Inhibition of Captopril-induced Increase in Plasma Renin Activity by Propranolol

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SUMMARY
The inhibition of renin release by angiotensin II (AII) is well documented. However, the interaction of this short loop feedback mechanism of AII with the sympathetic nervous system is still unclear. This study was designed to investigate the possible functional relationship between AII and the beta-adrenergic receptors with respect to renin release in vivo. First, the effect of propranolol on captopril-induced renin release was examined in conscious rats. Secondly, the effect of AII on isoproterenol-induced renin release was determined. Captopril (1 mg/Kg) increased plasma renin activity (PRA) from 1.6±0.3 ng/ml/hr to 4.5±0.6 ng/ml/hr (p<0.01). In contrast, there was no significant change in PRA in rats which received both captopril and propranolol (before 0.9±0.2 ng/ml/hr, after 1.3±0.3 ng/ml/hr). Thus, propranolol attenuated the increase in PRA caused by captopril. Isoproterenol infusion (0.1 µg/Kg/min) provoked a significant increase in PRA (before 1.3±0.4 ng/ml/hr, after 6.6±1.7 ng/ml/hr, p<0.01). AII infusion in combination with isoproterenol also increased PRA from 1.6±0.4 ng/ml/hr to 5.2±0.3 ng/ml/hr (p<0.01). AII in this dose did not suppress isoproterenol-induced renin release. These results suggest that the beta-adrenergic receptor mediating renin release is functionally located distal to the AII receptor in the short loop mechanism controlling renin release.

Additional Indexing Words:
Renin release  Isoproterenol  Angiotensin II

ANGIOTENSIN II (AII), the effector of the renin-angiotensin-aldosterone system, suppresses the release of renin by a negative feedback mechanism resulting from its direct action on the juxtaglomerular cells. Saralasin, an
angiotensin antagonist, by blocking this negative feedback of AII, stimulates renin release in normal and sodium-depleted rats, dogs, rabbits and man.\(^1\) Since saralasin-induced renin release is inhibited by the beta-adrenergic blocking agent, propranolol, some functional connections between the sites of action of AII and the sympathetic nervous system in the juxtaglomerular cells have been proposed.\(^2\)

The present study was undertaken to evaluate further the possible interaction of AII with the sympathetic nervous system with regard to renin release. First, the effect of propranolol on the release of renin induced by an angiotensin converting enzyme inhibitor, captopril, was examined in conscious rats. Secondly, the effect of AII on renin release stimulated by the beta agonist isoproterenol was investigated, also in conscious rats.

**METHODS**

Male Wistar rats weighing 250–350 Gm were used in this study. They were fed a regular rat chow containing 104 mEq/Kg of sodium and 177 mEq/Kg of potassium. All rats were anesthetized with ether. A venous catheter was placed in the jugular vein and an arterial catheter was introduced into the carotid artery. Both catheters, filled previously with 0.9% saline containing heparin 100 U/ml, were tunnelled subcutaneously to exit from the skin between the scapulae. Following the surgical procedure, all rats were allowed to recover for 24 hours.

We performed a preliminary experiment in individual rats to examine the effect of blood replacement immediately following blood sampling on plasma renin activity (PRA), blood pressure and heart rate, since blood sampling in small animals could lead to a decrease in blood volume and influence these parameters. Blood pressure and heart rate were recorded on a Nihon-Kohden multipurpose recorder through the arterial catheter connected to a pressure transducer. Two ml of blood were collected through the venous catheter

![Fig. 1. Plasma renin activity (PRA), blood pressure (BP) and heart rate (HR) before (B) and after (A) blood sampling and replacement in conscious rats (n=11). Values are mean±SE.](image-url)
and replaced by an equal volume of blood prepared from another donor rat. After 60 min blood pressure and heart rate were measured and an additional 2 ml of blood were withdrawn. As shown in Fig. 1, there were no significant changes in any of the measured parameters. Blood pressure, heart rate and PRA returned to the baseline levels 60 min later. This method was applied to the subsequent studies to examine drug-induced effects on these parameters in the same rats.

