POLYMORPHISMS OF SCD-1 GENE, INCREASED OXIDATIVE STRESS AND INSULIN RESISTANCE IN PERSONS WITH ELEVATED CONCENTRATIONS OF APOLIPOPROTEIN B-48

B. STAŇKOVÁ, J. MACÁŠEK, M. ZEMAN, M. VECKA, E. TVRZICKÁ, L. VÁVROVÁ, J. RYCHLÍKOVÁ, M. JÁCHYMOVÁ, A. SLABÝ, A. ŽÁK

IVth Department of Medicine and IMBLD, 1st Faculty of Medicine, Charles University and GUH in Prague, Czech Republic

Introduction

Increased concentrations of apoprotein (apo) B-48, which is a specific structural component of chylomicrons and chylomicron remnants, represent a risk factor for cardiovascular disease (Mori 2013, Valdivielso 2010) that is independent of conventional risk factors. High apo B-48 levels are a marker for clinical complications of atherosclerosis and probably have proatherogenic potential linked to prolonged presence of triglyceride rich lipoproteins in the circulation that is connected with subsequent formation of sdLDL and oxLDL. It was proved that apoB-48 is a risk factor for clinically manifested or subclinical atherosclerosis in diabetic as well as in nondiabetic population and associated with endothelial dysfunction. Furthermore, increased concentrations of apo B-48 were observed in diabetic nephropathy, end stage renal disease, obesity, metabolic syndrome (MS), and some hyperlipidemic phenotypes.

A key enzyme of fatty acid (FA) metabolism associated with cardiometabolic risk is stearoyl-CoA desaturase (SCD-1, D9D). SCD-1 is the rate-limiting enzyme that catalyzes the conversion of SFA to MUFAs. Activity of SCD-1 is influenced by genetic, exogenous and metabolic factors (Merino 2011, Mauvoisin 2011). Higher activity of SCD-1 are connected with dyslipidemia, ischaemic heart diseases and MS. The minor alleles are supposed to be associated with lower activity of SCD-1.

Aim of study

The aim of the study was to analyze associations of clinical, laboratory and genetic parameters with concentrations of apoB-48 in patients at high cardiometabolic risk. A cut-off value for apoB-48 concentration was set at 8.1 mg/L, representing 90th percentile of control group and being in concordance with the literature data (Nakajima 2014).

Materials and methods

Subjects. 220 probands with at high cardiometabolic risk. Metabolic syndrome (MS) was diagnosed according to IDF criteria (IDF 2005) and persons included into the study were further stratified into groups according to the total number of present MS component.

Biochemical parameters. Blood samples were collected from an antecubital vein into Vacutainer® tubes containing EDTA after 12-h overnight fast. The standard enzymatic-colorimetric procedures were used for routine biochemical parameters.

Specialized parameters: apo B-48, oxidized LDL, hsCRP - ELISA commercial kits
LDL subfractions - gel electrophoresis with Lipoprint® LDL System
conjugated dienes in LDL - spectrophotometric method (Ahotupa 1996)
fatty acid profile – TLC with GC-FID (Tvrzická 2002)

Genetic analyses: DNA was isolated from the peripheral blood leukocytes with modified salting out procedure. Genetic analyses were performed by PCR – RFLP or direct sequencing methods.

