PHARMACOLOGY AND TOXICOLOGY

Role of I_1-Imidazoline Receptors and α_2-Adrenoceptors in Hemodynamic Effects of Moxonidine Administration into the Rostroventrolateral Medulla

O. N. Khokhlova, A. N. Murashev, and O. S. Medvedev

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 131, No. 4, pp. 399-402, April, 2001
Original article submitted September 27, 2000

Local injection of 4 nmol moxonidine (unilaterally) into the rostroventrolateral medulla of spontaneously hypertensive rats (SHR-SP) decreased mean blood pressure and heart rate by 24±3 and 3±4%, respectively. Pretreatment with the I_1/α_2-receptor antagonist efaroxan abolished the moxonidine-induced decrease in mean blood pressure, but had no effect on heart rate. Yohimbine blocked hypotension, delayed bradycardia (8 nmol), or completely inhibited the effects of moxonidine (16 nmol). Our results indicate that both I_1-imidazoline receptors and α_2-adrenoceptors of the rostroventrolateral medulla are involved in the realization of moxonidine-induced changes.

Key Words: *moxonidine; I_1-imidazoline receptors; α_2-adrenoceptors; rostroventrolateral medulla*

Moxonidine is a new hypotensive drug, whose action is mediated via the central nervous system. Moxonidine cause much lower side effects compared to first-generation drugs [12], which is probably related to its selective interaction with I_1-imidazoline receptors localized primarily in the rostroventrolateral medulla (RVLM) [1,5]. According to the "I_1 theory", the antihypertensive effect of moxonidine is related to activation of I_1-imidazoline receptors in RVLM [3,4, 7]. Other authors reported that α_2-adrenoceptors play the major role in moxonidine-induced hypotensive effects [6,13]. Here we studied the effect of blockade of I_1-imidazoline receptors and α_2-adrenoceptors in RVLM on hemodynamic changes caused by moxonidine administration into this brain area.

M. M. Shemyakin and Yu. A. Ovchinnikov Institute of Bioorganic Chemistry (Pushchino Branch), Russian Academy of Sciences; Department of Pharmacology, Faculty of Fundamental Medicine, M. V. Lomonosov Moscow State University. ***Address for correspondence:*** khohlova@fibkh.serpukhov.su. Khokhlova O. N.

MATERIALS AND METHODS

Experiments were performed on 6-7-month-old male spontaneously hypertensive stroke-predisposed rats (SHR-SP, Institute of Bioorganic Chemistry). The animals were intraperitoneally narcotized with 1.25 g/kg urethane.

Heart rate (HR) and mean blood pressure (BP_m) were measured after the injection of 4 nmol moxonidine (Farmzashchita) into RVLM after blockade of I_1-imidazoline receptors or α_2-adrenoceptors with efaroxan (4 nmol, Sigma) or yohimbine (8 and 16 nmol, Sigma), respectively. Control rats were injected with moxonidine after pretreatment with Ringer solution or received receptor antagonists without moxonidine. BP_m and HR were measured for 10 min before (control) and after administration of efaroxan or yohimbine and for 60 min after injection of moxonidine.

BP was measured using a polyethylene catheter implanted into the femoral vein and connected to a SR-01 electromanometer (STS). The results were ana-

