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EFFECT OF CLONIDINE AND NALOXONE ON HEMODYNAMIC REACTIONS IN DECEREBRATE CATS

O. S. Medvedev, A. G. Vylegzhanin,*
and D. D. Matsievskii

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Activation of central α-adrenoreceptors by clonidine is accompanied by changes in functions of the cardiovascular system — hypotension, bradycardia, facilitation of the baroreceptor reflex [11, 15], and by modulation of the mechanisms of regulation of the muscular system — facilitation of the flexor reflex, stimulation of central generators of locomotion or of scratching movements [3, 10].

Considering data on the connection between the hypotensive and certain other effects of clonidine and activation of the endogenous opiate system [8, 9], the investigation described below was carried out to study interaction between the effects of clonidine and naloxone, which blocks opiate receptors, on the mechanism of central somato-autonomic stress, which is one of the components leading to development of adaptive hemodynamic reactions to physical exertion [2].

EXPERIMENTAL METHOD

Experiments were carried out on unanesthetized decerebrate cats. During preparation of the animals for the experiments, under ether anesthesia the carotid artery and jugular vein were catheterized and a bipolar electrode was applied to the central end of the divided nerve to the gastrocnemius muscle. Miniature sensors of an ultrasonic Doppler flowmeter were applied to the ascending arch of the aorta and the common iliac artery, and the dorsal surface of the two upper segments of the spinal cord was exposed. After intercollicular decerebration, by means of a comb electrode and EN-57 electrocoagulation apparatus flaxedil was injected into the animal (3-5 mg/kg, intravenously) and it was artificially ventilated. To facilitate evocation of the scratch reflex, a solution of D-tubocurarine was applied to the exposed surface of the spinal cord [7].

The scratch reflex was evoked by mechanical stimulation of the concha auriculae [5, 6], which was accompanied by the appearance of bursting activity in the nerve to the gastrocnemius muscle. The signal from the nerve was led to a type EMT-12B universal amplifier (Sweden), after which it underwent full-wave rectification and smoothing by means of an active RC integrator. The arterial blood pressure (BP) was measured with an EMT-34 electromanometer. All parameters were recorded on the N 338-6N instrument.

Clonidine (Boehringer Ingelheim) was tested in a dose of 30 μg/kg, naloxone (Endo Laboratories) in a dose of 0.12-0.15 mg/kg. The drugs were diluted with physiological saline.

The results were subjected to statistical analysis by Student's t-test for paired comparison.

*Deceased.

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TABLE 1. Changes in Background Hemodynamic Parameters under the Influence of Clonidine (30 μg/kg) and Naloxone (0.12-0.15 mg/kg) in Decerebrate Curarized Cats (M ± m)

Conditions	BP, mm Hg	Cardiac output, ml/min	Total peripheral vascular resistance, mm Hg/ml/min	Blood flow in common iliac artery, ml/min	Vascular resistance in common iliac artery, mm Hg/ml/min
Initial responses (n=6)	126,8±10,03	281,2±52,5	0,51±0,08	18,7±2,4	7,55±1,34
Changes after clonidine (n = 6)	—2,0±5,2	—11,1±34,4	—0,034±0,038	—2,8±1,4*	2,42±0,98*
Changes after naloxone (n=5)	14,0±5,9*	15,9±36,5	0,085±0,066	2,9±5,9	—0,49±0,65

Legend. Changes in hemodynamic parameters under the influence of clonidine given relative to initial level; changes in hemodynamic parameters under the influence of naloxone given relative to level after clonidine.
*Differences significant at $P \leq 0.05$ level.

TABLE 2. Changes in Hemodynamic Responses during Central Command for Movement under the Influence of Clonidine (30 μg/kg) and Naloxone (0.12-0.15 mg/kg) in Decerebrate Curarized Cats (M ± m)

Experimental conditions	Response of BP, mm Hg	Time taken for BP to reach maximum, sec	Response of cardiac output, ml/min	Response of total peripheral vascular resistance, mm Hg/ml/min	Response of blood flow in common iliac artery, ml/min	Response of vascular resistance in common iliac artery, mm Hg/ml/min
Initial responses	57±10,6	10,3±1,5	10,0±23,4	0,232±0,08	—1,6±1,7	18,6±15,6
Changes after clonidine	—8±3,5*	4.0±1,3*	1,1±20,8	0,017±0,04	3,5±1,9	—16,4±15,4
Changes after naloxone	2±3,9	—2,1±0,8	—18,6±25,9	0,033±0,077	—2,3±1,6	0,6±0,46

Legend. Changes in hemodynamic responses after clonidine given relative to initial responses; changes in hemodynamic responses after naloxone given relative to magnitude of response after clonidine.
*Differences significant at the $P \leq 0.05$ level.

