Effect of Moderately Increased Intrapelvic Pressure on Renal Tissue Pressure and Vasopressin Release in Rabbits

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Electrical stimulation of afferent renal nerves and activation of intrarenal receptors increases plasma vasopressin concentration, but the role of afferent renal nerves in the control of vasopressin secretion is not clear. Recently, we reported that activation of renal mechanoreceptors stimulates the release of vasopressin. However, intrapelvic pressure was increased to 50 mmHg, and this increase is above the normal physiological range. Therefore, in the present study, we investigated the effect of moderately increased intrapelvic pressure on plasma vasopressin concentration in anesthetized rabbits. First, we measured renal tissue pressure while intrapelvic pressure was increased stepwise in 10-mmHg increments. Basal renal tissue pressure was 17 ± 2 mmHg. Renal tissue pressure increased only when intrapelvic pressure was higher than the basal tissue pressure of each animal. Usually, increases in intrapelvic pressure less than 20 mmHg did not increase renal tissue pressure. This finding suggests that only increases in intrapelvic pressure more than 20 mmHg can activate renal mechanoreceptors. Based on this finding, the effects of moderate increases in intrapelvic pressure (15 and 30 mmHg) were studied. With a 15-mmHg increase in intrapelvic pressure, plasma vasopressin concentration did not change significantly. However, when intrapelvic pressure was increased to 30 mmHg, plasma vasopressin concentration increased from 5.6 ± 1.4 to 9.5 ± 2.8 pg/ml at 5 min ($p < 0.05$) and to 8.8 ± 2.0 pg/ml at 10 min ($p < 0.05$). Plasma renin activity and mean arterial pressure also increased when intrapelvic pressure was increased to 30 mmHg. We conclude that moderate increases in intrapelvic pressure stimulate vasopressin secretion. This provides further evidence that the kidneys participate in the physiological control of vasopressin release by way of afferent renal nerves. (*Hypertens Res* 1995; 18: 197-202)

Key Words: afferent renal nerves, renal mechanoreceptors, renal tissue pressure, plasma vasopressin concentration, rabbit

Electrical stimulation of afferent renal nerves and activation of intrarenal receptors have been reported to increase plasma vasopressin (AVP) concentration (1-7) and the activity of the neurosecretory cells in the supraoptic (8, 9) and paraventricular (10) nuclei of the hypothalamus. These observations suggest that afferent renal nerves may participate in the control of the release of AVP.

Intrarenal receptors of afferent renal nerves have been classified as mechanoreceptors and chemoreceptors. Renal mechanoreceptors are activated when intrarenal pressure is increased by maneuvers including increased renal perfusion pressure, renal vein occlusion, and increased ureteral pressure (11, 12). There appear to be two types of renal chemoreceptors. R1 chemoreceptors are activated by renal ischemia, while R2 chemoreceptors are activated by increases in the osmolality or ionic concentration in

the renal pelvis (11, 12).

In a previous study, we observed that activation of renal mechanoreceptors increased the release of AVP in anesthetized rabbits and that prior renal denervation attenuated this effect (5). This evidence supports the hypothesis that the kidneys participate in the control of AVP secretion by way of afferent renal nerves. However, intrapelvic pressure was increased to 50 mmHg in that study, and this increase is above the normal physiological range. Therefore, in the present study, we investigated the effect of smaller increases in intrapelvic pressure on plasma AVP concentration. We also investigated the relationship between renal tissue pressure and intrapelvic pressure, because activation of renal mechanoreceptors presumably occurs via increased renal tissue pressure.

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Fig. 1. Schematic drawing of the method used to increase the intrapelvic pressure.

Materials and Methods

Animal Preparation

Male New Zealand white rabbits weighing 2.5–3.4 kg were used. The rabbits were premedicated with acepromazine maleate (1 mg/kg, i.m.) and anesthetized with pentobarbital sodium (20–30 mg/kg, i.v.). Catheters were placed in the right femoral artery for monitoring arterial blood pressure and for sampling blood, and in the right femoral vein for injection of saline or anesthetic. Isotonic saline was infused at 0.1–0.2 ml/min via the femoral vein catheter to prevent dehydration during surgery. The saline infusion was stopped 30 min before the start of the experiment. Each rabbit was placed on its right side. The left kidney and left ureter were exposed retroperitoneally through a paravertebral incision. The left ureter was cannulated below the ureteropelvic junction with a double-lumen catheter (Fig. 1). The outer sheath of this catheter (branch 1) consisted of a PE-160 catheter (Clay Adams, Parsippany, NJ) through which a PE-10 catheter (Clay Adams) was passed into the renal pelvis (branch 2). Branch 2 was connected to a Statham pressure transducer for monitoring intrapelvic pressure. The catheter tip was advanced to just above the ureteropelvic junction and fixed in place with silk threads. The distal end of the ureteral catheter was connected to a saline column via a three-way stopcock, and urine flowed freely. Special care was taken not to touch the renal hilus and to leave the renal nerves intact. In some rabbits, a 25 gauge needle connected to polyethylene tubing was inserted to the renal cortex for monitoring renal tis-