0007-4888/04/1314-0336$25.00

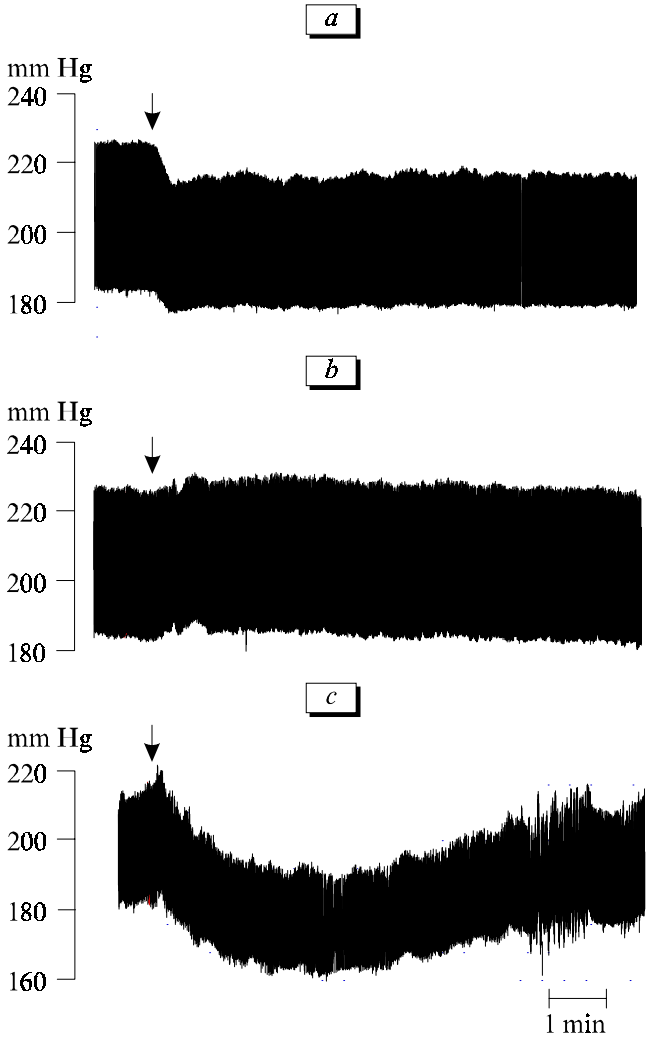

Fig. 1. Effects of 8 nmol yohimbine (*a*) and 4 (*b*) or 8 nmol efaroxan (*c*) injected into the rostroventrolateral medulla in SHR-SP rats. Arrows: injection.

lyzed on MacLab/4S (ADInstruments Ltd.) setup using Chart 3.5.6/s software.

Microinjections (0.5 μl) into RVLM were performed in a stereotaxic apparatus using glass capillaries (tip diameter 25-30 μ). The capillaries were introduced through the atlantooccipital membrane (dorsal approach) according to stereotaxic coordinates of this brain area [9]. Proper position of capillaries was confirmed by hypertensive reactions to 2 nmol sodium L-glutamate (Fluka) [7,10]. Histological verification was performed in cryostat sections of the medulla oblongata by the location of neutral red spot: 2% dye (Sigma) was injected via capillaries after the experiment [10].

Preliminary experiments with various doses of I_1/α_2 receptor antagonist efaroxan and α_2-adrenoceptor antagonist yohimbine showed that yohimbine decreased BP (Fig. 1). This effect was also observed after injection of other α_2-adrenoceptor antagonists into RVLM [7,11]. Efaroxan in a dose of 8 nmol caused hypotension (similarly to other α_2-adrenoceptor antagonists), while in a dose of 4 nmol this drug slightly increased BP. Therefore, efaroxan in a dose of 4 nmol selectively blocked I_1-imidazoline receptors.

The data are presented as *M*±*SEM*. Intergroup differences were evaluated using Mann—Whitney and ANOVA-2 tests. The differences were significant at $p<0.05$.

RESULTS

In control rats injected with Ringer solution, moxonidine decreased BP_m and HR (Table 1). These effects were most pronounced on minutes 60 (24±3%) and 20 (13±4%) postinjection, respectively (Fig. 2). Efaroxan did not change BP_m, but increased HR by 8±2% 30 min after injection (Fig. 2). Pretreatment with efaroxan completely blocked moxonidine-induced hypotension, but had no effect on heart rate: bradycardia was most pronounced 10 min after moxonidine administration (18±6%). Yohimbine in a dose of 8 nmol slightly decreased BP_m and HR (insignificant, Fig. 3, *a*, *b*). Pretreatment with 8 nmol yohimbine decreased the degree of moxonidine-induced hypotension; bradycardia was delayed by 10 min. Yohimbine in a dose of 16 nmol more significantly decreased BP_m and HR (Fig. 3, *c*, *d*) and completely inhibited the effect of moxonidine.

Efaroxan in doses much lower than those of yohimbine inhibited the moxonidine-induced decrease in BP. These results indicate that I_1-imidazoline receptors play the major role in the realization of hypotensive effects of moxonidine. It should be emphasized that efaroxan displayed different potencies in preventing hypotension and bradycardia, which indicated that the moxonidine-induced decrease in HR is primarily me-

TABLE 1. Initial BP_m and HR in SHR-SP Rats

Experimental conditions	BP_m, mm Hg	HR, bpm
Ringer solution+moxonidine (4 nmol), *n*=7	210±2	298±9
Efaroxan (4 nmol)+ moxonidine, *n*=7	216±4	297±17
Efaroxan (4 nmol)+ Ringer solution, *n*=6	210±8	305±17
Yohimbine (8 nmol)+ moxonidine, *n*=7	212±4	299±14
Yohimbine (8 nmol)+ Ringer solution, *n*=7	207±4	310±10
Yohimbine (16 nmol)+ moxonidine, *n*=7	207±5	292±10
Yohimbine (16 nmol)+ Ringer solution, *n*=6	206±5	294±15

