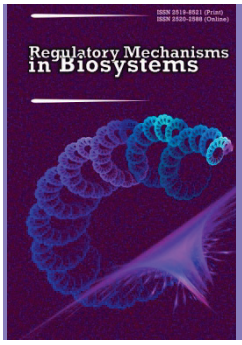

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Effects of germanium organic compound and α-lipoic acid on STZ-induced diabetes manifestations in rats

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Deep deteriorations accompany diabetes in all types of metabolism with an impact on functional activity and complications that are of vital significance. Damage to the liver contributes to the worsening of the patient's state. Oxidative stress makes a pivotal contribution to diabetes-induced liver disorders. The investigation of niacin-oxyethylidene-diphosphonate germanate (MIGU-4) and α-lipoic acid treatment, which both possessed antioxidative properties upon the functional state in rats with streptozotocin induced diabetes, was the aim of the investigation. Treatment with MIGU-4 (25.0 mg/kg, i.p.) and α-lipoic acid (50.0 mg/kg, i.p.) started six weeks after induction of diabetes by streptozotocin (65.0 mg/kg, i.p.) administration.and was performed daily for a further six weeks. The results obtained revealed that combined administration of MIGU-4 (25.0 mg/kg) and α-lipoic acid (50.0 mg/kg) caused an increase of albumin and a decrease in bilirubin when compared with streptozotocin-treated rats. Administration of MIGU-4 and α-lipoic acid caused the reduction of cholesterol and triglycerides, low-density lipoproteins, while high-density lipoproteins increased. Besides, treatment with MIGU-4 and α-lipoic acid reduced the malone dialdehyde, increased reduced glutathione content, increased the superoxide dismutase and catalase activity compared with the streptozotocin-diabetes. The protective effect of combined treatment is more pronounced than that caused by α-lipoic acid (50.0 mg/kg) treatment alone. There is ample scope for further investigations of MIGU-4 in treating diabetes-induced liver damage and complex metabolic disorders.

Keywords: organic germanium; liver function; lipid metabolism; diabetes; α-lipoic acid; malone dialdehyde.

Introduction

Diabetes is a chronic and dangerous disease that is characterized as the most severe epidemic all over the world, with serious consequences for both the health of patients and the economy of countries (Kharroubi & Darwish, 2015; GBD 2021 Diabetes Collaborators, 2023). According to the WHO and the International Diabetes Federation, the number of people with diabetes mellitus (DM) among adults aged 20 to 79 worldwide is almost 537 million. It will reach 643 million by 2030 and 783 million by 2045. Three out of four people with diabetes are citizens of low- and middle-income countries. It is forecast that by 2050, more than 1.31 billion people will suffer from diabetes. A 14-year reduction in life expectancy is assumed for patients 50 years old with a second type of diabetes diagnosed at 30. Life expectancy is six years shorter among those diagnosed at age 50 (GBD 2021 Diabetes Collaborators, 2023).

Debilitating complications such as heart insufficiency, acute heart failure, retinopathy, and kidney failure are not properly controlled (Yu et al., 2015; Thomas et al., 2019; Zborovska et al., 2019). Diabetes retinopathy is the first causative among newly diagnosed blindness in 18–64 years adults. Pronounced vision reduction or blindness was registered in 11.8% of U.S. 18 years and older patients suffering from diabetes in 2019. Similarly, the estimated ratio for chronic kidney diseases was 39.2%. Over 9% of the population in Ukraine is living with diabetes mellitus (Alifanov et al., 2019; Alifanov & Sakovych, 2022). Hence, drastic complications such as heart attacks, strokes, kidney failure, blindness, and lower-limb amputations, if untreated, are expected (Alessi & Yankiv, 2022).

The main links in the pathogenesis of diabetes are glucotoxicity with impaired microcirculation, pathological increase in blood-tissual barrier permeability with angiogenesis, and chronic inflammation (Ramesh et al., 2017; Zhang et al., 2022; Zhang et al., 2024). Tissues and organs with high metabolic activity suffer most from such a spectrum of pathogenic impacts. Complex deterioration of main metabolic processes, involving a huge number of cellular signaling pathways, results in the deterioration of functionality of parenchyma cells in different tissues, and the liver is among the most affected ones (Martín-Carro et al., 2023; Xie et al., 2023).

To model diabetes, the administration of streptozotocin (STZ) is the most widely used technique, whose action is characterized by selective toxicity to pancreatic β-cells (Kottaisamy et al., 2021; Dinić et al., 2022). Under the influence of STZ, insulin secretion by β-cells is disrupted by DNA methylation, which leads to an increase in the activity of adenosine-diphosphate-ribose polymerase (PARP) with a subsequent critical decreese in the production of nicotinamide adenine dinucleotide and ATP. At the final stage, intracellular nitric stress occurs with excessive nitric oxide production, which causes DNA fragmentation, making insulin production impossible (Dinić et al., 2022). Thus, the critical events in the onset and further development of diabetes are associated with production of free radicals (Kresyun & Godlevskii, 2014; Dinić et al., 2022).

It has been established that using organic germanium compounds effectively treats diseases in the pathogenesis of which there are mechanisms of inflammation, oxidative stress, and a decrease in immunological reactivity, including manifestations of diabetes mellitus (Luo et al., 2023). Accordingly, the antioxidant effectiveness of niacin-oxyethylene diphosphonate germanate (MIGU-4) provides a corrective effect on the manifestations of experimental diabetes mellitus (Al-Nadawi, 2023; Kresyun & Al-Nadawi, 2023). It is worth noting that niacin, as a component of MIGU-4, can also block the manifestations of alloxan-induced diabetes in rats when administered at doses of 10.0 and 15.0 mg/kg (Abdullah et al., 2018). These properties of the components of MIGU-4 indicate significant prospects for its use in diabetes mellitus.

