Contributions of AMPA/kainate receptors in the rostral ventrolateral medulla to the hypotensive and sympathoinhibitory effects of clonidine

Wei-Zhong Wang,¹,² Li-Gang Wang,¹ Lie Gao,² and Wei Wang²

¹Department of Physiology, Second Military Medical University, Shanghai, China; and ²Department of Cellular and Integrative Physiology, University of Nebraska Medical Center, Omaha, Nebraska

Submitted 5 April 2007; accepted in final form 15 June 2007

Wang W-Z, Wang L-G, Gao L, Wang W. Contribution of AMPA/kainate receptors in the rostral ventrolateral medulla to the hypotensive and sympathoinhibitory effects of clonidine. Am J Physiol Regul Integr Comp Physiol 293: R1232–R1238, 2007. First published June 20, 2007; doi:10.1152/ajpregu.00233.2007.—The depressor and sympathoinhibitory effect of the imidazoline drug clonidine is related to the glutamate receptors in the RVLM, which has been recognized as a specific target area for mediating the central depressor mechanism of clonidine. The objective of this study was to determine the role of the glutamate receptor subtype α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA)/kainate receptor within the RVLM in clonidine-induced depressor and sympathoinhibitory action in anesthetized normotensive rats. Unilateral microinjection of 200 pmol of 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX), a potent AMPA/kainate receptor antagonist, into the RVLM completely abolished the pressor effect evoked by AMPA (5 pmol) without affecting the pressor action of N-methyl-D-aspartate (20 pmol). Pretreatment with intra-RVLM injection of CNQX (20 and 200 pmol) dose-dependently attenuated the reduction in blood pressure (BP), heart rate (HR), and renal sympathetic nerve activity (RSNA) elicited by intra-RVLM clonidine (5 nmol) or intravenous clonidine (10 μg/kg), while 2 pmol of CNQX did not alter clonidine-induced cardiovascular action. Furthermore, the decreases in BP, HR, and RSNA evoked by intravenous clonidine (10 μg/kg) or intra-RVLM clonidine (5 nmol) were reversed when CNQX (20 and 200 pmol) was subsequently injected into the RVLM. In conclusion, these data show that blockade of AMPA/kainate receptors in the RVLM significantly antagonizes decreases in BP, HR, and sympathetic activity induced by clonidine, suggesting that the AMPA/kainate receptors within the RVLM contribute to the depressor and sympathoinhibitory effect of clonidine.

imidazoline-like drug; 6-cyano-7-nitroquinoxaline-2,3-dione; blood pressure; renal sympathetic nerve activity; α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid

THE IMIDAZOLINE-LIKE DRUG clonidine, a mixed agonist for I1-imidazoline receptors (I1R) and α2-adrenoceptors (α2AR), is suggested to reduce blood pressure (BP) primarily by an action within the central nervous system (7, 9). It is well known that the rostral ventrolateral medulla (RVLM) is a key region regulating cardiovascular function (5), and the RVLM I1R and/or α2AR is also suggested to mediate the central mechanism responsible for sympathoinhibition of clonidine (6, 11, 28). The effects induced by activation of I1R and α2AR are related to the glutamate receptors (21, 31). For example, the hypotension of systemic clonidine is abolished by glutamate receptor blockade in spontaneously hypertensive rats (SHR) (15).

Glutamate within the central nervous system plays an important role in cardiovascular regulation by acting on two families of glutamate receptors, the metabotropic and the ionotropic glutamate receptors (4, 5, 29). The N-methyl-D-aspartate (NMDA) and α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA)/kainate receptors are two subtypes of ionotropic glutamate receptors that play an important role in cardiovascular control (2). The functional states of NMDA receptors in the RVLM have been reported to be closely related to the central depressor mechanism of imidazoline-like drugs (20). For example, the blockade of NMDA receptors within the RVLM prevents the hypotension of the imidazoline-like drug rilmenidine in SHR (38) and attenuates the inhibitory effect of clonidine on activity of the RVLM presympathetic neurons in normotensive rats (36, 37). However, there is no evidence concerning the role of AMPA/kainate receptors in modulating the central depressor mechanism of clonidine. According to previous studies, clonidine can modulate glutamate release through a presynaptic mechanism (15, 31). In addition to NMDA receptors, glutamate effects also depend on the functional states of AMPA/kainate receptors. Furthermore, it is suggested that, compared with NMDA receptors, the AMPA/kainate receptor in the RVLM plays a major role in controlling cardiovascular activity (1). Thus it is reasonable to hypothesize that the AMPA/kainate receptors in the RVLM would be involved in the central depressor mechanism of clonidine.

