Consequences of Progestational Hypoxia in Pregnant Rats and Its Peptide Correction (EEG Data)

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We studied immediate and delayed changes in ECG in female albino rats subjected to acute hypobaric hypoxia on days 4-5 of pregnancy and evaluated the possibility of correction of hypoxia-produced disturbances with some peptides. Acute hypoxia lengthened the mean *RR* interval, increased heart rate variability, and decreased the monotony index. Hypoxia induced considerable changes in the structure of ECG that reflected the development of arrhythmias and conduction disturbances. These changes in ECG persisted during the posthypoxic period. Intranasal administration of heptapeptides Semax and β-casomorphin-7 to pregnant females promoted recovery from acute hypoxia and normalized ECG in the posthypoxic period.

Key Words: *progestational hypoxia; pregnant females; ECG; regulatory peptides*

Hypoxia developed at various stages of pregnancy attracted much attention of physiologists and physicians over many years. The progestational period (days 4-5 of pregnancy) received little attention in this respect. Acute hypoxia at this term can promote the development of extragenital pathologies in pregnant females, violate the normal course of pregnancy and labor, and cause abnormalities of fetuses and newborns.

Peptide complexes (constellations) consisting of potentially protective components hold promise for the correction of posthypoxic consequences [4]. For example, peptides Semax and β-casomorphin-7 produce an antihypoxic and anxiolytic effects, respectively [3,5].

Here we modeled acute hypobaric hypoxia (AHH) in female rats on days 4-5 of pregnancy, studied immediate and delayed changes in ECG and autonomic regulatory balance, and evaluated the possibility of correcting the negative consequences of hypoxia with a peptide complex consisting of Semax and β-casomorphin-7.

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MATERIALS AND METHODS

Female albino rats were subjected to AHH on days 4-5 of pregnancy. The rats were maintained in an altitude chamber at 145 mm Hg, which corresponded to an altitude of 11,500 m above sea level (the ascent took 1 min). Physiological saline or peptide complex was administered intranasally 15 min before hypoxia. This treatment directly affected functional activity of the central nervous system [6]. Controls received an equivalent volume of physiological saline or peptide complex, but were not subjected to hypoxia. The peptide complex contained Semax (Met-Glu-His-Phe-Pro-Gly-Pro, $ACTH_{4-7}$-Pro-Gly-Pro) and β-casomorphin-7 (Tyr-Pro-Phe-Pro-Gly-Pro-Ile) synthesized at the Institute of Molecular Genetics (Russian Academy of Sciences). Heptapeptides were administered in a dose of 0.1 mg/kg (20 μl).

Electrodes for ECG monitoring were subcutaneously implanted to nembutal-narcotized females (30 mg/kg) 1 day before hypoxia modeling. ECG in a standard lead was recorded before, during, and after AHH. ECG recordings made in the altitude chamber over 2 min before hypoxia were taken as baseline. The rats were considered to recover from hypoxia when the length of *RR* intervals returned to the initial values.

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TABLE 1. Parameters of ECG in Pregnant Rats during and after Acute Hypobaric Hypoxia (*M*±*m*)

Parameter	Normal	High-altitude exposure	Termination of AHH	Recovery from AHH
Group 1 (*n*=17)				
RR interval, msec	145.5±2.9	120.5±2.8*	334.2±15.8*	147.8±5.1
heart rate variability, msec	35.3±3.9	31.4±5.2	178.8±23.8*	21.2±3.0*
monotony index	1.1±0.3	1.9±0.3*	0.2±0.1*	2.9±0.5*
Group 2 (*n*=6)				
RR interval, msec	158.7±6.5	122.9±3.4*	258.7±35.8*	148.7±2.2
heart rate variability, msec	30.5±4.3	30.0±5.8	191.1±20.4*	44.0±14.8
monotony index	1.1±0.2	1.5±0.4	0.1±0.0*	1.6±0.4

Note. Here and in Table 2: *$p<0.01$ compared to normal.

ECG was recorded using CONAN software and looked like a curve of the dynamics of the analog signal digitized at 273 Hz [2]. Statistical treatment of the results was performed using RR software (D. D. Vorontsov). We evaluated the mean *RR* interval and heart rate variability (ΔX, msec). The monotony index reflecting autonomic regulation of cardiac activity was calculated as $K_M=AM_0/\Delta X$, where AM_0 and ΔX are the amplitude of mode (percent of most common *RR* intervals in the sample) and scatter of data, respectively [1].

