CONTRIBUTION OF VASOPRESSIN TO HYPERTENSION CAUSED BY BARORECEPTOR DENERVATION AND NUCLEUS TRACTUS SOLITARIUS LESIONS IN RATS

TAKAO KUBO* AND MINORU KIHARA

Department of Pharmacology, Yokohama City University School of Medicine Urafunecho, Minami-ku, Yokohama, 232, Japan

(Received January 20, 1986)

The purpose of this study was to examine, using pentobarbital-anesthetized rats, whether vasopressin contributes to hypertension caused by denervation of baroreceptors, in comparison to the contribution of vasopressin to hypertension caused by nucleus tractus solitarius (NTS) lesions. Bilateral baroreceptor denervation caused acute hypertension which was inhibited by intravenous injection of an antagonist of the vasoconstrictor action of vasopressin. Lesions of the NTS also produced an increased in blood pressure which was reduced by the vasopressin antagonist. The magnitude of the antagonist-induced hypotension was significantly greater in NTS hypertension than that in baroreceptor denervation hypertension. It is concluded, that in rats, vasopressin contributes to the neurogenic hypertension caused by baroreceptor denervation and NTS lesions. Thus, it appears that interruption of baroreceptor afferents at the NTS level is responsible for the vasopressin-mediated hypertension caused by NTS lesions but that it is not the only factor involved.

Keywords — baroreceptor denervation; NTS lesion; vasopressin; hypertension; vasopressin antagonist

INTRODUCTION

In previous reports, Kubo *et al.*^{1,2)} demonstrated that bilateral chemical lesions of the nucleus tractus solitarius (NTS) elicited arterial hypertension mediated by vasopressin in rats after spinal transection. A similar contribution of vasopressin has also been shown in hypertension caused by bilateral electrolytic lesions of the NTS in rats with the spinal cord intact.³⁾ Since the NTS is the site of termination of baroreceptor afferents,^{4,5)} the increase of vasopressin release has been attributed to removal of tonic inhibitory influences on vasopressin release, exerted by baroreceptors.^{6,7)} However, exact mechanisms responsible for the enhanced release of vasopressin remain unknown. In the present study we have examined, in pentobarbital-anesthetized rats, whether vasopressin indeed contributes to hypertension caused by acute denervation of baroreceptors, in comparison to the contribution of vasopressin to the NTS hypertension.

METHODS AND MATERIALS

Experiments were performed on male Wistar rats weighing 260—310 g. Rats were anesthe-

tized with pentobarbital (50 mg/kg, i.p.). Plastic cannulae were inserted into the left femoral artery and vein. The arterial cannula was connected to a strain gauge transducer. Pulsatile or mean arterial pressure and heart rate derived by a cardiometer (Sanei-2140) were displayed by channels of a polygraph. Methylatropine (2 mg/kg, i.v.) was injected in order to block peripheral muscarinic receptors. Body temperature was maintained 36—37 °C with a heating pad and a heating lamp.

The carotid sinus and aortic baroreceptors were denervated bilaterally using a procedure based on the technique described by Krieger.⁸⁾ A 5-mm segment of the cervical sympathetic trunk was cut and removed immediately caudal to the superior cervical ganglion. A 5-mm segment of the superior laryngeal nerve was also cut and removed from the redundant loop. The common carotid, internal carotid and external carotid arteries were then thoroughly cleared of all connective tissues. In sham-operated rats an extensive midline incision was made in the neck, to expose the neurovascular sheaths enclosing the common carotid arteries, the vagi, and the sympathetic trunk but the sino-aortic nerves

* To whom should be addressed.

were left intact.

Electrolytic lesions were made in a stereotaxic apparatus with a monopolar Teflon-coated stainless steel electrode (0.15 mm, o.d.) with a tip exposure of 0.2 mm. The dorsal surface of the lower brainstem was exposed as described ⁹⁾ and lesions of the NTS were produced bilaterally by passing of an anodal direct current (DC) of 5 mA for 1–2 s with the electrode placed, at the level of the obex, laterally 0.6 mm and vertically 0.8 mm beneath the dorsal surface of the brain. The cathode was an alligator clamp attached to a neck muscle. Sham-operated rats were treated identically except that no DC current was applied. The localization and site of lesions were later controlled histologically.

The following drugs were used: pentobarbital sodium (Abbott, IL, U.S.A.), (–)-phenylephrine hydrochloride, atropine methylbromide (Sigma, MO, U.S.A.) and $d(\text{CH}_2)_5\text{Tyr}(\text{Me})$ arginine-vasopressin (Kruszyski *et al.*).¹⁰⁾ All drugs were dissolved in saline (0.9% NaCl).

Data are reported as mean $\pm$ S.E. and the student's *t*-test was used for statistical analysis of the results.

