Changes in Erythrocyte Potassium Concentration in Goldblatt Hypertension of the Rabbit

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SUMMARY

Changes in potassium (K) concentration in erythrocytes (RBC) were investigated during the early developing stage (1 and 2 weeks after) and the chronic established stage (12 to 16 weeks after the renal artery constriction) in two-kidney (group 2H) and one-kidney Goldblatt hypertension (group 1H) of the rabbit. In both group 2H and group 1H, blood pressure was already elevated significantly during week 1, and reached a level about 70 mmHg higher than the pre-constriction level at the chronic stage. It did not change in the control group (group C). During week 2, the change in RBC K concentration showed a significant negative correlation with the change in blood pressure in group 2H (r = -0.529, n = 26, p < 0.01). During the chronic stage, the RBC K concentration was lower in group 2H (103.5 ± 1.7 mEq/l RBC, n = 10, p < 0.01 compared with group C) and in group 1H (102.1 ± 1.6, n = 7, p < 0.001) than in group C (111.6 ± 2.2, n = 9). The change in this parameter from the pre-constriction value was -11.9 ± 2.1 mEq/l RBC (p < 0.001) in group 2H, -13.0 ± 1.8 (p < 0.001) in group 1H, and -2.4 ± 2.6 (not significant) in group C. The results suggest that the intracellular electrolyte metabolism is altered in both types of chronic Goldblatt hypertension.

Additional Indexing Words:
Two-kidney Goldblatt hypertension One-kidney Goldblatt hypertension Developing stage Established stage

Previous studies of Goldblatt hypertension in the rabbit have shown that the serum potassium (K) concentration is decreased in the two-kidney model, but not in the one-kidney model.1,2) However, it is not clear whether the change in serum K concentration is associated with any change in intracellular...
cellular K concentration. In 1943, Eichelberger\textsuperscript{3)} estimated the electrolyte contents of the skeletal muscle of dogs made hypertensive by bilateral renal artery constriction, and reported an increase in sodium (Na) and a decrease in K contents. Subsequently, a number of studies on the electrolyte content of the arterial wall have been performed in various forms of experimental hypertension.\textsuperscript{4)-13)} Although measurements of tissue electrolyte contents provide direct information on the intracellular electrolytes, the necessity to sacrifice the animals precludes the longitudinal assessment of changes in individual animals.

The measurement of electrolyte concentration in erythrocytes (RBCs) offers a useful tool for investigating the changes in intracellular electrolyte content at different stages of experimental hypertension. In this study, we examined changes in K concentration in RBCs during an early developing and a chronic established stage of two-kidney and one-kidney Goldblatt hypertension of the rabbit.

\textbf{MATERIALS AND METHODS}

Male albino rabbits weighing 2.5 to 3.0 Kg were fed 100 Gm of pellets per day (CR-1, Clea Japan, Inc, Tokyo), which provides 14 mEq of Na and 26 mEq of K, and tap water \textit{ad libitum}. The rabbits were divided into 3 groups: 1) a group with two-kidney Goldblatt hypertension (group 2H), in which hypertension was produced by a constriction of the left renal artery with a silver clip (0.9 mm internal diameter), while the right kidney was left untouched; 2) a group with one-kidney Goldblatt hypertension (group 1H), in which hypertension was produced by a constriction of the left renal artery with a silver clip (1.2 mm internal diameter), 4 weeks after the removal of the right kidney; and 3) a control group (group C), in which a sham operation was performed on the left renal artery with the right kidney untouched. Systolic blood pressure in conscious animals was measured weekly by an indirect method\textsuperscript{14)} in the central ear artery dilated by application of a small amount of xylol to the tip of the ear.

