ЕКСПЕРИМЕНТАЛЬНА МЕДИЦИНА

UDC 616.378-008.64:616.092.9

Awad Ali Riyadh

THE INFLUENCE OF ALKILSELENONAFTIRIDIN ON LEVELS IN BLOOD OF LIPOPROTEINS HIGH, LOW AND VERY LOW DENSITY ON A BACKGROUND OF EXPERIMENTAL DIABETES

Lugansk National University Taras Shevchenko, Starobelsk, Ukraine

the_kingdom@mail.ua

Correction of disturbances in lipid metabolism by diabetes remains a problem today. The aim of this work was to study the impact alkilselenonaftiridin (ASNR) on the dynamics of changes in the blood serum of levels lipoproteins of high (HDL), low (LDL) and very low (VLDL) density on the background of experimental streptozotocin-induced diabetes (DM). ASNR (180 mg / 100 g) were administered daily from the first day and 21^{ht} day experiment in two different groups. It is shown that diabetes leads to changes in the lipid composition of blood – reducing the level of HDL and, conversely, increased LDL and VLDL levels. Introduction ASNR positively influenced the change of levels these of lipoproteins. Especially in the case when ASNR administered on the first day of the experiment. In particular, the introduction ASNR the first day of the experiment prevents significant reduction of HDL levels, such as 8.3 and 9.9% on the 20th and 40th day of the experiment, respectively. ASNR prevents increasing of LDL levels on 13.3 and 10.4% on the 20th and 40th days experiment, respectively. Simultaneously, ASNR reduces the negative impact of DM on VLDL levels on 7.4, 11.1 and 21.4 % on the 20th, 40th and 60th day experiment in accordance. However, the introduction of ASNR with 21st day of the experiment has little effect on changes of levels lipoproteins, caused by the development of DM. Except reduction on 13.3 % the negative impact of diabetes on levels of VLDL on the 60th day of the experiment. Thus, the introduction of ASNR positively influenced the change of levels lipoproteins of high, low and very low density on the background of experimental streptozotocin-induced diabetes.

Keywords: experimental diabetes; lipoproteins; alkilselenonaftiridin.

This article is part of the research work of the department of anatomy, physiology of human and animal in «The University of Lugansk» «Mechanisms of adaptation to environmental factors» under the state registration number 0198U002641.

Introduction. Diabetes mellitus (DM) is one of the most pressing biomedical problems, which is a priority the direction of national health systems [1]. In 2010 the total number of patients with all forms of diabetes in the world was about 239 million people. According to forecasts of the International Diabetes Federation (IDF), the number of patients with diabetes in the adult population (20–79 years) by 2030 will increase to 439 million [2]. The highest percentage of patients with diabetes mellitus belongs to the second type (80–95%) [3, 4]. Among the European population prevalence of diabetes is 7.8%. Most of them suffer from diabetes in Germany (10.2% of the population) and in Belgium (10% population), at least in the United Kingdom (4.2% of the population), significantly at least – in the Western Pacific Region [5, 6]. In Ukraine, according to Ministry of Health, about 1.5 million peoples are suffering with diabetes. It is estimated that among the inhabitants of different countries over 65 years, every 20th person suffers from diabetes, and this figure shows only those who know about the disease, and is registered in endocrinologist [7]. Diabetes accompanied by dangerous development of acute and chronic related disorders that lead to early disabilities and reducing life expectancy. Diabetes is a high risk for development blindness, renal failure, diabetic cardiomyopathy and encephalopathy [8].

Concerning experimental studies, the **aim** was to studying diagnosis and treatment of diabetes, that is

important and timely. The purpose of this work was to study the impact alkilselenonaftiridin on the dynamics of changes in the blood serum of levels lipoproteins of high, low and very low density on the background of experimental streptozotocin-induced diabetes.

