High Salt Intake Attenuates the Development of Hypertension in Two-Kidney, One-Clip Goldblatt Rats (43243)

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Abstract. **Effects of high salt intake on the early onset of hypertension were examined in two-kidney, one-clip rats. They were divided into high salt and control groups which were supplied with 1.0% NaCl and tap water, respectively, as a drinking solution for 12 days after clipping the left renal artery. The high salt group showed a lower plasma renin concentration and a higher plasma atrial natriuretic peptide (ANP) along with an attenuation of the magnitude of early hypertension, as compared with the control group. A significant positive correlation between blood pressure and plasma renin concentration and an inverse correlation between plasma renin concentration and ANP were shown. Cortical renal renin content was comparable between the two groups. In another two groups of sham-clipped rats, the high salt group did not differ from the tap water-drinking group in any of the parameters examined, except that ANP was significantly higher. These results demonstrate that high salt intake attenuates the developmental phase of hypertension in two-kidney, one-clip rats by increasing the ANP and suppressing the release of renin.** [P.S.E.B.M. 1991, Vol 197]

The hypertension in two-kidney, one-clip (2K1C) rats has been attributed to an enhanced activity of the renin-angiotensin system (RAS). Its renin dependency is supported by observations obtained using inhibitors or blockers of the various stages of the cascade of the RAS (1–3). Furthermore, a few studies have shown that a high sodium intake decreases renal renin content (RRC), renin secretory rate, plasma renin activity, and angiotensin II levels (4–7).

Therefore, it might be expected that a high sodium intake would suppress the activity of the RAS and, as a consequence, attenuate the hypertensive response to unilateral renal artery constriction. In fact, high sodium intake has been reported to delay the onset, but not the final magnitude, of 2K1C hypertension (8). Although high salt intake may directly inhibit the RAS in this model of hypertension, the precise mechanism by which the hypertension is delayed has not been studied extensively.

Atrial natriuretic peptide (ANP) has been suggested to act as a safety valve protecting against the pressor and sodium-retaining effects of an overactive RAS (9). In addition, it is known that ANP is secreted in response to various stimuli, including a high salt diet (10) and volume load (11). Therefore, an increase in plasma ANP may be responsible, at least in part, for the delayed hypertension due to the high salt intake.

The study presented here was done to test the hypothesis that a high salt intake attenuates the developmental phase of 2K1C hypertension by increasing plasma ANP levels.

Materials and Methods

Two-kidney, one-clip hypertension was made in ether-anesthetized male Sprague-Dawley rats (150–190 g) by constriction of the left renal artery with a 0.2-mm silver clip. The rats were then divided into two groups, so that one, Clip/NaCl, received 1.0% sodium chloride as a drinking solution and the other, Clip/Water, received tap water *ad libitum*.

Two additional groups of rats, subjected to a sham operation, were also provided as a control. One (Sham/NaCl) was supplied with 1.0% sodium chloride and the other (Sham/Water) with tap water, as were the clipped rats.

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Systolic blood pressure (SBP) was measured indirectly by means of a tail cuff method in conscious, prewarmed (37°C for 10 min) rats. The basal blood pressure was calculated as an average of the values taken on 3 or 4 consecutive days before clipping the renal artery. After the clipping, SBP was measured on Days 3, 6, 9, and 12.

On Day 12, after measuring SBP, the animals were decapitated and blood was collected for measurement of plasma concentrations of renin (PRC) and ANP. Both kidneys were removed and weighed, and cortical slice homogenates were prepared, as described by previous authors (12).

RRC, PRC, and ANP were measured by radioimmunoassay as described previously (13, 14).

Results are expressed as mean ± SE. Each datum point was compared by unpaired Student's *t* test between water-drinking and saline-drinking groups. Likewise, RRC, PRC, and ANP were compared between the groups.

Results

Figure 1 shows the development of hypertension in 2K1C rats. The Clip/NaCl group showed a lesser degree of increase in blood pressure than did the Clip/Water group. No significant changes in blood pressure were observed in sham-clipped rats.

