We are IntechOpen,
the world's leading publisher of
Open Access books
Built by scientists, for scientists

6,900
Open access books available

185,000
International authors and editors

200M
Downloads

154
Countries delivered to

Our authors are among the

TOP 1%
most cited scientists

12.2%
Contributors from top 500 universities

BOOK CITATION INDEX
CLARIVATE ANALYTICS
INDEXED

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

The Assessment of the Atherogenic Lipoprotein Profile in Cardiovascular Diseases by Lipoprint System Analysis

Stanislav Oravec, Kristina Gruber, Andrej Dukat,
Peter Gavornik, Ludovit Gaspar and Elisabeth Dostal

Additional information is available at the end of the chapter

Abstract

Research focus: Identification of incidence of an atherogenic lipoprotein pheno-type B in four representative diagnoses of cardiovascular diseases: a) arterial hypertension, b) coronary heart disease, c) lower extremity arterial disease, d) ischemic stroke Research methods: A clinical study included 366 patients with a diagnosis of arterial hypertension (n=107), coronary heart disease (n= 104), lower extremity arterial disease (n= 100) and ischemic stroke (n= 55). The control group consisted of 150 healthy normotensive and normolipemic volunteers, all non-smokers, without signs of cardiovascular disease. In all tested individuals (or subjects) lipid parameters in serum: cholesterol and triglycerides were analyzed, using the enzymatic CHOD-PAP method, Roche Diagnostics Germany. Lipopro-teins in serum lipoprotein spectrum by Lipoprint LDL system were analyzed and an atherogenic and a non-atherogenic lipoprotein profile idetified. The Score of the Anti-Atherogenic Risk (SAAR) was calculated as the ratio between non-atherogen-ic and atherogenic lipoproteins. Results: More than 80 percent of tested patients with cardovascular diseases have an atherogenic lipoprotein profile, with a high level of strongly atherogenic small dense LDL. The aterogenic profile was found in arterial hypertension 78.5%, in coronary heart disease in 81.7%, in lower extremity arterial disease in 80 %, and in patients who survived an ischemic stroke in 85%. Main conclusion: The atherogenic lipoprotein profile was found to be the overwhelming lipoprotein profile in tested cardiovascular diseases A new phenomenon- atherogenic normolipidemia - as a risk factor for the development

of cardiovascular disease, would be established as a new term used in the diagnostics of dyslipoproteinemias

Keywords: atherogenic lipoproteins, atherogenic lipoprotein profile, small dense LDL, cardiovascular diseases

---

1. Introduction

In the last few decades, lipoprotein research has focused on the phenomenon of atherogenic and non-atherogenic lipoproteins, specifically, atherogenic and non-atherogenic lipoprotein profiles phenotype A and phenotype B [7, 18, 60] after it was reported that more than 75% of patients with an acute coronary syndrome or myocardial infarction had normal plasma values of cholesterol, Low Density Lipoprotein cholesterol (LDL cholesterol) and High Density Lipoprotein cholesterol (HDL cholesterol) [15 - 17].

Thus, it was necessary to look for other risks factors in plasma, the presence of which in relevant quantities could cause damage to endothelial cells and resultant endothelial dysfunction [59]. This called into question whether an increased total cholesterol level, or increased LDL-cholesterol, as a criterion for the degree of atherogenic risk, provided a universal explanation for the origin of atherogenesis. A reasonable explanation was found in atherogenic lipoprotein subpopulations, the presence of which in plasma, even in very low concentrations, could impair the integrity of the vessel wall and lead to endothelial dysfunction with its fatal consequences [Table 1]: formation of atherothrombotic plaques, acute myocardial infarction, ischemic stroke, or sudden death [39, 59, 64].

The predominance of atherogenic lipoproteins in plasma is characteristic for the atherogenic lipoprotein spectrum, phenotype B. When present in plasma in high concentrations, these lipoproteins contribute to ischemic vascular impairment [6, 8, 57, 60]. The process of degen-erative changes in vessels results in the formation of atheromatous vascular plaques. These later play an important role in the formation of stable or unstable angina (pectoris), and critical ischemia of peripheral and/or cerebral arteries as well [39,56]. When atherogenic lipoproteins in plasma are present in small quantities, we obtain a picture of a non-atherogenic lipoprotein profile, phenotype A.

Various methods have been developed (gradient gel electrophoresis, ultracentrifugation, magnetic resonance spectroscopy, endothelial models for testing lipoprotein cytotoxicity) to identify atherogenic lipoproteins [2, 26, 45, 48], but because of technical and financial issues, long-term analyses and high operating costs, the previously mentioned methods were used primarily in basic research. Simple analytical procedures for routine distribution were lacking and the possibility of their implementation in every day laboratory practice remained limited.

An electrophoretic method by which to separate lipoproteins on polyacrylamide gel (PAG) with the use of Lipoprint LDL System [29, 41] has become a milestone in routine laboratory

analysis and in diagnosing metabolism disorders of lipoproteins. It enables the analysis of 12 lipoprotein subfractions: VLDL, IDL 1-3, LDL 1-7, and HDL.

1. The Lipoprint LDL system identifies and quantifies

    a. Atherogenic lipoproteins (VLDL, IDL1, IDL2, and LDL3-7, so-called small dense LDL)

    b. Non-atherogenic lipoprotein entities (IDL3, HDL)

    c. Lipoproteins with uncertain atherogenicity (LDL1, LDL2)

2. And determines

    a. The atherogenic vs. non-atherogenic lipoprotein spectrum, phenotype B vs. phenotype A

Atherogenic lipoprotein spectrums are characterized according to the predominance of atherogenic lipoproteins: very low density (VLDL); intermediate density IDL1 and IDL2; and by the presence of small dense-low density lipoproteins (sd-LDL). The last represented small dense LDL are highly atherogenic LDL subfractions that form fractions LDL3-7. As the name implies, they are smaller than the other types of LDL with a diameter < 26.5 nm (265 Angströms) and they float within the density range of 1.048–1.065 g/ml, that is, higher than LDL1 and LDL2. On the separating polyacrylamide gel (PAG) sd-LDL are detected as subtle bands on the anodic end of the gel, right behind HDL, that migrate to the head of separated lipoproteins.

Small dense LDL are highly atherogenic for [11, 14, 46]:		
* Low recognition by LDL-receptors (configuration alterations Apo B)	→	
* Enhanced aptitude for oxidation and acetylation	→	
* Oxide-LDL	→ release of pro-inflammatory cytokines → muscle cell apoptosis	
* Oxide-LDL	→ release of metalloproteinase → collagen degradation	
* Oxide-LDL	→ enhanced aptitude for trapping by macrophages (scavenger-receptors) → stimulation of foam cell formation	
* Easier penetration into the subendothelial space and formation of cholesterol deposits		

Table 1. Atherogenicity of small dense LDL

In our studies were analyzed serum lipoprotein spectrums in patients with newly recognized a) arterial hypertension, b) coronary heart disease, c) lower extremity arterial disease, and d) in patients who survived a stroke. As mentioned earlier, an analytical method for a quantitative evaluation of lipoprotein fractions was used, and the incidence of an atherogenic lipoprotein

spectrum phenotype B (vs. phenotype A) in these four representatives of cardiovascular diseases was identified. At the same time, a lipoprotein spectrum of a control group of healthy individuals was examined and tested for the incidence of phenotype B.

2. Arterial hypertension

Arterial hypertension (AH) (Fig. 3) is one of the most serious cardiovascular diseases. More than 20% of the adult population suffers from this disease. AH is one of the risk factors for atherosclerosis development of coronary, brain, and peripheral arteries, together with the main cardinal risk factors, that is, dyslipoproteinemia and tobacco smoking [14,20,38]. Atherogenic LDL subfractions also play a role in the development of the arterial hypertension [32, 43].

AH is a permanent, long-lasting increase in blood pressure of more than 140/90 mmHg in people of middle age. In people older than 70 years of age, values higher than 160/95 mmHg are considered increased. For more extensive guidelines see the Statement of WHO/ISH (International Society of Hypertension) on the management of hypertension [61].

Dyslipoproteinemia, which frequently accompanies AH and multiplies the risk of athero-sclerosis development, can also be considered one of the multiple sources that give rise to AH [35, 64].

Atherogenic lipoproteins in plasma cause endothelial dysfunction, increase vessel tone, and support the development of AH, which terminates in organ ischemia [8,50,51,55,57,59].

2.1. Patients

In our study 107 patients with newly diagnosed arterial hypertension were examined. Repeated blood pressure (BP) examination confirmed an increased blood pressure more than 150 mmHg for systolic and more than 90 mmHg for diastolic blood pressure in all hypertensive patients. Average systolic blood pressure was 172 ±19 mmHg and average diastolic blood pressure was 102 ±10 mmHg. The group of hypertensive patients comprised 66 men and 41 women. The average age of the men was 50 ± 17.6 years and the average age of the women was 51.0 ± 13.4 years.

The control group consisted of 150 healthy normotensive and normolipemic volunteers, all non-smokers, without signs of cardiovascular disease and without biochemical signs of lipid metabolism disorders. The average age of the subjects was 21 years, and the control group involved 50 males and 100 females. Volunteers were recruited from medical students at the Medical Faculty, who gave written, informed consent, and the study was approved by the local ethics committee.

