Effects of Euphylline on Breathing Pattern and Chemosensitivity of the Respiratory System after Activation of $GABA_b$-Receptors

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 149, No. 4, pp. 383-387, April, 2010
Original article submitted August 3, 2009

We studied changes in breathing pattern in nembutal-anaesthetized mongrel rats after administration of euphylline against the background of preliminary treatment with lithium hydroxybutyrate. Two types of external respiration responses to euphylline were observed; they depended on the initial blood pressure in systemic circulation and on its drop after euphylline administration. Thus, the reaction of the respiratory system to adenosine receptors blockade against the background of hydroxybutyrate pretreatment was associated with not only the effect of euphylline, but also the state of brain hemodynamics. The effects of euphylline on chemosensitive contour of the respiratory system regulation were also investigated. It was found that euphylline did not abolish desensitization of respiratory system to hypercapnia, but smoothed the response to hypoxia under conditions of $GABA_b$-receptor activation.

Key Words: *euphylline; respiration; chemsensing; hemodynamics; oxybutirate*

Adenosine, a product of degradation of macroergic compounds formed during brain hypoxia, acts as a tonic neuromodulator of respiration and may produce various effects depending on the site of its action and type of adenosine receptors to be activated [6,13]. The purinergic system considerably modulates respiration in not only fetuses and newborns [4], but also adults [7]. Hypoxia increases adenosine release in the brain [8,9]; endogenous adenosine is probably involved in respiratory depression during hypoxia, which follows the phase of initial activation. Respiratory suppression caused by hypoxia was eliminated by adenosine receptor blockers [10]. Theophylline, an adenosine receptor blocker, abolished suppression of respiration after administration of adenosine analogue and even improved lung ventilation when administered systemically [7,14]. Available data suggest that the purinergic system is actively involved in central regulation of respiration: in respiratory rhythm generation and in response of the respiratory system to hypoxia.

The objective of this study was to evaluate the involvement of adenosine receptors in respiratory rhythm disturbances caused by activation of $GABA_b$-receptors and the effects of adenosine receptor antagonist euphylline on the function of chemosensitive contour of respiratory system regulation under these conditions.

MATERIALS AND METHODS

The animals were kept in a vivarium on standard diet with food and water *ad libitum* under 12-hour light regimen. The experiments were carried out on 32 nembutal-anesthetized (40 mg/kg intraperitoneally) mongrel albino male rats weighting 350-400 g. Body temperature was maintained with 1°C accuracy within the range 37.0-38.5°C using an infrared lamp. Tracheotomy was performed at the level of upper third of the trachea, thereafter a plastic tube with suitable diameter was inserted and fixed. Then, this respiratory cannula

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0007-4888/10/1494-0400

was connected to a transducer to measure external respiration parameters. Distal end of the transducer was also connected to a valve for air and gas mixture supply. The main respiratory parameters, such as minute volume of respiration (MVR), respiration rate (RR), respiratory volume (RV), and pneumotachogram were recorded using block for respiration parameter registration on an MX-01 polygraph under BTPS conditions. Systemic blood pressure (BP) and heart rate (HR) were measured with a catheter inserted into the femoral artery and connected to tensiometric transducer. Intraesophageal pressure (IEP) was measured with another catheter with a water-filled elastic balloon introduced into the esophagus and connected to an MX-01 transducer. Pressure in the esophageal catheter was set in such a way that during passive expiration it was 0 mm Hg.

Blood flow velocity in the ascending aorta was measured using a miniature ultrasonic transducer fixed at the end of a catheter 0.6 mm in diameter [2]. Blood flow and BP data were inputted into an analog-computing device to assess peripheral resistance (PR) and cardiac output dynamics.

Registration of experimental data was carried out using a N3031-6 ink-recorder. The data was statistically analyzed using Student's t test ($p<0.05$). Chemosensitivity of the respiratory system was tested by inhalation of gas mixtures for 5 min. First, the animal breathed a mixture with 5% CO_2 and after 10 min hypoxic mixture with 10% O_2 in nitrogen. Then lithium hydroxybutyrate was administered (750 mg/kg intravenously) and changes in respiratory rhythm were observed after intravenous administration of 20 mg/kg euphylline (aminophylline). The effects of euphylline on chemosensitive pathway of respiratory system regulation were also investigated by repeated testing with gas mixtures.

RESULTS

The pattern of respiratory rhythm changes after systemic euphylline administration was investigated. In control animals, euphylline produced no significant changes in breathing pattern, increase in RR was noted in some animals. Euphylline effect was more pronounced after preliminary systemic administration of $GABA_b$-agonist lithium hydroxybutyrate inducing pathological periodic breathing in rats [3].

