

Lipids Peroxidation Products in Young Men with Type 1 Diabetes Mellitus

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Abstract

The aim of this research was to assess the level of lipid oxidation products level and the antioxidant defense state in young male patients with type 1 diabetes mellitus (T1DM) depending on the disease duration.

Methods and Results: A total of 57 men of young reproductive age (average age of 30.25±8.51 years) with T1DM and an unsatisfactory glycemic profile, depending on the disease duration, were divided into 2 groups. Group 1 included 29 men (average age of 27.69±6.92 years) with a T1DM duration <5 years (2.72±1.61 years) and HbA1c level of 11.37±2.74%. Group 2 included 28 men (average age of 32.89±9.28 years) with a T1DM duration ≥5 years (12.93±5.69) and HbA1c level of 10.19±2.18%). The control group consisted of 28 men of the same age (29.71±4.59 years). Spectrophotometric/spectrofluorometric methods and enzyme immunoassay were used. We found a significant increase in the values of CDs (by 2.04 times, $P<0.0001$), KD and CT (by 2.38 times, $P<0.0001$), TBARs (by 1.17 times, $P=0.001$), SB (by 2.6 times, $P<0.0001$), and retinol (by 1.44 times, $P<0.0001$) in Group 1 compared to the control group. In Group 2, there was a statistically significant increase in the levels of CDs (by 2.59 times, $P<0.0001$), KD and CT (by 2.94 times, $P<0.0001$), TBARs (by 1.49 times, $P=0.001$), SB (by 3.27 times, $P<0.0001$), and retinol (by 1.4 times, $P=0.001$) compared to the control group. The differences between the two groups with different duration of T1DM were characterized only by the CDs level, which was increased in Group 2 patients with a T1DM duration of ≥5 years (by 1.27 times, $P=0.048$) compared to Group 1 patients with a T1DM duration of <5 years.

Conclusion: LPO parameters can serve as additional laboratory markers that characterize the course of T1DM and can be used to develop potential prevention and therapy strategies. (**International Journal of Biomedicine. 2022;12(2):232-236.**)

Key Words: type 1 diabetes mellitus • men • diabetes duration • lipid peroxidation • antioxidant defense

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Abbreviations

AOD, antioxidant defense; **CDs**, conjugated dienes; **DM**, diabetes mellitus; **KD-CT**, ketodienes and conjugated trienes; **LPO**, lipid peroxidation; **MDA**, malondialdehyde; **OS**, oxidative stress; **RBC**, red blood cells; **ROS**, reactive oxygen species; **SOD**, superoxide dismutase; **TAA**, total antioxidant activity; **T1DM**, type 1 diabetes mellitus; **TBARs**, thiobarbituric acid reactants; **SB**, Schiff bases; **T2DM**, type 2 diabetes mellitus.

Introduction

Diabetes mellitus (DM) is one of the most critical problems of our time due to its high prevalence, early disability, and a decrease in the patients' life expectancy.⁽¹⁾ According to WHO (2021), between 2000 and 2016, there was a 5% increase in premature mortality from diabetes. More

than 10 million patients with DM are officially registered in the Russian Federation, of which more than 300 thousand are patients with type 1 diabetes mellitus (T1DM).⁽²⁾

Reactions of free radical oxidation, including lipid peroxidation (LPO), play an essential role in the pathogenesis of T1DM and its complications.⁽³⁾ LPO is a universal metabolic process represented in all organs and tissues. LPO reactions,

having the ability to modify the structure and functions of cell membranes, can determine the nature of intercellular and inter-organ relationships within a certain functional system, as evidenced by the data on the direct participation of LPO in the xenobiotic metabolism, in the regulation of the immune response, cell proliferation, vascular permeability, receptor sensitivity, etc.^(4,5) Under physiological conditions, the LPO reaction state can inform us about the nature of adaptive reactions.⁽⁶⁾ At the same time, the LPO intensification and the insufficiency of antioxidant response define the pathogenesis of many diseases, including T1DM.^(7,8)

The main molecular mechanisms associated with oxidative damage reactions in T1DM, associated with glucose and lipid metabolism, have been identified.⁽⁹⁾ They include the glycolytic pathway, enhanced formation of advanced glycation end products, the hexosamine pathway, protein kinase C activation, polyol pathway, and insulin signaling pathway deactivation.^(7,9) Oxidative stress (OS) can play a dual role concerning T1DM, contributing not only to its manifestation, but also to the exacerbation of the disease and related complications.⁽¹⁰⁾ Despite numerous studies of these reactions in T1DM, the age and gender of the subjects usually were not taken into account.

