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PREDICTIVE ROLE OF ANALYTICAL INVESTIGATIONS ON THE BIOCHEMICAL HOMEOSTASIS IN YOUTH NOTE I. LIPID METABOLITES RELATED TO SOMATOMETRY

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ABSTRACT: Research concerning the homeostasis of lipid metabolites elicits interest due to its predictive value of the pathobiochemical risk.

In this context the investigations were performed on a number of 423 subjects aged between 10-18 years (174 boys and 248 girls), pursuing the status of lipid metabolites in blood serum, i.e. triacylglycerols and total cholesterol. Also, the analytical data were correlated with the body mass index (BMI) – a somatometric index. Thus, it was possible to identify the overweight subjects (BMI over 95 percentile) as well as the overweight subjects with homeostatic disturbances (dyshomeostasis). As these adolescents might develop cardiovascular and/or other related diseases in adulthood, they were included in a preventive monitoring system.

Keywords: chemical investigations on lipid metabolites in human blood serum – dyshomeostasis, pathobiochemistry

INTRODUCTION

Biochemical homeostasis in human organism is fundamental for the maintenance of its morphophysiological status [1], [2]. Analytical and bio-analytical chemistry offer means for the investigation of various metabolites, e.g.: carbohydrates, lipids, proteins and electrolytes.

In metabollomics theoretical concepts and applicative chemical investigations are more and more developed in relation with biomarkers. In biochemistry and clinical chemistry such markers define the normal functioning of tissues and organs.

Dyshomeostasis is defined as a decrease or increase in the concentration of various metabolites. These changes can occur, among others, as a consequence to malnutrition (lack or excess of nutrients), the presence of different chemical xenobiotics from the environment. [3], [4].

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The problem of lipid metabolites presents interest from different point of view: e.g.: nutritional (hypercaloric foods), medical (pathobio-chemistry of cardiovascular system), social (stress in human activity) and others.

Investigations were performed successively on the concentration of the main serum lipid metabolites: triacylglycerols, total cholesterol and correlated with the body mass index - BMI (Note I); on HDL-cholesterol, LDL-cholesterol and extended to blood metallograms - regarding Ca and Mg (Note II). On the whole, peculiarities of the biochemical homeostasis in young people were monitored.

MATERIALS AND METHODS

Investigations included a group of 423 healthy young people from a school collectivity and is the first study in this domain in Romania. Youth were divided in two groups: group n_1 – including 174 boys and group n_2 – including 249 girls. The age limits of subjects were between 10-18 years which, according to WHO, covers the period of adolescence.

In the first step the weight and height of young people were measured, thus obtaining the somatometric data. Then blood samples were collected for biochemical analysis (lipids, lipoproteins and some metals).

This paper presents the results obtained on serum lipid fractions: triacylglycerols (TAG) and total cholesterol (T-Chol). The analytical results were correlated with the somatometric data expressed in body mass index (BMI) in order to evidence the subjects with risk to develop cardiovascular or other diseases [5], [6].

Analytical determinations were made by using a LABSYSTEM-901 analyzer and specific Clinilab reagents. The method of Fossatti and Prencipe [7] was used for triacylglycerols and the method of Allain et al. [8] for total cholesterol determination.

Serum triacylglycerols determined by the enzymatic spectrophoto-metric method has the following principle: serum triacylglycerols are transformed, in the presence of lipoproteinlipase, into glycerine-3-phosphate. Finally, the measurements were performed at the wavelength 540 nm.

Total cholesterolemia determined by the enzymatic spectrophotometric method was based on the following principle: the transformation of cholesterol ester in cholesterol and fatty acids in the presence of cholester-esterase. Final determinations were made at the wavelength 500 nm.

The analytical data were statistically processed; for each age group the mean value (X) and the standard deviation (SD) were calculated.

RESULTS AND DISCUSSIONS

Blood triacylglycerols result mainly from the catabolism of dietary glycerides and in a more reduced amount from the conversion of carbohydrates and of some aminated acids (in the frame of specific biochemical pathways of the tricarboxylic acid cycle).

