UDC 616.379-008.64-092

N. V. Mukha*, A. V. Govorin, E. B. Perevalova, D. N. Zaytsev

Chita State Medical Academy, Department of Internal Medicine, intermediate course, Chita, Russia

FATTY ACID COMPOSITION OF SERUM LIPIDS IN PATIENTS WITH DECOMPENSATED TYPE 1 DIABETES MELLITUS DEPENDING ON THE DIABETIC KETOACIDOSIS SEVERITY

Abstract

Summary. In diabetic ketoacidosis, significant metabolic disorders develop in all organs and tissues, including the myocardium. The main energy substrate in the myocardium are fatty acids. **The objective** was to study the fractional composition of serum in patients with T1DM, complicated by diabetic ketoacidosis (DKA), depending on the severity of DKA. **Materials and methods.** Determination of the fatty acid spectrum of serum lipids was carried out in the following groups of patients: 68 patients with compensated type 1 diabetes mellitus (group 1); 54 patients with type 1 diabetes complicated by mild ketoacidosis (group 2); and 42 patients with diabetes mellitus complicated by moderate and severe ketoacidosis (group 3). Extraction of lipids from serum was performed according to the method developed by J. Folh et al. (1957), after which methylation of fatty acids was carried out according to the method proposed by K. M. Sinyak et al., with subsequent analysis using the Crystal-2000M gas chromatograph (Russia). **Results.** All patients with type 1 diabetes mellitus compared with the group of healthy individuals showed an increase in the total content of saturated fatty acids, a decrease in the total concentration of unsaturated fatty acids, as well as an increase in the ratio of saturated / unsaturated fatty acids. At the same time, a significant difference between the studied groups was revealed. The most pronounced changes were found in the group of patients with type 1 diabetes mellitus complicated by moderate to severe ketoacidosis. **Conclusions.** These changes develop as part of a general systemic metabolic disorder in a given cohort of patients.

Key words: *type 1 diabetes mellitus, diabetic ketoacidosis, fatty acids*

For citation: Mukha N. V., Govorin A. V., Perevalova E. B., Zaytsev D. N. FATTY ACID COMPOSITION OF SERUM LIPIDS IN PATIENTS WITH DECOMPENSATED TYPE 1 DIABETES MELLITUS DEPENDING ON THE DIABETIC KETOACIDOSIS SEVERITY. The Russian Archives of Internal Medicine. 2019; 9(3): 182-187. [In Russian]. DOI: 10.20514/2226-6704-2019-9-3-182-187

DOI: 10.20514/2226-6704-2019-9-3-182-187

DKA — diabetic ketoacidosis, FAs — fatty acids, DM — diabetes mellitus, T1DM — type 1 diabetes mellitus, SFAs — saturated fatty acids, USFAs — unsaturated fatty acids

In recent years, there has been an increasing downward trend in the mortality rate due to true diabetic causes, such as diabetic coma as a result of ketoacidosis or hypoglycemia [2, 3]. According to the Register of Diabetes Mellitus (DM), as of 2017 this figure fell to 1.5 % [2, 3]. However, among other causes of death in patients with type 1 diabetes mellitus (T1DM), cardiovascular morbidity remains dominant (39.8 %): myocardial infarction, cerebrovascular disorders, chronic heart failure and acute cardiovascular events, such as cardiac rhythm disorders, pulmonary embolism, thrombosis, sudden cardiac death, cardiogenic shock, and cerebral edema [2, 3].

* Contacts. E-mail: mushanatasha@inbox.ru

In diabetic ketoacidosis (DKA), significant metabolic disorders develop in all tissues and organs. And myocardium is no exception [4, 5, 6]. It is well known that the main energy substrate for the cardiac muscle is fatty acids [4, 5, 7]. According to literature, they account for 60–70 % of the myocardium energy demands [4, 5, 7]. Medical literature extensively covers the changes occurring in the fatty acid composition of serum lipids in conditions, such as ischemic heart disease, hypertension, inflammatory and dystrophic conditions of the myocardium, acute alcohol-induced cardiac damage, etc. [1], and the study of fractional composition of serum lipids in patients with T1DM complicated by diabetic ketoacidosis (DKA), depending on DKA severity. However, the issue of how the fatty acid composition changes in patients with T1DM complicated by DKA, depending on DKA severity, remains poorly studied. One of the proposed mechanisms of development of cardiovascular disorders in patients with T1DM complicated by DKA can be impaired metabolism and composition of fatty acids.

