Loss of Carbon Dioxide Sensitivity by the Respiratory System in Cats with Activated Brain GABAergic Structures

I. A. Tarakanov

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 123, No. 4, pp. 385-390, April, 1997
Original article submitted March 6, 1996

Activation of GABAergic system by the agonist sodium oxybutyrate (200 mg/kg body weight) or phenibut (100 mg/kg) results in drastic reduction or complete loss of sensitivity to carbon dioxide by the central regulator of respiration. It is concluded that respiration in this case is regulated by oxygen but not by carbon dioxide.

Key Words: *GABAergic system; hypocapnia; hypercapnia; periodic respiration; chemoreception; cats; acetazolamide*

Carbon dioxide is the major stimulus for the central apparatus regulating respiration. Under normal conditions, respiration is controlled in accordance with arterial blood P_{CO_2} which is determined by chemoreceptors located in the carotid bodies and on the ventral surface of the medulla oblongata. Afferent pulses traveling from central and peripheral chemoreceptors to the respiratory center are influenced by the GABAergic system acting on these chemoreceptors [9,13] and their afferent projections in the medulla oblongata [10]. Stimulation of respiration in rats during hypercapnia is weakened by high doses of sodium oxybutyrate, an agonist of GABAergic receptors [11,12]. We have found that systemic administration of the GABAergic receptor agonists sodium oxybutyrate and phenibut increases the respiratory system sensitivity to oxygen and decreases it to carbon dioxide [8].

In the present study we evaluated the responsiveness of the respiratory system to hyper- and hypocapnia in cats with activated GABAergic receptors.

Laboratory of Respiratory Pathophysiology, Institute of General Pathology and Pathophysiology, Russian Academy of Medical Sciences, Moscow

MATERIALS AND METHODS

The study was performed on 28 random-bred cats of both sexes (body weight 2.2-4.1 kg) under pentobarbital anesthesia (40 mg/kg intraperitoneally). The GABAergic agonists sodium oxybutyrate (200 mg/kg) and phenibut (100 mg/kg), were injected intravenously. Similar results obtained in experiments with sodium oxybutyrate and phenibut enabled us to consider these compounds as belonging to the same group. Hypercapnic gas mixture (5% CO_2 in air) was prepared using the standard set of rotameters supplied with the anesthetic apparatus. This procedure for preparing gas mixture ensured an accuracy of about 1% (i.e., 5±1% CO_2). A small amount of O_2 was then added to the gas mixture so as to keep the O_2 concentration constant at a level of 21±1%. Gas concentrations in the mixture were monitored with an ABL-330 instrument (Radiometer Int.). The mixture was delivered to cats only after saturating it with water vapor at 38°C. Hypocapnia was attained by hyperventilating with an apparatus for artificial respiration, which permitted step-by-step regulation of the respiratory rate and continuous adjustment of the respiratory depth. Hyperventilation regime for each cat was selected so that respiratory rate was close to

