Studies on Sympathetic Nerve, Renin-Angiotensin and Renal Kallikrein-Kinin Systems, and Water-Sodium Balance in Essential Hypertension

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In order to clarify the relationships among sympathetic nerve, renin-angiotensin and renal kallikrein-kinin systems, and the water-sodium balance in essential hypertension, plasma noradrenaline concentration (pNA), plasma renin activity (PRA), plasma volume (PV), extracellular fluid volume (ECFV), total exchangeable sodium (Nae), urinary kinin excretion, and fractional excretion of sodium (FENa) and of inorganic phosphorus (FEP) were measured during the 4 days immediately after (the first period) and the last 4 days (the last period) of 2 weeks rest following admission with regular diet (Na; 256–300 mEq/day) in 209 patients with mild essential hypertension.

In the first period, the resting level of pNA correlated positively with diastolic blood pressure ($p < 0.02$), mean arterial pressure (MAP; $p < 0.05$) and PRA ($p < 0.001$), and negatively with PV ($p < 0.05$), ECFV ($p < 0.001$) and Nae ($p < 0.001$), respectively. In addition, a significantly inverse correlation was found between MAP and PV ($p < 0.01$), ECFV ($p < 0.01$) and Nae ($p < 0.01$).

Following 2 weeks rest after admission, most patients showed a spontaneous blood pressure fall. In 32 patients with MAP reduction more than 5 mmHg (average: 14 mmHg), PV ($p < 0.001$), ECFV ($p < 0.01$) and Nae ($p < 0.001$) significantly increased, while FENa ($p < 0.001$) and FEP ($p < 0.01$) decreased after 2 weeks rest.

In the last period of 2 weeks rest, no significant correlation was observed between pNA level and MAP, and MAP significantly correlated not negatively but positively with PV ($p < 0.02$) or Nae ($p < 0.05$).

In all of the values obtained in both the first and the last period of 2 weeks rest, there found significantly positive correlations between pNA and FENa ($p < 0.02$) and 24-hour urinary excretion of kinin ($p < 0.05$), respectively. Moreover, urinary excretion of kinin correlated positively with FENa ($p < 0.01$) and negatively with PV ($p < 0.01$). However, following intravenous infusion of noradrenaline (0.1–0.2 $\mu\text{g/kg/min.}$, 120 minutes), urinary sodium excretion ($p < 0.01$) and FENa ($p < 0.05$) significantly decreased and a similar tendency was found in FEP ($0.05 < p < 0.1$).

Key Words:

Essential hypertension
Plasma noradrenaline
Plasma volume
Extracellular fluid volume
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Urinary kinin

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These findings suggest that an increase of sympathetic nerve activity might be an important mechanism which maintains a high level of blood pressure, particularly, during the first period after admission in patients with mild essential hypertension. In addition, this activity might be closely related to body fluid-sodium balance, and to renin-angiotensin and renal kallikrein-kinin systems. On the other hand, after 2 weeks rest, when the sympathetic nerve activity declined the blood pressure level appeared to be more dependent on body fluid and sodium balance which might be regulated, in part at least, by sodium reabsorption in the renal tubule, including the proximal part, possibly via change in the renal kallikrein-kinin system.

RECENTLY, the plasma noradrenaline (pNA) level was reported to be elevated in some patients with essential hypertension¹⁻³ and Louis et al⁴ described the pNA level at supine rest positively correlated with diastolic blood pressure in this disease. On the other hand, Dustan et al⁵ reported that an inverse relationship in the patients was found between plasma volume (PV) and diastolic blood pressure. More recently, Simon et al⁶ observed that extracellular fluid volume (ECFV) positively correlated with diastolic blood pressure (DBP) in patients with sustained essential hypertension. Moreover, it has been suggested by some laboratories⁷⁻⁹ that the activity of both the sympathetic nervous system and the renin-angiotensin system might be influenced by body fluid volume and sodium balance. Thus, many studies have been reported on the role of sympathetic nerve activity, the renin-angiotensin system and water sodium balance with regard to the pathogenetic mechanism of essential hypertension. However, the significance of relationships among these systems in maintaining a high level of blood pressure in essential hypertensives still remains unclear.

