Effect of Certain Drugs on Perfused Human Placenta VII. Serotonin *Versus* Angiotensin-II

By RONALD F. GAUTIERI and RALPH T. MANCINI*

Serotonin produced vasoconstriction in perfused human placental vessels primarily *via* stimulation of α-adrenergic receptors and secondarily *via* direct stimulation of the vascular smooth muscle, whereas angiotensin-II produced vasoconstriction primarily *via* direct stimulation of the vascular smooth muscle and secondarily *via* stimulation of α-adrenergic receptors. In this system, serotonin was shown to cause a wider variation in its pressor action than angiotensin-II.

PREVIOUS INVESTIGATIONS in this laboratory have shown that serotonin is the most powerful vasoconstrictor of human placental vessels (1–6) and that part of this action is due to α-adrenergic cell stimulation (4). Recently, in testing other known pressor agents in this preparation, it was observed that angiotensin-II produced vasoconstriction which frequently was comparable in degree to that produced by serotonin. Therefore, the purpose of the present investigation was to compare the vasoconstrictor activity of angiotensin-II and serotonin as well as to discern whether angiotensin-II, like serotonin, caused part of its vasoconstricting action *via* stimulation of α-adrenergic receptors.

MATERIALS AND METHODS

Full term human placentas, obtained from the hospital 15–20 min. after normal delivery, were used throughout this investigation. Each was transported to the laboratory in a glass container filled with 1 L. of Tyrode's solution maintained at 38°.

The apparatus employed and the procedures involved in the preparation and perfusion of the placentas, in recording and maintaining the perfusion pressure, and in measuring the inflow and outflow volumes of the perfusate have been described in previous investigations (3, 4). As in previous papers, umbilical cords of placentas measuring more than 5.5 cm. in length were designated as long cords; those under 5.5 cm. as short cords. In 56 successful placental preparations, the duration of each experiment was approximately 1 hr.

The dosage of the agonist serotonin chosen to obtain the desired pressor response was always 50 mcg., because this dosage was formerly shown to yield consistent results (3, 4). The same dosage was used for the agonist angiotensin-II in order to be able to compare its vasoconstricting properties with those of serotonin.

Astrom and Samelius (7) have reported that 1–3 mg. of phentolamine abolished the pressor response of serotonin. However, this laboratory has shown that 3 mg. of phentolamine only partially blocked the vasoconstricting action of serotonin (4). Subsequently, a dosage of 3 and 6 mg. of the antagonist, phentolamine, was utilized after each agonist not only to confirm our previous results with serotonin and observe the effects of the lower dosage of phentolamine upon the activity of angiotensin-II, but also to observe whether doubling the dosage could affect more profoundly the pressor action of either of the agonists.

The experimental regimen was divided into three parts which were: (*a*) obtaining a control response with either agonist, (*b*) administering one of the phentolamine doses, when the agonist's initial pressor action subsided, and (*c*) administering two additional doses of either agonist approximately 3 and 15 min. after the antagonist.

The following drugs were injected into the arterial side of the perfusion: 5-hydroxytryptamine creatinine sulfate (serotonin), 0.01%; angiotensin-II,[1] 0.04%; and phentolamine methanesulfonate,[2] 0.2%.

RESULTS AND DISCUSSION

The following results, which are summarized in Tables I and II, were obtained on full-term placental vasculature perfused at the pressure range of 50–90 mm. Hg which corresponded to inflow rates of 30–60 ml. of perfusion fluid (Tyrode's solution modified by the addition of 0.525% polyvinylpyrrolidone[3]) per minute.

In a total of 42 placental preparations, regardless of the cord length, phentolamine always inhibited the normal pressor action of serotonin, with the 6-mg. dose producing the greatest degree of serotonin antagonism (Table I, Figs. 1 and 2). Angiotensin-II produced increases in perfusion pressure that, based on the mean responses in short cord placental preparations, were not significantly different from those caused by serotonin (Tables I and II). Phentolamine always reduced the vasoconstriction produced by angiotensin-II, though the 6-mg. dose was not more effective in producing antagonism than the 3 mg. dose (Table II, Fig. 3). In addition, within 15 min. after the phentolamine treatment, serotonin and angiotensin-II again produced their normal responses (Figs. 1–3). Also, it can be observed from Tables I and II, that, of the two pressor agents utilized, serotonin caused the widest variation in its pressor action.

Angiotensin-II has been shown to constrict innervated blood vessels (aorta and carotid arteries of pigs) *via* the release of norepinephrine from its stores at sympathetic nerve endings (8). Also, angiotensin-II has been shown to constrict guinea pig intestine both indirectly by stimulating ganglion

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* Present address: Smith Kline & French Laboratories, Philadelphia, Pa.

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[1] Hypertensin. Valyl-5-angiotensin-II amide, lot No. B-5578, supplied through the courtesy of Ciba Pharmaceutical Co., Summit, N. J.

[2] Marketed as Regitine by Ciba Pharmaceutical Co., Summit, N. J.