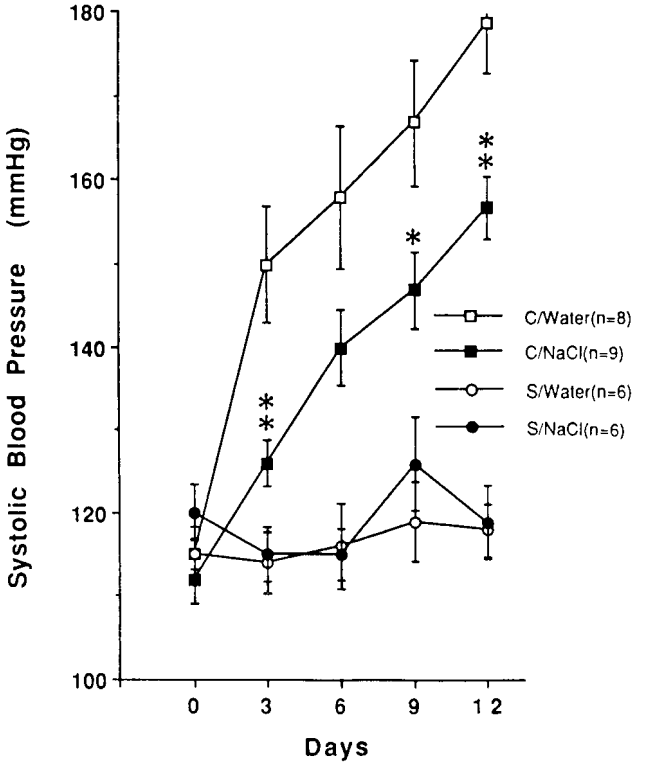

Figure 1. Effect of high salt intake on the development of hypertension in 2K1C rats. Blood pressure was not modified by the high salt intake in sham-clipped rats. *$P < 0.05$ and **$P < 0.01$, compared with C/water group on the corresponding day after clipping the left renal artery. C and S, clipped and sham-clipped rats, respectively.

PRC was significantly lower and ANP concentration higher in the Clip/NaCl than in the Clip/Water group (Table I). In the sham-clipped rats, PRC was not significantly different between Sham/NaCl and Sham/Water groups, although ANP was higher in the former (Table I).

Correlations between these parameters in the clipped rats are shown in Figure 2. SBP significantly and positively correlated with PRC. A significant inverse correlation was obtained between ANP and PRC. There was also an inverse correlation between ANP and SBP; however, it did not reach statistical significance.

RRC of the clipped kidney was significantly higher than that of the contralateral nonclipped kidney (Fig. 3). Comparing the two groups, Clip/NaCl versus Clip/Water, there was no difference in RRC between clipped and between nonclipped kidneys. In sham-clipped rats, no difference in RRC was noted between the kidneys. The clipped kidney weighed less than did the contralateral nonclipped kidney (Table II).

Discussion

The high salt intake attenuated the development of hypertension in association with an increase of ANP and a decrease of PRC in 2K1C rats. A positive correlation between PRC and SBP suggests that the pressor response was renin dependent, as expected.

On the other hand, the regression analysis between ANP and SBP failed to reach statistical significance, although a negative correlation was noted. Although we cannot completely rule out a direct action of ANP to result in a depression, it is unlikely that the increased ANP is a reflection of the attenuated pressor response, inasmuch as the hypertension is associated with an increased level of ANP (15, 16).

Table I. Plasma Concentrations of Atrial Natriuretic Peptide and Renin in the Clipped and Sham-Clipped Rats 12 Days after Clipping[a]

Group	ANP	PRC
Clip/water ($n = 8$)	266.4 ± 28.8	127.3 ± 17.1
Clip/NaCl ($n = 9$)	422.4 ± 40.4[b,c]	49.5 ± 5.6[b]
Sham/water ($n = 6$)	150.7 ± 22.2	47.9 ± 7.0
Sham/NaCl ($n = 6$)	264.1 ± 38.2[d]	36.9 ± 3.7

[a] Units for ANP and PRC are pg/ml and ngAI/ml/hr, respectively.
[b] $P < 0.01$, compared with the corresponding group drinking tap water.
[c] $n = 8$ because one sample is missing due to a technical problem.
[d] $P < 0.05$, compared with the corresponding group drinking tap water.