2.2. Methods

A blood sample from an antecubital vein was obtained in the morning after a 12-hour fasting period. Total cholesterol and triglycerides in serum were analyzed from lipid parameters,

using the enzymatic CHOD-PAP method, Roche Diagnostics Germany. To determine the non-atherogenic lipoprotein phenotype A and the atherogenic lipoprotein phenotype B, the Lipoprint LDL System Quantimetrix CA, USA, was used.

The Score of the Anti-Atherogenic Risk (SAAR) was calculated as the ratio between non-atherogenic and atherogenic lipoproteins in serum [42]. SAAR values over 10.8 represented a non-atherogenic lipoprotein profile, whereas values under 9.8 represented an atherogenic lipoprotein profile. The cut off values for a non-atherogenic lipoprotein profile and an atherogenic lipoprotein profile were calculated from the results of 940 Lipoprint LDL analyses. Using the Quantimetrix Lipoprint LDL system interpretation, all 940 individuals were examined (general group of subjects) and tested for the occurrence of atherogenic vs. non-atherogenic lipoprotein profile and were divided into the two subgroups of subjects with an LDL profile:

• Indicative of Type A, that is, a non-atherogenic lipoprotein profile phenotype A

• Not indicative of Type A, that is, an atherogenic lipoprotein profile, phenotype B [29]

Statistical evaluation of obtained values was performed with an unpaired student's t-test. The level of significance was accepted at $p < 0.05$.

2.3. Results

In the control group shown in Table 2, along with the individuals with non-atherogenic normolipidemia, that is, an ideal lipoprotein profile (Fig. 1), a subgroup of normolipidemic individuals with an atherogenic lipoprotein profile was also identified. This group represented people with an atherogenic normolipidemia (Fig. 2). These people are clinically healthy, without clinical or laboratory signs of cardiovascular diseases, but with a positive familial history for cardiovascular diseases (myocardial infarction) in the parents' or grandparents' generation. The triglycerides and LDL3-7 concentrations in the control group with the atherogenic profile, compared to the individuals with a non-atherogenic lipoprotein profile, were increased ($p < 0.05$, respectively, $p < 0.0001$). The Score of the Anti-Atherogenic Risk (SAAR) for a non-atherogenic lipoprotein profile is a sensitive indicator by which to differentiate between an atherogenic and non-atherogenic plasma lipoprotein constellation (non-atherogenic vs. atherogenic: $p < 0.0001$).

	Chol	TAG	VLDL	LDL1, 2	LDL3-7	LDL	HDL	Score
	(mmol/l±SD)							
Control	4.28	1.15	0.60	1.29	0.03	2.31	1.35	37.8
	±0.60	±0.39	±0.16	±0.38	±0.003	±0.53	±0.32	±19.7
(non atherogenic profile n =140)								
Control	4.25	1.44	0.68	1.16	0.22	2.24	1.32	6.0
	±0.54	±0.40	±0.14	±0.24	±0.08	±0.36	±0.31	±2.0

	Chol	TAG	VLDL	LDL1, 2	LDL3-7	LDL	HDL	Score
	(mmol/l±SD)							
(atherogenic profile n = 10)								
Control	4.27	1.17	0.61	1.28	0.04	2.30	1.34	35.8
	±0.60	±0.39	±0.16	±0.37	±0.004	±0.52	±0.32	±18.5
(total number n=150)								
Non-atherogenic vs. atherogenic								
		p<0.05			p< 0.0001			p< 0.0001

Non-atherogenic profile, 93.4 % vs. atherogenic profile, 6.6 %, in control group

Table 2. Serum concentration of lipids, lipoproteins, and SAAR-score in the control group
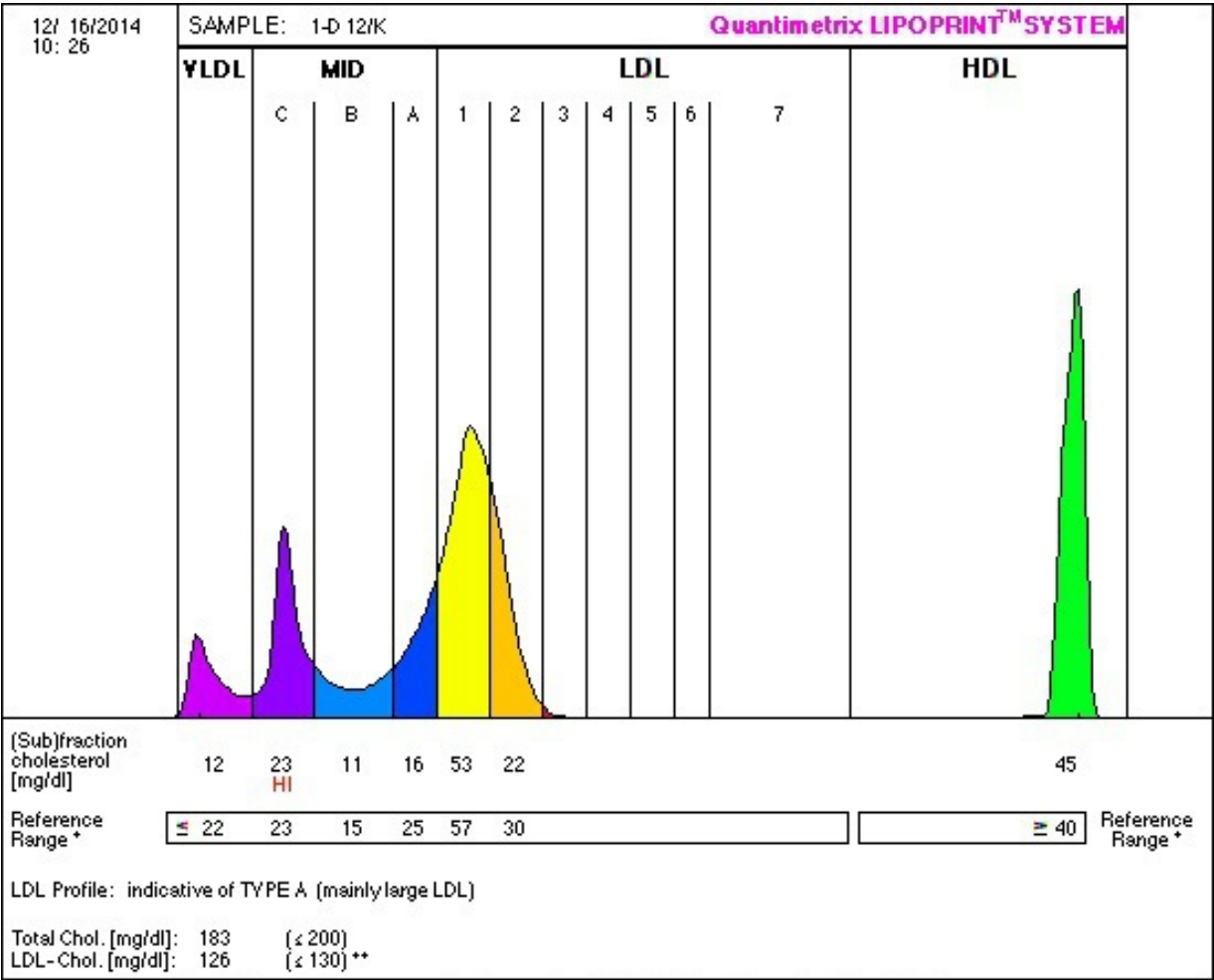
*Reference ranges derived from 125 serum samples that met the NCEP ATPIII guidelines for desirable lipid status
**LDL-C comprised of the sum of cholesterol in Md bands C through A as well as all the subfractions

Figure 1. Non-atherogenic normolipidemia – Control group, SAAR score: 62.5
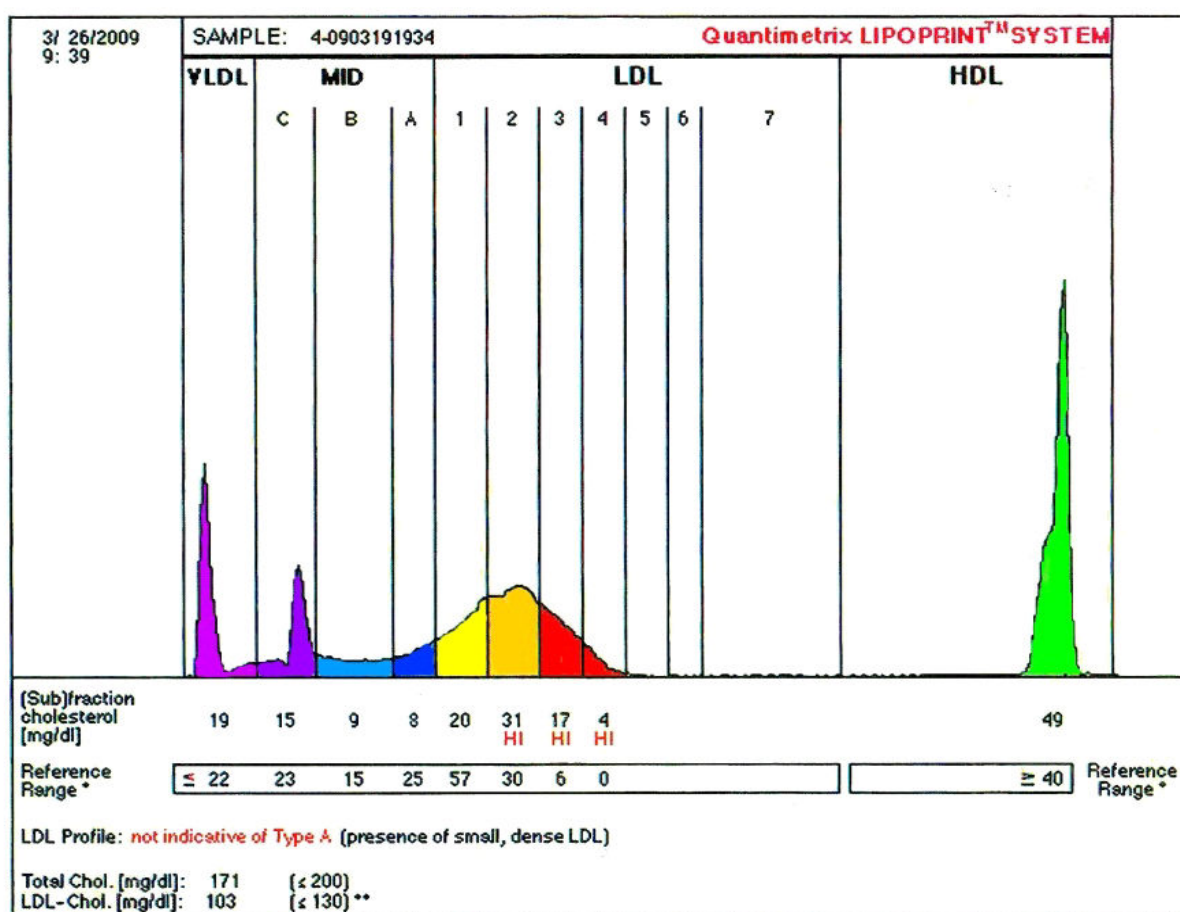
*Reference ranges derived from 125 serum samples that met the NCEP ATPIII guidelines for desirable lipid status
**LDL-C comprised of the sum of cholesterol in Md bands C through A as well as all the subfractions

