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Influence of Serotonin on Myocardial Blood Flow in the Presence and Absence of a Coronary Arterial Stenosis: Observations in Domestic Swine[1] (42682)

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Abstract. This study tested the hypothesis that 5-HT may impair coronary flow regulation by inappropriately increasing *arteriolar* tone in the coronary circulation. Ten closed chest, domestic swine were studied both in the presence and in the absence of a severe artificial intraluminal coronary stenosis. A 5-French micromanometer catheter with fluid lumen was placed in the left anterior descending coronary artery and used to record pressure and infuse 5-HT (40 and 100 μg/min) into the coronary circulation. For the stenosis phase of the protocol the catheter was embedded in the artificial stenosis. Hemodynamics, regional myocardial blood flow (microsphere technique), coronary vascular resistance, lactate consumption, and oxygen metabolism were measured at control and at 5 min of each 5-HT dose. In the absence of coronary artery stenosis (i.e., full vasodilatory reserve), there was no change in regional myocardial blood flow or coronary vascular resistance during 5-HT infusion. In the presence of a severe coronary stenosis (i.e., limited vasodilator reserve) 5-HT produced a significant ($P < 0.05$) decrease versus control in the distal left anterior descending:circumflex zone endocardial blood flow ratio (0.63 ± 0.19, mean ± 1 SD, to 0.55 ± 0.15) and a significant ($P < 0.05$) increase versus control in endocardial (50.6 ± 16.6 to 61.2 ± 19.8 mm Hg/ml/min/g) and transmural (49.9 ± 9.5 to 57.2 ± 12.8) coronary vascular resistance. Thus, 5-HT does not impair coronary flow regulation when full vasodilatory reserve is present. When coronary vasodilatory reserve is impaired by the presence of a severe proximal stenosis, 5-HT causes modest impairment of endocardial flow regulation.

Platelet aggregation and release of vasoactive compounds is known to occur at the site of atherosclerotic lesions in the human coronary circulation (1–4). Recent work from our laboratory has demonstrated that thromboxane-A2, released by aggregating platelets, can impair arteriolar vasodilation and worsen myocardial ischemia distal to a severe coronary arterial stenosis (5). Serotonin also is released by aggregating platelets and, therefore, may play a role in influencing arteriolar tone in vessels distal to the stenosis. Previous studies (6–8) have shown that the monoamine constricts large epicardial conductance vessels. Whether serotonin can compete with or inhibit appropriate regulation of myocardial blood flow particularly in the setting of a coronary arterial stenosis is not known. Accordingly, the present study was designed to test the hypothesis that infusion of serotonin into the coronary circulation of intact domestic swine may interfere with physiological regulation of regional myocardial blood flow by inducing constriction of vessels at the *arteriolar* level. In order to detect serotonin-induced changes in regional myocardial blood flow and coronary vascular resistance, heart rate and blood pressure were held constant while serotonin was selectively infused into the left anterior descending coronary artery of closed chest, sedated, domestic swine. The effect of serotonin was studied in the normal, unobstructed coronary circulation (full vasodilatory reserve) and in the presence of a severe proximal coronary stenosis (limited vasodilatory reserve). Measurements of regional myocardial blood flow, hemodynamics, oxygen metabolism, and lactate metabolism were made during each phase of the protocol.

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Materials and Methods. *Animal preparation.* Farm-bred pigs (N = 10, mean weight = 45.9 kg, range = 40.5 to 57.3) were premedicated with Ketamine (25 mg/kg) and then anesthetized with halothane (0.5–1.5%) and nitrous oxide. After this, each animal was systemically anticoagulated with heparin (225 IU/kg, intravenously). A double-lumen, 8-French catheter positioned in the arch of the aorta was used to monitor pressure and to obtain blood for determination of pH, PO_2, PCO_2, and regional myocardial blood flow. An 8-French angiographic catheter was passed in a retrograde manner under fluoroscopic guidance from the right femoral artery into the left atrium to allow administration of radioactive microspheres. A 7-French angiographic catheter, inserted into the right internal jugular vein, was advanced under fluoroscopic control to the proximal portion of the anterior interventricular vein to allow for selective blood sampling from myocardium perfused by the left anterior descending artery. A 7-French, bipolar pacing catheter inserted in the left internal jugular vein was positioned in the midcoronary sinus under fluoroscopic control to allow pacing at a constant rate (25 min^{-1} above resting rate) throughout the study. A 40-ml intraaortic balloon catheter was inserted into the left femoral artery, advanced to the descending aorta, and inflated as needed to reverse hypotension resulting from intracoronary serotonin infusion. A 7-French double-lumen catheter was inserted into the left femoral vein, advanced to the inferior vena cava, and used for administration of intravenous fluid and medications as required.