RESULTS

Effects of Baroreceptor Denervation

Following bilateral baroreceptor denervation in pentobarbital-anesthetized rats, blood pressure began to increase immediately and reached a maximal level within 5–10 min after the denervation (Fig. 1). The pressor response was maintained at this level for more than 20 min.

The vasopressin antagonist, $d(\text{CH}_2)_5\text{Tyr}(\text{Me})$ -arginine-vasopressin (20 $\mu\text{g}/\text{kg}$, i.v.), when given during the plateau level of the pressor response produced a reduction of blood pressure (Table

FIG. 1. Effects of Baroreceptor Denervation or NTS Lesion on Blood Pressure (BP) and Heart Rate (HR)

Pentobarbital-anesthetized rats received bilateral baroreceptor denervation (closed circles, $n = 9$) or sham denervation (open circles, $n = 3$), or NTS lesions (closed triangles, $n = 6$) or sham lesions (open triangles, $n = 3$). Values are the mean $\pm$ S.E. a) Significantly different from respective control ($p < 0.05$). b) Significantly different from baroreceptor denervation ($p < 0.05$).

TABLE I. Effects of $d(\text{CH}_2)_5\text{Tyr}(\text{Me})$ arginine-vasopressin (AVP Antagonist) on the Pressor Response Induced by Baroreceptor Denervation or NTS Lesions

Treatment	Mean arterial pressure (mm Hg)			Δ Arterial pressure (mm Hg) ^{b)}
	Initial level	Just before AVP antagonist ^{a)}	3 min after AVP antagonist	
Baroreceptor denervation				
Control ($n = 9$)	112.3 $\pm$ 2.5	135.6 $\pm$ 2.4	135.4 $\pm$ 2.2	-0.2 $\pm$ 1.1
AVP antagonist ($n = 7$)	107.1 $\pm$ 3.2	134.6 $\pm$ 2.6	119.6 $\pm$ 3.2 ^{c)}	-15.0 $\pm$ 3.3 ^{c)}
NTS lesion				
Control ($n = 6$)	113.8 $\pm$ 3.3	159.2 $\pm$ 8.7	158.5 $\pm$ 8.3 ^{d)}	-0.7 $\pm$ 0.4
AVP antagonist ($n = 5$)	111.0 $\pm$ 4.0	162.0 $\pm$ 10.0	124.2 $\pm$ 5.0 ^{c)}	-37.8 $\pm$ 8.9 ^{c,d)}

Values are the mean $\pm$ S.E. a) The vasopressin antagonist at 20 $\mu\text{g}/\text{kg}$ was intravenously injected 5–10 min after baroreceptor denervation or NTS lesion. b) Differences between the mean arterial pressure just before and 3 min after AVP antagonist. c) Significantly different from respective control ($p < 0.05$). d) Significantly different from baroreceptor denervation ($p < 0.05$).

FIG. 2. The Effect of Mean Blood Pressure (BP, mm Hg) and Heart Rate (HR, beats/min) of NTS Lesions and the Effect of $d(\text{CH}_2)_5\text{-Tyr(Me)arginine vasopressin}$ (AVP Antagonist, $20 \mu\text{g/kg}$, i.v.) on the Pressor Response to NTS Lesion

I). In contrast, the vasopressin antagonist ($20 \mu\text{g/kg}$, i.v.) did not affect blood pressure in sham-operated rats ($n = 3$). In 9 baroreceptor-denervated rats tested, phenylephrine ($16 \mu\text{g/kg}$, i.v.) produced little or no decrease (-4 ± 2 beats/min) in heart rate despite a marked increase in blood pressure (41.4 ± 3.1 mm Hg).

Effects of Nucleus Tractus Solitarius Lesion

Bilateral electrolytic lesions of the NTS also produced an increase in blood pressure. The pressor response to lesions reached a maximum within 5 min after lesions (Fig. 2) and was maintained at this level for more than 20 min. The pressor response to NTS lesions was significantly greater than that to baroreceptor denervation (Fig. 1 and Table I). The vasopressin antagonist ($20 \mu\text{g/kg}$, i.v.), when injected during the plateau phase, caused a decreased in blood pressure (Fig. 2 and Table I). The depressor response to the vasopressin antagonist in the NTS-lesioned rats was significantly greater than that in the baroreceptor-denervated rats (Table I).