Figure 2. Atherogenic normolipidemia – atherogenic subgroup of control group atherogenic small dense LDL are present in LDL 3,4 subfractions SAAR score: 2.7

A non-atherogenic lipoprotein profile in the control group was confirmed in 93.4% healthy normolipidemic individuals, and an atherogenic lipoprotein profile was found in 6.6%.

Table 3 shows high statistical significance for the analyzed lipid and lipoprotein parameters between the control group and the group of subjects with arterial hypertension ($p < 0.0001$, and for HDL, $p < 0.03$).

In Table 4, 78.5% of patients with arterial hypertension have an atherogenic lipoprotein profile. There is a highly significantly increased concentration of small dense LDL (subfractions LDL3-7) in a subgroup of AH-patients, who have an atherogenic profile, compared to the concentration of small dense LDL in the subgroup of AH-patients with a non-atherogenic profile, which confirms the predominance of atherogenic lipoproteins in AH-patients and the creation of atherogenic lipoprotein profile, phenotype B, as well. SAAR in patients with AH is low, that is, 9.2 (cut off is 10.8), and confirms also the predominance of atherogenic lipoproteins in serum.
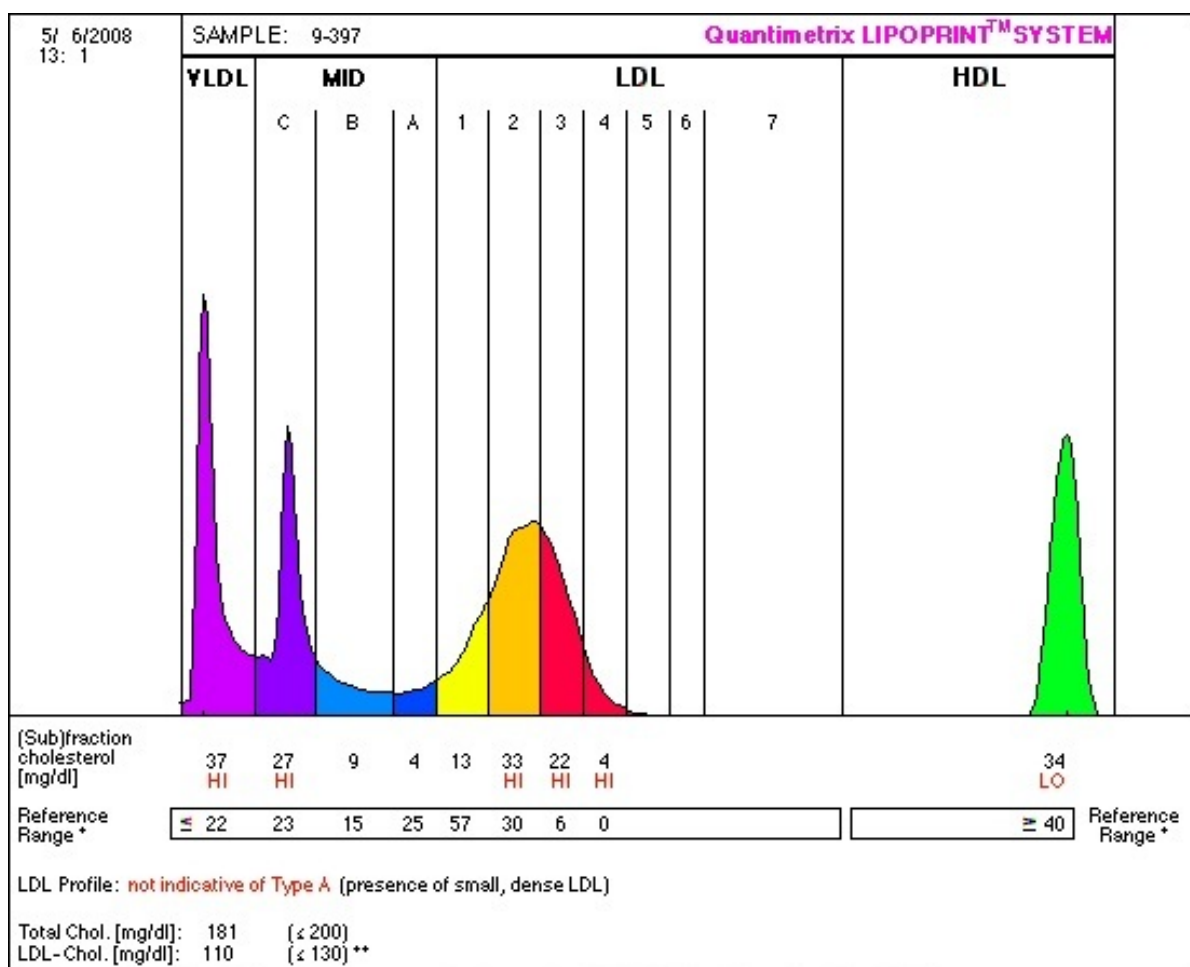
*Reference ranges derived from 125 serum samples that met the NCEP ATPIII guidelines for desirable lipid status
**LDL-C comprised of the sum of cholesterol in Md bands C through A as well as all the subfractions

Figure 3. Arterial hypertension with a borderline hypertriglyceridemia, small dense LDL are present in LDL3, 4 subfractions, SAAR score: 0.9

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l SD)							
Control	4.27	1.17	0.61	1.28	0.04	2.30	1.34	35.8
	±0.60	±0.39	±0.16	±0.37	±0.004	±0.52	±0.32	±18.5
(total number n=150)								
AH	5.19	2.28	0.97	1.54	0.35	3.00	1.25	9.2
	±1.10	±1.07	±0.34	±0.55	±0.25	±0.91	±0.34	± 4.5
(total number n= 107)								
Control vs. AH								
	p<0.0001						p< 0.03	p< 0.0001

Table 3. Serum concentration of lipids, lipoproteins, and SAAR-score in AH patients vs.

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l SD)							
AH	5.32	1.56	0.84	1.78	0.08	3.02	1.49	24.2
	±0.98	±0.55	±0.31	±0.44	±0.04	±0.71	±0.34	±13.6
(non-atherogenic profile n= 23)								
AH	5.15	2.48	1.01	1.47	0.42	2.99	1.18	5.1
	±1.14	±1.34	±0.35	±0.58	±0.31	±0.96	±0.34	± 2.0
(atherogenic profile n= 84)								
AH	**5.19**	**2.28**	**0.97**	**1.54**	**0.35**	**3.00**	**1.25**	**9.2**
	±1.10	±1.07	±0.34	±0.55	±0.25	±0.91	±0.34	± 4.5
(total number n=107)								
Non-atherogenic vs. atherogenic								
	n.s.	**p<0.002**	**p<0.05**	**p<0.02**	**p< 0.0001**	**n.s.**	**p< 0.001**	**p< 0.0001**

Atherogenic 78.5% vs. non-atherogenic 21.5% – arterial hypertension

Table 4. Serum concentration of lipids, lipoproteins, and SAAR-score in patients with arterial hypertension

3. Coronary heart disease

Coronary heart disease (CHD) (Fig. 4) is a common manifestation of cardiovascular diseases and is frequently associated with lipid and lipoprotein metabolism disorders. Hypercholesterolemia and hypertriglyceridemia, as well as combined hyperlipoproteinemia are regular features that accompany CHD [22,50,51]. Pathophysiologically, the cause of myocardial ischemia is a disproportion, or imbalance, between myocardial oxygen supply and oxygen demand. Ischemia in stable angina is generally due to fixed atheromatous stenosis of one or more coronary arteries as a consequence of impaired lipoprotein metabolism and the formation of lipid atheromas in the coronary arteries [5, 33, 34,49].