Each animal was used as its own control with respect to flow responses to intracoronary serotonin in the presence and absence of a stenosis. Accordingly, each animal was assigned to serotonin first with or without the stenosis by flip of a coin. If stenosis was first, a 7.5-mm-long, outer diameter 3.5 mm, inner diameter 0.625 mm, stenosis was placed in the proximal one-third of the left anterior descending coronary artery (9). The stenosis contained a second lumen into which the distal end of a 5-French Millar catheter with an infusion port had been attached before placement of the stenosis. The distal end of the catheter was open to the distal end of the stenosis and was used to infuse drugs into and record pressures from the distal arterial bed. After instrumentation, the first part of the study protocol (see below) was executed. Following this the stenosis was removed and the animal was reinstrumented with a 5-French Millar catheter (without the stenosis) positioned in the same approximate location as the previous stenosis. The protocol was then repeated. If no stenosis was first, the sequence was reversed.

After instrumentation was accomplished, all cutdown sites were closed and anesthesia was discontinued. Low-dose, constant intravenous infusion of sodium thiamylol was given throughout the study to ensure that the animal was comfortable and rested quietly. Although sedated, each animal breathed spontaneously, was awake, and had intact corneal reflexes. All animals remained intubated and were given supplemental oxygen (2–3 liters/min) during the study. Arterial blood gases were monitored frequently and remained at appropriate levels (pH 7.38–7.45; PCO_2 = 35–45; PO_2 = 100–125 mm Hg) throughout each experiment.

Study Protocol. *Intracoronary serotonin in the presence and absence of a severe coronary stenosis.* After the animal was stabilized for at least 30 min, Tris buffer (0.3 M, pH 7.4 at 37°C) was infused into the left anterior descending at 0.34 ml/min. At the end of 5 min, control measurements of hemodynamic parameters, regional myocardial blood flow, oxygen, and lactate metabolism were obtained (see below). Next, serotonin (5-hydroxytryptamine creatinine sulfate complex, Sigma Chemical Co., St. Louis, MO) made up in 0.3 M Tris buffer was infused at a rate of 40 μg/min (flow rate = 0.34 ml/min) for 5 min into the left anterior descending coronary artery. Repeat measurements of all experimental parameters were obtained at the end of the infusion. A second infusion of serotonin at 100 μg/min (flow rate = 0.34 ml/min) was then initiated, again for 5 min, and experimental parameters were remeasured at the end of that time. Systemic hypotension was induced by intracoronary setotonin in 35/40 trials. This effect was promptly reversed by inflation of the intraaortic balloon (average 10-ml volume) in each case. It should be noted that the form and dose range of serotonin used was similar to that employed by others (6, 10). After

completion of this phase of the study, the stenosis was either removed or inserted depending on the random order previously determined. The next phase of instrumentation was completed and the animal was allowed to stabilize for 15 to 20 min. At the end of this time a second control, again with a 5-min intracoronary infusion of Tris buffer, was documented. The protocol, as outlined above with intracoronary infusion of first 40 and then 100 μg/min of serotonin, was repeated.

After completion of the first sequence of interventions, approximately 300,000 radioactive microspheres (total activity ~ 1.4 μCi) were injected into the coronary circulation via the left anterior descending catheter to objectively label the myocardium that had been exposed to serotonin. Methylene blue was used in the same manner and for the same purpose after completion of the second sequence of interventions. Each animal was then given a large intravenous dose of sodium thiamylol (200–300 mg) and 3 to 5 min later a lethal dose of KCl was administered. The heart was then removed and sectioned for determination of microspheres activity.

