Effect of Physical Training on Blood Level of Endogenous Modulators of β-Adreno- and m-Cholinoreactivity in Patients with a History of Myocardial Infarction

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The relative content of myocyte-active factors and endogenous muscarinic receptor blocker in the blood increased, while the concentration of endogenous β-adrenoceptor sensitizer decreased in coronary patients with a history of acute myocardial infarction. Physical training produced a therapeutic effect, normalized the content of these factors and, probably, improved β-adrenergic and muscarinic cholinergic regulation of the heart and vessels.

Key Words: *endogenous chemomodulators; acute myocardial infarction; physical training*

Previous studies showed that endogenous chemoreactivity modulators belong to the endogenous system regulating activity of peripheral autonomic nervous structures in humans [9-12]. Endogenous β-adrenergic receptor sensitizer (EBARS) and endogenous muscarinic cholinergic receptor blocker (EMCRB) modulate adrenergic and cholinergic regulation of internal organs [9-12]. Published data show that modulation of the heart and vessels is impaired during coronary heart disease (CHD) and acute myocardial infarction (AMI) [2,13]. The exact mechanisms of these changes remain unknown. They can result form variations in the content of EBARS and EMCRB. Here we tested this hypothesis.

MATERIALS AND METHODS

Experiments were performed on 186 longitudinal strips of the uterine horn from 31 nonpregnant rats [11,12]. We measured the relative contents of EBARS and EMCRB in the plasma of 40 patients with AMI. The patients were divided into 4 groups. Group 4 patients (n=10) were subjected to physical training. Group 5 included 10 conventionally healthy elderly donors. The blood was taken on day 1 (group 1) and 1.5-2 (group 2) and 8 months after AMI (groups 3 and 4). All patients received standard therapy including β-adrenoceptor blockers. Physical training (6 months) was performed according to the program developed at the Research Center for Preventive Medicine in the framework of All-Russia Multicenter Study "Physical Training in Patients with IHD With Acute Coronary Accidents during Post-hospital Rehabilitation" [1]. Physical training included morning exercises, walking on level terrain (2-6 km), and group (2-3 times a week) and individual exercises (1-2 times a week).

Blood plasma was diluted by 50, 100, 500, 1000, and 10,000 times with Krebs solution (pH 7.4) containing (in mM): 136 NaCl, 4.7 KCl, 2.52 $CaCl_2$, 1.2 $MgCl_2$, 0.6 KH_2PO_4, 4.7 $NaHCO_3$, and 11 $C_6H_{12}O_6$. Contractions of muscle strips were recorded on a Miothsitograf device at 38°C [11,12]. The strips were tested with 10^{-9} g/ml epinephrine or 10^{-6} g/ml acetylcholine before, during, and after treatment with the plasma in specified dilutions (Fig. 1, *b*, *c*, *d*). The probability for the development of plasma-produced changes was determined by visual examination and logical study of mechanograms. Intergroup differences

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were evaluated by Student's *t* test and Fischer's test. The differences were significant at $p<0.05$.

RESULTS

Our results confirmed previous observation that physical training improves the efficiency of rehabilitation in patients with AMI [15]. In group 4 patients we observed increased left ventricle output (from 52.9±2.2 to 59.7±2.3%, $p<0.05$) and better tolerance to physical exercises (from 85.7±7.4 to 112.5±8.1 W, $p<0.05$). Group 3 patients were characterized by slightly increased left ventricle output (from 53.3±2.8 to 55.4±1.9%) and impaired tolerance to physical exercises (from 88.3±6.6 to 66.7±7.2 W, $p<0.05$).

Strips from rat uterine horn displayed spontaneous contractile activity. Epinephrine (10^{-9} g/ml) reversibly inhibited, while acetylcholine (10^{-6} g/ml) increased contractile activity of strips (Table 1, Fig. 1). Blood plasma produced myocyte-stimulating, myocyte-inhibitory, β-adrenoblocking, β-adrenosensitizing, and m-cholinoblocking effects.

