Effects of Angiotensin II Infusion on Renal Excretion of Purine Bases and Oxypurinol

Yuji Moriwaki, Tetsuya Yamamoto, Zenta Tsutsumi, Sumio Takahashi, and Toshikazu Hada

The effect of angiotensin II infusion on the renal transport of purine bases and oxypurinol (a metabolite of allopurinol) was investigated in 5 healthy subjects who were orally given allopurinol (300 mg) 9 hours prior to the study. Angiotensin II was intravenously administered at 8 ng/min/kg for 2 hours. The fractional clearances of uric acid, xanthine, and oxypurinol were significantly decreased during angiotensin II infusion; however, that of hypoxanthine did not change. The urinary excretion levels of uric acid, xanthine, and oxypurinol were also significantly decreased during angiotensin II infusion. These results suggest that angiotensin II infusion affected the renal clearances of uric acid, xanthine, and oxypurinol through direct tubular transport and/or hemodynamic changes. Accordingly, the hypouricemic effect of allopurinol may be exaggerated in hypertensive gout patients with an enhanced renin-angiotensin system, since an increased biological half-life of oxypurinol is expected in these patients.

HYPERURICEMIA IS OFTEN observed in hypertensive patients,[1-3] and the use of diuretics, β-blockers, and hormones such as noradrenaline and/or angiotensin II is assumed to induce it in hypertensive patients. However, the underlying cause for the relationship between hyperuricemia and hypertension is not clearly understood.

Recently, the renal transport of oxypurines (hypoxanthine and xanthine), oxypurinol (allopurinol metabolite), and uric acid has been studied extensively.[4-6] In previous reports, lactate[5] and norepinephrine[7] were found to decrease the urinary excretion of uric acid, xanthine, and oxypurinol, suggesting a common renal transport pathway for these substances. The effect of angiotensin II on the renal transport of oxypurines and oxypurinol, except for uric acid, has not been previously investigated. Therefore, we conducted the present study to determine whether angiotensin II affects the plasma concentrations and urinary excretion of purine bases and oxypurinol.

MATERIALS AND METHODS

Chemicals

Allopurinol was obtained from GlaxoWellcome (Tokyo, Japan). Angiotensin II (human type) was purchased from Yamanouchi Pharmaceutical (Tokyo, Japan). All other reagents were of analytical grade and obtained from Wako Pure Chemical Industries (Osaka, Japan).

Subjects

After informed consent was obtained, 5 adult male volunteers aged 29 to 48 years (mean $\pm$ SD, 37 $\pm$ 8 years) were enrolled in the present study. Their height and body weight were 173.9 $\pm$ 8.2 cm and 64.6 $\pm$ 7.1 kg, respectively. Both physical and laboratory examinations found normal results.

Experimental Design

The experiment was performed in a quiet research laboratory after an overnight fast. Nine hours prior to the study, 300 mg of allopurinol was taken orally. The subjects remained seated, only standing to void urine, and fasted throughout the experiment, except for drinking water ad libitum to encourage adequate diuresis. Following a 60-minute control period (period 1), angiotensin II was infused intravenously into a forearm vein at 8 ng/min/kg (5 μg/mL in physiological saline) over 120 minutes. Urine was collected at 60 minutes (period 2) and 120 minutes (period 3) after beginning the infusion period. Blood samples were taken from the opposite arm at the midpoint of each urine collection period and collected into a tube containing EDTA-2Na, after which plasma was immediately separated to prevent hypoxanthine leakage from red blood cells. Concentrations of uric acid, oxypurines (hypoxanthine, xanthine), oxypurinol, and creatinine were measured in each urine and blood sample. As controls, 5 representative subjects received a saline infusion and followed the same experimental protocol with at least a 2-week interval.

