Role of the Central N-Cholinergic Receptors in Preventing the Inhibition of Respiration Following Activation of the GABA-Ergic System in the Brain

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Experiments with Nembutal-anesthetized cats, in which sodium oxybutyrate, tubocurarine, and ethyl alcohol are injected into the fourth ventricle of the brain, show that the respiratory disorders caused by sodium oxybutyrate or ethyl alcohol can be eliminated by tubocurarine. By blocking the central N-cholinergic receptors, tubocurarine abolishes the inhibition of respiratory movements induced through activation of the GABA-ergic system.

Key Words: *cholinergic system; regulation of respiration; sodium oxybutyrate; tubocurarine; ethanol; GABA*

The relatively high concentration of gamma-butyric acid (GABA) found at the respiratory center [2,12] and its pronounced inhibitory effects following systemic or central administration to warmblooded animals indicate that GABA participates in the regulation of respiration [3-5,10]. The inhibition of respiratory activity by ethanol is also believed to involve the GABA-ergic system [9]. The N-cholinergic system, too, takes part in the regulation of respiration, as is evidenced by the presence of N-cholinergic receptors in central structures of the respiratory apparatus, by the relationship of these receptors to GABA-ergic neurons, and by their strong inhibitory influence on respiration when these neurons are activated [4,10,11]. However, the involvement of the GABA-ergic system in the regulation of respiration in comparison with that of the cholinergic system remains unexplored.

This study was undertaken to examine the role of the central N-cholinergic receptors in preventing the inhibition of respiration that occurs after the activation of the central GABA-ergic receptors.

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MATERIALS AND METHODS

The study was conducted on 53 tracheotomized cats (body weight 2.5-3.5 kg) under Nembutal anesthesia (40 mg/kg intraperitoneally). Respiratory rate, minute volume (MV), arterial pressure, and heart rate were recorded and averaged using a surgical monitor. Parameters of systemic hemodynamics were registered by means of a catheter inserted into the femoral artery. Respiratory volume was calculated as the ratio of MV to respiratory rate. Electromyograms of the phrenic muscle were taken with an M-42 electromyograph (Hungary) using bipolar metal electrodes. The GABA-ergic system was activated with sodium oxybutyrate. Oxybutyrate at 60 or 180 μmol/kg, tubocurarine at 130 or 300 nmol/kg, and 96% ethanol at 330 nmol/kg were injected into the fourth ventricle of the brain in a volume of 50 μl (the oxybutyrate and tubocurarine were dissolved in isotonic NaCl solution and 48% ethanol, respectively). Control cats received isotonic NaCl solution and 48% alcohol in the same volume.

