Responses to seven vasorelaxant neuropeptides in small renal arteries isolated from stroke-prone spontaneously hypertensive rats

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ABSTRACT

Relaxation responses to seven vasodilator neuropeptides were examined in 3rd order branches of renal arteries isolated from 6-7-month-old SHRSP and age-matched WKY. Calcitonin gene-related peptide (CGRP) induced markedly increased endothelium-independent relaxation in SHRSP rats compared to that in WKY rats. Relaxations generated by atrial natriuretic peptide, brain natriuretic peptide, vasoactive intestinal peptide, peptide histidine isoleucine were all endothelium-independent, while bradykinin-induced relaxation was dependent on the presence of an intact endothelium. There were no significant differences between specimens from SHRSP and WKY in their relaxations. Substance P caused almost no relaxation in these arteries. Thus, enhanced reactivity to CGRP appears to be a specific change in SHRSP.

Key words : SHRSP, renal artery, calcitonin gene-related peptide, vasodilator peptide, endothelium

Introduction

Several vasoactive peptides, such as calcitonin gene-related peptide (CGRP), vasoactive intestinal peptide (VIP), atrial natriuretic peptide (ANP), bradykinin (BK), substance P (SP), are all known as vasorelaxants.[1-5] Immunohistochemical studies have demonstrated the presence of various vasoactive neuropeptides in perivascular nerves.[6,7] All of this evidence indicates the participation of vasoactive neuropeptides in the regulation of systemic or local circulation.

We previously reported that the relaxation response to CGRP was markedly increased in isolated small renal arteries from stroke-prone spontaneously hypertensive rats (SHRSP).[8] However, whether this hyperreactivity to CGRP shown in SHRSP is a specific change remains unclear. The present study investigated the relaxation response of VIP, ANP, brain natriuretic peptide (BNP), peptide histidine isoleucine (PHI), BK and SP, in comparison with that of CGRP.

Methods

Male 6-7-month-old SHRSP rats and age-matched Wistar-Kyoto rats (WKY) were used. The methods were described in our previous report.[8] Breifly, after being anesthetized with sodium pentobarbitol (40 mg/kg, i.p.), the rats were exanguinated from the abdominal aorta, and the kidneys were obtained. The 3rd order branches of the renal arteries with unstretched luminal diameters in the range of 0.1-0.2 mm were dissected from within the kidney under a dissecting microscope. Ring segments approximatly 1 mm long were mounted horizontally on two L-shaped tungsten wires (50 μm in diameter) in a microvascular chamber[9] filled with 2 ml physiological salt solution, maintained at 37℃, and bubbled with carbogen. To remove the endothelium, the lumen surface of the segments was gently rubbed with a stainless steel rod. Successful removal of the endothelium was confirmed by the absence of acetylcholine (10^{-6} M)-induced relaxation. The preparations were placed under a resting tension of 200 mg and allowed to equilibrate for at least 90 minutes. The responses were recorded on a myograph. The segments were constricted to 80% of their maximal tension with noradrenaline (6×10^{-7} M). Dilator responses to these peptides were tested when the precontraction stablized. Relaxation responses were expressed as a percentage of the maximal relaxation generated by papaverine (10^{-4} M).

Results and discussion

In this study, we examined the relaxation responses to ANP, BNP, VIP, PHI, BK and SP, in comparison with that of CGRP in small renal arteries from SHRSP and WKY. ANP, BNP, VIP, PHI and BK all caused relaxations of these arteries, although magnitudes of responses varied. On the contrary, SP caused almost no relaxation. Fig. 1 shows the magnitudes of relaxation in response to these peptides at a concentration of 10^{-7} M in endothelium-intact preparations. There were no significant differences between SHRSP and WKY in the responses to these peptides except for that to CGRP. Endothelium removal did not affect relaxation actions of ANP, BNP, VIP, PHI as well as CGRP, while it nearly abolished that of BK.

ANP, an important hormone controling the homeostasis of sodium and water, possesses potent vasorelaxant effects especially on renal vasculature.[5,10-13] BNP, another cardiac hormone, also relaxes smooth muscles,[14] and BNP-containing nerve fibers have been found along the renal arteries