Analysis of Blood and Urine Samples

High-performance liquid chromatography (HPLC) was employed to quantitate the concentrations of oxypurines (hypoxanthine, xanthine) and oxypurinol. Plasma levels of oxypurines were measured using a modified method of Yamamoto et al using HPLC[8] as follows. The column was a Wakosil C18 (inner diameter, 4.6 mm; length, 150 mm) (Wako Pure Chemical Industries), to which a 30 μL sample was applied. The mobile phase consisted of 0.02 mol/L potassium phosphate buffer (pH 2.2) and the flow rate was 1.0 mL/min. The urinary concentrations of oxypurines were determined using a Wakosil 5C-18-200 (inner diameter, 4.6 mm; length, 250 mm) by the method described above, except that the mobile phase was 0.02 mol/L potassium phosphate buffer (pH 4.25). Clearances of uric acid (Cua), hypoxanthine (Chx), xanthine (Cx), oxypurinol (Cox), and creatinine (Ccr), along with the percent ratios of Cua/Ccr (fractional uric acid clearance), Chx/Ccr (fractional hypoxanthine clearance), Cx/Ccr (fractional xanthine clearance), and Cox/Ccr (fractional oxypurinol clearance) were calculated. Uric acid was measured by the uricase method. Other substances such as creatinine, electrolytes, renin activity, aldosterone, and angiotensin II were measured in our laboratory.

Statistics

Data are expressed as means $\pm$ SD and were analyzed for differences using Student's paired *t* test. Differences were considered significant at a *P* value less than .05.

From the Division of Endocrinology and Metabolism, Department of Internal Medicine, Hyogo College of Medicine, Hyogo, Japan.

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Address reprint requests to Yuji Moriwaki, MD, Division of Endocrinology and Metabolism, Department of Internal Medicine, Hyogo College of Medicine, Mukogawa-cho 1-1, Nishinomiya, Hyogo 663-8501, Japan.

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Table 1. Effects of Angiotensin II Infusion on Blood Pressure, Plasma Renin Activity, and Aldosterone and Angiotensin II Concentrations After Taking Allopurinol

	Period 1	Period 2	Period 3
SBP (mm Hg)	111.4 ± 11.2	130.6 ± 15.3*	139.4 ± 12.6*
DBP (mm Hg)	76.0 ± 9.8	98.2 ± 12.3*	102.4 ± 5.3*
PRA (ng/mL/h)	2.08 ± 1.20	1.22 ± 0.61†	0.66 ± 0.26†
Aldosterone (pg/mL)	121.8 ± 23.0	180.0 ± 24.5†	232.0 ± 54.0*
Angiotensin II (pg/mL)	9.6 ± 4.8	33.8 ± 18.7†	37.0 ± 22.4†
Ccr (mL/min)	83.2 ± 8.6	82.2 ± 4.0	79.5 ± 12.9
UV (mL/hr)	265.6 ± 164.6	131.4 ± 40.2	136.4 ± 100.3

NOTE. Data are expressed as means ± SD.

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; PRA, plasma renin activity; Ccr, creatinine clearance; UV, urine volume; Period 1, 60 minutes before angiotensin II infusion; Period 2, 60 minutes after beginning angiotensin II infusion; Period 3, 120 minutes after beginning angiotensin II infusion;

*$P < .01$.

†$P < .05$.

RESULTS

Effect of Angiotensin II Infusion on Blood Pressure and Plasma Renin Activity, and Aldosterone and Angiotensin II Concentrations After Taking Allopurinol

As shown in Table 1, angiotensin II infusion caused significant rises in blood pressure (period 1, 111.4 ± 11.2/76.0 ± 9.8 mm Hg; period 2, 130.6 ± 15.3/98.2 ± 12.3 mm Hg; period 3, 139.4 ± 12.6/102.4 ± 5.3 mm Hg; $P < .01$, respectively); however, creatinine clearance was not changed (period 1, 83.2 ± 8.6 mL/min; period 2, 82.2 ± 4.0 mL/min; period 3, 79.5 ± 12.9 mL/min; differences not significant [NS]). Plasma concentrations of angiotensin II and aldosterone were also significantly increased during angiotensin II infusion (angiotensin II: period 1, 9.6 ± 4.8 pg/mL; period 2, 33.8 ± 18.7 pg/mL; period 3, 37.0 ± 22.4 pg/mL; $P < .05$, respectively; aldosterone: period 1, 121.8 ± 23.0 pg/mL; period 2, 180.0 ± 24.5 pg/mL; period 3, 232.0 ± 54.0 pg/mL; $P < .05$ and $P < .01$, respectively), whereas plasma renin activity (PRA) was significantly decreased (period 1, 2.08 ± 1.20 ng/mL/h; period 2, 1.22 ± 0.61 ng/mL/h; period 3, 0.66 ± 0.26 ng/mL/h; $P < .05$). Saline infusion at the same rate as that of angiotensin II caused no significant changes in blood pressure, PRA, plasma concentrations of aldosterone and angiotensin II, or creatinine clearance (data not shown).