EXPERIMENTAL RESULTS

Injection of clonidine into the decerebrate animals was accompanied by slight changes in the background hemodynamic parameters (Table 1). A significant decrease was observed in the blood flow in the common iliac artery by 2.82 ml/min, with an increase in its vascular resistance by 2.42 mm Hg/min/ml. Injection of naloxone in a dose of 0.12-0.15 mg/kg after administration of clonidine caused BP to rise by 14 ± 5.9 mm Hg ($P < 0.05$). The cardiac output (CO) and the blood flow in the iliac artery showed no significant change.

The pressor response of BP arising in the decerebrate animals in response to transmission of the motor central command to the skeletal muscles, was 57 mm Hg (Table 2, Fig. 1) and reached a maximum 10.27 ± 1.5 sec after the beginning of stimulation. Clonidine had a depriming effect on the BP response, lowering its value by 8 mm Hg. Under the influence of clonidine the pressor response reached a maximum after 14 ± 0.8 sec.

By contrast with the inhibitory effect of clonidine on the cardiovascular components, the somatic components of the response were stimulated. This was shown by the appearance of a scratch reflex to fewer stimulations, the appearance of spontaneous bursts of activity in the nerve to the gastrocnemius muscle, and the longer persistence of bioelectric activity after the end of mechanical stimulation of the concha auriculae. The amplitude of the bursts of activity, assessed from the integrated record of the spike discharge, was unchanged at 17 ± 3.16 mm before and 16 ± 4.02 mm after administration of clonidine. Naloxone did not significantly change the character of the muscular and hemodynamic reactions. Shortening the time taken for the pressor response to reach its maximum, from 14 to 12.2 sec, was all that was observed (Table 2).

