ISSN 2079-0570, Advances in Gerontology, 2016, Vol. 6, No. 3, pp. 231–237. © Pleiades Publishing, Ltd., 2016.
Original Russian Text © V.N. Perfilova, N.V. Sadikova, I.I. Prokofiev, O.V. Inozemtsev, I.N. Tyurenkov, 2016, published in Uspekhi Gerontologii, 2016, Vol. 29, No. 1, pp. 116–122.

Cardioprotective Effect of a New Glutamic Acid Derivative, Glufimet, under Conditions of Acute Immobilization and Painful Stress in Animals of Different Ages

V. N. Perfilova, N. V. Sadikova, I. I. Prokofiev, O. V. Inozemtsev, and I. N. Tyurenkov

Volgograd State Medical University, pl. Pavshikh Bortsov 1, Volgograd, 400131 Russia
e-mail: vnperfilova@gmail.com

Abstract—The effect of 24-hour immobilization and painful stress on myocardial contractility was studied in young (6-month), middle-aged (12-month) and old (24-month) female rats. A reduction of the functional heart reserves was revealed. This manifested itself in a lower growth of the myocardial contraction and relaxation rates (+dP/dt max and –dP/dt max), the left ventricular pressure (LVP), the maximum intensity of the functioning of the structures (MIFS) under conditions of increased pre- and afterload, and stimulation of the cardiac adrenergic receptors, which was especially pronounced in the group of 24-month-old rats. During the load tests, higher growth indicators of +dP/dt max, –dP/dt max, LVP, and MIFS were observed in animals of all age groups treated with glufimet at a dose of 29 mg/kg before and after stress; these indicators were most significant in older rats compared to young and middle-aged animals. A reference drug, Phenibut, improves the studied parameters equally in 6-, 12-, and 24-month-old females subjected to stress.

Keywords: immobilization and painful stress, myocardial contractility, animals of different ages, glutamic acid derivative, Phenibut

DOI: 10.1134/S2079057016030073

INTRODUCTION

Age-related morphological and functional changes in the cardiovascular system and disorders of the energy metabolism and neurohumoral regulation of cardiac performance leads to a decrease in its functional reserves and heart failure [3]. The rate of blood flow and oxygen supply to the tissues decreases with age due to morphological and functional changes in the blood vessels. Chronic hypoxia initiates reactive oxygen species (ROS) formation, which is caused by a leakage of electrons in the respiratory chain and the auto-oxidation of adrenaline that occurs due to increased activity of the sympathetic adrenal system [9, 11, 24]. Excessive amounts of ROS cause oxidative stress and damage to the cardiomyocytes. The age-dependent accumulation of collagen in the heart leads to a progressive increase in diastolic LV rigidity; regulation of the excitation–contraction cycle, which is associated with changes in intracellular Ca^{2+} metabolism, and the activity of the calcium channels in the sarcoplasmic reticulum are violated. This plays a significant role in the development of diastolic dysfunction and the depression of myocardial contractility [10, 23].

The adaptive body functions responsible for the increased sensitivity of the heart to the damaging effect of stress factors, which are the triggering mechanism of pathological changes in it, are reduced during aging, which causes a reduction of the myocardial inotropic and pump functions. The reduction is manifested by a decrease in the rate of contraction and relaxation, stroke and minute blood volume, and functional reserves of the heart [13, 20, 22]. In this regard, the search for compounds that selectively restrict the negative effects of stress on myocardial inotropic function in different age periods is expedient. We previously found that a new derivative of glutamic acid, glufimet, possesses antistress and cardioprotective effects in animals aged 4–6 months [5, 7].

The purpose of this work was to study the effect of glufimet on the heart inotropic and chronotropic reserves in rats of different age groups subjected to stress, i.e., in young (6 months), middle-aged (12 months), and old (24 months) rats.

MATERIALS AND METHODS

The experiments were carried out in outbred female rats. The animals were kept under standard vivarium conditions according to GLP regulations for conducting preclinical trials in the Russian Federation (GOST Z 51000.396 and 51000.4–96). The experimental study was approved by the Regional Independent Ethics Committee of the Volgograd Medical Scientific Center (protocol no. 12–2011).