The plasma in specified dilutions produced a myocyte-stimulating effect and reversibly increased contractile activity of 5-90% samples (Table 1, Fig. 1, *a*). The probability of these changes decreased with in-

TABLE 1. Myocyte-Active and Chemomodulatory Effects of the Plasma from Patients with Acute Myocardial Infarction (% of Samples, *M*±*m*, *n*=10)

Effects	Plasma dilution				
	1:50	1:100	1:500	1:1000	1:10,000
Myocyte-stimulating effect					
Control	90.0±6.7	80.0±8.9	25.0±9.7	5.0±4.9	0±0
1	75.0±9.7	50.0±11.2*	50.0±11.2	30.0±10.3*	30.0±10.3*#
2	90.0±6.7	85.0±8.0*	45.0±11.1	35.0±10.7*	20.0±8.9*#
3	90.0±6.7	65.0±10.7	45.0±11.1	45.0±11.1*	30.0±10.3*#
4	80.0±8.9	65.0±10.7	25.0±9.7	15.0±8.0°	0±0
Myocyte-inhibitory effect					
Control	0±0	0±0	20.0±8.9	30.0±10.3	55.0±11.1
1	0±0	15±8	15±8	20.0±8.9	10.0±6.7*
2	0±0	10.0±6.7	15±8	15±8	10.0±6.7*
3	0±0	25.0±9.7*	20.0±8.9	20.0±8.9	5.0±4.9*
4	0±0	5.0±4.9	20.0±8.9	20.0±8.9	25.0±9.7*
β-Adrenoblocking effect					
Control	10.0±9.5	10.0±9.5	10.0±9.5	0±0	0±0
1	50.0±15.8*	30.0±14.5	0±0	20.0±12.7	10.0±9.5
2	50.0±15.8*	40.0±15.5	0±0	10.0±9.5	0±0
3	60.0±15.5*	40.0±15.5	10.0±9.5	0±0	0±0
4	40.0±15.5	10.0±9.5	10.0±9.5	0±0	0±0
β-Adrenosensitizing effect					
Control	30.0±14.5	70.0±14.5	90.0±9.5	80.0±12.7	50.0±15.8
1	20.0±12.7	40.0±15.5	60.0±15.5	20.0±12.7*	0±0*#
2	10.0±9.5	20.0±12.7*	50.0±15.8*	60.0±15.5	40.0±15.5
3	40.0±15.5	50.0±15.8	70.0±14.5	40.0±15.5	10.0±9.5*
4	50.0±15.8x	40.0±15.5	70.0±14.5	90.0±9.5+°	70.0±14.5°
m-Cholinoblocking effect					
Control	80.0±9.5	80.0±9.5	30.0±14.5	20.0±12.7	0±0
1	80.0±12.7	90.0±9.5	40.0±15.5	40.0±15.5	20.0±12.7
2	80.0±12.7	80.0±12.7	60.0±15.5	10.0±9.5	0±0
3	100±0	90.0±9.5	60.0±15.5	40.0±15.5	20.0±12.7
4	90.0±9.5	60.0±15.5	20.0±12.7x°	0±0+°	0±0

Note. $p<0.05$: *compared to the control; +compared to group 1; xcompared to group 2; °compared to group 3; #compared to group 4.

creasing in plasma dilution. Taking into account published data [9-12], we hypothesized that the myocyte-stimulating effect is related to the presence of endogenous oligopeptide activator of myocyte contractility (EAMC) in the blood. In patients of groups 1, 2, and 3 with AMI myocyte-stimulating activity of the plasma (dilutions 1000 and 10,000) and EAMC content were higher than in group 4 and 5 patients ($p<0.05$). These data indicate that high content of EAMC is related to the development of AMI. The decrease in EAMC content after physical training improved the efficiency of rehabilitation after AMI.

