Role of Renal Prostaglandins in Sympathetically Mediated Renin Release in the Rat

WILLIAM B. CAMPBELL, ROBERT M. GRAHAM, and EDWIN K. JACKSON, *University of Texas Health Science Center, Department of Pharmacology, Division of Clinical Pharmacology, Dallas, Texas 75235*

ABSTRACT Renal prostaglandins (PG) appear to mediate renin release due to stimulation of the intrarenal baroreceptor, but not that due to activation of the macula densa. However, as the role of PG in sympathetically mediated renin release remains unclear, a possible interrelationship between these factors was examined in conscious rats. Hydralazine increased the serum renin levels from 3.1 ± 0.8 to 16.7 ± 3.0 ng/ml per h at a dose of 1 mg/kg. Indomethacin (5 mg/kg) suppressed urinary PGE_2 and $PGF_{2\alpha}$ excretion by 89 and 74%, respectively, arachidonate hypotension by 82%, and inhibited the elevated renin levels from hydralazine by 100% without altering the hypotensive effect of the drug. Another PG synthetase inhibitor, meclofenamate, was also effective in attenuating hydralazine-induced renin release, urinary PGE_2 and $PGF_{2\alpha}$ excretion, and arachidonate hypotension. Isoproterenol, a nonselective beta-adrenergic agonist, increased heart rate, lowered blood pressure, and also stimulated the release of renin when administered intraperitoneally. However, intrarenal infusion of the drug only resulted in increased renin release. Indomethacin inhibited isoproterenol-induced renin release by 66 and 67%, respectively, without altering the hemodynamic effects associated with the intraperitoneal administration of the drug. The selective $beta_1$ agonist, H133/22, increased the release of renin and heart rate in a dose-related manner without altering blood pressure. H133/22-induced renin release was inhibited by 80% by indomethacin pretreatment. Finally, intrarenal infusions of dibutyryl cyclic AMP (3 mg/kg per min) increased the serum activity from 4.1 ± 0.2 to 20.4 ± 3.9 ng/ml per h without altering mean arterial pressure. Indomethacin inhibited this renin response to dibutyryl cyclic

AMP by 96%. Thus, renal PG appear to be important mediators of sympathetically stimulated renin release acting as a site distal to the beta-adrenergic receptor.

INTRODUCTION

The renin-angiotensin system is involved in the homeostatic regulation of arterial blood pressure as well as the maintenance of the elevated blood pressure in several forms of hypertension. Recently, prostaglandins have been implicated as important mediators of the release of renin from the kidney thereby controlling the activity of this system. This conclusion is based on direct evidence obtained from the administration of the prostaglandin precursor, arachidonic acid, or the various prostaglandins themselves, as well as indirect evidence with prostaglandin synthesis inhibitors. Arachidonic acid increases the rate of renin secretion when infused into the renal artery of experimental animals (1–3) and when added to the incubation media of renal cortical slices (4, 5). Because this effect on renin release can be blocked in vivo and in vitro by indomethacin, an inhibitor of prostaglandin synthesis (6), arachidonic acid must be converted to one of its prostaglandin $(PG)^1$ intermediates to exert its action (1, 4, 5). In similar studies, intrarenal infusions of PGE_1 and PGE_2 (7–11), PGD_2 (9, 10), PGI_2 (11), and intravenous infusions of PGA_1 (12, 13) also stimulated the release of renin; however, only the PG endoperoxides, PGG_2 and PGH_2, or PGI_2 could increase the rate at which renin was secreted in vitro from renal cortical slices (4, 5). Because the endoperoxides can be converted to PGI_2 in the renal cortex (14, 15), PGI_2 would appear to exert a direct action on juxtaglomerular cells, and from the available evidence, would appear to be the arachidonic acid intermediate regulating the release of renin.

Additionally, drugs which block the synthesis of PG

will inhibit the release of renin. Indomethacin, for example, will inhibit supine plasma renin levels (16–18) as well as those stimulated by upright posture (16) and furosemide (17, 18). It will also reduce the hyper-reninemia associated with hemorrhage (19), Bartter's syndrome (20, 21), acute renal failure (22), and renal hypertension (23). Because the sympathetic nervous system is thought to mediate the elevated renin release associated with some of these aforementioned examples and because the beta-adrenergic blocker, propranolol, can also mitigate the renin release in those same examples (24–26), the contribution of the renal PG to sympathetically mediated renin release was investigated. In our studies, sympathetically mediated renin release was examined in three ways. First, reflex activation of the sympathetic nervous system was produced by the vasodilator, hydralazine (27, 28). Secondly, beta-adrenergic receptors were directly stimulated by the nonselective beta agonist, isoproterenol (5, 29, 30), and the $beta_1$ selective agonist, H133/22 (31). Finally, renin release was stimulated distal to the beta-adrenergic receptor by $N^6,O^{2'}$-dibutyryl adenosine, $3':5'$-cyclic monophosphoric acid (dibutyryl cyclic AMP) (32, 33). In addition, because hydralazine and isoproterenol have the potential to stimulate renin release by decreasing the blood pressure as well as by stimulating juxtaglomerular cell beta-adrenergic receptors, their effects on renin release and blood pressure were compared with those of chlorisondamine, a ganglionic blocking agent, that stimulates renin release by decreasing the blood pressure without increasing sympathetic nerve activity (34). In these studies, inhibitors of PG synthesis were found to inhibit all three forms of sympathetically mediated renin release tested.

METHODS

Male Sprague-Dawley rats (250–300 g) were used in these studies (Simonsen Labs, San Francisco, Calif.). The rats were maintained on a standard Purina rat chow (Ralston Purina Co., St. Louis, Mo.) diet containing 150 meq Na/kg and 220 meq K/kg and tap water ad libitum. All experiments were performed between 0900 and 1200 on conscious rats to eliminate the influence of diurnal variation, surgical stress, or anesthetic stress on renin release.

