Parameters of High-Density Lipoproteins in Patients with Arterial Hypertension in Combination with Other Components of Metabolic Syndrome

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 143, No. 3, pp. 289-292, March, 2007
Original article submitted July 4, 2006

Parameters of HDL (concentrations of cholesterol, apoprotein A1, and phospholipids and phospholipid composition) determining their functional properties were studied in patients with arterial hypertension in combination with other components of metabolic syndrome (abdominal obesity, hyperlipidemia, and impaired glucose tolerance). Patients with isolated arterial hypertension did not differ from the control group by the concentration of apoprotein A1 and HDL cholesterol, but had lower content of HDL phospholipids and changed phospholipid composition: lower ratio of phosphatidylcholine and higher relative contents of lysophosphatidylcholine, sphingomyelin, and phosphatidylethanolamine. Parameters of HDL in patients with arterial hypertension associated with other components of metabolic syndrome did not differ from those in patients with isolated arterial hypertension. The observed changes in HDL in patients with arterial hypertension alone or in combination with other components of metabolic syndrome can impair functional capacity of HDL in reverse cholesterol transport, which increases the risk of atherosclerosis.

Key Words: *high-density lipoproteins; phospholipids; apoprotein A1; arterial hypertension; metabolic syndrome*

Arterial hypertension (AH), excessive body weight (obesity), hyperlipidemia (HLP), and insulin resistance are serious risk factors for the development of atherosclerosis and its clinical complications (CHD and stroke) [1,3]. These interrelated signs received the name metabolic syndrome. Pathogenicity of this syndrome is aggravated by decreased blood content of HDL cholesterol (CH). A negative correlation was found between the development of atherosclerosis and blood HDL CH content.

Antiatherogenic function of HDL is associated with their ability to accept CH from cell membranes and transport it to the liver in the esterified form (reverse CH transport) [6,9]. HDL-mediated reverse CH transport is determined by a number of factors, in particular, by HDL components apoprotein A1 and phospholipids [6,8-10]. The ratio between individual phospholipids determines microviscosity of membranes and surface monolayer of HDL. Apoprotein A1 and phospholipids can form complexes with CH, which determines CH-acceptor properties of HDL.

Functional properties of cell membranes in hypertensive animals (e.g. rats) and AH patients are disturbed due to changes in their lipid composition [4,8,13]. Hypertension is associated with increased CH content and decreased phospholipid concentration in cell membranes (increased CH/phospholipid ratio) and changed phospholipid composition. Ery-

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0007-4888/07/1433-0320

throcyte membrane is the most adequate model of cell membrane, because erythrocytes are characterized by the lack of cell nucleus, universal structure of the membrane, and common mesenchymal origin with endothelial cells. This is why the state of erythrocyte membrane reflects the involvement of all cell membranes into the pathological process. Synthetic processes do not occur in the erythrocyte membrane and its lipids are easily exchanged with blood lipoprotein lipids. In light of this we assume that changes in the lipid composition of erythrocyte membranes during AH are determined by composition of blood lipoproteins. Changes in the lipid and phospholipid composition of various classes of lipoproteins were previously revealed in patients with essential hypertension [5].

Here we evaluated whether parameters of HDL determining their functional properties are altered in patients with isolated AH or AH associated with other components of metabolic syndrome.

MATERIALS AND METHODS

We examined 82 patients (men, 35-66 years, mean age 54.2±1.8 years) with 4-6-year history of AH (WHO criteria, 1996) and with other components of metabolic syndrome [3]. The patients were divided into 5 age-matched groups: group 1, isolated AH; group 2, AH in combination with abdominal obesity (AO); group 3, AH in combination with HLP; group 4, AH in combination with AO and HLP; and group 5, AH in combination with AO, HLP, and impaired glucose tolerance (IGT). The control group included 15 healthy volunteers in whom epidemiological examination for CHD risk factors at the State Research Center of Preventive Medicine revealed none signs of metabolic syndrome.

The blood from the ulnar vein was taken from fasting subjects in the morning. The contents of total CH, triglycerides, and HDL CH in blood serum were measured by enzymatic methods using Human diagnostic kits on an Airone 200 automatic analyzer after precipitation of low-density lipoproteins (LDL) with sodium phosphotungstate in the presence of $MgCl_2$. Quality control of lipid assay was performed according to the requirements of the Federal System for Evaluation of the Quality of Clinical Laboratory Tests.

Apoprotein A1 content was measured by the method of immunonephelometry on a Behring automatic analyzer using antisera and control calibration reagents purchased from the same company.

HDL phospholipid content was measured in the supernatant after precipitation of serum LDL. Phospholipids were extracted with a 2:1 (v/v) chloroform-methanol mixture by the method of Folch. The concentration of HDL phospholipids was estimated after mineralization by the reaction with ammonium molybdate and ascorbic acid. Individual phospholipids were separated by two-dimensional thin-layer chromatography on glass plates with silica gel in chloroform-methanol-water (65:25:4 v/v) and chloroform-methanol-ammonium hydroxide (14:6:1, v/v) systems. Phospholipid spots were developed in iodine vapors and the corresponding zones were scraped off. Phospholipid standards were obtained from Sigma. The relative content of phospholipids of each class was determined after mineralization by the reaction with hydrazine hydrochloride (by phosphorus content) and expressed in percents of HDL phospholipid content [2].

