Effect of pH Alterations on the Vascular Reactivity of Dog Isolated Perfused Mesenteric Arteries

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It is well recognized that the pH of the perfusing fluid is one of the important factors involved in the control of vascular reactivity. Respiratory acidosis diminishes the effect of vasoconstrictor substances on blood vessels (1). Rogers et al. (2) demonstrated that increasing pH potentiated the increase in perfusion pressure to administered catecholamines in isolated perfused mesenteric arteries. Recently, a new model of an isolated arterial preparation was developed by Hongo and Chiba (3) and modified by Tsuji and Chiba (4). By using this method, we have attempted to evaluate responses of dog isolated mesenteric arteries to several vasoconstrictor substances at low, normal and high pH levels of perfusing fluid.

Twelve mongrel dogs weighing 9–16 kg were anesthetized with sodium pentobarbital (30 mg/kg, i.v.) and sacrificed by rapid exanguination, after treatment with sodium heparin (500 units/kg, i.v.). The mesenteric artery was dissected and carefully removed. Isolated mesenteric arteries selected for study were 10–15 mm in length and 0.5–1.0 mm in outer diameter, and they were cannulated as described previously (3, 4). Briefly, a steel cannula (21–27 gauge, 0.40–0.83 mm in outer diameter and 3 cm in length) was prepared with one end sealed. Near the sealed end of the cannula, 1 to 3 small holes were opened on the wall of the cannula. The steel cannula was inserted into each artery so that the small holes were covered with the proximal end of the artery, and this proximal end was fixed to the cannula by a thin thread. The isolated, cannulated artery was placed in a bath, which was maintained at constant temperature of 37°C, and was perfused with mammalian Ringer’s solution ( Constituents of which were: 9.0 g NaCl, 0.42 g KCl, 0.24 g CaCl₂, 0.3 g NaHCO₃ per 1000 ml distilled water) by means of a peristaltic pump. The perfusing solution was perfused with or without altered bubbling of 95% O₂ and 5% CO₂ mixed gas for changing the pH levels of the perfusing fluid. The pH of the perfusing solution was measured using a Corning pH/blood analyzer (Model 165/2). With bubbling, the pH changed to 6.6–7.8, depending on the bubbling rate. The pH values were measured during each experiment. The flow rate was initially adjusted so that the perfusion pressure was 50–60 mm Hg, and then it was kept constant throughout the experiment (0.5–1.5 ml/min). The constrictor response was, therefore, observed as an increase in perfusion pressure. Drugs used were serotonin creatinine sulfate (5HT, Sigma), dl-norepinephrine hydrochloride (NE, Sankyo), prostaglandin F₂α (PGF₂α, Ono), disodium adenosine triphosphate (ATP, Kowa) and potassium chloride (KCl). The drug solution was administered into the rubber tubing close to the cannula in a volume of 0.01–0.03 ml.

At low pH levels of perfusing fluid, norepinephrine was injected into the cannulated arterial preparation, and vasoconstrictor responses were induced in a dose-related manner. The threshold dose for inducing constriction was approximately 0.1 μg. At 3 μg, norepinephrine usually induced an increase in perfusion pressure of over 200 mm Hg. 5HT increased perfusion pressure, but it usually never exceeded 50 mm Hg even at larger doses of 10 to 100 μg. PGF₂α also produced slight vasoconstriction at more than 30 μg. Histamine induced slight vaso-
constriction. ATP and KCl induced marked vasoconstriction at much higher doses. Summarized data are shown in Fig. 1A.

At high pH levels, norepinephrine induced marked vasoconstriction in a dose-related manner. The threshold dose for inducing vasoconstriction was approximately 0.1 μg. It seems that the norepinephrine-induced vasoconstriction was slightly potentiated. On the other hand, 5HT-induced constriction was markedly potentiated in all the preparations examined. The threshold dose for inducing vasoconstriction was approximately 0.03 μg. With increasing doses, 5HT usually induced greater increases in perfusion pressure of over 100 mm Hg. The effects of 5HT were roughly 10 times more potent than that of norepinephrine. PGF₂α also induced marked vasoconstriction at a dose of over 3 μg. Histamine induced almost the same degree of vasoreactivity as observed at low pH values. Both ATP and KCl had a similar grade of potency an efficacy compared with those at the low pH levels.

At a pH range of 7.2–7.4, effects of norepinephrine, ATP, histamine and KCl were similar to those at pH 6.6–7.1 or pH 7.5–7.8. Effects of 5HT were slightly smaller than those at high pH level. Effects of PGF₂α were smaller than those at high pH level.

In this study, the effects of 6 vasoactive substances were investigated at low, normal and high pH levels of perfused fluid. Bygdeman (1) reported an increased effect of norepinephrine on muscle vascular bed during alkalosis (7.80; range, 7.71–8.00). Rogers et al. (2) also reported that increasing the pH (6.6–8.1) of the vessel bath increased arterial smooth muscle tone and potentiated the pressor responses to injected catecholamine and to nerve stimulation. In the present study, we could not confirm a significant potentiation of norepinephrine-induced vasoconstriction by increasing the pH. On the other hand, 5HT induced strong vasoconstriction at high pH levels such as 7.5–7.8. The potency of 5HT at high pH levels was approximately 10 times greater than that of norepinephrine. In the mammalian mesenteric arteries, relative unresponsiveness to 5HT has been well known in isolated and in situ arterial preparations (5–7). Even in this study, at low pH levels, 5HT did not cause clear vasoconstriction in all examined preparations as shown in Fig. 1A. However, as shown in Figs. 1B and 1C, 5HT produced an increase in perfusion pressure in the condition of normal and high pH levels of perfusing fluid. PGF₂α was also markedly potentiated at normal and high pH levels. On the contrary, norepinephrine, ATP, KCl and histamine induced almost the same response patterns at both low and high levels of pH.

Halpern et al. (8) reported that respiratory

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**Fig. 1.** (A) Dose-response curves of vasoconstriction to 6 vasoactive substances in the isolated dog mesenteric artery under the condition where the pH of the perfusing fluid is 6.6–7.1. (B) Dose-response curves at 7.2–7.4 pH levels. (C) Dose-response curves at 7.5–7.8 pH levels. NE, norepinephrine; H, histamine.
acidosis (pH 6.8) decreased the contraction induced by histamine in the guinea-pig isolated ileum and by acetylcholine in the uterus from cat, rabbit and guinea-pig, but the effect of other stimulating substances tested was not affected. However, Bygdeman (1) did not give evidence for a decrease sensitivity of only one type of specific receptor since he used norepinephrine and angiotensin which were influenced to about the same extent. In the present study, we demonstrated that the actions of specific vasoactive substances might be influenced by changing the pH levels in a relatively large artery. Further studies are needed to determine whether the PO2 content in perfusing fluid has an important role in the appearance of effects of a specific vasoactive substance.

References