[3] Marketed as Plasdone C by General Aniline and Film Corp.

TABLE I.—EFFECTS OF PHENTOLAMINE ON THE ACTION OF SEROTONIN IN THE PLACENTAL VASCULATURE

Drug and Dose	Cord Length[a]	No. Expt.	Pressure Changes, mm. Hg: Increase	Mean Change	S.E.[b]	*P* Value
Before phentolamine, 3 mg.	S	16	+4 to +108	+35	±7.8	...
Serotonin, 50 mcg.	L	15	+43 to +145	+83	±9.5	...
After phentolamine, 3 mg.	S	16	+1 to +93	+26	±8.1	.25
Serotonin, 50 mcg.	L	15	+10 to +165	+62[c]	±11.9	.1
Before phentolamine, 6 mg.	S	9	+13 to +58	+33	±5.6	...
Serotonin, 50 mcg.	L	2	+35 to +66	+50	±15.2	...
After phentolamine, 6 mg.	S	9	+1 to +36	+7[c]	±3.5	.005
Serotonin, 50 mcg.	L	2	+4 to +20	+12	±7.8	...

[a] S, short; L, long. [b] S.E., standard error. [c] Significant reduction in pressor response compared to control serotonin.

cells causing the release of acetylcholine and directly by an action on the intestinal smooth muscle (9). Recently, Vogin and Buckley (10), using a β-adrenergic receptor blocking agent to potentiate the activity of angiotensin-II, noted α receptor cell stimulation as another mechanism whereby angiotensin-II produced vasoconstriction. The present

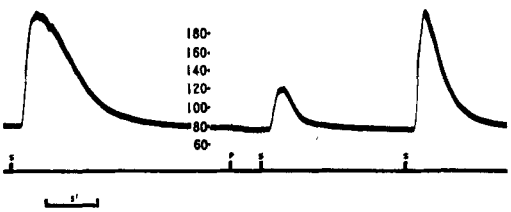

Fig. 1.—Effect of phentolamine on the action of serotonin in placental vasculature. Key: S, serotonin, 50 mcg.; P, phentolamine methanesulfonate, 3 mg.

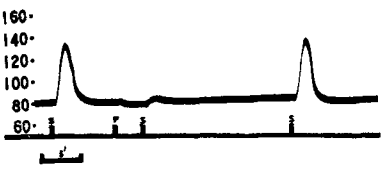

Fig. 2.—Effect of phentolamine on the action of serotonin in placental vasculature. Key: S, serotonin, 50 mcg.; P, phentolamine, 6 mg.

TABLE II.—EFFECTS OF PHENTOLAMINE ON THE ACTION OF ANGIOTENSIN-II IN THE PLACENTAL[a] VASCULATURE

Drug and Dose[b]	Pressure Changes, mm. Hg: Increase	Mean Change	S.E.[c]	*P* Value
Before phentolamine, 3 mg. Angiotensin, 50 mcg.	+21 to +51	+34	±4.2	...
After phentolamine, 3 mg. Angiotensin, 50 mcg.	+12 to +33	+23[d]	±2.4	.025
Before phentolamine, 6 mg. Angiotensin, 50 mcg.	+12 to +36	+25	±3.3	...
After phentolamine, 6 mg. Angiotensin, 50 mcg.	+11 to +30	+19[d]	±2.5	.1

[a] Short cord length in each case. [b] Seven experiments in each case. [c] S.E., standard error. [d] Significant reduction in pressor response compared to control angiotensin-II.

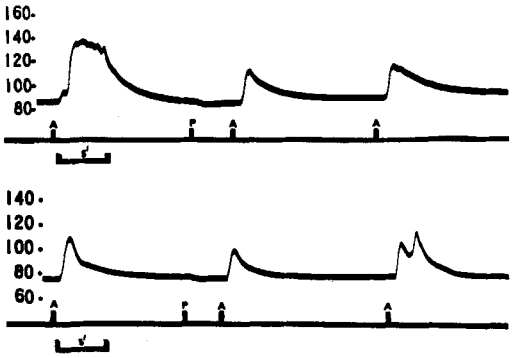

Fig. 3.—Effect of phentolamine on the action of angiotensin-II in placental vasculature. Key: A, angiotensin-II, 50 mcg.; P, phentolamine methanesulfonate. Top, 3 mg.; bottom, 6 mg.

investigation has confirmed the action of angiotensin-II on α adrenergic receptors in a noninnervated organ (11) by a different procedure—namely, the partial blockade of angiotensin-II's vasoconstrictor action by the use of the α-adrenergic receptor blocking agent, phentolamine.

Because phentolamine can diminish markedly the pressor action of serotonin, it follows that serotonin must produce most of its vasoconstriction, in these vessels, *via* stimulation of α receptors, with the remainder being due to direct stimulation of the vascular smooth muscle. On the other hand, angiotensin-II, whose vasoconstriction can be antagonized only partially by phentolamine, must produce most of its vasoconstriction, in these vessels, by a direct action on the vascular smooth muscle, with the remainder being due to stimulation of α receptors.

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