INTRARENAL ROLE OF RENIN-ANGIOTENSIN SYSTEM IN THE REGULATION OF RENAL HEMODYNAMICS

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Abstract—A reduction of renal arterial pressure in mongrel dogs to 70 mmHg resulted in marked increases in plasma renin activity and plasma levels of angiotensin I (AI) and angiotensin II (AII). Production of renin and AI but not AII in the kidney was observed. A reduction of renal arterial pressure also resulted in a redistribution of blood flow from the outer to inner cortex. An arterial infusion of AII (200 ng/min), however, failed to affect the intrarenal distribution of the blood flow. An intrarenal infusion of AII rather restored the normal pattern of the distribution of intrarenal blood flow altered by the pressure reduction. These results indicate that the renin-angiotensin system is probably not involved in the control of renal hemodynamics through the intrarenal formation of AII, and that the intrarenal hemodynamic changes caused by pressure reduction is due to the intrinsic difference in myogenic force in different cortical zones.

The possibility that the renin-angiotensin system is involved in the control of arterioglomerular balance has been reported (1-4). Schnermann et al. (4) postulated the presence of a glomerulo-tubulo-feedback mechanism presumably involving the renin-angiotensin system, the postulation was based on the observation that sodium concentrations in the distal tubule were inversely related to glomerular filtration rate (GFR) in rats kept on a low salt diet. Numerous investigators, however, reported that the intrarenal conversion from angiotensin I (AI) to angiotensin II (AII) is negligible (5-7). Moreover, little is known of the intrarenal formation of AI by renin.

In view of the lack of evidence for the local formation of AII, the present experiments were performed. In the anesthetized dog, the renal arterial pressure was reduced to the lower range of autoregulation. Following pressure reduction, the activity of renin in the systemic arterial and renal venous plasma, and plasma concentration of AI and AII were estimated concomitantly by radioimmunoassay (8, 9). The amount of renin released and the rate of AI and AII formation within the kidney were calculated. Additionally, correlations between intrarenal hemodynamics and the rates of renin, AI and AII formation were examined.

MATERIALS AND METHODS

Experiments were performed using 15 mongrel dogs weighing 12-19 kg which had been maintained on standard laboratory chow. They were anesthetized with sodium pentobarbital (30 mg/kg, i.v.) and a tracheostomy was performed. Catheters were placed in the right
brachial vein and artery for infusions and systemic blood sampling. The left kidney was exposed through a retroperitoneal flank incision. The kidney was denervated by dissecting all visible nerve fibers and the tissue connecting the renal hilum cephalad to the renal artery. An adjustable clamp was placed on the aorta proximal to the left renal artery and distal to the right renal artery. Renal blood flow (RBF) was measured by an electromagnetic flowmeter (Nihonkoden, Tokyo; model MF-25). Left renal arterial pressure was considered equal to the aortic pressure measured at the level of the left renal artery. Systemic arterial blood was collected from the right brachial artery and the renal venous blood was collected from a cannula introduced through the left spermatic or ovarian vein into the renal vein.

An intravenous infusion of 0.9% saline at the rate of 4.2 ml/min was started after anesthesia. A 23 gauge needle was introduced into the left renal artery proximal to the flow probe for intra-arterial administration of angiotensin II.

Distribution of cortical blood flow was determined with radioactive microspheres (3M Company, St. Paul, U.S.A.) by the technique described in a previous paper (10). The renal cortex was cut parallel to the surface into 4 zones of equal thicknesses. The cortical zones were numbered sequentially from the capsule to juxtamedullary cortex. The volume of each cortical zone was approximated by calculations based on the formula for an ellipsoid. Such a volume expressed as percent of total renal volume, was: zone 1, 27.0; zone 2, 21.9; zone 3, 17.3; and zone 4, 12.2.

Plasma renin activity (PRA), plasma concentration of Al and All were determined by radioimmunoassay (8, 9). All blood samples (5 ml) with 5 mg EDTA were centrifuged for 10 min at 4°C to obtain plasma. Dimercaprol and 8-hydroxyquinoline sulphate were added to inhibit angiotensinase and converting enzyme. Each sample was then divided. One aliquot was incubated at 37°C for 3 hours for renin assay, while the other served for assays of Al and All. Renin secretion rate (RSR) was calculated by multiplying the difference between renal venous and arterial plasma renin activities by the renal plasma flow. PRA was expressed as Al formed during a 3-hour incubation per ml of plasma. Plasma concentrations of Al and All were expressed as ng/ml and pg/ml, respectively.

