ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN — POLONIA
VOL. LV, 41 SECTIO D 2000

Katedra i Klinika Dermatologii Akademii Medycznej w Lublinie
Chair and Department of Dermatology, Medical University of Lublin

ALDONA PIETRZAK, BARBARA LECEWICZ-TORUŃ, JANUSZ URBAN

Comparison of serum lipid in girls affected with vitiligo and control group

Porównanie lipidów surowicy krwi u dziewczynek z vitiligo i z grupy kontrolnej

Vitiligo occurs worldwide in about 1% of the population, mostly between the ages of 10-30 years, as often in males as in females. The cause is unknown, but it might involve genetic factors, autoimmunity, toxic metabolites, and/or a higher vulnerability of melanocytes (7).

There are new reports on the etiology of vitiligo. Most papers deal with immunological theory, some contain information about biochemical disorders. Only single reports deal with disorders of lipid metabolism or some of its parameters; however, no significant deviations have been stated. Picardo et al. (5) suggested that abnormal release of catecholamines from autonomic nerve endings might play an etiological role in the onset and development of vitiligo through an overproduction of toxic (oxy)radicals in the microenvironment of melanocytes in the affected areas. Next they investigated whether this suggested increase in radicals might be associated with an oxidative stress in the blood of vitiligo. Their results show that blood levels of vitamin E, superoxide dismutase SOD, glutathione GSH, glutathione peroxidase GSH-Px activity, lipoperoxides LIP and polyunsaturated fatty acids of phospholipids PL-FA in patients affected with different forms of active vitiligo were not significantly different from those in healthy age matched controls, indicating that melanocyte damage in vitiligo is not linked with a generalized oxidative stress.

In our study we dealt at first with the behaviour of lipids of blood serum and with morphometric measurements of the abdominal cavity organs in ill children with psoriasis. Because we had problems with the selection of a suitably large age- and sex-matched control group, we decided to collect blood and measure organs only in children with vitiligo supposing that they should not differ from those in healthy children as regards

lipids. Because the research prolonged, we finally formed a control group of healthy children with a mild form of pityriasis capitis. A statistical analysis showed that there are statistical differences in the group of ill children with vitiligo and this group should be analysed separately. Therefore, we made a separate statistical analysis for the group of girls suffering from vitiligo.

MATERIAL AND METHODS

This study was conducted by the Pediatric Ward of the Clinic of Dermatology in Lublin. Eleven white girls (aged 7–15 years) with clinical and/or histopathologic diagnoses of vitiligo were evaluated; in mean age 10.8±2.0 years, the mean age at onset was 7.2 years. The children were compared with a control group of 19 girls in mean age 10.5±2.2 years. The enrollment excluded patients with disseminated vitiligo. The characteristics of the examined group are placed in Table 1. The type of vitiligo was type "A". The skin lesions covered the skin of the trunk and/or limbs and they did not affect more than 25% – 30% surface of the whole body.

In additional examinations there were found no deviations in the analysed population, there were no cases of hepatitis or any contagious diseases or infectious ones which might affect the immunological system and none of the children suffered from organic diseases, including endocrinological ones. The children were not previously treated with general or skin drugs. Prior to the examination (7 days) the children were either on a diet in hospital or on a low-fat diet at home. The blood was collected after overnight fasting. Children with a similar coefficient of body mass index were chosen for the study.

We examined the following lipid serum parameters: activity of EC 3.1.1.3 lipase, total cholesterol, HDL cholesterol, LDL cholesterol, total phospholipids, HDL phospholipids, LDL phospholipids, triglycerides, and all other routine parameters. Subjects with heart diseases, arterial hypertension, diabetes, thyroid gland disorders or any other organic diseases were excluded from the study.

Concentrations of lipid profile were assayed using the ready-made reagent kits by bio-Mérieux, France: LDL Cholesterol/Phospholipides; HDL Cholesterol/Phospholipides; Cholesterol enzymatique PAP; Phospholipides enzymatiques PAP; Triglycerides enzymatiques UV 250, in accordance with the producer's instructions. Methodological recommendations supplied by the manufacturer were stricly followed. In statistical analysis of the results we used Mann-Whitney U Test.