In this regard, the aim of our work was to assess the lipid oxidative damage products level and the antioxidant defense state in young men with T1DM, depending on the disease duration.

Material and Methods

Design of study

A total of 57 men of young reproductive age (average age of 30.25±8.51 years) with T1DM and an unsatisfactory glycemic profile, depending on the disease duration, were divided into 2 groups. Group 1 included 29 men (average age of 27.69±6.92 years) with a T1DM duration of <5 years (2.72±1.61 years) and HbA1c level of 11.37±2.74%. Group 2 included 28 men (average age of 32.89±9.28 years) with a T1DM duration of ≥5 years (12.93±5.69) and HbA1c level of 10.19±2.18%). The patients were treated in the endocrinology department of the Irkutsk Regional Clinical Hospital. The control group consisted of 28 men of the same age (29.71±4.59 years). Inclusion criteria for Groups 1 and 2 were confirmed diagnosis of T1DM, men aged 18-40 years, and residence in the specified territory. Exclusion criteria for Groups 1 and 2 were T2DM or other types of diabetes, severe DM complications (proteinuria, renal failure, and macrovascular complications), and other endocrine diseases, pronounced concomitant somatic pathology. Inclusion criteria for the control group were the absence of acute or exacerbation of chronic diseases at the time of the examination, normal indicators of glucose tolerance, absence of a hereditary predisposition to DM.

The study complied with the ethical principles of World Medical Association Declaration of Helsinki (1964, ed. 2013) and it was approved by the Biomedical Ethic Committee at the Scientific Centre for Family Health and Human Reproduction Problems (No. 8.2 dated November 2, 2018). Written informed consent was obtained from all participants.

Biochemical measurements

Plasma, serum and erythrocyte hemolysate were used as the material for the study. Plasma concentrations of primary/secondary/final products of LPO (CDs/KD-CT, TBARs/SB) were estimated.⁽¹¹⁾ TBARs content was detected by fluorometry.⁽¹²⁾

The state of the AOD system was determined by TAA in blood serum (using a commercial kit from Randox (UK)), the SOD activity in hemolysate (using a commercial kit from Randox (UK)), and the content of α -tocopherol and retinol in the blood plasma.⁽¹³⁾ The measurements were carried out using a Shimadzu RF-1501 spectrofluorophotometer (Japan) and Shimadzu RF-1650 spectrofluorophotometer (Japan). Enzyme immunoassay was performed using a MultiSkan ELX808 microplate reader (Biotek, USA).

Statistical analysis was performed using STATISTICA 10.0 software package (Stat-Soft Inc, USA). The normality of distribution of continuous variables was tested by the Kolmogorov-Smirnov test with the Lilliefors correction and Shapiro-Wilk test. The F test for equality of two variances was applied. For descriptive analysis, results are presented as mean±standard deviation (SD). For data with normal distribution, inter-group comparisons were performed using Student's t-test. Differences of continuous variables departing from the normal distribution, even after transformation, were tested by the Mann-Whitney U-test. The Spearman correlation coefficient r_s was calculated to measure the strength and direction of the relationship between two variables. A probability value of $P \leq 0.05$ was considered statistically significant.

Results and Discussion

We found a significant increase in the values of CDs (by 2.04 times, $P < 0.0001$), KD and CT (by 2.38 times, $P < 0.0001$), TBARs (by 1.17 times, $P = 0.001$), and SB (by 2.6 times, $P < 0.0001$) in Group 1 compared to the control group (Fig. 1). Among AOD parameters, in Group 1, only the retinol level was significantly greater (by 1.44 times, $P < 0.0001$) than in the control group.