Stress was simulated by hanging of the rats by the neck dorsal skin fold for 24 h [1]. To assess the functional state of the heart, we used the following tests: the volume load (preload), i.e., rapid, bolus, intravenous injection of saline solution at the rate of 0.3 mL per 100 g body weight; test for adrenoreactivity, i.e., intravenous injection of adrenaline at a dose of 0.0001 mg/kg; maximum isometric exercise (afterload), i.e., occlusion of the ascending aorta for 30 s [6]. Twelve groups of animals were formed: three groups of intact animals aged 6 (n = 6), 12 (n = 6), and 24 months (n = 6); three control groups of animals aged 6 (n = 8), 12 (n = 6), and 24 months (n = 7) that were subjected to stress and injected with saline (0.1 mL per 100 g body weight); three experimental groups of animals aged 6 (n = 6), 12 (n = 7), and 24 months (n = 7) that were subjected to stress and received the glutamic acid derivative glufimet at a dose of 29 mg/kg; and three experimental groups of animals aged 6 (n = 7), 12 (n = 6), and 24 months (n = 6) that were subjected to stress and received the reference drug Phenibut at a dose of 25 mg/kg. Test substances were administered intraperitoneally 10 min before and 10 min after stress. Glufimet and Phenibut at the indicated doses have the most pronounced cardioprotective effect, as was shown in our previous studies [5, 7].

The degree of the reduction in myocardial contractility during stress was assessed 30 min after the termination of immobilization. Anesthetized (chloral hydrate, 400 mg/kg) animals were intubated and transferred to the artificial lung ventilation; thoracotomy and pericardiotomy were then performed. A catheter connected to a pressure sensor (Elema, Sweden) was inserted through the apex of the heart into the left ventricle. The rate of myocardial contraction $+dP/dt$ max (mm Hg/s) and relaxation $-dP/dt$ max (mm Hg/s), left ventricular pressure (LVP, mm Hg), and heart rate (HR, bpm) were recorded with a computer hemodynamic analyzer based on BEAT software. The maximum intensity of the functioning of the structures (MIFS) was determined by calculation: (LVP mean ± HR mean)/weight of (LV + 1/3 of the interventricular septum).

Statistical processing of the results was performed in the Statistica 6.0 software package with preliminary verification of the samples for normal distribution by the Shapiro–Wilk test. The significance of differences was assessed by the Kruskal–Wallis and Siegel–Castellan tests.

RESULTS AND DISCUSSION

As a result of the study, we found lower initial rates of the myocardial contraction and relaxation, LVP, and heart rate in intact animals aged 12 and 24 months as compared with these indicators in young rats (Table 1). No differences in $+dP/dt$ max and $-dP/dt$ max in animals of different age groups subjected to stress were revealed; in this case, both values were higher than in intact female rats. LVP in 12- and 24-month-old animals was lower than that in 6-month-old and intact rats (Table 1). The initial indicators of the myocardial contraction and relaxation rates did not differ in different age groups of rats treated with glufimet; they corresponded to those in intact animals and were slightly lower than in the control group. Lower LVP was observed in old- and middle-aged females as compared with young rats, but it was not significantly different from that of the intact and control animals (Table 1). The heart rate in 24-month-old animals was lower than in 6- and 12-month-old rats of the control group. The initial indicators of myocardial contractility in the Phenibut-treated females subjected to stress slightly exceeded the values in the glufimet-treated and intact animals. LVP in the old and middle-aged rats was lower than in the young animals, higher than the values in the control group, and was equal to that of the intact animals (see Table 1).

The growth of indicators of myocardial contractility ($+dP/dt$ max and $-dP/dt$ max), LVP, heart rate, and MIFS in intact 6-month-old animals by the 20th second in response to an increase in pre- and afterload and cardiac adrenergic stimulation was significantly higher than that in the rats subjected to 24-hour stress exposure (Table 2 and Fig. 1). The growth of $+dP/dt$ max, $-dP/dt$ max, LVP, and heart rate at the 20th second did not significantly differ under the conditions of volume load in animals treated with glufimet and Phenibut before and after stress (see Table 2); it exceeded the indicators of the control group with the administration of adrenaline and clamping of the ascending aorta (see Table 2 and Fig. 1), which indicates the ability of test compounds to restrict the damaging effects of stress on the myocardium.