Study objective: to investigate the fractional composition of serum lipids in patients with T1DM complicated by diabetic ketoacidosis (DKA), depending on DKA severity.

Materials and Methods

The fatty acid composition of serum lipids was studied in the following groups of patients: Group 1 — 68 patients with compensated T1DM; Group 2 — 54 patients with T1DM complicated by mild DKA; Group 3 — 42 patients with moderate and severe DKA. Patients in Groups 2 and 3 were hospitalized at the decompensation stage. T1DM and severity of DKA were diagnosed on the basis of the relevant criteria of National Guidelines for the diagnosis and treatment of DM patients, which relied on the therapeutic standards established by the WHO [2]. The mean age of the patients was 29.2±8.9 years. Blood sampling for the analysis was performed on day 1 after admission. The compensation of carbohydrate metabolism in Group 1 patients was confirmed by daily glycemia and glycated hemoglobin level. The control group included 32 healthy individuals without impaired carbohydrate metabolism and risk factors for cardiovascular diseases, who were comparable in gender and age to patients in Groups 1, 2 and 3.

Venous blood was used as study material. Lipids were extracted from serum by the method developed by J. Fohl et al. (1957), and then fatty acids (FAs) were methylated according to the method proposed by K. M. Sinyak et al. (1976), followed by analysis using the Crystal-2000M gas chromatograph (Russia). Fatty Acid Methyl Ester (FAME) Standard produced by Sigma (USA) was used for calibration. Peaks were calculated and identified using the Analytica for Windows software and hardware package based on IBM Pentium IV 1800. Concentrations of the following higher FAs were determined: myristic (C14:0), palmitic (C16:0), stearic (C18:0), palmitoleic (C16:1), oleic (C18:1), linoleic (C18:2ω6), α-linolenic (C18:3ω3), γ-linolenic (C18:3ω6), dihomo-γ-linolenic (C20:3ω6), and arachidonic (C20:4ω6).

Statistica 6.0 software package was used for statistical data processing. The significance of differences among the study groups was assessed using the non-parametric Mann-Whitney test. Differences at $p<0.05$ were considered statistically significant. The data were presented as median, 25th and 75th percentiles (Me 25p; 75p).

Results and Discussion

When analyzing serum concentrations of fatty acids in T1DM patients depending on the severity of DKA, we identified regular changes in the fatty-acid profile, as presented in the table below.

What attracts attention is the significant increase in the total content of saturated fatty acids (SFAs), the decrease in the total concentration of unsaturated fatty acids (USFAs), as well as the increase in the SFA/USFA ratio in all T1DM patients as compared to healthy individuals. Statistically significant differences among the study groups were found. The total content of SFAs in the group of patients with T1DM complicated by mild DKA increased by 12.9 % as compared to those in T1DM patients without DKA ($p=0.003$). This figure in the group of patients with T1DM complicated by moderate and severe DKA increased by 198 % as compared to that in T1DM patients without DKA, and by 6 % as compared to the patients with T1DM complicated by mild DKA ($p<0.001$).

Table 1. *Fatty acid composition of serum lipids in patients with type 1 diabetes complicated by diabetic ketoacidosis (Me (25th; 75th))*