Two series of experiments described below were performed.

A) Renin, Al and All concentrations in renal arterial and venous blood at normal and reduced renal arterial pressure: This experiment was carried out in 8 dogs. Control blood samples from the artery and renal vein were taken at the end of a 1-hour stabilization period. Renal arterial pressure was reduced to the lower limit of the autoregulatory range of RBF, i.e., 70 mmHg, and kept at this level by manipulation of the aortic clamp. The RBF decreased transiently and then recovered to the control value within a few minutes, at which time blood samples from the artery and renal vein were simultaneously withdrawn. During the pressure reduction, two more blood samples were withdrawn at 10 and 15 minutes after the aortic constriction. The constriction was subsequently released and blood samples were taken 20 and 60 min later.

B) Effects of All on intrarenal hemodynamics at normal and reduced renal arterial pressure: Nine dogs were divided into two subgroups. All (200 ng/min) was infused into the renal
artery at normal pressure in 5 dogs and at reduced pressure (75 mmHg) in another 4 dogs. In both groups, AII infusion resulted in an approximate 50 percent decrease in RBF. Gamma-labelled microspheres (diameter 15 μm, labelled either 85Sr, 51Cr or 141Ce) were injected before AII infusion and 10 min after the AII infusion or before the pressure reduction, 15 min after the pressure reduction, and 10 min after AII infusion following the pressure reduction.

Student's t-test was used for statistical determinations.

RESULTS

Effects of pressure reduction on renin secretion and plasma concentration of AI and AII

PRA in the systemic artery and renal vein and plasma concentrations of AI and AII are shown in Fig. 1. At normal blood pressure, PRA in the systemic arterial and renal

![Graph showing effects of renal arterial pressure reduction on renin secretion and plasma concentrations of AI and AII.](image_url)

**Fig. 1.** Effects of renal arterial pressure reduction on renal blood flow, PRA and plasma concentrations of AI and AII. Renal arterial pressure was reduced by means of a clamp placed on the aorta. Each point represents the mean of 8 experiments. Vertical line indicates the standard error of the mean.
venous blood, expressed as AI formed during a 3-hour incubation, was 18.8±8.0 and 26.5 ±13.6 ng/ml, respectively. Concentrations of AI in arterial and renal venous plasma were 0.8±0.4 and 1.0±0.5 ng/ml, respectively, and the concentrations of AII were 75±15 and 55±11 pg/ml, respectively. Concentrations of AI in arterial and renal venous plasma were approximately 4.3% of renin activity (amount of AI formed during a 3-hr incubation). The ratio of plasma concentration of AII to that of AI was 0.051 in the systemic arterial blood and 0.029 in the renal venous blood, and the difference between these two ratios was statistically significant (p<0.05).

A reduction of renal arterial pressure from 125 mmHg to about 70 mmHg resulted in an increase in PRA in both the systemic arterial and renal venous blood, despite a complete autoregulation of RBF. Time course of the changes in AI concentrations in both the arterial and renal venous blood was identical to that of changes in PRA in each blood sample. Plasma AII concentration increased slightly during the pressure reduction. The time course of changes in AII, however, was different from that of changes in AI; a parallel change was observed between concentrations of AII and AI only in the systemic arterial blood, but not in the renal venous blood. At 50 min after the release of aortic constriction, these three parameters in both the arterial and renal venous PRA and also renal hemodynamics recovered to the control value (Fig. 1).

Figure 2 shows the relation between PRA and plasma concentration of AI in the systemic arterial and renal venous blood. There was a significant linear relationship between these two parameters (r=0.842, p<0.001 for the systemic arterial blood; r=0.957, p<0.001 for the renal venous blood). For a given plasma renin activity, the concentration of AI in renal venous blood was not significantly different from that of AI in arterial blood. Figure 3
shows the relation between AI and All concentrations in the systemic arterial and renal venous blood. There was a significant linear relationship between AI and All concentrations (r=0.851, p<0.001 for the systemic arterial blood; r=0.898, p<0.001 for the renal venous blood). A significant difference (p<0.05) was found between the slopes of two regression lines, indicating that the relation between these two parameters is different in arterial and renal venous blood.