However, clinically, stable angina is not the only form of manifestation of coronary heart disease. Stable angina, as an ischemia due to fixed atheromatous stenosis, can turn into a myocardial ischemia due to plaque rupture with thrombosis and spasm of the artery (instable angina). In addition, myocardial necrosis (myocardial infarction), caused by acute occlusion of a coronary artery (due to plaque rupture and thrombosis), can have fatal consequences for disabled persons. It can be supposed that the modified forms of lipoproteins can play an important role in any form of clinical manifestation of coronary heart disease. Recently, clinical studies reported that the atherogenic lipoprotein populations (lipoprotein subfractions), presented in the plasma lipoprotein spectrum in high concentrations, play an important role in the development of atherosclerotic changes in the arterial wall [14, 38, 39].
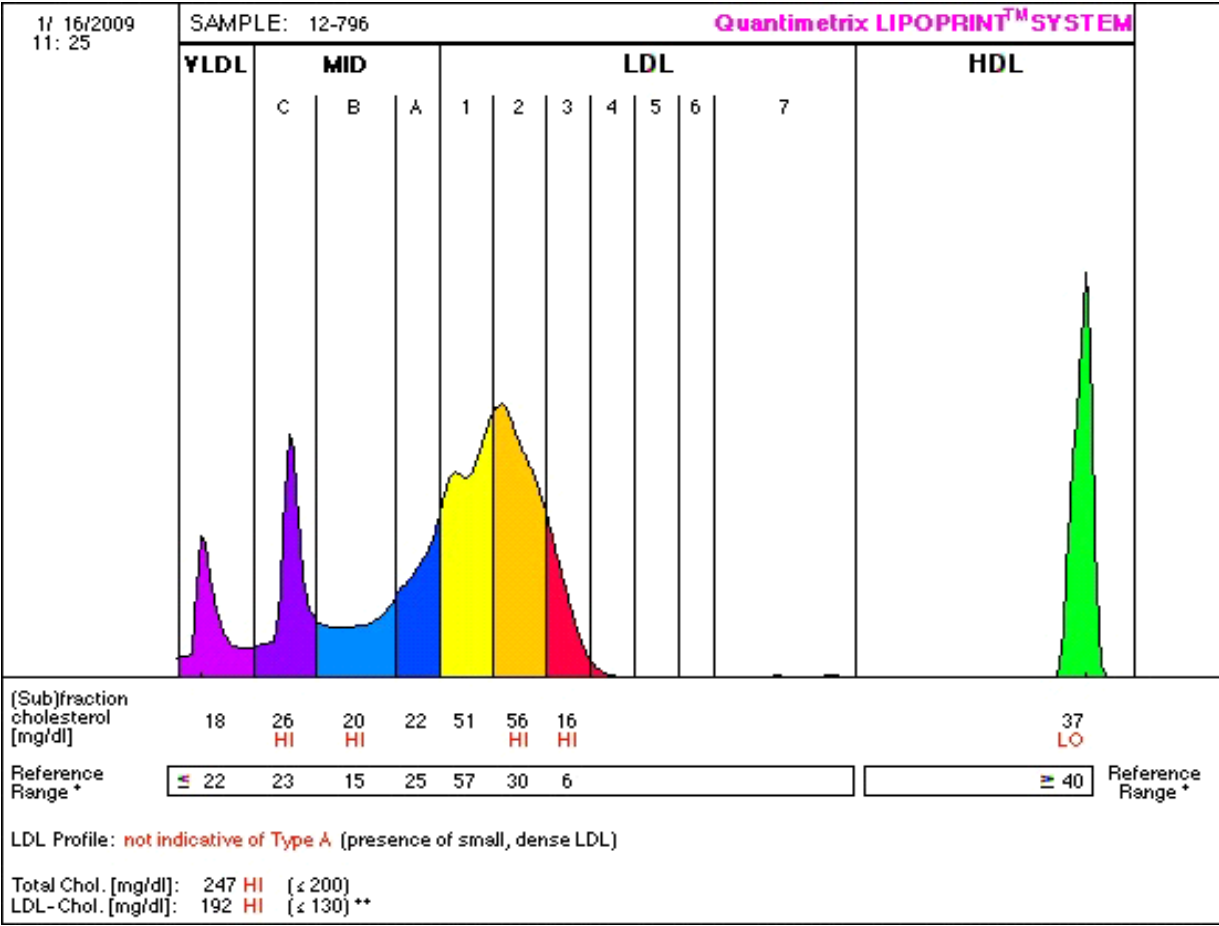
*Reference ranges derived from 125 serum samples that met the NCEP ATPIII guidelines for desirable lipid status
**LDL-C comprised of the sum of cholesterol in Md bands C through A as well as all the subfractions

Figure 4. Coronary heart disease combined with an atherogenic hypercholesterolemia. High concentration of atherogenic small dense LDL in LDL 3,4 subfractions SAAR score: 5.1

We distinguish facultative atherogenic very low density lipoproteins, VLDL, and their remnants, intermediate density lipoproteins, IDL, low density lipoproteins, LDL (considered a lipoprotein family with high atherogenicity), and high density lipoproteins, HDL. Modified lipoprotein entities in all these lipoprotein families can play a role in the formation of atherogenic lipoproteins, which accelerate the atherogenesis in the artery walls, including in the coronary arteries.

In our study, we focused on the determination of the incidence of an atherogenic lipoprotein phenotype in patients with coronary heart disease – in stable angina patients.

3.1. Patients

In our study, 104 patients with newly diagnosed coronary heart disease were examined. The diagnosis of CHD (stable angina pectoris grade I or II) was confirmed by medical examination, laboratory results, resting ECG, results of echocardiography, and duplex ultrasound of the carotid arteries.

3.2. Methods

See methods published in the section "Arterial hypertension (AH)."

3.3. Results

The results of lipid parameters presented in Table 5 confirm a highly significant increased concentration of analyzed lipid and lipoprotein parameters ($p<0.0001$) in CHD-patients, compared to control values and a low value of the SAAR. These low values (<10.8) are regularly found in atherogenic lipoprotein phenotype B.

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l SD)							
Control	**4.27**	**1.17**	**0.61**	**1.28**	**0.04**	**2.30**	**1.34**	**35.8**
	±0.60	±0.39	±0.16	±0.37	±0.004	±0.52	±0.32	±18.5
(total number n=150)								
CHD	**5.25**	**2.41**	**0.99**	**1.52**	**0.41**	**3.06**	**1.18**	**5.6**
	±1.15	**±1.72**	**±0.43**	**±0.47**	**±0.29**	**±0.85**	**±0.29**	**± 3.8**
(total number n= 104)								
Control vs. CHD								
				p<0.0001				

Table 5. Serum concentration of lipids, lipoproteins, and SAAR-score in CHD patients vs. control group

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l±SD)							
CHD	5.26	1.44	0.82	1.73	0.13	3.11	1.29	12.7
	±0.99	±0.50	±0.27	±0.49	±0.06	±0.80	±0.31	±5.1
(non-atherogenic profile = 19)								
CHD	5.25	2.63	1.02	1.47	0.48	3.05	1.16	4.0
	±1.19	±2.03	±0.47	±0.46	±0.35	±0.86	±0.28	±3.5
(atherogenic profile n= 85)								
CHD	**5.25**	**2.41**	**0.99**	**1.52**	**0.41**	**3.06**	**1.18**	**5.6**
	±1.15	±1.72	±0.43	±0.47	±0.29	±0.85	±0.29	±3.8
(total number n=104)								
Non-atherogenic vs. atherogenic profile								
		p<0.01		**p<0.05**	**p< 0.0001**	**n.s.**		**p< 0.0001**

Note: In the column without published p values, the differences in the evaluated parameter were not significant (n.s.)

Atherogenic 81.7% vs. non-atherogenic 18.3%, in coronary heart disease

Table 6. Serum concentration of lipids, lipoproteins, and SAAR-score in coronary heart disease

In Table 6, an atherogenic lipoprotein phenotype B is present in 81.7% of patients with CHD. An increased concentration of small dense LDL (LDL3-7) in the CHD-patient subgroup with an atherogenic lipoprotein profile, compared to the results of the CHD-patient subgroup with a non-atherogenic lipoprotein profile ($p<0.0001$), confirms a predominance of atherogenic lipoproteins in the serum of patients with CHD.