Parameter	Control (healthy) (n=31)	Type 1 diabetes mellitus		
		No DKA (n=68)	Mild DKA (n=54)	Moderate and severe DKA (n=42)
C14:0,%	0.92 (0.67;1.19)	1.15 (1.01;1.92) $p<0.001$	1.13 (1.09; 1.46) $p<0.001$	2.16 (2.13; 2.36) $p<0.001$ $p_1<0.001$ $p_2<0.001$
C16:0,%	24.37 (21.69; 27.29)	28.96 (25.97; 30.09) $p<0.001$	30.86 (26.37; 32.68) $p<0.001$ $p_1=0.008$	32.8 (29.94; 35.68) $p<0.001$ $p_1<0.001$ $p_2<0.001$
C18:0,%	7.17 (5.66; 8.37)	6.88 (6.12; 8.26)	8.68 (7.52; 9.24) $p<0.001$ $p_1<0.001$	9.29 (8.55; 9.55) $p<0.001$ $p_1<0.001$ $p_2=0.003$
C16:1,%	3.19 (2.89; 4.82)	3.23 (2.51; 4.64)	2.78 (2.58; 3.48) $p<0.001$ $p_1=0.017$	2.53 (2.31; 2.78) $p<0.001$ $p_1<0.001$ $p_2<0.001$
C18:1,%	23.66 (20.62; 25.2)	23.85 (21.95; 25.37)	21.71 (20.31; 24.54) $p_1<0.001$	19.77 (18.92; 21.4) $p<0.001$ $p_1<0.001$ $p_2<0.001$
C18:3ω3,%	1.72 (1.35; 2.91)	1.53 (1.1; 1.71) $p<0.001$	1.24 (0.74; 1.6) $p=0.004$ $p_1=0.024$	1.15 (0.8; 1.29) $p<0.001$ $p_1=0.007$
C18:2ω6,%	31.74 (26.95; 34.32)	26.33 (24.45; 30.59) $p<0.001$	26.22 (24.54; 28.59) $p<0.001$	24.21 (23.59; 25.38) $p<0.001$ $p_1<0.001$ $p_2<0.001$
C18:3ω6,%	0.43 (0.33; 0.71)	0.95 (0.50; 1.53) $p<0.001$	1.32 (0.76; 1.42) $p<0.001$	2.07 (1.82; 2.36) $p<0.001$ $p_1<0.001$ $p_2<0.001$
C20:3ω6,%	0.92 (0.78; 1.09)	1.19 (0.96; 1.65) $p<0.001$	1.63 (1.22; 1.76) $p<0.001$ $p_1=0.007$	1.99 (1.39;2.1) $p<0.001$ $p_1=0.003$ $p_2<0.001$
C20:4ω6,%	4.17 (3.77; 7.28)	3.42 (2.69; 5.41) $p<0.001$	3.58 (2.9; 5.15) $p=0.001$	2.88 (2.57; 3.22) $p<0.001$ $p_1<0.001$ $p_2<0.001$

Table 1. *(The end)*

Parameter	Control (healthy) (n=31)	Type 1 diabetes mellitus		
		No DKA (n=68)	Mild DKA (n=54)	Moderate and severe DKA (n=42)
Σ polyunsaturated fatty acids	39.77 (37.02; 45.55)	34.5 (32.0; 39.75) ρ<0.001	33.67 (31.24; 37.04) ρ<0.001	32.1 (30.84; 33.94) ρ<0.001 ρ_1<0.001 ρ_2=0.01
Σ monounsaturated fatty acids	27.51 (23.6; 29.03)	23.36 (25.78; 31.22) ρ<0.001	24.67 (23.03; 27.5) ρ<0.001	22.31 (21.5; 23.58) ρ<0.001 ρ_1<0.001 ρ_2<0.001
Σ unsaturated fatty acids	66.9 (65.9; 69.24)	63.47 (61.81; 65.08) ρ<0.001	59.93 566; 62.6) ρ<0.001 ρ_1<0.001	54.62 5368; 55.65) ρ<0.001 ρ_1<0.001 ρ_2<0.001
Σ saturated fatty acids	33.08 (30.76; 34.1)	36.64 (33.8; 37.7) ρ<0.001	41.4 (35.56; 43.66) ρ<0.001 ρ_1=0.003	43.9 (42.35; 47.34) ρ<0.001 ρ_1<0.001 ρ_2<0.001
Σ ω6 acids	38.45 (35.8; 43.59)	33.1 (30.47; 37.23) ρ<0.001	32.33 (30.74; 35.95) ρ<0.001	31.23 (29.96; 32.44) ρ<0.001 ρ_1<0.001 ρ_2<0.001
Σ ω3 acids	1.72 (1.35; 2.91)	1.53 (1.11; 1.71) ρ<0.001	1.24 (0.74; 1.6) ρ=0.004 ρ_1=0.024	1.11 (0.8; 1.29) ρ<0.001 ρ_1=0.007 ρ_2<0.001
Saturated fatty acids / unsaturated fatty acids, units	0.49 (0.44; 0.52)	0.57 (0.53; 0.61) ρ<0.001	0.7 058; 0.76) ρ<0.001 ρ_1=0.005	0.81 (0.75; 0.87) ρ<0.001 ρ_1<0.001 ρ_2<0.001
Poly-/ monounsaturated fatty acids, units	1.32 (1.04; 1.54)	1.32 104; 1.54) ρ=0.016	1.37 137; 1.51) ρ=0.001	1.42 137; 1.58) ρ<0.001 ρ_1=0.005 ρ_2=0.034
ω3/ω6, units	0.04 (0.03; 0.06)	0.05 003; 0.05)	0.03 003; 0.05) ρ=0.006 ρ_1=0.037	0.03 (0.03; 0.04) ρ=0.006 ρ_1=0.006
Saturated / polyunsaturated fatty acids, units	0.81 (0.72; 0.91)	1.06 (0.91; 1.16) ρ<0.001	1.13 103; 1.27) ρ<0.001 ρ_1=0.003	1.41 (1.26; 1.48) ρ<0.001 ρ_1<0.001