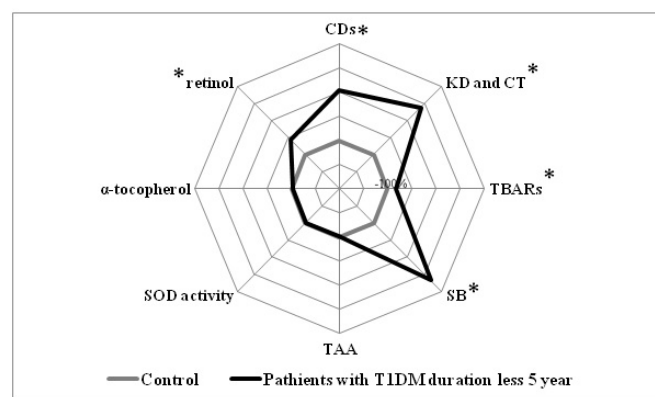


Fig. 1. State of the LPO-AOD system in patients with T1DM duration of <5 years (*- statistically significant differences compared to the control (values are taken as 100%).

Group 2 was characterized by similar changes in the LPO-AOD system. Thus, there was a statistically significant

increase in the levels of CDs (by 2.59 times, $P < 0.0001$), KD and CT (by 2.94 times, $P < 0.0001$), TBARs (by 1.49 times, $P = 0.001$), SB (by 3.27 times, $P < 0.0001$), and retinol (by 1.4 times, $P = 0.001$) in Group 2 compared to the control group (Fig. 2).

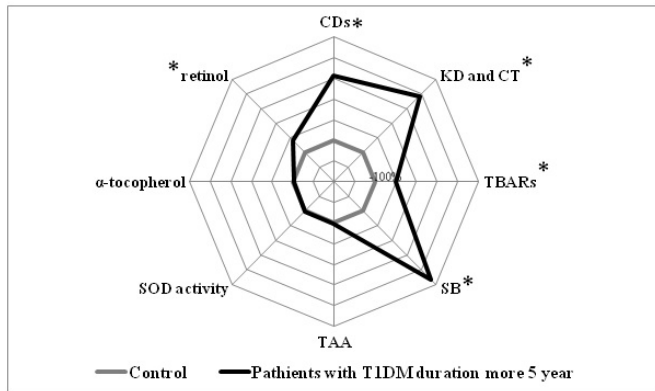


Fig. 2. State of the LPO-AOD system in patients with T1DM duration of ≥ 5 years (*- statistically significant differences compared to the control (values are taken as 100%).

The differences between two groups with different duration of T1DM have characterized only the CDs level, which was increased in Group 2 patients with a T1DM duration of ≥ 5 years (by 1.27 times, $P = 0.048$) compared to Group 1 patients with a T1DM duration of < 5 years.

Data analysis found that in patients with T1DM, regardless of the disease duration, a significant increase in the content of primary LPO products occurs at all stages of the LPO process. However, when the duration of T1DM was more than 5 years, the changes were more pronounced.

According to the literature data, the activation of LPO processes in erythrocyte membranes occurs at the T1DM debut. The presence of atherogenic dyslipidemia and LPO activation manifested by an increased content of primary and secondary LPO products in patients with newly diagnosed T1DM has been shown in numerous studies.⁽¹⁴⁾ At the same time, the assumption that the LPO-AOD system imbalance is closely related to the degree of compensation and severity of T1DM turned out to be significant.^(15,16) The state of metabolic decompensation in T1DM, defined as “metabolic stress,” also includes lipid metabolism disturbances.^(8,17,18) Many studies established a close positive relationship between the accumulation of MDA with the duration of diabetes and HbA1c and a negative association with the ferric reducing ability of plasma.^(19,20) Perhaps this is due to insulin insufficiency, which is involved in lipoperoxide utilization.⁽²¹⁾ It was also shown that the primary LPO product (CDs) accumulation might depend on the content of free fatty acids.^(3,14,22) With the enhanced LPO activity, the interactions between insulin and its receptors can be changed due to the MDA’s ability covalently binding to cell membranes of lipids and proteins with the crosslinking formation.⁽⁵⁾ All of the above leads to the alteration of insulin receptor internalization—the number of insulin-binding sites decreases, which can serve as one of the causes of insulin resistance.^(23,24) It was noted that under conditions of poorly controlled T1DM, there is a direct dependence of the

malondialdehyde level and SOD activity from the ketone bodies content due to their excess formation in T1DM.⁽²⁵⁾ The products of toxic MDA reactions with proteins, phospholipids, and nucleic acids are not destroyed due to strong bonds. They are accumulated in the body, leading to the violations of biopolymers properties, which can worsen a patient’s condition in conditions of AOD system poor functioning.⁽³⁾

