PHYSIOLOGY

Respiratory Effects of Stimulation of the Limbic Cortex in Rats and Their Modulation with Serotonin

O. A. Vedyasova

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 140, No. 9, pp. 244-246, September, 2005
Original article submitted November 16, 2004

Acute experiments on rats showed that the anterior cingulate gyrus contains 2 efferent regions of the functional respiratory system (inhibitory supragenual and excitatory infragenual areas). Stimulation of these cortical areas produced a respiratory effect, which depended on activity of serotoninergic mechanisms in the solitary tract nucleus.

Key Words: *limbic cortex; solitary tract nucleus; serotonin; respiration*

The cingulate gyrus (CG) and limbic cortex (LC) are the major suprabulbar structures involved in the respiratory regulation. The anterior part of CG mainly produces an the inhibitory effect on respiration [4]. Stimulation of LC produces a variety of respiratory reactions, which is determined by its viscerotopic organization and direct or indirect (via diencephalic structures) transduction of cingulofugal signals to nuclei of the vagosolitary complex [2,5]. Taking into account complex synaptology and polychemical composition of LC [14] and respiratory center [6,10,15] we can hypothesize that the respiratory response to CG stimulation depends on the involvement of various endogenous regulators in the cingulorespiratory relations [3]. Much attention is given to serotonin, which is present in all respiratory nuclei [9] and first modulates the respiratory rhythm during ontogeny [7,8]. Here we studied the role of serotoninergic mechanisms in the solitary tract nucleus (STN) in the realization of LC-mediated efferent influences on respiration in rats.

MATERIALS AND METHODS

Acute experiments were performed on 24 outbred rats intraperitoneally narcotized with 75 mg/kg nembutal. The respiratory effects of anterior CG electrostimulation were studied before and after local administration of 3×10^{-9} M serotonin into the ventrolateral subnucleus of STN. Serotonin (0.2 μl) was injected stereotactically through a glass micropipette (tip diameter 20 μ) using an MSh-1 microsyringe [13]. CG was stimulated with a series of electric pulses delivered through bipolar silver electrodes (100-300 μA, pulse 0.5 msec duration, 50-100 Hz frequency, 5-10 sec series duration). The respiratory effects were evaluated by a change in frequency-amplitude characteristics of external respiration and electromyogram (EMG) of the diaphragm. External respiration was recorded on a miniature spirograph connected to the tracheotomy tube. EMG was registered with bipolar electrodes, converted, and recorded on an H-338-6 automatic recorder (integrated curve). The results were analyzed by Student's *t* test (SigmaStat 2.0 software, Jandel Corp.).

Department of Human and Animal Physiology, Samara State University

RESULTS

Stimulation of anterior CG in rats produced 2 types of respiratory responses. Depending on changes in external respiration and diaphragm EMG, they can be divided into inhibitory and facilitating responses. The inhibitory responses were mainly observed during stimulation of the supragenual (dorsal) area in anterior

0007-4888/05/1403-0265

CG and manifested (judging from spirography) in significant lengthening of the respiratory cycle due to prolongation of the expiratory phase (by 41.8%, $p<0.001$, Fig. 1, *1*). Parameters of inspiration (duration of inspiration and respiratory volume) only little changed under these conditions. The study of integral parameters for the respiratory pattern revealed deceleration of breathing rhythm (by 40.2%, $p<0.05$) and decrease in cycle efficiency and pulmonary ventilation; minute respiratory volume decreased by 1.5 times in some animals. The inhibitory effect of supragenual stimulation on respiration was verified by changes in electrical activity of the diaphragm muscle (Fig. 1, *2*).

Electrostimulation of the infragenual (ventral) area in anterior CG had a more pronounced respiratory effect. It was mainly the excitatory response. The study of integral EMG from the diaphragm (Fig. 1, *3*) showed that typical response manifested in lengthening and increase in the amplitude of inspiratory bursts (by 25.2 and 53.4%, respectively, $p<0.01$). These changes reflect increased effectiveness of inspiration. By contrast, the interburst interval correlating with the expiratory phase decreased under these conditions. Changes in temporal characteristics of EMG were accompanied by an increase in estimated values of the inspiratory ratio and respiratory rate. It was associated with the increase in central inspiratory activity.