0007-4888/97/0004-0331$18.00

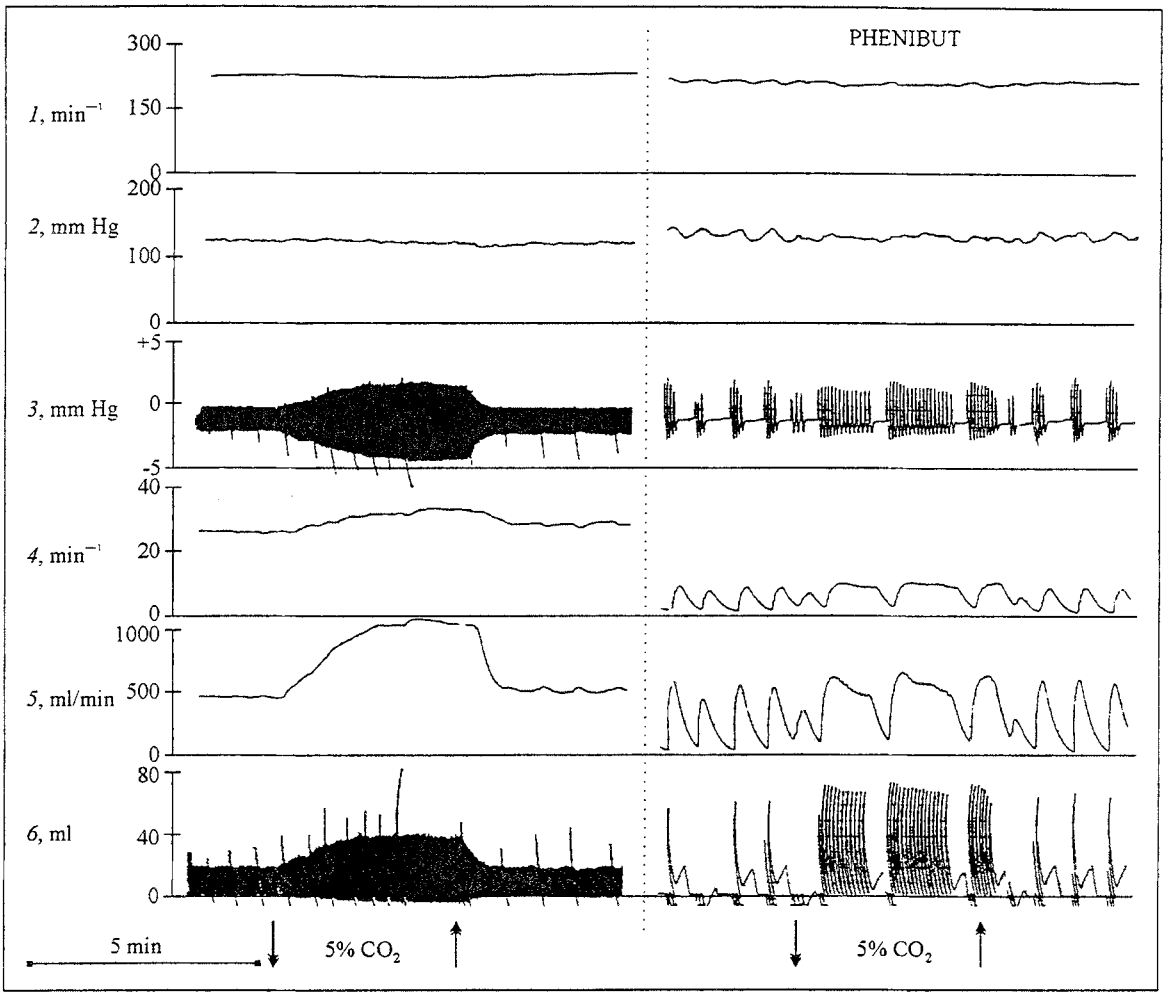

Fig. 1. Variations of respiration and systemic hemodynamics in anesthetized cat inhaling hypercapnic gas mixture during periodic apneustic respiration after phenibut administration.

Here and in Figs. 2, 3, and 4: *1*) heart rate; *2*) mean arterial pressure in the systemic circulation; *3*) intraesophageal pressure; *4*) respiratory rate; *5*) minute volume; *6*) respiratory volume.

Arrows indicate the moment when the cat started and finished breathing the gas mixture.

the control (baseline) level, while respiratory depth was at least twice above its control level; this regime remained unchanged throughout the experiment. Each cat inhaled the gas mixture and was hyperventilated for 5 min, with intervals of no less than 10 min between inhalation and hyperventilation sessions. The protocol included tests of the respiratory system for hyper- and hypocapnia before (control conditions) and after activation of the GABAergic system by its injected agonist following various changes in the respiratory rhythm (test conditions). In order to produce stable tissue hypercapnia of endogenous origin, cats were injected intravenously with acetazolamide (20 mg/kg; Serva), a blocker of carbonic anhydrase, 60 min before injection of sodium oxybutyrate or phenibut. All procedures were described in detail elsewhere [1,6,8].