On the other hand, our previous studies¹⁰⁻¹³ demonstrated that, following 2 weeks rest after admission, 1) a spontaneous reduction of blood pressure was observed in most cases of mild essential hypertension, 2) this blood pressure fall was associated with a decrease of urinary noradrenaline output and of plasma dopamine- β -hydroxylase (DBH) activity, 3) a significantly positive correlation was found between the changes in blood pressure and those in urinary noradrenaline or in plasma DBH activity, and 4) a remarkable increase of PV, ECFV and total exchangeable sodium (Nae) was observed.

In the present investigation, therefore, in order to further extend our previous studies and to clarify the relationships among sympathetic

nerve activity, the renin-angiotensin system, water-sodium balance and blood pressure in patients with essential hypertension, the plasma noradrenaline level, plasma renin activity, body fluid sodium status and urinary excretion of kinin were measured before and after 2 weeks rest following hospitalization.

MATERIALS AND METHODS

Two hundred and nine inpatients with uncomplicated essential hypertension, one hundred and thirty males and seventy-nine females, ranging from 18 to 60 years of age, were studied. Patients with a glomerular filtration rate less than 60 ml/min./1.48 m² body surface area (BSA) in endogenous creatinine clearance (Ccr) were excluded. All of these patients were either untreated or discontinued antihypertensive drugs at least 2 weeks prior to hospitalization. These patients were kept at rest with regular diet (in the northern part of Japan) containing 256-300 mEq of daily sodium and about 0.8g of calcium and 1.35g of inorganic phosphorus.

The investigation was performed during the 4 days immediately after (the first period) and the last 4 days (the last period) of 2 weeks rest following admission.

In the early morning when the patients remained in at least 1 hour of supine rest after overnight fasting, blood samples for measurement of plasma noradrenaline (pNA) and plasma renin activity (PRA) were taken from a forearm vein, at least 20 minutes after insertion of a butterfly needle without the use of a tourniquet.

After overnight fasting and emptying of the bladder, basal fractional excretions of sodium (FENa) and inorganic phosphorus (FEP) were measured by the 2 hour clearance method¹⁴ after ingestion of 400 ml/m² BSA of water. FENa and FEP were also measured before and after infusion of noradrenaline (NA: 0.1-0.2 μ g/kg/min., 120 minutes). Urinary kinin was

Fig.1. Correlation between plasma noradrenaline concentration (pNA) and diastolic (DBP), and mean arterial pressure (MAP) in the patients with mild essential hypertension, immediately after admission.

determined in both the urine of 2 hour-clearance ($\text{uKinin} \times 100/\text{Ccr}$) and a 24-hour urine which was collected on another day ($\mu\text{g}/\text{day}$).

Plasma noradrenaline concentration (pNA) and plasma renin activity (PRA) were determined by the method of Renzini et al.¹⁵ and Haber et al.¹⁶ respectively. Urinary kinin was measured by radioimmunoassay.¹⁷ Plasma volume (PV) was determined by the ^{125}I -RISA dilution method and was expressed as the percent of the mean value per height of normal men or women (ml/cm^2 normal).¹⁸ Extracellular fluid volume (ECFV: ml/cm^2) and total exchangeable sodium (Nae: mEq/cm^2) were determined by the $^{22}\text{NaCl}$ dilution method.^{19,20}

Blood pressure was measured at least twice or three times daily by the auscultatory method and mean arterial blood pressure (MAP) was calculated. The average of MAP during each 4 day period was calculated.

Mean values were expressed as the standard error of the mean, and all statistical analysis were assessed by Student's t-test.

RESULTS

During the four days immediately after hospital admission (the first period), in the patients with essential hypertension, a significantly positive correlation was found between supine-resting pNA and diastolic blood pressure (DBP: $r=0.361$, $p<0.02$), but not between pNA

Fig.2. Correlation between plasma noradrenaline concentration (pNA) and plasma renin activity (PRA) in supine bed rest, immediately after admission.

Fig.3. Correlation between plasma noradrenaline concentration (pNA) at supine rest and plasma volume (PV), extracellular fluid volume (ECFV) and total exchangeable sodium (Nae), immediately after admission.