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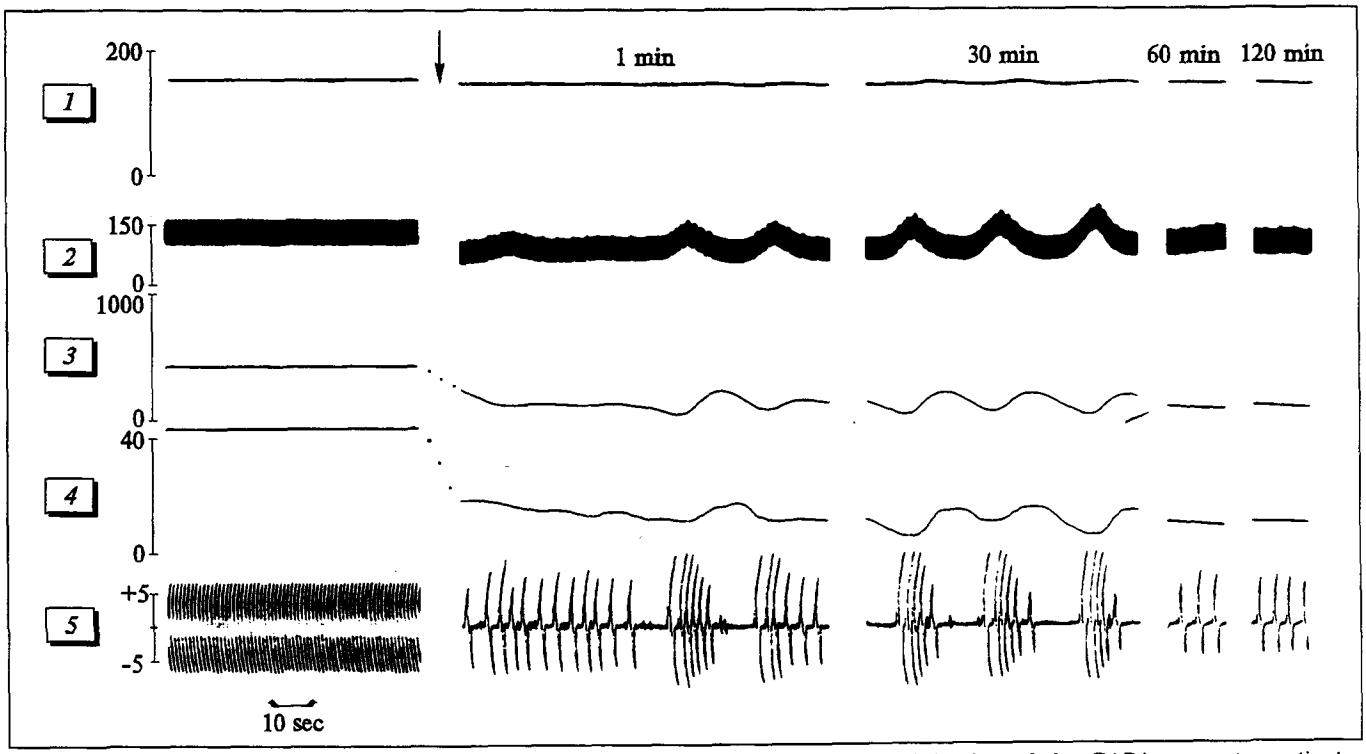

Fig. 1. Variations in respiration and hemodynamics in a cat after intraventricular injection of the GABA–receptor activator sodium oxybutyrate (60 μmol/kg). The arrow marks the time of sodium oxybutyrate injection. Here and in Fig. 2: *1*) heart rate, beats/min; *2*) arterial pressure, mm Hg; *3*) minute volume, ml/min; *4*) respiratory rate, min^{-1}; *5*) pneumotachogram, liters/min. At top: times after sodium oxybutyrate injection.

RESULTS

In complete agreement with our previous findings [3], sodium oxybutyrate in a dose of 60 μmol/kg (7 cats) caused periodic respiration, which lasted for 30-40 min and was followed by spontaneous restoration of a rhythmic pattern of respiratory movements, although the respiratory rate remained greatly reduced; MV was decreased while the respiratory volume was slightly increased. Such respiratory parameters were recorded for 2 to 3 h (Fig. 1).

In all the 9 cats administered tubocurarine at 130 nmol/kg 5 min after sodium oxybutyrate in the indicated dose, resumption of rhythmic respiration was observed 5-10 min later, followed (after approximately 60 min) by a return of both the respiratory rate and MV to their baseline values (Fig. 2).

In another group of 9 cats, given sodium oxybutyrate at 180 μmol/kg, respiratory movements ceased by the end of the first minute after its injection and the cats were transferred to artificial lung ventilation (ALV) but failed to regain spontaneous respiration for 90 min despite continued ALV. In similar tests, also on 9 cats, protracted inspirations followed by cessation of respiratory movements were observed for 30 min during periods when ALV was temporarily discontinued. The cats given tubocurarine at 300 nmol/kg at minute 30 after oxybutyrate began to breathe spontaneously as early as 1 or 2 min later; their respiration was periodic at first but then (after 4-5 min) became rhythmic, and the respiratory rate, MV, and respiratory volume all exceeded their baseline values (Fig. 3).

In further tests (16 cats), the activity of the respiratory system was found to be impaired also by ethanol. In all these animals, respiratory activity ceased less than 1 min after ethanol administration and they had to be ventilated artificially. During 30 min, at times when ALV was discontinued, a few protracted inspirations followed by respiratory arrest were observed. In other tests of this series, in which tubocurarine was administered to 9 cats at 130 nmol/kg 10 min after ethanol, normal respiratory movements were recorded at minutes 3-5, and both the respiratory rate and respiratory volume exceeded their baseline values (Fig. 4).