Before removal of the heart, the position of the distal end of the anterior interventricular vein catheter was noted with respect to the presence of any venous tributaries draining toward it from myocardium proximal to the stenosis. The position of the left anterior descending catheter and gross appearance of the myocardium distal to the left anterior descending catheter were also noted.

Determination of regional myocardial blood flow and coronary vascular resistance. For each experimental condition approximately 4×10^6 radiolabeled microspheres (15 μDIA, 84–105 μCi total radioactivity) were injected via the left atrial catheter in order to determine regional myocardial blood flow (11). A different radioisotope was chosen at random for each flow determination. A complete description of microsphere methods employed in our laboratory has been published (9).

Coronary arteriolar resistance in endocardium distal to the stenosis was calculated by dividing distal coronary mean diastolic pressure minus mean left atrial pressure plus 7 mm Hg (5, 12) by distal zone endocardial blood flow. Distal zone epicardial and transmural resistances were calculated in the same fashion save for the fact that distal coronary mean pressure and distal zone epicardial and transmural flows, respectively, were employed in the computation. Resistances in the circumflex zone were calculated using aortic instead of distal coronary pressure.

Determination of regional myocardial oxygen metabolism. Paired samples (2–3 ml) of arterial and anterior interventricular venous blood were obtained for determination of oxygen content (Lex-O_2-CON instrument, Lexington Instruments, Waltham, MA) during each phase of the study. Oxygen content (vol%) was determined in duplicate for each sample and values were accepted only if the difference between them was ≤0.2 ml O_2/dl. Regional myocardial oxygen consumption (ml/min/100 g) was calculated as the product of transmural regional myocardial blood flow distal to the stenosis and the arterial–anterior interventricular venous oxygen difference.

Determination of regional lactate consumption. Lactate concentration in arterial and AIV blood was determined by a spectrophotometric method with commercially available kits (Calbiochem rapid lactate reagents, Calbiochem–Behring, La Jolla, CA). Samples of blood (5 ml) were immediately deproteinized by placing them in cold perchloric acid (8%, v/v). The samples were centrifuged and the supernatant was frozen for subsequent analysis in duplicate. Regional lactate consumption was calculated as the product of transmural regional myocardial blood flow distal to the stenosis and the arterial-AIV lactate difference.

Statistical methods. The significance of group mean changes (versus control) in hemodynamic parameters, regional myocardial blood flow, and metabolic parameters in response to drug infusion were assessed by means of a blocked one-way analysis of variance and Dunnett's test (13). Results were considered statistically significant when $P < 0.05$. All values are expressed as mean ± SD.

Results. *Hemodynamics. (A) Without stenosis.* Heart rate was controlled by atrial pacing and thus did not change significantly (versus control 111 ± 10) during serotonin infusion. Mean aortic pressure (mm Hg) also

was maintained at control levels (117 ± 12) by inflation of an intraaortic balloon during serotonin infusion, and so did not change versus control during drug infusion.

(B) With left anterior descending stenosis. Heart rate (111 ± 8) again was held constant by atrial pacing. Aortic pressures were maintained at control levels (116 ± 12) during the study. Diastolic pressure distal to the stenosis rose significantly versus control (27 ± 6 mm Hg) during both the 40 μg/min (33 ± 10 mm Hg, $P < 0.05$) and 100 μg/min (37 ± 11 mm Hg, $P < 0.01$) infusions. Mean pressure distal to the stenosis did not change significantly versus control (64 ± 11 mm Hg) during the 40 μg/min serotonin infusion but did rise significantly at the 100 μg/min dose (71 ± 10 mm Hg, $P < 0.05$).

Regional myocardial blood flow. (A) Without stenosis. Compared with control values, there was no significant change in the endocardial (1.50 ± 0.28), epicardial (1.24 ± 0.24), or transmural (1.39 ± 0.25) blood flow (ml/min/g) in the left anterior descending zone at the 40 or 100 μg/min serotonin doses. Regional myocardial blood flow of the circumflex zone (1.51 ± 0.26, 1.24 ± 0.28, 1.41 ± 0.28; endo, epi, transmural, respectively) remained stable during selective serotonin infusion into the left anterior descending. Accordingly, the ratio of left anterior descending to circumflex blood flow showed no significant change throughout this phase of the protocol.