Figure 4A shows the distribution of intrarenal blood flow in control kidney and during the intrarenal-arterial infusion of All (Hypertensin-CIBA, CIBA Pharmaceutical, Summit, U.S.A.). Figure 4B shows the distribution of RBF at reduced pressure and during All infusion at reduced pressure. At normal pressure, the fraction of total RBF distributed to each zone was significantly different. The inner zones were characterized by a low distribution of blood flow. The reduction of renal arterial pressure from 125 mmHg to 70 mmHg resulted in a significant alteration in the distribution of blood flow, i.e., a significant decrease (p<0.01) in zone 1 and a significant increase (p<0.05) in zone 3 and 4, although total RBF was not altered (Fig. 4B). Renal-arterial infusion of All (0.2 μg/min) decreased RBF from 3.01 to 1.52 ml/g·min. However, such an All infusion had no significant effect on the mean arterial pressure or the distribution pattern of blood flow in control animals (Fig. 4A). Reduction of arterial pressure from 140 to 70 mmHg resulted in a significant decrease in blood flow in zone 1 and significant increase in that in zones 3 and 4. Total RBF was unchanged (autoregulation). Angiotensin II infusion during the period of reduced arterial pressure resulted in the same decrease in RBF as was seen at normal pressure. During the period of reduced blood pressure, however, zonal distribution patterns were significantly

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**Fig. 4.** A) Effects of intrarenal-arterial infusion of All (200 ng/min) on percent distribution of blood flow to each zone of renal cortex. All infusion decreased total RBF from 3.01 to 1.52 ml/g·min without any change in mean arterial pressure. Mean of 5 experiments. Vertical line indicates standard error of the mean. B) Effects of arterial pressure reduction (from 125 to 70 mmHg) and All (200 ng/min) infusion at the reduced pressure (70 mmHg) on percent distribution of blood flow of renal cortex. All infusion at reduced arterial pressure resulted in the same decrease in RBF as seen at normal pressure. Mean of 4 experiments. Vertical line indicates the standard error of the mean.
altered by the AII infusion, i.e., a significant increase in the percent blood flow in zone 1, and a significant decrease in that in zones 3 and 4 (Fig. 4B). Thus, AII infusion restored the distribution pattern which was altered by the reduction of arterial blood pressure.

DISCUSSION

It has been postulated that the renin-angiotensin system is one of the factors controlling renal hemodynamics. It is well recognized that the injection of AII is capable of altering renal function (11–13). Although it has been suggested that endogenous angiotensin might have similar effects, experimental evidence is lacking. Schnermann et al. (4) and Granger et al. (14) suggested a local action of the renin-angiotensin system and local formation of AII in the kidney. These same authors reported the presence of enzymes required for the formation of AII in the juxtaglomerular apparatus. However, there is apparently no direct evidence for the local action of the renin-angiotensin system. Our present findings indicate clearly that the intrarenal formation of AII is minute and the amount physiologically insignificant.

Arterial pressure reduction resulted in marked increases in PRA and plasma concentration of AI. A linear correlation was observed between PRA and plasma concentration of AI in the systemic arterial and renal venous blood. Since no significant difference in AI concentrations was observed between arterial and venous blood, it appears that renin produces AI in the renal circulation exactly at the same rate as AI production in the systemic circulation.

During the arterial pressure reduction, plasma concentration of AII in the systemic arterial blood was elevated from 75 to 161 pg/ml with a concomitant increase in PRA and plasma concentration of AI. AII concentration in the renal venous blood was lower than that in the systemic arterial blood during the control period. During the period of reduced renal arterial pressure, AII in the renal venous blood was elevated with an increase in RSR, but AII in the renal venous blood did not significantly exceed the concentration of AII in the systemic arterial blood (Fig. 1). The ratio of AII to AI was 0.051 in the arterial blood and 0.029 in the renal venous blood, the difference being statistically significant. This indicates that the formation of AII from AI in the renal circulation was significantly less than that in the systemic circulation, although the rate of AI formation was the same. Ng and Vane (5) also reported that the conversion of AI to AII was negligible in the renal circulation.

