic potassium content has been reported to be unchanged.

Regarding technical differences, it is possible that the degree of renal involvement in the hypertension may be different in abdominal and thoracic coarctation. Although there was no evidence for renal dysfunction in our rats, the normal serum electrolyte and creatinine concentrations certainly do not rule out occult renal abnormalities at the time the vascular tissue was sampled. Thus it is possible that our rats had elevated levels of renin, angiotensin, and aldosterone. Chronic angiotensin infusions elevate arterial sodium contents in dogs, therefore it is possible that the increased vascular sodium we found in our rats may have been related to an action of angiotensin. On the other hand, such angiotensin infusions have not been found to elevate vascular wall water content. Other renal factors not involving the renin-angiotensin-aldosterone system may, of course, also have played a role in the vascular changes we observed.
Whether or not renal factors were involved, we found no good evidence that the vascular changes we observed were causally related to elevated intravascular pressures. Thus, in this regard rats and dogs appear to differ. It should be noted especially that the composition of vessels from normotensive vascular beds of humans with hypertension has not yet been studied. Thus it is certainly too early to conclude on the basis of available data that changes in vascular wall ionic and water composition in humans with hypertension are the result only of elevated intravascular pressure.

In hypertension abnormal water and ionic contents reportedly occur in many other tissues in addition to arteries, including myocardium,10,11 skeletal muscle,10,12,13 submaxillary gland,10 brain,10,14 liver, gut, spleen, and skin.10 In the only previous study of veins, normal wall composition was found in spontaneously hypertensive rats.16 We believe ours is the first report that venous wall composition is abnormal in hypertension.

This abnormal venous composition is noteworthy because recently there has been increased interest in veins in hypertension; elevated venous return to the heart due to decreased venous compliance may help to account for the increased cardiac output in early stages of hypertension.17 We have previously reported that femoral18 and mesenteric19 venous compliances are decreased in peripheric hypertensive dogs. We suggested that abnormal venous wall ionic and water composition may underlie this decreased venous compliance.18 It is also possible that abnormalities in ionic composition of veins may be related to the enhanced responses of veins to norepinephrine and nerve stimulation observed in coarctation hypertension.20 Furthermore, if abnormalities in ionic and water composition in hypertension reflect underlying defects in ion metabolism, as suggested by Tobian,1 our present data would suggest that such underlying defects may be present not only in arterial but also in venous tissue.

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References

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