Serotonin Modulates Glutamate Action in the Medulla to Regulate Cardiovascular Functions in Cats

Wun-Chin Wu[1], Ming-Ren Wang[2], Ching-Yue Yang[3], Chun-Kuei Su[4], and Chok-Yung Chai[4*]

[1]*Department of Electronic Engineering*
National Penghu Institute of Technology
Penghu, Taiwan, R.O.C.
[2]*Institute of Aerospace Medicine*
National Defense University
Taipei, Taiwan, R.O.C.
[3]*Department of Anaesthesiology*
Chang-Gung Memorial Hospital
Taoyuan, Taiwan, R.O.C.
and
[4]*Institute of Biomedical Sciences*
Academia Sinica, Taipei
Taiwan, R.O.C.

Abstract

We examined the effects of serotonin (5-HT) on cardiovascular responses and blood flows in the right common carotid artery (RCCA), superior mesenteric artery (SMA) and right femoral artery (RFA), stimulated by glutamate (Glu) in the dorsomedial medulla (DM), rostral ventrolateral medulla (RVLM) and caudal ventrolateral medulla (CVLM). Microinjection of Glu into the DM produced increases in systemic arterial pressure (SAP) and flows in the RCCA and RFA, and decrease in flow in the SMA. Microinjection of Glu into the RVLM produced increases in SAP and decreases in flows in the RCCA, SMA and RFA. Prior microinjections of 5-HT into the same sites attenuated all the Glu-induced responses. Microinjection of Glu into the CVLM produced decreases in SAP and flows in the RCCA, SMA and RFA. These decreases were potentiated by prior injection of 5-HT. These findings suggest that 5-HT modulates the cardiovascular and blood flow responses induced by Glu in the medulla.

Key Words: caudal ventrolateral medulla, dorsomedial medulla, rostral ventrolateral medulla, systemic arterial pressure, glutamate, serotonin

Introduction

Serotonin (5-HT) plays an important role as a neuromodulator of cardiovascular functions including vasoconstriction and dilatation (20). 5-HT-immunoreactive cells are distributed not only in the midline raphe nuclei of the midbrain, but also in the pons and medulla (12, 22, 25). Nervous structures that may be responsible for the maintenance of vasomotor tone through the regulation of sympathetic outflow activity are located mainly in the medullary region. The rostral ventrolateral medulla (RVLM) area is a major source of sympathetic vasomotor drive that provides an excitatory bulbospinal pathway to the sympathetic preganglionic cells in the intermediolateral column of the spinal cord (6). The RVLM plays an important role in the maintenance of sympathetic tone and SAP, while the dorsomedial medulla (DM) shares these functions (2, 6, 30). Moreover, the caudal ventrolateral medulla (CVLM) is important for sympathetic inhibition leading to depressor actions (5).

Corresponding author: Dr. Chok-Yung Chai, Institute of Biomedical Sciences, Academia Sinica, Taipei 11529, Taiwan, R.O.C. Tel: +886-2-2789-9105; Fax: +886-2-2782-9224; E-mail: wcwu@npit.edu.tw
Received: September 6, 2004; Revised: November 30, 2004; Accepted: December 7, 2004.

Gong *et al*. have suggested that 5-HT and Glu appear to be tonically and endogenously released from nerve terminals in DM, and the 5-HT-induced decrease in systemic arterial pressure (SAP) could be attributed to the decreased Glu release resulting from the inhibitory action of 5-HT in DM (9). In addition, 5-HT inhibited glutamate (Glu) release in the dorsal facial area of the medulla and decreased the blood flow of the common carotid artery (8). Besides, the Glu release from the dorsal facial area was dose-dependently decreased by perfusion of 5-HT agonist to the same area (14, 15). This prompted us to investigate the modulatory effects of 5-HT on cardiovascular integration in medulla.

In this study we examined effects of 5-HT on the Glu action not only in the RVLM, DM, but also in CVLM about the regulation of cardiovascular functions i.e., SAP, HR, and blood flows of the right common carotid artery (RCCA), superior mesenteric artery (SMA) and right femoral artery (RFA).

Materials and Methods

General Procedures

Thirty-seven cats of either sex, weighing 2.5-4.0 kg, were anesthetized intraperitoneally with urethane (400 mg/kg) and α-chloralose (40 mg/kg). All experimental procedures followed were approved by the Committee of Animal Care and Use of the Institute of Biomedical Sciences under the guideline of National Science Council. The general procedures of experimentation have been described previously (3). These included cannulation of the left femoral artery for monitoring SAP, mean SAP (MSAP), heart rate (HR) and indirect cardiac contractile force (dp/dt); cannulation of the left femoral vein for saline or drug injection; intubation of the trachea for artificial ventilation to maintain the end-tidal CO_2 concentration at 4%; maintenance of the animal's rectal temperature at 37.5±1.0°C by a thermostatically controlled infrared light.

