Adrenergic Stimulation–Released Histamine Taken-up in Adrenergic Nerves Induces Endothelium-Dependent Vasodilation in Rat Mesenteric Resistance Arteries

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Abstract. The present study investigated whether histamine was taken up by perivascular adrenergic nerves and released by periarterial nerve stimulation (PNS) to induce vascular responses. In rat mesenteric vascular beds treated with capsaicin to eliminate calcitonin gene–related peptide (CGRP)ergic vasodilation and with active tone, PNS (1 – 4 Hz) induced only adrenergic nerve–mediated vasoconstriction. Histamine treatment for 20 min induced PNS-induced vasoconstriction followed by vasodilation without affecting CGRP-induced vasodilation. Chlorpheniramine, guanethidine, combination of histamine and desipramine, and endothelium-removal abolished PNS-induced vasodilation in histamine-treated preparations. These results suggest that histamine taken up by and released from adrenergic nerves by PNS causes endothelium-dependent vasodilation in rat mesenteric arteries.

Keywords: histamine, perivascular adrenergic nerve, rat mesenteric artery
Vasoconstrictor activity (transient maximum value) was pressure at maximum relaxation induced by papaverine. Values) was expressed as a percentage of the perfusion relaxation. Vasodilator activity (maximum vasodilation with histamine for 20 min. Then, the preparations were with normal Krebs solution for 30 min; and perfused previously (9, 10) to remove vascular endothelium; washed with deoxycholate (1.8 mg/mL) for 30 s as described previously. To produce active tone and elevate perfusion pressure, mesenteric vascular beds were perfused with methoxamine (α1-adrenoceptor agonist) and PNS was applied as a control (S1). To observe vascular reactivity, a bolus of CGRP (50 pmol, 100 μL/12 s) as a control (I1) was directly injected into the perfusate proximal to the arterial cannula with an infusion pump (model 975; Harvard Apparatus, Inc., Holliston, MA, USA). Thereafter, Krebs solution containing methoxamine was switched to normal Krebs solution to return to basal perfusion levels. Then, the preparation was perfused with Krebs solution containing histamine (100 μM) for 20 min and washed by normal Krebs solution for 30 min. Thereafter, the preparation was re-perfused with Krebs solution containing methoxamine to elevate perfusion pressure, and then PNS and CGRP injection were performed.

To assess the effect of histamine treatment, preparations treated with histamine was perfused with Krebs solution containing methoxamine and chlorpheniramine (histamine H1-receptor antagonist, 1 μM) or guanethidine (adrenergic neuron blocker, 5 μM) and then subjected to PNS and CGRP injection. In experiments using desipramine (selective catecholamine-transporter inhibitor, 0.1 μM), perfusion of desipramine was started 10 min before the combined perfusion of histamine and desipramine for 20 min, and then perfusion of desipramine alone was continued for 10 min during the washing-out period. Thereafter, the preparation was perfused with Krebs solution containing methoxamine and then subjected to PNS and CGRP injection. Preparations with resting tension were perfused with saline solution containing sodium deoxycholate (1.8 mg/mL) for 30 s as described previously (9, 10) to remove vascular endothelium; washed with normal Krebs solution for 30 min; and perfused with histamine for 20 min. Then, the preparations were perfused with Krebs solution containing 2 μM methoxamine and subjected to PNS and CGRP injection.

At the end of each experiment, preparations were perfused with 100 μM papaverine to produce complete relaxation. Vasodilator activity (maximum vasodilation values) was expressed as a percentage of the perfusion pressure at maximum relaxation induced by papaverine. Vasoconstrictor activity (transient maximum value) was expressed as a percentage of the perfusion pressure before PNS. Experimental results are expressed as the mean ± S.E.M. The statistical analysis was performed using Student’s paired t-test. A P < 0.05 was considered statistically significant.

The following drugs were used: acetylcholine (ACh) chloride (Daiichi-Sankyo Pharmaceutical, Tokyo), rat CGRP (Peptide Institute, Osaka), and papaverine hydrochloride (Dainippon-Sumitomo Pharmaceutical, Osaka). Capsaicin, chlorpheniramine hydrochloride, methoxamine hydrochloride, guanethidine sulphate, histamine dihydrochloride, sodium deoxycholate, and desipramine hydrochloride were purchased from Sigma Aldrich Japan (Tokyo).

In the preparation with endothelium and with active tone, ACh injection produced a transient decrease in perfusion pressure due to endothelium-dependent vasodilation (Fig. 1A). As shown in Fig. 1A, the first series of PNS caused a transient increase in perfusion pressure due to vasoconstriction followed by secondary sustained vasoconstriction, which gradually returned to pre-stimulation levels without vasodilation. The second series of PNS induced reproducible vasoconstrictor responses, which were slightly greater, but not significantly, than those to the first PNS except for 2 Hz (Figs. 1B and 2A). The first and second series of CGRP injections induced reproducible long-lasting vasodilations (Figs. 1A, 1B, and 2B).

As shown in Fig. 1E, in the preparation treated with histamine, PNS induced initial vasoconstrctor responses similar to non-treated preparations (Figs. 1E and 2C), while PNS-induced sustained vasoconstrictions almost disappeared (Fig. 1E). Additionally, PNS (2 and 4 Hz) induced a frequency-dependent vasodilation after vasoconstriction (Figs. 1E and 2D). However, histamine treatment did not affect CGRP-induced vasodilation (Figs. 1E and 2D).