RESULTS

Two-phase changes in respiratory rhythm after systemic administration of GABA-positive substance were observed previously [7]. Phase 1 was characterized by periodic apneustic breathing with marked breath holdings upon inspiration. After 90-120 min, this breathing pattern was gradually succeeded, in about one-third of the cats (depending on their individual sensitivity to the agonist used), by respiratory movements of constant frequency and amplitude (phase 2).

During both phases, the respiratory system sensitivity to hypercapnia and hyperventilatory hypocapnia caused by artificial ventilation was determined.

Under the control conditions (i.e., before the GABAergic system was activated), the respiratory system responded to the hypercapnic gas mixture by

an increase in pulmonary ventilation 2- to 2.5-fold; in most animals the depth and rate of breathing also increased. Change from air to hypercapnic gas mixture during periodic apneustic breathing (phase 1) led to augmented pulmonary ventilation, often as a result of shortened duration or temporary disappearance of breath holdings (Fig. 1). As shown in Fig. 1, the respiratory rhythm prior to inhalation of gas mixture is characterized by a well-defined periodicity with 30-sec or longer periods of breath holding. The intraesophageal pressure curve shows that these breath holdings correspond to the inspiratory phase, negative pressure being maintained. This respiratory pattern is also reflected by the minute volume of respiration