Microinjection of serotonin into STN modulated the respiratory response to stimulation of LC. It should be emphasized that this mediator had different modulatory effects on the influences from various areas of CG. Activation of serotoninergic structures in STN was accompanied by an increase in the inhibitory effect of the supragenual area on the respiratory cycle (Fig. 2, *1* and *2*). Pharmacological treatment reduced the facilitating effect of infragenual stimulation. Stimulation of the infragenual area inhibited respiration

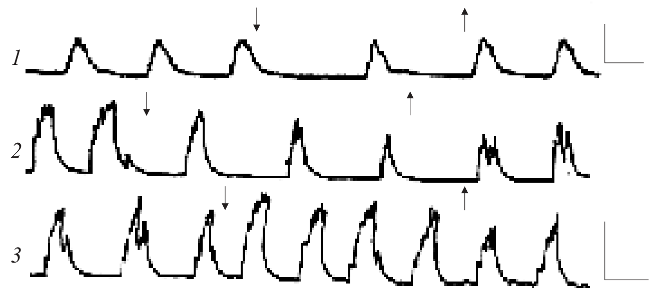

Fig. 1. Respiratory response to stimulation of rat limbic cortex before microinjection of serotonin into the solitary tract nucleus. Here and in Fig. 2: spirogram and integrated EMG from the diaphragm during stimulation of the supragenual area (*1*, *2*); integrated EMG from the diaphragm during stimulation of the infragenual area (*3*). Arrows: start and end of stimulation. Calibration: spirogram, 1 ml and 1 sec; EMG, 100 μV and 1 sec.

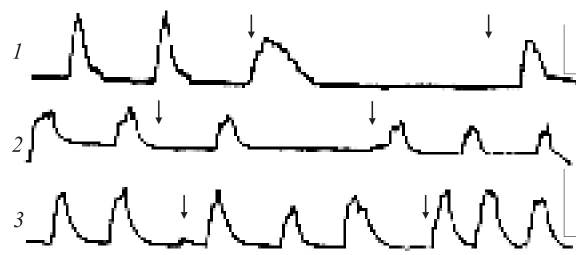

Fig. 2. Respiratory response to stimulation of rat limbic cortex after microinjection of serotonin into the solitary tract nucleus.

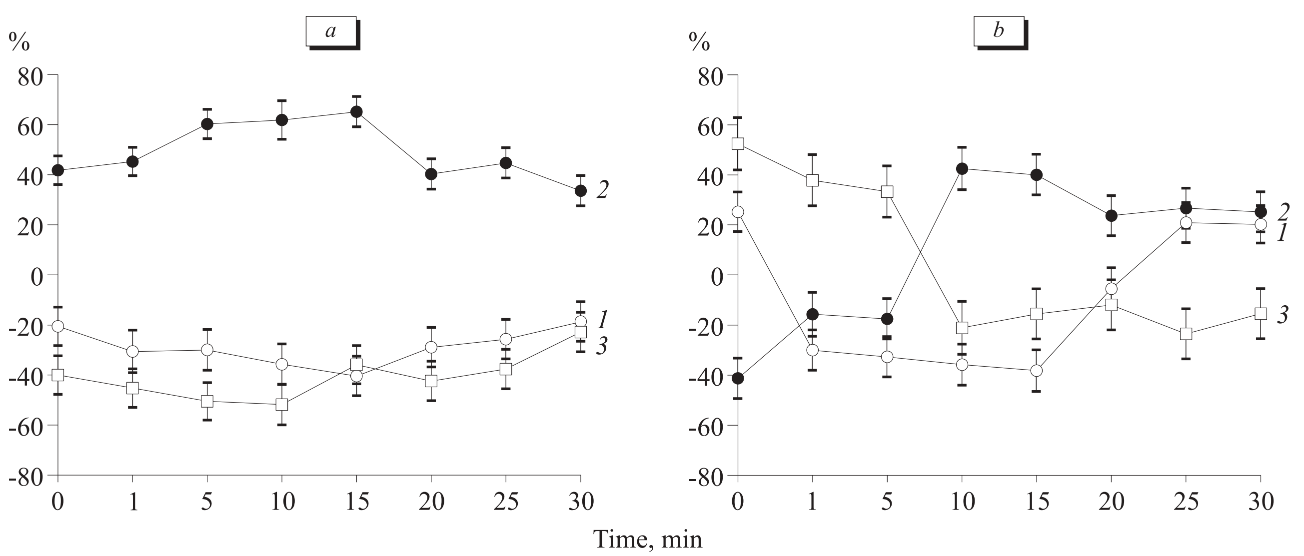

Fig. 3. Changes (% of the basal level) in parameters of external respiration (*a*) and diaphragm EMG (*b*) produced by stimulation of supragenual and infragenual areas in the limbic cortex, respectively, before and over 30-min period after serotonin microinjection into the solitary tract nucleus. *a*: duration of inspirition (*1*), duration of expiration (*2*), and respiratory rate (*3*); *b*: Duration of inspiratory burst (*1*), interburst interval (*2*), and respiratory rate (*3*).

