Different Sensitivities of Vasoconstrictor Responses to Serotonin and KCl of Isolated and Perfused Dog Mesenteric Arteries with and without Endothelia

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Abstract—The stainless steel cannula inserting method was used to investigate the effects of serotonin on isolated and perfused dog mesenteric arteries with and without intraluminal saponin treatment. By intraluminal administration, serotonin and potassium chloride caused dose-related vasoconstrictions. After intraluminal treatment with 3 mg of saponin, the potassium chloride-induced vasoconstrictor response was significantly enhanced, whereas the serotonin-induced one was not potentiated but rather reduced slightly.

Although serotonin reaches all parts of the body because mammalian blood contains it, it has no recognized physiological role on blood vessel walls. Recently, we demonstrated that potassium chloride (KCl)-induced vasoconstriction was potentiated by removal of the endothelium in isolated and perfused arteries produced by intraluminal administration of saponin (1, 2). Moreover, CaCl_2 -induced vasoconstriction was also significantly enhanced after removal of the endothelium (2). On the other hand, phenylephrine-induced constriction was not potentiated by removal of the endothelium (2). In isolated arterial ring preparations, it was reported that serotonin-induced constriction was potentiated by removal of the endothelium (3, 4). Thus, they considered serotonin may contribute to certain pathological conditions by inducing strong vasoconstriction after the damage of the endothelium. In the present study using isolated and perfused mesenteric arteries which was developed by Hongo and Chiba (5) and modified by Tsuji and Chiba (6), we investigated whether the constrictor responses to serotonin were different with and without the endothelium in the same arterial vasculature.

Six mongrel dogs of either sex, weighing

8–18 kg, were anesthetized with sodium pentobarbital (30 mg/kg, i.v.), and the dogs were sacrificed by rapid exsanguination from the right common carotid artery. Arteries (which supplied the large middle portion of the small intestine) which are median branches of the cranial mesenteric artery were carefully isolated. Isolated arteries selected for study were 10–15 mm in length and 0.8–1.3 mm in outer diameter, and they were cannulated as described previously (5, 6). The isolated, cannulated artery was placed in a bath maintained at 37°C and perfused with Krebs solution by means of a peristaltic pump. The perfusion solution was bubbled with 95% O_2 and 5% CO_2 which maintained the pH between 7.2–7.4. The flow rate was initially adjusted so that the perfusion pressure was between 50–100 mmHg; subsequently, it was kept constant throughout the experiment (0.5–2 ml/ml). The vasoconstrictor response was, therefore, observed as an increase in perfusion pressure. Drugs used were saponin (Kanto Chem. Co.), serotonin creatinine sulfate (5-hydroxy-tryptamine, Sandoz), and potassium chloride. The drug solution was administered into the rubber tubing close to the cannula in a volume of 0.01–0.03 ml by use of a microinjector (Terumo Co.). The data

are presented as the mean $\pm$ S.E.M. in the text and illustrations.

Intraluminal injection of serotonin and KCl immediately induced transient vasoconstriction in a dose-related manner as reported previously (7, 8). After intraluminal treatment with 3 mg of saponin, serotonin-induced constriction was slightly suppressed. On the other hand, the KCl-induced constriction was apparently potentiated in the same pre-

parations. Figure 1 shows a typical experiment of vasoconstrictor responses to serotonin and KCl before and after intraluminal treatment with 3 mg of saponin in the same arterial preparation. As shown in Fig. 1, the vasoconstrictor response to 1 mg of KCl was markedly enhanced from 20 to 150 mmHg in a maximum increase, but that to 0.1 μ g of serotonin was rather depressed from 200 to 160 mmHg. Summarized data are shown in Fig. 2.

Previously, the cannula inserting method for isolated vessels was developed and modified (5, 6). By use of this method, various regions of arterial vessels were perfused, and effects of serotonin were examined. Serotonin usually caused a transient constriction in a dose-related manner, although the potencies were different in different regional vessels (5–10). More recently, we demonstrated that a bolus of intraluminal saponin (1–3 mg) readily caused removal of the endothelium in the isolated arterial vessels (1, 2). After removal of the endothelium, vasoconstrictor responses to CaCl_2 and KCl were significantly enhanced

Fig. 1. Vasoconstrictor responses to 0.1 μ g of serotonin (5-HT) and 1 mg of potassium chloride (KCl) in an isolated and perfused dog mesenteric arterial preparation before and after treatment with 3 mg of saponin.

Fig. 2. Dose-response curves of vasoconstrictor responses for serotonin (5-HT) and potassium chloride (KCl) before and after treatment with 3 mg of intraluminal saponin. Each point is the average. Vertical bars signify $\pm$ S.E.M. The number for each point indicates the number of preparations perfused.

in the same preparations (1, 2), suggesting that calcium ions may readily enter into the vascular smooth muscle cell in the absence of the endothelium. On the other hand, phenylephrine-induced vasoconstriction was not potentiated by removal of the endothelium (2). In the present study, serotonin-induced constriction was not potentiated, although the KCl-induced one was obviously enhanced by treatment with intraluminal saponin. Cocks and Angus (3) reported that removal of the endothelium from both dog and pig coronary artery rings shifted the concentration-contraction curves to serotonin and norepinephrine to the left and increased their respective maxima compared with those of rings in which the endothelium remained intact. They also reported that the concentration-contraction curves for KCl were decreased by the removal of the endothelium. They considered that serotonin released a vasodilator substance from endothelial cells. Cohen et al. (4) also reported that serotonin-induced contractions were larger in the absence of the endothelium in isolated canine coronary artery rings, whereas those caused by phenylephrine and KCl were not. They considered that the vasodilator response to serotonin mediated by the endothelium was initiated at serotonergic receptors on endothelial cells. Thus, it is postulated that serotonin-induced constriction may be enhanced by disappearance of endothelium-dependent relaxations after removal of the endothelium. However, they could not explain the reason for decreasing constriction for KCl by the removal of the endothelium. In the present experiments, our results were different from those. We considered the reasons of the differences from previous reports (3, 4). Since Cocks and Angus and Cohen et al. (3, 4) used the coronary arteries from greyhound, mongrel dogs and pigs, it is not ruled out that different kinds of arteries cause different results in addition to different methods for making the isolated arterial preparations. We need to confirm the effects of serotonin on the coronary arterial preparation by the present method in the future. As KCl- or CaCl_2 -induced constrictions were potentiated in the absence of the endothelium in perfused

preparations (1, 2), we consider that serotonin-induced constriction may be due to an increase in the release of intracellular Ca ions from intracellular stores, but not due to an increase in entry of Ca ions from the extracellular space, and a relatively large amount of saponin may exert its action directly on vascular smooth muscles which in turn causes slight suppression of the serotonin-induced constriction.

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