4. Lower extremity arterial disease

Lower extremity arterial disease (LEAD) (Fig. 5) is a common atherogenic disease of the cardiovascular system. Patients with LEAD exhibit normal to high atherogenic dyslipoproteinemia [8, 31, 50 -52, 62].
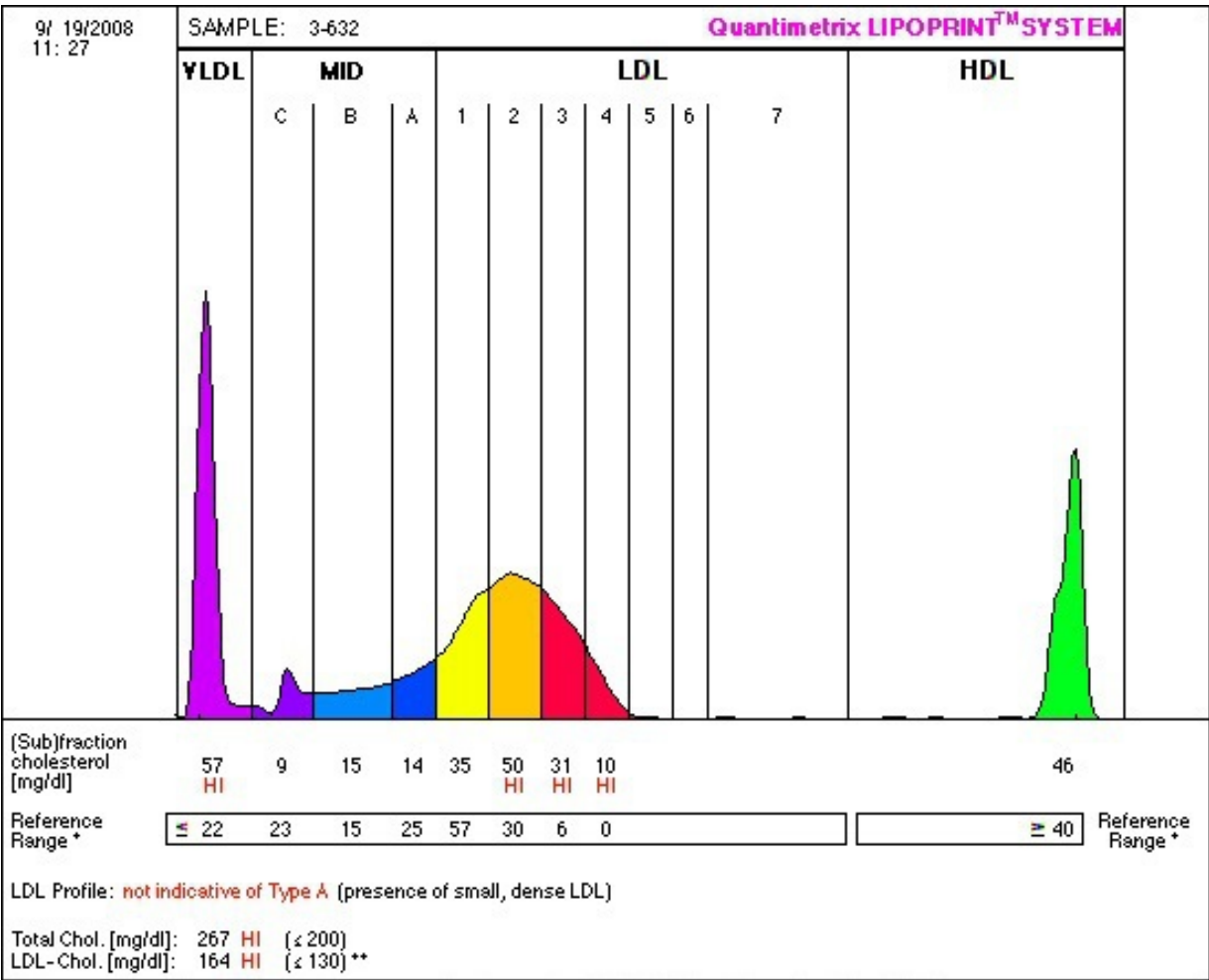
*Reference ranges derived from 125 serum samples that met the NCEP ATPIII guidelines for desirable lipid status
**LDL-C comprised of the sum of cholesterol in Md bands C through A as well as all the subfractions

Figure 5. Lower extremity arterial disease with combined atherogenic hyperlipoproteinemia with high concentration of atherogenic small dense LDL (LDL3,4 subfractions) SAAR score: 1.5

Almost all lower extremity arterial disease is due to atherosclerotic changes in artery vessels, and the pathology of LEAD is also similar to coronary heart disease. The most important risk factor for the development and progression of atherosclerotic LEAD are tobacco smoking, arterial hypertension, and hyperlipidemia. Other risk factors include diabetes mellitus, low physical activity, and a diet rich in lipids and carbohydrates. However, dyslipidemia plays an important role. Increased lipid levels of cholesterol and triglycerides are generally accepted as important risk factors for the development of atherosclerosis [14,25, 47].

In the last few decades, there has been much discussion about which atherogenic lipoproteins participate in the formation of the atherogenic lipoprotein profile, phenotype B. Atherogenic lipoproteins in relevant concentration in the blood serum are responsible for the acceleration of the development of atherogenic cardiovascular diseases, including the development of LEAD. The LDL subpopulations of small dense LDL are considered to be strongly atherogenic lipoprotein entities in the plasma/serum lipoprotein spectrum [38,59] with consequent acceleration of endothelial dysfunction and formation of the atheromatous subendothelial plaques in the arteries [21]. In the present study, we have focused on determining the incidence of an atherogenic lipoprotein phenotype, along with determining the role of atherogenic serum lipoproteins, in patients with lower extremity arterial disease.

4.1. Patients

In the clinical study, 100 patients with newly diagnosed lower extremity arterial disease were examined. The study included 55 males and 45 females: the average age of males was 56.0 years ±11 years and the average age of females 52.5 years ± 14 years. The patients had C2a degree, according to the Claudication classification: [proximal type (AP), the first degree (P1) with dyslipidemia]. Patients were ex-smokers.

LEAD was diagnosed according to the history of disease, intermittent claudication, the medical examination, including physical examination (Ratschow's test in the modification according to Linhart, see the Angiological Section of Slovak Medical Chamber) [23, 24, 27, 28] and examination of the ankle-brachial (pressure) index (ABPI) [40, 55, 57].

4.2. Methods

See methods published earlier in the section "Arterial hypertension (AH)."

4.3. Results

Results of lipid parameters presented in Table 7 confirm the highly significant increased concentration of analyzed lipid and lipoprotein parameters in LEAD-patients (p<0.0001), compared to control values. The low values of the SAAR, which is generally low (< 10.8) in the atherogenic lipoprotein phenotype, also confirms the atherogenic lipoprotein constellation in the serum of LEAD-patients.

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l SD)							
Control	**4.27**	**1.17**	**0.61**	**1.28**	**0.04**	**2.30**	**1.34**	**35.8**
	±0.60	±0.39	±0.16	±0.37	±0.004	±0.52	±0.32	±18.5
(total number n=150)								
LEAD	**5.29**	**2.21**	**0.96**	**1.58**	**0.39**	**3.11**	**1.21**	**7.2**
	±1.21	±1.05	±0.37	±0.51	±0.28	±0.96	±0.31	± 4.5
(total number n= 100)								
Control vs. LEAD								
	«..............................p<0.0001................................»							

Table 7. Serum concentration of lipids, lipoproteins, and SAAR-score in LEAD-patients vs. control group

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l±SD)							
LEAD	5.37	1.81	0.86	1.82	0.10	3.18	1.33	17.4
	±0.95	±0.51	±0.26	±0.54	±0.03	±0.82	±0.29	± 6.5
(non-atherogenic profile n= 20)								
LEAD	5.28	2.31	0.98	1.52	0.46	3.09	1.18	4.6
	±1.28	±1.18	±0.39	±0.50	±0.34	±0.99	±0.32	± 4.0
(atherogenic profile n= 80)								
LEAD	**5.29**	**2.21**	**0.96**	**1.58**	**0.39**	**3.11**	**1.21**	**7.2**
	±1.21	±1.05	±0.37	±0.51	±0.28	±0.96	±0.31	± 4.5
(total number n=100)								
Non-atherogenic vs. atherogenic								
	p<0.01	p<0.001		p<0.05	p< 0.0001			p< 0.0001

Atherogenic 80% vs. non-atherogenic 20% in lower extremity arterial disease

Table 8. Serum concentration of lipids, lipoproteins, and SAAR-score in lower extremity arterial disease

In 80% of patients (Table 8), LEAD was associated with an atherogenic lipoprotein phenotype. An increased concentration of small dense LDL (LDL3-7 subgroups) in the LEAD-patients with an atherogenic lipoprotein profile, compared to the results from the LEAD-patients with a non-atherogenic lipoprotein profile (p<0.0001), confirms the predominance of atherogenic lipo-proteins in serum in the subgroup of patients with an atherogenic lipoprotein profile.