Effect of Angiotensin II Infusion on the Plasma Concentrations of Purine Bases and Oxypurinol After Taking Allopurinol

Plasma concentrations of uric acid, oxypurines (hypoxanthine, xanthine), and oxypurinol did not change significantly after angiotensin II infusion (Table 2). Moreover, saline infusion also caused no significant changes in the plasma concentrations of purine bases and oxypurinol (data not shown), as has been reported previously.[5]

Effect of Angiotensin II Infusion on the Urinary Excretion of Purine Bases and Oxypurinol After Taking Allopurinol

As shown in Table 2, urinary excretion of uric acid, xanthine, and oxypurinol decreased significantly during angiotensin II infusion (uric acid: period 1, 0.16 ± 0.03 mmol/h; period 2, 0.10 ± 0.02 mmol/h; period 3, 0.07 ± 0.02 mmol/h; $P < .01$, respectively; xanthine: period 1, 19.99 ± 8.78 μmol/h; period 2, 12.65 ± 7.22 μmol/h; period 3, 11.38 ± 6.18 μmol/h; $P < .01$ and $P < .05$; oxypurinol: period 1, 46.75 ± 10.47 μmol/h; period 2, 30.18 ± 6.05 μmol/h; period 3, 21.74 ± 3.66 μmol/h, $P < .05$ and $P < .01$). In contrast, the urinary excretion of hypoxanthine did not change significantly (period 1, 8.95 ± 4.36 μmol/h; period 2, 8.43 ± 3.34 μmol/h; period 3, 8.09 ± 3.66 μmol/h; NS). Saline infusion caused no significant changes in the urinary excretions of purine bases and oxypurinol (data not shown), as has been reported previously.[5]

Effect of Angiotensin II Infusion on the Fractional Clearances of Purine Bases, Oxypurinol, and Sodium After Taking Allopurinol

As shown in Table 2, angiotensin II infusion caused significant decreases in the fractional clearances of uric acid, xanthine, and oxypurinol (uric acid: period 1, 9.3 ± 1.5; period 2, 6.1 ± 1.0; period 3, 4.5 ± 1.0, $P < .01$; xanthine: period 1, 71.8 ± 8.1; period 2, 49.0 ± 8.7; period 3, 50.6 ± 13.2, $P < .01$; oxypurinol: period 1, 22.2 ± 2.2; period 2, 15.0 ± 2.1; period 3, 11.8 ± 1.6, $P < .01$, respectively). In contrast, the fractional clearance of hypoxanthine was not changed after angiotensin II infusion (period 1, 66.6 ± 22.4; period 2, 76.4 ± 17.3; period 3, 77.4 ± 19.4; NS); however, the fractional excretion of sodium (FENa) decreased significantly during the time of infusion (period 1, 1.04 ± 0.08; period 2, 0.49 ± 0.06; period 3, 0.31 ± 0.12; $P < .01$, respectively). Saline infusion caused no significant changes in the fractional clearances of

Table 2. Effects of Angiotensin II Infusion on the Plasma Concentrations, Urinary Excretion, and Fractional Clearances of Purine Bases and Oxypurinol After Taking Allopurinol