In another preliminary experiment we investigated the effect on PRA of the dosage of captopril (1–8 mg/Kg) administered using the method outlined above. As shown in Fig. 2, captopril increased PRA in a dose-dependent fashion after 60 min. Even the smallest dose of captopril (1 mg/Kg) caused a substantial rise in PRA (about 200%).

**Experiment 1**

Groups of 6–7 rats were used. Captopril (1 mg/Kg, 1 ml/Kg) or propranolol (1 mg/Kg, 1 ml/Kg) was injected intravenously 60 min after the initial determination of blood pressure, heart rate and PRA. Blood pressure and heart rate were remeasured and 2 ml of blood were collected 1 hour after drug administration. In 1 group of rats propranolol was injected 20 min after captopril administration. Blood pressure, heart rate and PRA were determined similarly 1 hour later. Control rats received an injection of 0.9% saline.

**Experiment 2**

Groups of 5–7 rats were used. Isoproterenol (0.5–0.1 μg/Kg/min) alone, or in combination with AII (5–10 ng/Kg/min), was given by constant infusion (infusion rate 0.5 ml/30 min) for 30 min. Controls received 0.9% saline infusion. Thirty minutes after the start of the infusion, blood pressure and heart rate were recorded and 2 ml of blood were withdrawn for PRA measurement.
PRA was measured by radioimmunoassay of angiotensin I generated during 6 hours of incubation of plasma at 37°C, pH 6.0, using a Becton-Dickinson angiotensin I radioimmunoassay kit and expressed as ng angiotensin I/ml/hr.

Results are presented as mean±SEM. Paired t-test was used for statistical analysis. A p-value of less than 0.05 was considered significant.

**Results**

*Experiment 1*

In control rats, PRA was unchanged (Fig. 3). In propranolol-treated rats PRA tended to decrease, but this change was not significant. Captopril administration caused a significant increase in PRA from 1.6±0.3 ng/ml/hr to 4.5±0.6 ng/ml/hr (p<0.01). Administration of propranolol in addition to captopril clearly suppressed the increase in PRA induced by captopril. There was no alteration in PRA during combined treatment (before 0.9±0.2 ng/ml/hr, after 1.3±0.3 ng/ml/hr) (Fig. 3).

Fig. 3. Alteration in plasma renin activity (PRA) following administration of captopril (SQ, 1mg/Kg) and/or propranolol (Pr, 1mg/Kg). Values are mean±SE. n indicates number of rats.

* p<0.01.

Fig. 4. Change in plasma renin activity (PRA), blood pressure (BP) and heart rate (HR) following administration of captopril (SQ, 1mg/Kg) and/or propranolol (Pr, 1mg/Kg). Values are mean±SE.

※ p<0.05, * p<0.01.
No significant changes in blood pressure were found in either the control or the captopril-treated rats. In contrast, administration of propranolol with and without captopril caused rises in blood pressure of 8 mmHg and 18 mmHg, respectively (Fig. 4).

Heart rate in control rats was unchanged. Propranolol decreased heart rate significantly. Neither captopril nor combined treatment had any significant effect on heart rate (Fig. 4).

Experiment 2

PRA in control rats was unchanged. Isoproterenol infusion (0.1 µg/Kg/min) considerably increased PRA from 1.3 ± 0.2 ng/ml/hr to 6.6 ± 1.7 ng/ml/hr (p<0.01, Fig. 5). Simultaneous infusion of AII (5 ng/Kg/min) and isoproterenol (0.1 µg/Kg/min) also increased PRA from 1.6 ± 0.4 ng/ml/hr to 4.2 ± 0.3 ng/ml/hr (p<0.01). A similar rise in PRA was observed when we infused a larger dose of AII (10 ng/Kg/min) with a smaller dose of isoproterenol (0.05 µg/Kg/min).

There were no significant changes in blood pressure in any group of rats (Fig. 5). Heart rate in control rats was unchanged. Isoproterenol infusion alone or in combination with AII increased heart rate significantly (Fig. 5).