(B) With left anterior descending stenosis. As expected, the control left anterior descending endocardial blood flow distal to the stenosis (0.93 ± 0.32) declined significantly compared to the control left anterior descending endocardial blood flow without a stenosis (1.50 ± 0.28, $P < 0.01$). Transmural and epicardial blood flow distal to the left anterior descending stenosis did not change significantly compared to control values (1.39 ± 0.25 and 1.50 ± 0.28, respectively) during serotonin infusion. There was a downward trend, compared to control (0.93 ± 0.32) in endocardial blood flow distal to the stenosis at both the 40 μg/min (0.85 ± 0.27) and 100 μg/min (0.89 ± 0.28) doses of serotonin. Although flow declined in 8/10 animals, the change was not statistically significant. However, the left anterior descending to circumflex zone endocardial blood flow ratio decreased significantly compared to control (0.63 ± 0.19) at the 100 μg/min (0.55 ± 0.15, $P < 0.05$) dose of serotonin. Transmural and epicardial left anterior descending to circumflex zone blood flow ratios did not change significantly compared to control. Finally, circumflex zone regional myocardial blood flow did not change versus control (1.53 ± 0.40, 1.26 ± 0.26, 1.43 ± 0.32; endo, epi, transmural, respectively) at any time during the study.

Coronary vascular resistance. (A) Without stenosis. There was no significant change compared to control values (70.0 ± 11.0, 86.9 ± 18.4, 77.0 ± 12.7; endo, epi, transmural, respectively) in either the left anterior descending zone or the circumflex zone coronary vascular resistance (mm Hg/ml/min/g) during infusion of serotonin. Resistances did not differ significantly between zones.

(B) With left anterior descending stenosis (Table I). The presence of a severe left anterior descending stenosis caused a significant ($P < 0.01$) decline in the control values of transmural, endocardial, and epicardial coronary vascular resistance compared to control values in the absence of the stenosis. There was a significant increase in the left anterior descending zone endocardial resistance compared to control (50.6 ± 16.6) at both the 40 μg/min (62.8 ± 20.2, $P < 0.05$) and 100 μg/min (61.2 ± 19.8, $P < 0.05$) doses of serotonin. The epicardial and transmural coronary vascular resistance increased in 8/10 animals at both doses of serotonin compared to control but only the transmural coronary vascular resistance at the 100 μg/min serotonin dose (57.2 ± 12.8, $P < 0.05$) was significantly higher than that of control (49.9 ± 9.5). It should be noted that distal zone resistance responses of animals in whom the stenosis was placed first were comparable to those in whom the stenosis was placed second. Circumflex zone resistance showed no significant change versus control during serotonin infusion.

Regional myocardial oxygen metabolism. (A) Without stenosis. There was no significant change versus control in aortic (13.1 ± 1.5 ml/dl) or AIV (3.0 ± 0.8) oxygen content during serotonin infusion. Since transmural regional myocardial blood flow also remained stable during serotonin infusion, there was no significant change versus con-

TABLE 1. CORONARY VASCULAR RESISTANCE (mm Hg/ml/min/g; MEAN ± 1 SD) RESPONSE TO INTRACORONARY SEROTONIN INFUSION: STENOSIS PRESENT

		Serotonin	
	Control	40 μg/min	100 μg/min
Distal zone			
Transmural	49.9 ± 9.5††	55.2 ± 15.5	57.2 ± 12.8*
Endocardial	50.6 ± 16.6†	62.8 ± 20.2*	61.2 ± 19.8*
Epicardial	46.6 ± 11.7††	49.5 ± 16.2	52.1 ± 13.5
Circumflex zone			
Transmural	77.6 ± 17.6	76.8 ± 18.6	73.2 ± 15.9
Endocardial	71.7 ± 17.4	71.9 ± 17.1	66.5 ± 14.4
Epicardial	87.7 ± 19.4	87.6 ± 21.6	84.6 ± 19.4

* $P < 0.05$ versus control.
† $P < 0.05$ versus circumflex zone at control.
†† $P < 0.01$ versus circumflex zone at control.

trol in regional myocardial oxygen consumption (14.3 ± 4.3 ml/min/100 g) during serotonin infusion.

