Serotonin Infusion in Rats with Experimental Renovascular Hypertension

By EDWIN R. FISHER and PAUL O. KEVERLINE

Simultaneous, constant infusion of serotonin (100 mcg./kg./min.) and angiotensin (2 mcg./kg./min.) for 2 hr. in normotensive rats resulted in a diminution of the pressor response noted with angiotensin alone. A more marked depression occurred following infusion of serotonin in rats with established renovascular hypertension and this agent failed to influence blood pressure in rats with unilateral renal artery constriction that did not develop hypertension. Serotonin and/or angiotensin in the doses employed did not affect the degree of granularity of renal juxtaglomerular cells in these situations. These findings fail to support the view relating some instances of hypertension in man to the combined effects of serotonin and subpressor amounts of angiotensin II.

SUBTOTAL OCCLUSION of the superior mesenteric artery in dogs has been observed to augment hypertension resulting from unilateral renal artery constriction (1). The former procedure alone has no effect on blood pressure (2). A high incidence of renal artery insufficiency or intrinsic renal disease has recently been reported in patients with occlusive disease of the superior mesenteric artery (3). Some normotensive individuals with known renal or renovascular alterations have been observed to develop arterial hypertension following onset of insufficiency of the superior mesenteric artery (3). It has been suggested that the hypertension in this situation may be induced by serotonin by increasing the pressor response to relatively small increases in angiotensin or by decreasing its inactivation by angiotensinase (3). Experimental occlusion of the superior mesenteric artery results in an increased release of intestinal serotonin into the portal circulation (4). However, no data are available concerning the pressor effect of simultaneous infusions of serotonin and angiotensin in normotensive rats or of serotonin in rats in which renovascular hypertension was successfully or unsuccessfully induced by unilateral renal artery constriction. Such information would appear pertinent to considerations relating some instances of renovascular hypertension to hyperserotonemia resulting from mesenteric insufficiency.

MATERIALS AND METHODS

Female Wistar rats weighing 200–250 g. were used in all experiments. These were housed in individual cages and allowed water and a standard ratio *ad libitum*.

All infusions were performed for 2 hr. using a Harvard pump adjusted to deliver 0.0247 ml./min. into a PE-50 polyethylene catheter whose tip was directly placed into the left femoral vein. Animals were lightly anesthetized with ether during the experiment and an airway was maintained with a small indwelling tracheal catheter. Groups of 10 rats received either isotonic saline; 2 mcg./kg./min. of angiotensin[1]; 100 mcg./kg./min. serotonin (5-hydroxytryptamine creatinine sulfate)[2]; or similar concentrations of both simultaneously. All solutions were prepared in isotonic saline. Preliminary studies revealed that smaller doses of angiotensin (1 mcg./kg./min.) failed to consistently evoke a pressor response in untreated rats even when perfused for as long as 5 hr. Infusions of larger doses of serotonin (200 mcg./kg./min.) result in prompt bronchospasm, severe vasodepression, episodes of apnea, and death in 90% of untreated rats, those simultaneously infused with 2 mcg./kg./min. of angiotensin, or those with established renovascular hypertension. Smaller doses of serotonin (50 mcg./kg./min.) failed to influence the blood pressure of untreated animals and did not alter the pressor responses of rats to simultaneous infusion of 2 mcg./kg./min. of angiotensin. Infusion of this dose of serotonin and/or angiotensin for 5 hr. resulted in subsequent decline in blood pressure, apnea, and death.

Rats subjected to unilateral renal artery constriction with a silver clip also received an infusion of isotonic saline or serotonin in the concentrations noted above, regardless of whether induction of hypertension was successful. Rats whose constricted kidneys exhibited infarction at the time of sacrifice were excluded from analysis.

Blood pressure was constantly monitored from the right femoral artery during the infusion. A PE-50 polyethylene catheter was inserted into the artery and attached to a pressure transducer in sequence with a transducer amplifier and Viso-Cardiette recorder.