Germanium-containing organic compounds can prevent and suppress neuroinflammation (Kim et al., 2017; Wada et al., 2018; Lee, 2023). Among anti-inflammatory and neurodegenerative action mechanisms, antioxidant effects play a leading role (Wada et al., 2018). The perspectives for clinical usage originated from the low toxicity and high biological activity of certain germanium-containing derivatives, such as germanium bound to oxyethylidene-diphosphonate acid (Wada et al., 2018; Lee, 2023). Also, the widening spectrum of the effectiveness of germanium organic compounds that was observed recently has attracted attention. Thus, anticancer and antiinfectuous properties, ability to regulate electrolyte homeostasis, along with mentioned antiinflammatory action compose the grounds for facilitation of introducing germanium-containing drugs in clinical practice (Godovan & Kresyun, 2007; Al-Nadawi & Kresyun, 2023).

It has been established that niacin bound to oxyethylidene-diphosphonate germanate offers a wide range of beneficial effects, providing reassurance about its potential. It is characterized by easy penetration into the blood, high accessibility to tissues, demonstrates protection of hepatocytes on the model of toxic hepatitis, normalizes mitochondrial function and energetic metabolism, prevents seizures, and causes a pronounced antioxidant action (Godovan & Kresyun, 2007; Al-Nadawi & Kresyun, 2023).

α-Lipoic acid (α-LA), a well-known drug, has proven effective in treating diabetes neuropathy (Rochette et al., 2015). Due to its antioxidant potency, it is supposed to be effective against diabetes complications, including laver lipodystrophy (Ko et al., 2021; Genazzani et al., 2024).

Meanwhile, the effectiveness of germanium organic compound in STZ diabetes upon a typical complex of diabetes manifestation has not been investigated systematically. Hence, the emergence of numerous complications, in particular from the functional state of the liver and metabolic disorders associated with hepatocyte activity, indicates the need to study methods of pharmacological correction of diabetes at the systemic level and requires further research (Kresyn & Godlevskii, 2014; Wada et al., 2018).

The aim of the study is to comprehensively identify the effectiveness of niacin-oxyethylidene-diphosphonate germanate (MIGU-4) in relation to hyperglycemia, insulin content, as well as the content of proteins, bilirubin, cholesterol, triglycerides, low and high-density lipoproteins (LDL and HDL, respectively) in the blood serum, superoxide dismutase activity (EC 1.15.1.1; SOD), catalase (EC 1.11.1.6; CT), malondialdehyde (MDA) and reduced glutathione levels (GSH) in the liver tissue of rats with STZ-induced diabetes after a course of MIGU-4 administration. A separate task was to compare the effectiveness of MIGU-4 with the use of α-LA.

Material and methods

All animal experiments adhered to the National Institutes of Health (NIH) Guidelines for the Care and Use of Laboratory Animals and the Helsinki Declaration and ARRIVE demands. The study was approved by the Bioethics Committee of Odesa National Medical University (Meeting Minutes No. 3 dated March 14, 2018). 32 Wistar male rats, aged 6 to 9 months, were used in this experiment. They were maintained in an environmentally controlled room (23 ± 2 °C, 60% humidity) on a 12:12-h light-dark cycle, and fed and watered ad libitum.

Diabetes was induced using i.p. streptozotocin (STZ) (Sigma Aldrich, USA) (65.0 mg/kg, i.p.), which was dissolved in a sodium-citrate buffer solution (pH 4.5). Rats that exhibited glucose levels higher than 16.7 mMol/L in venous blood one week after STZ administration were included in the study. Glucose levels were determined in a fasted state at 9.00 AM. Rats received insulin injections (0.5–1.0 IU s.c., two to five injections per week) during all observation periods (Al-Nadawi & Kresyun, 2023).

The control group consisted of intact rats (n = 10). Animals with STZ administration were randomly assigned to the following groups: Rats (n = 9), with i.p.administration of 0.9% NaCl solution; Group (n = 8) received i.p., (±)-α-LA (50.0 mg/kg, i.p.) (Sigma Aldrich, USA). Rats (n = 8) were treated daily with MIGU-4 (25.0 mg/kg, i.p.). Rats (n = 8) were treated daily with MIGU-4 (25.0 mg/kg, i.p.) and α-LA (50.0 mg/kg, i.p.). Treatment started six weeks after STZ, and the animals were administered the drug 30–40 minutes before eating daily until the end of the observation period (12 weeks) (Fig. 1).