Blood Flow Measurements

The right common carotid artery (RCCA), superior mesenteric artery (SMA) and right femoral artery (RFA) were isolated, and each vessel was put on a proper sized flow-probe (blood flow transducer) through which the instantaneous pulsatile blood flow of blood was monitored through the blood flow meter (Spectramed, SP2204B). The actual blood flow (in ml/min) was calibrated using a calibrator (Spectramed, SP7014). All recordings were made on a polygraph (Gold ES2800) and all data were stored on a tape recorder system (Neuro Data DR-890, Sony slv-400) for later analysis.

Brain Stimulation

The head of each cat was fixed in a David-Kopf stereotaxic apparatus. The dorsal surface of the brain stem was exposed and the obex was used as the reference point. Three areas in the medulla were stimulated, namely the DM, RVLM and CVLM. The stereotaxic coordinates of these structures were: DM, 2.0-4.0 mm rostral to the obex, 1.5-3.0 mm lateral to the midline and 0.5-2.0 mm ventral to the dorsal surface of the medulla; RVLM, 3.5-5.0 mm rostral to the obex, 3.0-4.5 mm lateral to the midline and 3.5-4.5 mm ventral to the dorsal surface of the medulla; CVLM, 1.0 mm rostral to the posterior obex, 2.8-4.2 mm lateral to the midline and 3.0-4.5 mm ventral to the dorsal surface of the medulla.

Chemical stimulation was applied through a double-barrel micropipettes (outside tip diameter of 20-30 μm) inclined at 34° from the stereotaxic frame. Each micropipette contained Glu or 5-HT in saline with 0.1% pontamine sky blue, at pH 7.4. The other ends of the barrel were connected to the pneumatic pressure system (PPS-2, PPM-2, Medical Systems Corp., Great Neck, NY, USA) for chemical injection under a microscope (Wild M650) fitted with a reticule. Control injection of the vehicle alone did not produce any discernible effects. The cardiovascular sites in the brain were first activated by microinjections of Glu. Thirty minutes later, 5-HT was microinjected, and about 5 min later, Glu was microinjected again at the same site.

Histology

At the end of each experiment, the animal was euthanized by an overdose of pentobarbital. The brain was removed and immersed in 10% formalin-saline for 8 hrs. After fixation, frozen transverse sections (50 μm) of the brain were stained with Cresyl violet for identifying the injection sites.

Data Analysis

The resistance of arteries was calculated by the formula: (Blood pressure) / (Blood flow). Percent changes in SAP, HR, differential of blood pressure (dp/dt), resistance and blood flow volume of RCCA, SMA and RFA in response to microinjections of Glu and 5-HT were measured with the following formula: (Response value – Control value)/(Control value) × 100%. Changes in all data were analyzed by Student's paired *t*-tests and considered statistically significant at $P < 0.05$.

Results

Dose-response curve of SAP to stimulation of

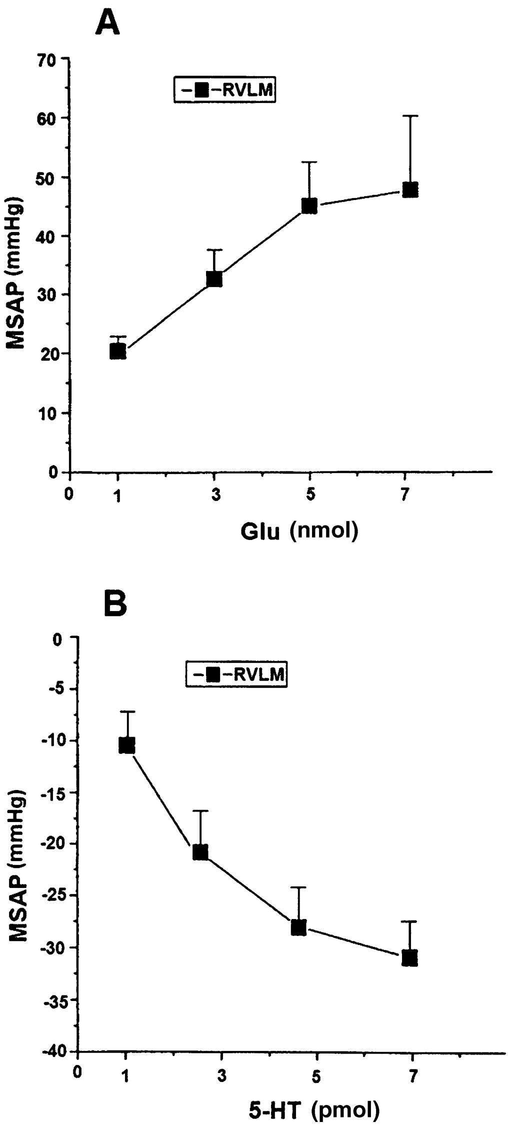

Fig. 1. Dose-response curves showing the changes of MSAP elicited by Glu and 5-HT in RVLM. (A) The response of MSAP produced by Glu. (B) The response of MSAP produced by 5-HT. In each group 5 points were studied. Vertical lines are SEM.