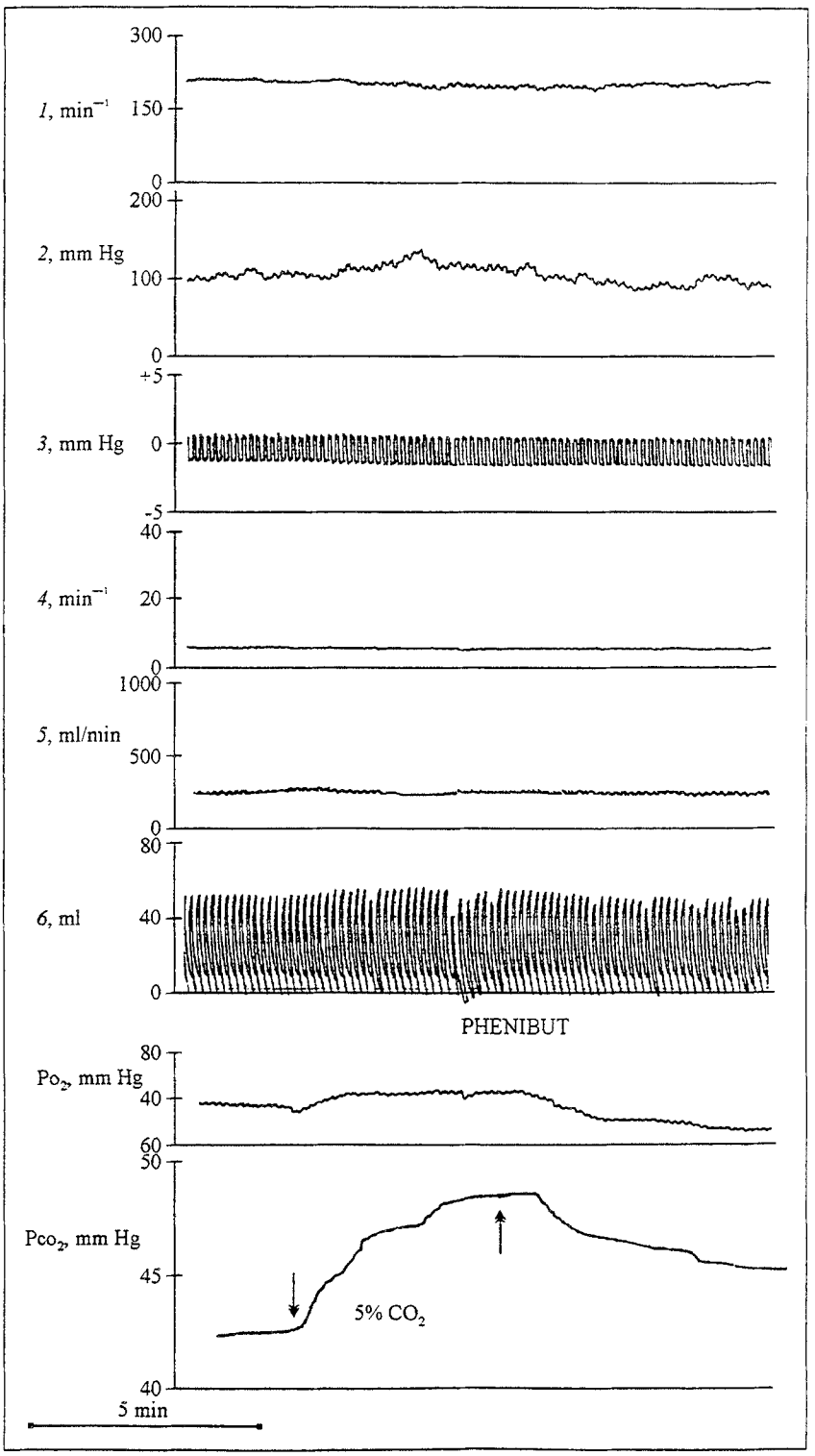

Fig. 2. Variations of respiration, gaseous composition of arterial blood, and systemic hemodynamics in anesthetized cat inhaling hypercapnic gas mixture during "machinelike" respiration after phenibut administration. Figures 2 and 4 represent a montage of three recordings made at equal speeds of tape winding, with parameters of respiration and circulation being recorded in oblique coordinates and those of blood gases in rectangular coordinates.