5. Stroke

Stroke (Fig. 6) is the leading cause of mortality and of long-term morbidity in the populations of developed industrialized countries in the world. The atherogenic serum lipoproteins in high concentrations create an atherogenic lipoprotein profile, which plays a key role in the acute onset of cardiovascular and cerebrovascular events, that is, stroke [54,55]. Cerebral stroke attack remains a frequent medical problem and is the third most frequent cause of mortality all over the world. It represents a heterogeneous group of diseases with more than 150 known causes. In 25–39% of strokes, the cause leading to the acute cerebrovascular event cannot usually be definitively explained [4].
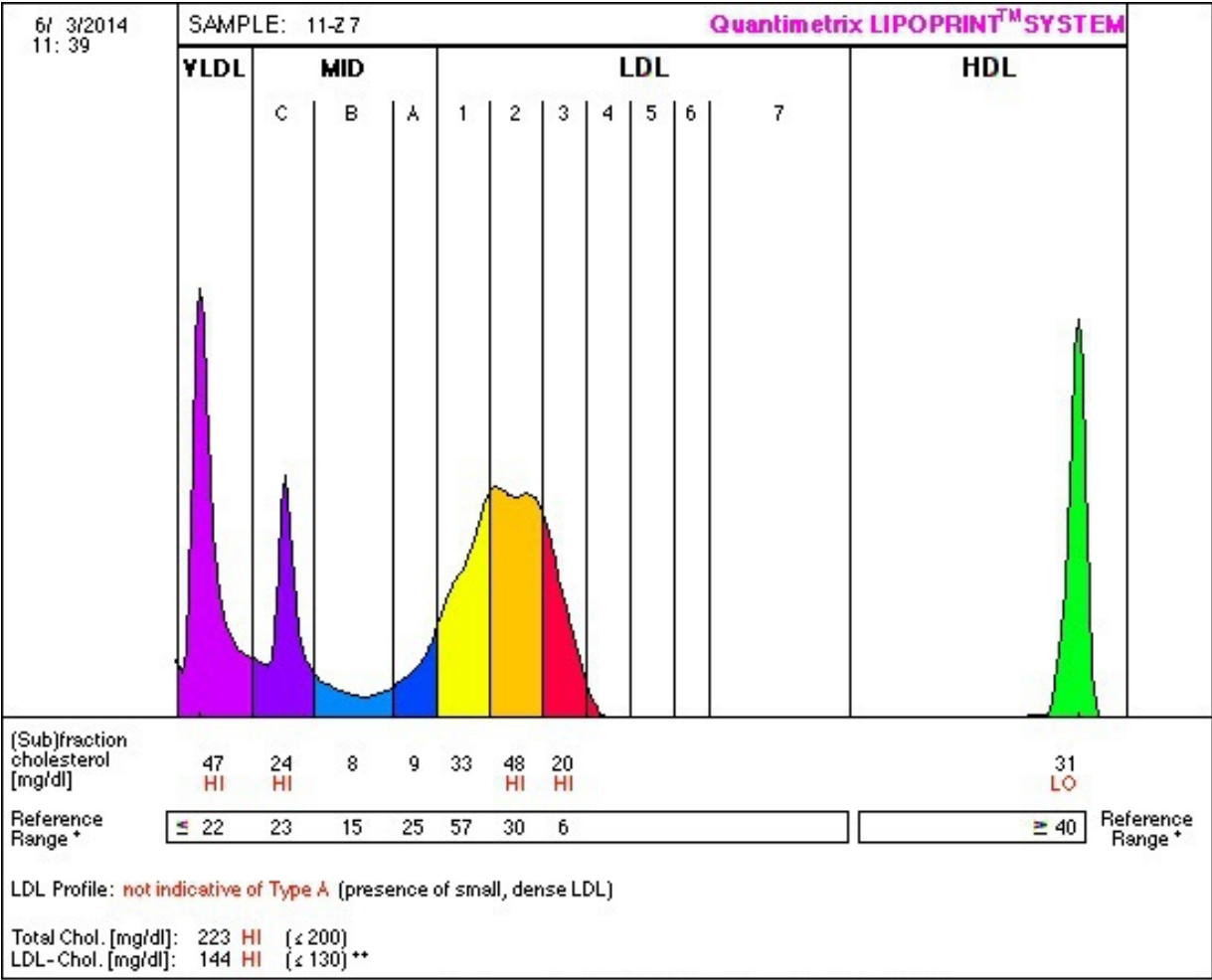
Figure 6. Patient survived an ischemic stroke with combined atherogenic hyperlipoproteinemia high concentration of VLDL, VLDL remnants and atherogenic small dense LDL, i.e. LDL 3, 4 subfractions. SAAR score: 2.4*Reference ranges derived from 125 serum samples that met the NCEP ATPIII guidelines for desirable lipid status**LDL-C comprised of the sum of cholesterol in Md bands C through A as well as all the subfractions

Dyslipidemia represents a risk factor for the development of cardiovascular disease, and thus dyslipidemia has been classified as an atherogenic phenomenon. The goal of the treatment of

hyperlipoproteinemia, that is, of dyslipidemia, is to reduce the lipid concentration in serum to established target values of lipids (cholesterol and triglycerides), but the primary goal is to reduce the atherogenic potential of serum lipids [9, 21, 53]. Dyslipoproteinemia is also the key phenomenon in the pathogenesis of the onset of atherosclerotic alterations in brain vessels [64]. Accompanied by high cholesterol levels – a classic risk factor for the development of cardio-vascular diseases – an increased concentration of triglycerides in the blood serum can also play an important role in atherogenesis [3,58].

There are several studies that have provided evidence for the relation between carotid artery stenosis and an ischemic cerebral event [55]. However, the causal inter-relation between dyslipidemia and stroke has not been explained sufficiently [3, 4, 63]. Relapsing ischemic strokes account for one-fourth of all strokes in a year and are a strong evidence for a failure of secondary prevention [10]. This hard reality leads rightly so to the idea of optimal stroke prevention through the selection of individuals, who are at risk of stroke [13]. The aim of this pilot study was to identify the atherogenic lipoproteins and determine the lipoprotein profile in subjects who had suffered an ischemic cerebrovascular event, that is, stroke.

5.1. Patients

The study included 55 patients, 23 men, with an average age of 64 years ± 13 years, and 32 women, average age 74 years ± 13 years, who survived an ischemic cerebrovascular event, that is, a large-artery atherosclerosis subtype of stroke. To determine the subtype of ischemic stroke, the original TOAST (Trial of ORG 10172 in Acute Stroke Treatment) [1] criteria were used. The diagnosis of subtype was based on the risk factor profiles, clinical features, and results of diagnostic tests, including CT scan/MRI, vascular imaging (carotid duplex, transcranial Doppler), EEG – electroencephalography, echocardiography (transesophageal/ transthoracic), assessment of prothrombotic syndromes [1,30], activated partial thromboplastin time (aPTT), and international normalized ratio (INR).

5.2. Methods

See methods published in the section "Arterial hypertension (AH)."

A blood sample from the antecubital vein was obtained throughout the 24 hours after the onset of cerebrovascular event.

5.3. Results

The results of lipid parameters presented in Table 9 confirm a highly significantly increased concentration of analyzed lipid and lipoprotein parameters (p<0.0001) in people who survive a stroke, compared to control values, and also a low value on the SAAR, which is generally low (< 10.8) in an atherogenic lipoprotein phenotype.

In Table 10, an atherogenic lipoprotein phenotype was identified in 85.5 % of the patients who survive a cerebral ischemic stroke. The increased concentration of small dense LDL (LDL3-7

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score
	(mmol/l SD)							
Control	**4.27**	**1.17**	**0.61**	**1.28**	**0.04**	**2.30**	**1.34**	**35.8**
	±0.60	±0.39	±0.16	±0.37	±0.004	±0.52	±0.32	±18.5
(total number n=150)								
Stroke	**5.19**	**2.21**	**1.08**	**1.56**	**0.29**	**2.91**	**1.09**	**6.40**
	±1.10	±0.91	±0.34	±0.57	±0.24	±0.82	±0.32	±4.22
(total number n= 55								
Control vs. stroke								
	«..p<0.0001...»							

Table 9. Serum concentration of lipids, lipoproteins, and SAAR-score in stroke patients vs. control group.

	Chol	TAG	VLDL	LDL1,2	LDL3-7	LDL	HDL	Score	LDL1	LDL2
	(mmol/l±SD)									
Stroke	5.54	1.70	0.93	2.19	0.14	3.30	1.31	13.74	1.22	0.92
	±1.30	±0.44	±0.14	±0.86	±0.07	±1.01	±0.35	±1.36	±0.45	±0.44
(non-atherogenic profile n= 8)										
Stroke	5.14	2.29	1.11	1.48	0.31	2.86	1.06	5.33	0.72	0.76
	±1.11	±0.94	±0.37	±0.41	±0.23	±0.72	±0.29	±3.32	±0.26	±0.26
(atherogenic profile n = 47)										
Stroke	**5.19**	**2.21**	**1.08**	**1.56**	**0.29**	**2.91**	**1.09**	**6.40**	**0.78**	**0.78**
	±1.06	±0.91	±0.35	±0.53	±0.23	±0.77	±0.31	± 4.22	±0.33	±0.28
(total number n= 55)										
Non-atherogenic vs. atherogenic										
	n.s.	**n.s.**	**n.s.**	**p<0.002**	**p<0.05**	**n.s.**	**n.s.**	**p< 0.0001**	**n.s.**	**p< 0.0001**

Atherogenic 85.5 % vs. non-atherogenic 14.5 % in stroke patients.

Table 10. Serum concentration of lipids, lipoproteins, and SAAR-score in patients with stroke.

subgroups) in the atherogenic lipoprotein profile of patients with stroke, compared to the results in a non-atherogenic lipoprotein profile, is mild, but significant (p<0.05). The difference in the SAAR between the two subgroups was highly significant (p<0.0001), which also confirmed the overwhelming atherogenic lipoprotein constellation in patients who survived a stroke. The concentration of LDL1 was significantly higher in the subgroup of stroke-patients with a non-atherogenic lipoprotein profile (p<0.0001), however, the difference in the LDL2 lipoprotein subfraction was not significant.