Note: n — number examined; ρ — level of significance of differences compared to control; ρ_1 — level of significance of differences compared with the group without ketoacidosis; ρ_2 — level of significance of differences compared with the group of mild ketoacidosis

In analyzing concentrations of individual saturated fatty acids, it can be seen that statistically significant changes as compared to T1DM patients without DKA were observed only in concentrations of palmitic (C16:0) and stearic acids (C18:0) in the group of patients with T1DM complicated by mild DKA. These figures increased by 6.6 % (p=0.008) and 26.1 % (p<0.001), respectively, as compared to similar parameters in Group 1. It should be noted that the concentrations of stearic acid showed no statistically significant difference between the group of T1DM patients without DKA and healthy individuals. The most pronounced changes in all the SFAs studied were identified in the group of patients with T1DM complicated by moderate and severe DKA. The content of myristic acid (C14:0) was 87.8 % higher than in the group of T1DM patients without DKA, and 91.1 % higher than that in patients with T1DM complicated by mild DKA (p<0.001). As for palmitic (C16:0) and stearic (C18:0) acids, their concentrations in Group 3 was much higher than these figures in Groups 1 and 2. Concentrations of palmitic acid (C16:0) were 13.2 % and 6.3 % higher than those in the groups of T1DM patients without DKA (p<0.001) and patients with T1DM complicated by mild DKA (p=0.008), respectively. Levels of stearic acid (C18:0) were 35 % and 7 % higher than in the above-mentioned study groups, respectively (p<0.001; p=0.003 for Groups 2 and 3). Myristic acid (C14:0) concentrations in all of the study groups were similar to those in the control group. No statistical difference was found for this parameter between T1DM patients without DKA and with mild DKA. In contrast, the concentration of myristic acid (C14:0) in the group of patients with moderate and severe DKA was 87.8 % and 91.1 % higher than that in Groups 1 and 2, respectively, and the difference with the control group was 163.4 % (p<0.001).

The total content of polyunsaturated fatty acids in T1DM patients without DKA was 94.8 % of the level reported in the control group. The figure was 6.6 % higher than in patients with T1DM complicated by mild DKA, and 13.9 % higher than in patients with T1DM complicated by moderate and severe DKA. Differently directed changes were noticeable when considering concentrations of individual USFAs of serum lipids.

It should be noted that no statistically significant difference was identified, based on concentrations of palmitoleic (C16:1) and oleic acids (C18:1), between the control group and T1DM patients without DKA. On the contrary, concentrations of linoleic (C18:2ω6), α-linolenic (C18:3ω3) and arachidonic (C20:4ω6) acids in Group 1 were lower by 17 %, 11 % and 17.9 %, respectively, as compared to healthy individuals (p<0.001), and levels of γ-linolenic (C18:3ω6) and dihomo-γ-linolenic (C20:3ω6) acids were higher by 120 % and 29.3 %, respectively.

Statistically significant changes, as compared to the control group and T1DM patients without DKA, were observed in concentrations of palmitoleic (C16:1), oleic (C18:1), linoleic (C18:2ω6), α-linolenic (C18:3ω3), and dihomo-γ-linolenic (C20:3ω6) acids in both groups of patients with T1DM complicated by DKA. The concentrations of palmitoleic acid (C16:1) decreased by 12.8 % and 14 %, respectively, in the group of patients with T1DM complicated by mild DKA, and by 20.6 % and 217 %, respectively, in patients with T1DM with moderate and severe DKA (p<0.001). The concentration of oleic acid (C18:1) in serum lipids in Groups 2 and 3 was lower than that in T1DM patients without DKA by 9 % and 17.1 %, respectively (p<0.001). The reduction in α-linolenic acid (C18:3ω3) was 19 % and 24.8 %, respectively, for patients with T1DM complicated by mild DKA (p=0.024) and moderate and severe DKA (p=0.007). The most pronounced changes were identified in the USFA pool with respect to concentrations of γ-linolenic (C18:3ω6) and dihomo-γ-linolenic (C20:3ω6) acids. On the contrary, their concentrations increased in the groups of patients with T1DM complicated by mild DKA and moderate to severe DKA. The concentration of γ-linolenic acid (C18:3ω6) was 398 % and 117.8 % higher than that in T1DM patients without DKA, and 206.9 % and 3813 % higher than that in healthy individuals (p<0.001). Examinations of dihomo-γ-linolenic acid (C20:3ω6) concentrations in these groups showed a difference of 36.9 % and 67.2 %, respectively, as compared to Group 1 patients, and 77.1 % and 116.3 %, respectively, as compared to healthy individuals.