Methods. The study was carried out in the autumn-winter period on 92 male rats of Wister weighing 220–280 g, which were kept in a standard diet of the vivarium of the department of anatomy and physiology of animal of «Lugansk National University Taras Shevchenko» [9]. Selection of rats for experiments was associated with the peculiarities of the methodological approach to the solution of the objectives and tasks. The number of experimental animals was determined according to the methods of statistical analysis [10]. The content and care of rats was carried out on compliance with the principles of bioethics and the «European Convention for the Protection of Vertebrate Animals», which are used for experimental and other scientific purposes (Strasbourg, 1985), as well as the decisions of the «First National Congress on Bioethics» (Kiev, 2001) [11].

The control group consisted of 23 rats. In the 69 research groups of animals was modeled experimental streptozotocin-induced diabetes. All animals of the experimental group were divided into three subgroups (23 rats) each one. The animals of the first sub-group (1-EG) simulated of streptozotocin diabetes (DM) without introducing Alkilselenonaftiridin – ASNR (number 7498352, «Brillstein Handbook»). The animals of second sub-group (2-EG) ASNR started to enter 21st days from the beginning of the experiment and the animals of the third experimental subgroups (3-EG) ASNR started to enter the first day of the experiment. Daily dose ASNR (180 mg /100 g) was calculated in accordance with M.A. Ansari et al. (2004) and N. Stanishovski (2008) [12, 13].

The animals were injected intraperitoneally research group streptozotocin («Sigma-Aldrich», USA) at a dose of 50 mg/kg body weight in 0.1mol citrate buffer (pH=4.5) once. To confirm the playback diabetes in rats under administration streptozotocin photometrically determined glucose in the blood serum using *glucoseoxydase* method («Agat-Med», Ukraine) and whole blood using a glucometer «Glucofort» (Ukraine) and glucose in urine – by using diagnostic strips «Pentafan» («Lachema», Czech Republic).

Results and Discussion. The level of high density lipoproteins (HDL) in the blood serum of animals control group before the experiment was 0.90± 0.11 mmol/l. By the 20^{th} day of the experiment HDL was 0.99±0.19 mmol/l, after a 40-day experiment 0.95±0.18 mmol/l and after 60-days 1.0±0.22 mmol/l.

Baseline level of HDL in animals with experimental diabetes (1-EG) was 1,06±0,07 times higher than the control (0.95±0.17 mmol/l). After a 20^{th} day experiment HDL level decreased in 1.29±0.03 times than exposure control and amounted to 0.77±0.14 mmol/l. On 40^{th} and 60^{th} day experiment HDL levels increased with relative 20^{th} day rate to 0.82±0.15 mmol/l (P<0.05) and 0.92±0.19 mmol/l (P<0.01) in accordance. In comparison with the exposure control revealed a decrease in HDL levels in 1.16±0.02 and 1.08±0.02 times accordingly. Thus in animals with experimental diabetes levels of HDL in the blood serum versus control animals were significantly reduced (**Fig. 1**).

In comparison with control animals 2-EG baseline in HDL was above 1.01±0.08 times (0.91±0.17 mmol/l). Average level of HDL on a 20^{th} day experiment was reduced in 1.30±0.06 times, to 0.76±0.16 mmol/l (P <0.05). On a 40^{th} day exposure experiment HDL levels dropped relative to the exposure control in 1.13±0,02 times. In comparison to the 20^{th} day exposure experiment, the average of the level of HDL increased to 0.84±0.15 mmol /l (P<0.05). After a 60^{th} day experiment the level of HDL increased to 0.97± 0.20 mmol/l (1.02±0.03 times lower than the exposure control), P<0.05 (**Fig. 1**).