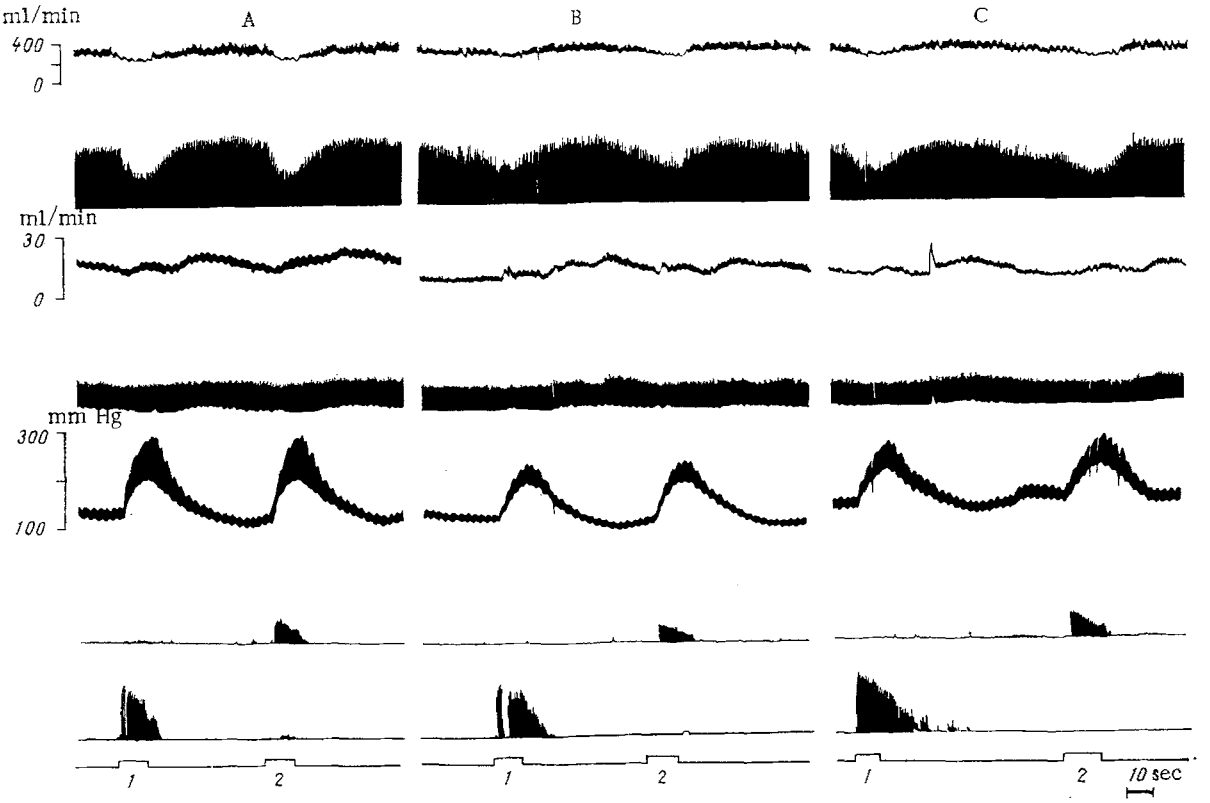

Fig. 1. Effect of 30 μg/kg clonidine (B) and 0.12-0.15 mg/kg naloxone (C) on adaptive responses of hemodynamics in decerebrate cat during transmission of central motor command to skeletal muscles (A — initial responses). From top to bottom: cardiac output (integrated recording), velocity of blood flow in aorta, blood flow in common iliac artery (integrated), phasic blood flow in common iliac artery, BP, spike discharge in nerve to right gastrocnemius muscle, spike discharge in nerve to left gastrocnemius muscle, marker of mechanical stimulation of concha auriculae. 1) Stimulation of left ear (ipsilateral relative to common iliac artery), 2) stimulation of right ear.

The results of this investigation indicate very slight inhibition of adaptive hemodynamic responses evoked by a central command for movement under the influence of clonidine. This conclusion is in full agreement with data on the small effect of clonidine on hemodynamic responses during actual physical exertion [1].

The decrease in amplitude of the pressor response, incidentally, was combined with the activating effect of clonidine on the appearance of the central command to the skeletal muscles. This property of clonidine and of L-dopa, administered after monoamine oxidase inhibitors, is known and has been used in experimental physiology to facilitate activation of the spinal locomotion generator [11, 15]. The locomotion generator formed in the region of Rexed's lamina 7 has many features in common with the generator of scratching movements and, in the opinion of Berkinblit et al. [4], the two central generators may be formed by the same neurons.

Naloxone, which blocks opiate receptors, if injected after clonidine, did not significantly change the hemodynamic responses during the central command for movement (Table 2), but it significantly raised BP from its background level by 14 mm Hg (Table 1). In this case naloxone did not affect the magnitude of the pressor response, when depressed by clonidine. Data in the literature suggest that one cause of the rise of BP may be antagonism between the effects of clonidine and naloxone [9, 10]. The basis of this antagonism, in the opinion of Farsang et al. [9], is the connection of the hypotensive effect of clonidine with activation of the endogenous peptide system of the brain, most probably by liberation of β-endorphin. Comparison with our own data suggests that this explanation is unlikely, for clonidine did not cause any significant fall in BP in the experimental model used. The rise

in BP against the background of naloxone may perhaps reflect one of its actions and may explain its antishock properties, for elevation of BP has been demonstrated under the influence of naloxone in spinal, endotoxin, and other types of shock [8, 13, 14]. It can be tentatively suggested that electrolytic decerebration may activate the release of endogenous opiate peptides, whose effect is blocked by the naloxone administered later. Considering our own data and the results of experiments conducted by Farsang et al. [9, 10], it is suggested that an endogenous opiate mechanism is of great importance for the regulation of the background parameters of the hemodynamics, but it evidently does not participate in processes of somato-autonomic stress.

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GENETIC DIFFERENCES IN THE ANTISEIZURE EFFECT AND METABOLIC RATE OF PHENAZEPAM

S. B. Seredenin, V. G. Zin'kovskii, B. A. Badyshtov, N. Ya. Golovenko, and I. V. Rybina

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KEY WORDS: phenazepam; antiseizure effect; metabolic rate.

Previous experiments have shown that the antiseizure effect of phenazepam and the rate of its oxidation *in vitro* are genetically dependent [1, 2]. The object of this investigation was to study the metabolism of the tranquilizer *in vivo* in C57BL/6 and BALB/c mice and also the character of inheritance of the antiseizure effect and the metabolic rate of the drug in first generation hybrids.

EXPERIMENTAL METHOD

Male inbred mice of lines C57BL/6 (B6) and BALB/c (C) lines and (C × B6)F_1 hybrids weighing 18-20 g were used in the experiments. To study metabolism, ^{14}C-phenazepam was injected intraperitoneally with Tween-80 in a dose of 14 mg/kg. The animals were decapitated 0.5, 1, 2, 3, and 6 h after injection of the substance. The total radioactivity of the initial compound (I), of its 3-hydroxy metabolite (II), of the total derivatives hydroxylated in the aromatic rings of the phenazepam molecule (III), and protein-bound radioactivity (IV) in the blood plasma, brain, and liver were determined [3]. Antiseizure activity of compound I against metrazol was estimated by the method described previously [2]. For statistical analy-

Laboratory of Pharmacologic Genetics, N. I. Pirogov Second Moscow Medical Institute.

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