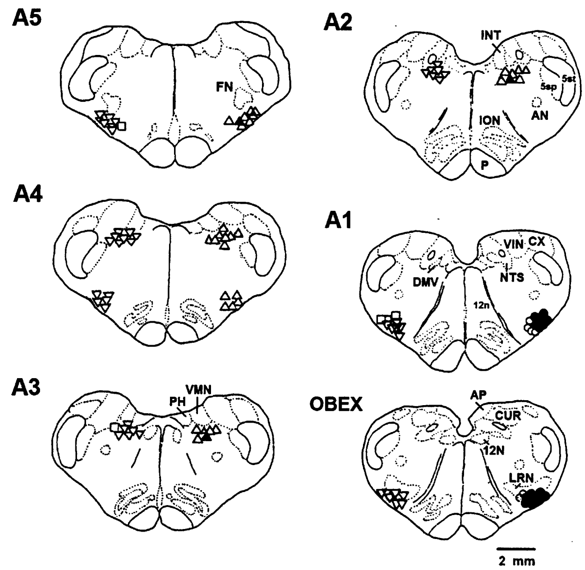

Fig. 2. Distribution of the pressor and depressor sites in medulla from the level of obex (A0) to 5 mm (A5) rostral to the obex consequent to microinjection of Glu or 5-HT. The drawing on the right side maps brain sites that responded first to microinjections of Glu (5.0 nmol), and 30 min later to 5-HT (5.0 pmol), and then 5 min after 5-HT again a second dose of Glu. Sites that responded to Glu and 30 min later followed by 5-HT alone are mapped on the left side. On the right side, solid triangles (▲) indicate that microinjection of Glu increased SAP, and microinjection of 5-HT later to the same point produced increases in SAP over 30 mm Hg. Empty triangles (△) indicate that Glu increased SAP. Glu again produced lower increases in SAP, below 30 mm Hg. Solid circles (●) indicate that Glu decreased SAP; 5 min after 5-HT, Glu again produced more marked decreases in SAP. Empty circles (○) indicate that Glu produced a SAP decrease; at 5 min after 5-HT, Glu again produced less SAP decrease. On the left side, empty squares (□) and inverse empty triangles (▽) indicate sites that showed no change or decreases in SAP induced by 5-HT, respectively. Abbreviations: AN, nucleus of ambiguous; CUR, cuneate nucleus, rostral division; CX, external cuneate nucleus; DMV, dorsomotor nucleus of vagus; FN, facial nucleus; INT, nucleus intercalates; ION, inferior olivary nucleus; NTS, nucleus tractus solitarius; P, paramidal tract; PH, nucleus praepositus hypoglossi; VIN, inferior vestibular nucleus; VMN, medial vestibular nucleus; 5sp, spinal trigeminal nucleus; 5st, spinal trigeminal tract; 12N, hypoglossal nucleus; 12n, hypoglossal nerve.

RVLM in 5 cats with different concentrations of Glu (1, 3, 5, 7 nmol) and 5-HT (1, 3, 5, 7 pmol) are shown in Fig.1. These results were obtained when Glu or 5-HT alone was microinjected into the pressor areas of RVLM through a four-barrel micropipette. Following these response curves, we selected a dose of 5 nmol of Glu and a dose of 5 pmol of 5-HT for chemical activations.

Changes in cardiovascular responses, namely the SAP, HR and blood flows in the RCCA, SMA and RFA, induced by microinjecting Glu (5.0 nmol in 50 nl) in the DM, RVLM or CVLM were compared with those after microinjecting 5-HT (5.0 pmol in 50 nl) in the same point. The reactive sites of Glu and 5-HT are mapped in Fig. 2. Judging from the locations and diameters of the dye spread, the DM sites are distributed in the dorsal recticular formation, medial vestibular nucleus and ventral praepositus hypoglossi nucleus. The RVLM sites are distributed in the caudal retrofacial nucleus and rostral Pre-Botzinger complex, while the CVLM sites are distributed around the external division of lateral recticular nucleus and the noradrenergic A1 area. Under control conditions, the resting MSAP and HR were 122.8±11.4 mmHg and 205±13.6 bpm, and the blood flows in RCCA, SMA and RFA were 24.7±4.8, 27.6±5.3 and 5.4±1.3 ml/min, respectively.