Therefore, we investigated the effects of the RVLM injected with three doses (20, 200, and 200 pmol) of 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX) on cardiovascular responses to either intra-RVLM or intravenous clonidine. We further determined the effects of subsequent injection of CNQX into the RVLM in response to the cardiovascular action of intra-RVLM or intravenous injection of clonidine.

MATERIALS AND METHODS

Animal preparation. All experiments were performed on adult male Sprague-Dawley rats weighing between 310 and 360 g. Experimental protocols were approved by the Institutional Animal Care and Use Committee of the University of Nebraska Medical Center and were carried out under the guidelines of the American Physiological Society and the National Research Council’s Guide for the Care and Use of Laboratory Animals. We have previously described the methods for general surgery, renal sympathetic nerve activity (RSNA) recording, and microinjection (8, 34).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
General surgery. Rats were anesthetized with urethane (800 mg/kg, ip) and α-chloralose (40 mg/kg, ip). The trachea was cannulated, and the rats were paralyzed with pancuronium bromide (1 mg/kg, iv; 0.1 mg/kg thereafter as needed) and ventilated artificially with room air supplemented with 100% oxygen. The left common carotid artery was cannulated, and the BP was measured with a pressure transducer (model PT300; Grass Instruments, Quincy, MA) for measurement of mean arterial pressure (MAP). Heart rate (HR) was derived from the BP pulse using a Powerlab model 16S (AD instruments, Colorado Springs, CO). The femoral vein was cannulated for intravenous injections. Rats were placed in a stereotaxic frame (Stoelting, Chicago, IL), and the dorsal surface of the medulla was surgically exposed by occipital craniotomy and partial cerebellectomy. Supplemental doses of α-chloralose (20 mg/kg, iv) were administered to maintain an appropriate level of anesthesia. Depth of anesthesia was gauged by the stability of BP and HR and the absence of a pressor response to paw pinch. Body temperature was maintained at ~37°C by an animal temperature controller (ATC1000, World Precision Instruments).

Microinjections into the RVLM. Microinjections were made from four-barrel micropipettes with total tip diameters of 20–50 μm and performed by a four-channel pressure injector (PM2000B, World Precision Instruments). The injections were made over 10–20 s, and the 100-nl injection volume was measured by observing the movement of the fluid meniscus along a reticule in a microscope. The coordinates for RVLM were 2.3–2.8 mm rostral to the obex, 1.8–2.0 mm lateral to the midline, and 3.0–3.4 mm below the dorsal surface of the medulla. Chemical identification of the RVLM was based on obtaining a pressor response elicited by microinjection of l-glutamate (5 nmol) at the start of each experiment, as in our previous study (36). The time interval between bilateral injections was within a 3-min period. At the end of the experiments, 100 nl of 2% pontamine sky blue were injected for marking the injection sites for subsequent histological identification.

Injected drugs. All chemicals including clonidine, l-glutamate, NMDA receptor agonist NMDA, AMPA/kainate receptor agonist AMPA, and AMPA/kainate receptor antagonist CNQX were obtained from Sigma Chemical. Clonidine, l-glutamate, NMDA, and AMPA were directly dissolved in artificial cerebrospinal fluid (aCSF) (in mM: 133.3 NaCl, 3.4 KCl, 1.3 CaCl2, 1.2 MgCl2, 0.6 Na2HPO4, 32.0 NaHCO3, and 3.4 glucose, pH 7.4). CNQX was initially dissolved in 0.1 M phosphate buffer solution and then diluted in aCSF to the final concentration. CNQX was chosen as the antagonist for AMPA/kainate receptors based on its specificity to block the respective receptor agonist (12). The doses of injected clonidine and CNQX into the RVLM were based on previous studies (6, 22). In addition, clonidine for intravenous injection (10 μg/kg) was dissolved in 0.9% normal saline.