Animals were exsanguinated at the conclusion of the infusion and blood urea nitrogen (BUN), hematocrit, and serum Na, K, and Cl determined by standard methods. Kidneys were fixed in Helly's fluid and sections stained for the estimation of juxtaglomerular cell granularity (JGI) (5).

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[1] Hypertensin, Ciba Pharmaceutical Co., Summit, N. J.
[2] Sigma Chemical Co., St. Louis, Mo.

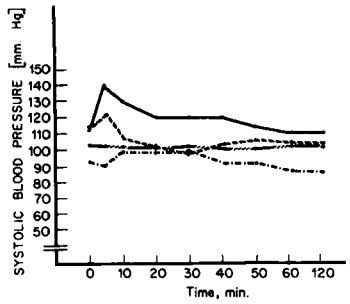

Fig. 1—Effect of constant infusion of serotonin and/or angiotensin on blood pressure of normotensive rats (average of 10 rats/group). Key: —···—, saline; ——, angiotensin; — · —, serotonin; - - -, angiotensin and serotonin.

RESULTS

As indicated in Fig. 1 slow infusion of isotonic saline failed to significantly alter blood pressure in normotensive controls. Administration of angiotensin resulted in an immediate pressor response of 30 ± 5 mm. Hg ($p < 0.01$) which lasted for approximately 5–10 min. Subsequent readings were slightly but not significantly ($p > 0.05$) elevated above control values for approximately 50 min., at which time pressure returned to normal. Administration of serotonin was followed by an immediate, insignificant ($p > 0.05$) depression of blood pressure followed by a slight (10 mm. Hg), statistically insignificant ($p > 0.05$) increase that was maintained for approximately 40 min., at which time pressures returned to control values (Fig. 1). Infusion of both angiotensin and serotonin together resulted in a prompt, slight (10 mm. Hg) insignificant ($p > 0.05$) increase in pressure followed within 5 min. by slight depression of blood pressure which was most marked (14 mm. Hg) at 0.5 hr. (Fig. 1).

The effect of serotonin on blood pressure of rats with established renal hypertension was characterized by an immediate, significant ($p < 0.01$) depression to normotensive levels followed by a significant ($p < 0.05$) rise reaching its maximum of 20–48 mm. Hg at 15 min. This elevation, which was subsequently sustained at this level, was never noted to exceed the initial hypertensive pressure (Fig. 2). Animals with unilateral renal artery constriction that did not develop hypertension exhibited a response to serotonin almost identical to that observed in unoperated, normotensive rats receiving this agent (Fig. 2). No change was noted in rats with unilateral renal artery constriction infused with saline.

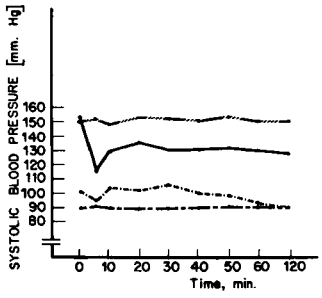

Fig. 2—Effect of constant infusion of serotonin on blood pressure of hypertensive and normotensive rats with unilateral renal artery constriction (average of 10 rats/group). Key: —···—, hypertensive-saline; ——, hypertensive-serotonin; — · —, normotensive-serotonin; — - —, normotensive-saline.

No significant differences in hematocrit, BUN, serum Na, K, or Cl were noted among the various groups of infused rats. All values were also comparable to those obtained from unoperated, normotensive controls in this laboratory. Similarly, no statistically significant difference ($p > 0.05$) in JGI was observed in rats infused with angiotensin and/or serotonin and those receiving only saline (Table I).

JGI of unclipped kidneys of rats with established renal hypertension were significantly ($p < 0.01$) less than those of clipped kidneys (Table I). These latter were significantly greater than control values ($p < 0.01$). No difference in JGI of clipped or unclipped kidneys from rats with unilateral renal artery constriction that did not develop renal hypertension was evident. Serotonin infusion in these latter animals failed to influence JGI.