Table 1. Effects of 5-HT (5.0 pmol) on the cardiovascular and blood flow responses induced by Glu (5.0 nmol) in the medulla.

					RCCA		SMA		RFA	
		MSAP (%)	HR (%)	dp/dt (%)	Flow (%)	Res (%)	Flow (%)	Res (%)	Flow (%)	Res (%)
DM	A	45.3±6.4	-32.7±4.6	114.5±36.1	52.5±12.1	-47.9±8.3	-34.6±6.1	92.3±11.9	173.6±13.4	-67.5±8.2
	B	4.0±2.9	-13.1±4.5	-4.8±1.0	6.1±2.2	-5.3±3.5	-8.1±3.7	9.9±3.4	7.3±2.4	-8.2±3.6
	C	16.2±4.3*	-15.4±4.7*	15.6±3.4*	28.6±8.8*	-19.5±4.7*	-15.8±4.4*	52.9±11.6*	82.6±9.5*	-33.5±6.4*
RVLM	A	53.7±8.7	-42.4±7.5	18.7±6.2	-35.2±7.2	32.9±6.5	-38.4±6.9	43.4±7.2	-24.5±5.6	17.1±3.9
	B	-7.8±1.6	-9.4±2.3	-2.9±6.4	7.3±2.8	-4.2±1.8	9.2±2.7	-3.3±2.4	3.8±2.5	- 2.5±5.3
	C	28.5±4.7*	-38.3±7.6*	33.8±10.6*	-18.5±5.4*	15.3±9.8*	-24.7±9.3*	27.4±4.1*	-12.5±3.2	8.4±5.9
CVLM	A	-23.6±5.5	-32.2±6.2	5.5±6.2	-17.5±5.1	18.3±4.5	-23.7±5.8	19.2±5.3	-12.3±4.5	8.5±4.9
	B	-15.4±4.4	-34.4±5.6	14.3±6.9	-7.8±4.3	9.9±4.3	-8.4±2.5	5.8±1.8	-9.9±3.3	5.1±1.2
	C	-48.6±7.3*	-36.7±4.1	-3.0±4.7*	-24.8±5.5*	25.4±6.4*	-38.6±4.4*	26.1±7.4*	-28.4±6.4*	23.4±7.9*

Values are mean ± S.E.M.; A: effects of Glu; B: effects of 5-HT; C: after 5-HT, effects of Glu again; DM: dorsomedial medulla; RVLM: rostral ventrolateral medulla; CVLM: caudal ventrolateral medulla; MSAP: mean systemic arterial pressure; HR: heart rate; dp/dt: differential of blood pressure; RCCA: right common carotid artery; SMA: superior mesenteric artery; RFA: right femoral artery; Res: resistance of artery. The symbol * indicates the values-induced by Glu before and after 5-HT activation are statistically significant compared to the control value calculated by Student's paired t-test with $P < 0.05$.

In 22 DM points of 10 cats, microinjection of Glu (5.0 nmol) into the DM produced increase in MSAP, dp/dt, and decrease in HR associated with flow increases in RCCA and RFA, but decreases in SMA (Table 1). After injecting 5-HT (5.0 pmol) at the same site, the cardiovascular and blood flow responses to Glu were attenuated (Fig. 3). Injection of 5-HT alone produced slight decreases in the responses of SAP, HR and blood flow.

In 24 RVLM points of 10 cats, microinjection of Glu produced increase in MSAP and decrease in HR associated with flow decreases in RCCA, SMA and RFA (Table 1). After injecting 5-HT, the Glu-induced cardiovascular responses and decrease in flows were attenuated (Fig. 4). Microinjection of 5-HT alone produced decreases in MSAP and HR and increases in the flow of RCCA and SMA.

In 27 CVLM points of 12 cats, microinjection of Glu produced decreases in MSAP and HR associated with flow decreases in RCCA, SMA and RFA (Table 1). After injecting 5-HT, the Glu-induced decreases of MSAP, HR and blood flows were enhanced (Fig. 5). Microinjection of 5-HT alone produced decreases in MSAP, HR, RCCA, SMA and RFA.

