Onpi-tô administration increases renal function in rats with renal failure

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Abstract

Onpi-tô extract and adenine were given orally at the same time to investigate their effects on renal function. Glomerular filtration rate, renal plasma flow and renal blood flow were markedly and significantly increased in rats given Onpi-tô at a dose of 40 mg or 80 mg/rat/day for 12 or 24 days, and a similar, significant increase was also found in rats given a dose of 80 mg for 36 days. The filtration fraction value was significantly decreased in rats given 80 mg of Onpi-tô for 24 days and 40 mg or 80 mg for 36 days.

Key words Onpi-tô, glomerular filtration rate, renal plasma flow, renal blood flow, renal failure, rat.

Abbreviation Onpi-tô (Wen-Pi-Tang), 温脾湯.

Introduction

We have previously reported that our method for preparing model rats with renal failure allows easy preparation of rats with mild, moderate or severe renal failure according to the period of adenine administration, the animals exhibiting azotemia, increased levels of uremic toxins such as methylguanidine and guanidinosuccinic acid, decreased renal function, and hormonal abnormalities.[1-5] When we investigated the action of rhubarb-containing prescriptions in Chinese medicine and those used for renal diseases, we found that Onpi-tô (Wen-Pi-Tang) produced a very potent improvement of nitrogen metabolism in the living body.[6-9] These actions were also observed when Onpi-tô was orally administered to rats on consecutive days in combination with simultaneous adenine administration, or when given to rats in which renal failure had been induced by a preliminary diet. Treatment using rhubarb-containing prescriptions, including Onpi-tô, has been tried on a clinical basis, and its usefulness as a drug therapy is now becoming established.[10,11] In the present study, in order to get further insight into the action of this prescription on the kidney, we investigated its effects on various parameters of renal function.

Materials and Methods

Animals and treatment : Male rats of the LWH : Wistar strain with a body weight of 200－210 g, were placed in metabolic cages and kept at a temperature of 23±1℃ under a 12-hr dark-light cycle. They were allowed an adaptation period of several days, during which they were fed on a commercial feed (type CE-2, CLEA Japan Inc., Tokyo, Japan). They were then fed *ad libitum* orally for 36 days as drinking water, while control rats received tap-water. On the 12th, 24th animals.[1-5] During the adenine-feeding period, an aqueous solution of Onpi-tô was administered orally for 36 days as drinking water, while control rats received tap-water. On the 12th, 24th and 36th days of the experimental period, renal function tests were performed. Throughout the

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experimental period, there were no statistically significant differences between the control and Onpi-tô-treated rats with regard to changes in body weight. The food intake of each rat was essentially proportional to weight change. In addition, daily intake of drinking water showed no appreciable changes in the three groups. No case of diarrheal symptoms was found. Six to seven rats were used for each experimental group. Values were expressed as means±S.E.

Onpi-tô : The Onpi-tô preparation was the same as that previously described.[6] The composition of Onpi-tô used in this experiment was as follows : 15 g of Rhei Rhizoma (*Rheum officinale* BAILLON), 3 g of Ginseng Radix (*Panax ginseng* C. A. MEYER), 5 g of Glycyrrhizae Radix (*Glycyrrhiza glabra* LINN. var. *glandulifera* REGEL et HERDER), 3 g of Zingiberis Rhizoma (*Zingiber officinale* ROSCOE) and 9 g of Aconiti Tuber (*Aconitum japonicum* THUNBERG). Ginseng Radix was a product of Korea, Aconiti Tuber was from Japan, and the other ingredients were from China. The extract was obtained as follows : the above-mentioned crude drugs were boiled gently in 1000 ml of water for 65 min and about 500 ml of decoction was obtained. The extract was then concentrated under reduced pressure to leave a brown residue with a yield of about 30%.

Examination of renal function : Glomerular filtration rate (GFR) and renal plasma flow (RPF) were measured by renal clearance test using a single intravenous administration of sodium thiosulfate or sodium *para*-aminohippurate, respectively, as an indicator.[12,13] At 25 min after intravenous administration of either of these agents, the bladder was reflexly emptied by having each rat inhale ether for 3—5 sec. The urine thus voided was discarded. During the next 30 min, the urine was collected, and collection was terminated after the bladder had again been emptied reflexly by ether inhalation. Blood samples were taken from conscious rats by heart puncture in the middle of the period used for the clearance test. Thiosulfate and *para*-aminohippurate were determined by titrimetry and colorimetry, respectively. Renal blood flow (RBF) was calculated on the basis of RPF and hematocrit (Ht) using the equation shown below. Ht was determined with a hematocrit measurement apparatus, model KH-120A (Kubota Co., Ltd., Tokyo, Japan).

$$RBF=\frac{RPF}{1-Ht}\ (ml/min)$$

Statistics : The significance of differences between the control and Onpi-tô-treated groups was tested using Student's t test. Differences at p values greater than 0.05 were considered to be statistically insignificant.