DISCUSSION

Serotonin is generally regarded as a vasodepressor (6, 7). Such activity was observed only in preliminary studies with larger doses than that employed in the definitive investigations and was invariably accompanied by severe bronchospasm, apnea and death in 90% of the animals. The significance of dose-response relationships with this agent has been emphasized (8). Although pressor responses to serotonin vary in different species, the immediate, slight depression of blood pressure promptly followed by restoration to normal levels noted in this study coincides with the pattern observed by Page (9) in the rat. Development of compensatory cardiovascular reflexes or tachyphylaxis following constant infusion of angiotensin noted in rats in this study has also been noted previously in dogs (10). On the other hand, a depressor effect of serotonin was more perceptible when ad-

TABLE I—JUXTAGLOMERULAR INDEXES (JGI) FOLLOWING INFUSION OF SEROTONIN AND/OR ANGIOTENSIN IN RATS WITH AND WITHOUT UNILATERAL RENAL ARTERY CONSTRICTION

Infusion	No.	JGI	Clipped Kidney	Unclipped Kidney
Nonconstricted				
Saline	10	33 ± 12[a]		
Angiotensin	10	29 ± 10		
Serotonin	10	26 ± 14		
Angiotensin + serotonin	10	33 ± 11		
Constricted-hypertensive				
Saline	10		55 ± 19	10 ± 4
Serotonin	10		57 ± 17	7 ± 5
Constricted-normotensive				
Saline	10		32 ± 11	30 ± 6
Serotonin	10		29 ± 7	28 ± 4

[a] Standard deviation.

ministered simultaneously with angiotensin or to rats with established renovascular hypertension. These findings were consonant with evidence indicating a preferential inhibitory or vasodilating effect of serotonin on constricted arterioles, the net effect of which is vasodepression (8, 11). The slight, insignificant increase in blood pressure noted in normotensive rats infused with serotonin appears to be the result of the vasoconstriction of large vessels induced by this agent in the absence of arteriolar constriction (11).

This information, as well as the failure to observe significant elevation of blood pressure in rats with a degree of unilateral renal artery constriction that was insufficient to produce hypertension indicate that serotonin does not augment renovascular hypertension in the rat as has been suggested to occur in man (3). Indeed, the converse was obtained in this species. Although renal blood flow and glomerular filtration rate may be decreased following administration of serotonin (12), its lack of effect on JGI suggests that any action of serotonin on blood pressure is not mediated by renal juxtaglomerular cells, the purported sites of renal renin formation. The failure to observe any significant effect of angiotensin infusions on JGI is unlike the effect of daily subcutaneous injections of pressor doses of this agent. Katz *et al.* (13) observed increased granularity during the first week of treatment after which time return to control values was noted.

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Keyphrases

Serotonin infusions—rats
Renovascular hypertension—experimental
Angiotensin effect—serotonin activity
Renal artery constriction—serotonin activity

Toxicity of Emetine to Isolated Embryonic Chick-Heart Cells

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Isolated beating embryonic chick-heart cells were cultured and observed *in vitro* to investigate the toxic specificity of emetine for the myocardium. The results of this investigation indicated that emetine exerts its toxic effects by at least two modes of action: at high concentrations, a physical disruption of the plasma membrane of the cell; and, at lower concentrations, an intracellular inhibition of the process of forming high-energy phosphate compounds. The site of this inhibition is thought to occur at the enzymatic oxidation of substrates, other than succinate, mediated by nicotinamide-adenine dinucleotide (NAD).

EMETINE HYDROCHLORIDE is a compound which has classically been used in the treatment of amebic dysentery. Although its use for this purpose has been largely superceded by more effective and less toxic drugs, it is still used to treat certain types of amebiasis which are refractory to treatment by the therapeutic alternatives.

The predominance of toxicities resulting from emetine involve the cardiovascular system, and most investigators today agree that the typical cardiovascular symptoms of emetine toxicity are due to a direct myocardial depression. Attention has been called to the apparent hypersensitivity of the heart toward emetine (1).

Wenzel (2) has recently presented a relatively comprehensive review of the literature concerning emetine cardiotoxicity. Significant toxic mani-

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