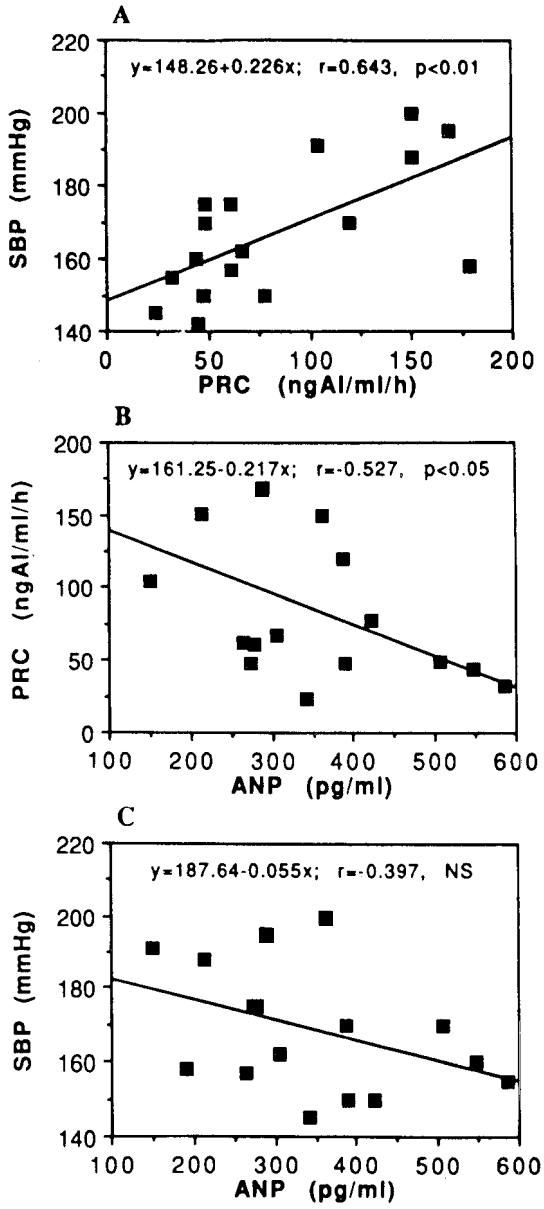

Figure 2. Correlations between PRC and SBP (A), between plasma ANP and PRC (B), and between ANP and SBP (C).

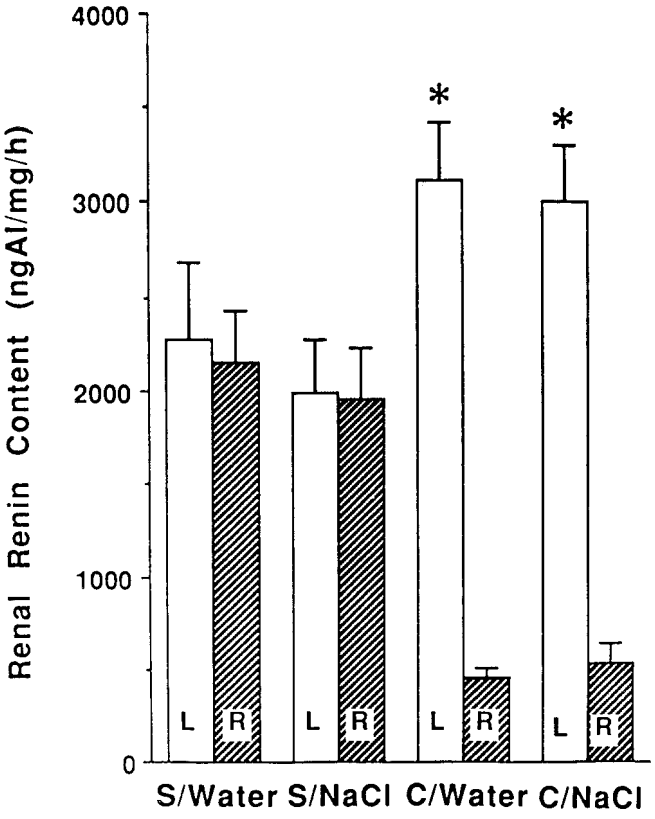

Figure 3. Cortical renal renin content in the four groups of rats. Open columns (L) represent the clipped kidneys and hatched columns (R) represent the contralateral nonclipped. *$P < 0.001$, compared with the contralateral kidneys. The values were not significantly different between the clipped kidneys. The values were not significantly different between the clipped kidneys comparing the C/NaCl versus C/water groups. For details, see legend to Figure 1.