A similar tendency was retained in 12-month-old rats: under conditions of volume load, stimulation of cardiac adrenergic receptors, and occlusion of the ascending aorta in animals of the intact group, the growth of $+dP/dt$ max, $-dP/dt$ max, LVP, heart rate, and MIFS relative to the initial data was higher than that in the control animals subjected to stress; however, it was not significantly different from that in young rats (see Table 2 and Fig. 1). An increase in the indicators with volume load and adrenaline administration was more significant in glufimet-treated female rats than in 12-month-old control rats and young animals treated with the test compound. In the test for adrenoactivity, the growth of myocardial contraction and relaxation rates, LVP, heart rate, and MIFS in Phenibut-treated rats also exceeded that in the 6- and 12-month-old animals of the control group subjected to stress (see Table 2 and Fig. 1).

An increase in $+dP/dt$ max, $-dP/dt$ max, LVP, heart rate, and MIFS in response to the loads was significantly higher in intact 24-month-old female rats than in control animals exposed to immobilization-painful stress. When comparing the studied indicators

Table 1. Initial indicators of the myocardial contraction and relaxation rates, LVP, and heart rate in intact animals and animals of different ages subjected to stress

Indicator	Groups of animals of different ages		
	6 months	12 months	24 months
Intact			
+dP/dt max, mm Hg/s	3854.3 ± 590.1	3562.5 ± 336.5	3259.4 ± 324.2
−dP/dt max, mm Hg/s	3763.6 ± 343.2	3382 ± 290.1	3576.3 ± 481.5
LVP, mm Hg	115.7 ± 6.3	88.4 ± 7.5$^{(2)}$*	86.5 ± 4.1$^{(2)}$*
HR, bpm	272 ± 48.4	222.5 ± 45.1	249.8 ± 59.6
Stress + saline (control)			
+dP/dt max, mm Hg/s	4326.8 ± 545.7	4322.9 ± 1020.0	4496.4 ± 659.8$^{(1)}$*
−dP/dt max, mm Hg/s	4218.7 ± 767.7	3899.2 ± 1137.8	4730.5 ± 1024.3
LVP, mm Hg	105.5 ± 16.3	74.4 ± 14.9	77.7 ± 18.1
HR, bpm	324.7 ± 50.1	331.3 ± 50.7$^{(1)}$*	299.1 ± 25.7
Stress + glufimet			
+dP/dt max, mm Hg/s	4326.8 ± 545.7	4322.9 ± 1020.0	4496.4 ± 659.8$^{(1)}$*
−dP/dt max, mm Hg/s	4218.7 ± 767.7	3899.2 ± 1137.8	4730.5 ± 1024.3
LVP, mm Hg	105.5 ± 16.3	74.4 ± 14.9	77.7 ± 18.1
HR, bpm	324.7 ± 50.1	331.3 ± 50.7$^{(1)}$*	299.1 ± 25.7
Stress + Phenibut			
+dP/dt max, mm Hg/s	4620.9 ± 1220.6	4822.2 ± 1150.5	4476.4 ± 1336.4
−dP/dt max, mm Hg/s	5002.5 ± 1180.5	4852.2 ± 1012.0	4817.2 ± 1314.0
LVP, mm Hg	113.2 ± 16.9	82.9 ± 9.4	85.2 ± 21.4
HR, bpm	305.1 ± 50.7	285.2 ± 39.0	292.5 ± 34.5

$^{(1)}$* The changes are significant with respect to the intact group at $p < 0.05$ (Kruskal–Wallis test, Siegel–Castellan test); $^{(2)}$* changes are significant with respect to the group of animals aged 6 months at $p < 0.05$ (Kruskal–Wallis test, Siegel–Castellan test).

in two groups with those in 6- and 12-month-old rats, we found a considerably smaller growth in 24-month-old female rats (see Table 2 and Fig. 1).