Animals with 3-EG before experiment had level of HDL 0.97±0.18 mmol/l which is 1.08±0.09 fold over control was. After a 20^{th} day experiment HDL level decreased to 0.84±0.18 mmol/l (in 1.18±0.06 times lower than the exposure control), P<0.05. On the 40th and 60^{th} day of the experiment revealed increasing of levels of HDL relative to 20th-day rate to 0.91±0.16 mmol/l (P<0.05) and 0.96±0.20 mmol/l (P<0.05), and were reduced versus the control in 1.04±0.01 and 1.01±0.25 times in accordance (**Fig. 1**). However, positive can be considered as mitigating negative impact of diabetes on the level of HDL. Namely, prevent a significant reduction in their levels on 8.3 and 9.9% on the 20th and 40th day experiment, respectively.

We can conclude that the level of HDL in animals of all experimental subgroups dropped to 20^{th} day of the experiment and then increased until the 60th day experiment, exposure, but remained below the exposure control. The changes were more in the animals of 1st and 2nd experimental groups. Thus, the introduction of animals of ASNR with first day of the experiment (group 3-EG) reduces the negative impact of DM on levels of HDL. Simultaneously, the introduction of ASNR with 21st day (group 2-EG) has little effect on changes in levels of HDL, caused by the development of DM.

The level of low-density lipoproteins (LDL) in the animals of the control group before the experiment was 0.24±0.05 mmol/l. On the 20th day of the experiment the level of LDL was within 0.23±0.05 mmol/l. On 40th day experiment was 0.26±0.06 mmol/l and on 60th day was 0.22±0.07 mmol/l.

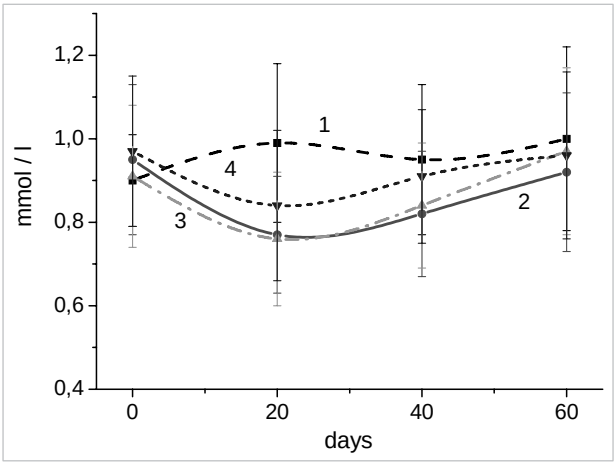

Figure 1. The level of high density lipoprotein in the blood serum of animals in the control (1) and the experimental group 1-EG (2), 2-EG (3) and 3-EG (4).

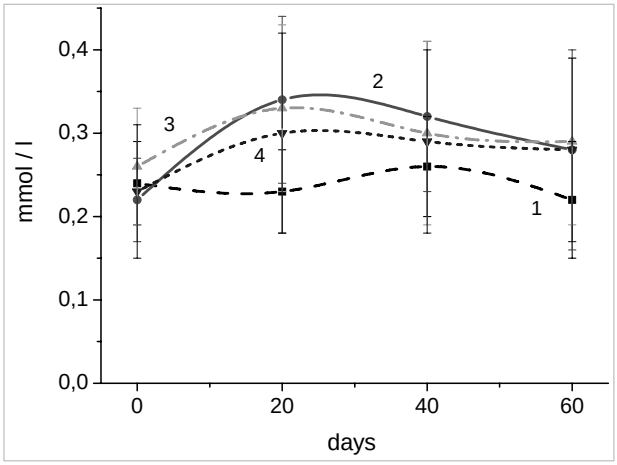

Figure 2. The level of low density lipoprotein in the blood serum of animals in the control (1) and the experimental group 1-EG (2), 2-EG (3) and 3-EG (4).