In the seven control cats that received tubocurarine at 130 nmol/kg, the respiratory rate dropped during the first 4 min, after which the respiratory parameters reverted to their baseline values by minute 5 or so; 20-25 min later, the animals fully awoke, the awakening being preceded by slight increases in respiratory rate and volume.

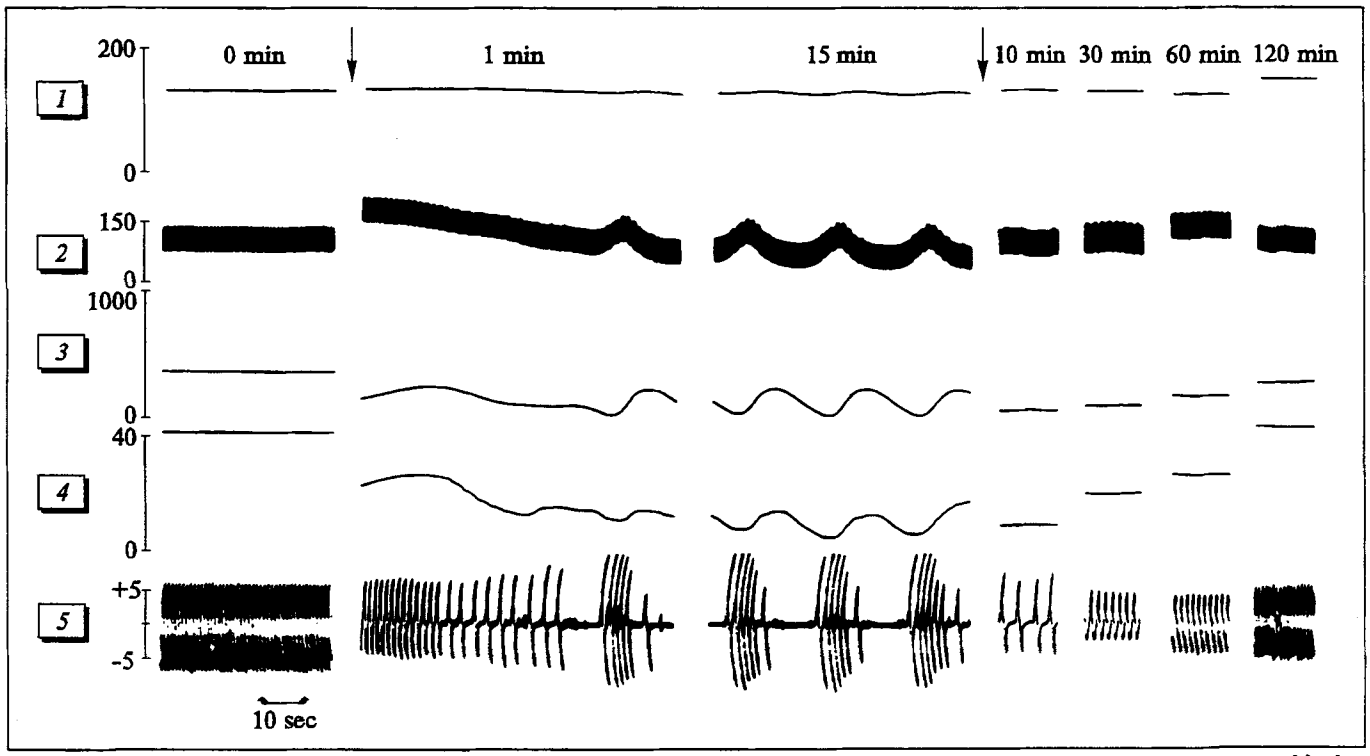

Fig. 2. Variations in respiration and hemodynamics in a cat after intraventricular injection of the H–cholinergic receptor blocker tubocurarine (130 nmol/kg), following activation of central GABA receptors by sodium oxybutyrate (60 μmol/kg). Here and in Fig. 3: the left arrow marks the time of sodium oxybutyrate injection and the right arrow, the time of tubocurarine injection.