A pronounced increase in +dP/dt max, −dP/dt max, LVP, heart rate, and MIFS was observed in glufimet-treated old animals compared with the control group and the Phenibut-treated animals, as well as young and middle-aged female rats treated with the substance under investigation (see Table 2 and Fig. 1).

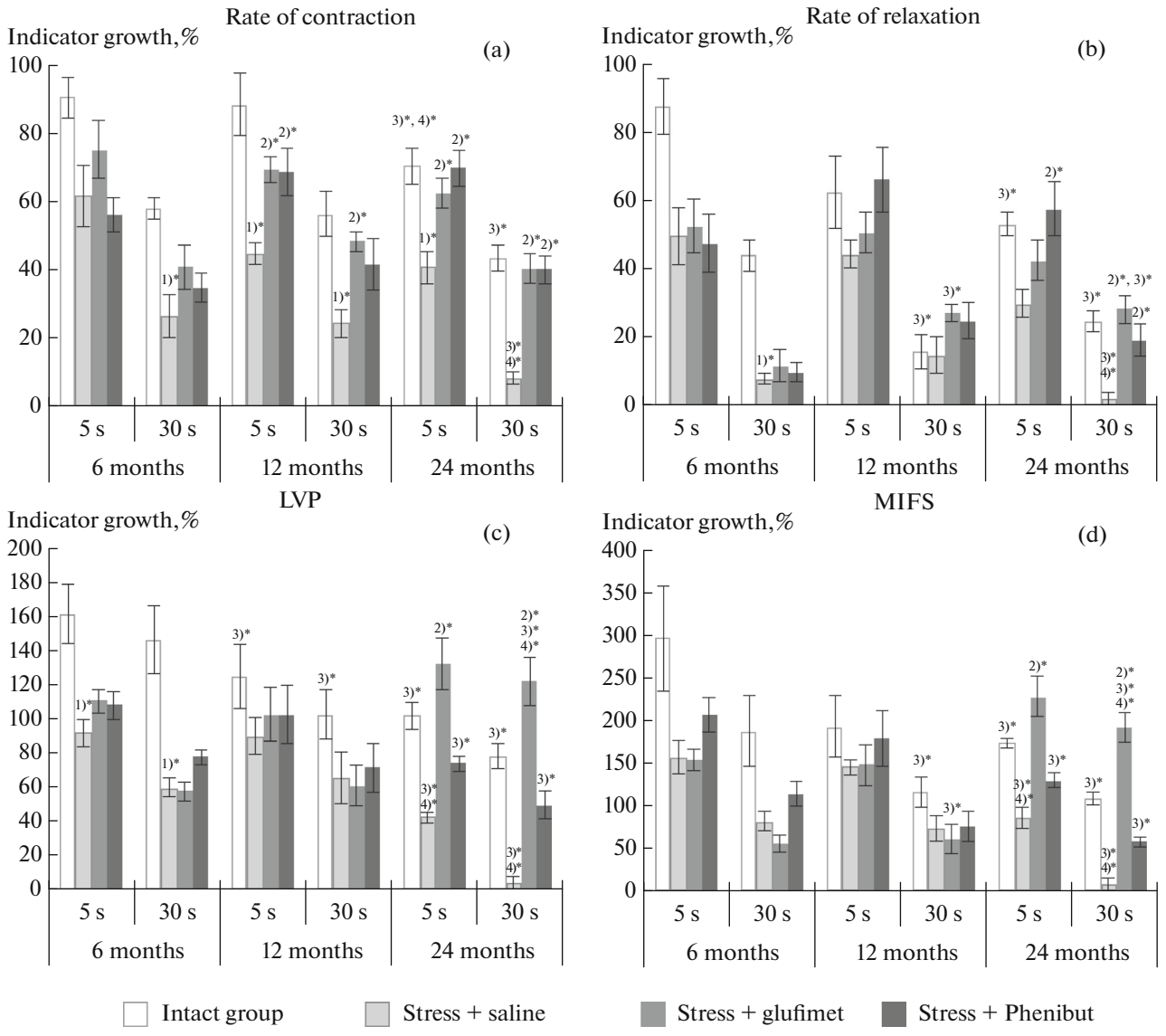
Thus, we observed a lower starting LVP in 12- and 24-month-old females compared to 6-month-old rats in all of the studied groups; the heart rate became significantly lower in old rats. A drop in LVP is likely to be due to the age-dependent decrease in the elastic properties of arteries and veins tone, the increased volume of deposited blood, and a decrease of the venous return to the heart. The sympathetic control of heart rhythm is disturbed with age, which is manifested by a decrease in the heart rate and ejection fraction [18]. The initial indicators of inotropic function (the myocardial contraction and relaxation rates) in intact animals and rats exposed to stress may not differ if there are no visible manifestations of disorders of the cardiomyocyte structure. Only when increased demands are placed on the heart are decreased ino- and chronotropic reserves and defects of contraction and relaxation detected. In the experiment, this is achieved with the use of functional tests (volume load, dosed stimulation of the cardiac adrenergic receptors, and the maximum isometric load) [1, 4, 7]. We found an age-dependent decrease in the inotropic function of the heart in animals of intact groups; statistically significant differences in indicators were revealed in 24-month-old females as compared with those in rats aged 6 and 12 months during the load tests. These results are consistent with published data: a decrease in the cardiac contractility with age was demonstrated in humans at rest; this limits the adaptive capabilities of the heart during the load [2].