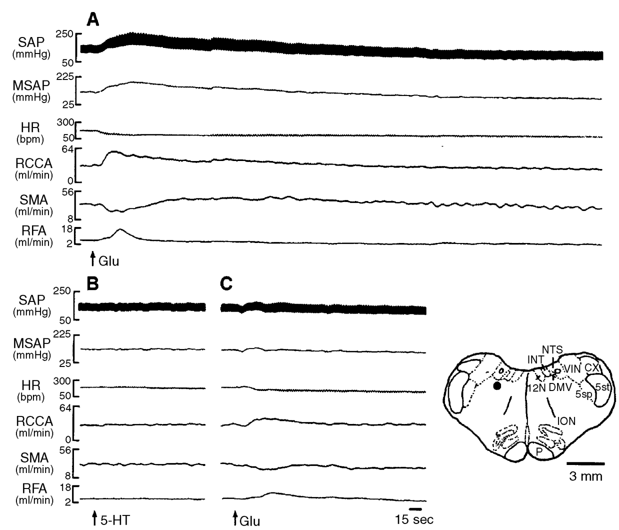

Fig. 3. Microinjection of 5-HT attenuates the DM responses induced by Glu. (A) Microinjection of Glu (5.0 nmol) into the DM produced a marked increase of SAP (47.5%) and decrease in HR (27.7%). Blood flows of RCCA increased 76.5% and RFA 290%, while SMA decreased 25.3%. (B) Thirty minutes later, microinjection of 5-HT (5.0 pmol) into the same point produced slight change in SAP and blood flows of the three arteries. (C) Five minutes after 5-HT, microinjection of Glu again to the same site produced an increase in SAP (8.7%) and a decrease in HR (20.5%). Blood flows of RCCA and RFA increased by 31.5% and 130%, while the SMA decreased by 17.4%. The arrow (↑) under the tracings indicates the time of chemical injection. Dot (●) in the brain drawing shows the point of stimulation. In this and following figures, from top to bottom are tracings of SAP, systemic arterial pressure; MSAP, mean SAP; HR, heart rate; RCCA, right common carotid artery; SMA, superior mesenteric artery; RFA, right femoral artery.

Discussion

Microinjection of Glu into the RVLM, DM and CVLM produced cardiovascular responses and changes in blood flow of the RCCA, SMA and RFA. The effects of Glu in the DM and RVLM on the

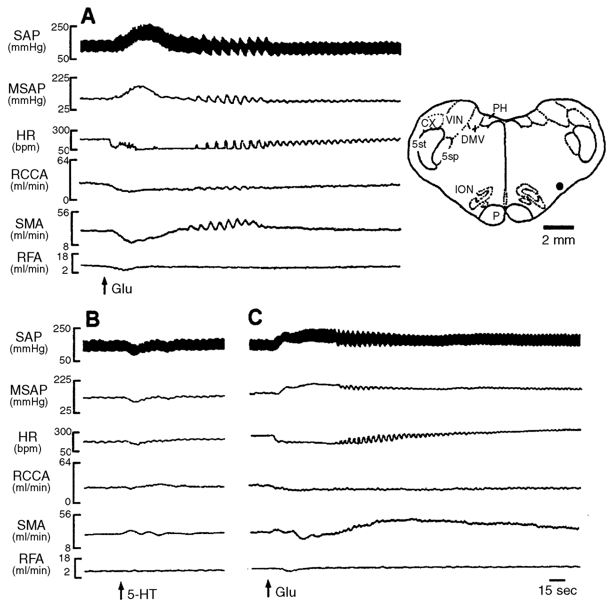

Fig. 4. Microinjection of 5-HT attenuates the Glu-induced cardiovascular and blood flow responses in RVLM. (A) Microinjection of Glu (5.0 nmol) into the RVLM increased the SAP (58.3%) and decreased the HR (54.2%). Blood flows in the RCCA and RFA were decreased by 32.8% and 26.5%. The SMA was decreased by 43.2% following an increase of 35.6%. (B) Thirty minutes later, microinjection of 5-HT (5.0 pmol) produced a decrease in SAP (12.5%) and increases in the RCCA (9.2%) and SMA (15.3%). (C) Five minutes after 5-HT, microinjection of Glu produced an increase in the SAP (27.3%) and a decrease in HR (56.3%). Blood flow in the RCCA and RFA were decreased by 18.2% and 12.3%. The SMA was decreased by 27.6% following an increase of 42.3%.