The obtained data on triacylglycerolemia are presented in Table 1. These are mentioned for gender groups (noted n_1 the boys and n_2 the girls) and within gender groups for age categories – see Table 1. Comparative data regarding triacylglycerolemia (in mg / dL) in the studied boy and girl groups.

Table 1. Comparative data regarding triacylglycerolemia (in mg/dL) in the studied boy and girl groups

Age groups (years)	Group (boys)		Group (girls)		ΔX (X_1-X_2)
	n_1	$X_1 \pm DS$	n_2	$X_2 \pm DS$	
10	15	66.07 ± 33.92	14	69.50 ± 35.65	+ 3.43
11	23	86.09 ± 37.25	33	78.21 ± 42.28	- 7.88
12	29	76.72 ± 44.14	45	78.82 ± 48.77	+ 2.10
13	35	59.06 ± 25.14	26	87.73 ± 59.48	+ 28.77
14	30	52.83 ± 23.88	36	78.86 ± 45.03	+ 26.03
15	13	62.46 ± 26.41	36	73.77 ± 31.27	+ 11.31
16	18	67.56 ± 23.33	25	72.48 ± 34.32	+ 4.92
17	8	76.75 ± 20.44	16	91.56 ± 35.92	+ 14.81
18	3	80.33 ± 46.48	18	76.22 ± 32.52	- 4.11

According to the method for triacylglycerolemia determination the normal values range between 50-160 mg/dL in boys and between 45-140 mg/dL for girls. In this context the resulted mean values were in normal limits.

As seen in Table 1 the highest values of triacylglycerolemia in boys were found at 11 years age and the lowest at 14 years age. In girls the values were higher than in boys excepting age groups of 11 and 18 yrs.

A similar study was performed in Greece by Schulpis and Karikas [9] on a number of 7767 subjects (3980 boys and 3785 girls) aged between 6-14 years. Their results showed a maximum level for triacylglyceridemia in boys aged 11 yrs and a diminishing to age13 yrs. The same results were revealed in girls with the observation that in the group of 10 yrs and 14 yrs old girls the values were over the normal limit.

Another study performed by Khalil et al. [10] on the serum lipid profile in a number of 410 healthy Indian subjects aged 3-12 years revealed no significant difference between genders. The mean value for triglycerides was 91.1 mg/dL and for total cholesterolemia 134.5 mg/dL. It is to mention that the mean value of triacylglycerols was higher than in our study but total cholesterolemia was much lower.

Bistrizer et al. [11] evaluated the serum lipids profile in 107 children and their parents (40 families) in which the father had a myocardial infarction or coronary heart disease (CHD) before age 40 years. Regarding the results for children, significant hyperlipidaemia were found in 42%.

Studies performed by Tamir et al. [12] and Williams et al. [6], evidenced the complex influence of morphophysiologic factors in adolescents.

The American Heart Association set guidelines for serum triacylglycerol levels (values were expressed in mg/dL), thus: normal < 150 (no risk); borderline high 150-199; high 250-499; very high > 500 (high risk)

In the human organism augmented levels of triacylglycerols in the bloodstream have been linked to atherosclerosis, even in the absence of hypercholesterolemia, and, by extension the risk of heart disease and stroke.

Between the causes of hypertriacylglycerolemia one can mention obesity, high intakes of fat and concentrated sweets, genetic predisposition and others. The values found by us indicate changes related to age and maturation which could reflect the influence of sex hormones on serum lipoprotein metabolism [13], [14].

Homeostasis changes in cholesterolemia originate in the metabolic disturbances of sterides – macronutrients present in diet [15], [16]. In table 2 there are presented the values of the T-Chol in the studied groups.

Normal values according to the used method for cholesterolemia range between 160-199 mg/dL. Thus, in our subjects the mean values did not exceed normal limits, but in girls the values are higher, except for girls aged 14 and 15 years.

The 1987 report of National Cholesterol Education Program, suggest the total serum cholesterol level should be: < 200 mg/dL normal serum cholesterol, 200–239 mg/dL borderline-high, > 240 mg/dL high cholesterol [17]. Nowadays, the link between cholesterol and atherosclerosis is accepted by the majority of medical scientists [18].