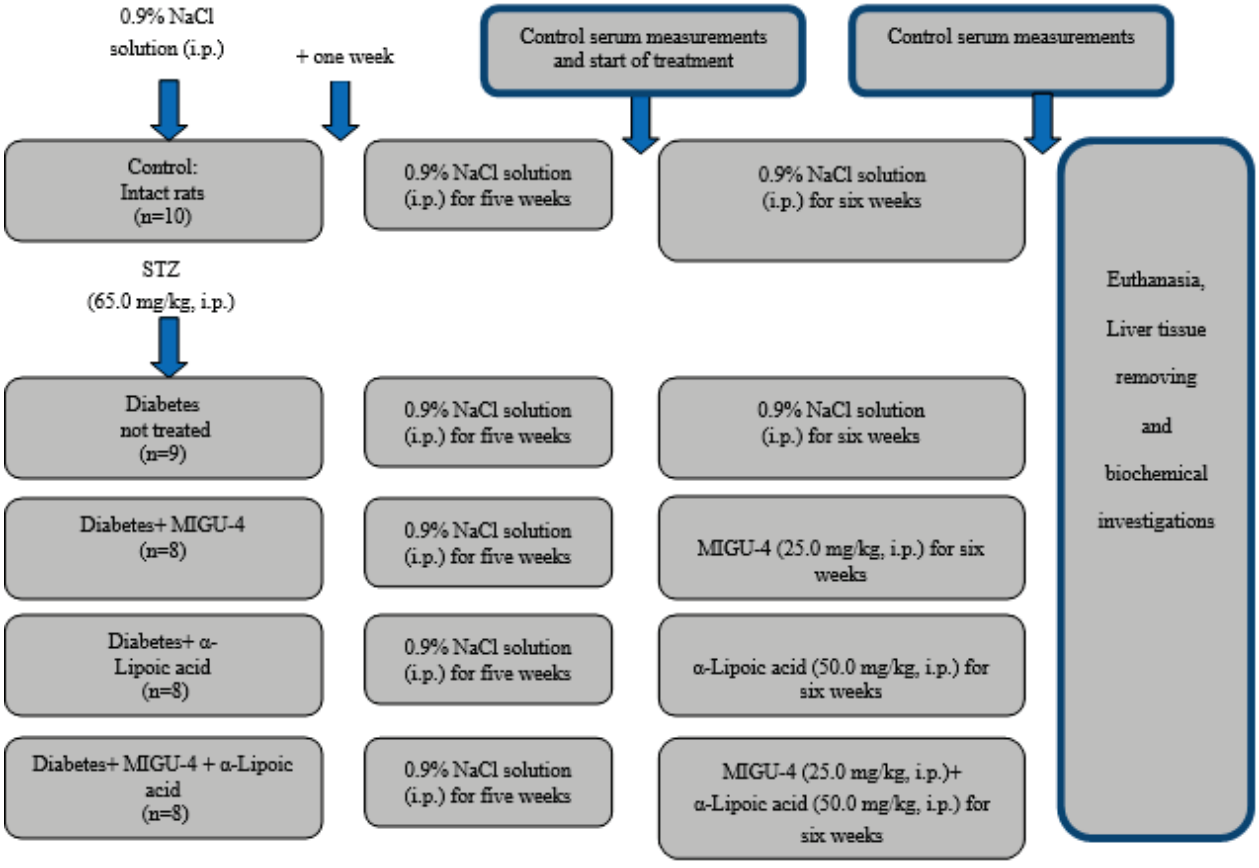

Fig. 1. Design of investigations

After the administration of STZ, the experimental animals were observed for six weeks, after which the treatment was applied for the next six weeks. 24 hours after the last application of the study drugs, the studied parameters were measured.

The insulin (Actrapid NM, Novo Nordisk, Denmark) was administered to animals 30–40 minutes before meals once every two days. Niacinoxyethylene diphosphonate germanate (MIGU-4, synthesized under the supervision of Prof. I. Y. Seifullina, Odesa I. I. Mechnikov National Medical University) was administered at doses of 25.0 mg/kg, i.p. MIGU-4 was administered for six weeks, starting on the thirtieth day from the inclusion of rats in the observation. In a separate group, (±)-α-LA (Sigma Aldrich) was administered at 50.0 mg/kg, i.p. The control rats were injected with 0.5 mL of 0.9% sodium chloride saline under similar conditions.

Food intake was determined by subtracting food residues from the diet given to the rats at 2-day intervals; the rats' daily body weight gain was also calculated, and daily water intake was taken into account (Abdullah et al., 2018).

24 h after the last injection, after euthanasia, which was performed by i.p. injection of pentobarbital (100.0 mg/kg), liver tissue was removed from the rats for further studies.

Total cholesterol, triglycerides, HDL, and LDL were measured calorimetrically at 520 nm. In particular, the content of HDL and LDL was determined by the method, which consisted of preliminary precipitation of serum LDL using polyethylene glycol and expressed in mg/dL (Al-Muzafar et al., 2021). The serum insulin content was determined by enzyme-linked immunosorbent assay (ELISA) using reagent kits from Diaclone (France) (Nithiya & Udayakumar, 2018). The state of lipid peroxidation in the blood serum was determined by the content of malondialdehyde (MDA), one of the most important end products. MDA was determined by the method (Kresyun & Al-Nadawi, 2023) and expressed as μmol/mg protein. Protein content was determined by Lowry's method (Stalnaya & Harishvili, 1977). Albumin was determined by the bromocresol method and expressed as g/dL (Moreira et al., 2018). The most informative criteria for the functional state of the enzymatic part of antioxidant defense are the activity of superoxide dismutase (SOD), catalase (CT), and reduced glutathione (GSH) content, which were determined by the previously described methods (Kresyun, & Godlevskii, 2014) and expressed as U/mg protein, nmol H_2O_2/min/mg protein, and mmol/g protein, respectively.

Values were compared using one-way ANOVA with the Bonferroni post hoc test. Results were presented as x ± SE (main ± standard error). The Shapiro-Wilk test for normality was used. P values < 0.05 were considered significant.

Results

The results showed that in rats with diabetes, in the twelfth week after STZ, blood glucose was 4.7 times higher than in intact rats, and insulin levels were 36.0% lower (P < 0.05) (Table 1). In addition, an increase in water and food consumption was observed – by 4.2 times and 30.4%, respectively, and a decrease in body weight growth by 37.2% (P < 0.05).

The use of MIIGU-4 at a dose of 25.0 mg/kg was accompanied by a significant decrease in glucose levels – by 42.8% (P < 0.05), which at the same time remained 72.7% higher than in the control group (Table 1). The insulin content significantly increased by 22.3% and remained 17.5% lower than in the control group (P < 0.05). Daily water intake decreased by 52.9% (P < 0.05), food intake – by 22.2% (P > 0.05), and daily body weight gain increased by 15.7% (P < 0.05). In the background of α-LA (50.0 mg/kg) administration, glucose content decreased by 31.7% and, at the same time, was higher than in the control group by 27.3% (P < 0.05). There was also a decrease in water and food consumption by 20.9% and 14.5%, respectively (P < 0.05). Daily body weight gain exceeded that of diabetic rats by 6.9% (P > 0.05, Table 1).