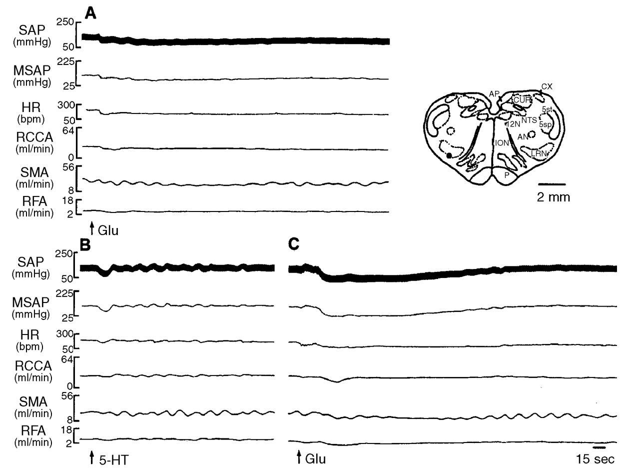

Fig. 5. Microinjection of 5-HT potentiates the depressor and blood flow response in the CVLM induced by Glu. (A) Microinjection of Glu (5.0 nmol) decreased the SAP (29.7%) and HR (25.3%) in association with decreases in blood flow in the three arteries; RCCA (18.2%), SMA (22.4%) and RFA (12.5%). (B) Thirty minutes after Glu, microinjection of 5-HT (5.0 pmol) into the same point produced more apparent decreases in the SAP (35.6%) and HR (25.2%). (C) Five minutes after 5-HT, Glu at the same site again produced more marked responses in the reduction of SAP (58.6%), HR (42.5%) and blood flow of the three arteries; RCCA (35.8%), SMA (34.3%) and RFA (27.8%).

cardiovascular and flow responses were attenuated but those in the CVLM were enhanced by prior treatment of 5-HT in the same point. This suggests that 5-HT modulates the effects of Glu not only in the DM, RVLM, but also in the CVLM to regulate cardiovascular functions including SAP, HR, dp/dt and blood flows of different vascular beds.

Previous studies showed that microinjections of 5-HT into the NTS and the dorsal motor nucleus of vagus (DMV) produced hypotension, bradycardia and modulation of gastrointestinal functions (7, 23, 26). In addition, microinjection of 5-HT into the RVLM produced decreases in SAP and HR associated with vasodilation (16). The pressor areas of the DM which plays an important role on cardiovascular regulation are located close to the vagal complex area. Microinjection of Glu into DM produced pressor and bradycardic responses associated with sympathetic vertebral nerve activity (2, 30, 31). In general, increase in SAP always induces change in blood flow. Besides measuring blood flow, changes in the vessel resistance were calculated in this study. Vasoconstriction induces an increase in vessel resistance resulting in a decrease in blood flow, whereas vasorelaxation induces a decrease in vessel resistance with increases in blood flow. Changes of blood flow in the RCCA, SMA and RFA have been examined in the studies of behavioral defense reactions and cardiovascular regulation (3, 27). In the present study, microinjection of Glu into DM produced increases in SAP and flow in the RCCA and RFA but decrease in flow in the SMA. Although 5-HT alone produced slight changes in cardiovascular and blood flow responses, it attenuated these effects of Glu on cardiovascular response and blood flow.

Microinjections of Glu into the RVLM produced an increase in SAP, while bilateral microinjections of 5-HT receptor agonist into the same area produced decreases in SAP, HR and renal sympathetic nerve activity (13). In this study, microinjection of Glu into the RVLM produced pressor and bradycardic responses associated with blood flow decreases in the RCCA, RFA and SMA. Although 5-HT alone induced slight decrease in SAP, it attenuated effects of Glu on the cardiovascular and blood flow responses. Based on these observations, 5-HT might inhibit sympathetic neurons in the RVLM, and then it reduced the responses to Glu. This is consistent to the reports that microinjection of 5-HT into RVLM produced hypotension, bradycardia and peripheral vasodilatation (1, 16), and inhibited the activation of sympathoexcitatory neurons evoked by amino acid

(28). Furthermore, 5-HT decreases Glu-induced excitation of neurons in rat cerebral slices (10) and attenuates the excitation of locus coeruleus neuron discharges evoked by Glu (4).

The CVLM plays a significant role in the inhibitory regulation of sympathetic nerve activity and SAP (11, 32). Depressor neurons in this region form a serial link in the central baroreceptor-vasomotor pathway, constituting an inhibitory connection between the NTS and sympathoexcitatory bulbospinal neurons in the RVLM (21). Microinjection of Glu into the CVLM elicits decreases in SAP, HR and mesenteric and hindquarter vascular resistance (29). In this study, microinjection of Glu into the CVLM produced decreases in SAP, HR and blood flows of the RCCA, SMA and RFA. Microinjection of 5-HT alone produced decreases in SAP and HR. It enhanced the effects of Glu on cardiovascular and blood flow responses. According to the results of the present study, 5-HT might activate parasympathetic neurons in the CVLM, subsequently it increased its responses to Glu. These are similar to the findings that activation of the 5-HT_{2A} receptor potentiates the depolarizing responses mediated by Glu and NMDA receptors in neocortical neurons (19).

CNS serotonergic projection regulates the extracellular Glu concentration and consequently modulates the sensitivity of neurons to excitatory inputs. For instance, the extracellular concentration of Glu in the suprachiasmatic nucleus, which contains NMDA receptors, is significantly reduced after treating with 5-HT (24), and activation of 5-HT_3-like receptors in the hippocampal CA1 region effectively reduces the efficacy of glutamatergic neurotransmitters (33). In addition, 5-HT and DOI inhibit Glu release in the dorsal facial area (8, 14, 15). Besides, 5-HT and other neurotransmitters are also frequently found to coexist in axon terminals along with other hormones or neuropeptides. There is evidence implying a role of 5-HT in modulating the release of other neurotransmitters and hormones such as catecholamine (18) and dopamine (17).