Biosynthesis of nonessential fatty acids

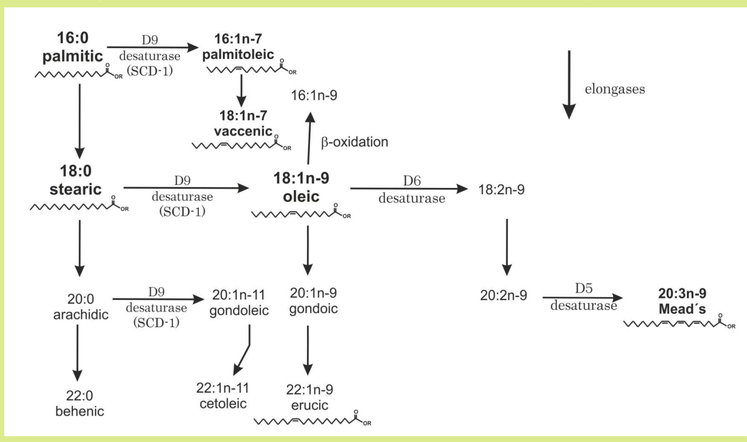


Table 1 Clinical characteristics of groups

	Apo B-48 < 8.1 (mg/L) n = 93	Apo B-48 ≥ 8.1 (mg/L) n = 127	p value (t-test)
Sex (male/female)	(35/58)	(69/58)	0.021[a]
Age (years)	50.1 ± 15.2	52.4 ± 13.8	0.259
Probands with MSn≥3 (%)	32 (34.4)	70 (55.1)	0.004[a]
BMI (kg/m^2)	28.5 ± 5.4	29.4 ± 5.9	0.294
Waist circumference (cm)	94.3 ± 15.8	98.7 ± 14.7	0.876
Fat mass (kg)	33.4 ± 10.6	32.5 ± 11.0	0.566
SBP (mm Hg)	130 ± 14	135 ± 19	0.015
DBP (mm Hg)	87 ± 10	87 ± 11	0.660

Values are expressed as the mean ± SD, [a/] χ^2 test

Results

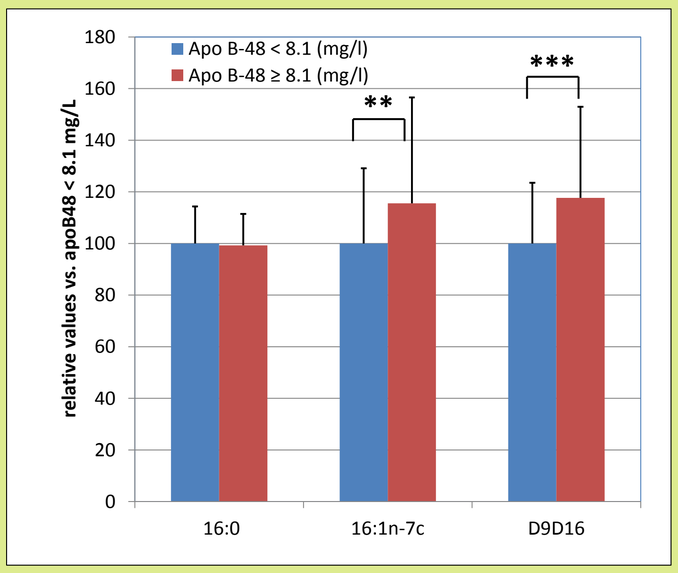
Table 2 Laboratory parameters

	Apo B-48 < 8.1 (mg/L)	Apo B-48 ≥ 8.1 (mg/L)	p value (t-test)
TC (mmol/L)	4.99 ± 1.02	5.75 ± 1.32	0.001
TG (mmol/L)	0.97 (0.72 – 1.28)	1.78 (1.29 – 2.51)	0.001[b]
HDL-C (mmol/L)	1.57 ± 0.46	1.40 ± 0.39	0.003
LDL-C (mmol/L)	2.93 ± 0.87	3.41 ± 1.06	0.001
nonHDL-C (mmol/L)	3.42 ± 0.96	4.35 ± 1.20	0.001
Apo A1 (g/L)	1.50 ± 0.33	1.41 ± 0.28	0.047
Apo B-100 (g/L)	1.04 ± 0.29	1.27 ± 0.34	0.001
Apo B-48 (mg/L)	4.3 (2.9 – 6.3)	22.8 (12.5 – 36.7)	0.001[b]
sd-LDL-C (mg/dL)	2.0 (0.0 – 4.0)	5.0 (2.0 – 17.0)	0.001[b]
LDL size phenotype B n (%)	16 (17.2)	58 (45.7)	0.001[a]
NEFA (mmol/L)	0.55 (0.37 – 0.71)	0.51 (0.38 – 0.70)	0.394[b]
Glucose (mmol/L)	5.09 ± 0.67	5.18 ± 0.64	0.310
HOMA-IR (index)	1.67 (1.31– 3.18)	2.32 (1.53–3.64)	0.013[b]
Insulin (μU/mL)	7.79 (6.09–13.53)	10.10 (7.07–15.22)	0.024[b]
KD (μmol/L)	60.5 ± 15.4	67.2 ± 16.8	0.003
oxLDL (U/L)	31.7 (17.8 – 55.0)	54.5 (29.3 – 70.8)	0.001[b]
hsCRP (mg/L)	2.22 (0.92 – 4.24)	3.08 (1.16 – 4.05)	0.671
Uric acid (μmol/L)	288 ± 79	337 ± 91	0.001