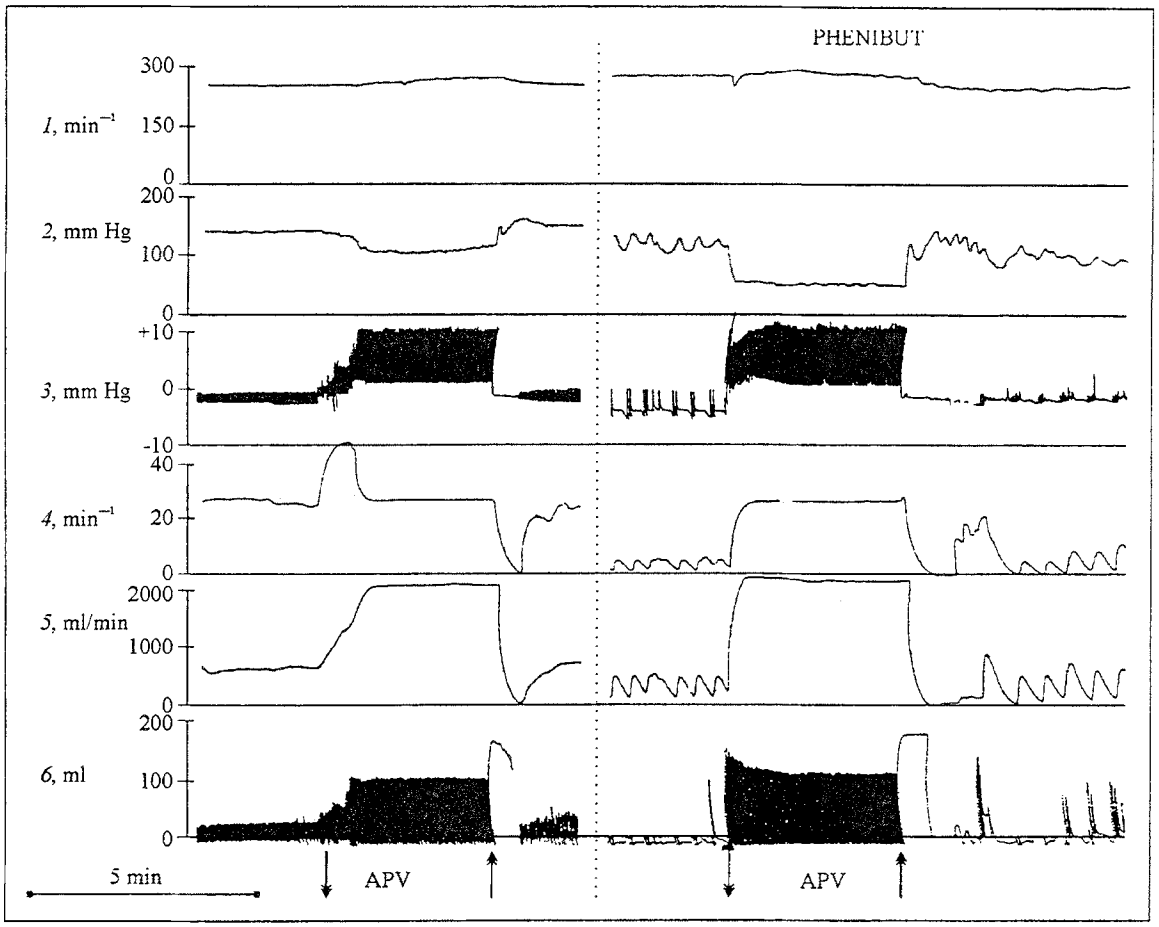

Fig. 3. Variations of respiration and systemic hemodynamics in anesthetized cat subjected to artificial pulmonary ventilation (APV) in a large volume during periodic apneustic respiration after phenibut administration.

and the frequency of respiratory movements. Under such conditions compensatory reactions of the circulatory system were obvious: both heart rate and systemic arterial pressure (SAP) were elevated during the period of apneusis. After transition to hypercapnic gas mixture, the respiratory rate and minute volume curves became smoother, but the depth of inspiration remained essentially unchanged. Approximately 3-5 min after cessation of breathing this mixture, the initial respiratory pattern was restored.

The transition to hypercapnia during "machine-like" breathing (phase 2) usually resulted in nonresponsiveness of the respiratory system, and in some cases led to inhibition of pulmonary ventilation (Fig. 2). Respiratory system virtually did not respond to hypercapnia despite a 10 mm Hg increase in arterial blood Pco_2. Under these conditions heart rate slightly decreased without noticeable changes in SAP. Slight elevation of Po_2 in arterial blood (Fig. 2) was due to the presence of O_2 in gas mixture.

Thus, the GABA agonists sodium oxybutyrate and phenibut caused a progressive decrease in the sensitivity of the respiratory system to CO_2, making this system less responsive to it during phase 1 and completely nonresponsive during phase 2.

The response of pulmonary ventilation to hyperventilatory hypocapnia was then investigated in order to confirm that the impact of CO_2 on respiration is markedly weakened after activation of the GABAergic system. Under the action of phenibut spontaneous respiration ceased much sooner after the start of hyperventilation and was restored slower after its discontinuation than under the control conditions (Fig. 3). The cardiovascular system generally responded to hyperventilation by an increase in heart rate and by pronounced decrease in SAP. A comparison of these results with our previous data on the effect of O_2 on the respiratory system under the same conditions [8] confirms that the GABAergic receptor agonists sodium oxybutyrate and phenibut decrease the contribution of Pco_2-sensitive chemoreceptors to the formation of respiratory rhythm. Such an activation of the GABAergic system during phase 1 (i.e., the phase of periodic apneustic respiration) is characterized by gradual reduction in the CO_2 sensitivity of the central regulator of re-

spiration and by the development of hypoventilatory hypercapnia.

In order to determine the contribution of hypercapnic factor to the respiratory rhythm after injection of a GABA-positive substance, we used a model similar to that employed in our previous studies [2,5], namely, stable endogenous hypercapnia developing after inactivation of enzyme carbonic anhydrase by acetazolamide. Carbonic anhydrase is known to accelerate the CO_2—bicarbonate conversion, thus promoting the removal of CO_2 from cells into the blood and from the blood to alveolar air. Inhibition of this enzyme hinders CO_2 clearance from the body and leads to a long-lasting tissue