First, the dynamics of respiratory and hemodynamic parameters was assessed for 45 min after lithi-

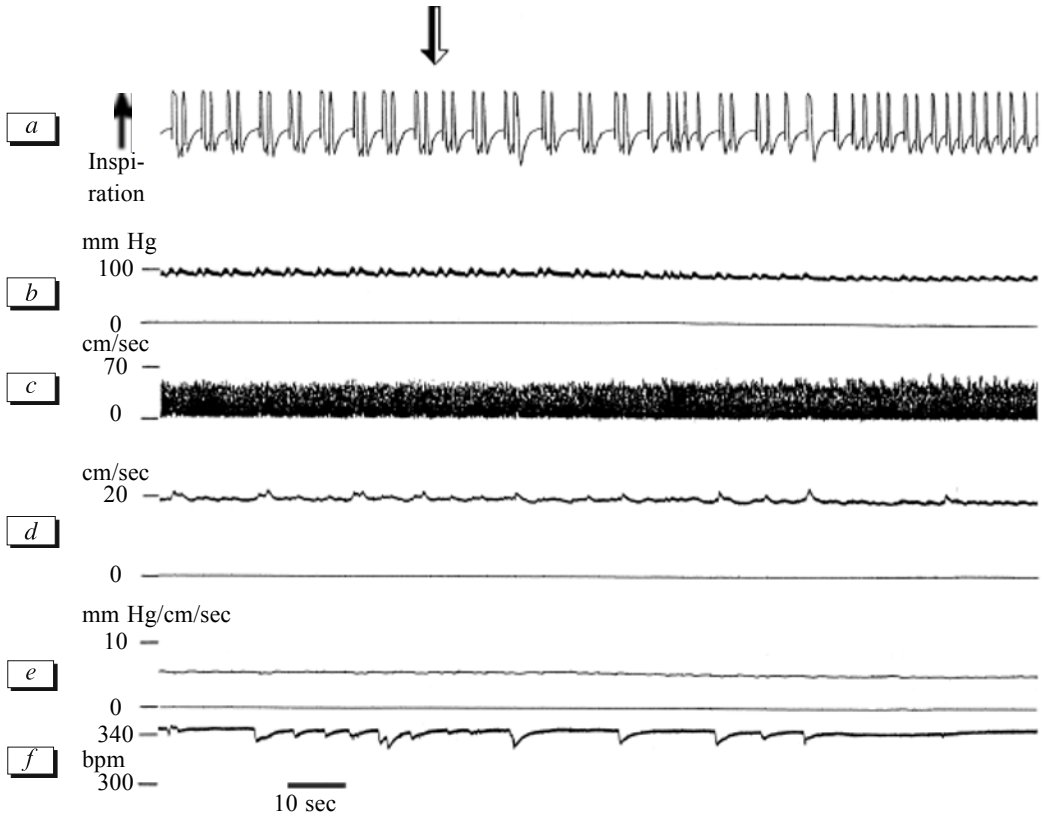

Fig. 1. Parameters of hemodynamics and respiration during development of periodic breathing 30 min after lithium hydroxybutyrate administration and during recovery of rhythmic breathing after euphylline administration (arrow). Here and on Fig. 2: *a*) respiratory movements, *b*) BP, *c*) linear blood flow velocity in the ascending aorta, *d*) cardiac output, *e*) peripheral resistance, *f*) HR. Lines under the curves: baseline levels.