(B) With left anterior descending stenosis. There was a small but significant fall in the oxygen content (ml/dl) of aortic blood compared to that of control (13.2 ± 1.05) during the 40 μg/min (12.5 ± 1.03, $P < 0.05$) serotonin infusion. Regional myocardial oxygen consumption (ml/min/100 g) also fell significantly ($P < 0.05$) compared to that of control (12.8 + 4.0) during serotonin infusion at 40 μg/min (11.7 ± 3.4). However, at the 100 μg/min dose regional myocardial oxygen consumption returned to a level (12.6 ± 3.2) which did not differ significantly from that of control.

Regional myocardial lactate metabolism. (A) Without stenosis. Arterial lactate content did not change compared to that of control (0.93 ± 0.45 m*M*) during serotonin infusion. The AIV lactate content (0.74 ± 0.34) also showed no significant change during serotonin infusion. Lactate consumption (regional transmural flow × arterial-AIV lactate concentration difference (mmole/min/100 g) showed no significant change compared to that of control (0.03 ± 0.02) during serotonin infusion.

(B) With left anterior descending stenosis. The presence of a severe left anterior descending stenosis changed regional lactate metabolism significantly. Lactate was consumed 0.03 ± 0.02 under control conditions in the absence of a stenosis. In the presence of a severe left anterior descending stenosis, AIV lactate rose and lactate was produced (−0.03 ± 0.06). Serotonin infusion did not change regional myocardial lactate metabolism significantly at either dose tested. Thus, arterial lactate (0.82 ± 0.31 m*M*), AIV lactate (1.12 ± 0.54), and overall lactate consumption (−0.03 ± 0.06) all remained unchanged compared to those of control during serotonin infusion.

Discussion. The purpose of this study was to test the hypothesis that serotonin may interfere with appropriate regulation of *arteriolar* tone and hence blood flow in the coronary bed. We tested this hypothesis in both the presence and the absence of full coronary vasodilatory reserve. The results of the study indicate that serotonin does not impair coronary flow regulation when full vasodilatory reserve is present. In contrast, when flow reserve is impaired by severe, proximal coronary arterial stenosis, stimulation with serotonin results in modest impairment of myocardial flow regulation particularly in endocardial layers of the heart. Interference with appropriate regulation of arteriolar tone in endocardium distal to the stenosis was manifested by a significant decrease in the distal:circumflex zone endocardial blood flow ratio, a significant increase in distal coronary diastolic pressure, and a significant increase in distal zone endocardial (and transmural) coronary vascular resistance.

The fact that lactate production did not increase (versus baseline) and regional myocardial oxygen consumption did not decline during serotonin infusion distal to the steno-

sis likely reflects the fact that its direct constrictor effect was modest and confined primarily to the endocardium. Accordingly, while serotonin may impair coronary flow regulation in the stenosis setting, its capacity to do so is limited. Indeed, the monoamine may contribute more in its role as a stimulator of platelet aggregation and release of other vasoactive products (e.g., thromboxane-A2) than as a direct arteriolar constrictor.

As noted above, serotonin causes platelet aggregation in many mammalian species including canines and humans. Accordingly, it is possible that platelet aggregation and release of other vasoactive compounds (e.g., thromboxane) could have contributed (in whole or part) to the modest coronary vasoconstriction observed during serotonin infusion in the present study. Several observations, however, argue against this hypothesis. None of the animals experienced complete occlusion of the stenosis as might be expected if intense platelet aggregation occurred on it. On gross examination, the stenosis was always widely patent. Second, cyclic decreases in distal coronary perfusion pressure were not observed as would be expected if platelet aggregates were forming on and then dislodging from the stenosis (14–16). Finally, whole blood platelet aggregation studies conducted in our laboratory (Appendix) demonstrate that serotonin is only a very weak stimulator of platelet aggregation and does not cause ATP release in domestic swine. Since this study was designed to assess the effects of serotonin on coronary arteriolar tone, the insensitivity of porcine platelets to the monoamine represents a distinct advantage of the model.