Table 1
Markers of diabetes at the end of treatment (x ± SE)

Groups	Glucose, mg/dL	Insulin, mg/L	Water consumption, mL/day	Food consumption, g/day	Increase of body weight, g/day
Control (n = 10)	933 ± 31	50.1 ± 1.9	26.3 ± 2.1	17.9 ± 1.2	4.3 ± 0.2
STZ-diabetes (n = 9)	4373 ± 110*	32.1 ± 0.8*	110.3 ± 4.8*	25.7 ± 1.6*	2.7 ± 0.2*
STZ-diabetes + MIGU-4 (25.0 mg/kg) (n = 8)	2502 ± 99 *#	41.3 ± 1.7*#	52.0 ± 3.5*#	20.0 ± 1.4#	3.2 ± 0.2*#
STZ-diabetes + α-LA (50.0 mg/kg) (n = 8)	2985 ± 121*#	36.4 ± 1.2*	87.3 ± 5.1*#	22.0 ± 1.8*#	2.9 ± 0.1*
STZ-diabetes + MIGU-4 (25.0 mg/kg)+ α-LA (50.0 mg/kg) (n = 8)	1530 ± 68 *#&	44.1 ± 1.3*#	52.3 ± 3.6*#	19.2 ± 1.6#	3.5 ± 0.1#

Notes: * – P < 0.05 vs 1; # – P < 0.05 vs 2, and & – P < 0.05 vs MIGU-4 and α-LA (ANOVA followed with Bonferroni correction).

Under the condition of simultaneous administration of MIGU-4 (25.0 mg/kg) and α-LA (50.0 mg/kg), the glucose content decreased by 31.7% (P < 0.05) and was simultaneously higher than in control by 40/1% (P < 0.05). It should be noted that glucose levels also decreased significantly compared to rats treated with MIGU-4 (by 38.8%) and α-LA (by 48.7%, P < 0.05). The insulin content remained lower than the control by 12.2% (P < 0.05) and exceeded the corresponding value in diabetic rats by 27.1% (P < 0.05). There was also a decrease in water and food consumption by 52.6% and 25.3%, respectively (P < 0.05, Table 1).

In the control group of rats, the level of proteins in the blood serum as the baseline for comparison, was 81.13 ± 2.72 g/L (Fig. 2). In rats with STZ diabetes, the level of proteins was significantly reduced by 16.9% (P < 0.05).

In the control group of rats, the level of albumin was 46.32 ± 1.93 g/L (Fig. 3). In rats with STZ diabetes, the level of albumin fraction was significantly reduced by 25.5% (P < 0.05). The impact of the treatments is evident as in STZ rats treated with MIGU-4 (25.0 mg/kg), albumin content exceeded that in the STZ rats by 17.7% (P < 0.05). The combined administration of MIGU-4 (25.0 mg/kg) and α-LA (50.0 mg/kg) caused a significant increase of albumin by 19.5% when compared with STZ-treated rats (Fig. 3).

In the control group of rats, the level of bilirubin was 18.91 ± 1.23 mg/L (Fig. 4). By comparison, bilirubin was dramatically raised by 84.7% in STZ-treated rats (P < 0.05). The treatment with MIGU-4 (25.0 mg/kg) was followed by a decrease in bilirubin content pertaining to the diabetes rats by 24.4% (P < 0.05). At the same time, bilirubin content remained higher (by 32.3%) than in control (P < 0.05, Fig. 4). Similarly, α-LA-treated rats (50.0 mg/kg) demonstrated a reduction of bilirubin by 33.0% (P < 0.05) in comparison with STZ-treated animals. Still, it was higher than in the control by 19.2% (P < 0.05, Fig. 4). The combined administration of MIGU-4 (25.0 mg/kg) and α-LA (50.0 mg/kg) caused a significant decrease of bilirubin by 41.5% (P < 0.05) when compared with STZ-treated rats (Fig. 4).

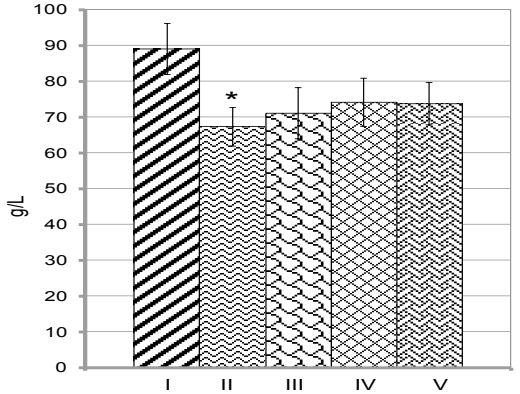

Fig. 2. Protein level in serum of rats treated with MIGU-4 and αLA: abscissa: I – control (rats with 0.9% solution of NaCl, i.p.); II – STZ-treated rats; III – MIGU-4 (25.0 mg/kg); IV – α-LA (50.0 mg/kg); V – MIGU-4 (25.0 mg/kg) + IV – α-LA (50.0 mg/kg); ordinate: units of substance content; * – P < 0.05 vs control (one-way ANOVA with Bonferroni correction)