6. Discussion

In the last few decades, lipoprotein research has focused on the phenomenon of atherogenic and non-atherogenic lipoproteins, and on the phenotype A vs. phenotype B characterization, as a consequence of the published evidence that the majority of the patients with an acute coronary syndrome or patients who survive a myocardial infarction had normal plasma values of cholesterol, LDL-cholesterol, and HDL-cholesterol [15-17]. A reasonable explanation for this fact was to posit a new, active atherogenic substance in plasma, an atherogenic lipoprotein subfraction, the presence of which in plasma, even in very low concentrations, could impair the integrity of the vessel wall and lead to endothelial dysfunction with its fatal consequences. Several clinical studies reported observations that in the plasma of patients with coronary heart disease there are subfractions of lipoproteins, which could play a crucial role in atherodegenerative processes and form the atherothrombotic plaques [5, 33, 34, 37, 39,49]. The Quebec Cardiovascular Study, a prospective study of 2,103 men [33,34] concluded that " a significant proportion of the risk for heart disease associated with small, dense LDL particles may be independent of variations in plasma lipid concentrations. Small LDL particles and elevated apo B levels were found to be the most predictive indications for ischemic heart disease ".

For this reason, patients who were suffering from cardiovascular diseases were examined in order to quantify the atherogenic lipoproteins in serum and to determine the incidence of an atherogenic lipoprotein profile in patients who had a diagnosis of cardiovascular diseases.

The clinical studies included 366 patients with a diagnosis of arterial hypertension (n=107), coronary heart disease (n= 104), lower extremity arterial disease (n= 100), and ischemic stroke (n= 55). Patients were tested with the diagnostic method Lipoprint LDL System, which quantifies atherogenic lipoproteins and identifies an atherogenic and a non-atherogenic lipoprotein profile [29]. This was a fundamental methodological contribution of this new analytical and diagnostic method.

Our study confirmed that more than 80% of tested patents with cardiovascular diseases have an atherogenic lipoprotein profile, with a high level of strongly atherogenic small dense LDL. The atherogenic lipoprotein profile was found to be the overwhelming lipoprotein profile in tested cardiovascular diseases. Such a profile was found in arterial hypertension in 78.5%, in coronary heart disease in 81.7%, in lower extremity arterial disease in 80%, and in patients who survived an ischemic stroke in 85%. The average atherogenic lipoprotein profile in all these tested diagnoses in the study was found to be of 81.3%.

This study also highlights the observation that, in the atherogenic lipoprotein profiles, in all diagnoses, compared to the non-atherogenic profiles, the concentration of total cholesterol is lower (n.s.) and the concentration of triglycerides is higher (even statistically significant; in AH, CHD, LEAD, as well as in the control group, up to $p<0.002$). Hypertriglyceridemia accompanied the hypercholesterolemia in all tested diagnoses, that is, in AH, CHD, LEAD, and stroke). The concentration of triglycerides, compared to the control group, was significantly increased ($p<0.0001$) and proportionally even higher than cholesterol. From this result,

it can be assumed that triglycerides/hypertriglyceridemia can play a much more important role than was generally accepted, as until now the most important role in the pathogenesis of vascular degenerative atherosclerotic injury was attributed to cholesterol and hypercholester-olemia. Our present results are in agreement with other authors, who have called attention to hypertriglyceridemia as a risk factor for cardiovascular diseases [8,12,19, 36,58], as triglyceride-rich lipoproteins can generate small dense LDL in high quantities [46].

The strong atherogenic lipoproteins – small dense LDL – have been found in the lipoprotein profile of all diagnostic groups [25,52,55,56]. Their presence is decisive for an atherogenic profile declaration. This is a rule that is valid not only for a hyperlipidemia, but also for a normolipidemia.

In the case of normolipidemia (see the atherogenic lipoprotein profile in the control group), a new phenomenon could be established – atherogenic normolipidemia [44] – as a risk factor for the development of cardiovascular disease. A special form of normolipidemia can also be atherogenic. This is new knowledge, and this new knowledge could help in the prevention and treatment of cardiovascular disease.

Acknowledgements

Author details

Stanislav Oravec[1*], Kristina Gruber[2], Andrej Dukat[1], Peter Gavornik[1], Ludovit Gaspar[1] and Elisabeth Dostal[3]

*Address all correspondence to: stanislavoravec@yahoo.com

1 2nd Department of Internal Medicine, Faculty of Medicine, Comenius University, Bratislava, Slovak Republic

2 Department of Internal Medicine, Landesklinikum, Thermenregion Baden, Austria

3 Krankenanstalten Dr. Dostal, Vienna, Austria

References

[1] Adams HP, Benedixen BH, Kapelle LJ, Biller J, Love BB, Gordon DL, Marsh EE. Classification of subtype of acute ischemic stroke. Definition for use in a multicenter clinical trial:TOAST Trial of Org 10172 in Acute Stroke Treatment. Stroke 1993; 24(1): 35–41

[2] Alabakovska SB, Todorova BB, Labudovic DD, Tosheska KN. Gradient gel electrophoretic separation of LDL and HDL subclasses on BioRad Mini Protean II and size phenotyping in healthy Macedonians. Clin Chim Acta 2002; 317: 119–123

[3] Amarenco P, Labreuche J, Touboul PJ. High-density lipoprotein cholesterol and risk of stroke and carotid atherosclerosis. A systemic review. Atherosclerosis 2008; 196: 489–496

[4] Amarenco P, Bogousslavsky J, Caplan LR, Donnan GA, Hennerici MG. Classification of stroke subtypes. Cerebrovasc Dis 2009; 27: 493–501

[5] Arsenault BJ, Lemieux I, Despres J-P, Wareham NJ, Luben R, Kastelein JP, Khaw K-T, Boekholdt SM. Cholesterol levels in small LDL particles predict the risk of coronary heart disease in the EPIC-Norfolk prospective population study. Eur Heart J 2007; 28(22): 2770–2777

[6] Austin MA, Breslow JL, Henekens CH, Buring JE, Willett WC, Krauss RM. Low-density lipoprotein subclass patterns and risk of myocardial infarction. JAMA 1988; 260:1917–1921

[7] Austin MA, King MC, Vranizan KM, Krauss RM. Atherogenic lipoprotein phenotype. A proposed genetic marker for coronary heart disease risk. Circulation 1990; 82: 495–506

[8] Austin MA, Hokanson JE, Edwards KL. Hypertriglyceridemia as a cardiovascular risk factor. Am J Cardio 1994; 81: 7B–12B

[9] Backers J. Effect of lipid lowering drug therapy on small-dense low density lipoprotein. Ann Pharmacol 2005; 39(3): 523–526

[10] Bejot Y, Benatru I, Roaud O, Fromont A, Besancenot JP, Moreau T, Giroud M. Epidemiology of stroke in Europe: Geographic and environmental differences. J Neurol Sci 2007; 262(1–2): 85–88

[11] Berneis KK, Krauss RM. Metabolic origins and clinical significance of LDL heterogeneity. J Lipid Res 2002; 43: 1363–1379

[12] Brunzell JD. Hypertriglyceridemia. N Engl J Med 2007; 357(10): 1009–1017

[13] Bulas J. Interdisciplinárny prístup k liečbe náhlej cievnej mozgovej príhody zo strany Internistu.(A complex approach to the treatment of an acute cerebral-vascular event from the position of internist). Interna Medicina 2006; 6(9): 1363–1379

[14] Carmena R, Duriez P, Fruchart JC. Atherogenic lipoprotein particles in atherosclerosis. Circulation 2004; 109: III2–III7

[15] Castelli WP. Cholesterol and lipids in the risk of coronary artery disease – The Framingham Heart Study. Can J Cardiol 1988; (Suppl A): 5A–10A

[16] Castelli WP. Epidemiology of triglycerides; a view from Framingham. Am J Cardiol 1992; 70: 43–49

[17] Castelli WP. The new pathophysiology of coronary artery disease. Am J Cardiol 1998; 82(Suppl 2): 60–85

[18] Chait A, Brazo RL, Tribble DL, Krauss RM. Susceptibility of small, low-density lipoproteins to oxidative modification in subjects with the atherogenic lipoprotein phenotype, pattern B. Amer J Med 1993; 94: 350–356

[19] Cullen P. Evidence that triglycerides are an independent coronary heart disease risk factor. Am J Cardiol 2000; 86: 943–949

[20] Felmeden DC, Spencer CGC, Blann AD, Beewers DG, Lip YH. Low-density lipoprotein subfractions and cardiovascular risk in hypertension. Relationship to endothelial dysfunction and effects of treatment. Hypertension 2003; 41: 528–533

[21] Fruchart JC, Sacks FM, Hermans MP et al. The residual risk reduction initiative: A call to action to reduce residual vascular risk in dyslipidemic patients. Diab Vasc Res 2008; 5(4): 319–335

[22] Gardner CD, Fortman SP, Krauss, RM. Association of small low-density lipoprotein particles with the incidence of coronary artery disease in men and women. JAMA 1996; 276: 875–881

[23] Gavorník P. Všeobecná angiológia. Angiologická propedeutika. Cievne choroby-2. vydanie (General angiology, propedeutics of angiology, Vessel diseases, 2nd ed). Comenius University, Bratislava, 2001, 268 p.

[24] Gavorník P. Etiopatogenéza a nová klinicko-etiologicko-anatomicko-patologická (CEAP) klasifikácia končatinovej choroby (Ethiopathogenesis and a new clinical ethio-anatomico-pathological (CEAP) classification of the low extremity arterial disease). Interna Medicina 2010; 10: 115–123

[25] Gazi IF, Tsimihodimos V, Tselepis AD, Elisaf M, Mikhailidis DP. Clinical importance and therapeutic modulation of small dense low-density lipoprotein particles. Expert Opin Biol Therapy 2007; 7(1): 53–72

[26] Hirany SV, Othman Y, Kutscher P, Rainwater D, Jialal I, Devaraj S. Comparison of low-density lipoprotein size by polyacrylamide tube gel electrophoresis and polyacrylamide gradient gel electrophoresis. Am J Clin Pathol 2003; 119: 439–445

[27] Hirsch AT, Criqui MH, Treat-Jacobson D, Regensteiner JG, Creager MA, Olin JN, Krook SH, Hunninghake DB, Comerota AJ, Walsh ME, McDermott MM, Hiatt WR.