The potential role of serotonin in influencing coronary vascular tone has been the subject of several recent investigations (6, 7, 14, 15, 17, 18). In a study involving isolated, epicardial, canine coronary vessels Cohen *et al.* (17) demonstrated that the vascular response to serotonin depended importantly on the endothelial cell layer of the vessel. Intact coronary endothelium was associated with relaxation of the vessel in response to serotonin stimulation whereas contraction was elicited when the vessel was stimulated by serotonin in the absence of an intact endothelial cell layer. Others, however, have demonstrated constriction of large conductance vessels when the monoamine has been infused directly into either the coronary circulation (6) or the hind limb (10) of intact dogs. Unless one postulates that endothelial cell damage occurred in the experiments involving intact animals (6, 10), it is difficult to reconcile these results with the *in vitro* data of Cohen *et al.* (17).

The role of serotonin as a potential dilator or constrictor of precapillary *arteriolar resistance vessels* in the intact coronary circulation has been examined in only one previous study (6). The results of the investigation in dogs *without* coronary arterial stenosis were similar to those observed in domestic swine in the present study (i.e., no significant change in myocardial blood flow or calculated coronary vascular resistance in the absence of a coronary arterial stenosis). It is possible, however, that arteriolar vasoconstriction in response to serotonin administration might not be observed in animals with intact vasodilator reserve because of compensatory, metabolic vasodilation. Furthermore, the vasoconstrictor response to serotonin may be increased under ischemic conditions (19). Data obtained in the present study provide new information (vide infra) concerning both hypotheses.

The rigid intraluminal stenosis employed in our study substantially impaired coronary flow reserve and induced myocardial ischemia as evidenced by (i) reduction in myocardial blood flow distal to the stenosis and (ii) change from consumption to production of lactate in myocardium distal to the device. Under these circumstances (i.e., reduced flow with lactate production) stimulation with serotonin resulted in a small increase in coronary arteriolar resistance and a modest relative decline in blood flow in endocardial layers distal to the stenosis. Thus, under conditions of myocardial ischemia serotonin released from platelets aggregating at the site of a coronary arterial stenosis could contribute to reducing myocardial blood flow by increasing arteriolar tone distal to the stenosis. However, since the coronary arteriolar constrictor response to serotonin was modest, it is likely that local effects of serotonin at the stenosis site (e.g., spasm and/or augmentation of platelet aggregation and release) would contribute more to interference with

coronary flow regulation distal to the stenosis. Indeed, administration of ketanserin to dogs with external coronary constrictors improves coronary flow regulation by decreasing the frequency of cyclic coronary flow reductions (14, 15), an observation that supports the hypothesis that serotonin is more likely to interfere with coronary flow regulation primarily via effects at the stenosis site.

Several methodological issues bearing on the conclusions of the present study should be addressed. First, there is the question of damage to the endothelial lining of the stenosed coronary vessel. In an earlier study we demonstrated in our model that placement of the artificial stenosis does not result in damage to the endothelial lining of the vessel *downstream* of the stenosis (5). Accordingly, the data obtained in the present study are applicable to clinical conditions in which endothelial cell function is preserved at the arteriolar level of the coronary circulation. Second, there is the question of the appropriateness of the serotonin doses employed in the study. In an isolated canine coronary vessel system Lorentz and Vanhoutte (20) measured serotonin concentration in superfusate collected from vascular rings which had been exposed to aggregating platelets in concentrations of roughly 25×10^5 platelets/ml (i.e., 100-fold lower than the normal platelet concentration of human blood). Measured serotonin concentration in the emerging superfusate was on the order of 10^{-6} *M*. Thus, serotonin concentrations as high as 10^{-4} *M* conceivably could occur under clinical conditions. In our model system we infused a maximum serotonin dose of 100 μg/min into a coronary stream flowing at roughly 60 ml/min (40-g distal zone with flow of 1.5 ml/min/g) and so at most may have achieved blood serotonin concentrations on the order