In the studies involving renin release, olive oil or a suspension of a PG synthesis inhibitor (indomethacin or meclofenamate, 5 mg/kg) in olive oil was injected subcutaneously at time zero. 90 min later saline, hydralazine, isoproterenol, H133/22, or chlorisondamine was administered intraperitoneally, or subcutaneously as in the case of the latter compound, and the animals were sacrificed at 110 min. The animals were killed by decapitation, and aortic blood was collected in siliconized tubes kept on ice (27, 28). The blood samples were allowed to clot at 4°C, centrifuged at 4°C, and the serum separated at 4°C and stored at −20°C until assayed for serum renin activity.

In the studies involving isoproterenol and dibutyryl cyclic AMP, chronic Weeks' catheters were surgically placed in the abdominal aorta with the tip of the catheter extending above the bifurcation of the renal arteries (35). After a 2-d recovery period, the protocol followed was similar to that previously described with oil or indomethacin administered at time zero followed in 90 min by a saline, isoproterenol (100 ng/kg per min), or dibutyryl cyclic AMP (3 mg/kg per min) intraarterial infusion at 50 μl/min for 20 min. After the infusion, the animals were decapitated, and the blood was collected as previously described. Additionally, the trunks of the animals were retained, and the surgical placement of the catheter was verified.

In the studies in which changes in mean arterial blood pressure and heart rate were measured, chronic Weeks' catheters were placed in the abdominal aorta (35). After allowing 2–5 d for the animals to recover from the surgical stress, the conscious rats were placed in individual, closed cages in a quiet room while mean arterial pressure and heart rate were monitored with a Narco RP-1500 pressure transducer (Narco Scientific Industries, Inc., Fort Washington, Pa.) and recorded by a Grass model 7 polygraph (Grass Instrument Co., Quincy, Mass.). After allowing 1 h for the animals to equilibrate, a protocol identical to those described above was followed. In some animals, sodium arachidonate (8 mg/kg) was administered intravenously before and 110 min after pretreatment with a PG synthesis inhibitor, and changes in mean arterial pressure were monitored.

An additional group of 12 rats was placed in metabolism cages to allow the collection of urine. After 2 d of equilibration, urine was collected for 8 h from 0900 to 1700 from rats injected with either olive oil or a PG synthesis inhibitor. The urine was then measured, filtered, and stored frozen at −20°C until assayed for PG.

Serum renin activity was measured by the antibody trapping method of Poulsen and Jorgensen (36), and the results were expressed as nanograms of angiotensin I generated at 37°C per milliliter of serum per hour (ng angiotensin I/ml per h). Urinary PG were measured according to the method of Dray et al. (37) involving acid-lipid extraction, silicic acid chromatography, and the radioimmunoassay of PGE_2 and $PGF_{2\alpha}$. The PGE_2 antibody cross-reacted 14% with PGE_1, but <0.5% with PGA_2, PGB_2, PGD_2, 6-keto $PGF_{1\alpha}$, $PGF_{2\alpha}$, or its 15-keto metabolites. The $PGF_{2\alpha}$ antibody cross-reacted 100% with $PGF_{1\alpha}$, but <0.5% with PGA_2, PGB_2, PGD_2, 6-keto $PGF_{1\alpha}$, PGE_2, or its 15-keto metabolites. The sensitivity of the assays was <5 pg/0.3 ml.

The drugs and their sources of supply were as follows: isoproterenol (Sigma Chemical Co., St. Louis, Mo.), dibutyryl cyclic AMP (Sigma Chemical Co.), H133/22 (A. B. Hassle, Mölndal, Sweden), meclofenamate (Parke-Davis), hydralazine (Ciba-Geigy Corp., Summit, N. J.), arachidonic acid (Sigma Chemical Co.), chlorisondamine (Ciba-Geigy Corp.), indomethacin (Sigma Chemical Co.), PGE_2 and $PGF_{2\alpha}$ (The Upjohn Co., Kalamazoo, Mich.), angiotensin I (Beckman Instruments Inc., Fullerton, Calif.), carrier free ^{125}I (New England Nuclear, Boston, Mass.), and [^3H]PGE_2 and [^3H]$PGF_{2\alpha}$ (New England Nuclear).

Statistical analyses were performed with an unpaired Student's t test for single comparisons and analysis of variance for multiple comparisons.

RESULTS

Hydralazine caused an increase in serum renin activity (Table I), which had been found previously to result from reflex activation of the sympathetic nervous system (27, 28). Indomethacin reduced the control renin levels by 32% ($P > 0.1$, NS) from 3.1 ± 0.8 to 2.13 ± 0.2 ng/ml per h. Additionally, hydralazine-in-

TABLE I

Effect of PG Synthetase Inhibition on Hydralazine and Chlorisondamine-Induced Changes in Renin Release, Mean Arterial Pressure, and Heart Rate in Conscious Rats

Treatment	SRA	MAP	HR
	ng AI/ml/h	mm Hg	beats/min
Saline	3.1±0.8	108±3	400±18
Indomethacin, 5 mg/kg s.c.	2.1±0.1	107±3	393±10
Meclofenamate, 5 mg/kg s.c.	2.8±0.5		
Hydralazine, 1 mg/kg i.p.	16.7±3.0*s	85±5*s	413±34
Indomethacin + hydralazine	2.0±0.3*h	80±5*s	382±26
Meclofenamate + hydralazine	7.4±1.5‡h		
Chlorisondamine, 10 mg/kg s.c.	5.6±0.4*s	63±3*s	289±19§s
Indomethacin + chlorisondamine	2.1±0.4*c	68±4*s	271±25§s

Each value represents the mean±SEM for six to nine rats measured at 20 min after saline, hydralazine, or chlorosondamine administration. SRA, serum renin activity; MAP, mean arterial pressure; and HR, heart rate. s, compared with saline control; h, compared with hydralazine; c, compared with chlorisondamine; AI, angiotensin I.
* $P < 0.001$.
‡ $P < 0.01$.
§ $P < 0.05$.

duced renin release was inhibited by indomethacin by 100% ($P < 0.001$). Blood pressure and heart rate remained unchanged after indomethacin alone (Table I). However, hydralazine significantly decreased mean arterial pressure by 21% ($P < 0.001$) at 20 min after its administration. At this time, heart rate was not changed from control values. In the same rats pretreated with indomethacin, hydralazine produced a similar decrease in blood pressure.