RESULTS

By the resistance to AHH all pregnant females during the progestational period were divided into 2 groups. In group 1, the first agonal inspiration serving as the signal for termination of AHH was observed 2.8 min after the onset of AHH, while in group 2 it occurred after 10 min or latter (in this case AHH was stopped after 10 min). Chronotropic changes in cardiac activity produced by AHH were similar in animals of both groups (Table 1).

Shortening of *RR* intervals and increase in the monotony index during AHH indirectly reflect activation of adrenergic regulation of cardiac chronotropism and adaptive mechanisms. The opposite effects accompanied by a considerable increase in heart rate variability and decrease in the monotony index were observed by the end of hypoxia and were associated with activation of the parasympathetic regulation and inhibition of adrenergic mechanisms. Changes in the structure of ECG during AHH were manifested in the development of arrhythmias and alteration in myocardial conduction (Fig. 1).

Test parameters in group 2 rats returned to the baseline level after AHH. In group 1 animals the duration of *RR* intervals returned to normal, while the monotony index increased, and the degree of heart rate variability increased. The changes were opposite to those observed during hypoxia and reflected stabilization of the heart rate.

ECG was recorded in freely moving rats during the posthypoxic period (days 1-5 after AHH). Changes in ECG were typical only for group 2 animals. It was probably related to a long period of oxygen deficiency in these rats (Fig. 2). These changes were manifested in lengthening of the mean *RR* interval, increase in heart rate variability, and decrease in the monotony index and reflected "freezing behavior" of nocturnal rodents in a new environment. In group 2 rats the heart rate decreased more significantly than in group 1 animals. Therefore, group 2 rats were stressed more significantly than group 1 animals.

Intranasal administration of the peptide complex to group 1 rats reduced heart rate variability from 178.8±23.8 to 100.1±19.0 msec ($p<0.05$) and increased the monotony index from 0.2±0.1 to 0.4±0.1 ($p<0.01$) by the end of AHH. However, this treatment had no effect on ECG in group 2 animals. The peptide complex stabilized cardiac chronotropism in rats of both groups during recovery from AHH. The mono-

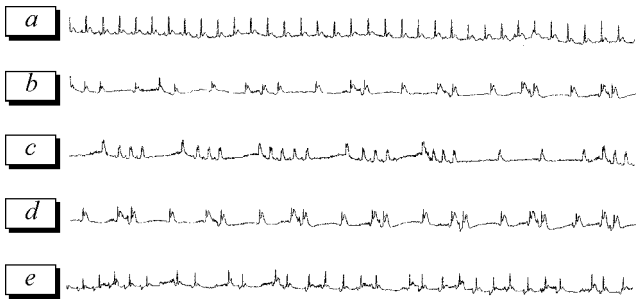

Fig. 1. Disturbances in heart rate and myocardial conduction in pregnant rats (days 4-5 of pregnancy) during acute hypobaric hypoxia (AHH): normal ECG (*a*); episode of single extrasystole and bigeminy during bradycardia (*b*); episode of recurrent tachycardia during bradycardia (*c*); episode of trigeminy with increased T waves (*d*); episode with deep Q waves (abnormal cardiac conduction).

TABLE 2. Effect of the Peptide Complex on ECG in Pregnant Rats during Recovery from Acute Hypobaric Hypoxia (*M*±*m*)

Group	*RR* interval, msec		Heart rate variability, msec		Monotony index	
	normal	recovery	normal	recovery	normal	recovery
1st						
PS+AHH (*n*=17)	145.5±2.9	147.8±5.1	35.3±3.9	21.2±3.0*	1.06±0.2	2.9±0.5*
PC+AHH (*n*=19)	147.0±2.8	150.4±5.0	32.6±3.8	19.7±4.1*	1.2±0.2	4.3±0.9*+
2nd						
PS+AHH (*n*=6)	158.7±6.5	148.7±2.2	30.5±4.3	44.0±14.8	1.1±0.2	1.6±0.5
PC+AHH (*n*=5)	149.6±8.1	137.3±1.9+	36.6±4.2	18.3±2.4*+	0.8±0.1	3.1±0.7*+

Note. PS: physiological saline; PC: peptide complex. *$p<0.01$ compared to PS+AHH group.

tony index increased in group 1 and 2 animals. Moreover, in group 2 rats we observed by shortening of the *RR* interval and decrease in heart rate variability (Table 2). Changes in ECG were not observed in group 2 animals on days 1-5 of the posthypoxic period (Fig. 2).