The myocyte-inhibitory effect of blood plasma in low dilutions (1:100) was observed in 5-25% samples

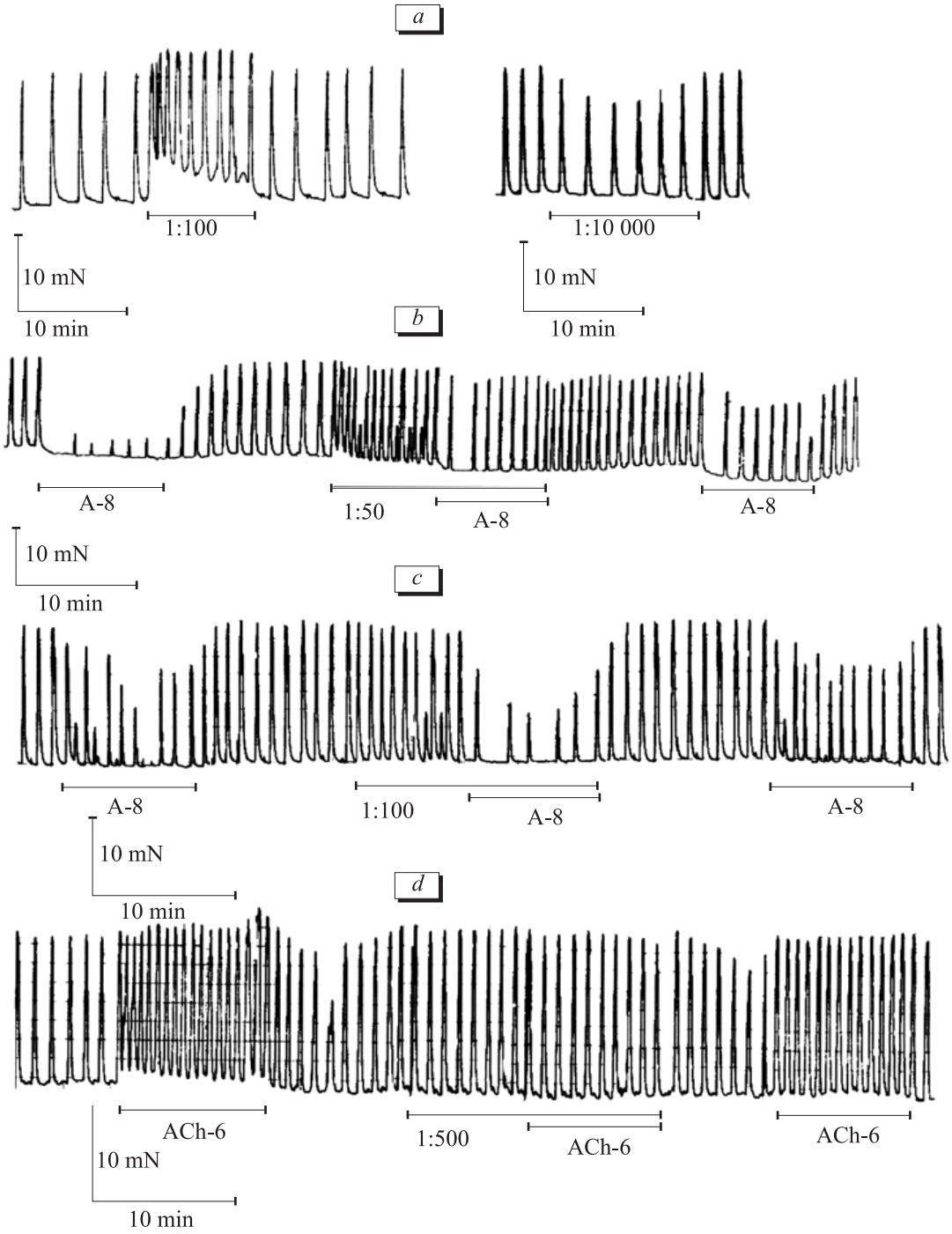

Fig. 1. Mechanogram of longitudinal strips of the uterine horn from nonpregnant rats demonstrating myocyte-stimulating (*a*), myocyte-inhibitory (*b*), β-adrenoblocking (*c*), β-adrenosensitizing (*d*), and m-cholinoblocking (*e*) reactions to the plasma from patients with acute myocardial infarction. Horizontal lines: moment of treatment with the plasma in dilutions of 1:50, 1:100, 1:500, and 1:10,000, epinephrine (10^{-8} g/ml, E-8), and acetylcholine (10^{-6} g/ml, ACh-6). Calibration: 10 mN, 10 min.

and was associated with the presence of the endogenous inhibitor of myocyte contractility (EIMC, Table 1, Fig. 1, *a*). This compound probably acts as a vasodilator. In patients of groups 1, 2, and 3 myocyte-inhibitory activity of the plasma in a dilution of 1:100 and EIMC content were higher than in group 4 and 5 patients ($p<0.05$). Published data show that production of NO and other vasodilating substances decreases during AMI [4], while physical exercises activate NO synthesis [14]. It can be hypothesized that the increase in EIMC plays a compensatory role. The demands for these changes decrease during physical training.

The myocyte-inhibitory effect of blood plasma in high dilutions (1:1000 and 1:10,000) observed in 5-55% samples can be explained by the presence of bound β-adrenoceptor agonists in the blood [10]. Their amount reflects the content of free β-adrenoceptor agonists and storing function of blood proteins. In patients of groups 1, 2, and 3 myocyte-inhibitory activity of the plasma in a dilution of 1:10,000 was lower than in group 4 and 5 patients ($p<0.05$). It cannot be excluded that catecholamine synthesis and storing function of plasma proteins decrease in patients with myocardial infarction due to hypodynamia and treatment with β-adrenoceptor blockers. Physical training normalizes these characteristics and improves the efficiency of rehabilitation after AMI.