Results

As shown in Fig. 1, among rats given 12 days of the adenine diet, the GFR was 0.71 ml/min/kg in controls (2.15 ml/min/kg in normal rats), whereas the values were significantly higher by 66% and 256% in rats given 40 mg and 80 mg of Onpi-tô, respectively. Among rats given 24 days of the adenine diet, the corresponding values were 1.05 and 1.39 ml/min/kg in rats given 40 mg and 80 mg of Onpi-tô, respectively, which were higher than the control value at 12 days (0.71 ml/min/kg). Among rats given 36 days of the adenine

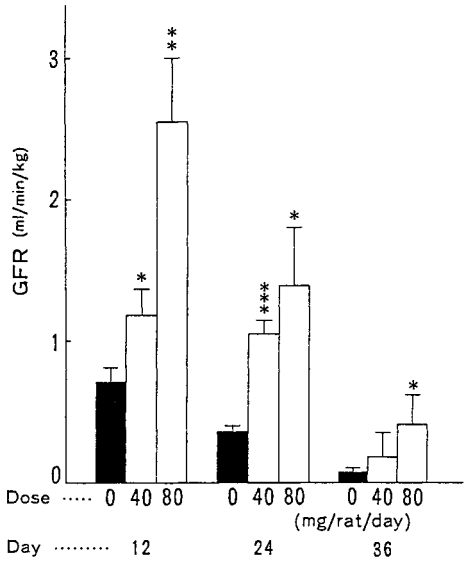

Fig. 1 Effect of Onpi-tô extract on glomerular filtration rate.
Significantly different from the control value, $*p<0.05$, $**p<0.01$, $***p<0.001$.

diet, control rats showed a very low GFR value corresponding to 3.5% of that for normal rats, whereas the value tended to be increased in rats given 40 mg of Onpi-tô, and was significantly increased in those given 80 mg, approximating the control value at 24 days. The RPF was 4.32 ml/min/kg in control rats given 12 days of the adenine diet (10.68 ml/min/kg in normal rats), whereas the corresponding values in rats given 40 mg and 80 mg of Onpi-tô were markedly increased to 7.83 and 14.25 ml/min/kg, respectively (Fig. 2). The latter value was 230% higher than the control value. At 24 days of the adenine diet, the RPF values were also markedly and significantly increased in rats given 40 mg and 80 mg of Onpi-tô, the values being comparable to or exceeding the control value at 12 days. In rats given 36 days of the adenine diet and 80 mg of Onpi-tô, the RPF value was also markedly increased, approximating the control value at 24 days. The RBF decreased gradually with the progress of renal failure, in a similar manner to the case of GFR or RPF. In rats given 12, 24 and 36 days of the adenine diet, the RBF values were 7.75, 1.83 and 0.12 ml/min/kg, respectively (19.73 ml/min/kg in normal rats). On the other hand, in rats given both 12 and 24 days of Onpi-tô administration, the RBF value was increased significant-

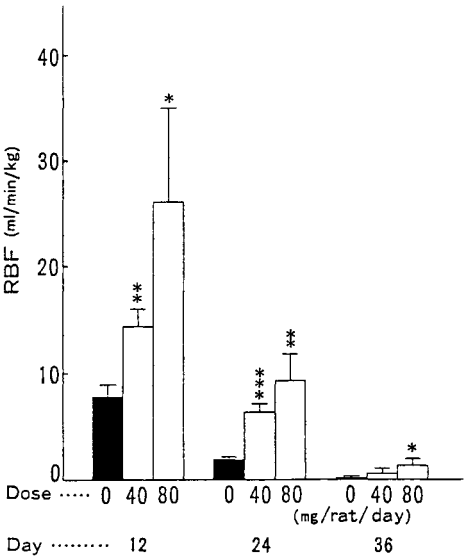

Fig. 3 Effect of Onpi-tô extract on renal blood flow.
Significantly different from the control value, $*p<0.05$, $**p<0.01$, $***p<0.001$.

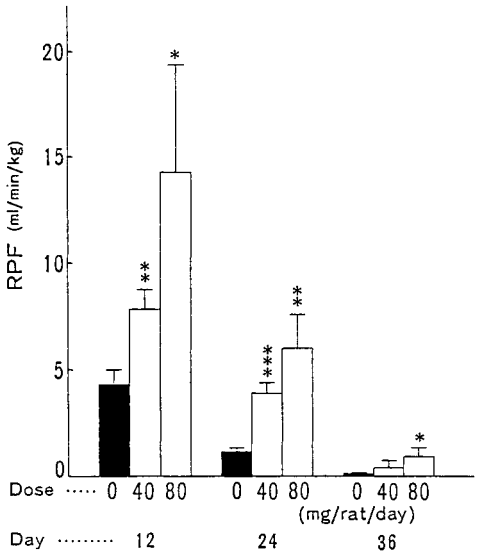

Fig. 2 Effect of Onpi-tô extract on renal plasma flow.
Significantly different from the control value, $*p<0.05$, $**p<0.01$, $***p<0.001$.