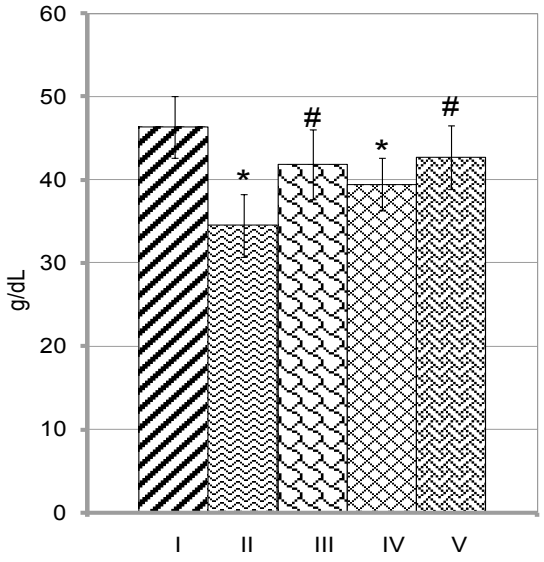

Fig. 3. Albumin level in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

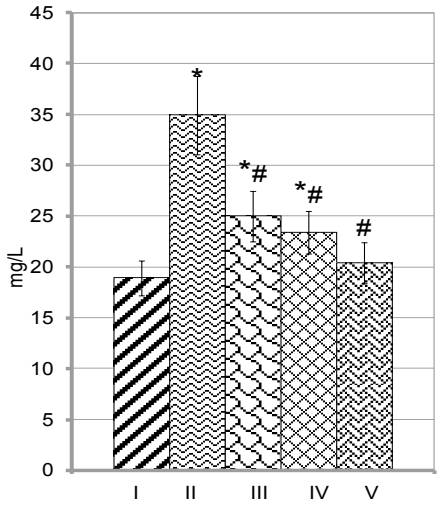

Fig. 4. Bilirubin level in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

The cholesterol content in the serum of intact rats was 621.3±28.2 mg/L (Fig. 5). The cholesterol level in STZ-diabetes increased significantly by 33.6%. In STZ rats treated with α-LA (50.0 mg/kg), cholesterol levels decreased when compared with the STZ-diabetes by 19.4% ($P < 0.05$) but exceeded control by 17.5% ($P < 0.05$) (Fig. 5). Combined administration of MIGU-4 and α-LA caused the reduction of cholesterol by 47.0% ($P < 0.05$) in comparison with STZ-diabetes (Fig. 5).

The triglyceride content in the serum of intact rats was 354.0±17.8 mg/L (Fig. 6). The level of triglycerides in STZ-diabetes increased significantly by 55.4% ($P < 0.05$). α-LA (50.0 mg/kg) treatment caused triglycerides significant reduction by 41.4% ($P < 0.05$) but it continued to be higher than in control – by 23.9% ($P < 0.05$) (Fig. 6). Combined administration of MIGU-4 and α-LA caused the reduction of triglycerides by 37.6% ($P < 0.05$) in comparison with STZ-diabetes (Fig. 6).

The LDL content in the serum of intact rats was 987.3±62.4 mg/L (Fig. 7). The level of LDL in STZ-diabetes increased significantly by 70.3% ($P < 0.05$). Treatment with MIGU-4 (25.0 mg/kg) caused a significant decrease in LDL by 57.4% ($P < 0.05$) when compared with STZ-diabetes (Fig. 7). At the same time, LDL level exceeded control by 57.3% ($P < 0.05$). α-LA (50.0 mg/kg) treatment reduced LDL content significantly by 39.6% ($P < 0.05$), but both continued to be higher than in control –by 50.8% ($P < 0.05$) (Fig. 7). Combined administration of MIGU-4 and α-LA caused the reduction of LDL by 60.2% ($P < 0.05$) ($P < 0.05$) in comparison with STZ-diabetes (Fig. 7).

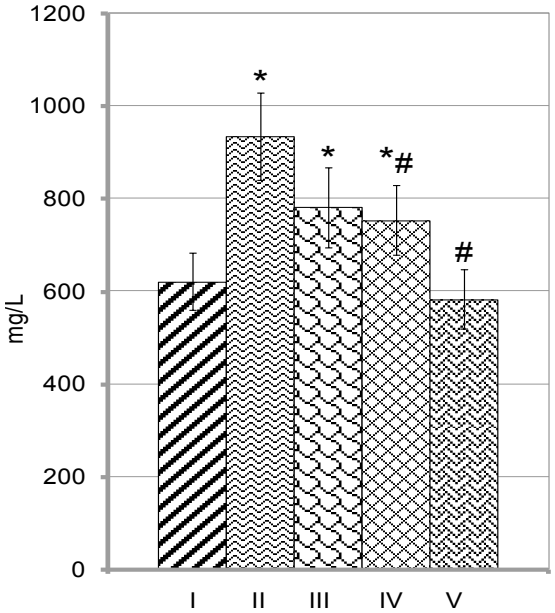

Fig. 5. Cholesterol level in STZ-diabetes rats under condition of treatment with MIGU-4 and α-LA: the same as in Figure 2

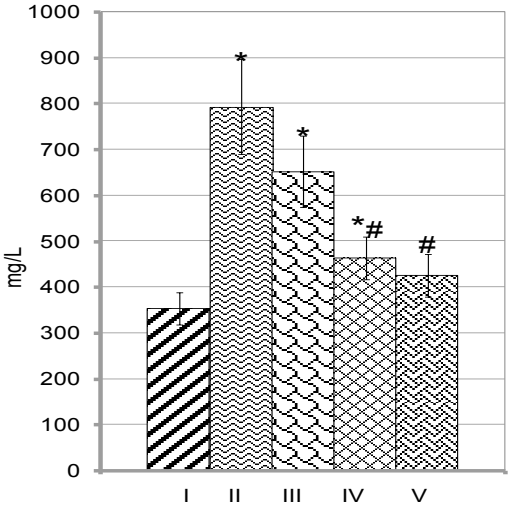

Fig. 6. Triglycerides level in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

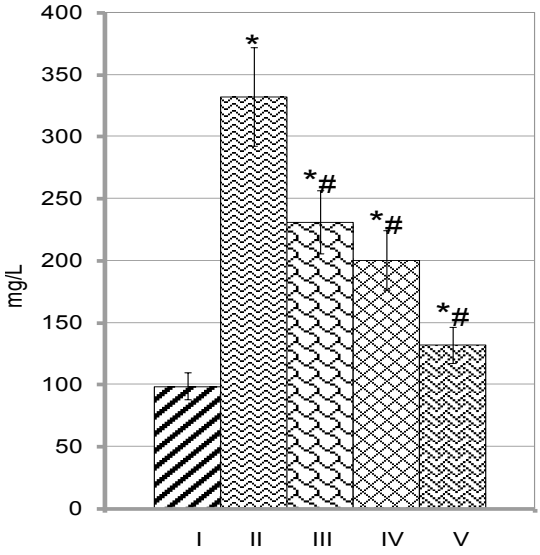

Fig. 7. Low-density lipoproteins level in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