Table 2. Comparative data regarding cholesterolemia (in mg/dL) in the studied boy and girl groups

Age groups (years)	Group n_1 (boys)		Group n_2 (girls)		ΔX (X_1-X_2)
	n_i	$X_1 \pm DS$	n_i	$X_2 \pm DS$	
10	15	150.50 ± 24.79	14	159.14 ± 34.87	+ 8.64
11	23	143.60 ± 29.54	33	155.82 ± 30.10	+ 12.22
12	29	147.70 ± 32.35	45	155.27 ± 32.57	+ 7.57
13	35	131.70 ± 20.29	26	141.42 ± 24.08	+ 9.72
14	30	140.50 ± 29.25	36	138.11 ± 20.25	- 2.39
15	13	147.80 ± 27.66	36	143.81 ± 29.18	- 3.99
16	18	136.10 ± 24.36	25	157.40 ± 22.61	+ 21.30
17	8	122.40 ± 26.12	16	146.56 ± 32.25	+ 24.16
18	3	137.70 ± 16.29	18	146.06 ± 35.42	+ 8.36

Investigations concerning cholesterolemia is of great importance because this steroid compound is present in blood, bile and brain tissue. In metabolic processes cholesterol is a precursor in the synthesis of bile acids, steroid hormones and vitamine D [19], [20]. The level of T-Chol in blood serum is associated with metabolic processes and their disturbances in some cardiovascular disease and infections [21]. There are also experimental studies on young subjects [22], [23].

Genetics play an important role in determining adolescents`s cholesterol level. Also, the family lifestyle and diet will have an important effect on an adolescent's cholesterol.

Cholesterol and other cardiovascular risk factors often persist from childhood to adulthood. Autopsy studies demonstrated that aortic and coronary atherosclerosis are commonly seen before age 20 years. High total cholesterolemia, LDL-Chol, VLDL-Chol and low HDL-Chol are correlated with extent of early atherosclerotic lesions in adolescent and young adult [12], [24].

Usually, dyshomeostasis of TAG and T-Chol in youth give evidence of „metabolic syndrome" which develops in time to atherosclerotic processes [25].

Based on somatometric data (weight and height) the body mass index (BMI) - an anthropometric index, defined as the individual`s body weight in kilograms divided by the square of his or her height, were calculated. BMI is widely used to define overweight and obesity, because it correlates well with accurate measures of body fatness.

Using the BMI-for-age gender specific charts recommended by Center for Disease Control and Prevention – Atlanta, USA, the adolescents were classified as: under-weighted - BMI < 5th percentile; normal weighted - BMI between 5 < 85 percentile; at risk of overweight - BMI between 85 < 95 percentile or overweighted - BMI > 95 percentile [26]. The obtained results are presented in Table 3.

Having in view the somatometric criteria, exactly the BMI and assessing the distribution of subjects in the percentile limits, there were revealed 14 boys and 16 girls between 85-95 percentile, and 6 boys and 12 girls over 95 percentile.

Recent studies indicate an increasing prevalence of overweight and obesity in childhood in developed and developing countries, e.g. the prevalence among children and adolescents in USA has doubled in the past two decades [27], [28].

It is known that obesity is associated with significant health problems in childhood and adolescence and is an important early risk factor for much of adult morbidity and mortality [29], [30]. In obese children and adolescents the cardiovascular system is affected (e.g. hypercholes-terolemia, dyslipidemia, hypertension) – see [31], [32]. At the same time, perturbations were found in endocrine system (hyperinsulinism, insulin resistance, impaired glucose tolerance, type 2 diabetes mellitus, menstrual irregularity) and mental health (depression).