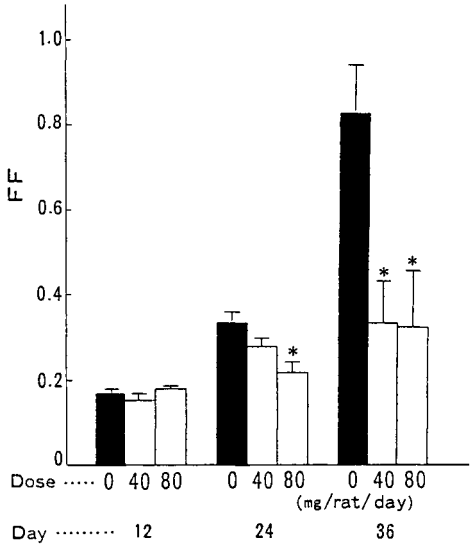

Fig. 4 Effect of Onpi-tô extract on filtration fraction.
Significantly different from the control value, $*p<0.01$.

ly ; in particular, the values were 237% and 407% higher in rats given 80 mg of Onpi-tô for 12 and 24 days, respectively, than in control rats. Rats given 80 mg of Onpi-tô for 36 days also showed a significant increase in RBF, being comparable to the value in control rats at 24 days (Fig. 3). As shown in Fig. 4, there was no significant difference in the filtration fraction (FF) value calculated from the GFR and RPF between the control group (0.168), the 40 mg treatment group (0.151) and the 80 mg treatment group (0.178) at 12 days. In contrast, at 24 days, the value was significantly decreased in rats given 80 mg of Onpi-tô, and there was a tendency toward a decreased value in rats given 40 mg of the preparation, in comparison with the control rats. At 36 days, the value was significantly decreased in rats given both 40 mg and 80 mg of Onpi-tô.

Discussion

Chronic renal failure is characterized by various clinical symptoms generically known as uremia, that parallel the deterioration of renal function. Under these conditions, uremic toxins, which are thought to induce uremia, are retained in the body because of a decrease in their excretion from the kidneys resulting from impaired renal function.[14] In a previous study, by determining renal function in rats administered adenine, we found that GFR began to decrease significantly at 6 days of adenine feeding, and RPF and RBF at 12 days, and pointed out that these parameters of renal function decreased gradually with the increased retention of uremic toxins in the body.[2,3] In the present experiment, we confirmed that these parameters markedly decreased as the period of adenine administration lengthened. On the other hand, in rats given the adenine diet together with oral Onpi-tô administration, there was a suppressive effect on the decrease in renal function. This effect was observed in both groups given 40 mg and 80 mg of Onpi-tô at 12 and 24 days. A dose of 80 mg was associated with a greater facilitatory action on renal function than was the case for a 40-mg dose. However, the effect of 40 mg Onpi-tô treatment on renal function was only a slight increase in the group of rats with severe renal impairment induced by 36 days of adenine ingestion. These findings seem to indicate that rats with renal failure have a reversible alteration in renal vascularization that is modulated, in part, by treatment with Onpi-tô extract. Further studies are therefore in progress to clarify the morphological changes. Onpi-tô is a prescription composed of Rhei Rhizoma (classified as a "cold" drug in Chinese medicine) as the main ingredient, together with Aconiti Tuber, Ginseng Radix, Glycyrrhizae Radix and Zingiberis Rhizoma. Of these components, Rhei Rhizoma alone does not exert any of the renal function-improving actions of Onpi-tô (unpublished data). The activation of renal function by Onpi-tô is thus probably based on the action of its warm-drug components such as Aconiti Tuber, Ginseng Radix and Zingiberis Rhizoma, rather than Rhei Rhizoma. These warm drugs are considered to neutralize the "cold" property of Rhei Rhizoma by exerting a general body-warming action (facilitating blood circulation), thus maintaining the homeostasis of the kidney. In support of this speculation, Ginseng Radix has been found to produce a significant increase in renal tissue blood flow.[15] On the other hand, FF is used as an index of the GFR/RPF ratio. In particular, great importance is attached to the pressure difference between the afferent and efferent arterioles, as a factor that exerts a strong influence on the GFR. It has been reported that angiotensin-converting enzyme inhibitors, which are widely used in clinical diagnosis, cause a decrease in angiotensin II, resulting in decreased GFR due to a greater expansion of the efferent arterioles than that of the afferent arterioles. Since GFR was increased and FF decreased after Onpi-tô administration, it seems that the increase in the afferent-efferent arteriole pressure difference was not proportional to the increase in blood flow. At present, a conservative therapy which does not cause a decrease in GFR, but reduces the afferent-efferent arteriole pressure difference (*i.e.*, a decrease in FF), is considered to be desirable for the improvement of intraglomerular hypertension. Further future

studies will be required in order to obtain more detailed information. In this regard, Onpi-tô is an interesting prescription. We have previously reported the effects of intraperitoneal administration of Onpi-tô after induction of renal failure.[16] The effects of oral administration under such conditions are now being investigated.