	Period 1	Period 2	Period 3
Plasma concentration			
Uric acid (mmol/L)	0.29 ± 0.05	0.29 ± 0.05	0.29 ± 0.05
Hypoxanthine (μmol/L)	2.41 ± 1.24	2.15 ± 1.67	1.96 ± 1.27
Xanthine (μmol/L)	4.76 ± 2.17	4.30 ± 2.01	3.82 ± 1.54
Oxypurinol (μmol/L)	30.74 ± 1.92	29.95 ± 3.91	28.61 ± 3.97
Urinary excretion			
Uric acid (mmol/h)	0.16 ± 0.03	0.10 ± 0.02*	0.07 ± 0.02*
Hypoxanthine (μmol/h)	8.95 ± 4.36	8.43 ± 3.34	8.09 ± 3.66
Xanthine (μmol/h)	19.99 ± 8.78	12.65 ± 7.22*	11.38 ± 6.18†
Oxypurinol (μmol/h)	46.75 ± 10.47	30.18 ± 6.05†	21.74 ± 3.66*
Fractional clearance			
Uric acid (%)	9.3 ± 1.5	6.1 ± 1.0	4.5 ± 1.0*
Hypoxanthine (%)	66.6 ± 22.4	76.4 ± 17.3	77.4 ± 19.4
Xanthine (%)	71.8 ± 8.1	49.0 ± 8.7*	50.6 ± 13.2
Oxypurinol (%)	22.2 ± 2.2	15.0 ± 2.1*	11.8 ± 1.6*

*$P < .01$.

†$P < .05$.

purine bases and oxypurinol (data not shown), or FENa, as has been reported previously.[5]

DISCUSSION

Several epidemiologic studies have demonstrated a positive relationship between hypertension and hyperuricemia[1-3]; however, the underlying cause remains poorly understood. Decreased uric acid clearance in hypertensive patients has also been found,[2,3] and it is assumed that increased sympathetic nerve tone, and/or the renin-angiotensin-aldosterone and kinin-kallikrein systems may be involved in the development of hypertension. Further, a correlation between serum uric acid and the renin-angiotensin system in hypertension has been described.[9] Accordingly, in a previous study we investigated the effect of norepinephrine on the urinary excretion of oxypurines and oxypurinol, as well as uric acid, and found that the urinary excretion and fractional clearance of uric acid, xanthine, and oxypurinol were significantly decreased by norepinephrine infusion.[7]

The effect of angiotensin II infusion on uric acid clearance has been tested in healthy male subjects by Ferris and Gordon,[10] as well as in pregnant and nonpregnant women by Brown et al.[11] Ferris and Gordon[10] suggested that the decreased renal clearance of uric acid by angiotensin II infusion was due to a reduction in filtered urate and enhanced tubular urate reabsorption. However, the effect of angiotensin II on the urinary excretion and fractional clearance of oxypurines and oxypurinol has not been previously reported. The present study results confirmed those of Ferris and Gordon[10] which showed that angiotensin II infusion decreased the renal clearance and urinary excretion of uric acid. In addition, the present results suggested that angiotensin II has an effect on the common renal transport pathway for uric acid, xanthine, and oxypurinol, resulting in a decrease in the fractional clearance and urinary excretion of these substances. Further, a cause-effect relationship between an enhanced renin-angiotensin system (hypertension) and hyperuricemia is suggested, although the angiotensin II infusion may have decreased the effective renal plasma flow, leading to a decrease in uric acid excretion, as suggested by Ferris and Gordon.[10]

Renal blood flow was not measured in the present study; however, creatinine clearance was not changed, although a significant decrease in FENa was observed during angiotensin II infusion. Johnson and Malvin[12] described a more direct antinatriuretic action of angiotensin II on renal tubules, and, further, a cotransport of uric acid and sodium in proximal renal tubules has been suggested.[13,14] Therefore, the direct effect of angiotensin II on the common renal transport pathway of uric acid, xanthine, and oxypurinol seems more likely to cause a decrease in uric acid excretion. Regardless of the mechanism of angiotensin II that induces a decrease in the urinary excretion of uric acid, xanthine, and oxypurinol, exaggerated hypouricemic effect of allopurinol in hypertensive gout patients with an enhanced renin-angiotensin system may be expected, although an enhanced renin-angiotensin system is unlikely to result in very high plasma oxypurinol levels.

In conclusion, the results of the present study suggested for the first time that angiotensin II decreases the fractional clearance of uric acid, xanthine, and oxypurinol through direct tubular transport and/or hemodynamic change. In addition, it is suggested that the hypouricemic effect of allopurinol in hypertensive gout patients with an enhanced renin-angiotensin system may be exaggerated, since the observed plasma concentrations of angiotensin II in the present study were nearly within the physiological range. Further studies are necessary to clarify the mechanism that causes a decrease in the fractional clearance of uric acid, xanthine, and oxypurinol by angiotensin II.

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