The disease duration can also aggravate the LPO-AOD system disturbances in T1DM patients. Thus, a pronounced LPO process activation can increase in patients with diabetes duration of more than 10 years.^(3,20) Many studies have reported that in patients with T1DM complications, the content of LPO products was more pronounced, suggesting their participation in the vascular damage initiation.⁽²⁶⁻²⁸⁾ Toxic products of LPO can cause microvascular complications through various mechanisms. In DM, the increased oxidative stress, the alteration of lipogenesis, the reduction of nitric oxide, and the alteration of endothelial progenitor cell function create damage to the vessel wall leading to the pathogenesis of arterial thrombus.⁽²⁹⁾ The pathogenesis of endothelial dysfunction in T1DM is complex and involves metabolic and hormonal changes. In particular, insulin deficiency leads to decreased number of endothelial progenitor cells, decreased nitric oxide production, increased oxidative stress in the vascular milieu, and a consequent decrease in the ability to promote vessel dilation.⁽³⁰⁾ It was found that the cell membranes of T1DM patients undergo significant structural changes. Lee et al.⁽³¹⁾ demonstrated that hyperglycemia in T1DM patients severely impairs RBC deformability by remodeling the mechanical properties of the cell membrane. Currently, several impaired biochemical pathways such as glycolytic, hexosamine, protein kinase C, polyol, and advanced glycation end-product (AGE) pathways have been identified as pro-oxidative processes in the diabetics.⁽³²⁻³⁸⁾ Inhibition of glyceraldehyde-3-P dehydrogenase by poly-ADP-ribose polymerase 1 and subsequent accumulation of the enzyme substrate (glyceraldehyde-3-P) appears to be central to diabetes-associated oxidative stress.⁽⁹⁾

Any impact that causes an increase in the peroxidation process exerts a different effect depending on the activity of the antioxidant system. We found no significant differences in most of the studied parameters, with the exception of retinol, the values of which were elevated in both two groups with T1DM. Insulin insufficiency may be the main cause of the antioxidant status changes in T1DM.⁽³⁹⁾ It was noted that the vascular pathology progression in T1DM is associated with increasing AOD insufficiency, manifested in the form of the main antioxidants concentration decrease.⁽⁴⁰⁻⁴³⁾

Thus, in patients with T1DM and diabetic nephropathy (decompensated form), against the background of metabolic disorders, the LPO activation and the antioxidant system inhibition are noted.⁽¹⁰⁾ Very contradictory information was obtained while studying the activity of antioxidant enzymes in the RBC of patients with T1DM. Analysis of serum TAA using bioluminescence revealed a significant decrease in this indicator in patients with T1DM decompensation stage compared with healthy people.⁽⁷⁾ In our study, we observed an increase in retinol values regardless of the disease duration. It can be assumed that retinol plays the role of both an

independent antioxidant that ensures the preservation of the cell membrane functional stability, blocking the LPO processes in the cell membrane, and serve as a synergist of α -tocopherol—the main fat-soluble antioxidant.

Thus, an increase in retinol content can be regarded as a compensatory phenomenon. However, despite the increase in its values, patients with T1DM have a significant LPO activity at all disease stages, which can be characterized as a shift in the redox balance toward pro-oxidant factors.

Conclusion

We identified the significant increase in the LPO product content and the retinol level with longer disease duration in young men with T1DM. LPO parameters can serve as additional laboratory markers that characterize the course of T1DM and can be used to develop potential prevention and therapy strategies.

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Competing Interests

The authors declare that they have no competing interests.

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