As for concentrations of monounsaturated acids, T1DM patients without DKA were not significantly

different from Group 2, and these figures were lower by 15.1 % and 10.3 %, respectively, as compared to healthy individuals. The concentrations of polyunsaturated acids were also lower in patients from the above-mentioned study groups and averaged 86.7 % and 88.3 % of the similar figures in healthy individuals ($p<0.001$). Changes in monounsaturated and polyunsaturated acids were also determined in the group of patients with T1DM complicated by moderate and severe DKA. The decrease in monounsaturated acids was 9.6 %, 4.5 % and 18.9 %, respectively, as compared to patients with T1DM complicated by mild DKA, T1DM patients without DKA, and healthy individuals ($p<0.001$). The decrease in polyunsaturated acids in Group 3 was 4.6 %, 6.9 % and 192 %, respectively, as compared to the above-mentioned groups.

The ratio of polyunsaturated and monounsaturated fatty acids in blood lipids increased in all of the study groups as compared to the control group. The most pronounced changes were identified in patients with T1DM complicated by moderate and severe DKA. These figures in patients of Group 3 were 3.6 % and 7.5 % higher as compared to patients with T1DM and mild DKA and T1DM patients without DKA and healthy individuals. It should be noted that no statistically significant difference was identified for this parameter between the groups of patients with T1DM complicated by mild DKA and those without DKA.

At the same time, there was a statistically significant increase in the SFA/USFA ratio due to the increase mainly in myristic acid (C14:0) and, to a lesser degree, in stearic acid (C18:0), and decrease in γ-linolenic (C18:3ω6) and dihomo-γ-linolenic acids (C20:3ω6).

The ratio of ω-3 to ω-6 polyunsaturated fatty acids in blood lipids showed statistically significant decrease in patients with T1DM complicated by DKA, irrespective of the severity of DKA, as compared to T1DM patients without DKA and the healthy individuals. No differences were found in the group of T1DM patients with DKA depending on DKA severity. The ω3/ω6 ratio was reported mainly due to reduction in concentrations of ω-3 fatty acids and relative increase in proportions of γ-linolenic (C18:3ω6) and dihomo-γ-linolenic (C20:3ω6) fatty acids.

Thus, our study identified significant changes in the fractional blood fatty acid composition in patients with T1DM complicated by DKA. The identified disorders were based on the imbalance of FAs expressed as a decrease in the pool of unsaturated fatty acids and an increase in that of saturated fatty acids. These changes develop as a part of general systemic metabolic disorders in the study cohort of patients.

Conflict of interests

The authors declare no conflict of interests.

References:

1. Govorin A.V. Non-coronary myocardial damage. Novosibirsk: «Science». 2010; 231 p. [in Russian].
2. Dedov I.I., Shestakova M.V., Vikulova O.K. The prevalence, morbidity, mortality, parameters of carbohydrate metabolism and the structure of glucose-lowering therapy according to the federal diabetes mellitus registry, status 2017. Diabetes mellitus. 2018; 21 (3): 144-159. [in Russian].
3. Dedov I.I., Shestakova M.V., Galstyan G.R. The prevalence of type 2 diabetes in the adult population of Russia (NATION study). Diabetes. 2016; 19 (2): 104-112. [in Russian].
4. Kotkina, T.I., Titov, V.N., Parkhimovich R.M. Other ideas on the β-oxidation of fatty acids in peroxisomes, mitochondria and ketone bodies. Diabetic, acidotic coma as an acute deficiency of acetyl CoA and ATP. Clinical laboratory diagnosis. 2014; 3: 14-23. [in Russian].
5. Dedov I.I., Melnichenko G.A. Endocrinology: national leadership. M.: GEOTAR-Media. 2008; 1072 p. [in Russian].
6. Enert A.V., Kravets E.B., Ivanov S.N. Cardiovascular disorders in type 1 diabetes mellitus in children and adolescents (literature review). Siberian Medical Journal. 2009; 2 (4): 77-84. [in Russian].
7. Horrabin D., Mancy M., Hyang Y. Effect of essential fatty acids on prostaglandin biosynthesis. Biomed. Biohim. Acta. 1984; 72(8/9): 114 — 120.

Article received on 16.01.2019
Accepted for publication on 26.02.2019