sue pressure using a Statham pressure transducer. Arterial blood pressure and heart rate were continuously recorded throughout each experiment with a Statham pressure transducer and a Grass polygraph (Grass Instruments, Quincy, MA). An equilibration period of at least 60 min was allowed before the start of an experiment.

Experimental Protocols

Three series of experiments were performed.

1. Effect of increased pelvic pressure on renal tissue pressure (n=5)

After hemodynamics and urine flow had stabilized, basal intrarenal tissue pressure was determined. Intrapelvic pressure was then increased stepwise in 10 mmHg increments to a maximum of 50 mmHg by raising the saline column. At each increment of intrapelvic pressure, intrarenal tissue pressure was measured. The position of the tip of the needle was determined when the kidney was excised at the end of each experiment.

Based on the results of these measurements (see below), we chose 15 and 30 mmHg increments of intrapelvic pressure for the following protocols.

2. Effect of increased pelvic pressure (15 mmHg) on plasma AVP concentration (n=5)

After blood pressure and heart rate had stabilized, a 3.5 ml control arterial blood sample was collected. Intrapelvic pressure was then increased for 10 min by connecting the ureteral catheter to the saline column in which the fluid level had been adjusted to 15 mmHg. Additional blood samples were collected 5, 10, and 30 min after pelvic pressure was increased. From each sample, a 2.7-ml aliquot of blood was placed immediately in a chilled tube containing 0.3 ml 0.3 M EDTA. Plasma was separated by centrifugation at 4°C and was frozen until it was analyzed for plasma renin activity (PRA) and plasma AVP concentration. The remaining 0.8-ml aliquot of blood was placed in a tube containing heparin for the determination of plasma osmolality. Blood samples were replaced with an equal volume of sterile isotonic saline.

At the end of experiment, the left kidney was removed together with the ureteral catheter. The kidneys were opened to confirm that the catheter tip was within the renal pelvis and that there was no damage to the mucosa membrane.

3. Effect of increased pelvic pressure (30 mmHg) on plasma AVP concentration (n=7)

For this series of experiments, the fluid level in a saline column was adjusted to 30 mmHg. Ureteral cannulation, increasing pelvic pressure, and blood sampling were performed as described in protocol 2.

Analytical Methods

Plasma AVP concentration was determined by radioimmunoassay after extraction with bentonite (13). PRA was measured using a radioimmunoassay for angiotensin I and expressed as nanograms angiotensin I generated per ml plasma during a two-hour incubation at 37°C and pH 6.5 (14). Plasma osmolality was determined by freezing-point de-

Fig. 2. Relationship between intrapelvic pressure (PP) and renal tissue pressure (RTP). Curvilinear regression analysis was performed using the polynomial method. $y = 16.561 + 0.048x + 0.006x^2$, $r = 0.9$, $p < 0.01$.

pression (Advanced Model 3W).

Statistical Analysis

All results are expressed as the means $\pm$ SE. Multiple comparisons were performed subsequent to one-way analysis of variance for repeated measures by the Scheffe *F*-test (15). Analysis of plasma AVP concentration was made after logarithmic transformation of the data. Curvilinear regression analysis was performed using polynomial method. $p < 0.05$ was used as the level of statistical significance.

Results

1. Effect of Increased Intrapelvic Pressure on Renal Tissue Pressure ($n = 5$)

Figure 2 shows the relationship between renal tissue pressure and intrapelvic pressure. Basal renal tissue pressure was 17 ± 2 mmHg. Raising intrapelvic pressure increased renal tissue pressure only when intrapelvic pressure was higher than the basal tissue pressure of each animal. Usually, increases in intrapelvic pressure of less than 20 mmHg did not increase renal tissue pressure. Renal tissue pressure increased in proportion to intrapelvic pressure ($y = 16.561 + 0.048x + 0.006x^2$, $r = 0.9$, $p < 0.01$). After the period of increased pelvic pressure, renal tissue pressure returned promptly to its basal level.