Experimental groups and protocols. First, a total of seven rats were used to confirm that 200 pmol of CNQX unilaterally injected into the RVLM can adequately and selectively block AMPA/kainate receptors without affecting NMDA receptors. We observed the cardiovascular effects of NMDA receptor or AMPA/kainate receptor activation by injection of their responding agonist, NMDA (20 pmol) or AMPA (5 pmol), respectively, into the RVLM 5 min after intra-RVLM injection of 200 pmol of CNQX. The interval between repeated injections of NMDA or AMPA into the same site was at least 60 min. Second, six groups of rats (n = 4–7 each) were used to investigate the effects of intra-RVLM pretreatment with AMPA/kainate receptor blockade on clonidine actions. Five minutes after unilateral injection of 100 nl of vehicle (n = 4) or three doses of CNQX (2, 20, and 200 pmol; n = 6 each), we observed the responses of BP, HR, and RSNA to injection of 5 nmol of clonidine into the same site, and the responses were continuously recorded for 60 min after intra-RVLM clonidine. Furthermore, we examined the responses (60 min) of cardiovascular values to intravenously injected clonidine (10 μg/kg) 5 min after bilateral microinjection of 100 nl of vehicle (n = 5) or three doses of CNQX (2, 20, and 200 pmol; n = 6 each) 20 min after unilateral injection of clonidine (5 nmol). We also determined the cardiovascular responses of bilateral injection of 100 nl of vehicle (n = 3) or 200 pmol of CNQX (n = 6) into the RVLM 20 min after intravenously applied clonidine (10 μg/kg). In the above experiments, the cardiovascular values of BP, HR, and RSNA at the 10-min interval after injection of clonidine were collected for further statistical analysis.

Histological analysis. At the end of the experiment, the rat was given a lethal injection of pentobarbital sodium (100 mg/kg, iv) and perfused with 10% formaldehyde solution (100 ml) intracardially. The brain stem was then quickly removed and fixed in 10% buffered Formalin. Frozen 50-μm coronal sections were made on a freezing microtome and mounted on slides. The dye spot for injection sites was identified and plotted on standardized sections according to the atlas of Paxinos and Watson (24). Data were excluded if the injection sites were not located in the RVLM. Figure 1 shows the locations of the centers of microinjection sites in the RVLM.

RESULTS

Effects of 200 pmol of CNQX on BP, HR, and RSNA response to the RVLM NMDA or AMPA/kainate receptor agonists. A total of seven rats [baseline MAP and HR; 92 ± 4 mmHg and 381 ± 16 beats/min (bpm), respectively] were used to determine whether unilateral injection of 200 pmol of CNQX into the RVLM adequately blocks AMPA/kainate receptors without affecting NMDA receptors. Figure 2A shows the original tracings of effects of pretreatment with CNQX (200 pmol) on the pressor action induced by AMPA or NMDA injected into the RVLM unilaterally. Unilateral injection of 20 pmol of NMDA or 5 pmol of AMPA into the RVLM markedly (P < 0.01) increased BP by 34 ± 4 or 28 ± 4 mmHg,
respectively. The maximal changes in cardiovascular values after NMDA or AMPA injection were reached within 2–5 min and gradually returned to control level. Unilateral injection of 200 pmol of CNQX did not significantly alter basal BP but completely abolished increases in BP evoked by intra-RVLM AMPA injection. However, prior injection of the same dose of CNQX failed to affect the NMDA-induced increase in BP. The agonist-evoked changes in BP before and after pretreatment with CNQX are shown in Fig. 2B.

Effects of pretreatments with CNQX on clonidine-induced cardiovascular responses. A total of 34 rats (baseline MAP and HR: 96 ± 4 mmHg and 376 ± 17 bpm, respectively) were used to determine the role of the AMPA/kainate receptors in the RVLM in modulating the effect of clonidine. Figure 3 shows original tracings of BP, HR, and RSNA response to clonidine microinjected unilaterally into the RVLM 5 min after pretreatment with aCSF or CNQX. Unilateral injection of clonidine (5 nmol) into the RVLM following an equal volume (100 nl) of aCSF caused gradual reductions in BP, HR, and RSNA. Notably, the maximal decreases in BP, HR, and RSNA after RVLM clonidine were reached within 20 min and gradually returned to preinjection values within 40–60 min. The magnitudes of reductions in BP, HR, and RSNA evoked by intra-RVLM clonidine were significantly lower in the bilateral RVLM pretreated with 200 pmol of CNQX than in the RVLM pretreated with 100 nl of aCSF (Fig. 5). We also noted that, compared with intra-RVLM clonidine, the CNQX-induced (200 pmol) decreased responses of MAP (−3 ± 3 vs. −9 ± 3 mmHg), HR (−2 ± 5 vs. −15 ± 5 bpm), and RSNA (−9 ± 5 vs. −21 ± 5%) to intravenous clonidine were slightly lower.