DISCUSSION

In the present study we have shown that in pentobarbital-anesthetized rats, bilateral baroreceptor denervation produces an increase in blood pressure and such pressor responses were diminished by a vasopressin antagonist, $d(\text{CH}_2)_5\text{Tyr(Me)arginine vasopressin}$. The vasopressin antagonist did not affect the blood pressure of sham-operated control rats. The vasopressin antagonist abolished the pressor response to vasopressin but not to noradrenaline or angio-

tensin II,¹¹⁾ suggesting that the antagonist is specific to vasopressin. These findings suggest that a component of the neurogenic hypertension caused by baroreceptor denervation is mediated by vasopressin. These results also corroborate the suggestion that arterial and cardiopulmonary baroreceptors tonically inhibit vasopressin release.^{6,7,12)}

NTS lesions produced an increase in blood pressure and such hypertension was also diminished by the vasopressin antagonist, suggesting the involvement of vasopressin in mediation of this type of hypertension. These results are compatible with our previous reports^{1,2)} showing that in rats with spinal transection, chemical lesions of the NTS cause an increase in vasopressin release to the extent of eliciting a pressor response. These results are also in general agreement with those of Sved *et al.*³⁾ who used conscious or chloralose-anesthetized rats. Since the NTS is the site of termination of primary afferent fibers from all the baroreceptors,^{4,5)} the vasopressin-mediated hypertension induced by NTS lesions may be, at least in part, due to interruption at the NTS level of the baroreceptor afferents.

In the present experiments, the magnitude of the NTS lesion-induced increase in blood pressure was significantly greater than that of the baroreceptor denervation-induced increase in blood pressure. In addition, the magnitude of the depressor response to the vasopressin antagonist in the NTS hypertension was significantly greater than that in the baroreceptor denervation hypertension. In the baroreceptor-denervated rats, phenylephrine failed to induce bradycardic reflex responses, which confirms the efficacy of the denervation procedure. Thus, it seems likely that not only interruption of baroreceptor afferents but also other additional mechanisms in the NTS are involved in mediation of the vasopressin-mediated hypertension caused by NTS lesion. Further studies are needed to elucidate the mechanisms.

In conclusion, the results of the present study indicate that, in anesthetized rats, vasopressin contributes to the neurogenic hypertension caused by baroreceptor denervation and NTS lesions. It appears that the baroreceptor afferent interruption is responsible for the vasopressin-mediated hypertension caused by NTS lesions but is not the only factor involved.

REFERENCES

- 1) T. Kubo and Y. Misu: Vasopressin-induced pressor responses following microinjection of kainic acid into the nucleus tractus solitarii of the rat, *Jpn. J. Pharmacol.*, **36**, 118–120 (1984).
- 2) T. Kubo, H. Amano, M. Katsumata and Y. Misu: Further studies on vasopressin-induced pressor responses to kainic acid injected into the nucleus tractus solitarii of the rat, *Arch. Int. Pharmacodyn.*, **276**, 73–81 (1985).
- 3) A. F. Sved, T. Imaizumi, W. T. Talman and D. J. Reis: Vasopressin contributes to hypertension caused by nucleus tractus solitarius lesions, *Hypertension*, **7**, 262–267 (1985).
- 4) M. Miura and D. J. Reis: Termination of secondary projections of carotid sinus nerve in the cat brainstem, *Am. J. Physiol.*, **217**, 142–153 (1969).
- 5) M. Palkovits and L. Zaborszky: Neuroanatomy of central cardiovascular control. Nucleus tractus solitarii: afferent and efferent neuronal connections in relation to the baroreceptor reflex arc, *Prog. Brain Res.*, **47**, 9–34 (1977).
- 6) L. Share and M. N. Levy: Cardiovascular receptors and blood titer of antidiuretic hormone, *Am. J. Physiol.*, **203**, 425–428 (1962).
- 7) M. D. Thames and P. G. Schmid: Cardiopulmonary receptors with vagal afferents tonically inhibit ADH release in the dog, *Am. J. Physiol.*, **237**, H299–H304 (1979).
- 8) E. M. Krieger: Neurogenic hypertension in the rat, *Circ. Res.*, **15**, 511–521 (1964).
- 9) T. Kubo and Y. Misu: Cardiovascular response to microinjection of physostigmine and choline into the dorsal medullary site of the rat, *Neuropharmacology*, **20**, 1091–1095 (1981).
- 10) M. Kruszynski, B. Lammek, M. Manning, J. Seto, J. Halder, and W. H. Sawyer: [1-(β -Mercapto- β , β -cyclopentamethylenepropionic acid), 2-(*o*-methyl) tyrosine]arginine-vasopressin and [1-(β -mercapto- β , β -cyclopentamethylenepropionic acid)] arginine-vasopressin, two highly potent antagonists of the vasopressor response to arginine-vasopressin, *J. Med. Chem.*, **23**, 364–368 (1980).
- 11) T. Kubo, H. Amano and Y. Misu: Vasopressin-induced pressor responses to carotid occlusion in the rat, *Naunyn-Schmiedeberg's Arch. Pharmacol.*, **327**, 332–335 (1984).
- 12) H. Yamashita and K. Koizumi: Influence of carotid and aortic baroreceptors on neurosecretory neurons in supraoptic nuclei, *Brain Res.*, **170**, 259–277 (1979).