The HDL content in the serum of intact rats was 389.2 ± 32.4 mg/L (Fig. 8). HDL content dropped by 45.6% when compared with the control rats ($P < 0.05$). Treatment with MIGU-4 (25.0 mg/kg) caused a significant HDL increase by 24.6% ($P < 0.05$) when compared with STZ-diabetes (Fig. 8). At the same time, HDL was lower than in control by 28.5% ($P < 0.05$). α-LA (50.0 mg/kg) treatment caused an increase in HDL level, which was higher than in STZ-diabetes by 41.8% ($P < 0.05$). Combined administration of MIGU-4 and α-LA caused the HDL to rise by 37.1% ($P < 0.05$) in comparison with STZ-diabetes (Fig. 8).

Comparison of the effectiveness of combined and separate usage of MIGU-4 and LA- revealed that only the LDL level exceeded that of the control by 25.4% ($P < 0.05$), while the remaining indices did not differ from the control under conditions of combined administration. Besides, the cholesterol level in rats with combined administration was reduced by 25.4% compared to MIGU-4 treatment and by 22.6% compared with α-LA ($P < 0.05$). Triglyceride content was 35.5% less than in MIGU-4 treated group ($P < 0.05$) under combined treatment. LDL level was reduced by 42.9% and by 34.1% compared to MIGU-4 and α-LA ($P < 0.05$).

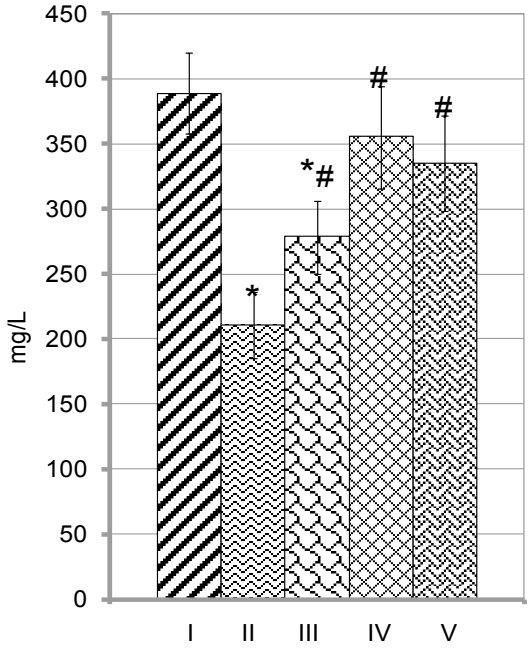

Fig. 8. High-density lipoproteins level in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

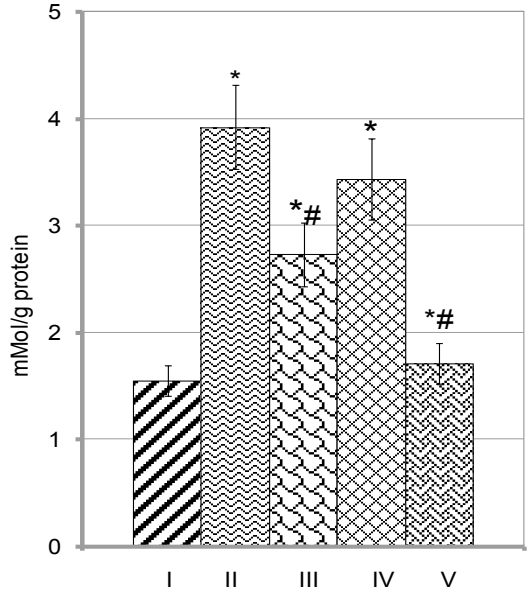

Fig. 9. Malondialdehyde level in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

In the control group of rats, the oxidative stress marker MDA was 3.92 ± 0.23 mMol/g protein (Fig. 9, I). In STZ-diabetes, the MDA level exceeded control by 60.5% ($P < 0.05$, Fig. 9, II). MIGU-4 (25.0 mg/kg) treated diabetic rats demonstrated a decrease of MDA by 30.4% ($P < 0.05$) when compared with STZ diabetes, while they demonstrated an increase by 43.2% ($P < 0.05$) in comparison with the control (Fig. 9, III). Combined administration of MIGU-4 and α-LA caused the reduction of MDA by 56.3% ($P < 0.05$) in comparison with the STZ-diabetes (Fig. 9, V). At the same time, the level of MDA was less in comparison with that of rats treated separately with MUGU-4 – by 37.2% ($P < 0.05$) and in rats treated with α-LA by 50.1% ($P < 0.05$, Fig. 9, V).

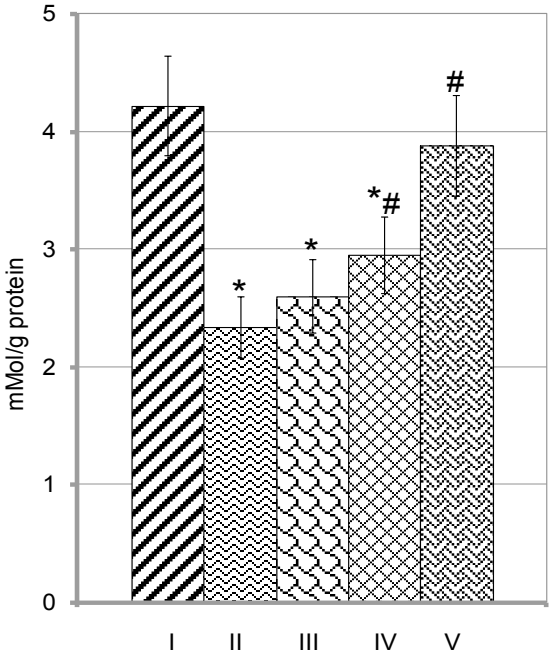

Fig. 10. Glutathione reduced content in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