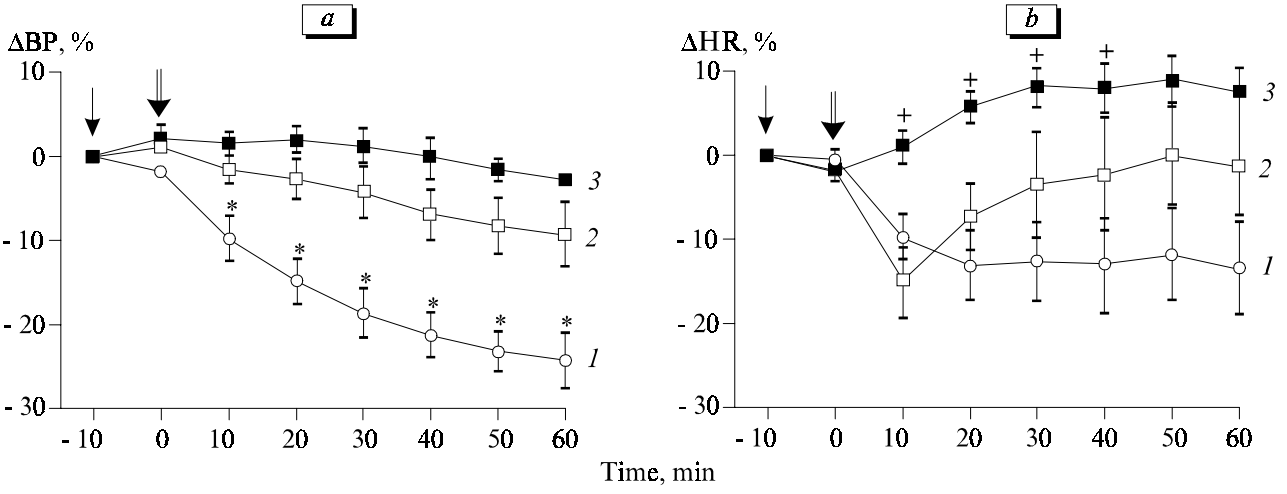

Fig. 2. Changes in mean BP (*a*) and HR (*b*) after injection moxonidine into the rostroventrolateral medulla in rats pretreated with efaroxan: Ringer solution+moxonidine (*1*), efaroxan+moxonidine (*2*), and efaroxan+Ringer solution (*3*). Arrows: injection of efaroxan or Ringer solution. Double arrows: injection of moxonidine or Ringer solution. Here and in Fig. 3: **p*<0.05 compared to group 2.