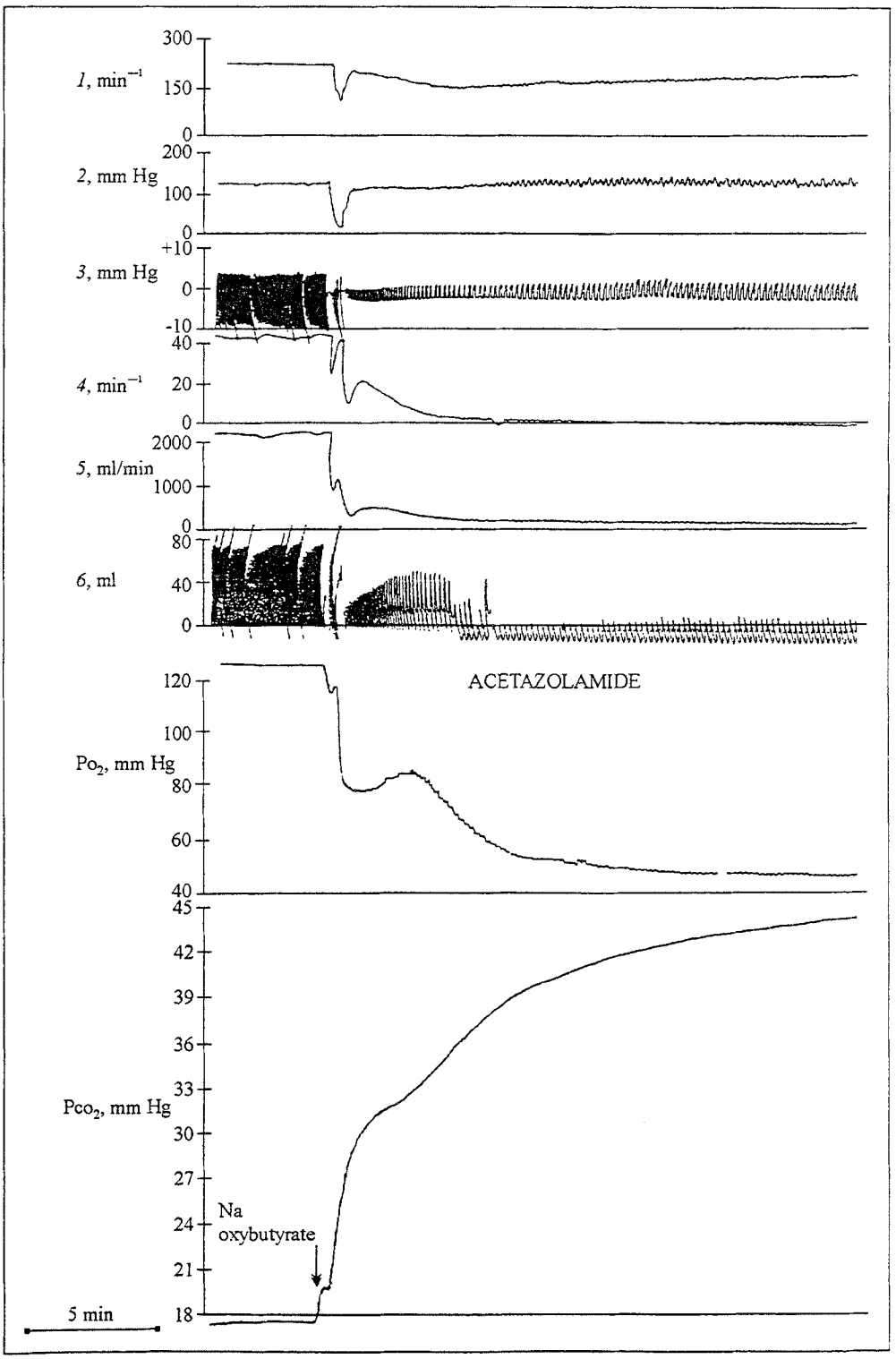

Fig. 4. Variations of respiration, gaseous composition of arterial blood, and systemic hemodynamics in anesthetized cat injected with sodium oxybutyrate when carbonic anhydrase activity was blocked by acetazolamide.

hypercapnia. Hypercapnia is usually accompanied by greatly increased (2- to 2.5-fold) pulmonary ventilation, primarily as a result of increased breathing depth [2,5].