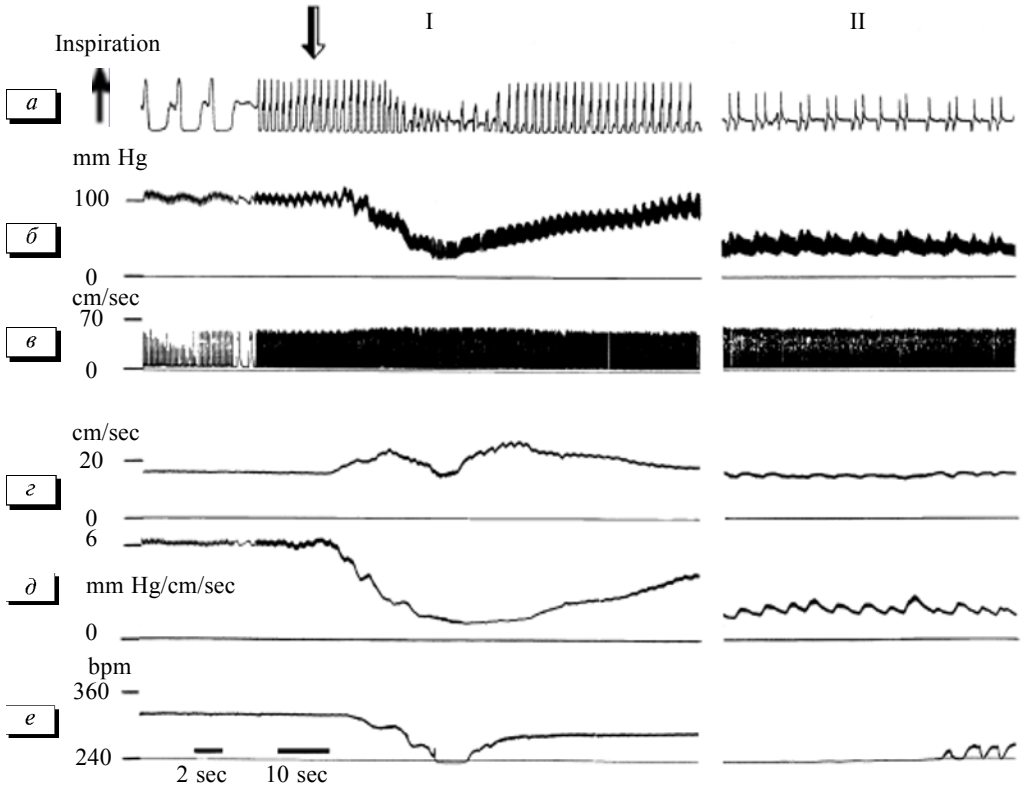

Fig. 2. Parameters of hemodynamics and respiration 40 min after lithium hydroxybutyrate administration without pathological periodic breathing development (*I*) and during subsequent euphylline administration (*II*; BP drop and periodic breathing development).

um hydroxybutyrate administration. If periodic breathing with expiratory breath-holding developed before this time (15-20 min), euphylline was administered to correct respiration rhythm disturbances.

Lithium hydroxybutyrate produced an increase in HR during the first minute, then from the 5th to 10th minute, and on the 30th minute. By the end of observation period (45 min) HR did not exceed control values. BP significantly decreased during the first minute of agent action, heart output increased and peripheral resistance decreased. These changes persisted from the 15th to 30th minutes, while by the 45th minute the parameters returned to baseline values. MVR did not change. In addition, all the period of lithium hydroxybutyrate action was associated with RR decrease and RV increase (both parameters changed by more than 100%). By minute 45, these parameters significantly differed from control values. Then, euphylline was administered and registration of the studied parameters was continued.

TABLE 1. Effects of Euphylline on Respiratory and Hemodynamic Parameters during Inhalation of Hypercapnic Gas Mixture against the Background of $GABA_b$-Receptor Activation with Lithium Hydroxybutyrate

Parameter	Control		Lithium hydroxybutyrate		Euphylline	
	baseline	4 min	baseline	4 min	baseline	4 min
HR, min^{-1}	401.6±10.4	407.2±13.3	414.7±15.3	432.0±25.3	428.0±12.7	386.2±33.7
BP, mm Hg	114.1±3.2	97.7±3.8*	80.0±8.9	41.7±2.5^{**+++}	61.5±9.4	51.5±10.2^{+++}
MVR, ml/min	21.1±1.3	46.0±5.4**	17.3±1.7	16.5±1.9^{++}	21.9±1.7	25.5±3.5^{+}
RR, min^{-1}	66.0±1.1	68.2±3.6	31.2±5.3	19.8±3.2^{+++}	40.0±6.6	35.2±6.3^{+++}
RV, ml	0.32±0.02	0.71±0.11*	0.63±0.10	0.88±0.08	0.64±0.07	0.86±0.11

Note. Here and in Table 2: *p<0.05, **p<0.01, ***p<0.001 in comparison with baseline values; ^{+}p<0.05, ^{++}p<0.01, ^{+++}p<0.001 in comparison with control.

TABLE 2. Effects of Euphylline on Respiratory and Hemodynamic Parameters during Inhalation of Hypercapnic Gas Mixture against the Background of $GABA_b$-Receptor Activation with Lithium Hydroxybutyrate