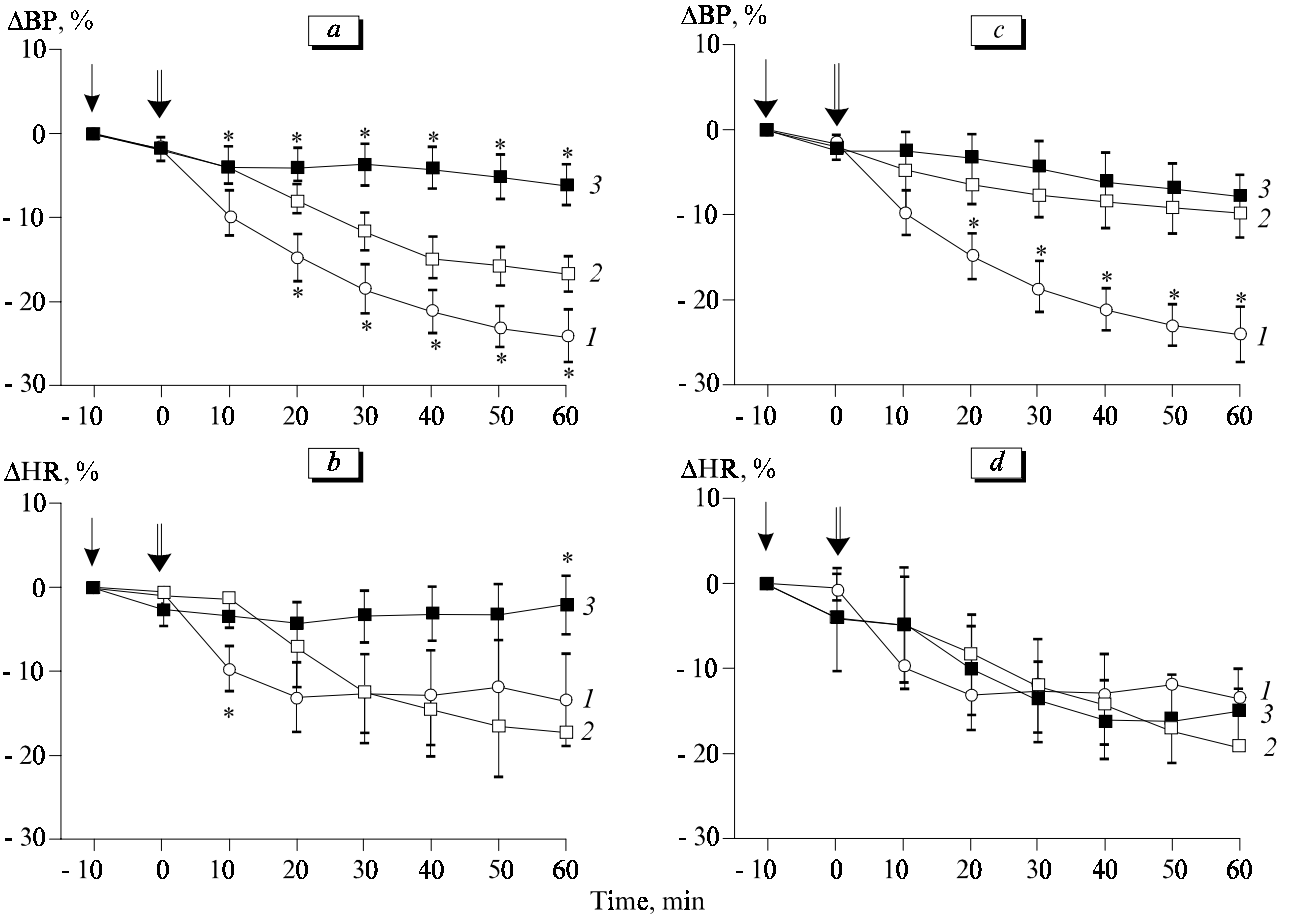

Fig. 3. Changes in mean BP (*a, c*) and HR (*b, d*) after injection moxonidine into the rostroventrolateral medulla in rats pretreated with yohimbine in doses of 8 (*a*, *b*) and 16 nmol (*c*, *d*): Ringer solution+moxonidine (*1*), yohimbine+moxonidine (*2*), and yohimbine+Ringer solution (*3*). Arrows: injection of yohimbine or Ringer solution. Double arrows: injection of moxonidine or Ringer solution.

diated via activation of α_2-adrenoceptors. Previous studies showed that RVLM performs selective and differential control over effectors involved in the cardiovascular regulation [2]. Therefore, it can not be excluded that vasodepressor and negative chronotropic effects of moxonidine in RVLM are realized via different pathways involving neurons with various pharmacological properties. It was also hypothesized that

I_1-imidazoline receptors and α_2-adrenoceptors are involved in the realization of effects of imidazoline derivatives [8]. Our experiments do not demonstrate the type of interaction between these receptors in RVLM, but our findings suggest that both I_1-imidazoline receptors α_2-adrenoceptors are involved in the realization of hemodynamic changes caused by moxonidine administration into RVLM.

This work was supported by the Russian Foundation for Basic Research (grant No. 98-04-49413).

REFERENCES

1. G. Bricca, M. Dontenwill, A. Molines, *et al*., *Eur. J. Pharmacol*., **162**, 1-9 (1989).
2. R. A. L. Dampney, *Physiol. Rev*., **74**, No. 2, 323-364 (1994).
3. P. Ernsberger, T. H. Damon, L. M. Graff, *et al*., *J. Pharmacol. Exp. Ther*., **264**, 172-182 (1993).
4. P. Ernsberger, J. E. Friedman, and R. J. Koletsky, *J. Hypertens*., **15**, Suppl.. 1, S9-S23 (1997).
5. P. Ernsberger, M. P. Meeley, J. J. Mann, and D. J. Reis, *Eur. J. Pharmacol*., **134**, 1-13 (1987).
6. P. G. Guyenet, *Am. J. Physiol*., **273**, R1580-R1584 (1997).
7. M. A. Haxhiu, I. Dreshaj, S. G. Schafer, and P. Ernsberger, *J. Cardiovasc. Pharmacol*., **24**, Suppl. 1, S1-S8 (1994).
8. G. A. Head, C. K. Chan, and S. L. Burke, *J. Auton. Nerv. Syst*., **72**, Nos. 2-3, 163-169 (1998).
9. G. Paxinos and C. Watson, *The Rat Brain in Stereotaxic Coordinates*, San Diego (1997).
10. S. Sesoko, H. Muratani, M. Yamazato, *et al*., *Am. J. Physiol*., **274**, R1119-R1124 (1998).
11. R. L. Stornetta, D. Huangfu, D. L. Rosin, *et al*., *Ann. N. Y. Acad. Sci*., **763**, 541-551 (1995).
12. J. Webster and H. F. Koch, *J. Cardiovasc. Pharmacol*., **27**, Suppl. 3, S49-S54 (1996).
13. Q. M. Zhu, J. D. Lesnick, J. R. Jasper, *et al*., *Br. J. Pharmacol*., **126**, No. 6, 1522-1530 (1999).