The β-adrenoblocking effect (attentuation of epinephrine-produced inhibition) was observed in 10-60% samples treated with low dilutions of the plasma (1:50 and 1:100) from patients with AMI receiving atenolol or metoprolol. This effect was related to the presence of the exogenous β-adrenoceptor blocker (Table 1, Fig. 1, *b*). Our results suggest that the endogenous β-adrenergic receptor blocker (EBARB) is absent in the blood of healthy elderly donors, and its content does not increase after AMI or physical training.

Blood plasma in various dilutions (especially in dilutions of 1:500 and 1:1000) produced a β-adrenosensitizing effect in 10-90% samples and reversibly potentiated the inhibitory action of epinephrine (10^{-9} g/ml). Therefore, the blood from elderly donors contains EBARS that consists of histidine, tryptophan, and tyrosine (Table 1, Fig. 1, *c*) [10]. In patients of groups 1, 2, and 3 β-adrenosensitizing activity of the plasma in various dilutions and EBARS content were lower than in group 4 and 5 patients ($p<0.05$). Our results are consistent with published data that stress and AMI are accompanied by a decrease in the contents of histidine, tryptophan, and tyrosine in the blood [5]. Low content of EBARS in patients with myocardial infarction probably contributes to a decrease in the efficiency of β-adrenergic modulation of cardiomyocytes and vascular myocytes [2]. These changes are observed in patients with AMI [2]. The increase in EBARS content during physical training recovers this modulation, which probably contributes to the positive effect of exercises on rehabilitation after AMI. Our suggestion is confirmed by published data that preductal and mildronat [8] possessing β-adrenosensitizing activity are efficient in the therapy of patients with myocardial infarction [9,11,12].