Parameter	Control			Lithium hydroxybutyrate			Euphylline		
	baseline	1 min	4.5 min	baseline	1 min	4.5 min	baseline	1 min	4.5 min
HR, min^{-1}	406.9±9.5	396.9±6.6	395.3±21.1	433.5±15.7	425.2±11.6^{+++}	398.2±14.3	444.9±12.4	441.1±14.1^{+++}	429.3±19.0
BP, mm Hg	116.8±2.6	59.1±4.5**	41.8±2.8*	97.5±10.1	98.3±3.8^{+++}	81.7±6.9^{+++}	68.1±12.7	66.9±12.4	64.3±11.6^{+}
MVR, ml/min	22.8±1.3	39.2±2.1**	30.8±2.7*	18.7±1.4	43.2±2.6***	37.8±3.5***	24.4±2.4	47.0±2.7*$^{++}$	36.7±2.3*
RR, min^{-1}	68.7±2.2	64.0±2.8	58.8±2.5*	35.3±5.1	62.2±2.8***	68.0±2.2*$^{+++}$	46.0±6.4	65.5±3.3*	60.9±3.1
RV, ml	0.34±0.03	0.63±0.04**	0.53±0.05*	0.58±0.08	0.70±0.05	0.56±0.04	0.72±0.04*	0.61±0.05	

Two qualitatively different external respiration reactions were observed in response to euphylline administration. The effect depended on baseline systemic BP and degree of its decrease after drug administration.

In animals with periodic respiration induced by lithium hydroxybutyrate administration, euphylline insignificantly decreased systemic blood pressure, increased respiratory rate, and restored regular breathing (Fig. 1). Euphylline probably blocked pre- and postsynaptic A_1-receptors responsible for RR drop during adenosine action [9] and located on respiratory neurons exposed to $GABA_b$-agonist, what resulted in compensation of the developed pathological breathing. Although, some investigators believe that these adenosine receptor do not play significant role in respiratory rhythm generation [6].

If BP drop after euphylline administration was significant, euphylline induced periodic breathing with expiratory breath-holding in animals exhibiting no respiration rhythm disturbances after $GABA_b$-receptor activation (Fig. 2). The response of the respiratory system to euphylline administration against the background of periodic breathing is probably associated with compound effects on brain blood supply [5,11,12]. Probably, the effects of adenosine receptor antagonist, like effects of adenosine itself, differ in animals with predominance of sympathetic and parasympathetic activity, and with predominance of hyper- or hypodynamic blood circulation [1].

Chemosensitivity of respiratory system was evaluated in tests with inhalation of gas mixtures at the start of the experiment and after exposure to each compound.

In our experiments we typically observed a single-phase respiratory response to hypercapnia and a biphasic response to hypoxia, therefore respiratory reaction to 5% CO_2 was assessed after attaining the plateau 4 min after onset of gas mixture inhalation and response to 10% O_2 was evaluated at the end of the first minute (maximum ventilation) and 4.5 min after attaining the plateau.

Adenosine receptor antagonists differently affected the response of the respiratory system to acute and chronic hypoxia and hypercapnia [13,14].

Under control conditions, hypercapnia reduced BP by almost 15% and increased MVR and respiration depth by almost 120% on minute 4. HR and RR values remained unchanged under these conditions (Table 1). Preliminary hydroxybutyrate administration resulted in additional BP reduction by 50%, RR reduction by 35%, and RV increase only by 39%. The reaction of the respiratory system was weakened. BP remained low after euphylline administration, but there was no significant BP drop in response to hypercapnia (16%).

RR slightly decreased (12%) and MVR and RV slightly increased (16 and 34%).

Thus, the obtained results showed that euphylline did not abolish the decrease in respiratory system sensitivity to hypercapnia caused by $GABA_b$-receptor agonist.

Under control conditions, hypoxia significantly decreased BP by almost 50% during the first phase of the response (Table 2) and then by 74% during the second phase. RR decreased only on minute 5 (by 14%). Volumetric parameters were above the baseline values during the entire period of hypoxic gas mixture inhalation (by 80%). Against the background of hydroxybutyrate action, RR did not decrease, but even increased by 92% on minute 5. MVR was above the baseline level by 131%, *i.e.* the response to hypoxia became more pronounced.

Hypoxia did not affect HR and BP decrease after euphylline administration on minutes 1 and 4.5. Hence, cardiovascular system did not react to hypoxia after euphylline administration against the background of hydroxybutyrate action. MVR was changed by 92%, which slightly exceeded the control reaction to hypoxia, but was significantly lower than the reaction of the respiratory system against the background of hydroxybutyrate action. RV increased by 28% during the first phase of the response to hypoxia, while RR increased by 42%. The results suggest that the increase in lung ventilation in response to hypoxia under control conditions was provided mainly by increase in RV, and after hydroxybutyrate administration by the increase in both RR and MVR. Euphylline slightly diminished the response of the respiratory system to hypoxia against the background of hydroxybutyrate.

Thus, euphylline did not restore the sensitivity of the respiratory system to hypercapnia after $GABA_b$-receptor activation, but partially reduced the intensity of the response to hypoxia. Euphylline administration against the background of hydroxybutyrate-induced pathological periodic breathing restored regular breathing in animals with minor BP reduction.

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