Fig. 2. Effects of prior injection of 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX; 200 pmol) into the rostral ventrolateral medulla (RVLM) on the pressor action evoked by N-methyl-D-aspartate (NMDA; 20 pmol) and α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA; 5 pmol). A: representative tracings show that the pressor action evoked by AMPA but not by NMDA is abolished by treatment with CNQX injected into the RVLM. B: bar graphs summarize the mean data from 7 rats. BP, blood pressure; MAP, mean arterial pressure; HR, heart rate; bpm, beats/min; aCSF, artificial cerebrospinal fluid. Results are presented as means ± SE.
Effects of subsequent CNQX injection into the RVLM on clonidine-induced cardiovascular responses. A total of 32 rats (baseline MAP and HR: 96 ± 5 mmHg and 375 ± 16 bpm, respectively) were used to obtain further evidence to support the role of RVLM AMPA/kainate receptors in the effect of clonidine. Figure 6 shows original tracings of BP, HR, and RSNA responses to subsequent injection of CNQX into the RVLM 20 min after intra-RVLM or intravenous injection of clonidine. At the higher doses (20 and 200 pmol), subsequent injection of CNQX into the RVLM dose dependently reversed BP and RSNA 20 min after intra-RVLM clonidine (5 nmol) injection. The increases in BP and RSNA evoked by subsequent CNQX injection persisted for at least 20 min. Only at the highest dose did CNQX reverse clonidine-induced decreased HR. Interestingly, the highest dose (200 pmol) of CNQX injected after clonidine produced a strong excitatory effect on RSNA in three rats that significantly exceeded the baseline value. Subsequent injection of the lowest dose of CNQX (2 pmol) did not modify RVLM clonidine action compared with an equal volume of aCSF. The time course of changes in BP, HR, and RSNA elicited by subsequent injection of CNQX or aCSF after intra-RVLM clonidine is shown in Fig. 7. Similar to when clonidine was unilaterally injected into the RVLM, potent elevations in BP, HR, and RSNA were produced by
bilateral injection of CNQX (200 pmol, n = 6) but not 100 nl of vehicle (n = 3) 20 min after intravenous injection of clonidine (10 μg/kg) (mean data not shown).

**DISCUSSION**

The principal findings in this work are that pretreatment with injection of CNQX into the RVLM prevented the reductions in BP, HR, and RSNA evoked by RVLM injection of clonidine as well as by intravenous injection of clonidine. Furthermore, the reductions in BP, HR, and RSNA evoked by intra-RVLM or intravenous clonidine were significantly reversed by subsequent injection of CNQX into the RVLM. These results suggest that the AMPA/kainate receptors in the RVLM play an important role in modulating the central mechanism responsible for clonidine-induced cardiovascular and sympathetic inhibition.

It is well known that the glutamatergic system within the RVLM plays an important role in cardiovascular regulation (5, 29). Clonidine presynaptically modulates glutamate release in the RVLM but also attenuates the cardiovascualr responses evoked by RVLM NMDA receptor activation (20, 31). Recently, results from our laboratory (36, 37) and other laboratories (38) suggest that blockade of NMDA receptors in the RVLM significantly attenuates the decrease in BP and inhibition of RVLM presynaptic neurons induced by imidazoline-like drugs (clonidine and rilmenidine) in hypertensive or normotensive rats. These previous studies mainly focused on the role of NMDA receptors in modulating the effects of imidazoline-like drugs. It has been reported that, compared with NMDA receptors, the RVLM AMPA/kainate receptors may play a major role in control of cardiovascular function (1).