It should be pointed out that although N-cholinergic receptors are present in brainstem structures participating directly in the regulation of respiration, some authorities consider the N-cholinergic system of the medulla oblongata to be involved in this regulation [4,8], whereas others believe that the

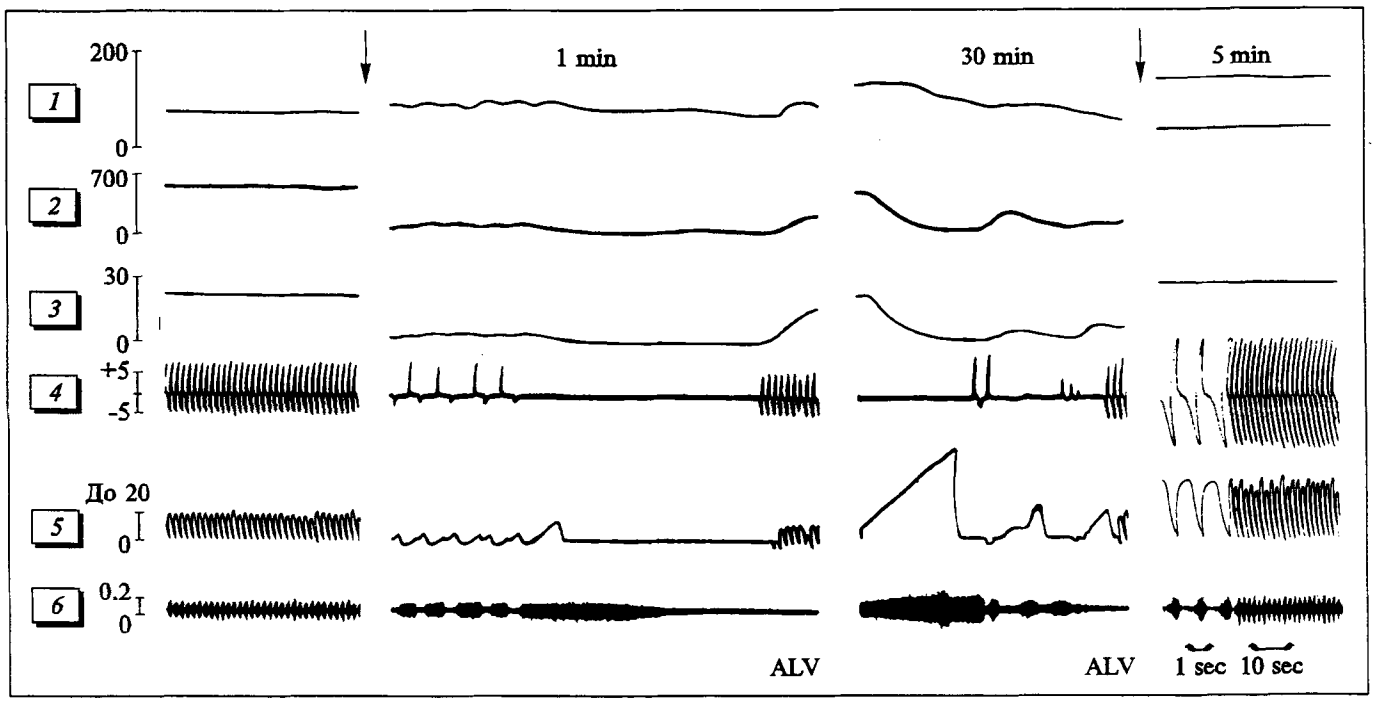

Fig. 3. Respiratory arrest in a cat following intraventricular injection of the GABA–receptor activator sodium oxybutyrate (180 μmol/kg) and resumption of respiration after blockade of central N–cholinergic receptors with tubocurarine (300 nmol/kg). Here and in Fig. 4: *1*) arterial pressure, mm Hg; *2*) minute volume, ml/min; *3*) respiratory rate, min^{-1}; *4*) pneumotachogram, liters/min; *5*) respiratory volume, ml; *6*) electromyogram of phrenic muscle, mV. At top are the times postinjection. *ALV*: artificial lung ventilation.