Table 1. Comparison of girls with vitiligo with control group♀

Variable	group	n	Mean	Minimum	Maximum	SD	SE	p
Age	C	19	10.05	7.00	15.00	2.25	0.51	>0.2
Age	V	11	10.91	7.00	15.00	2.38	0.72	
Weight	C	19	35.29	19.50	75.00	14.65	3.36	>0.4
Weight	V	11	36.32	21.50	50.00	9.19	2.77	
Height	C	19	138.95	117.00	167.00	16.98	3.89	>0.6
Height	V	11	142.64	115.00	170.00	17.17	5.18	
BMI	C	19	17.51	13.54	28.58	3.34	0.76	>0.7
BMI	V	11	17.62	15.20	21.53	1.96	0.59	
TC	C	19	167.95	117.60	187.60	19.91	4.57	>0.8
TC	V	11	169.30	125.80	191.60	19.52	5.88	
HDLC	C	19	54.42	38.70	75.50	7.99	1.83	<0.03*
HDLC	V	11	47.93	35.30	68.30	9.40	2.83	
LDLC	C	19	98.21	64.00	123.40	16.68	3.83	>0.2
LDLC	V	11	105.80	70.80	126.60	16.92	5.10	
TPh	C	19	175.04	140.40	212.00	20.21	4.63	>0.8
TPh	V	11	171.79	137.60	203.20	18.27	5.50	
HDLPh	C	19	82.30	63.20	102.30	11.11	2.55	>0.08
HDLPh	V	11	76.57	59.50	100.20	11.48	3.46	
LDLPh	C	19	77.08	54.60	111.00	12.87	2.95	>0.6
LDLPh	V	11	76.22	51.40	103.70	15.61	4.70	
TG	C	19	93.30	65.60	145.30	21.03	4.82	<0.001*
TG	V	11	121.34	101.60	140.00	13.72	4.14	

C – controls, V – vitiligo, N – number, SD – standard deviation, SE – standard error, p – probability

RESULTS

The results of the study are presented in Table 1. In the upper part there is a characteristics of the examined groups. No statistical differences as regards age, weight, height and BMI in the examined groups were stated (with $p>0.2$, $p>0.4$, $p>0.6$, $p>0.7$ respectively). No statistically significant differences in the concentrations of total cholesterol (TC), total phospholipids (TPh), fraction LDL phospholipids (LDLPh) as well as fraction LDL cholesterol in the examined groups were stated. However, there were slightly higher concentrations here than in controls (105.8 mg% vs 98.21mg%), insignificant statistically though ($p>0.2$). The concentrations of HDL cholesterol were lower in girls with vitiligo in relation to the concentrations in the control group (47.93 ± 9.40vs 54.42 ± 7.99mg%) and the difference was statistically significant ($p<0.03$). In girls with vitiligo the concentrations of HDL phospholipids (HDLPh) tended to be lower in relation to the control group. However, they were not statistically significant (76.57 ± 11.48 vs 82.30 ± 11.11mg%, $p>0.08$). Instead, the concentration of blood serum triglycerides in ill children in relation to the concentration in the control group of healthy girls (121.34 ± 13.72mg%), the increase was highly statistically significant and it was $p<0.001$).

DISCUSSION

Although it is known that vitiligo is a hypopigmentary dermatosis of a probable autoimmune origin, there are also other theories on the origin of this disease considered. Other scientists, like Cucchi et al. (2) stated an increase in the concentration of catecholamine and its derivatives in patients with vitiligo. They measured the plasma levels of the following substances in 35 healthy subjects and in 70 patients suffering from nonsegmental vitiligo at different stages of the disease: catecholamines [norepinephrine (AD), epinephrine (E) and dopamine (DA)], their precursor/3,4-dihydroxyphenylalanine (DOPA), their metabolites [3-methoxy-4-hydroxyphenylglycol (MHPG), normetanephrine (NMN), metanephrine (MN) and homovanillic acid (HVA)] and 5-hydroxyindolacetic acid (5-HIAA) as the major metabolite of serotonin.They found out that the levels of norepinephrine AD, epinephrine E, normetanephrine NMN, metanephrine MN, homovanillic acid HVA and 5-hydroxyindolacetic acid 5-HIAA were significantly higher in patients compared to controls. Concentrations of parameters varied depending on whether the form of vitiligo was active or progressive. The higher catecholamine and metabolite levels in the early phase of the disease may reflect an increased activity by monoaminergic systems, probably due to stressful events, including the onset of vitiligo itself. Picardo et al. (5) previously reported that patients with active vitiligo (AVP) have elevated urinary levels of catecholamine metabolites, such as homovanillic and vanilmandelic acids, irrespective of the form of the disease (acrofacial, segmental, gener-