In this work, we observed the effects of blockade of the AMPA/kainate receptors in the RVLM on clonidine-induced decreases in BP, HR, and RSNA. At a high dose, CNQX is a potent AMPA/kainate receptor antagonist that also blocks the glycine modulatory site on the NMDA receptor complex (19). The present data showed that prior injection of 200 pmol of CNQX into the RVLM completely abolished the pressor action induced by AMPA (5 pmol) but had no effect on that of NMDA (20 pmol), suggesting that the dosage of CNQX (200 pmol) can selectively and adequately block AMPA receptors without affecting NMDA receptors. Although we did not further test the effect of CNQX on kainate receptors, a previous study (17) reported that a similar dose of CNQX did block the pressor action induced by kainate (4.7 pmol). Taken together, we believe that these studies indicate that 200 pmol of CNQX can adequately block the AMPA/kainate receptors without affecting NMDA receptors within the RVLM. In addition, the present data showed that injection of CNQX (2–200 pmol) into the RVLM had no effect on basal BP, HR, and RSNA. The fact that we only took 5 min to observe the effect of intra-RVLM CNQX may be a limitation. A longer period of time (60 min) for the control test of CNQX alone would be useful to determine its effects and for comparison with the clonidine actions. However, it has been demonstrated that, in the normal state, injection of glutamate receptor antagonists into the RVLM does not affect baseline cardiovascular activity (10, 16). It appears that excitatory amino acid receptors in the RVLM do not participate in generation of tonic cardiovascular activity. Interestingly, it has been suggested that, in normal baseline, the balance between excitatory and inhibitory influences may prevent the change in BP following simultaneous removal of these two influences by glutamate antagonists (29). Following removal of inhibitory input to the RVLM or in the hypertensive state, injection of glutamate receptor antagonists into the RVLM significantly decreases basal BP (13, 14).

Similar to previous studies (6, 7), we found that clonidine injected unilaterally into the RVLM produced long-lasting hypotensive, bradycardiac, and sympathoinhibitory effects. Importantly, the decreases in BP, HR, and RSNA evoked by clonidine injected into the RVLM were significantly attenuated after pretreatment with CNQX in a dose-dependent manner.
Although 2 pmol of CNQX did not affect the clonidine action, it is possible that this dosage of CNQX was too low to effectively block the AMPA/kainate receptors. Similar to RVLM injection of clonidine, we further verified that hypotensive, bradycardic, and sympathoinhibitory effects evoked by systemic clonidine were significantly attenuated by blockade of the AMPA/kainate receptors in the RVLM. We also found that, compared with the intra-RVLM clonidine group, the degree of CNQX-induced (200 pmol) attenuation of MAP, HR, and RSNA in response to intravenous clonidine was slightly lower. Although the RVLM is a major area involved in the central depressor mechanism of clonidine, the RVLM may be not the sole target area modulating the central effects of systemic clonidine. It has been reported that other central areas such as the locus ceruleus are also sensitive to clonidine (27). It is possible that, in addition to the RVLM, AMPA/kainate receptors in other areas contribute to intravenous clonidine. Taken together, these findings strongly suggest that the cardiovascular and sympathetic inhibition evoked by local RVLM or systemic clonidine depended on the functional state of the AMPA/kainate receptors in the RVLM. In support of this idea, we also determined whether the cardiovascular and sympathetic inhibition evoked by clonidine can be reversed by subsequent injection of CNQX into the RVLM. Subsequent injection of CNQX into the RVLM after intra-RVLM or intravenous injection of clonidine produced a strong excitatory effect on cardiovascular and sympathetic activity. Because blockade of the AMPA/kainate receptors in the RVLM did not affect the basal BP and RSNA, these data raised the suggestion that clonidine significantly changed the function of the RVLM AMPA/kainate receptors and then caused the changes in cardiovascular and sympathetic activity. Notably, these results are very similar to the role of NMDA receptors in the RVLM in modulating the cardiovascular inhibition evoked by the imidazoline-like drugs clonidine and rilmenidine (15, 35, 36, 38). We speculate that both NMDA and AMPA/kainate receptors in the RVLM contribute to cardiovascular inhibition evoked by imidazoline-like drugs. In fact, the clonidine action was mostly abolished following the high-dose CNQX injection into the RVLM. The relative strength or importance of NMDA and AMPA/kainate receptors in modulating the central depressor mechanism is not clear from the present data. It has been suggested that, compared with NMDA receptors, AMPA/kainate receptors in the RVLM play a major role in controlling cardiovascular activity (1). This may be a reason why the RVLM CNQX seems to exhibit a stronger blockade effect on clonidine action. Another possibility is that there exist different distribution densities of NMDA or AMPA receptors in the sub-area of the RVLM. Therefore, it is possible that the relative strength of NMDA or AMPA receptors in modulating clonidine action is dependent on the distribution density of NMDA or AMPA receptors in the RVLM. Confirmation of this hypothesis requires additional study.