和文抄録

温脾湯エキスをアデニン投与と同時に経口投与し，腎機能に及ぼす影響を検討した。糸球体濾過値(GFR)，腎血漿流量 (RPF)，腎血流量 (RBF) はいずれも温脾湯 40 mg，80 mg/rat/day 12日間並びに24日間投与したラットにおいて著しく有意に増加し，36日間投与したラットの 80 mg 投与群においても有意な増加作用が認められた。また濾過率(FF) も24日間投与ラットの温脾湯 80 mg群，36日間投与ラットの 40 mg，80 mg 投与群において有意に低下していた。

References

1) Yokozawa, T., Zheng, P.D., Oura, H. and Koizumi, F.: Animal model of adenine-induced chronic renal failure in rats. *Nephron* **44**, 230-234, 1986.
2) Yokozawa, T., Chung, H.Y. and Oura, H.: Urinary constituents and renal function in rats administered with adenine. *Jap. J. Nephrol.* **29**, 1129-1135, 1987.
3) Yokozawa, T. and Oura, H.: Distribution of guanidino compounds in rats with chronic renal failure induced by adenine. *Jap. J. Nephrol.* **29**, 1137-1143, 1987.
4) Yokozawa, T., Oura, H. and Nakada, T.: Blood flow in renal tissue, blood pressure, and blood hormone levels in rats with adenine-induced renal failure. *Jap. J. Nephrol.* **29**, 1145-1151, 1987.
5) Yokozawa, T., Mo, Z.L. and Oura, H.: Comparison of toxic effects of methylguanidine, guanidinosuccinic acid and creatinine in rats with adenine-induced chronic renal failure. *Nephron*, in press.
6) Oura, H., Zheng, P.D. and Yokozawa, T.: Effect of Onpi-tô in rats with chronic renal failure. *J. Med. Pharm. Soc. WAKAN-YAKU* **1**, 209-217, 1984.
7) Zheng, P.D., Yokozawa, T. and Oura, H.: Effect of Onpi-tô in adenine-induced chronic renal failure rats. *J. Med. Pharm. Soc. WAKAN-YAKU* **3**, 83-88, 1986.
8) Yokozawa, T., Zheng, P.D., Mo, Z.L. and Oura, H.: The effect of Onpi-tô on urinary excretion of methylguanidine in rats with chronic renal failure. *J. Med. Pharm. Soc. WAKAN-YAKU* **3**, 198-201, 1986.
9) Yokozawa, T., Mo, Z.L., Wu, X.Q. and Oura, H.: The actions of various prescriptions on rats with experimental renal failure. *J. Med. Pharm. Soc. WAKAN-YAKU* **4**, 164-171, 1987.
10) Mitsuma, T., Terasawa, K., Yokozawa, T. and Oura, H.: Rhubarb therapy in patients with chronic renal failure (Part 1). *J. Med. Pharm. Soc. WAKAN-YAKU* **1**, 266-278,1984.
11) Mitsuma, T., Yokozawa, T., Oura, H. and Terasawa, K.: Rhubarb therapy in patients with chronic renal failure (Part 2). *Jap. J. Nephrol.* **29**, 195-207, 1987.
12) Brun, C.: Thiosulfate determination in kidney function tests. *J. Lab. Clin. Med.* **35**, 152-154, 1950.
13) Brun, C.: A rapid method for the determination of para-aminohippuric acid in kidney function tests. *J. Lab. Clin. Med.* **37**, 955-958, 1952.
14) Mujais, S.K., Sabatini, S. and Kurtzman, N.A.: Pathophysiology of the uremic syndrome. In "The Kidney" (Eds. by B.M. Brenner and F.C. Rector), W.B. Saunders Company, Philadelphia, pp. 1587-1630, 1986.
15) Yokozawa, T., Zheng, P.D., Chung, H.Y., Fukumoto, J. and Oura, H.: Effect of red ginseng powder in rats with chronic renal failure. *J. Med. Pharm. Soc. WAKAN-YAKU* **3**, 136-142, 1986.
16) Yokozawa, T., Orihashi, M., Chung, H.Y., Wu, X.Q. and Oura, H.: Effects of Onpi-tô extract on renal function in rats with renal failure. *J. Med. Pharm. Soc. WAKAN-YAKU* **5**, 98-103, 1988.