Baseline LDL animals with DM (group 1-EG) was (0.22±0.05 mmol/l) on 1.09±0.13 times lower than the control. After a 20th day experiment the level of LDL increased in 1.42±0.20 times than the exposure control and amounted to 0.34±0.12 mmol/l (P<0.05). On 40th and 60th day experiment the level of LDL dropped relative to 20th day rate to 0.32±0.09 mmol/l (P<0.05) and 0.28±0.12 mmol/l (P<0.01). In comparison with the exposure control found an increase in the level of LDL in 1.23±0.13 and 1,27±0.13 times in accordance (**Fig. 2**). Thus in animals with experimental diabetes levels of LDL in the blood serum versus control animals were increased.

In comparison with the control animals in 2-EG baseline LDL was lower in 1.08±0.19 times (0.26± 0.07 mmol/l). On a 20th day experiment the average of level of LDL was increased to 0.33±0.13 mmol/l (in 1.43±0.227 fold) when P<0.05. On 40th and 60th day experiment exposure LDL level was higher than the control in 1.15±0.16 and 1.32±0.35 times. But the average of level of LDL was less than in 20th day – 0.30± 0.11 mmol/l (at P<0.05) and 0.29±0.10 mmol/l (P<0.05) in accordance (**Fig. 2**).

In animals 3-EG LDL level before the experiment was 0.23±0.08 mmol/l, which in 1.04±0.231 times was lower than the control. On a 20th day experiment LDL level increased to 0.30±0.12 mmol/l (in 1.30±0.23 times the exposure control) when P<0.05. On the 40th and 60th day of the experiment revealed a decrease in LDL level relative to 20th day rate to 0.29±0.11 mmol/l (P<0.05) and 0.28±0.11 mmol/l (P< 0.05) in accordance, and remained above the exposure control in 1.12±0.17 and 1.27±0.37 times in accordance (**Fig. 2**).

Thus, the introduction of ASNR with first day of the experiment (3-EG) reduces the negative impact of DM on levels of LDL. Namely, prevent increase their level on 13.3 and 10.4% on the 20th and 40th day experiment, respectively. Simultaneously, the introduction of ASNR with 21st day (3-EG) has little effect on changes in levels of LDL, caused by the development of DM (**Fig. 2**).

The level of very low density lipoproteins (VLDL) the animals of the control group before the experiment the VLDL level in serum was 0.34±0.10 mmol/l. By the 20th day of the experiment the level of VLDL was within 0.36±0.09 mmol/l, on a 40th day experiment was 0.34±0.06 mmol/l and on 60th day was 0.32±0.12 mmol/l.

Baseline level of VLDL animals with experimental diabetes (1-EG) was in 1.03±0.10 time increase over control (0.35±0.10 mmol/l). On a 20th day experiment the VLDL level increased in 1.61±0.12 times than the exposure control and amounted to 0.58±0.15 mmol/l (P<0.05). On 40th and 60th day experiment the VLDL level dropped relative to 20th day rate to 0.50± 0.12 mmol/l (P<0.05) and 0.51±0.21 mmol/l (P<0.05) in accordance. In comparison with the exposure control found an increase in the level of VLDL in 1.47± 0.16 and 1.59±0.44 times accordingly (**Fig. 3**). Thus in animals with experimental diabetes levels of VLDL in the blood serum versus control animals were significantly increased.

In comparison with the control animals in 2-EG animals, the VLDL baseline level was below in 1.06± 0.09 times (0.32±0.11 mmol/l). After a 20th day experiment the average of the level of VLDL in 2-EG animals increased in 1.84±0.01 times to 0.59±0.14 mmol/l (P<0.05), and in 1.64±0.02 (P<0.05) and 1.02±0.01 times compared to the control and diabetic animals respectively. On 40th and 60th days of exposure experiment the VLDL level dropped relative to 20th day and was 0.48±0.09 mmol/l (P<0.05) and 0.45± 0.19 mmol/l (P<0.05) in accordance (**Fig. 3**).

