Cold Paralysis of the Thermoregulatory Center and Its Recovery at Paralyzing Temperature

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Cooling of albino rats in water to brain temperature of 19.6±0.2°C causes almost complete suppression of thermoregulatory muscle tone and shivering because of cold paralysis of the thermoregulatory center. Intravenous injection of 15-20 μM EDTA restores these thermoregulatory reactions at the same temperature.

Key Words: *hypothermia; thermoregulation; heat production*

Cold paralysis of the thermoregulatory center during deep hypothermia means the absence of the most potent defense reaction in the form of thermoregulatory tone and shivering. Cold paralysis occurs in men at rectal temperature of 27-28°C [7,10] and in rats at body temperature of 17-18°C [2].

It has been recognized that the rise of cytosolic Ca^{2+} during hypothermia is the primary cause of functional cell disturbances in homoiothermic vertebrates [8]. Normal Ca^{2+} concentration in cytosol and intercellular space is 10^{-7} M and 10^{-3} M, respectively. So, translocation of an excess of Ca^{2+} from the cytosol into the extracellular space against the huge concentration gradient requires much energy. In particular, translocation of one calcium ion requires energy of one ATP molecule [5]. At a low ambient temperature, activity of ATP-synthesizing enzymes sharply decreases, which suppresses Ca^{2+} transport and accumulation of calcium ions in the cytosol. This impairs cell metabolism and paralyzes cell functions [8,9]. Reduction of blood Ca^{2+} concentration with EDTA results in a decrease of calcium concentration in the medium and, consequently, less energy is required for translocation of calcium ions from the cell against concentration gradient. This promotes normalization of cytosolic Ca^{2+} concentration and functional recovery of the cell [8]. Using this approach we have recently restored the function of skin thermoreceptors at a skin temperature of about 0°C, i.e., at the temperature of their cold paralysis [1].

In the present study we attempted to restore function of the thermoregulatory center at a temperature of cold paralysis.

MATERIALS AND METHODS

Experiments were carried out on male Wistar rats (240-270 g) under Nembutal anesthesia (35 mg/kg, intraperitoneally). The animals were fixed in special stands. Fine (0.2 mm in diameter) copper-constantan thermocouple was inserted into anterior hypothalamus to a depth of 7 mm. Other thermocouples were introduced into the rectum to the depth of 3-4 cm and under the derm and epidermis on the back parallel to skin surface. The thermometer accuracy was 0.05°C.

For quantitative evaluation of muscular electrical activity (thermoregulatory muscular tone and shivering) needle electrodes were introduced into dorsal muscles. Amplified potentials were input into an integrator, which determined electrical activity (S, μV, sec) [3,4]. All parameters were recorded with a strip-chart potential recorder throughout the experiment, in some experiments respiratory rate was determined.

After measuring the baseline parameters at room temperature, the animals were immersed into cold

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water (9-10°C) at an angle of about 45° so that water reached the ears without disturbing respiration. Muscular electrical activity abruptly decreased or completely disappeared at cerebral and rectal temperatures of 18 and 20.5°C, respectively. This was attained during 35-40 min, after which the rats were drawn out of water and covered with wet cloth with ice to maintain cerebral and rectal temperature. After stabilization of temperature 0.4-0.6% EDTA was infused for 4 min into the femoral vein through a polyethylene catheter. Each animal received 5-7 mg (15-20 μM) EDTA. Ten-fifteen minutes after the effect of first infusion had decayed, a second dose of EDTA was infused. Seven 1.5-2-h experiments were performed with 7 rats, all of them survived.

RESULTS

Muscular electrical activity rapidly increased during cooling. Shiver appeared as periodic bursts of electrical activity with an amplitude of 300-500 μV; interburst activity also increased to 20-50 μV (thermoregulatory muscular tone) [3,4]. Activity peaked at cerebral and rectal temperatures of 28-29°C and 25-26°C, respectively, and then sharply decreased and attained a minimum, so-called cold paralysis of the thermoregulatory center, at cerebral and rectal temperatures of 20-21°C and 17-18°C, respectively. Infusion of EDTA at the same temperature increased muscular electrical activity and restored shivering. Maximum effect was observed for 8-10 min, then EDTA was infused, and infused muscular electrical activity decreased again. The second infusion induced a more pronounced effect, and electrical activity approximated the level observed at the start of cooling. Figure 1 presents the data of a typical experiment.

Data of all experiments are summarized in Table 1. EDTA induced strong reaction at stable low temperatures in the brain, rectum, and deep and superficial skin layers. Respiratory rate at these temperatures was 20-25 inspirations per minute.