Morphological functional changes in the myocardium, metabolic disorders, and disturbances of neurohumoral regulation of the heart are the most common

Table 2. Effect of glufimet and Phenibut on the myocardial contraction (+dP/dt max) and relaxation (−dP/dt max) rates, LVP, and HR in animals of different ages that were subjected to stress at carrying out of load tests

Group	Volume load, % of indicator growth				Test for adrenoactivity, % of indicator growth			
	+dP/dt max	−dP/dt max	LVP	HR	+dP/dt max	−dP/dt max	LVP	HR
Female rats aged 6 months								
Intact, $n = 6$	58.7 ± 10.3	54.4 ± 3.3	47.9 ± 4.2	6.9 ± 4.8	94.0 ± 8.6	69.6 ± 11.5	74.1 ± 12	21.9 ± 5.3
Stress + saline, $n = 8$	31.4 ± 6.1(1)*	29.0 ± 7.7(1)*	30.8 ± 6.7	−5.5 ± 2.3	38.7 ± 5.3(1)*	31.0 ± 8	31.3 ± 7.2	39.9 ± 6.3
Stress + glufimet, $n = 6$	48.8 ± 6	33.6 ± 8.2	19.2 ± 6.6	6.7 ± 5.2	44.3 ± 4	41.3 ± 5.5	43.6 ± 6.3	43.7 ± 11.9
Stress + Phenibut, $n = 7$	50.3 ± 6.2	29.8 ± 7.2	31.8 ± 11.4	4.8 ± 2.7	46.4 ± 6.1	25.6 ± 3.9	48.4 ± 9.7	28.8 ± 9.3
Female rats aged 12 months								
Intact, $n = 6$	52.1 ± 6.9	59.0 ± 7.6	39.7 ± 8.5(3)*	10.9 ± 8.8	69.9 ± 6.8	66.1 ± 6.7	59.8 ± 8.0	37.0 ± 9.7
Stress + saline, $n = 6$	31.7 ± 3.4(1)*	44.3 ± 10.7(1)*	19.8 ± 9.6	7.3 ± 6.4	32.3 ± 4.2(1)*	25.6 ± 2.9(1)*	21.4±3.9*(1)*	31.3 ± 8.0
Stress + glufimet, $n = 7$	70.0 ± 6.4(2)*, (3)*	44.9 ± 9.3	23.0 ± 4.9	6.2 ± 1.0	68.2 ± 1.2(2)*, (3)*	68.2 ± 4.3(2)*.3)*	55.9 ± 6.7(2)*	17.2 ± 5.6
Stress + Phenibut, $n = 6$	57.4 ± 7.5	43.5 ± 8.4	32.0 ± 7.9	9.5 ± 4.8	70.9 ± 9.8(2)*, (3)*	57.7±9.5(2)*, (3)*	60.2 ± 5.6(2)*	34.8 ± 17.2
Female rats aged 24 months								
Intact, $n = 6$	35.3 ± 8.2(3)*, (4)*	35.9 ± 11.5(3)*, (4)*	37.9 ± 5.6(3)*	8.9 ± 5	79.7 ± 6(3)*	44.4 ± 7.9(3)*	33.8 ± 5.8(3)*	39.7 ± 5.1
Stress + saline, $n = 7$	9.1 ± 1.6(1)*, (3)*	10.2 ± 2.4(3)*	8.5 ± 5(1)*.3)*	0.3 ± 2.1	13.9±2.1(1)*, (3)*	19.7 ± 5.9(3)*	12.5 ± 2.3(3)*	15.1 ± 7.2
Stress + glufimet, $n = 7$	79.2 ± 6.5(2)*, *3)*	64.5 ± 5.1(2)*, (3)*	51.6 ± 6.6(2)*	22.9±7.8(2)*, (3)*, (4)*	65.0 ± 6.9(2)*, (3)*	50.0±6.1(2)*, (3)*	61.4 ± 8.3(2)*	22.3 ± 8.3
Stress + Phenibut, $n = 6$	51.7 ± 8.5(2)*	57.1 ± 9(2)*	36.6 ± 9.3(2)*	16.7 ± 6.2(2)*	48.1 ± 8.3(2)*	46.5 ± 7.6(2)*	58.5 ± 9.2)2)*	39 ± 7.9