Baseline level of VLDL before the experiment in 3-EG animals (0.35±0.11 mmol/l) was in 1.031± 0.099 times increase in comparison with control. On a 20th day experiment VLDL levels increased to 0.54±

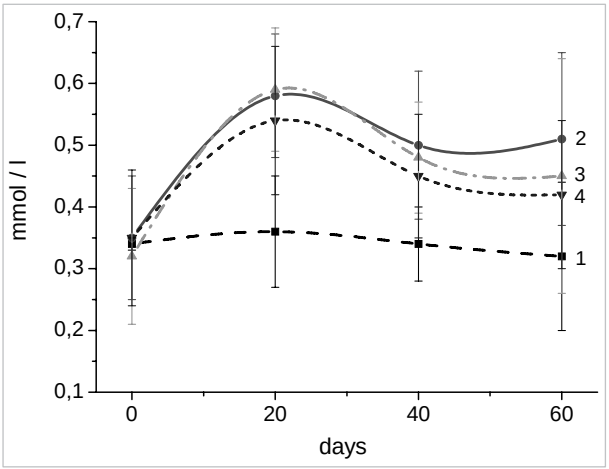

Figure 3. The level of very low density lipoproteins in the blood serum of animals in the control (1) and the experimental group 1-EG (2), 2-EG (3) and 3-EG (4).

0.12 mmol/l (in 1.478±0.087 times relatively control, P<0.05), however, less than in animals with diabetes (in 1.07±0.01 times). On the 40th and 60th days of the experiment revealed lowering VLDL with respect to 20th day rate to 0,45±0.10 mmol/l (P<0.05) and 0.42± 0.19 mmol/l (P<0.05). VLDL level remained above in comparison with control in 1.32±0.08 and 1.31±0.25 times, respectively. Simultaneously, VLDL levels on the 40th and 60th days of the experiment in this group were significantly lower VLDL levels in animals with diabetes, namely in 1.32±0.08 and 1.31±0.25 times in accordance (**Fig. 3**).

Thus, the introduction ASNR with 21st day of the experiment (2-EG) reduces (on 13.3 %) the negative impact of DM on levels of VLDL only on the 60th day of the experiment. Simultaneously, the administration of ASNR with first day of the experiment (group 3-EG) reduces the negative impact of DM on VLDL levels on 7.4, 11.1 and 21.4 % on the 20th, 40th and 60th day experiment, respectively.

Conclusions and prospects of further researches. Thus, it is shown that diabetes leads to changes in the lipid composition of blood – reducing the level of HDL and, conversely, increased LDL and VLDL levels [14, 15]. Introduction ASNR positively influenced the change of levels these of lipoproteins. Especially in the case when ASNR administered on the first day of the experiment (3-EG). In particular, the introduction ASNR the first day of the experiment prevents significant reduction of HDL levels, such as 8.3 and 9.9% on the 20th and 40th day of the experiment, respectively. ASNR prevents increasing of LDL levels on 13.3 and 10.4% on the 20th and 40th days experiment, respectively. Simultaneously, ASNR reduces the negative impact of DM on VLDL levels on 7.4, 11.1 and 21.4 % on the 20th, 40th and 60th day experiment in accordance. However, the introduction of ASNR with 21st day of the experiment (2-EG) has little effect on changes of levels lipoproteins, caused by the development of DM. Except reduction on 13.3 % the negative impact of diabetes on levels of VLDL on the 60th day of the experiment. Thus, the introduction of ASNR positively influenced the change in the level of HDL, LDL and VLDL on the background of DM.