In conclusion, Glu produced cardiovascular responses associated with changes in the blood flow of the RCCA, SMA and RFA. 5-HT alone produced change in cardiovascular responses and blood flow. It modulated the SAP, HR and blood flow effects induced by Glu. Thus, 5-HT attenuated the Glu-induced increases in SAP in the DM and RVLM, while it potentiated the Glu-induced decreases in SAP in the CVLM; all associated with changes in blood flow.

Acknowledgments

This study was supported in part by the Foundation of Biomedical Sciences, the Shih-Chun Wang Research Fund, and the National Science Council, Taiwan, ROC, Grant No. NSC 92-2320-B-016-045.

References

1. Bago, M., Sprtel, B.M. and Dean, C. Modulation of sympathetic nerve activity by microinjection of the 5-HT_{1A} receptor agonist 8-OH-DPAT into the rostroventrolateral medulla. *J. Auton. Nerv. Syst*. 76: 127-134, 1999.
2. Chai, C.Y., Lin, R.H., Lin, A.M.Y., Pan, C.M., Lee, E.H.Y. and Kuo, J.S. Pressor responses from electrical or glutamate stimulations of the dorsal or ventrolateral medulla. *Am. J. Physiol*. 255: R709-R717, 1988.
3. Chai, C.Y., Wu, W.C., Hwang, J.J., Wang, M.R. and Kuo, J.S. Attenuation of cardiovascular reactions of vocalized and non-vocalized defence areas of periaqueductal grey following lesions in dorsomedial or ventrolateral medulla of cats. *J. Auton. Nerv. Syst*. 64: 115-127, 1997.
4. Chouvet, G., Akaoka, H. and Aston-Jones, G. Selective reduction by serotonin, of neuronal excitation in the locus coeruleus evoked by glutamate. *C. R. Acad. Sci. III*., 306: 339-344, 1988.
5. Cravo, S.L. and Morrison, S.F. The caudal ventrolateral medulla is a source of tonic sympathoinhibition. *Brain Res*. 621: 133-136, 1993.
6. Dampney, R.A.L. Functional organization of central pathways regulating the cardiovascular system. *Physiol. Rev*. 74: 323-364, 1994.
7. Feldman, P.D. and Galiano, F.J. Cardiovascular effects of serotonin in the nucleus of the solitary tract. *Am. J. Physiol*. 269: R48-R56, 1995.
8. Gong, C.L., Lin, N.N. and Kuo, J.S. Glutamatergic and serotonergic mechanisms in the dorsal facial area for common carotid artery blood flow control in the cat. *Auton. Neurosci*. 101: 85-90, 2002.
9. Gong, C.L., Chiu, Y.T., Lin, N.N., Lin, S.Z., Cheng F.C. and Kuo, J.S. Inhibitory actions of serotonin on glutamate release in dorsal medulla suppress systemic arterial pressure of cats. *Neurosci. Lett*. 355: 73-76, 2004.
10. Hicks, T.P., Krupa, M. and Crepel, F. Selective effects of serotonin upon excitatory amino acid-induced depolarizations of purkinje cells in cerebellar slices from young rat. *Brain Res*. 492: 371-376, 1989.
11. Hsieh, J.H., Chang, Y.C., Chung, J.L., Hsiao, M.C., Chen, S.C., Yen, C.T. and Chai, C.Y. The relationship between FTL and NA, DMV or CVLM in central cardiovascular control. *Chinese J. Physiol*. 44: 169-179, 2001.
12. Jacobs, B.L. and Azmitia, E.C. Structure and function of the brain serotonin system. *Physiol. Rev*. 72: 165-229, 1992.
13. Kubo, T., Taguchi, K., Ozaki, S., Amano, M. and Ishizuka, T. 8-OH-DPAT-induced hypotensive action and sympathoexcitatory neurons in the rostral ventrolateral medulla of the rat. *Brain Res. Bull*. 36: 405-411, 1995.
14. Kuo, J.S., Li, H.T., Lin, N.N., Yang, C.S. and Cheng, F.C. Dorsal facial area of cat medulla: 5-HT_2 action on glutamate release in regulating common carotid blood flow. *Neurosci. Lett*. 266: 137-140, 1999.
15. Li, H.T., Chen, W.Y., Liu, L., Yang, C.S., Cheng, F.C., Chai, C.Y. and Kuo, J.S. The dorsal facial area of the medulla in cats: inhibitory action of serotonin on glutamate release in regulating common carotid blood flow. *Neurosci. Lett*. 210: 193-196, 1996.
16. Lovick, T.A. Systemic and regional haemodynamic responses to microinjection of 5-HT agonists in the rostral ventrolateral medulla in the rat. *Neurosci. Lett*. 107: 157-161, 1989.
17. Muramatsu, M., Tamaki-Ohashi, J., Usuki, C., Araki, H., Chaki, S.