(1)* The changes are significant with respect to the intact group; (2)* changes are significant with respect to the control group of animals subjected to stress; (3)* changes are significant with respect to a similar group of animals aged 6 months; (4)* changes are significant with respect to a similar group of animals aged 12 months; $p < 0.05$ (Kruskal–Wallis test, Siegel–Castellan test).



Effect of glufimet and Phenibut on (a) the rate of myocardial contraction, (b) the rate of myocardial relaxation, c) LVP, and (d) maximum intensity of the functioning of the structures (MIFS) in animals of different ages subjected to stress under conditions of isometric load. $^{(1)}$* The changes are significant with respect to the intact group; $^{(2)}$* changes are significant with respect to the control group of rats subjected to stress; $^{(3)}$* changes are significant with respect to a similar group of animals aged 6 months; $^{(4)}$* changes are significant with respect to a similar group of animals aged 12 months; $p < 0.05$ (Kruskal–Wallis test, Siegel–Castellan test)

cause of a reduction of the functional reserves [2, 12, 14]. The cardiomyocytes number decreases with age; the content of slow myosin isoforms increases; focal degeneration of muscle fibers and myocardial fibrosis are developed; a shortage of AMP-activated protein kinase (which controls the cellular energy balance) is observed; calcium exchange between the sarcoplasmic reticulum and mitochondria is disturbed; and oxidative stress is developed [10, 15–17, 21, 24]. One more essential cause of the decreased contractile function of myocardium during aging is hypoxia, which develops with age and is associated with impaired oxygenation of the blood in the lungs, reduced coronary blood flow, and myocardial oxygen uptake. Chronic hypoxia aggravates the development of metabolic changes in cardiomyocytes and promotes contractile dysfunction of myocardium in elderly and old people [11].

It was shown that the range of compensatory reactions is limited in elderly and senile patients during stressful situations and the development of pathological processes, since the system of physiological adaptation mechanisms in older age is functioning under normal conditions of vital activity without additional loads [2]. In this regard, heart failure in elderly and

senile patients arises earlier and progresses faster. The results of our study are consistent with the published data. We observed a smaller increase in the myocardial contraction and relaxation rates, LVP, and heart rate in response to loads in the animals of control groups of different ages that were exposed to stress compared with these indicators in intact rats. The increase of the studied parameters was significantly lower in old female rats than in rats aged 6 and 12 months.