EDTA does not penetrate the blood-brain barrier and its effect on the brain is realized through the blood. Blood Ca^{2+} concentration was 1.09±0.03 μmol/ml. This concentration is in equilibrium with intracellular calcium [6]. In a rat weighing 250 g peripheral blood contains approximately 19 mmol Ca^{2+}. In a single infusion the rat received 15-10 μmol EDTA interacting with Ca^{2+} in a mol:mol ratio. Thus, one dose of EDTA is sufficient to chelate all calcium ions in the blood. This, due to exchange with the extracellular medium, reduces the concentration of ionized calcium and restores functional activity of skin thermoreceptors. Blood Ca^{2+} concentration increases 10-20 min postinjection (due to dissociation of calcium-containing compounds) and diminishes the effect of EDTA. The conception proposed by Hochachka [8] and previous experimental data [1] allowed us to assume a positive effect of EDTA in cold paralysis of the thermoregulatory center; doses of EDTA (per kg body weight) used in our experiments did not exceed therapeutical doses for humans.

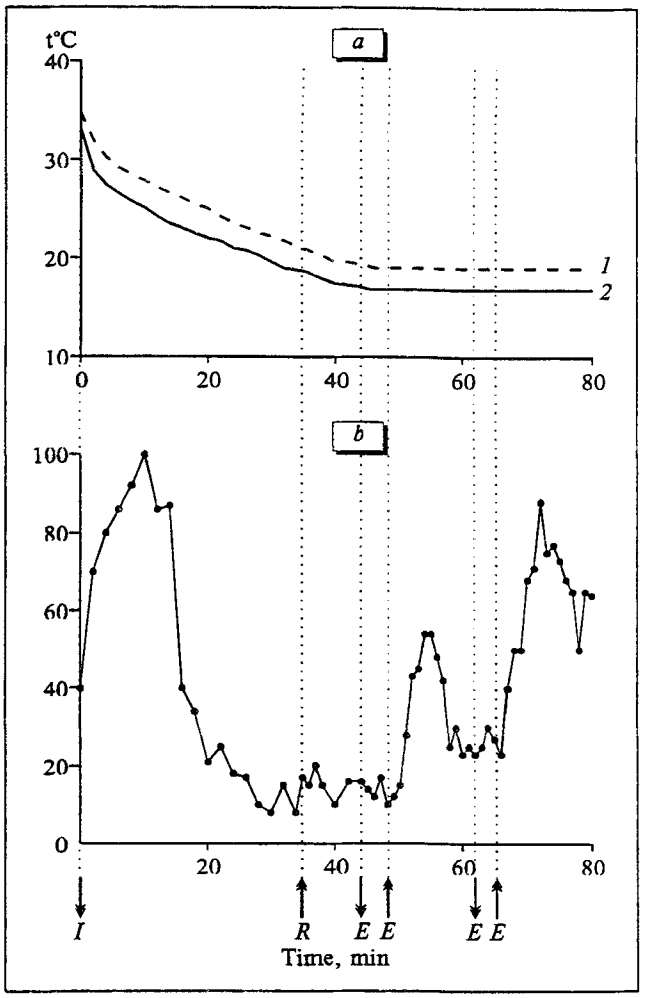

Fig. 1. Cold-induced activation of chemical thermoregulatory reaction (thermoregulatory muscular tone and shivering) followed by its suppression during cooling in water. Restoration of this reaction at the same temperature by intravenous injection of EDTA (data of typical experiment). *a*) temperature of the brain (hypothalamic area near the thermoregulatory center, *1*) and rectum (*2*). *b*) muscular electrical activity reflecting the intensity of thermoregulatory muscular tone and shivering. *I*: immersion; *R*: removal from cold water: *E*: start (upward arrow) and end (downward arrow) of intravenous infusion of EDTA. Ordinate: muscular electrical activity, % of maximum activity after the start of cooling.

It is often impossible to restore thermoregulation and vital functions in victims of accidental deep hypothermia (core body temperature below 26-27°C) [7,8]. The principal conclusion of the present study is that in cold-induced paralysis of the thermoregula-

TABLE 1. Cold-Induced Stimulation and Inhibition of Chemical Thermoregulation and Its Restoration with EDTA (*M±m*, *n*=7)

Parameter	Temperature, °C				Muscular electrical activity, %
	rectal	cerebral	skin		
			deep	superficial	
Before cooling	35.7±0.7	35.3±0.8	34.1±0.7	31.5±0.6	50±7
Intensity of reaction					
Maximum	25.8±0.5	28.5±0.4	16.6±1.3	13.9±0.9	100
Minimum	18.2±0.3	20.7±0.3	13.6±0.7	11.9±0.7	16±4
Before the first EDTA injection	17.2±0.3	19.6±0.2	15.4±0.6	15.1±0.5	12±4
8-10 min postinjection	17.1±0.2	19.2±0.3	15.9±0.5	15.4±0.5	60±8
Before the second EDTA injection	17.1±0.3	19.2±0.3	16.0±0.6	15.7±0.6	22±1
8-10 min postinjection	17.2±0.3	19.3±0.2	16.2±0.6	15.9±0.5	79±7

tory center it does not loose functional activity and can be stimulated with EDTA.

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