2. Effect of Increased Pelvic Pressure (15 mmHg) on Plasma AVP concentration ($n = 5$)

In this group, we investigated the effect of a 15-mmHg increase in pelvic pressure, which did not affect renal tissue pressure, on plasma AVP concentration. Basal plasma AVP concentration was 5.6 ± 0.5 pg/ml. Plasma AVP concentration was 6.8 ± 1.3 pg/ml at 5 min and 6.6 ± 1.1 pg/ml at 10 min after pelvic pressure was increased to 15 mmHg (Fig. 3). These changes in plasma AVP concentration were not statistically significant. PRA tended to increase from 8.2 ± 1.6 to 10.1 ± 2.2 ng/ml/2 h at 5 min and

to 11.7 ± 3.0 ng/ml/2 h at 10 min (Fig. 3), but these changes were not statistically significant. The resting basal values of mean arterial pressure (MAP) and heart rate (HR) were 73 ± 2 mmHg and 201 ± 12 beats/min, respectively. Raising intrapelvic pressure to 15 mmHg did not change MAP and HR (Fig. 3). There was no change in plasma osmolality (Table 1).

3. Effect of Increased Pelvic Pressure (30 mmHg) on Plasma AVP Concentration ($n = 7$)

The renal tissue pressure measurements indicated that a 30-mmHg increase in intrapelvic pressure increased renal tissue pressure by approximately 6 mmHg (Fig. 2). When pelvic pressure was increased to 30 mmHg, plasma AVP concentration increased from 5.6 ± 1.4 to 9.5 ± 2.8 pg/ml at 5 min ($p < 0.05$) and to 8.8 ± 2.0 pg/ml at 10 min ($p < 0.05$) (Fig. 4). PRA increased from 11.1 ± 2.6 to 19.9 ± 3.4 ng/ml/2 h at 5 min ($p < 0.01$) and to 20.1 ± 3.7 ng/ml/2 h at 10 min ($p < 0.01$) (Fig. 4). MAP increased from 79 ± 3 to 81 ± 3 mmHg at 5 min ($p < 0.01$) and to 81 ± 3 mmHg at 10 min ($p < 0.05$) (Fig. 4). Plasma AVP concentration, PRA and MAP all returned to their basal levels 20 min after intrapelvic pressure was decreased. HR gradually increased in some rabbits but the changes were not statistically significant. Plasma osmolality did not change significantly (Table 1).

Discussion

Several studies have provided evidence that afferent renal nerves contribute to the control of vasopressin release and the regulation of arterial blood pressure and renal function (1-7, 12). In a previous study, we examined the effects of activation of renal mechanoreceptors on the release of vasopressin (5). Plasma vasopressin concentration increased three-fold when intrapelvic pressure was increased to 50 mmHg. Prior denervation of the kidneys blocked the increase in plasma vasopressin concentration produced by activation of mechanoreceptors. These results support the hypothesis that the kidneys may participate in the control of the release of vasopressin by way of afferent renal nerves. However, because a 50-mmHg increase in intrapelvic pressure is not physiological, these results do not permit conclusions concerning the physiological role of renal receptors in the control of vasopressin release. Therefore, in the present study, we investigated the effects of smaller increases in intrapelvic pressure on plasma vasopressin concentration.

There have been several reports concerning the distribution of mechanoreceptors within the kidney. Astrom and Crafoord (16) observed an increase in the activity of afferent renal nerves when renal vein pressure was elevated in rats. They concluded that an adequate stimulus for activation of mechanoreceptors is increased intrarenal pressure and perhaps also distension of the intrarenal veins. They also observed increased afferent renal nerve activity after elevation of renal perfusion pressure and mechanical pressure applied to the hilus area in

Table 1. Effect of Increased Intrapelvic Pressure on Plasma Osmolality

		Plasma osmolality (mOsm/kgH ₂ O)			
		0 min	5 min	10 min	30 min
15 mmHg	(n=5)	292±1	294±3	294±2	295±1
30 mmHg	(n=5)	294±4	293±3	291±2	292±1

Values are means±SE.

Fig. 3. Effects of increased intrapelvic pressure (15 mmHg) on plasma vasopressin concentration, plasma renin activity, mean arterial pressure and heart rate. Values are means±SE, n=5.

cats (17). Nijima (18) reported that mechanoreceptors responded with an increased afferent discharge rate when the arterial pressure in the kidney of rabbits increased, and that mechanoreceptors did not respond to an increase in venous pressure. He suggested that mechanoreceptors are present around the arterial walls in the kidney. He also observed that the receptors are also located in the parenchyma and in the wall of the renal pelvis (19). Recently Kopp *et al.* (20) found that intrapelvic administration of lidocaine did not affect the afferent renal nerve response to elevated renal venous pressure in rats, but abolished the increase in afferent renal nerve activity elicited by elevated ureteral pressure. They suggested that the mechanoreceptors activated by elevated renal venous pressure are located in an

Fig. 4. Effects of increased intrapelvic pressure (30 mmHg) on plasma vasopressin concentration, plasma renin activity, mean arterial pressure and heart rate. Values are means±SE, n=7. *p<0.05; **p<0.01 vs. 0 min.

anatomically different area than those activated by elevated pelvic pressure. The precise localization of mechanoreceptors within the kidney remains to be elucidated. Therefore, the surest method of activating renal mechanoreceptors at the present time is to increase intrarenal tissue pressure.