under these conditions (Fig. 2, *3*). The modulatory effect of serotonin depended on the time of exposure and peaked 10-15 min after its injection into STN. In this period the effect of LC stimulation was opposite to that observed before microinjection (Fig. 3).

Our results are consistent with published data on spatial distribution and relations between visceral systems in the cerebral cortex [1,2]. It is necessary to revise the notion that anterior CG serves as a source of mainly inhibitory influences on the respiratory center. The anterior part of the cingulate cortex in rats and cats [2] probably includes two efferent regions of the functional respiratory system (inhibitory supragenual and excitatory infragenual areas). Stimulation of these areas in LC had different effects on respiration. It can be hypothesized that the infragenual area is characterized by higher density of efferent neurons sending impulses to premotor structures in STN. These structures provide the integral output to spinal centers of respiratory muscles. The respiratory response to LC stimulation depended on activity of serotoninergic mechanisms in STN. Serotonin acting as a synaptic transmitter in respiratory neurons [7-9] is probably involved in the realization of suprabulbar influences on the respiratory center. Activation of inspiratory neurons in STN and increase in the respiratory rate with serotonin are realized via 5-HT_{1A} receptors [11]. Serotonin causes depolarization in expiratory and post-inspiratory neurons and decreases the respiratory rate. These effects are mediated by 5-HT_2 receptors [12]. Our findings suggest that that serotonin modulates basal activity of respiratory neurons in STN, changes their sensitivity to external regulatory influences, and affects the respiratory response to LC stimulation. It cannot be excluded that cingulofugal projections enter the area of STN with high density of 5-HT_2 receptors, which determines mainly the inhibitory effect of CG.

REFERENCES

1. V. G. Aleksandrov and N. P. Aleksandrova, *Ros. Fiziol. Zh.*, **84**, No. 4, 316-322 (1998).
2. O. G. Baklavadzhyan, L. B. Nersesyan, E. A. Avetisyan, *et al.*, *Usp. Fiziol. Nauk*, **31**, No. 4, 11-23 (2000).
3. O. A. Vedyasova and Yu. P. Senyutina, *Physiology of the Organism under Normal and Extreme Conditions* [in Russian], Tomsk (2001), pp. 352-354.
4. N. L. Mikhailova, *Modern Problems of Respiratory Physiology* [in Russian], Kuibyshev (1987), pp. 114-122.
5. S. S. Panteleev, V. A. Bagaev, and O. A. Lyubashina, *Ros. Fiziol. Zh.*, **83**, No. 4, 33-44 (1997).
6. I. A. Tarakanov, V. A. Safonov, and L. N. Tikhomirova, *Byull. Eksp. Biol. Med.*, **128**, No. 9, 274-278 (1999).
7. E. Di Pasquale, R. Monteau, and G. Hilaire, *Dev. Brain Res.*, **8**, Nos. 1-2, 222-232 (1994).
8. M. A. Haxhiu, F. Tolentino-Silva, G. Pete, *et al.*, *Respir. Physiol.*, **129**, 191-209 (2001).
9. G. Hilaire and B. Duron, *Physiol. Rev.*, **79**, No. 2, 325-360 (1999).
10. D. D. Kline, K. N. Takacs, E. Ficker, and D. L. Kunze, *J. Neurophysiol.*, **88**, 2736-2744 (2002).
11. P. Lalley, R. Benacka, A. Bischoff, and D. Richter, *Brain Res.*, **747**, 156-159 (1997).
12. P. Lalley, A. Bischoff, S. Schwarzacher, and D. Richter, *J. Physiol. (Lond.)*, **487**, 653-661 (1995).
13. G. Paxinos and G. Watson, *The Rat Brain in Stereotaxic Coordinates*, San Diego (1997).
14. J. A. Rosenkranz and A. A. Grace, *J. Neurosci.*, **22**, No. 1, 324-337 (2002).
15. E. J. Zuperku and D. R. McCrimmon, *Respir. Physiol. Neurobiol.*, **131**, 121-133 (2002).