Although the present study shows that the hypotensive and sympathoinhibitory effects of clonidine are closely related to the functional states of the AMPA/kainate receptors in the RVLM, its physiological significance and exact mechanism remain to be defined. Because CNQX and clonidine act on their specific receptors, the interaction between CNQX and clonidine probably is a physiological antagonism rather than a pharmacological antagonism. This interaction may occur at many levels such as presynaptic, postsynaptic, acting receptor, and intracellular signal conduction mechanism, etc. There are several possible mechanisms underlying the interaction between AMPA/kainate receptors and clonidine. First, it has been reported that clonidine effectively stimulates the spontaneous release of the sympathoinhibitory transmitter γ-aminobutyric acid (GABA) in the RVLM, which is associated with enhancement of glutamate release (31). It is well known that increased GABA within the RVLM produces a significant sympathoinhibition (4, 5, 18). Acute and chronic clonidine application is also suggested to enhance the GABA content in rat brain, and GABA receptor blockade can attenuate the hypotension elicited by clonidine (3, 15). Stimulation of glutamate receptors increases GABA release and facilitates the GABAergic synaptic activity (25). Furthermore, activation of AMPA/kainate receptors is able to trigger GABA release (25, 26). The view that the subsequent release of GABA is associated with activation of AMPA/kainate receptors may explain why the cardiovascular inhibition evoked by clonidine is converted to an excitation after RVLM injection of the highest dose of CNQX (200 pmol) in the present study. However, there is no direct evidence to clarify the interaction between the functional states of the RVLM GABA receptor and clonidine action. Second, it has been reported that the apparent desensitization caused by the dopamine D2 receptor agonist quinpirole is specific to certain sympathetic vasomotor pathways, including those that mediate the antihypertensive action of clonidine, rilmenidine, and α-methyl dopa (32, 33). Therefore, it is not clear whether CNQX produces the desensitization of neuronal excitation in response to clonidine and subsequently attenuates the effects of clonidine. However, there is a significant difference between these two pretreatments, because the basal BP can be elevated by quinpirole but not by CNQX. Finally, it has been reported that the imidazoline compounds interact with a specific site (such as the phencyclidine-binding site) on glutamate receptors (23). Zhang et al. (38) also suggest that a common binding site on the NMDA receptor recognized by NMDA and I1R agonist contributes to the central antihypertensive mechanism. The possibility is that, on AMPA/kainate receptors, there exists a similar binding site recognized by CNQX and clonidine. Another important question is whether I1R or α2AR contributes to mediation of processing of the interaction between glutamate receptors and clonidine actions. Clonidine is a mixed agonist for I1R and α2AR, but their specific antagonists were not used in this work to further characterize which receptor is responsible for mediating the interaction between clonidine and AMPA/kainate receptors. It is reported that the effect of clonidine on the release of glutamate and GABA is prevented by the selective α2AR antagonist yohimbine (15). On the other hand, the hypotension induced by injection of the selective I1R agonist rilmenidine into the RVLM is abolished by an NMDA receptor antagonist (38). Considering the evidence, it is likely that clonidine action is dependent on the glutamatergic system via a mechanism of I1R or α2AR or both. The relationship between AMPA/kainate receptors and the effects of imidazoline-like drugs is more complex and awaits further investigation.

In summary, the present results show that the decreases in BP, HR, and RSNA evoked by clonidine are significantly prevented by blockade of AMPA/kainate receptors in the RVLM. It is concluded that AMPA/kainate receptors in the
RVLM contribute to clonidine-induced cardiovascular and sympathetic inhibition. However, because of the present data obtained from the normotensive rats, it is not known whether the same relationship exists in the hypertensive model. Some receptors, such as glutamate receptors in the brain, are upregulated in the hypertensive model, and therefore it is also important to determine the relationship between glutamate receptors and the effects of imidazole-like drugs in the hypertensive state.

ACKNOWLEDGMENTS

We are deeply grateful to Allison Kleiber for kind assistance with English writing.

GRANTS

This work was supported by the National Natural Science Foundation of China (grant nos. 30470636 and 30670759). This study was also funded by National Heart, Lung, and Blood Institute Grants RO-1-HL-077691 and PO-1-HL-62222 and a postdoctoral fellowship to W.-Z. Wang from the American Heart Association, Heartland Affiliate (no. 0720066Z).

REFERENCES