Potassium concentration in RBCs was determined before, 1 and 2 weeks after (early stage), and 12 to 16 weeks after (chronic stage) the renal artery constriction. Blood was drawn without anesthesia from the marginal ear vein into a syringe moistened with heparin calcium. Hematocrit was measured in duplicate by centrifuging a blood sample in a microhematocrit tube at 11,000 rpm for 5 min (RC-24B, Tomy-Seiko, Tokyo). The hematocrit data were corrected for trapped plasma by 3%. This correction value was based on the relationship between the mean corpuscular volume (MCV) of
RBCs and trapped plasma,\(^{15}\) where the MCV was calculated from the hematological data for the rabbit.\(^{18}\) The plasma was separated from a portion of the blood sample, and the concentration of K was measured with a flame spectrophotometer (Model 143, Instrumentation Laboratory, Boston, Massachusetts, USA). The K concentration in the whole blood was determined from the remainder of the sample in triplicate, according to the method of Bernstein.\(^{17}\) One-tenth ml of the blood was diluted with 8.9 ml of distilled water, and 1 ml of 40% trichloracetic acid solution was added to precipitate protein. After addition of 10 ml of lithium solution (30 mEq/l) as an internal standard, the sample was centrifuged. Potassium in the supernatant was measured with a flame spectrophotometer. A standard solution containing 100 mEq/l of K was treated in the same way as the blood sample for calibration. Since preliminary experiments had shown that the precipitated protein constituted 1.25% of the final solution in volume, the K concentration in whole blood was corrected by this factor. The K concentration in RBCs was calculated from the hematocrit (Ht), and the K concentration in the whole blood (WK) and plasma (PK) using the formula,

\[
RBC \text{ K} = \frac{(100 \times WK - (100-Ht) \times PK)}{Ht}.
\]

Statistical evaluations were made by Student's t test, and the p values less than 0.05 were taken to indicate statistical significance.

**RESULTS**

The changes in blood pressure and RBC K concentration in the 3 groups are shown in Table I and Fig. 1. The blood pressure in both hypertensive groups was significantly elevated by week 1, and it attained a level about 70 mmHg higher than the pre-constriction level during the chronic stage. There was no significant change in blood pressure in group C.

The RBC K concentration did not change in either hypertensive or control groups during weeks 1 and 2. During the chronic stage, it decreased by 11.9±2.1 mEq/l RBC in group 2H and by 13.0±1.8 mEq/l RBC in group 1H. By contrast, the RBC K concentration remained unchanged in group C, even during the chronic stage.

Confirming the previous results,\(^{11,2}\) the plasma K concentration decreased only in group 2H, falling from 4.2±0.08 mEq/l before the operation to 3.9±0.08 mEq/l during week 2 (n=26, p<0.001). It also tended to decrease during week 1 and at the chronic stage, but the differences from the control values were not statistically significant.

Fig. 2 illustrates the correlation between the changes in RBC K con-
Table I. Blood Pressure and Erythrocyte Potassium Concentration during Early and Chronic Stages of Goldblatt Hypertension

<table>
<thead>
<tr>
<th>Stage</th>
<th>n</th>
<th>Blood pressure (mmHg)</th>
<th>RBC K (mEq/l RBC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group C</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>control</td>
<td>10</td>
<td>93.3±3.6</td>
<td>113.8±0.7</td>
</tr>
<tr>
<td>week 1</td>
<td>10</td>
<td>92.8±3.8</td>
<td>115.7±1.8</td>
</tr>
<tr>
<td>week 2</td>
<td>10</td>
<td>95.5±3.5</td>
<td>114.1±1.1</td>
</tr>
<tr>
<td>chronic</td>
<td>9</td>
<td>102.2±5.3</td>
<td>111.6±2.2</td>
</tr>
<tr>
<td>Group 2H</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>control</td>
<td>26</td>
<td>88.6±1.4</td>
<td>114.6±1.0</td>
</tr>
<tr>
<td>week 1</td>
<td>18</td>
<td>109.7±3.8*</td>
<td>114.5±1.2</td>
</tr>
<tr>
<td>week 2</td>
<td>26</td>
<td>115.4±2.9*</td>
<td>114.2±1.1</td>
</tr>
<tr>
<td>chronic</td>
<td>10</td>
<td>156.1±5.9*</td>
<td>103.5±1.7*</td>
</tr>
<tr>
<td>Group 1H</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>control</td>
<td>30</td>
<td>92.3±1.5</td>
<td>114.5±0.9</td>
</tr>
<tr>
<td>week 1</td>
<td>18</td>
<td>121.7±3.3*</td>
<td>114.8±1.3</td>
</tr>
<tr>
<td>week 2</td>
<td>30</td>
<td>128.9±2.7*</td>
<td>114.6±1.1</td>
</tr>
<tr>
<td>chronic</td>
<td>7</td>
<td>159.6±10.7*</td>
<td>102.1±1.8*</td>
</tr>
</tbody>
</table>

Values are means±SEM.

