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The Assessment of Oxidative Stress Intensity in Adolescents with Obesity by the Integral Index

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Abstract

The aim of this research was to assess lipid peroxidation (LPO) and antioxidative defense (AOD) changes in adolescent boys with obesity using the integral index.

Materials and Methods: We examined 19 adolescent boys with obesity of the first degree (the study group). The control group included 23 healthy adolescent boys. The study included the collection of anamnestic data, physical examination, and anthropometric data analysis (body weight (BW), height, waist circumference (WC), hip circumference (HC), and body mass index (BMI)). Laboratory analysis included an assessment of the blood levels of total cholesterol, triglycerides, HDL, LDL, and glucose, as well as the intensity of LPO and AOD in blood plasma and primary and secondary products of LPO. To measure the intensity of OS, the oxidative stress index (OSi) was calculated (the ratio of the LPO-AOD system indicators in the study group to average indicators in the control group).

Results: We found a statistically significant increase in BW, BMI, SDS BMI, WC, and HC in the study group compared to the control group. The obese patients had higher values of blood glucose, total cholesterol, triglycerides and LDL compared to the control group. In the study group, we found a significant decrease in the concentration of diene conjugates and an increase in the level of ketodienes and conjugated trienes. The values of α-tocopherol and retinol, and SOD activity were significantly decreased in the study group compared to the control group. There were no statistically significant changes in total antioxidant activity and glutathione status components. According to the data received, the OSi level in the group of obese patients increased approximately 7 times, which confirms the results on the development of antioxidant insufficiency in this pathology.**(International Journal of Biomedicine. 2018;8(1):37-41.)**

Key Words: lipid peroxidation • antioxidative defense • oxidative stress index • adolescent boys • obesity

Abbreviations

AOD, antioxidative defense; **BMI,** body mass index; **BW**, body weight; **CDs,** conjugated dienes; **GSH,** reduced glutathione; **GSSG,** oxidized glutathione; **HC**, hip circumference; **HDL,** high-density lipoprotein; **KD-CT,** ketodienes and conjugated trienes; **LDL**, low-density lipoprotein; **LPO,** lipid peroxidation; **OS,** oxidative stress; **SDS BMI,** the standard deviation score of BMI; **SOD,** superoxide dismutase; **TAA,** total antioxidant activity; **TBARs**, thiobarbituric acid reactants; **WC**, waist circumference.

Introduction

Childhood obesity is a serious medical condition that affects children and adolescents. In the developed countries, 25% of adolescents have excessive body weight and 15% have obesity.[1-4] The incidence of obesity is 6.8% in boys 5 to 17 years old and 5.3% in girls.[5] Childhood obesity usually progresses and often leads to health problems in adulthood: hypertension, metabolic syndrome, diabetes, and cardiovascular and fatty liver diseases.

WHO experts connect the prevalence of obesity in childhood with economic and social changes of life in a

modern society, unhealthy diets, and low physical activity.[6,7] The study of molecular mechanisms of obesity formation in adolescence is very important.[8-10] One of the pathogenetic mechanisms of obesity development is OS activation and a decrease in AOD activity.[11-15] Endogenous aldehydes generated during OS can act as mediators of damage, which precedes the emergence of metabolic shifts.[14]

The aim of this research was to assess LPO and AOD changes in adolescent boys with obesity using the integral index.

Materials and Methods

We examined 19 adolescent boys (mean age of 4.41±0.45 years) with obesity of the first degree (the study group) according to the WHO classification.[16] The control group included 23 healthy boys (mean age of 15.12±0,32 years). The study included the collection of anamnestic data, physical examination, and anthropometric data analysis (BW, height, WC, HC, and BMI). BMI was calculated using Quetelet's formula:

$BMI = body\ weight(kg)/height(cm)^2$

Laboratory analysis included an assessment of the blood levels of total cholesterol, triglycerides, HDL, LDL, and glucose. The quality, quantity and regularity of food intake were evaluated. Hereditary anamnesis included the presence of relatives with type 2 diabetes, obesity, ischemic heart disease, and hypertension. All patients did not take vitamins during the blood sampling period.

Blood was taken from the ulnar vein in accordance with the existing requirements in the morning after an overnight fast. Blood samples were centrifuged for 5 min at 1.500g at 4°C; erythrocytes were rinsed three times with 0.9% NaCI. Aliquots of the separated EDTA plasma and washed erythrocytes were used immediately or kept frozen at -40°C (not longer than one month).