Values are expressed as the mean ± SD or median (25th – 75th percentile)
[a/] χ^2 test; [b/] Mann-Whitney U-test

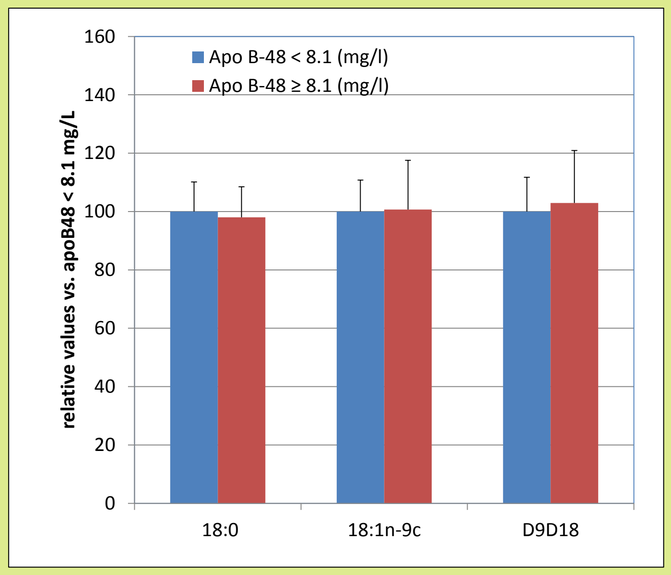
Fatty acid analyses in plasma phosphatidylcholines

Graph 1
16 carbon fatty acids



** - p < 0.01, *** - p < 0.001 (t-test); D9D16 = 16:0/16:1n-7c

Graph 2
18 carbon fatty acids



D9D18 = 18:0/18:1n-9c

Table 3 SCD1 Genotype and allele distributions

Polymorphism	Genotype				Allele			HWE (all p NS)	Comparison of groups (* - p < 0.05)
rs2167444	TT	TA	AA	total	T	A	total		
Apo B-48 < 8.1 mg/L	57	31	5	93	145	41	186	χ^2=0.084	χ^2=8.732* (genotype)
Apo B-48 ≥ 8.1 mg/L	93	34	0	127	220	34	254	χ^2=3.033	χ^2=5.095* (alleles)
rs508384	CC	AC	AA	total	C	A	total		
Apo B-48 < 8.1 mg/L	56	32	5	93	144	42	186	χ^2=0.023	χ^2=8.848* (genotype)
Apo B-48 ≥ 8.1 mg/L	92	35	0	127	219	35	254	χ^2=3.244	χ^2=5.167* (alleles)

Conclusion

1. Our results support the importance of increased concentrations of apo B-48 (above 8.1 mg/L) as an indicator of higher cardiometabolic risk. Higher concentrations of apo B-48 are associated with phenotype B of LDL particle size, increased oxidative stress, abdominal distribution of adipose tissue, and with insulin resistance.
2. In plasma phosphatidylcholines, we found higher index of delta-9 desaturase (SCD-1) for 16:0.
3. We observed different genotype and allele frequencies in groups stratified according to the cut-off value of apoB-48. Lower frequency of minor alleles for SCD-1 polymorphisms (rs2167444, rs508384) in patients with higher concentrations of apo B-48 implies a role of genetic factors in pathogenesis of MS in patients with high concentrations of apo B-48.

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