Injection of sodium oxybutyrate or phenibut into cats with tissue hypercapnia markedly inhibited respiration (Fig. 4). The frequency and depth of respiratory movements dropped to values usually observed in phase 2 of alterations in the respiratory rhythm after activation of the GABAergic system. Under these conditions, Po_2 fell and Pco_2 rose more than 2-fold. The most important finding is the absence of periodic apneustic breathing during tissue hypercapnia after the GABAergic system had been activated. This may be due to the continued ability of preformed hypercapnia to stimulate respiration initially and/or to the requirement for fairly high CO_2 levels in the body so that transition from phase 1 to phase 2 is possible.

From these results it follows that activation of GABAergic receptors by their agonist even in a moderate dose (200 mg/kg body weight sodium oxybutyrate compared with 70-120 mg/kg used in clinical practice) markedly reduces or even abolishes the sensitivity of the respiratory system to CO_2. The results of this study together with our previous findings [8] suggest that the chemoreceptor circuit involved in the regulation of respiration [4] is so reorganized under such conditions that the respiration-controlling system as a whole starts to control respiration in terms of O_2 rather than CO_2 [3]. Transition from phase 1 to phase 2 observed after activation of GABAergic system indicates that the respiratory system has lost its sensitivity to CO_2, hypoventilatory hypercapnia has developed, and the respiratory center no longer responds to afferent stimuli from lung mechanoreceptors [1]. However, since bilateral vagotomy reproduced during periodic apneustic breathing (phase 1) does not contribute to the rapid transition to "machinelike" breathing (phase 2), factors other than the loss of sensitivity to pulmonary chemoreceptors are probably implicated in the transition from phase 1 to phase 2.

REFERENCES

1. G. N. Kryzhanovskii, I. A. Tarakanov, and V. A. Safonov, *Fiziol. Zh.*, No. 11, 13-23 (1993).
2. A. M. Kulik, L. N. Kondrat'eva, and I. A. Tarakanov, *Byull. Eksp. Biol. Med.*, **81**, No. 2, 152-156 (1976).
3. L. L. Shik and N. N. Kanaeva (Eds.), *A Handbook on Clinical Physiology of Respiration* [in Russian], Leningrad (1980), pp. 209-232.
4. V. A. Safonov, V. N. Efimov, and A. A. Chumachenko, in: *Neurophysiology of Respiration* [in Russian], Moscow (1980), p. 222.
5. I. A. Tarakanov, "Protective effects of gaseous and tissue hypercapnia on the body in hypoxic hypoxia," Author's Synopsis of Dissertation [in Russian], Moscow (1984).
6. I. A. Tarakanov, E. A. Golovatyuk, E. R. Turskaya, and V. A. Safonov, *Byull. Eksp. Biol. Med.*, **115**, No. 6, 583-587 (1993).
7. I. A. Tarakanov and V. A. Safonov, *Byull. Eksp. Biol. Med.*, **119**, No. 6, 606-609 (1995).
8. I. A. Tarakanov, *Byull. Eksp. Biol. Med.*, **123**, No. 3, 264-268 (1997).
9. A. M. T. Da Silva, B. Hartley, P. Hamosh, *et al.*, *J. Appl. Physiol.*, **62**, 2264-2272 (1987).
10. W. D. Dietrich, O. H. Lowry, and A. D. Loewy, *Brain Res.*, **237**, 254-260 (1982).
11. J. Hedner, J. Jonason, and D. Lundberg, *J. Neural Transm.*, **49**, 179-186 (1980).
12. R. A. Mueller, D. B. A. Lundberg, G. R. Breese, *et al.*, *Pharmacol. Rev.*, **34**, No. 3, 255-285 (1982).
13. K. A. Yamada, J. M. Moerschbaecher, P. Hamosh, and R. A. Gillis, *J. Pharmacol. Exp. Ther.*, **226**, No. 2, 349-355 (1983).