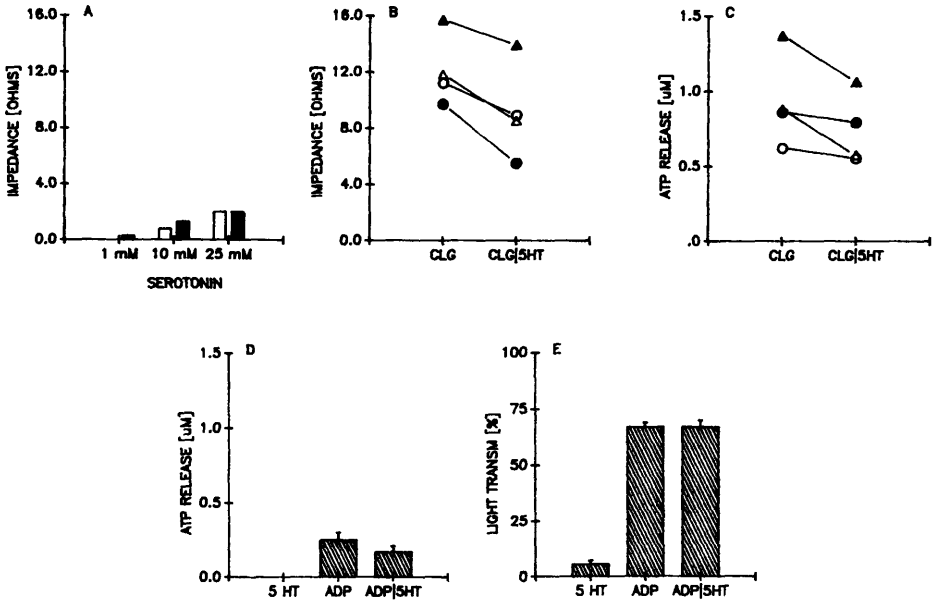

FIG. 1. (A) Platelet aggregation responses of two swine (open and filled bars) to serotonin. (B and C) Platelet aggregation and ATP release responses, respectively, of four additional swine whose platelets were stimulated with 1 μg/ml collagen (CLG) and with 1 μg/ml CLG plus 1 m*M* serotonin (5-HT). It is apparent that (i) in comparison with collagen, 5-HT alone does not provide a strong stimulus for porcine platelet aggregation, and (ii) 5-HT does not enhance and, indeed, may inhibit the release reaction seen during collagen stimulation. (D and E) Mean (+ SE) values for ATP release and aggregation, respectively, of another group of swine ($N = 8$) whose platelets were stimulated with 5-HT (40 μ*M*), ADP (5 μ*M*), and ADP (5 μ*M*) plus 5-HT (40 μ*M*). Note that (i) 5-HT is a much weaker platelet agonist than ADP and (ii) 5-HT does not potentiate either platelet aggregation or release in response to ADP. Platelet aggregation was measured by light transmission in this group of animals.

of 10^{-6} or 10^{-5} *M*. It is likely, therefore, that the doses of serotonin employed in the present study are physiologically relevant ones. Finally, computation of coronary vascular resistance required use of an assumed backpressure (i.e., measured left atrial pressure plus 7 mm Hg). Since distal coronary pressure remained relatively high (i.e., ~70 mm Hg) and constant during the study in relation to the assumed backpressure (i.e., ~10 mm Hg), the assumption employed is unlikely to have had an important influence on the results.

In conclusion, the present study demonstrates that (i) serotonin does not interfere with coronary flow regulation when coronary vasodilatory reserve is fully intact and (ii) under conditions of myocardial ischemia and very limited flow reserve, serotonin is a modest constrictor of coronary arterioles. Local effects of the monoamine at the site of coronary arterial stenosis, however, probably play a more important role in inhibiting appropriate regulation of myocardial blood flow in the stenosis setting.

Appendix. The aggregation responses of porcine platelets to serotonin (5-HT) were assessed by both electrical impedance aggregometry and by light transmission aggregometry. Release of ATP by platelets in response to agonist administration was measured by the Luciferin–Luciferase method (21). Impedance aggregometry studies were conducted with diluted whole blood as previously described (22). Light transmission aggregometry studies employed platelet-rich plasma with platelet-poor plasma used as a reference for assessing percentage change in light transmission in response to various agonists. Since each animal served as its own control with respect to platelet responses to serotonin versus other agonists such as ADP or collagen, no effort was made to standardize platelet concentration from animal to animal. Results of these studies are shown in Fig. 1.

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