In the control group of rats, the content of GSH was 4.22 ± 0.15 mMol/g protein (Fig. 10, I). In the STZ-diabetes group, GSH decreesed by 44.4% ($P < 0.05$, Fig. 10, II). α-LA (50.0 mg/g) treatment resulted in an increase of GSH compared to the STZ-diabetes by 20.7% ($P < 0.05$), but it still was less than in control by 30.1% ($P < 0.05$, Fig. 10, IV). Combined administration of MIGU-4 and α-LA caused an increase of GSH content by 39.7% ($P < 0.05$) in comparison with the STZ-diabetes group (Fig. 10, V). The level of GSH after combined administration of MIGU-4 and α-LA was greater in comparison with that in rats treated with MUGU-4 by 33.0% ($P < 0.05$), and in rats treated with α-LA by 24.0% correspondently ($P < 0.05$).

The enzymatic activity of SOD in the control was 19.44 ± 1.06 U/mg protein (Fig. 11, I). SOD activity was 57.0% less than in the STZ-diabetes group ($P < 0.05$, Fig. 11, II). MIGU-4 (25.0 mg/kg) treatment caused an increase of SOD by 41.7% ($P < 0.05$) when compared with the STZ-diabetes group (Fig. 11, III). The enzyme activity was reduced by 26.2% ($P < 0.05$) compared to the control. The treatment with α-LA (50.0 mg/kg) did not significantly affect the investigated enzyme activity (Fig. 11, IV). In contrast, combined treatment with MIGU-4 and α-LA induced the increase of SOD activity by 52.4% ($P < 0.05$) compared with the STZ-diabetes group (Fig.11, V).

Enzymatic activity of CT in the control was 65.22 ± 2.65 H_2O_2/min/mg protein, nMol (Fig. 12, I). In STZ-diabetes, CT activity was by 50.8% less than in control ($P < 0.05$, Fig. 12, II). MIGU-4 (25.0 mg/kg) treatment caused the increase of CT activity by 30.3% ($P < 0.05$) when compared with the STZ-diabetes group (Fig. 12, III). At the same tine,the CT activity was reduced by 29.7% ($P < 0.05$) compared to the control (Fig. 12, III). The CT activity was not significantly impacted by α-LA (50.0 mg/kg), while the combined treatment with MIGU-4 and α-LA induced the increase of CT activity by 47.0% ($P < 0.05$) compared with the STZ-diabetes group (Fig. 12, IV, V).

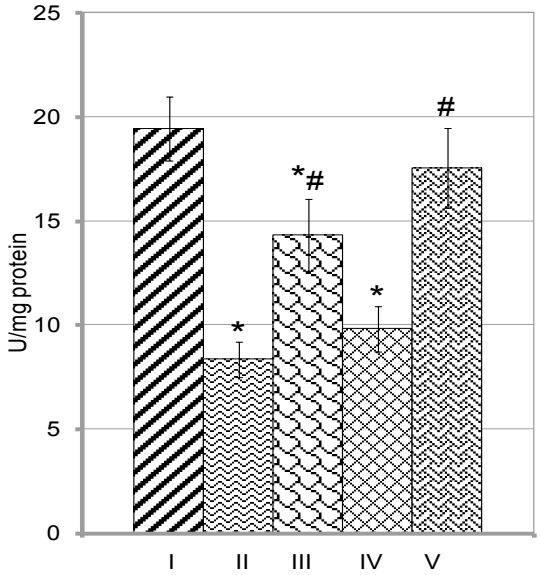

Fig. 11. Superoxide dismutase activity in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

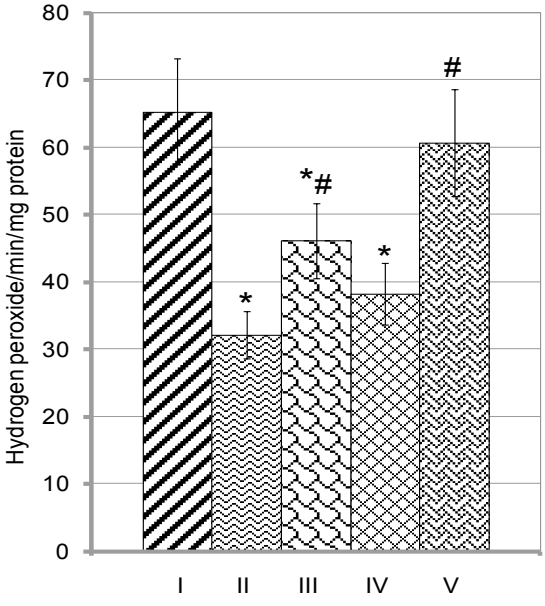

Fig. 12. Catalase activity in serum of rats treated with MIGU-4 and αLA: the same as in Figure 2

It should be noted that combined administration of MIGU-4 and α-LA caused a more significant elevation of SOD activity in comparison with the effects of α-LA (by 44.0%, $P < 0.05$). Besides, the CT activity exceeded those observed after MIGU-4 (by 24.0%) and α-LA (by 26.9%) administration ($P < 0.05$).

Discussion

Thus, the results showed that the development of experimental diabetes caused by STZ is accompanied by increased food and water intake with a simultaneous decrease in daily body weight gain in rats against the background of a reduction in insulin content and hyperglycemia. In addition, a decrease in blood protein levels, albumin content, and lipid metabolism disorders with a characteristic increase in cholesterol, triglycerides, and LDL were detected against a decrease in HDL content. Also, an increase in MDA content was recorded in liver tissue against a reduction of the activity of antioxidant enzymes – SOD, CT, and a decrease in reduced glutathione (Nithiya & Udayakumar, 2018; Al-Muzafar et al., 2021). It is important to note the contribution of oxidative stress to neuroinflammation during diabetes development (Forrester et al., 2020; Sohail et al., 2022).