![Fig. 5. The change in plasma renin activity (PCA), blood pressure (BP) and heart rate (HR) during isoproterenol (Isopr, 0.1 µg/Kg/min) infusion with or without angiotensin II (AT, 5 ng/Kg/min). Values are mean ± SE. n indicates number of rats. * p<0.05, ** p<0.01.](attachment:image)

**DISCUSSION**

The sympathetic nervous system, macula densa and the intrarenal baroreceptors are the three major factors which control the release of renin. Several other mechanisms are also involved in the regulation of renin release. Among others, AII has been shown to inhibit renin release in vivo and in vitro. Injection of AII intravenously or into the renal artery of intact animals reduces...
renin activity in the renal venous plasma.\(^3\),\(^4\) AII can inhibit renin release from the nonfiltering kidney without producing detectable changes in renal blood flow or renal perfusion pressure. These observations have been interpreted as evidence of a direct effect of AII on the juxtaglomerular cells. Further support for the direct negative feedback by endogenous AII on renin release comes from experiments with the AII antagonist, saralasin,\(^5\) and the converting enzyme inhibitors, teprotide\(^5\) and captopril. In these experiments blockade of AII generation or of AII receptors led to an increase in PRA in the intact animals.

Capponi et al\(^6\) demonstrated the inhibition of isoproterenol-stimulated renin release from rat kidney slices by both AII and a number of beta-adrenergic blockers. They pointed to the possibility that AII may act directly on the juxtaglomerular cells via a beta-adrenergic mechanism. Moreover, in a clinical study with hypertensive patients, Pettinger et al\(^2\) showed that saralasin-induced renin release was prevented by the administration of propranolol. This observation also indicated that the inhibition of renin release by AII is somehow related to the adrenergic control of renin secretion.

In the present study captopril increased PRA in a dose-related manner. This increase in PRA provoked by captopril was presumably due to the interruption of the so-called "short-loop" negative feedback mechanism of AII on renin release since blood pressure was unaffected by captopril administration. Propranolol administered 20 min following captopril injection definitely blunted the increase in PRA induced by captopril. This finding is in agreement with others.\(^7\),\(^8\) Although we cannot exclude the influence of the slight rise in blood pressure on renin release via the baroreceptor mechanism, these results strongly suggest possible connections between AII and the sympathetic nervous system in terms of renin release. It has been demonstrated that the addition of propranolol to captopril produces no further fall in blood pressure.\(^8\) However, propranolol may be useful in preventing the accumulation of large quantities of angiotensin I in plasma during long-term use of captopril. The rapid conversion of large quantities of accumulated angiotensin I could take place following discontinuation of captopril resulting in an abrupt elevation of blood pressure.\(^9\)

In our study AII infused in combination with isoproterenol did not inhibit the increase in PRA induced by isoproterenol. In contrast with our findings, Meyer et al\(^10\) and Vandongen et al\(^11\) found that the increase in PRA associated with isoproterenol infusion was markedly suppressed by simultaneous infusion of AII in rats. The latter authors interpreted their finding as evidence of a close functional relationship between the AII receptor and the beta-receptor in the kidney and proposed that the site of action of
AII might be distal to the beta-receptor. It is noteworthy that this positional relationship is totally different from that which Pettinger et al\textsuperscript{2} proposed. They postulated that the AII receptor inhibiting renin release is functionally located proximal to the beta-adrenergic receptor mediating renin release. Our initial finding that propranolol inhibited the increase in PRA following captopril administration also supports the hypothesis that the AII receptor in the short-loop mechanism controlling renin release is functionally proximal to the beta-receptor mediating renin release. Our second finding that AII did not reduce the increase in PRA stimulated by isoproterenol neither refutes nor supports this hypothesis. The fact that we used a much smaller dose of AII relative to that of isoproterenol than did previous investigators might explain the discrepancy between our results and theirs.\textsuperscript{10,11} However, our second finding may indicate that beta-adrenergically mediated renin secretion and the AII short-loop feedback mechanism are independent.

Clarification of the relation between the AII receptor and the beta-receptor with regard to renin release will require the use of a pure juxtaglomerular cell preparation and a better definition of the pharmacologic properties of receptors for AII and beta-adrenergic agonists on these cells.\textsuperscript{19}

**References**