RBC K = erythrocyte potassium; group C = sham-operated control group; group 2H = two-kidney Goldblatt hypertensive group; group 1H = one-kidney Goldblatt hypertensive group.

*p<0.001 compared with group C.

Fig. 1. Changes in blood pressure (ΔBP) and potassium concentration in erythrocytes (ΔRBC K) after renal artery constriction in the sham-operated control (group C), the two-kidney Goldblatt hypertensive (group 2H), and the one-kidney Goldblatt hypertensive groups (group 1H) of rabbits. Values are expressed as means±SEM. * p<0.005 compared with the pre-constriction value. ** p<0.001 compared with the pre-constriction value.
Fig. 2. Correlation between the changes in potassium concentration in erythrocytes (RBC K) and blood pressure 2 weeks after renal artery constriction in the sham-operated control (group C), the two-kidney Goldblatt hypertensive (group 2H), and the one-kidney Goldblatt hypertensive groups (group 1H) of rabbits.

centration and blood pressure during week 2, when the largest number of animals were examined. In group 2H, the change in RBC K concentration had a significant negative correlation with the change in blood pressure, although the average K concentration in the group did not differ from the pre-constriction value (vide supra). There was no correlation between these parameters in either group 1H or group C.

DISCUSSION

The present study revealed that the RBC K concentration decreases at the chronic stage of Goldblatt hypertension of the rabbit. Regardless of the presence (two-kidney) or absence (one-kidney Goldblatt hypertension) of the contralateral kidney, almost identical results were obtained. On the other hand, no significant change was noted in RBC K concentration during the early stage of hypertension. These findings on RBC K contrast with the results of studies of serum K. The serum K concentration decreases in the two-kidney model and is normal in the one-kidney model.11,2) Furthermore, the decrease in serum K concentration begins during the early developing stage of two-kidney Goldblatt hypertension, which was partly confirmed also in this study.

The time course of the changes in RBC K concentration is rather similar to that of changes in Na content of the arterial wall in one-kidney Goldblatt hypertension of the dog.18) It was shown that the arterial Na content remains normal during the early stage and that it increases during the chronic stage. In a preliminary experiment, we also observed that the RBC Na concentration is unchanged during the early stage, but that it increases
during the chronic stage in both two-kidney and one-kidney Goldblatt hypertension in the rabbit. The disparity between changes in K concentration in serum and RBCs, with respect to models of hypertension and time course, indicates differences in the regulation of extra- and intracellular K.

The processes influencing RBC K concentration were not defined in this study. A negative correlation between the changes in RBC K concentration and blood pressure in the two-kidney model during week 2 may be due to varying degrees of secondary aldosteronism: the higher the plasma renin activity becomes, to the greater extent the blood pressure and plasma aldosterone elevate and the RBC K concentration decreases. This is possible because plasma renin activity is high in this model,1,2) and the RBC K concentration is decreased in primary aldosteronism.19) However, secondary aldosteronism is not a sufficient explanation for the whole picture. There may also be an alteration of the electrolyte transport across the plasma membrane. In essential hypertension18) and genetic hypertension in animals,24)-28) derangements of electrolyte transport in the erythrocyte membrane have been reported. However, studies of secondary hypertension have not shown a similar membrane abnormality.29) Haddy et al.30) have suggested the existence of a humoral factor which suppresses the membrane Na+K+-ATPase activity in several models of experimental hypertension. The ultimate result of a reduction in enzyme activity should be an increased Na and a decreased K concentration in the cell. It is possible that such a humoral factor is responsible for the decrease in RBC K concentration in chronic Goldblatt hypertension. The fact that the elevation of blood pressure preceded the changes in RBC K concentration may indicate that the mechanism involved in these changes is associated with the maintenance rather than the development of hypertension.

It is generally agreed that the Na content of the arterial wall is high in various models of experimental hypertension4)-13) and essential hypertension.31) On the contrary, the results concerning the K content of arterial tissues in hypertension are not unequivocal; some studies have shown an increase,4),6)-8),10),11),32) and others have shown no alteration or a decrease.3),5),9),12) Since most K in tissues is located intracellularly, an increased value of K per unit dry weight of arterial specimens may merely reflect medial hypertrophy, and the intracellular K concentration may in fact be reduced. Therefore, reports of an increase in the K content of arteries do not necessarily contradict the present results.
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