Table II. Right and Left Kidney Weights of Clipped and Sham-Clipped Rats 12 Days after Clipping

Group	Right (g)	Left (g)
Clip/water ($n = 8$)	1.01 ± 0.06	0.83 ± 0.06[a]
Clip/NaCl ($n = 9$)	1.00 ± 0.05	0.84 ± 0.04[a]
Sham/water ($n = 6$)	0.93 ± 0.09	0.92 ± 0.08
Sham/NaCl ($n = 6$)	0.94 ± 0.08	0.94 ± 0.09

[a] $P < 0.001$, compared with the right (nonclipped) kidney.

Therefore, it is hypothesized that the high salt intake increased ANP, which, in turn, caused renin to decrease and blood pressure to be attenuated. The speculation was substantiated by the inverse correlation between PRC and ANP, although a direct inhibition of sodium on the cascade of the RAS may be of primary importance.

In fact, a role for ANP to antagonize the development of renin-dependent hypertension has been suggested. The hypotensive response to ANP has been reported as greater in, or observed only in, saralasin-sensitive 2K1C rats (17, 18). Furthermore, ANP is known to suppress the release of renin from the kidney (19).

Interestingly, changes in plasma and kidney renin levels were unproportional. Although PRC was lower in the high salt group than in the control group, RRC of the clipped kidney was comparable between the two groups. Nakamura *et al.* (20) have also observed a decrease of PRC without changes in either RRC or mRNA content after inhibition of the highly stimulated RAS.

Although these observations support the possibility that renin secretion was inhibited at the posttranslational stage, it is unlikely that the high salt intake (or ANP) affects the synthesis of renin. The lower PRC

may be attributable to a decreased release. No changes in RRC may then be a reflection of increased intracellular destruction, for which we have no explanation at present and for which further studies will be needed.

Jackson and Navar (8), on the contrary, observed a suppression of the kidney renin content of the clipped kidney as well as a reduced PRA to normal values after the high salt intake, along with a delayed development of 2K1C hypertension. The discrepancy may not be fully accounted for, except that they used whole kidney instead of cortical slices. In addition, they attributed the failure to prevent totally the development of hypertension despite the suppression of plasma renin activity to a diminished angiotensin dependency. Although likely, such a speculation demands further clarification.

Sodium chloride is known to be an important factor in the development of hypertension (21). Therefore, one may argue that sodium and water retention is more likely to exacerbate, rather than to ameliorate, the development of hypertension. However, an excess of sodium chloride intake does not always result in elevation of blood pressure (22). The present study may explain part of the mechanisms for a difference in sodium chloride sensitivity.

High salt intake has been reported to have no sustained effect on ANP release (23). If sustained, however, it is doubtful whether the increase of ANP is of physiologic significance in the established phase of renal hypertension. In other words, whether ANP can chronically sustain a blood pressure reduction remains to be determined, although we speculate that the long-term effects of ANP on blood pressure is mediated via interaction with RAS.

Additionally, the clipped kidney weighed less than the contralateral nonclipped kidney, not only in the water-drinking, but also in the saline-drinking rats. The lower PRC in the saline-drinking rats cannot be ascribed to any technical problem in clipping the renal artery.

ANP also increased in the sham-clipped and high salt-fed rats, by which, however, blood pressure was not modified. This finding suggests that ANP is of significance only in that the activity of the RAS is already stimulated. Alternatively, the increment of ANP was not enough to affect the blood pressure for which the ANP level was as low as 60% of that in the corresponding high salt group of clipped rats.

High salt intake does not prevent, but does attenuate, the developmental phase of 2K1C hypertension. The attenuation may be attributable to the associated increase of ANP and decrease of PRC.

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