We estimated the intensity of LPO and AOD in blood plasma (TAA, SOD, GSH, GSSG, α-tocoferol, retinol) and primary and secondary products of LPO (CDs, ketodienes, KD-CT and TBARs). The concentrations of CDs and KD-CT were evaluated at 232 nm in plasma heptane extracts.[17] The coefficient of molar absorption (K=2.2 • 105 M-1 C-1) for conversion of absorption units to μmol/L was used. TBARs levels were detected by fluorometry[18] and estimated in μmol/L.

Plasma levels of GSH, GSSG, α-tocopherol and retinol, as well as SOD activity in hemolysate were detected by fluorometry.[19-21] Plasma TAA levels were detected photometrically.[22] The measurements were conducted by a spectrophotometer consisting of two blocks: a UV-1650PC spectrophotometer and an RF-1501 spectrofluorimeter. To measure the intensity of OS, the oxidative stress index (OSi) was calculated (the ratio of the LPO-AOD system indicators in the study group to average indicators in the control group).[23]

The study was conducted in accordance with ethical principles of the WMA Declaration of Helsinki (1964, ed. 2000) and approved by the Ethics Committee of Scientific Centre for Family Health and Human Reproduction Problems. Written informed consent was obtained from all participants.

Statistical analysis was performed using the statistical software «Statistica». (v6.1, StatSoft, USA).The mean (M) and standard deviation (SD) were calculated. 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. A probability value of $P<0.05$ was considered statistically significant.

Results and Discussion

We found a statistically significant increase in BW (1.48 times, $P<0.001$), BMI (1.5 times, $P<0.0001$, SDS BMI (12.43 times, $P<0.0001$), WC (1.35 times, $P<0.0001$, and HC (1.21 times, $P<0.0001$) in the study group compared to the control group. In obese patients, we found an increase of 1.4 times in glucose level ($P=0.028$), which can be explained by impaired glucose tolerance.[24] The obese patients had higher values of total cholesterol, triglycerides and LDL compared to the control group (Table 1). According to a number of authors, changes in the blood lipids in children with obesity depend on the severity of the disease.[25]

Table 1.

Clinical characteristics and biochemical parameters in adolescents of the study and control groups

Variable	Control group	Study group
Age, years	15.69±1.49	14.58±1.98
Weight, kg	64.83±9.17	95.82±15.06*
Height, cm	174.20±9.17	173.40±8.93
BMI	21.25±1.01	31.79±3.61*
SDS BMI	0.23±0.30	2.86±0.51*
WC, cm	73.20±4.53	99.10±8.49*
HC, cm	88.00±4.00	106.2±6.65*
Total cholesterol, μmol/L	3.43±0.44	4.83±0.64*
Triglycerides, μmol/L	0.43±0.16	1.63±0.52*
HDL, μmol/L	1.10±0.24	1.18±0.38
LDL, μmol/L	2.75±0.39	2.83±1.06*
Glucose, μmol/L	4.02±1.66	5.64±1.29*

** - P<0.05 vs. the control group*

In the study group, we found a significant decrease in the concentration of diene conjugates (primary products of LPO) and an increase in the level of ketodienes and conjugated trienes (secondary products of LPO) on the background of the absence of statistically significant changes in the content of TBA-active products (Table 2). The obtained results partly agree with multiple research data showing that obesity stimulates the processes of free radical oxidation, and the resulting OS acts as one of the pathogenetic mechanisms of obesity. Over-expression of OS damages cellular structures together with under-production of anti-oxidant mechanisms,

leading to the development of obesity-related complications. [26] It was found that the increased content of ketodienes and conjugated trienes might provoke a multifaceted deleterious effect on biopolymers and cellular structures. In obesity, increased OS in plasma is due to increased ROS production from accumulated fat Shigetada Furukawa and colleagues suggested that obesity per se may induce systemic OS stress and that increased OS in accumulated fat is, at least in part, the underlying cause of dysregulation of adipocytokines and development of metabolic syndrome.[11] OS plays a crucial role in disorders related to obesity, such as dyslipidemia.[26] In addition to a pro-inflammatory process, ROS can also directly damage lipids, proteins or DNA and modulate intracellular signalling pathways, such as mitogen activated protein kinases and redox sensitive transcription factors, causing changes in protein/lipid expression and, therefore, irreversible oxidative damage.[27] Due to ROS-mediated changes in lipid expression, further oxidation-derived products, including oxidative LDL (Ox-LDL), can play a further critical role in CVD. Additionally, Ox-LDL alters the production of adipokines which can lead to further OS.[26,28] Increased Ox-LDL in obese patients with dyslipidemia may be due to loss of antioxidant capacity caused by low serum activity of the antioxidant enzyme (SOD).[29]

Table 2.