Blood plasma in various dilutions produced the m-cholinoblocking effect in 10-100% samples and reversibly blocked the ability of acetylcholine (10^{-6} g/ml) to increase contractile activity of strips (Table 1, Fig. 1, *d*). These changes reflect the presence of blood EMCRB, which contains lysophosphatidylcholine [7]. The probability of this effect decreased with increasing plasma dilution. Intergroup differences indicate that in patients of groups 1, 2, and 3 m-cholinoblocking activity of the plasma (dilutions 1:500 and 1:1000) and EMCRB content were higher than in group 4 and 5 patients. Our results agree with published data that blood lysophosphatidylcholine concentration increases during stress and AMI [3]. It contributes to the reduced efficiency of m-cholinergic modulation of the heart and vessels during myocardial infarction, which is manifested in low power of rapid HF waves in the heart rhythm [13]. Physical training increases the power of rapid HF waves in healthy people [6] and, probably, promotes m-cholinergic modulation of the heart in patients with AMI and improves their rehabilitation. This effect is mediated by a decrease in the contents of EMCRB and lysophosphatidylcholine.

Our results suggest that impairment of β-adrenergic and m-cholinergic modulation during AMI [2, 13] results from the decrease in EBARS content and excessive accumulation of EMCRB. EBARS, EMCRB, EAMC, and EIMC play a role in the pathogenesis of ischemic heart disease and AMI. These data should be taken into account in developing methods for prevention and therapy of AMI based on physical training and treatment with histidine, tryptophan, tyrosine, preductal, mildronat, antioxidants, and lysophosphatidylcholine synthesis inhibitors.

REFERENCES

1. D. M. Aronov, *Kardiologiya*, No. 8, 69-80 (1998).
2. T. L. Krasnikova and S. A. Gabrusenko, *Usp. Fiziol. Nauk*, **31**, No. 2, 35-50 (2000).
3. E. S. Mazur, G. M. Zubareva, and A. V. Kargapolov, *Kardiologiya*, No. 4, 65-68 (1996).
4. E. B. Manukhina, I. Yu. Malyshev, and Yu. V. Arkhipenko, *Vestn. Ros. Akad. Med. Nauk*, No. 4, 16-21 (2000).
5. A. A. Nikolaeva, G. I. Lifshits, I. Sh. Shterental', *et al*., *Kardiologiya*, No. 1, 41-44 (1997).
6. A. D. Nozdrachev and Yu. V. Shcherbatykh, *Fiziol. Chel*., **27**, No. 5, 95-101 (2001).

7. N. V. Prokazova, N. D. Zvezdina, I. V. Suslova, *et al*., *Ros. Fiziol. Zh*., **84**, No. 10, 969-978 (1998).
8. B. A. Sidorenko and D. V. Preobrazhenskii, *Kardiologiya*, **40**, No. 9, Appl., 106-119 (2000).
9. E. N. Sizova, V. I. Tsirkin, and S. A. Dvoryanskii, *Ros. Fiziol. Zh*., **88**, No. 7, 856-864 (2002).
10. V. I. Tsirkin and S. A. Dvoryanskii, *Contractile Activity of the Uterus (Regulatory Mechanisms)* [in Russian], Kishinev (1997).
11. V. I. Tsirkin, A. D. Nozdrachev, E. N. Sizova, *et al*., *Dokl. Ros. Akad. Nauk*, **383**, No. 5, 698-701 (2002).
12. V. I. Tsirkin, E. N. Sizova, A. D. Podtetenev, *et al*., *Ros. Kardiol. Zh*., **33**, No. 1, 45-52 (2002).
13. I. S. Yavelov, A. D. Deev, E. E. Travina, *et al*., *Kardiologiya*, **39**, No. 6, 6-15 (1999).
14. P. Ennezat, S. Malendowicz, M. Testa, *et al*., *J. Am. Coll. Cardiol*., **38**, No. 1, 194-198 (2001).
15. M. Sullivan, A. La Croix, C. Buum, *et al*., *Am. J. Med*., **103**, No. 5, 48-356 (1997).