and Aihara, H. 5-HT_2 antagonists and minaprine block the 5-HT-induced inhibition of dopamine release from rat brain striatal slices. *Eur. J. Pharmacol*. 153: 89-95, 1988.
18. Nosjean, A., Hamon, M. and Darmon, M. 5-HT_{2A} receptor are expressed by catecholaminergic neurons in the rat nucleus tractus solitarii. *Neuroreport* 13: 2365-2369, 2002.
19. Rahman, S. and Neuman, R.S. Activation of 5-HT_2 receptors facilitates depolarization of neocortical neurons by N-methyl-D-aspartate. *Eur. J. Pharmacol.* 231: 347-354, 1993.
20. Saxena, P.R. Cardiovascular effects from stimulation of 5-hydroxytryptamine receptors. *Fundam. Clin. Pharmacol*. 3: 245-265, 1989.
21. Sevoz, C., Hamon, M. and Laguzzi, R. Medullary pathways of cardiovascular responses to 5-HT_2 and 5-HT_3 receptor stimulation in the rat nucleus tractus solitarius. *Neuroreport* 7: 1965-1969, 1996.
22. Sevoz-Couche, C., Maisonneuve, B., Hamon, M. and Laguzzi, R. Glutamate and NO mediation of the pressor response to 5-HT_3 receptor stimulation in the nucleus tractus solitarii. *Neuroreport* 13: 837-841, 2002.
23. Sporton, S.C., Shepheard, S.L., Jordan, D. and Ramage, A.G. Microinjections of 5-HT_{1A} agonists into the dorsal motor vagal nucleus produce a bradycardia in the atenolol-pretreated anesthetized rat. *Br. J. Pharmacol*. 104: 466-470, 1991.
24. Srkalovic, G., Selim, M., Rea, M.A. and Glass, J.D. Serotonergic inhibition of extracellular glutamate in the suprachiasmatic nuclear region assessed using *in vivo* brain microdialysis. *Brain Res*. 656: 302-308, 1994.
25. Tallaksen-Greene, S.J., Elde, R.J. and Wessendorf, M.W. Regional distribution of serotonin and substance P co-existing in nerve fibers and terminals in the brainstem of the rat. *Neuroscience* 53: 1127-1142, 1993.
26. Varanasi, S., Chi, J. and Stephens, R.L. Jr. 5-CT or DOI augments TRH analog-induced gastric acid secretion at the dorsal vagal complex. *Am. J. Physiol.* 273: R1607-R1611, 1997.
27. Wang, M.R., Kuo, J.S. and Chai, C.Y. Cardiovascular and vocalization reactions elicited by N-methyl-D-aspartate in the pretentorial periaqueductal grey of cats. *Clin. Exp. Pharmacol. Physiol*. 29: 759-771, 2002.
28. Wang, W.H. and Lovick, T.A. Inhibitory serotonergic effects on rostral ventrolateral medullary neurons. *Pflugers Arch.* 422: 93-97, 1992.
29. Willette, R.N., Punnen-Grandy, S., Krieger, A.J. and Sapru, H.N. Differential regulation of regional vascular resistance by the rostral and caudal ventrolateral medulla in the rat. *J. Auton. Nerv. Syst*. 18: 143-151, 1987.
30. Wu, W.C., Chen, S.Y., Kuo, J.S. and Chai, C.Y. Glycine produced pressor responses when microinjected in the pressor areas of pons and medulla in cars. *J. Auton. Nerv. Syst*. 59: 1-11, 1996.
31. Wu, W.C. and Chai, C.Y. Spike responses of sympathetic vertebral nerve activities during stimulation of the pressor dosomedial and rostral ventrolateral medulla differ in cats. *Chinese J. Physiol*. 41: 45-52, 1998.
32. Wu, W.C., Su, C.K., Yang, C.Y. and Chai, C.Y. The nNOS/cGMP mediation of the depressor response to NMDA receptor stimulation in the caudal ventrolateral medulla. *Chinese J. Physiol.* 46: 175-179, 2003.
33. Zhang, J.Y., Zeise, M.L. and Wang, R.Y. Serotonin 3 receptors agonists attenuate glutamate-induced firing in rat hippocampal CA1 pyramidal cells. *Neuropharmacology* 33: 483-491, 1994.