References

1. Balabolkin MI. Diabetology. Moscow, Medicine; 2000. 672 p.
2. Shaw JE, Shaw RA, Sicree P, Zimmet Z. Global estimates of the prevalence of diabetes for 2010 and 2030. Diabetes Res Clin Pract. 2010;87:4–14.
3. International Diabetes Federation. Diabetes Atlas Second Edition. 2003;17–71.
4. Gregg EW, Cadwell BL, Cheng YJ, Cowie CC, Williams DE, Geiss L, Engelgau MM, Vinicor F. Trends in the prevalence and ratio of diagnosed to undiagnosed diabetes according to obesity levels in the U.S. Diabetes Care. 2004; 27(12): 2806–12.
5. Sekikawa A, Eguchi H, Tominaga M, Igarashi K, Abe T, Manaka H, Sasaki H, Fukuyama H, Kato T, Kiyohara Y, Fujishima M. Prevalence of type 2 diabetes mellitus and impaired glucose tollerantce in a rural area of Japan. The Funagata diabetes study. J Diabetes Complications. 2000 Mar–Apr;14(2):78–83.
6. Dunstan DW, Zimmet PZ, Welborn TA, De Courten MP, Cameron AJ, Sicree RA, Dwyer T, Colagiuri S, Jolley D, Knuiman M, Atkins R, Shaw JE. The rising prevalence of diabetes and impaired glucose tolerance: the Australian Diabetes, Obesity and Lifestyle Study. Diabetes Care. 2002 May;25(5):829–34.
7. Mayakov AI, Beacons AI, Intercession TG, Pokrovsky MV. Endothelial dysfunction in the background fructose indutsirovannoy models of metabolic disorders in rats. XVII Russian National Congress «Man and medicine» 2010; 676–7.
8. Donnelly R, Emslie-Smith AM, Gardner ID, Morris AD. Vascular complications of diabetes. BMJ. 2000 Apr 15;320 (7241):1062–6.
9. Zapadnyuk IP, Zacharias AE, Vishcha wk. Laboratory animals. Breeding, maintenance, use in the experiment. 3rd. Kyiv: Vyscha shkola;1983; 243–76.
10. Polyakov LE. Statistical methods for research in medicine and health. Leningrad: Medicine; 1971. 200 p.
11. European convention for the protection of vertebral animals used for experimental and other scientific purpose: Council of Europe 18.03.1986. Strasbourg, 1986; European Treaty Series.123.

12. Ansari MA, Ahmad AS, Ahmad M. Selenium protects cerebral ischemia in rat brain mitochondria. Biol Trace Elem Res. 2004;101(1):73–86.
13. Stanishovski NV. Effect of ischemic appear on morph functional adaptation to cardiac myocardial necrosis at introduction Alkil Selenonaftiridin (experimental research). Kharkov, 2008.
14. Panfilova V, Taranushenko TE, Salmin AB.et al .High-density lipoprotein as an indicator of vascular dysfunction in children with type 1 diabetes. Saratov Journal of Medical Science. 2011;7(4):973–5.
15. Bozhko G, Pertseva T, Chursina V, Sokolik V. Features dislipoproteinemia in patients with encephalopathy distsirkulyatornoy diabetes mellitus type II Ukrainian Journal Neuropsychiatries. 2004;12(1):107–109.

УДК 616.378-008.64:616.092.9

ВПЛИВ АЛКІЛСЕЛЕНОНАФТІРІДІНА НА ВМІСТ В КРОВІ ЛІПОПРОТЕЇДІВ ВИСОКОЇ, НИЗЬКОЇ І ДУЖЕ НИЗЬКОЇ ЩІЛЬНОСТІ НА ТЛІ ЕКСПЕРИМЕНТАЛЬНОГО ЦУКРОВОГО ДІАБЕТУ

Авад А.Р.