Meclofenamate, another PG synthesis inhibitor (6), similarly reduced the control serum renin levels from 3.1±0.8 to 2.8±0.5 ng/ml per h ($P > 0.1$, NS) and inhibited hydralazine-induced renin release by 77% from 16.7±3.0 to 7.4±1.5 ng/ml per h ($P < 0.01$) (Table I).

Further examination of sympathetically mediated renin release used isoproterenol, a nonselective beta-adrenergic agonist known to directly stimulate juxtaglomerular cells to release renin (5, 29, 30). Isoproterenol caused a dose-related increase in serum renin activity, which was inhibited by 66% ($P < 0.001$) at the 10 μg/kg dose and by 44% ($P < 0.05$) at the 30 μg/kg dose after pretreatment with indomethacin (Fig. 1). Isoproterenol produced similar increases in heart rate and decreases in blood pressure at both the 10 and 30 μg/kg doses ($P < 0.001$) (Fig. 2). However, unlike its effect on renin release, indomethacin failed to alter the fall in blood pressure or the rise in heart rate after isoproterenol.

To eliminate the systemic effects of the drug, isoproterenol was infused intraarterially, and its effect on renin release was examined (Table II). Isoproterenol increased the serum renin levels from 4.1±0.8 to 18.7±2.7 ng/ml per h ($P < 0.001$) without altering blood pressure or heart rate (Table II). Indomethacin inhibited this isoproterenol-induced renin release by 67% ($P < 0.01$) in the absence of hemodynamic changes.

To further eliminate the involvement of hypotension in beta-adrenergically mediated renin release, the effect of a beta$_1$ selective agonist, H133/22, was similarly tested. H133/ 22 increased serum renin levels from 3.1±0.4 to 10.4±1.1 ($P < 0.001$) and 25.0±2.8 ng/ml per h ($P < 0.001$) with 0.3 and 1 mg/kg, respectively (Fig. 3). Pretreatment with indomethacin inhibited this release of renin by 80% at the 0.3 mg/kg dose and 71% at the 1 mg/kg dose. Additionally, H133/22 increased heart rate by 33% without altering blood pressure at the 0.3 and 1 mg/kg dose (Fig. 4). Similar hemodynamic changes were observed after indomethacin with the 0.3 mg/kg dose. However, with the 1 mg/kg dose, after indomethacin, H133/22 caused a transient, 6-mm Hg decrease in blood pressure ($P < 0.05$), which was not observed in the absence of indomethacin.

To further characterize beta-adrenergically mediated renin release, an agent dibutyryl cyclic AMP was used which bypasses the juxtaglomerular beta-receptor to directly stimulate the release of renin (32, 33). Fig. 5 illustrates the effect of indomethacin on renin release stimulated by intraarterial infusions of dibutyryl cyclic AMP. Dibutyryl cyclic AMP increased serum renin levels from 4.1±0.2 to 20.4±3.9 ng/ml per h ($P < 0.001$), and indomethacin pretreatment inhibited renin by 96% to 4.7±1.2 ng/ml per h ($P < 0.001$). The
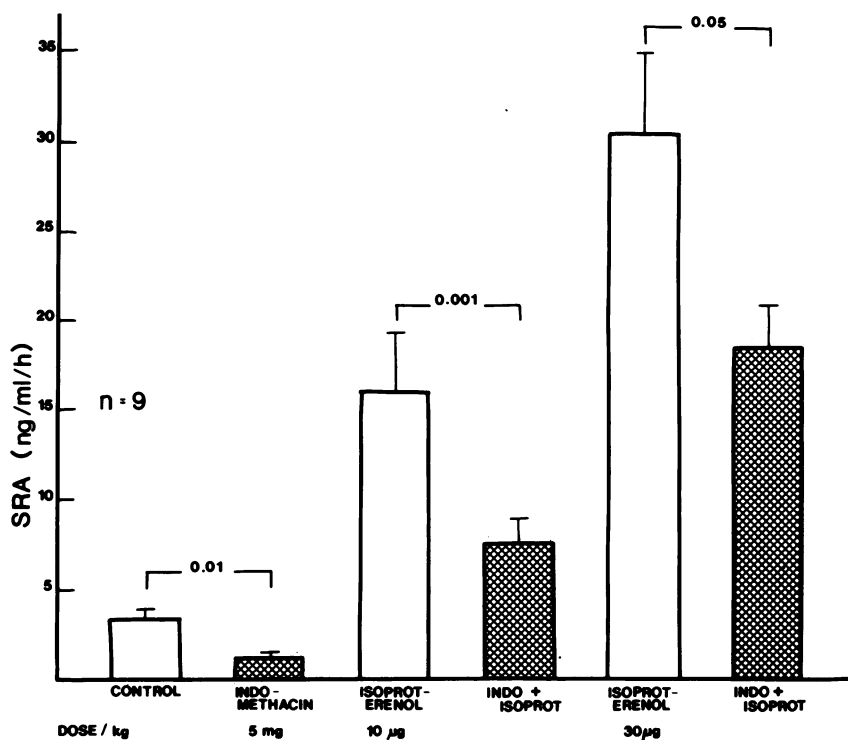
FIGURE 1   Effect of indomethacin (indo) on renin release stimulated by intraperitoneal isoproterenol (isoprot). Each point represents the mean±SEM for nine rats. Statistical significance is indicated by the brackets connecting the compared bars. SRA, serum renin activity.
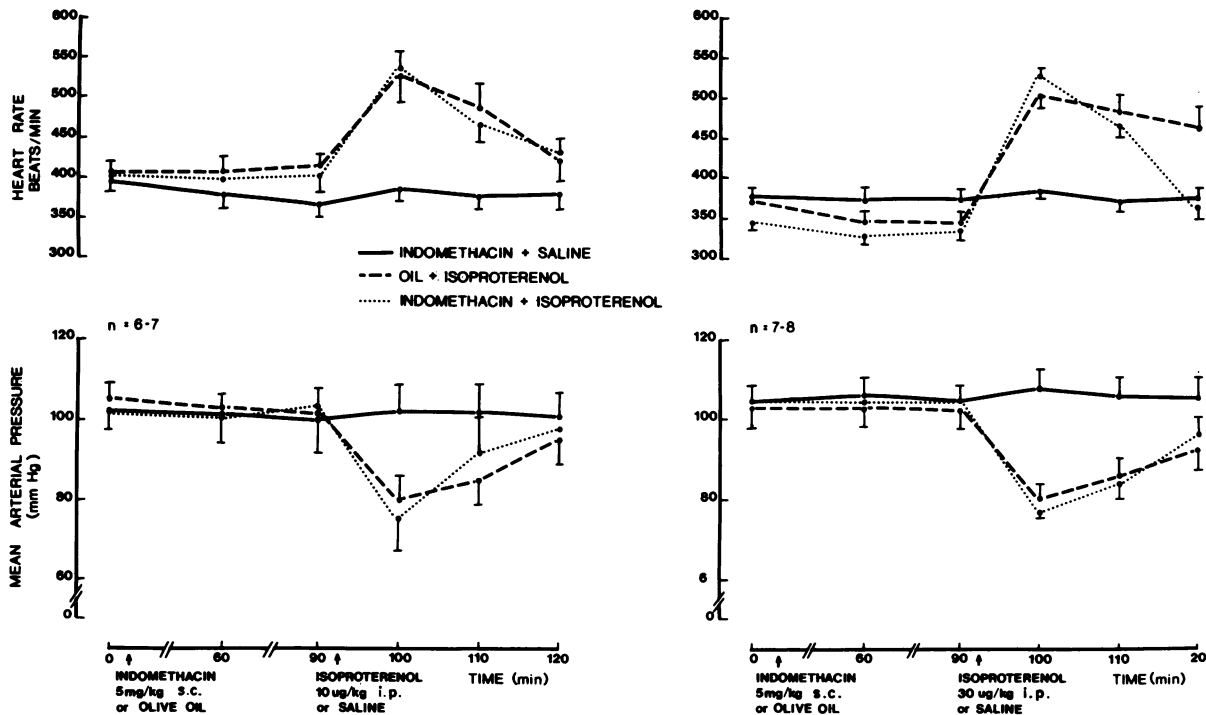