The results were analyzed by Student's t test. The differences were significant at $p<0.05$.

RESULTS

The mean content of total CH and triglycerides in blood samples of group 1 patients did not differ from the control (Table 1). However, the patients with isolated AH were characterized by lower content of HDL phospholipids. This was associated with higher ratio of HDL CH to HDL phospholipids (0.630±0.060 *vs*. 0.400±0.037 in the control, $p<0.05$) and changes in phospholipid composition: the relative content of phosphatidylcholine (lecithin) decreased and the relative contents of lysophosphatidylcholine (lysolecithin), sphingomyelin, phosphatidylethanolamine, and cardiolipin increased.

Groups 2 and 1 did not differ by all test parameters. HDL CH content in group 3 patients was similar to that in group 1 patients. In group 3 patients, the level of HDL CH did not differ from that in group 1 patients, but the concentrations of apoprotein A1 and HDL phospholipids were higher. The composition of HDL in group 4 patients was similar to that in group 1 patients. Group 5 patients were characterized by highest concentration of triglycerides and lower content of HDL CH, but the contents of apoprotein A1 and HDL phospholipids and phospholipid composition in these patients did not differ from those in group 1 and 4 patients.

Our results suggest not only lower content of HDL phospholipids, but also variations in the phospholipid composition of HDL (lower percent of phosphatidylcholine and higher percent of sphingomyelin and lysophosphatidylcholine) observed in patients with isolated AH or AH associated with other components of metabolic syndrome probably determine decreased activity of processes of re-

TABLE 1. Serum Lipid Content and HDL Parameters in Patients with Isolated AH and AH Associated with Various Components of Metabolic Syndrome (*M*±*m*)

Parameter, mg/dl	Groups					
	control (*n*=15)	1 (*n*=10)	2 (*n*=14)	3 (*n*=12)	4 (*n*=26)	5 (*n*=20)
Total CH	174±3.7	178±3.9	175±5.4	239±1.4*x	235±7.0*x	231±6.4*x
Triglycerides	67±6.4	84±10.4	84±7.1	104±16.2*	181±17.0*x	200±18.9*x
HDL CH	50±3.1	46±1.8	46±1.7	48±2.4	44±1.4	41±1.6*x
Apoprotein A1	132±4.2	123±6.8	119±8.4	147±6.8*x	128±3.7	127±5.0
HDL phospholipids	126±5.4	77±5.1*	78±57*	95±5.5*x	94±4.2*x	85±5.6*
Percent of total phospholipid content						
phosphatidylcholine	74.1±1.03	62.3±2.40*	60.5±1.55*	62.4±2.06*	61.2±1.18*	61.4±1.54*
lysophosphatidylcholine	10.8±0.73	14.1±1.17*	16.4±1.04*	16.1±1.72*	16.1±1.02*	14.6±0.79*
sphingomyelin	11.3±0.48	14.9±0.56*	15.6±1.14*	13.6±0.79*	15.1±0.70*	14.8±0.94*
phosphatidylethanolamine	2.9±0.41	5.3±0.78*	4.2±0.63*	4.6±0.46*	4.6±0.45*	5.3±0.40*
cardiolipin	0.9±0.19	3.4±0.88*	3.3±0.50*	3.5±0.50*	3.3±0.38*	3.9±0.49*

Note. $p<0.05$: *compared to the control group; xcompared to group 1.

verse CH transport (acceptance of CH from cell membranes, its esterification, transport of CH esters to LDL, and interaction with liver cells). These data are of considerable importance, because the role of phospholipids in HDL-mediated reverse transport of CH is proven [9-10] and a correlation was found between HDL phospholipid content and the severity of coronary atherosclerosis [11,12]. Moreover, taking into account the data on phospholipid exchange between serum lipoproteins and cell membranes, it can be hypothesized that changes in the content and composition of HDL phospholipids lead to changes in phospholipids of cell membranes, which can modify their physicochemical properties (increase in microviscosity or destruction with resultant dysfunction of cell membranes). Changes in phospholipid composition of HDL were detected in patients with essential AH [5] and were considered as the fact leading to changes in physicochemical properties of cell membranes and ion channels and impairment of transport of Na^+, K^+, and Ca^{2+}, which contributes to the development of AH. Parameters of HDL that determine their CH-acceptor and CH-transport properties did not differ in patients with isolated AH and AH associated with other components of metabolic syndrome, probably because isolated AH is accompanied by disturbances in phospholipid metabolism in the liver, where HDL are synthesized.

Thus, changes in phospholipid composition of HDL in patients with isolated AH and AH associated with other components of metabolic syndrome can led to structural changes in the cell membrane and impairment of HDL function, which increases the risk of atherosclerosis development.

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