In preparations treated with histamine, guanethidine (5 μM) abolished both vasoconstrictor and vasodilator responses to PNS (Figs. 1G, 2E, and 2F). Guanethidine did not affect CGRP-induced vasodilation (Fig. 2F).

In preparations treated with histamine, PNS in the presence of chlorpheniramine (1 μM) caused only a vasoconstrictor response without producing a vasodilator response (Fig. 3: A and B). However, chlorpheniramine had no effect on the vasoconstrictor response to PNS (Fig. 3A) and CGRP-induced vasodilation (Fig. 3B).

In preparations treated with histamine in the presence of desipramine (0.1 μM), PNS caused only vasoconstrictor responses similar to control responses before the combined treatments (Fig. 3C). The combined treatment did not affect PNS- and CGRP-induced vasodilation (Fig. 3D).
Histamine Uptake Into Adrenergic Nerves

Fig. 1. Typical recordings showing vascular responses to periarterial nerve stimulation (PNS; 1, 2, and 4 Hz; closed triangles) before and after histamine treatment in perfused rat mesenteric vascular beds with endothelium and with active tone produced by methoxamine. A, B, C, and F: Control responses without treatment with histamine. D: Treatment with histamine. E: Responses after histamine treatment. G: Responses in the presence of guanethidine. S1 and S2: responses to the first and second PNS, respectively. I1 and I2: responses to the first and second CGRP injection (50 pmol), respectively. ACh: the injection of acetylcholine (1 nmol, closed circle). PPV: the perfusion of papaverine.
In preparations without endothelium, ACh injection did not cause vasodilation. In denuded preparations pretreated with histamine, PNS at 2 Hz caused an augmented vasoconstrictor response (Fig. 3E), and a significant vasodilator response to PNS at 4 Hz was observed (Fig. 3F). Endothelium-removal had no effect on CGRP-induced vasodilation (Fig. 3F).

In rat perfused mesenteric vascular beds with active tone, PNS has been shown to produce an initial vasoconstriction followed by a long-lasting vasodilation (11). Since guanethidine abolished the PNS-induced vasoconstriction (Fig. 1G) (10, 11), perivascular adrenergic nerves mediate the response. By contrast, perivascular CGRPergic nerves mediate the PNS-induced vasodilation, since capsaicin (Fig. 1A) and CGRP8-37 (CGRP-receptor antagonist) abolished the response (5, 12). Therefore, in capsaicin-treated mesenteric vascular beds, PNS induces adrenergic nerve-mediated vasoconstriction without producing CGRPergic nerve-mediated vasodilation.

The present study is the first to demonstrate that when mesenteric vascular beds without CGRPergic nerve

Fig. 2. Bar graphs showing changes in PNS (1, 2, and, 4 Hz)- and CGRP (50 pmol)-induced responses without and with temporary histamine treatment and effect of guanethidine in perfused rat mesenteric vascular beds with endothelium and with active tone produced by methoxamine. A and B: Responses without histamine treatment. C and D: Responses after histamine treatment. E and F: Effect of guanethidine on responses after histamine treatment. Panels A, C, and E indicate vasoconstrictor responses. Panels B, D, and F indicate vasodilator responses. *P < 0.05, **P < 0.01 vs. the first PNS.
function were temporarily treated with histamine, the PNS-induced vasodilation appeared after vasoconstriction even in the absence of histamine. However, the vasodilator response to CGRP injection was not affected by histamine pretreatment, suggesting that histamine pretreatment has no effect on vascular reactivity. Since CGRPergic nerve function was eliminated by capsaicin, the PNS-induced vasodilation observed after histamine treatment is unlikely to be mediated by CGRPergic nerves. Furthermore, in preparations treated with histamine, PNS did not induce vasodilation after vasoconstriction in the presence of chlorpheniramine, suggesting that histamine is involved in the PNS-induced vasodilation observed after histamine treatment. Additionally, guanethidine abolished vasodilator responses to PNS observed after histamine pretreatment. Therefore, PNS-induced vasodilation after histamine treatment is likely to be mediated by histamine, which is released by stimulation of perivascular adrenergic nerves.

In the present study, combined treatment with histamine and desipramine abolished the PNS-induced vasodilation. Therefore, it is likely that histamine is taken up
by and stored in perivascular adrenergic nerves via catecholamine transporters. Since PNS-induced vasodilation observed after temporary histamine treatment was abolished by guanethidine, it appears that stored histamine is neuronally released from adrenergic nerves to induce vasodilation.

The present study demonstrated that endothelium-removal markedly inhibited PNS-induced vasodilation after temporary treatment with histamine. Histamine has been shown to induce endothelium-dependent vasodilation, which is mediated by histamine H₁ receptors located in endothelium (1). Thus, it is assumed that neuronally released histamine reaches the endothelium and acts on histamine H₁ receptors, resulting in endothelium-dependent vasodilation.

In conclusion, the present findings suggest that histamine is taken up by and accumulated in perivascular adrenergic nerves via catecholamine transporters in rat mesenteric arteries, and released by nerve stimulation, resulting in endothelium-dependent vasodilation through mainly the activation of histamine H₁ receptors.

References