FIGURE 2   Effect of indomethacin on the vasodilation and tachycardia associated with the intraperitoneal administration of 10 μg/kg (left) and 30 μg/kg (right) of isoproterenol. Each point represents the mean±SEM for six to eight rats.

Treatment	MAP	HR	SRA
	mm Hg	*beats/min*	*ng AI/ml/h*
Saline	109±2	385±14	4.1±0.8
Indomethacin, 5 mg/kg s.c.	108±2	352±8	3.4±0.5
Isoproterenol, 100 ng/kg/min i.a.	111±3	435±24	18.7±2.7*
Indomethacin + isoproterenol	110±2	412±19	8.9±1.9‡

Each value represents the mean±SEM for six rats. Mean arterial pressure (MAP), heart rate (HR), and serum renin activity (SRA) were measured 20 min after beginning the infusion of saline or isoproterenol. AI, angiotensin I; i.a., intra-arterial administration.
* $P < 0.001$ compared with control.
‡ $P < 0.01$ compared with isoproterenol.

intraarterial infusions failed to alter the blood pressure (109±2.5 vs. 109±2.8 mm Hg); however, heart rate was increased from 338±18 to 402±16 beats/min ($P < 0.05$).

Chlorisondamine, a ganglionic blocking agent, has been found to reduce sympathetic outflow and to stimulate renin release by activation of the intrarenal baroreceptor (29, 34). In our studies, it lowered blood pressure from 108±3 to 63±4 mm Hg ($P < 0.001$) as well as the heart rate (Table I). Similar results were obtained in the indomethacin-pretreated animals. Chlorisondamine alone significantly increased the release of renin. Indomethacin inhibited the serum renin levels in both the control and chlorisondamine-treated groups by 21 and 100%, respectively ($P < 0.001$).

Finally, the effects of indomethacin and meclofenamate on the urinary excretion of PGE_2 and $PGF_{2\alpha}$ were studied (Table III). Indomethacin (5 mg/kg) reduced

urinary PGE_2 excretion by 89% and the $PGF_{2\alpha}$ excretion by 74% ($P < 0.001$). Meclofenamate (5 mg/kg) produced similar reductions of 68 and 65% for PGE_2 and $PGF_{2\alpha}$, respectively ($P < 0.001$). Additionally, whereas in normal rats, arachidonic acid (8 mg/kg i.v.) decreased mean arterial pressure by 28.5±3.6 mm Hg, in animals pretreated with indomethacin or meclofenemate mean arterial pressure was reduced by only 5.1±0.9 and 4.2±1.4 mm Hg ($P < 0.001$), respectively.

DISCUSSION

The sympathetic nervous system, macula densa, and the intrarenal baroreceptor are the three major factors which control the release of renin (38). A number of investigators have used inhibitors of PG synthesis to probe the degree of involvement of the renal PG in these forms of renin release. In such studies, indomethacin was found to inhibit hemorrhage-induced renin release in rabbits (19). Also in dogs with a single
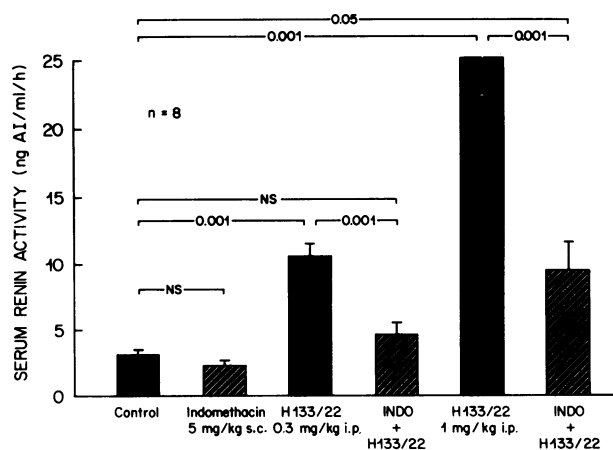