Our results show that pretreatment with the peptide complex produced a pronounced normalizingef-

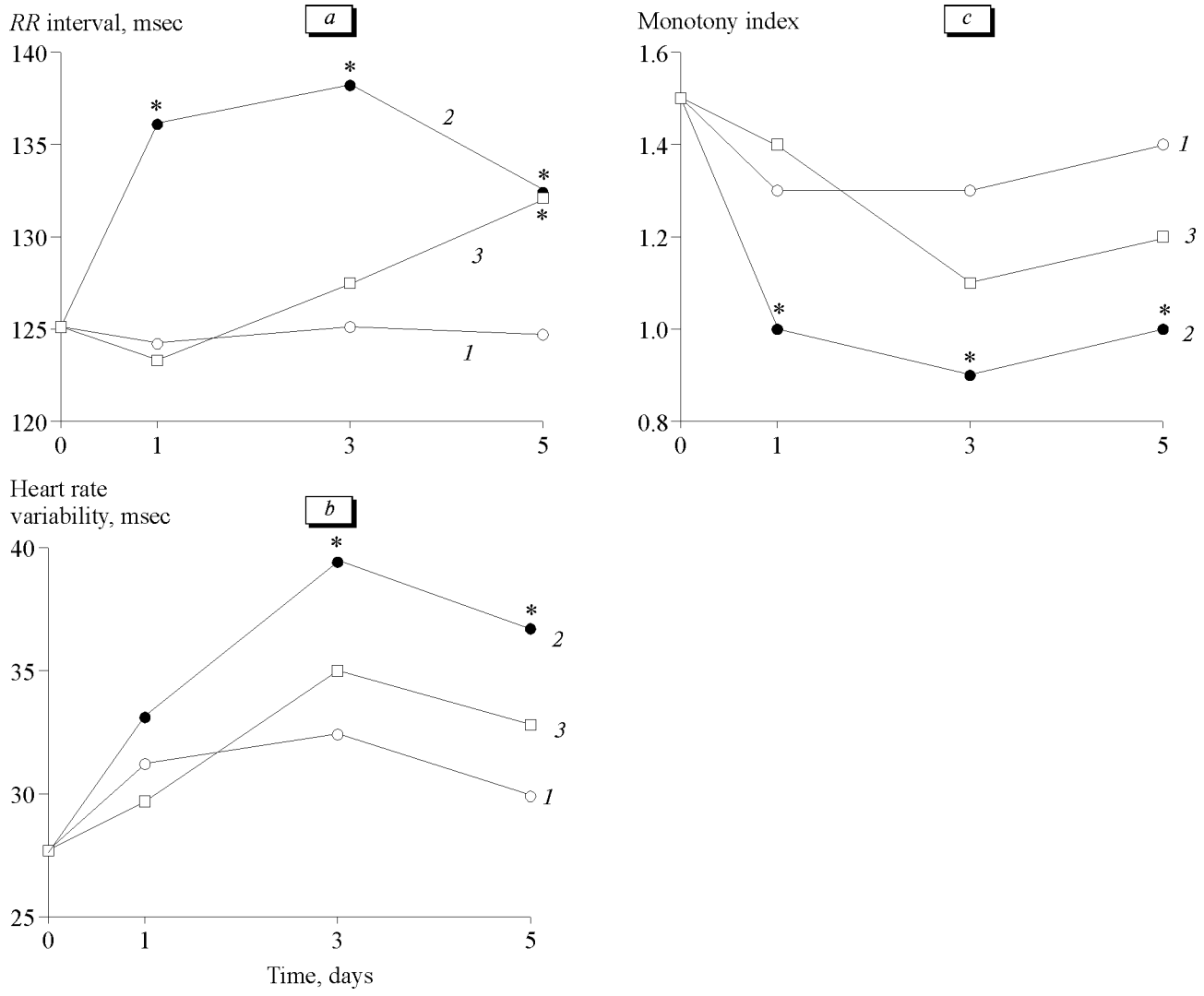

Fig. 2. Delayed effects of AHH on ECG in freely moving pregnant rats of group 2 subjected to the open-field test. Correction with the peptide complex. Control (*1*), physiological saline and AHH (*2*), peptide complex and AHH (*3*). *$p<0.05$ compared to the control.

fect related to antihypoxic and anxiolytic properties of Semax and β-casomorphin-7. High efficiency of intranasal treatment with peptides indicates that their protective effects are realized via the central mechanisms.

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