Fig.4. Correlation of mean arterial pressure (MAP) with plasma volume (PV), extracellular fluid volume (ECFV) and total exchangeable sodium (Nae), immediately after admission.

and systolic blood pressure, which were measured simultaneously with the blood sampling for pNA in the early morning. In addition, there also was found a positive correlation between pNA level and calculated MAP ($r = 0.291$, $p < 0.05$), as shown in Fig. 1. The pNA level

significantly correlated with simultaneously determined PRA ($r = 0.528$, $p < 0.001$) as shown in Fig. 2. On the other hand, the pNA level negatively correlated with the values of PV ($r = -0.252$, $p < 0.05$), ECFV ($r = -0.445$, $p < 0.001$), and Nae ($r = -0.593$, $p < 0.001$), respec-

Fig.5. Comparison of mean arterial pressure (MAP), plasma volume (PV), extra-cellular fluid volume (ECFV) and total exchangeable sodium (Nae) immediately after (before) and following 2 weeks rest after admission (after) in 32 patients whose mean arterial pressure spontaneously reduced more than 5 mmHg after 2 weeks rest.

Fig.6. Comparison of fractional excretion of sodium (FENa), inorganic phosphorus (FEP) and endogenous creatinine clearance (Ccr) immediately after (before) and following 2 weeks rest after admission (after) in 25 patients whose mean arterial pressure spontaneously reduced more than 5 mmHg after 2 weeks rest.

tively (Fig. 3). Moreover, a significantly negative correlation between MAP and PV ($r = -0.436$,

$p < 0.01$), ECFV ($r = -0.493$, $p < 0.01$) or Nae ($r = -0.532$, $p < 0.001$), respectively, in this

Fig.7. Correlation between plasma noradrenaline concentration (pNA) at supine rest and mean arterial pressure (MAP) following 2 weeks rest after admission.

Fig.8. Correlation between mean arterial pressure (MAP) and plasma volume (PV) and total exchangeable sodium (Nae) following 2 weeks rest after admission.

period was observed (Fig. 4).

Following 2 weeks rest after admission, most patients with mild essential hypertension showed a spontaneous blood pressure fall in association with a decrease of urinary NA and plasma DBH

activity as reported previously^{11,12}. A similar tendency was seen in the lowering of the pNA level in relation to MAP reduction. In 32 patients, whose MAP spontaneously was reduced more than 5 mmHg (average; from 123 ± 2.6 to

Fig.9. Correlation between plasma noradrenaline concentration (pNA) at supine rest and fractional excretion of sodium (FENa), and 24-hour urinary excretion of kinin including all of those measured in both the first and the last period of 2 weeks rest.

Fig.10. Correlation between fractional excretion of sodium (FENa) and urinary kinin in the same urine sample, and between plasma volume (PV) and urinary kinin excretion in 24-hour urine.

109 ± 2.7 mmHg, $p < 0.001$) after 2 weeks rest, PV (from 90.1 ± 2.3 to 100.7 ± 2.4 ml/cm-% normal, $p < 0.01$), ECFV (82.8 ± 3.2 to 89.3 ± 3.2 ml/cm, $p < 0.01$) and Nae (13.7 ± 0.5 to 15.4 ± 0.6 mEq/cm, $p < 0.001$) significantly in-

creased following 2 weeks rest, as compared with each in the first period (Fig. 5). On the other hand, a significant decrease was found in FENa (1.53 ± 0.10 to $1.11 \pm 0.09\%$, $p < 0.001$) and FEP (12.8 ± 0.9 to $9.8 \pm 0.9\%$, $p < 0.01$), while

Fig.11. Changes of urinary sodium excretion (U_{NaV}), fractional excretion of sodium (FENa) and inorganic phosphorus (FEP) before and after infusion of noradrenaline ($0.1-0.2 \mu\text{g/kg/min.}$).

no significant change in endogenous creatinine clearance appeared, following 2 weeks rest (Fig. 6).

In the last four days, of 2 weeks rest, the last period, no significant correlation existed between pNA and MAP (Fig. 7). Moreover, in this period, a significantly positive correlation between MAP and PV ($r = 0.200$, $p < 0.02$) or Nae ($r = 0.212$, $p < 0.05$) was observed as shown in Fig. 8.