FIGURE 3 Effect of indomethacin (indo) on renin release stimulated by the beta$_1$ selective adrenergic agonist, H133/22. Each point represents the mean±SEM for seven rats. Statistical significance is indicated by the brackets connecting the compared bars. AI, angiotensin I.
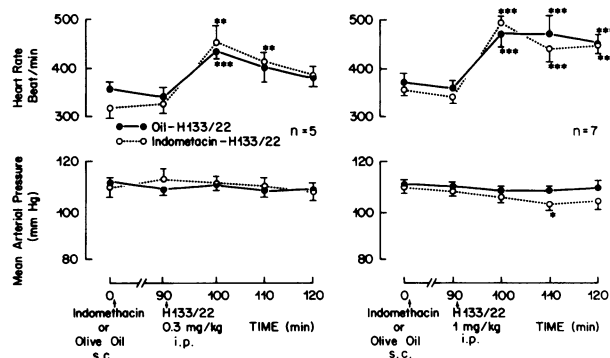

FIGURE 4 The effect of the beta$_1$ selective adrenergic agonist, H133/22, and indomethacin on blood pressure and heart rate. Each point represents the mean±SEM for five to seven rats. **$P < 0.01$; ***$P < 0.001$ compared with the value at 90 min.
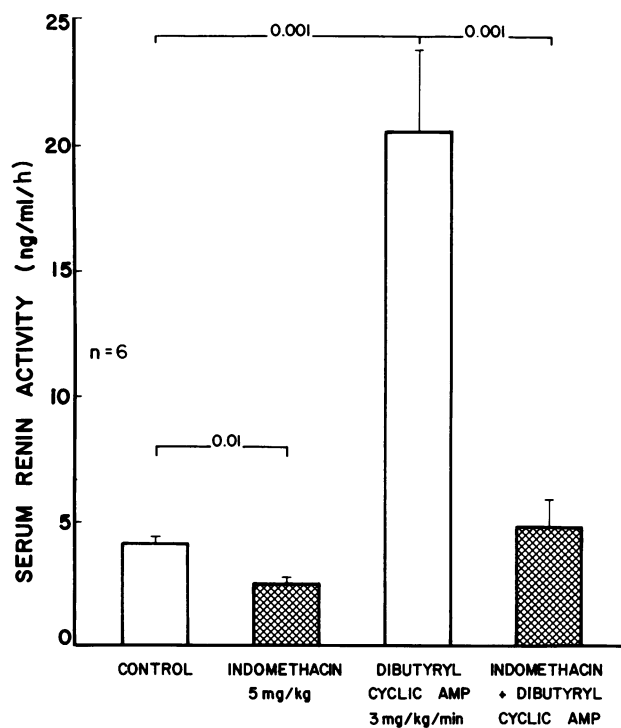
FIGURE 5 Effect of indomethacin on renin release stimulated by intraarterial infusions of dibutyryl cyclic AMP. Each point represents the mean±SEM for six rats. Statistical differences are indicated by the brackets connecting the compared bars.

denervated, nonfiltering kidney, indomethacin blocked the renin release associated with renal arterial hypotension (39). Thus, PG are thought to participate as mediators of intrarenal baroreceptor-stimulated renin release. PG synthesis inhibitors will not, however, inhibit the renin release due to sodium depletion, which is dependent upon a macula densa mechanism (40, 41). Whereas PG participation in sympathetically mediated renin release has not been directly studied, indomethacin was found to inhibit several forms of renin release which were known to possess a beta-adrenergic

component to their mechanism (hemorrhage, upright posture, etc.) (16, 19, 23–26). Furthermore, renal nerve stimulation and exogenous catecholamines were found to enhance the release of PG from the kidney (42, 43). With this in mind, these studies were designed to examine the role of PG in sympathetically mediated renin release and also to indicate the approximate site at which they may participate in this action.

The vasodilator, hydralazine, is known to stimulate renin release by reflex activation of the sympathetic nervous system (27, 28). In this study, the decrease in blood pressure produced by hydralazine was associated with a reflex increase in heart rate and a fivefold increase in renin release. In contrast, the ganglionic blocker, chlorisondamine, caused a decrease in blood pressure similar to that produced by hydralazine; however, because the sympathetic ganglia were blocked, there was no reflex tachycardia and only a twofold increase in renin release. This small increase in renin release by chlorisondamine was produced by the reduced blood pressure which activated the intrarenal baroreceptor (34). Thus, when the release of renin produced by hydralazine and chlorisondamine were compared, the data would indicate that the major component of the hydralazine-induced renin release is sympathetically mediated, and only a small component is baroreceptor mediated.

The PG synthetase inhibitors, indomethacin and meclofenamate, blocked this sympathetically mediated renin release produced by hydralazine without altering its effect on blood pressure. Because hydralazine has been found to decrease PG biosynthesis in rat renal medullary tissue in vitro (44), it is unlikely that the drug could directly stimulate renin release by an action on the synthesis of PG. Thus, these findings indicate that renal PG participate in sympathetically mediated renin release by an action on renal nerve activity or by a direct action on the juxtaglomerular cells.