Indicators of LPO and AOD in blood plasma

Variable	Control group	Study group
CDs, μmol/L	2.32±0.74	1.67±0.71*
KD-CT, units	0.26±0.12	0.43±0.26*
TBA-active products, μmol/L	0.86±0.45	0.97±0.48
TAA, units	15.73±3.52	15.39±4.43
α-tocopherol, μmol/L	8.15±2.8	5.75±3.38*
retinol, μmol/L	0.68±0.21	0.45±0.24*
SOD activity, units	1.69±0.1	1.42±0.29*
GSH, mmol/L	2.29±0.22	2.12±0.45
GSSG, mmol/L	1.94±0.22	1.89±0.47

** - P<0.05 vs. the control group*

The AOD system involving special antioxidant enzymes (SOD, catalase, enzymes of the glutathione redox system, water- and fat-soluble vitamins) plays an important role in protecting the body from oxidative damage.[26,30] Dysfunction of the AOD system is characterized by the development of the LPO syndrome and can lead to a number of negative consequences for the cell, such as membrane damage, inactivation or transformation of enzymes, suppression of cell division, and accumulation of inert polymers.[11,26] We found a decrease in the values of α-tocopherol (1.42 times, P=0.0158) and retinol (1.51 times, P=0.0025), and SOD activity (1.19 times, P=0.0001) in the study group compared to the control group (Table 2). There were no statistically significant changes in TAA and glutathione status components.

It has been established that even a slight decrease in SOD activity is an important signal of a metabolic shift towards the prevalence of prooxidant processes. α-tocopherol and retinol are the strongest bioantioxidants and the necessary nutritive factors.[30] Moreover, α-tocopherol shows membrane-protective and antimutagenic activity and is the most important regulator of oxidative homeostasis of cells. The in vitro and in vivo evidence of the function of vitamin E as a peroxyl radical-scavenging antioxidant and inhibitor of lipid peroxidation is presented in study by E.Niki.[31] According to many studies, vitamin A in large doses increased the antioxygenic potential of the tissues, and it was suggested that retinol also might be considered as a potential antioxidant similar to tocopherol. [32-34]

It seems optimal to use OSi because multidirectional changes occur in the LPO-AOD system with the development of various pathological conditions. For that purpose, the formula for OSi calculation was developed in our previous study [23]:

$$OSi=(CDs_i/CDs_n)\times(KD\text{-}CT_i/KD\text{-}CT_n)\times(TBA\text{-}active\ products_i/TBA\text{-}active\text{-}products_n)/(SOD_i/SOD_n)\times(GSH_i/GSH_n)\times(\alpha\text{-}tocopherol_i/\alpha\text{-}tocopherol_n)\times(retinol_i/retinol_n),$$

where i - the indicators levels in obese patients, n - the indicators levels in the control group.

OSi >1 shows the presence of OS. This formula takes into account not only the accumulation of LPO products at various stages, but also the activity of various parts of the AOD system. According to the data received, the OSi level in the group of obese patients increased approximately 7 times, which confirms the results on the development of antioxidant insufficiency in this pathology (Fig.1).

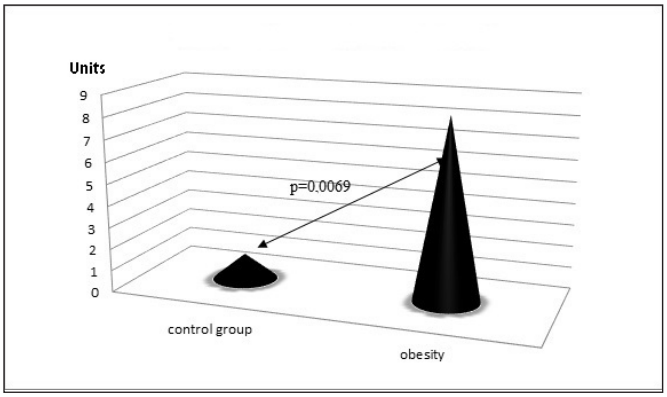

Fig.1. Oxidative stress index (OSi)

Conclusion

Thus, our study showed certain features of the changes in the LPO-AOD system in adolescent boys with obesity: a decrease in levels of primary LPO products and an increase in the level of secondary LPO products on the background of reducing the fat-soluble vitamins and SOD activity. The use of OSi confirmed the presence of antioxidant deficiency in adolescents with obesity.

Competing interests

The authors declare that they have no competing interests.

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