Peripheral arterial disease detection, awareness and treatment in primary care. JA-MA 2001; 286(11): 1317–1322

[28] Hirsch AT, Haskal ZJ, Hertzer NR et al. ACC/AHA 2005 Guidelines for the management of patients with peripheral arterial disease. J Am Coll Cardiol 2006; 47: 1239–1312

[29] Hoefner DM, Hodel SD, O'Brien JF, Branum EL, Sun D, Meissner I, McConnell JP. Development of a rapid quantitative method for LDL subfraction with use of the Quantimetrix Lipoprint LDL system. Clin Chem 2001; 472: 266–274

[30] Kalita Z. Akutní cévní mozkové príhody. Diagnostika, patofyziologie, management (The acute cerebral-vascular events. Diagnostics, pathophysiology, management). Maxdorf, Praha, 2006, 623 p

[31] Kasko M, Gaspar L, Dukat A, Gavornik P, Oravec S. High-density lipoprotein profile in newly-diagnosed lower extremity artery disease in Slovak population without diabetes mellitus. Neuroendocrinol Lett 2014; 35(6): 531–535

[32] Kim YK, Seo HS, Lee EM, Na JO, Choi CU, Lim HE, Kim EJ, Rha SW, Park CG, Oh DJ. Association of hypertension with small, dense low-density lipoprotein in patients without metabolic syndrome. J Hum Hypertension 2012; 26: 670–676

[33] Lamarche B, Tchernof A, Moorjani S, Cantin B, Dagenais RG, Lupien PJ, Despres J-P. Small dense low-density lipoprotein particles as a predictor of the risk of ischemic heart disease in men. Prospective results from the Quebec Cardiovascular Study. Circulation 1997; 95: 69–75

[34] Lamarche B, Lemieux I, Despres J-P. The small, dense LDL phenotype and the risk of coronary heart disease: Epidemiology, pathophysiology and therapeutic aspects. Diabetes Metab 1999; 25: 199–211

[35] Landray MJ, Edmunds E, Li Saw Hee FL, Hughes BA, Beewers DG, Lip Gyh. Abnormal low density lipoprotein subfraction profile in patients with untreated hypertension. Q J Med 2002; 95: 165–171

[36] McBride PE. Triglycerides and risk for coronary artery disease. JAMA 2007; 298(3): 336–338

[37] Mudd JO, Borlaug BA, Johnston PV, Kral BG, Rouf R, Blumenthal RS, Kwiterovich PO, Jr. Beyond low-density lipoprotein cholesterol: Defining the role of low-density lipoprotein heterogeneity in coronary artery disease. J Am Coll Cardiol 2007; 50(18): 1735–1741

[38] National Cholesterol Education Program (NCEP), Expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III) final report. Circulation 2002; 106: 3143–3421

[39] Nicholls S, Lundmann P. The emerging role of lipoproteins in atherogenesis: Beyond LDL cholesterol. Semin Vasc Med 2004; 4: 187–195

[40] Norgren L, Hiatt WR, Dormandy JA, Nehler MR, Harris KA, Fowkes FG. TASC II Working Group. Inter-society consensus for the management of peripheral arterial disease. J Vascul Surg 2007; 45(1 Suppl): S5–S67

[41] Oravec S. Identifikácia subpopulácií LDL triedy – Aktuálny prínos v diagnostike porúch metabolizmu lipoproteínov a ochorení kardiovaskulárneho systému (Identification of LDL subfractions – A contribution in the diagnostics of lipoprotein metabolism diseases and cardiovascular diseases). Med Milit Slov 2006; 8: 32–34

[42] Oravec S. Nové perspektívy v diagnostike porúch metabolizmu lipoproteínov-prínos v interpretácii výsledkov (New perspectives in the diagnostics of lipoprotein metabolism diseases – a contribution in the interpretation of results). Med Milit Slov 2007; 9: 42–45

[43] Oravec S, Dukat A, Gavornik P, Caprnda M, Kucera M, Ocadlik I. Contribution of the atherogenic lipoprotein profile to the development of arterial hypertension. Bratisl Lek Listy 2011; 12(1): 4–7

[44] Oravec S, Dukat A, Gavornik P, Lovasova Z, Gruber K. Atherogenic normolipidemia – A new phenomenon in the lipoprotein profile of clinically healthy subjects. Neuroendocrinol Endocrinol Lett 2011; 32(3): 317-321

[45] Otvos JD, Jeyarajah EJ, Bennet SW, Krauss RM. Development of a proton nuclear magnetic resonance spectroscopic method for determining plasma protein concentrations and subspecies distribution from a single, rapid measurement. Clin Chem 1992; 38: 1632–1638

[46] Packard CJ. Triacylglycerol-rich lipoproteins and the generation of small dense low-density lipoprotein. Biochem Soc Trans 2003; 31: 1066–1069

[47] Pollak AW, Kramer CM. LDL lowering in peripheral arterial disease. Are there benefits beyond reducing cardiovascular morbidity and mortality ? Clin Lipidol 2012; 7(2): 141–149

[48] Rainwater DL, Moore PH Jr, Shelledy WR, Dyer TD, Slifer SH. Characterization of a composite gradient gel for the electrophoretic separation of lipoproteins. J Lipid Res 1997; 38: 1261–1266

[49] Rajman I, Kendall MJ, Cramb R, Holder RL, Salih M, Gammage MD. Investigation of low density lipoprotein subfractions as a coronary risk factor in normotriglyceridemic men. Atherosclerosis 1996; 125: 231–242

[50] Rizzo M, Berneis K. Low density lipoprotein size and cardiovascular prevention Europe J Int Med 2006; 17: 77–80

[51] Rizzo M, Berneis K. Low-density-lipoprotein size and cardiovascular risk assessment QJM – Int J Med 2006; 99: 1–14

[52] Rizzo M, Pernice V, Frasheri A, Berneis K. Atherogenic lipoprotein phenotype and LDL size and subclasses in patients with peripheral arterial disease. Atherosclerosis 2008; 197: 237–241

[53] Rubins HB, Davenport J, Babikian V et al. Reduction in stroke with gemfibrozil in men with coronary heart disease and low HDL cholesterol: The veterans affairs HDL intervention trial (VA-HIT). Circulation 2001; 103(23): 2828–2833

[54] Sarkar PD, Rautaray SS. Oxidized LDL and paraoxonase status in ischemic stroke patients. Indian J Physiol Pharmacol 2008; 52(4): 403–407

[55] Shoji T, Hatsuda S, Tsuchikura S, Shinohara K, Komoto E, Koyama H, Emoto M, Nishizawa Y. Small dense low-density lipoprotein cholesterol concentration and carotid atherosclerosis. Atherosclerosis 2009; 202: 582–588

[56] St-Pierre AC, Cantin B, Daganais GR, Mauriege P, Bernard PM, Despres JP, Lamarche B. Low density lipoprotein subfractions and the long-term risk of ischemic heart disease in men: 13-year follow-up data from the Quebec Cardiovascular Study. Arterioscler Thromb Vasc Biol 2005; 25: 553–559

[57] Superko HR. Lipoprotein subclasses and atherosclerosis. Front Biosci 2001; 6: D355–D365

[58] Tanne D, Koren-Morgan N, Graff E, Goldbourt U. Blood lipids and first ever ischemic stroke, transient ischemic attack in the bezafibrate infarction prevention (BIP) registry: High triglyceride constitute an independent risk factor. Circulation 2001; 104: 2892–2897

[59] Vakkilainen J, Mäkimattila S, Seppälä-Lindroos A, Vehkavaara S, Lahdenperä S, Groop PH, Taskinen MR, Järvinen HY. Endothelial dysfunction in men with small LDL particles. Circulation 2000; 102: 716–721

[60] Van J, Pan J, Charles MA, Krauss R, Wong N, Wu X. Atherogenic lipid phenotype in a general group of subjects. Arch Pathol Lab Med 2007; 131: 1679–1685

[61] Whitworth JA. 2003 World Health Organisation (WHO), International Society of hypertension (ISH) Statement on management of hypertension. J Hypertension 2003; 21: 1983–1992

[62] Wierzbicki AS. Quality as well as quantity? Beyond low-density lipoprotein-cholesterol – The role of particle size. Int J Clin Pract 2007; 61: 1780–1782

[63] Vrablik M. Prevence cévních mozkových příhod hypolipidemickou léčbou. Remedia. Interetové stránky ceského farmakoterapeutického dvoumesícniku, rubrika PND, 2. duben 2009 (The prevention of the stroke by hypolipidemic treatment. Remedia). In-

ternet Website of Czech Pharmaco therapeutic two-monthly Journal, column PND, April 2, 2009

[64] Chun Xia Zhao, Ying Hua Cui, Qiao Fan, Pei Hua Wang, Ruitai Hui, Cianflone K, Dao Wen Wang. Small dense low-density lipoproteins and associated risk factors in patients with stroke. Cerebrovasc Dis 2009; 27: 99–104