In the rat, the role of PG in the control of norepinephrine release after sympathetic nerve stimulation is controversial. Some investigators have found that PGE inhibits the release of norepinephrine and reduces the

TABLE III
Effect of PG Synthesis Inhibitors on the Urinary Excretion of PG
and Arachidonate-Induced Hypotension in the Rat

Treatment	PGE_2	$PGF_{2\alpha}$	Arachidonate hypotension
	ng/8 h	ng/8 h	mm Hg
Olive oil, 1 ml/kg s.c.	34.9±4.6	109.0±6.2	28.5±3.6
Indomethacin, 5 mg/kg s.c.	3.8±0.7*	28.0±1.7*	5.1±0.9*
Meclofenamate, 5 mg/kg s.c.	11.2±4.4*	38.3±7.9*	4.2±1.4*

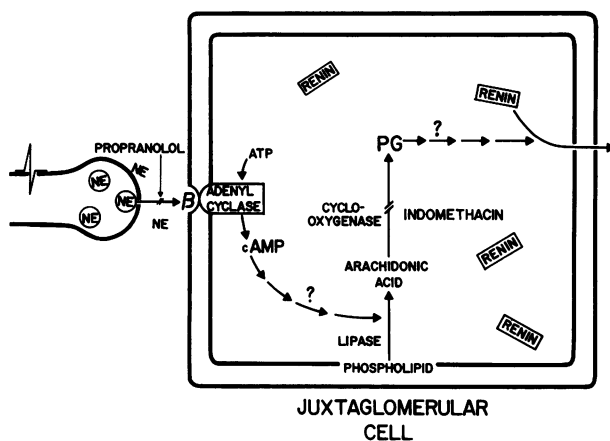Each value represents the mean±SEM for six rats.
* $P < 0.001$ compared with control.

FIGURE 6 A schematic representation of the role of PG in sympathetically mediated renin release. NE, norepinephrine; cAMP, cyclic AMP; β, beta-receptor.

vasoconstrictor responses to nerve stimulation and that indomethacin, by removing this tonic inhibitory influence, enhances these effects (45–48). In contrast, Malik and McGiff (49) reported that PGE enhanced the vasoconstrictor effects of sympathetic nerve stimulation and exogenous norepinephrine in the rat mesentery and suggested that in the rat, unlike other species, PG facilitate norepinephrine release. However, subsequent studies have indicated that PGE inhibits the release of norepinephrine after nerve stimulation in the rat mesentery and enhances the vasoconstrictor responses via a postsynaptic effect.[2] Thus, indomethacin in our studies should enhance the release of norepinephrine after sympathetic activation by hydralazine. This increased norepinephrine release should increase rather than decrease the release of renin unless indomethacin had an additional postsynaptic effect to block the release of renin at or distal to the juxtaglomerular beta-adrenergic receptor (Fig. 6).

With these possibilities in mind, isoproterenol, a beta-adrenergic agonist, was used to directly stimulate juxtaglomerular beta receptors, thus bypassing the renal nerves. As with hydralazine, indomethacin inhibited isoproterenol-induced renin release without altering the associated hypotension or tachycardia. Thus, because classical beta-adrenergic blocking drugs inhibit not only the release of renin but also the tachycardia and hypotension caused by the beta-adrenergic agonist (50, 51), it would appear unlikely that indomethacin is inhibiting renin release via blockade of beta-adrenergic receptors. Along these lines, Frolich et al. (52) also found that indomethacin could inhibit isoproterenol-induced renin release in normal subjects. These findings suggest that renal PG exert their

action on sympathetically mediated renin release at a point in the juxtaglomerular cell distal to the beta-adrenergic receptor (Fig. 6).

Because the systemic administration of isoproterenol was associated with a profound hypotension, the possibility must be considered that the enhanced rate of renin release after isoproterenol was the result of not only juxtaglomerular cell beta-receptor activation but also baroreceptor activation. To eliminate the baroreceptor contribution to renin release, two types of experiments were performed: (a) isoproterenol was infused intraarterially in conscious rats in doses which stimulated the release of renin without altering systemic blood pressure, and (b) a selective $beta_1$-adrenergic agonist, H133/22, (31) was used which released renin and increased heart rate through activation of $beta_1$ receptors yet failed to activate the $beta_2$ receptors necessary for hypotension. In the absence of hypotension, indomethacin pretreatment inhibited these two types of renin release, again suggesting a role for renal PG in sympathetically mediated renin release.

This contention was further examined by testing the effect of indomethacin on dibutyryl cyclic AMP-stimulated renin release. Because this agent does not reduce arterial blood pressure but stimulates renin release in vivo and in vitro by a mechanism which is not inhibited by propranolol (32, 33), it is thought to increase the release of renin by a direct action on juxtaglomerular cells. Indomethacin also inhibited dibutyryl cyclic AMP-stimulated renin release, which further suggested that PG mediated the sympathetically induced renin release via an action past the beta-adrenergic receptor (Fig. 6). Along these lines, Lindgren et al. (53) reported that dibutyryl cyclic AMP increased the release of arachidonic acid and the formation of PG in cultured 3T3 fibroblasts.

In these studies, indomethacin inhibited several forms of sympathetically mediated renin release. This inhibitory effect of the drug seems to be related to its ability to inhibit PG synthesis because a similar effect was observed with meclofenamate, a structurally dissimilar PG synthesis inhibitor, and because in the doses used both compounds markedly inhibited the urinary excretion of PGE_2 and $PGF_{2\alpha}$ and arachidonate-induced hypotension. Thus, these data indicate that renal PG play an important role in sympathetically mediated renin release in the rat and act at a site distal to the beta-adrenergic receptor of the juxtaglomerular cell (Fig. 6).

ACKNOWLEDGMENTS

---

[2] Malik, K. U. Personal communication.