On the other hand, the following results were obtained in relationships among pNA, water sodium balance and urinary kinin, including all of those measured in both the first and the last periods of 2 weeks rest. As shown in Fig. 9, the resting pNA level positively correlated with the value of FENa ($r = 0.410$, $p < 0.02$) and of the 24-hour urinary excretion of kinin ($r = 0.384$, $p < 0.05$), respectively. In Fig. 10, a significant correlation was observed positively between FENa and urinary kinin in the same urine sample ($r = 0.779$, $p < 0.01$), and negatively between PV and urinary kinin in 24-hour urine ($r = -0.496$, $p < 0.01$). However, following intravenous infusion of NA ($0.1-0.2 \mu\text{g/kg/min.}$) for 2 hours, urinary excretion of sodium (U_{NaV} : from 206.7 ± 25.5 to $158.0 \pm 20.6 \mu\text{Eq/min.}$, $p < 0.01$) and FENa (1.49 ± 0.18 to $1.09 \pm 0.11\%$, $p < 0.05$) were significantly reduced, and FEP (9.8 ± 1.3 to $8.1 \pm 1.1\%$, $0.05 < p < 0.1$) tended to decrease, as shown in Fig. 11.

DISCUSSION

Although plasma noradrenaline (pNA) concentration was extensively measured as an indicator of the activity of the sympathetic nervous system to assess the role of this system in the mechanism of essential hypertension, no definite conclusion has been established. Recently, many reports¹⁻³ have indicated that a higher pNA level exists in 15-50% of patients with mild essential hypertension as compared with that in normotensive subjects. Our previous study²¹ also demonstrated that 24% of young patients, under 39 years old, with this disease had higher values of resting pNA level than those in age-matched normotensive subjects. Furthermore, some laboratories have reported that a significantly positive correlation was observed between the pNA level and systolic (Cousineau et al²² and Hofman et al²³) or diastolic blood pressure (Eide et al²⁴ and Brecht & Schoeppe²⁵). However, Lake & Ziegler,²⁶ Kopin et al²⁷ and Christensen²⁸ indicated that they could not find any significant difference of pNA levels between essential hypertensive and normotensive subjects, and any correlation between pNA and blood pressure level in these patients.

On the other hand, our previous studies revealed that, in the patients with mild essential hypertension following 2 weeks rest after hospital admission, the blood pressures spontaneously

fell in association with the reduction of urinary NA excretion and plasma DBH activity. Furthermore, it was found that the patients with higher urinary excretion of NA during the first 4 days immediately after admission showed a more marked fall of blood pressure after 2 weeks rest.¹¹ Together with these spontaneous blood pressure falls, significant elevations of PV, ECFV and Nae were observed.¹² In addition, in comparing each of these in the patients with normal PRA, the patients with low PRA showed lower pNA, urinary NA and DBH, and higher PV, ECFV, Nae and blood pressor response to infused NA or angiotensin-II. All of these differences were particularly remarkable during the first period of the admission.¹³ From these findings, the possibility was suggested that pNA levels are partly influenced by changes in PRA and water sodium balance.

Therefore, the purpose of the present study was to establish the involvement of sympathetic nerve activity in the rise of blood pressure, and to clarify relationships among sympathetic nerve activity, the renin-angiotensin system and water sodium balance in essential hypertension.

In this study, immediately after admission, with a regular diet containing 256–300 mEq of sodium which is usual in the northern part of Japan, a significantly positive correlation was found between resting pNA and both DBP and MAP in the patients with mild essential hypertension (Fig. 1). These findings are fairly compatible with those of some previous reports. However, in the last period of 2 weeks rest, no correlation could be found between pNA and blood pressure levels in these patients (Fig. 7). Thus, as to these correlations, it is necessary to consider that a difference in the results will easily arise from a dissimilarity of the conditions for pNA estimation.