alized). The autotoxic theory states that an increased activity of melanocytes leads to their self-destruction. Autotoxicity can be caused by inhibition of thioredoxine reductase removing free radicals which is located in the membrane of melanocytes (3). This enzyme is inhibited by calcium in the cellular membrane and its concentration on keratinocytes in patients with vitiligo is greater than in the control group. The elevated concentration of extracellular calcium causes an increase in the number of peroxidate radicals which cause an inhibition of tyrosinase by disturbance of equilibrium between the processes of oxygenation and of thioredoxin reductase in epidermis (3). Elevated H_2O_2 levels can be demonstrated *in vivo* in patients compared with healthy controls (6). H_2O_2 accumulation is associated with low epidermal catalase levels (6). So far, four potential sources for epidermal H_2O_2 generation in vitiligo have been identified: (a) perturbed (6R)-L-erythro 5, 6, 7, 8 tetrahydrobiopterin (6BH4) *de novo* synthesis /recycling/ regulation; (b) impaired catecholamine synthesis with increased monoamineoxidase A activities; (c) low glutathione peroxidase activities; and (d) "oxygen burst" via NADPH oxidase from a cellular infiltrate.

H_2O_2 overproduction can cause inactivation of catalase as well as vacuolation in epidermal melanocytes and keratinocytes. The next phase after a cell vacuolation is its death (3). Vacuolation was also observed *in vitro* in melanocytes established from lesional and nonlesional epidermis of patients (n = 10) but it was reversible upon an addition of catalase. Schallreuter et al. (6) stated in his conclusion that there are several lines of evidence that the entire epidermis of patients with vitiligo is involved in the disease process and that the correction of the epidermal redox status is mandatory for repigmentation (1). It has recently been shown that patients with vitiligo can accumulate epidermal hydrogen peroxide (H_2O_2) in association with low catalase levels. This study examined serum selenium levels and blood glutathioneperoxidase activities in 61 patients and controls. The results showed high serum selenium levels in 56% of the patients. As at least one isoform of glutathioneperoxidase requires selenium for its activity, enzyme activities were also evaluated. As glutathione peroxidase can also efficiently degrade H_2O_2, the results of this study could indicate an additional impaired H_2O_2 metabolism in vitiligo (1). These processes may secondarily influence the concentration of numerous substances in blood serum, including lipids. The results we obtained indicate the existence of certain abberations in lipid metabolism in vitiligo. This may suggest the existence of certain disorders in liver cells where a part of lipid metabolism takes place. There are reports on the disorders of liver activity in the course of vitiligo. However, these disorders did not influence the size of liver in vitiligo patients examined by means of ultrasonography as we showed in another paper. Picardo et al. (5) did not state any abnormalities in other parameters of lipid metabolism in vitiligo patients in comparison with controls. Our results require further research on other lipids in vitiligo patients.

REFERENCES

1. Beazley W. D. et al.: Serum selenium levels and blood glutathione peroxidase activities in vitiligo. Br. J. Dermatol., 141, 301,1999.
2. Cucchi M. L. et al.: Higher plasma catecholamine and metabolite levels in the early phase of nonsegmental vitiligo. Pigment. Cell Res., 13, 28, 2000.
3. Kovacs S. O.: Vitiligo [see comments]. J. Am. Acad. Dermatol., 38, 647, 1998.
4. Mahmoud F. et al.: Peripheral T-cell activation in non-segmental vitiligo. J. Dermatol. 25, 637, 1998.
5. Picardo M. et al.: Antioxidant status in the blood of patients with active vitiligo. Pigment. Cell. Res., 7, 110, 1994.
6. Schallreuter K. U. et al.: *In vivo* and *in vitro* evidence for hydrogen peroxide (H_2O_2) accumulation in the epidermis of patients with vitiligo and its successful removal by a UVB-activated pseudocatalase. J. Investig. Dermatol. Symp. Proc., 4, 91, 1999.
7. Westerhof W. Vitiligo. Management Update. Skin Therapy Lett., 5, 1, 2000.

Otrz.: 2000.06.28

STRESZCZENIE

Przeprowadzono badania stężenia lipidów surowicy krwi u 11 dziewczynek chorych na bielactwo nabyte. Wyniki badań porównano z wynikami grupy kontrolnej złożonej z 19 zdrowych dziewczynek dobranych wiekiem i indeksem BMI. Stwierdzono obniżenie stężenia cholesterolu frakcji HDL u dziewcząt z bielactwem, obniżenie to było istotne statystycznie. Obserwowano jednoczesną tendencję do obniżenia stężenia fosfolipidów frakcji HDL. U chorych dziewcząt występowało również istotne statystycznie podwyższenie stężenia trójglicerydów w surowicy krwi. Występowała także tendencja do podwyższenia stężenia cholesterolu frakcji LDL.