Higher indicators during load tests were recorded in animals of different ages in test groups treated with glufimet before and after the stress as compared with the control groups. At the same time, the most pronounced growth of $+dP/dt$ max, $-dP/dt$ max, LVP, and MIFS was observed in 24-month-old rats as compared with rats aged 6 and 12 months. The reference drug Phenibut improved the studied indicators equally in rats of different ages exposed to stress. The cardioprotective effect of glufimet is likely to be due to its antioxidant activity, as was shown for glutamic acid and its derivatives [8]. Glutamic acid exhibits an antihypoxic activity; it is converted in the body to GABA, which, in turn, is transformed into succinic acid via succinic semialdehyde and enters the Krebs cycle, thus increasing the energy supply of the cells [19]. The interaction of glufimet with the GABA-ergic system and the activation of its stress-limiting action are also possible. The age-dependent increase of cardioprotective effect of glufimet can be due to the state of the liver microsomal system and reduced activity of cytochrome P450, which contributes to a more prolonged maintenance of its high concentration in tissues.

CONCLUSIONS

Exposure to stress for 24 h causes a decrease in the heart inotropic reserves, which was most pronounced in old rats, as indicated by the low growth of the myocardial contraction and relaxation rates, LVP, and heart rate in the control group as compared with these indicators in intact female rats during exercise testing.

A new derivative of glutamic acid, glufimet, restricts the damaging effect of stress on the myocardium and retains the heart functional reserves at a higher level. The most pronounced effect was observed in animals aged 24 months, as evidenced by the high growth of $+dP/dt$ max, $-dP/dt$ max, LVP, and the maximum intensity of the functioning of structures in the glufimet-treated rats compared with control animals.

REFERENCES

1. Kovalev, G.V., Gurbanov, K.G., and Tyurenkov, I.N., Influence of sedatives on functions of myocardium after stress damage, *Farmakol. Toksikol.*, 1983, vol. 46, no. 3, pp. 41–44.
2. Korkushko, O.V. and Yaroshenko, Yu.T., Reserves of general functions of cardiovascular system during aging, *Probl. Stareniya Dolgoletiya*, 2012, no. 2, pp. 119– 152.
3. Korkushko, O.V., Specific tissue oxygen metabolism in elderly patients with chronic heart failure, *Krovoobrashchenie Gemostaz*, 2012, no. 3, pp. 5–26.
4. Meerson, F.Z. and Pshennikova, M.G., *Adaptatsiya k stressornym situatsiyam i fizicheskim nagruzkam* (Adaptation to the Stress and Physical Trainings), Moscow: Meditsina, 1988.
5. Perfilova, V.N., Sadikova, N.V., Berestovitskaya, V.M., and Vasil'eva, O.S., Cardiac protectors of new derivatives of glutamic acid in stress, *Eksp. Klin. Farmakol.*, 2014, vol. 77, no. 9, pp. 13–17.
6. Tyurenkov, I.N. and Perfilova, V.N., Methodological recommendations for pre-clinical study of cardiotonic activity of medical drugs, in *Rukovodstvo po provedeniyu doklinicheskikh issledovanii lekarstvennykh sredstv* (Manual on Pre-Clinical Studies of Medical Drugs), Moscow: Grif i K, 2012, part 1.
7. Tyurenkov, I.N., Volotova, E.V., Kurkin, D.V., Mazina, N.V., Bakulin, D.A., Logvinov, I.O., and Antipova, T.A., Neuroprotective and antioxidant effects of neuroglutam in cerebral ischemia, *Bull. Exp. Biol. Med.*, 2015, vol. 159, no. 3, pp. 365–367.
8. Udintsev, N.A. and Ivanov, V.V., Antioxidant effect of glutamic acid, *Patol. Fiziol.*, 1984, no. 4, pp. 60–62.
9. Shustanova, T.A., Bondarenko, T.I., and Miliutina, N.P., Free radical mechanism of the cold stress development in rats, *Ross. Fiziol. Zh.*, 2004, vol. 90, no. 1, pp. 73–82.
10. Biernacka, A. and Frangogiannis, N.G., Aging and cardiac fibrosis, *Aging Dis.*, 2011, vol. 2, no. 2, pp. 158–173.
11. Cataldi, A. and Di Giulio, C., "Oxygen supply" as modulator of aging processes: hypoxia and hyperoxia models for aging studies, *Curr. Aging Sci.*, 2009, vol. 2, no. 2, pp. 95–102.
12. Hua, Y., Zhang, Y., Ceylan-Isik, A.F., et al., Chronic Akt activation accentuates aging-induced cardiac hypertrophy and myocardial contractile dysfunction: role of autophagy, *Basic Res. Cardiol.*, 2011, vol. 106, no. 6, pp. 1173–1191.
13. Ikeda, Y., Sciarretta, S., Nagarajan, N., et al., Insights into the role of mitochondrial dynamics and autophagy during oxidative stress and aging in the heart, *Oxid. Med. Cell. Longevity*, 2014, art. ID 210934. doi 10.1155/2014/210934
14. Khan, M., Steppan, J., Schuleri, K.H., et al., Upregulation of arginase-II contributes to decreased age-related myocardial contractile reserve, *Eur. J. Appl. Physiol.*, 2012, vol. 112, no. 8, pp. 2933–2941.
15. Kwak, H.B., Effects of aging and exercise training on apoptosis in the heart, *J. Exercise Rehab.*, 2013, vol. 9, no. 2, pp. 212–219.
16. Olivetti, G., Melissari, M., Capasso, J.M., and Anversa, P., Cardiomyopathy of the aging human heart. Myocyte loss and reactive cellular hypertrophy, *Circ. Res.*, 1991, vol. 68, no. 6, pp. 1560–1568.
17. Strait, J.B. and Lakatta, E.G., Aging-associated cardiovascular changes and their relationship to heart failure, *Heart Failure Clin.*, 2012, vol. 8, no. 1, pp. 143–164.