REFERENCES

1. Larsson, C., P. Weber, and E. Anggard. 1974. Arachidonic
   acid increases and indomethacin decreases plasma renin
   activity in the rabbit. Eur. J. Pharmacol. 28: 391–394.
2. Weber, P., H. Holzgreve, R. Stephan, and R. Herbst. 1975.
   Plasma renin activity and renal sodium and water excre-
   tion following infusion of arachidonic acid in rats. Eur.
   J. Pharmacol. 34: 299–304.
3. Bolger, P. M., G. M. Eisner, P. W. Ramwell, and L. M.
   Slotkoff. 1976. Effect of prostaglandin synthesis on renal
   function and renin in the dog. Nature (Lond.). 259: 244–
   245.
4. Weber, P. C., C. Larsson, E. Anggard, M. Hamberg, E. J.
   Corey, K. C. Nicolaou, and B. Samuelsson. 1976. Stimu-
   lation of renin release from rabbit renal cortex by arachi-
   donic acid and prostaglandin endoperoxides. Circ. Res.
   39: 868–874.
5. Whorton, A. R., K. Misono, J. Hollifield, J. C. Frolich,
   T. Inagami, and J. A. Oates. 1977. Prostaglandins and
   renin release. I. Stimulation of renin release from rabbit
   renal cortical slices by PGI_2. Prostaglandins. 14: 1095–
   1105.
6. Flower, R. J. 1974. Drugs which inhibit prostaglandin
   biosynthesis. Pharmacol. Rev. 26: 33–67.
7. Werning, C., W. Vetter, P. Weidmann, H. U. Schweikert,
   D. Stiel, and W. Siegenthaler. 1971. Effect of prosta-
   glandin E_1 on renin in the dog. Am. J. Physiol. 220: 852–
   856.
8. Yun, J., G. Kelly, F. C. Bartter, and H. Smith, Jr. 1977.
   Role of prostaglandins in the control of renin secretion
   in the dog. Circ. Res. 40: 459–464.
9. Bolger, P. M., G. M. Eisner, P. T. Shea, P. W. Ramwell,
   and L. M. Slotkoff. 1977. Effects of PGD_2 on canine renal
   function. Nature (Lond.). 267: 628–630.
10. Bolger, P. M., G. M. Eisner, P. W. Ramwell, and L. M.
    Slotkoff. 1978. Renal actions of prostacyclin. Nature
    (Lond.). 271: 467–469.
11. Gerber, J. G., R. A. Branch, A. S. Nies, J. F. Gerkens,
    D. G. Shand, J. Hollifield, and J. A. Oates. 1978. Prosta-
    glandins and renin release. II. Assessment of renin secre-
    tion following infusion of PGI_2, E_2 and D_2 into the renal
    artery of anesthetized dogs. Prostaglandins. 15: 81–89.
12. Krakoff, L. R., D. DeGuia, N. Vlachakis, J. Stricker, and
    M. Goldstein. 1973. Effect of sodium balance on arterial
    blood pressure and renal responses to prostaglandin A_1
    in man. Circ. Res. 33: 539–546.
13. Golub, M. S., P. F. Speckart, P. K. Zia, and R. Horton.
    1976. The effect of prostaglandin A_1 on renin and aldo-
    sterone in man. Circ. Res. 39: 574–579.
14. Zenser, T. V., C. A. Herman, R. R. Gorman, and B. B.
    Davis. 1977. Metabolism and action of the prostaglandin
    endoperoxide PGH_2 in rat kidney. Biochem. Biophys. Res.
    Commun. 79: 357–363.
15. Whorton, A. R., M. Smigel, J. A. Oates, and J. C. Frolich.
    1978. Regional differences in prostacyclin formation by
    the kidney–prostacyclin is a major prostaglandin of renal
    cortex. Biochim. Biophys. Acta. 529: 176–180.
16. Rumpf, K. W., S. Frenzel, H. D. Lowitz, and F. Scheler.
    1975. The effect of indomethacin on plasma renin activity
    in man under normal conditions and after stimulation of

17. Patak, R. V., B. K. Mookerjee, C. J. Bentzel, P. E. Hysert,
    M. Babej, and J. B. Lee. 1975. Antagonism of the effects
    of furosemide by indomethacin in normal and hyperten-
    sive man. Prostaglandins. 10: 649–659.
18. Frolich, J. C., J. W. Hollifield, J. C. Dormois, B. L. Frolich,
    H. Seyberth, A. M. Michelakis, and J. A. Oates. 1976.
    Suppression of plasma renin activity by indomethacin in
    man. Circ. Res. 39: 447–452.
19. Romero, J. C., C. L. Dunlap, and C. G. Strong. 1976. The
    effect of indomethacin and other anti-inflammatory drugs
    on the renin-angiotensin system. J. Clin. Invest. 58: 282–
    288.
20. Fichman, M. P., N. Telfer, P. Zia, P. Speckart, M. Golub,
    and R. Rude. 1976. Role of prostaglandins in the patho-
    genesis of Bartter's syndrome. Am. J. Med. 60: 785–797.
21. Gill, J. R., J. C. Frolich, R. E. Bowden, A. A. Taylor,
    H. R. Keiser, H. W. Seyberth, J. A. Oates, and F. C. Bartter.
    1976. Bartter's syndrome: a disorder characterized by high
    urinary prostaglandins and a dependence of hyperren-
    menia on prostaglandin synthesis. Am. J. Med. 61: 43–51.
22. Torres, V. E., C. G. Strong, J. C. Romero, and D. M.
    Wilson. 1975. Indomethacin enhancement of glycerol-
    induced acute renal failure in rabbits. Kidney Int. 7: 170–
    178.
23. Romero, J. C., and C. G. Strong. 1977. The effect of indo-
    methacin blockade of prostaglandin synthesis on blood
    pressure of normal rabbits and rabbits with renovascular
    hypertension. Circ. Res. 40: 35–41.
24. Tobert, J. A., J. D. H. Slater, F. Fogelman, S. L. Lightman,
    A. B. Kurtz, and N. N. Payne. 1973. The effect in man
    of (+)-propranolol on renin secretion stimulated by ortho-
    static stress. Clin. Sci. (Oxf.). 44: 291–295.
25. Michelakis, A. M., and R. G. McAllister. 1972. The effect
    of chronic adrenergic receptor blockade on plasma renin
    activity in man. J. Clin. Endocrinol. Metab. 34: 386–394.
26. Winer, N. 1972. Mechanism of increased renin secretion
    associated with adrenalectomy, hemorrhage, renal artery
    constriction, and sodium depletion. In Hypertension 72.
    J. Genest and E. Koiw, editors. Springer-Verlag New York,
    Inc., New York. 25–36.
27. Pettinger, W. A., W. B. Campbell, and K. Keeton. 1973.
    Adrenergic component of renin release induced by vaso-
    dilating antihypertensive drugs in the rat. Circ. Res. 33:
    82–86.
28. Pettinger, W. A., and K. Keeton. 1975. Altered renin re-
    lease and propranolol potentiation of vasodilatory drug
    hypotension. J. Clin. Invest. 55: 236–243.
29. Pettinger, W. A., L. Augusto, and A. S. Leon. 1972. Altera-
    tion of renin release by stress and adrenergic receptor
    and related drugs in unanesthetized rats. In Comparative
    Pathophysiology of Circulatory Disturbances. C. M.
    Bloor, editor. Plenum Publishing Corp., New York. 105–
    117.
30. Weinberger, M. H., W. Aoi, and D. P. Henry. 1975. Direct
    effect of beta-adrenergic stimulation on renin release by
    the rat kidney slice in vitro. Circ. Res. 37: 318–324.
31. Carlsson, E., C-G. Dahlof, A. Hedberg, H. Persson, and
    B. Tangstrand. 1977. Differentiation of cardiac chron-
    tropic and inotropic effects of β-adrenergic agonists.
    Naunyn-Schmiedeberg's Arch Pharmacol. 300: 101–105.
32. Michelakis, A. M., J. Caudle, and G. W. Liddle. 1969.
    In vitro stimulation of renin production by epinephrine,
    norepinephrine, and cyclic AMP. Proc. Soc. Exp. Biol.
    Med. 130: 748–753.
33. Okahara, T., Y. Abe, and K. Yamamoto. 1977. Effects of