Резюме. Корекція порушень у ліпідному обміні за цукрового діабету залишається актуальним завданням сьогодення. Метою даної роботи було вивчення впливу алкілселенонафтірідіна (АСНР) на динаміку змін в сироватці крові рівнів ліпопротеїдів високої (ЛПВЩ), низької (ЛПНЩ) та дуже низької (ЛПДНЩ) щільності на тлі експериментального стрептозотоцин-індукованого діабету (ЦД). АСНР (180 мг / 100 г) вводили щодня з першого і з 21-го дня експерименту в двох різних групах. Показано, що цукровий діабет призводить до змін ліпідного складу крові – знижується вміст ЛПВЩ і, навпаки, збільшуються рівні ліпідів низької і дуже низької щільності. Введення АСНР позитивно впливали на зміну вмісту цих ліпопротеїнів в крові на тлі ЦД. Зокрема, введення АСНР з першого дня експерименту запобігає значному зниженню рівнів ЛПВЩ на 8,3 і 9,9% на 20-й і 40-й день відповідно. Препарат також запобігає підвищенню рівня ЛПНЩ на 13,3 і 10,4% на 20-й і 40-й дні експерименту відповідно. Одночасно він знижує негативний вплив ЦД на рівні ЛПДНЩ. А саме, на 7,4, 11,1 і 21,4% на 20-й, 40-й і 60-й день експерименту відповідно. Проте, введення АСНР з 21-го дня експерименту не мало значного впливу на індуковані ЦД зміни в метаболізмі ліпопротеїнів. За винятком зниження на 13,3% негативного впливу діабету на рівні ЛПДНЩ на 60-й день експерименту. Таким чином, введення АСНР позитивно впливає на зміну рівнів ліпопротеїдів високої, низької і дуже низької щільності на тлі експериментального стрептозотоцин-індукованого діабету.

Ключові слова: експериментальний цукровий діабет; ліпопротеїни; алкілселенонафтірідін.

УДК 616.378-008.64:616.092.9

ВЛИЯНИЕ АЛКИЛСЕЛЕНОНАФТИРИДИНА НА СОДЕРЖАНИЕ В КРОВИ ЛИПОПРОТЕИДОВ ВЫСОКОЙ, НИЗКОЙ И ОЧЕНЬ НИЗКОЙ ПЛОТНОСТИ НА ФОНЕ ЭКСПЕРИМЕНТАЛЬНОГО САХАРНОГО ДИАБЕТА

Авад А.Р.

Резюме. Коррекция нарушений липидного обмена при сахарном диабете до сих пор остается важной задачей современности. Целью данной работы было изучение влияния алкилселенонафтиридина (АСНР) на динамику изменений в сыворотке крови уровней липопротеидов высокой (ЛПВП), низкой (ЛПНП) и очень низкой (ЛПОНП) плотности на фоне экспериментального стрептозотоцин-индуцированного диабета (СД). АСНР (180 мг / 100 г) вводили ежедневно с первого и с 21-го дня эксперимента в двух разных группах. Показано, что сахарный диабет приводит к изменениям липидного состава крови – снижается содержание ЛПВП и, наоборот, увеличиваются уровни липидов низкой и очень низкой плотности. Введение АСНР положительно влияли на содержание этих липопротеинов в крови на фоне СД. В частности, введение АСНР с первого дня эксперимента предотвращает значительное снижение уровней ЛПВП на 8,3 и 9,9 % на 20-й и 40-й день соответственно. Препарат также предотвращает повышение уровня ЛПНП на 13,3 и 10,4 % на 20-й и 40-й дни эксперимента соответственно. Одновременно он снижает негативное влияние СД на уровни ЛПОНП. А именно, на 7,4, 11,1 и 21,4 % на 20-й, 40-й и 60-й день эксперимента соответственно. К сожалению, введение АСНР с 21-го дня эксперимента мало влияет на изменение уровней липопротеинов, вызванных развитием СД. За исключением снижения на 13,3% негативного влияния диабета на уровни ЛПОНП на 60-й день эксперимента. Таким образом, введение АСНР положительно влияет на изменение уровней липопротеидов высокой, низкой и очень низкой плотности на фоне экспериментального стрептозотоцин-индуцированного диабета.

Ключевые слова: экспериментальный сахарный диабет; липопротеины; алкилселенонафтиридин.

Стаття надійшла 10.03.2017 р.
Рекомендована до друку на засіданні редакційної колегії після рецензування