On the other hand, in the first period, it was found that PV, ECFV and Nae negatively correlated with MAP (Fig. 4). Tarazi et al.¹⁸ previously reported that a remarkably inverse correlation was found between PV and DBP, and the reduction of intravascular volume might be caused by an increase of venous tone in uncomplicated essential hypertension. Venous tone will be regulated by various factors including sympathetic nerve activity. In this study, a significant correlation of pNA was observed positively with DBP or MAP (Fig. 1) and negatively with PV, ECFV and Nae (Fig. 3). Therefore, it is assumed that lower PV in patients with

higher blood pressure might partly result from an increase of venous tone due to an augmentation of sympathetic nerve activity.

According to the very recent report of Simon et al.,⁶ ECFV positively correlated with DBP. In their study, however, ECFV was measured by inulin space and expressed in ml/kg body weight in patients with sustained hypertension. Since obesity is often seen in patients with essential hypertension, it would be difficult to compare the results obtained in this study with those reported from Simon et al., in which ECFV was expressed by ml/kg of body weight.

In our studies, spontaneous blood pressure fall was observed in many patients with mild essential hypertension following 2 weeks rest after admission. As shown in Fig. 5 and 6, following 2 weeks rest, marked increases of PV, ECFV and Nae, and decreases of FENa and FEP were observed, while Ccr was not significantly changed, in the patients whose MAP was reduced more than 5 mmHg. The reduction of MAP is possibly based on a lowering of sympathetic nerve activity, at least in this study, and an augmentation of body fluid volume will result in reduction of MAP as in generally known. Therefore, the increases of PV, ECFV and Nae may be not primary but rather secondary to the reduction of MAP. Furthermore, these increases of body fluid volume and sodium might be thought of as a result of diminution of renal sodium excretion due to an enhanced sodium reabsorption in the renal tubule. In addition to, in the last period of 2 weeks rest, a significantly positive correlation between MAP and PV or Nae was found (Fig. 8), whereas no significant correlation had been observed between MAP and pNA (Fig. 7). This finding could be partly supported by results, reported by Dustan et al.,⁵ in which a positive relationship of both systolic or diastolic blood pressure and PV was significantly seen in hypertensive men treated with adrenergic blocking drugs or diuretics. Thus, the possibility is considered that the values of blood pressure in hypertensive patients mainly depend on body fluid volume or sodium following diminution of sympathetic nerve activity.

In addition to the above mentioned, in the present study, significantly positive correlations of pNA were found with FENa and the value in the 24-hour urinary excretion of kinin (Fig. 9). And, the fractional excretion of kinin (urinary kinin $\times$ 100/Ccr) also positively correlated with FENa, while the value of the 24-hour urinary

excretion of kinin correlated negatively with PV (Fig. 10). Moreover, our previous study¹⁴ has demonstrated that FENa inversely correlated with both ECFV and Nae. According to Margolius et al^{2,9} the activity of the renal kallikrein-kinin system is remarkably dependent on that of the renin-angiotensin-aldosterone system. Since pNA positively correlated with PRA, it can be considered that in the patients with a higher pNA level, the elevation of urinary kinin is caused at least in some part by an enhancement of the renin-angiotensin-aldosterone system. It is also suggested that the renal kallikrein-kinin system may regulate the body fluid and/or sodium balance via a modulation of sodium reabsorption in the renal tubule.

However, contrary to expectation, urinary sodium excretion and FENa clearly decreased following an acute administration of NA in dosis with 0.1–0.2 $\mu\text{g/kg/min}$ for 120 min (Fig. 11). Although it can be said that there are some remarkable relations among pNA, PV, FENa and urinary kinin, a precise solution to the cause or the mechanism could not be obtained.

From these findings, in the patients with mild essential hypertension, it may be concluded that an augmentation of sympathetic nerve activity is one of the important mechanisms in which a high level of blood pressure is maintained, particularly, in the first period immediately after admission. In addition, sympathetic nerve activity seems to be closely related to water sodium balance, and the renin-angiotensin-aldosterone and renal kallikrein-kinin systems. Following 2 weeks rest after admission, sympathetic nerve activity lowers, and the blood pressure level appears to be more dependent on body fluid and sodium volume, which may be partly regulated by sodium reabsorption in the renal tubule including the renal kallikrein-kinin system. At least, it may be considered that there are some remarkable relationships among sympathetic nerve activity, renin-angiotensin, water sodium balance and renal kallikrein-kinin, but this mechanism still remains unclear.

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