dibutyryl cyclic AMP and propranolol on renin secretion in dogs. *Proc. Soc. Exp. Biol. Med.* **156**: 213–218.

34. Keeton, T. K., and W. A. Pettinger. 1979. The dominance of adrenergic mechanism in mediating hypotensive drug-induced renin release in the conscious rat. *J. Pharmacol. Exp. Ther.* **208**: 303–309.

35. Weeks, J. R. 1973. Upjohn 1011. Available upon request from Dr. Weeks, Upjohn Co., Kalamazoo, Mich.

36. Poulsen, K., and J. Jorgensen. 1974. An easy radioimmunological microassay of renin activity, concentration, and substrate in human and animal plasma and tissue based on angiotensin I trapping by antibody. *J. Clin. Endocrinol. Metab.* **39**: 816–825.

37. Dray, F., B. Charbonner, and J. Maclouf. 1975. Radioimmunoassay of prostaglandins Fα, E₁, and E₂ in human plasma. *Eur. J. Clin. Invest.* **5**: 311–318.

38. Davis, J. O., and R. H. Freeman. 1976. Mechanisms regulating renin release. *Physiol. Rev.* **56**: 1–56.

39. Data, J. L., H. G. Gerber, W. J. Crump, J. C. Frolich, J. W. Hollifield, and A. S. Nies. 1978. The prostaglandin system. A role in canine baroreceptor control of renin release. *Circ. Res.* **42**: 454–458.

40. Campbell, W. B., E. K. Jackson, and R. M. Graham. 1979. Saralasin-induced renin release: blockade by prostaglandin synthetase inhibitors. *Hypertension.* In press.

41. Norbiato, G., M. Bevilacqua, U. Raggi, P. Micossi, C. Moroni, and A. Fasoli. 1978. Effect of prostaglandin synthetase inhibitors on renin and aldosterone in man on a normal or low sodium diet. *Acta Endocrinol.* **87**: 577–588.

42. Dunham, E. W., and B. G. Zimmerman. 1970. Release of prostaglandin-like material from dog kidney during nerve stimulation. *Am. J. Physiol.* **219**: 1279–1285.

43. Needleman, P., J. R. Douglas, Jr., B. Jakschik, P. B. Stoecklein, and E. M. Johnson, Jr. 1974. Release of renal prostaglandin by catecholamines: relationship to renal endocrine function. *J. Pharmacol. Exp. Ther.* **188**: 453–460.

44. Taube, C., H. J. Mest, H. U. Block, and W. Forster. 1976. Changes in prostaglandin synthesis in rabbit kidney medulla after repeated intravenous administration of antihypertensive drugs. *Artery.* **2**: 564–573.

45. Bergstrom, S., L-O. Farnebo, and K. Fuxe. 1973. Effect of prostaglandin E₂ on central and peripheral catecholamine neurons. *Eur. J. Pharmacol.* **21**: 362–368.

46. Needleman, P., G. R. Marshall, and E. M. Johnson. 1974. Determinants and modification of adrenergic and vascular resistance in the kidney. *Am. J. Physiol.* **3**: 665–669.

47. Fredholm, B. B., and P. Hedqvist. 1975. Indomethacin-induced increase in noradrenaline turnover in some rat organs. *Br. J. Pharmacol.* **54**: 295–300.

48. Jackson, E. K., W. B. Campbell, and W. G. Clark. 1979. Inhibition of angiotensin II enhancement of adrenergic transmission by propranolol: an antihypertensive mechanism. *Fed. Proc.* **38**: 678. (Abstr.)

49. Malik, K. U., and J. C. McGiff. 1975. Modulation by prostaglandins of adrenergic transmission in the isolated perfused rabbit and rat kidney. *Circ. Res.* **36**: 599–609.

50. Weber, M. A., I. R. Thornell, and G. S. Stokes. 1974. Effects of beta adrenergic blocking agents on plasma renin activity in the conscious rabbit. *J. Pharmacol. Exp. Ther.* **188**: 234–240.

51. Harms, H. H., L. Gooren, A. J. G. Spoelstra, C. Hesse, and L. Verschoor. 1978. Blockade of isoprenaline-induced changes in plasma free fatty acids, immunoreactivity insulin levels and plasma renin activity in healthy human subject, by propranolol, pindolol, practolol, atenolol, metoprolol and acebutolol. *Br. J. Clin. Pharmacol.* **5**: 19–26.

52. Frolich, J. C., J. W. Hollifield, and J. A. Oates. 1976. Effect of indomethacin on isoproterenol induced renin release. *Clin. Res.* **24**: 9A. (Abstr.)

53. Lindgren, J. A., A-E. Claesson, and S. Hammarstrom. 1978. Stimulation of arachidonic acid release and prostaglandin production in 3T3 fibroblasts by adenosine 3':5'-monophosphate. *Adv. Prostaglandin Thromboxane Res.* **3**: 167–174.