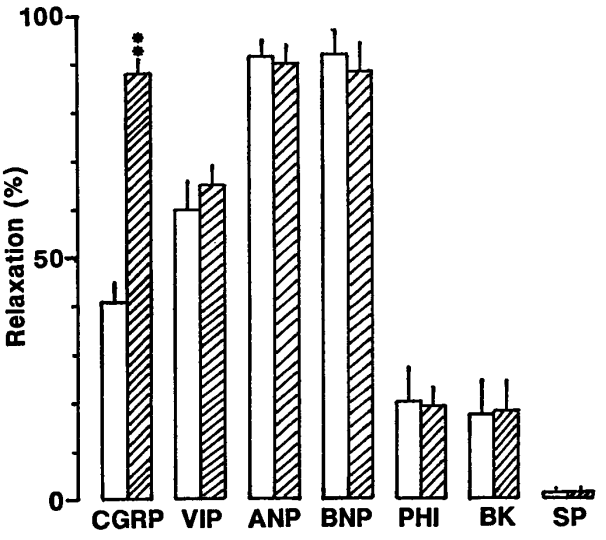

Fig. 1 Relaxant effects of seven vasorelaxant neuropeptides(10^{-7} M) on endothelium-intact small renal arteries from WKY (open columns) and SHRSP (shaded columns). Each column is the mean of 4-7 experiments. Vertical lines represent the S.E.M. **$p<0.01$, compared with WKY (t-test).

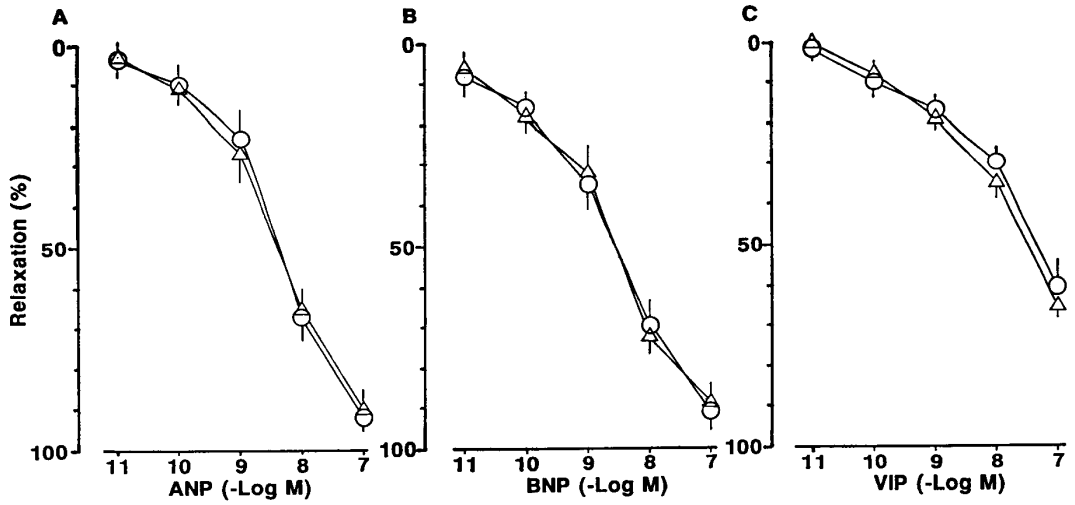

Fig. 2 Concentration-response curves for ANP (A), BNP (B), and VIP (C) on endothelium-intact segments of small renal arteries from WKY (circles) and SHRSP (triangles). Each point is the mean of 4-6 experiments. Vertical lines represent the S. E. M.

within the kidney.[15] In the present experiment, we found that both ANP and BNP dose-dependently relaxed precontracted rings of small renal arteries (Fig. 2A, B) in an endothelium-independent manner. However, there were no significant differences between SHRSP and WKY in their relaxations.

There is evidence that VIP-containing nerve fibers exist in blood vessels, and that VIP has vasodilator actions.[2,3,16,17] In this study, we demonstrated that VIP was also a potent dilator of the small renal arteries of rats (Fig. 2C), and this effect was not affected by endothelium removal. PHI, which coexists with VIP in parasympathetic nerve endings,[18] also dilated the arteries, but the magnitude of relaxation was smaller than that of VIP. There were no significant differences between the two groups in relaxation responses induced by VIP and PHI.

BK is known as an endothelium-dependent vasodilator.[4] We also found that its action was dependent on the existence of intact endothelium in the small renal arteries of rats. SP, the peptide that coexists with CGRP in sensory nerve endings,[19] caused almost no relaxation of the arteries. This is in accordance with the finding of Mulderry et al[20] that nerve fibers containing SP were sparse or completely absent from the renal arteries of rats.

In summary, the present study demonstrated that among the seven vasorelaxant neuropeptides tested, significant difference between SHRSP and WKY was found only in the relaxations induced by CGRP. Thus. the hyperreactivity to CGRP seems to be a specific change in SHRSP rats. We hypothesize that this enhancement is the result of up-regulation of CGRP receptors in vascular smooth muscles, but further investigation is needed.

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