Gocke et al. (9) found that AII concentration increased in the renal venous plasma of the ischemic kidney, concomitant with an increase in renin activity. In the plasma from the contralateral kidney, however, AII concentration was either normal or reduced despite an increase in plasma renin concentrations. These investigators thus suggested that the normal kidney is capable of inactivating or excreting circulating AII. Hodge et al. (15) and Bailie and Oparil (16) also demonstrated a large clearance rate of AII. Therefore, differences in AII concentration between the systemic arterial and renal venous blood observed in the present experiment may depend on the large capacity of the kidney to in-
activate or excrete circulating AII.

We attempted to determine whether the increased AII concentration in the systemic arterial blood, instead of the intrarenally formed AII, has a specific effect on renal hemodynamics. The released renin generates AI in the intrarenal circulation as well as in the systemic circulation, and the produced AI is converted further to AII by converting enzyme. In the present experiments, the concentration of AII in the systemic arterial blood increased from 75 to 161 pg/ml during arterial pressure reduction. If we may assume that the PRF is 100 ml/min, the load of AII on one kidney should be approximately 8 ng/min. Intrarenal arterial infusion of AII at a rate of 8 ng/min induced no change in total RBF and GFR. In addition, an arterial pressure reduction from 140 to 70 mmHg resulted in a marked dilation of renal blood vessels, especially in the afferent arteriole, with a marked increase in RSR. We previously reported that an intrarenal arterial infusion of angiotensin II antagonist, 1-Sar-8-Ile-angiotensin II, did not affect the resting vascular tone of renal vessels, and that the angiotensin II antagonist did not disturb the renal autoregulation and redistribution of blood flow by pressure reduction (17). In contrast, Itskovitz and McGiff (18) reported that infusions of AI in the presence or absence of converting enzyme inhibitor selectively diminished the inner cortical fraction of renal blood flow without affecting the outer cortical fraction. They concluded that AI is major determinant of inner cortical flow. However, in our previous experiments (17) the infusion of AII antagonist completely blocked the vasoconstrictor action of AI (0.2 μg). These dissociations might depend on the difference in experimental procedures: in situ kidney preparations or isolated perfused kidney preparation.

Schnermann et al. (4) demonstrated that increased Na concentration in Henle’s loop led to a decrease in the filtration rate of the glomerulus belonging to the same nephron unit and suggested that the collapse of tubules is due to the AII-induced afferent arterial constriction resulting from the sodium-induced increase in the renin activity. On the contrary, Hall et al. (19) recently reported a dissociation of renal blood flow and glomerular filtration rate autoregulation in renin-depleted dogs on a high-sodium diet and deoxycorticosterone acetate. They concluded that the renin-angiotensin system participates in the control of GFR, possibly by an efferent arteriolar mechanism. The present data do not support the hypothesis that both afferent and efferent arterioles are constricted as a result of an increase in AII concentration.

Itskovitz and McGiff (18) reported that infusion of AII in the isolated perfused kidney decreased outer cortical blood flow without producing consistent changes in inner cortical blood flow. In contrast, Rector et al. (20) reported that an intrarenal infusion of AII decreased total blood flow to about 40%, but there was no alteration in the zonal distribution of blood flow. Similarly in the present study, the arterial infusion of AII did not affect the intrarenal distribution of blood flow. A reduction of renal arterial pressure stimulated the renin release and altered the distribution of blood flow from the outer to the inner cortex. AII infusion at a reduced arterial pressure resulted in changing the distribution pattern of the blood flow to the opposite direction to that seen with reduction of arterial pressure,
whereas the RSR level remained high. Moreover, we previously reported a dissociation between renin release and changes in the intrarenal distribution pattern of blood flow during maximum vasodilation caused by an intrarenal acetylcholine infusion (21). Grandchamp et al. (22, 23) also reported such a dissociation. These investigators found that hemorrhagic hypotension resulted in a progressive and patchy hypoperfusion of the renal cortex with a marked renin release. Alpha-adrenergic blockade by phenoxybenzamine completely prevented alterations in distribution of RBF in hemorrhagic hypotension or restored the normal distribution pattern without any change in RSR. These findings indicate that the released renin does not induce changes in distribution of blood flow through the intrarenal formation of AI.

The present results suggest that released renin does not affect renal hemodynamics through the intrarenal formation of AI, and changes in intrarenal hemodynamics during pressure reduction are not specific for the autoregulatory process. A more general pattern which depends on intrinsic differences in the responsiveness of arterioles, such as differences in myogenic force of afferent arterioles in different cortical zones may be involved.

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