Table 3. Distribution of the studied subjects according to percentile (BMI-for-age) evaluation

Age group	Scale of BMI in boys					Scale of BMI in girls				
	< 5	**5-85**	**85-95**	**> 95**	**Total**	**< 5**	**5-85**	**85-95**	**> 95**	**Total**
10	0	12	1	2	**15**	0	13	1	0	**14**
11	2	15	3	3	**23**	1	30	0	2	**33**
12	3	23	3	0	**29**	3	35	6	1	**45**
13	4	28	3	0	**35**	0	24	1	1	**26**
14	4	24	1	1	**30**	1	26	5	4	**36**
15	2	9	2	0	**13**	3	29	2	2	**36**
16	1	16	1	0	**18**	1	22	1	1	**25**
17	1	7	0	0	**8**	1	15	0	0	**16**
18	0	3	0	0	**3**	2	15	0	1	**18**
Total	**17**	**137**	**14**	**6**	**174**	**12**	**209**	**16**	**12**	**249**

Detailed results on TAG and T-Chol determination for overweighted subjects (18 cases) with high risk for “metabolic syndrome” – i.e. category of subjects over 95 percentile of BMI, are presented in Table 4.

Table 4. Serum TAG and T-Chol (mg/dL) in boys and girls with BMI over 95

Gender	Age (years)	Identified subjects		TAG	T-Chol
		No.	**Cases**		
Boys (6)	10	1.	CS	**164**	167
		2.	TD	89	171
	11	3.	BA	**193**	176
		4.	CA	50	137
		5.	MA	83	116
	14	6.	SM	49	136
Girls (12)	11	1.	NI	**191**	117
		2.	CA	**194**	195
	12	3.	TA	56	**211**
	13	4.	CV	**259**	158
	14	5.	BS	**251**	173
		6.	PR	84	153
		7.	PM	68	147
		8.	KA	84	127
	15	9.	RB	101	146
		10.	MI	72	174
	16	11.	TM	48	162
	18	12.	FC	61	**201**

Regarding the obtained analytical results of TAG and T-Chol in overweighted subjects, high values (over the normal limits) were observed in 8 cases (2 boys and 6 girls). In boys 2 cases of hypertriacyl-glycerolemia were evidenced. In girls 4 cases of hypertriacyglicerolemia and 2 cases of hypercholesterolemia were found. The values over the normal limits are evidenced in table 4 by using bold.

Vanhala et al. [33] found that obese children who went on to be obese adults had a threefold increased risk of developing the metabolic syndrome compared with those who became obese as adults.

The probability of childhood obesity persisting into adulthood is estimated to increase from approximately 20% at 4 years of age to approximately 80% by adolescence [34]. In addition, it is probable that comorbidities will persist into adulthood [35].

In light of the literature data, in order to prevent the development of cardiovascular and other related diseases all the young subjects found with BMI over 95 and high values of TAG and T-Chol will be monitored.

CONCLUSIONS

Investigations on some serum lipid fraction by using methods based on bioanalytical chemistry, the values of TAG and T-Chol were determined in 423 adolescents (174 boys and 248 girls) aged 10-18 years. Also, the somatometric index, i.e. body mass index (BMI), for each subject was calculated. Thus, a number of 18 adolescents (6 boys and 12 girls) was detected with BMI over 95 (considered as overweight). This category of young people is exposed to pathobiochemical risk in time.

When correlated the BMI of the 18 overweight adolescents with their analytical findings only a number of 8 subjects were find with hyper-lipidemia: 2 boys and 4 girls with hypertriacylglycerolemia and 2 girls with hypercholesterolemia. The subjects with hyperlipidemia will be included in a monitoring system with a strict treatment (diet and medication) in order to reduce body weight and to decrease TAG and T-Chol values, thus preventing the development of cardiovascular and/or other related diseases.

The identified 10 subjects (4 boys and 6 girls) only with BMI over 95 percentile will be included also in a preventive monitoring program in order to reduce their body weight, e.g. with energy-restricted diet (lowered lipid and carbohydrate nutrients in ratio), increase of physical activity, and others.

Using methods which are specific for analytical chemistry in lipidology and calculations to obtain the somatometric index, permitted the identification of subjects with “metabolic syndrome”. These subjects have a major risk to develop biochemical injuries characteristic to atherosclerosis. In order to prevent adulthood diseases in these subjects they must be monitored.

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