These disorders are characteristic of experimental models of diabetes mellitus (Kottaisamy et al., 2021; Kresyn & Godlevskii, 2014). In particular, diabetes induced by a high-fat diet in rats is accompanied by severe dyslipidemia with hypertriglyceridemia, a decrease in high-density lipoprotein content, and an increase in high-density lipoprotein content (Al-Muzafar et al., 2021; Kottaisamy et al., 2021). Such changes are associated with morphological signs of hepatosteatosis, namely, the formation of lipid deposits in the liver tissue and an increase in serum bilirubin and glucuronic acid levels. The results obtained in our study indicate that lipid metabolism disorders are inherent in STZ-induced diabetes (Martín-Carro et al., 2023).

MIGU-4 at a 25.0 mg/kg dose caused a moderate hypoglycemic effect, increased blood insulin levels, reduced water and food consumption in rats with STZ-induced diabetes, and increased daily body weight gain. At the same time, these indicators remained significantly different from those in the control group. The directionality of the effects of MIGU-4 was similar to that of α-LA (50.0 mg/kg); hypoglycemia was more affected with MIGU-4, and the daily body weight gain in rats with STZ-induced diabetes was more pronounced.

The administration of MIGU-4 was accompanied by positive dynamics of proteins and lipids in the blood of rats with STZ-induced diabetes. The pronouncement of the effects of MIGU-4 (25.0 mg/kg) on the total protein and albumin content was similar to the impact of α-LA (50.0 mg/kg), but the effectiveness of MIGU-4-induced corrective effect on the studied parameters was somewhat higher, given the recovery of serum albumin levels. The combined use of drugs provided a more pronounced effect, particularly on lipid metabolism. Thus, the combined use of MIGU-4 and α-LA caused a more pronounced reduction in cholesterol and LDL and a significant decrease in triglyceride levels.

The obtained results also showed that the liver parenchyma of rats with STZ-induced diabetes showed signs of severe oxidative stress, which consisted of an increase in MDA content, a decrease in SOD and CAT activity, as well as the content of reduced glutathione. MIGU-4 ensured the reverse dynamics of these indicators, the intensity of which was higher than that in the group treated with α-LA, due to a significantly higher reduction in MDA content. The possible role of α-tocopherol in the blood raised by germanium organic compounds might be in charge of the antioxidant action (Nakamura et al., 2014). However, the antioxidant properties of organic germanium are realized in multiple ways, including direct scavenging of free radicals and activation of antioxidative enzymes (Dobrzyński et al., 2018; Baidya et al., 2021; Azumi et al., 2022).

A possible factor in the corrective effects of MIGU-4 may be niacin, which, when administered at a dose of 1–2 grams per day, causes a decrease in total cholesterol, apolipoprotein A, triglycerides and LDL, and an increase in HDL in the blood serum, which significantly reduces the risk of cardiovascular complications in patients with diabetes mellitus (Gordon et al., 2020). The use of niacin in patients with impaired statin tolerance provides positive dynamics of lipid metabolism, induces lipolysis (Wang et al., 2020), increases the activity of antioxidant enzymes – SOD, glutathione peroxidase in experimental diabetes (Tupe et al., 2011; Dou et al., 2013). It should also be noted that niacin causes activation of peroxisome proliferator-activated receptor-γ (PPAR-γ) (Knowles et al., 2006), which leads to the control of lipid metabolism, adipogenesis, and increased sensitivity to insulin (Tontonoz & Spiegelman, 2008; Ahmadian et al., 2013). This property is significant given the widespread use of PPAR-γ agonists for treating patients with diabetes mellitus.

In addition, an important mechanism of action of α-LA as a potent antioxidant (Viana et al., 2022) causes antiinflammatory not-specific effects (Skibska et al., 2023). Thus, it appears that LA or its reduced form, dihydrolipoic acid (DHLA), possesses many biochemical functions acting as biological antioxidants, able to regenerate endogenous antioxidants such as vitamins C and E, and modulator of the signaling transduction of several pathways. α-LA also is an insulin-mimetic and possesses an anti-inflammatory activity (Rochette et al., 2014). Besides, α-LA restores glutathione via activating Nrf2/ARE signaling pathway (Zhang et al., 2017). Such an effect explains the observed significant raising of GHS in STZ-rats serum treated with α-LA.

According to the results obtained, it is possible to assume that the antioxidant effects of MIGU-4 and α-LA provide further complex changes

in liver parenchyma metabolism. These changes manifest in the restoration of both general indicators of the diabetic state and metabolic activity of the liver parenchyma, which resulted in the restoration of lipid metabolism and a positive effect on blood glucose and protein levels.

Conclusions

The development of experimental diabetes mellitus caused by the use of STZ is accompanied by disorders of the liver's functional state, which indicate damage to hepatocytes, bilirubinemia, serum hypoprotein content, and hypoalbuminemia, increased cholesterol, triglycerides, and low-density lipoprotein, with a simultaneous decrease in high-density lipoprotein against the background of increased oxidative stress in the liver parenchyma.

Course administration of MIGU-4 causes a moderate hypoglycemic effect and a mild increase in insulin levels. The success of the corrective effect of MIGU-4 (25.0 mg/kg, i.p.) corresponded to that of α-LA (50.0 mg/kg, i.p.), except that the effectiveness of MIGU-4 was higher in terms of insulin, albumin, daily body weight gain, increased SOD activity, and CT in liver tissue. At the same time, the success of the corrective effect of α-LA was more pronounced in the correction of triglycerides, high-density lipoprotein in the blood serum, and the level of reduced glutathione in the liver tissue.

The combined use of MIGU-4 and α-LA enhances the corrective effect on the studied markers of STZ-induced diabetes, which is promising for use in patients with diabetes mellitus, prevention of diabetes complications, and lipodystrophic disorders of hepatocytes with simultaneous restoration of their function.

The author declares that there is no conflict of interest.

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