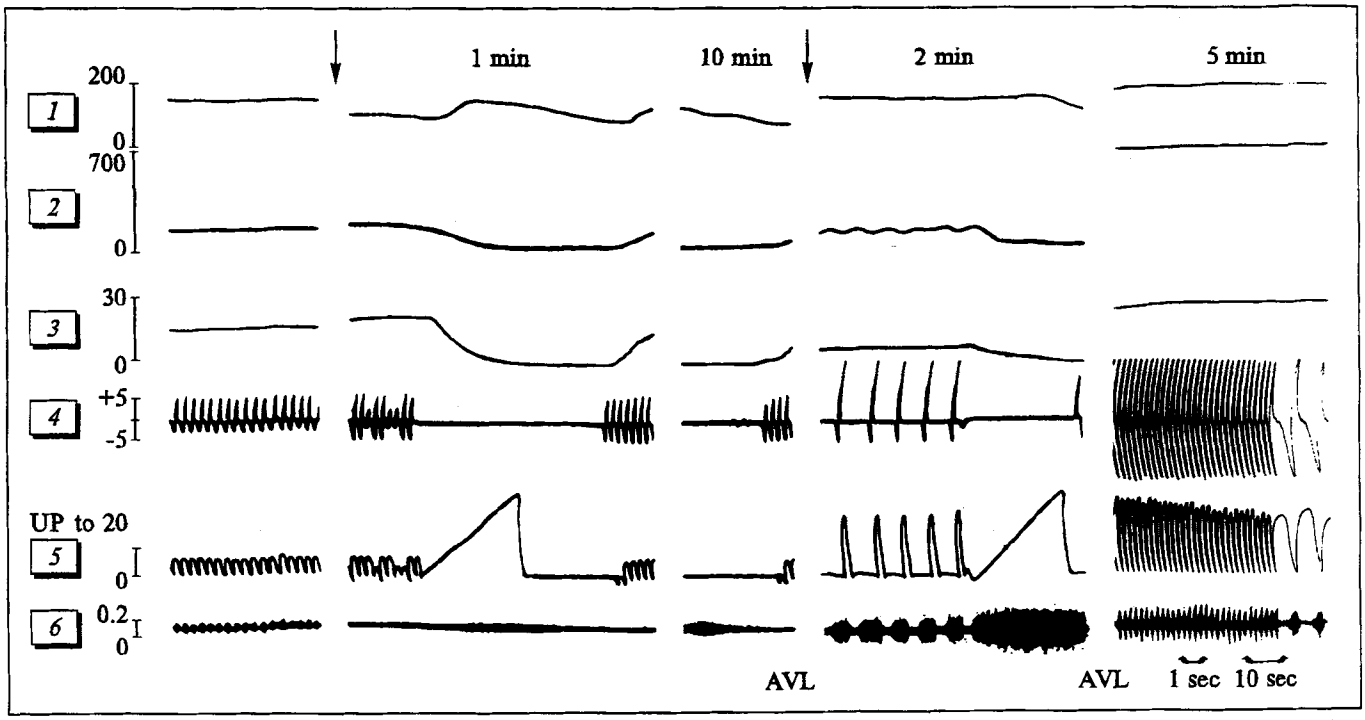

Fig. 4. Respiratory arrest in a cat following intraventricular ethanol injection (330 nmol/kg) and resumption of respiration after blockade of central N-cholinergic receptors with tubocurarine (130 nmol/kg). The left arrow marks the time of ethanol injection and the right arrow, the time of tubocurarine injection. At top are the times postinjection.

N-cholinergic receptors of the bulbar structures of the respiratory center do not significantly affect the respiration of an anesthetized cat.

Our analysis of the role the N-cholinergic system plays in the respiratory disorders caused in cats by sodium oxybutyrate or ethanol shows that blockade of the central N-cholinergic receptors by tubocurarine abolishes the inhibition of respiratory activity in such animals, a fact which supports the view that the cholinergic system does participate in the regulation of respiration. Previously, we demonstrated that the major receptors responsible for the occurrence of pathological periodic respiration of the apneic type are the $GABA_B$ receptors present in neurons of the dorsal respiratory group in the nucleus of the solitary tract [1]. The prevention of GABA effects via blockade of the central N-cholinergic receptors by tubocurarine thus attests to an undoubtedly antagonistic interaction of these with the $GABA_B$ receptors. This conclusion is supported by the ability of tubocurarine to prevent ethanol-induced respiratory arrest and eventually to increase the minute volume due to increases in both respiratory rate and respiratory volume.

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