Activation of renal mechanoreceptors apparently occurs by way of increased renal tissue pressure. Therefore, in the present study, we examined the relationship between intrapelvic pressure and renal tissue pressure. Renal tissue pressure was 17 ± 2 mmHg under basal conditions. Yoshitoshi *et al.* (21) have reported a similar value (18.4 ± 3.1 mmHg) in rabbits. Renal tissue pressure increased only when intrapelvic pressure was higher than the basal tissue pressure of each animal. Usually, increasing in-

trapelvic pressure by more than 20 mmHg increased renal tissue pressure. These results indicate that intrapelvic pressure must be increased above 20 mmHg to activate renal mechanoreceptors. Based on the present results (Fig. 2), renal tissue pressure would increase by about 6 and 17 mmHg after intrapelvic pressure was increased to 30 and 50 mmHg, respectively. We chose increases of intrapelvic pressure that would either not increase renal tissue pressure (15 mmHg), or that would slightly increase tissue pressure (30 mmHg) to examine the effect of increased intrapelvic pressure on plasma vasopressin concentration. A 30 mmHg increase in intrapelvic pressure may be above the normal physiological range. However, Schweitzer (22) studied intrapelvic pressure in dogs with unilateral nephrectomy. Intrapelvic pressure was about 5 mmHg under basal conditions and increased up to 15 mmHg under diuretic conditions without ureteral obstruction. During chronic partial ureteral obstruction, intrapelvic pressure was about 25 mmHg under non-diuretic conditions and was elevated up to 45 mmHg under diuretic conditions. In humans with partial ureteral obstruction due to calculi, the basal intrapelvic pressure was reported to be 25 mmHg and intrapelvic pressure increased to more than 50 mmHg during flank pain (23). Thus, a 30 mmHg increase in pelvic pressure can easily occur during partial ureteral obstruction.

With a 15 mmHg increase in intrapelvic pressure that would not increase renal tissue pressure, plasma vasopressin concentration did not change significantly. However, plasma vasopressin concentration increased significantly after intrapelvic pressure was increased to 30-mmHg when renal tissue pressure would increase. These results thus confirm that activation of renal mechanoreceptors can stimulate the release of vasopressin.

Plasma renin activity increased significantly when intrapelvic pressure was increased to 30 mmHg. Stimulation of vasopressin release by increased intrapelvic pressure could conceivably have been mediated via the renin-angiotensin system because angiotensin II is known to stimulate vasopressin release (24). However, in our previous study (5), renal denervation almost completely blocked the increase in plasma vasopressin concentration without preventing the increase in plasma renin activity. These results therefore suggest that the increase in plasma vasopressin concentration caused by increasing intrapelvic pressure to 30 mmHg is mediated by way of afferent renal nerves rather than by way of the renin-angiotensin system.

The mechanism of the increase in plasma renin activity by increasing intrapelvic pressure was not investigated. One possibility is that it was mediated via macula densa mechanism (25), because glomerular filtration would have been markedly reduced so that the delivery of sodium chloride to the distal tubule would have been decreased. Another possibility is that it was mediated via increased prostaglandin synthesis, because increasing intrapelvic pressure is known to increase renal prostaglandin synthesis, and prostaglandins stimulate renin secre-

tion (25). Further studies are required to investigate these possibilities.

In the present study, blood pressure increased when intrapelvic pressure was increased to 30 mmHg. It is known that electrical stimulation of the renal nerves or activation of renal receptors increases blood pressure and heart rate (2-4, 6, 12). These increases have generally been attributed to changes in sympathetic neural activity. However, Caverson and Ciriello (1) have recently shown that increased plasma vasopressin concentration contributes to the pressor response. Thus, the change in blood pressure in the present study may have been due both to increases in autonomic neural activity and plasma vasopressin concentration.

In summary, renal tissue pressure increased only when intrapelvic pressure was higher than the basal tissue pressure. A 30-mmHg increase in intrapelvic pressure that would slightly increase renal tissue pressure increased plasma vasopressin concentration, while a 15 mmHg increase that would not increase renal tissue pressure did not increase plasma vasopressin concentration. These results thus demonstrate that activation of renal mechanoreceptors by moderate increases in intrapelvic pressure increases the release of vasopressin.

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