18. Stratton, J.R., Levy, W.C., Caldwell, J.H., et al., Effects of aging on cardiovascular responses to parasympathetic withdrawal, *J. Am. Coll. Cardiol.*, 2003, vol. 41, no. 11, pp. 2077–2083.

19. Tiwari, V., Susmitha, A., and Patel, A.B., Glutamatergic and GABAergic TCA cycle and neurotransmitter cycling fluxes in different regions of mouse brain, *J. Cereb. Blood Flow Metab.*, 2013, vol. 33, pp. 1523–1531.

20. Townsend, D., Daly, M., Chamberlain, J.S., and Metzger, J.M., Age-dependent dystrophin loss and genetic reconstitution establish a molecular link between dystrophin and heart performance during aging, *Mol. Ther.*, 2011, vol. 19, no. 10, pp. 1821–1825.

21. Turdi, S., Fan, X., Li, J., et al., AMP-activated protein kinase deficiency exacerbates aging-induced myocardial contractile dysfunction, *Aging Cell*, 2010, vol. 9, no. 4, pp. 592–606.

22. Uchino, B.N., Holt-Lunstad, J., Bloor, L.E., and Campo, R.A., Aging and cardiovascular reactivity to stress: longitudinal evidence for changes in stress reactivity, *Psychol. Aging*, 2005, vol. 20, no. 1, pp. 134–143.

23. Unsöld, B., Schotola, H., Jacobshagen, C., et al., Age-dependent changes in contractile function and passive elastic properties of myocardium from mice lacking muscle LIM protein (MLP), *Eur. J. Heart Failure*, 2012, vol. 14, no. 4, pp. 30–37.

24. Zhang, Y., Mi, S.L., Hu, N., et al., Mitochondrial aldehyde dehydrogenase 2 accentuates aging-induced cardiac remodeling and contractile dysfunction: role of AMPK, Sirt1, and mitochondrial function, *Free Radicals Biol. Med.*, 2014, vol. 71, pp. 208–220.

Translated by G. Levit