PHARMACOLOGY

Dynamics of Interrelationships between the Content of Lipoprotein Particles, Fibrinogen, and Leukocyte Count in the Plasma from Patients with Coronary Heart Disease Treated with Kwai

I. F. Chernyad'eva, S. V. Shil'nikova, A. N. Rogoza, and V. V. Kukharchuk

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The hypolipidemic effect of Kwai, a preparation based on garlic powder (*Allium sativum*), depended on the initial content of cholesterol and/or triglycerides. This effect was most pronounced in patients with coronary heart disease with initial cholesterol >7.0 mmol/liter and triglyceride >1.92 mmol/liter. After treatment with Kwai the correlation between triglyceride content and leukocyte count remained unchanged, the correlation between cholesterol content and leukocyte count disappeared, fibrinogen concentration decreased by 11%, and no correlation was found between fibrinogen content and leukocyte count.

Key Words: *Allium sativum; garlic; lipoproteins; fibrinogen; leukocytes; inflammation; coronary heart disease*

There is no agreement regarding pharmacological activity of garlic preparations (*Allium sativum*). The discrepancy is related to the presence of various biologically active compounds in these preparations. The content of allicin (allyl-2-propene-thiosulfinate) formed from alliin (S-allyl cysteine sulfoxide or S-2-propenyl cysteine sulfoxide) in the reaction catalyzed by alliinase (alliin lyase or alliin alkyl-sulphenate-lyase, EC 4.4.1.4) serves as a quality criterion for preparations from finely ground and dried garlic. Here we evaluated the efficiency of biologically active compounds of Kwai preparation based on dry garlic powder (*Allium sativum*, Lichtwer Pharma) in normalizing the content of plasma lipoproteins, fibrinogen level, and leukocyte count in patients with coronary heart disease (CHD). The elationships between these processes was evaluated.

A. L. Myasnikov Institute of Cardiology, Russian Research-and-Production Center for Cardiology, Russian Ministry of Health, Moscow

MATERIALS AND METHODS

A clinical trial was performed in 28 men (39-62 years) with postinfarction cardiosclerosis and stable angina pectoris (functional class II-III). Inclusion criteria were CHD, postinfarction period >1 year, and triglyceride content <4 mmol/liter, exclusion criterion was hereditary hypercholesterolemia. The scheme of treatment, dosage of antianginal preparations, and diet remained unchanged throughout the study. Hypolipidemic preparations and probucol were withdrawn 2 and 6 months before the start of observations, respectively. Kwai preparation standardized by allicin level (300 mg, Lichtwer Pharma Gmbh, registration certifi-

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TABLE 1. Contents of Lipids and Fibrinogen and Leukocyte Count in the Plasma from CHD Patients before and after Kwai Therapy (*M*±*m*, *n*=28)

Parameter	Before therapy	After therapy	Decrease (%)
Total cholesterol, mmol/liter	6.75±0.87	6.45±0.81 (0.20)	3.81±9.84
Triglycerides, mmol/liter	2.11±0.89	1.84±1.07(0.31)	13.90±24.80
LDL cholesterol, mmol/liter	4.65±0.92	4.37±1.05 (0.29)	5.87±15.25
HDL cholesterol, mmol/liter	1.02±0.16	1.08±0.21 (0.27)	-5.23±10.38
Fibrinogen, g/liter	3.21±0.77	2.87±0.95 (0.15)	11.21±21.35
Leukocytes, $\times 10^{-9}$/liter	6.78±2.12	6.39±1.44 (0.09)	2.46±16.36

Note. Significant differences by Wilcoxon paired test are shown in brackets.

cation P-8-242 No. 00688835, Russian Ministry of Health) was given 3 times a day with meals for 12 weeks.

The blood was taken from the cubital vein after 14-h starvation at the beginning and by the end of observations. The concentrations of total cholesterol, high-density lipoprotein (HDL) cholesterol (after precipitation of apoB-containing lipoproteins), and triglycerides in blood plasma were measured by enzymatic methods on a biochemical analyzer. The content of low-density lipoprotein (LDL) cholesterol was estimated by the Friedewald formula. Fibrinogen content was measured photometrically by the method of Becker [2]. Blood leukocyte count was determined on a blood cell counter and expressed in IU.

The results were analyzed by paired Wilcoxon test.

RESULTS

The effect of Kwai on blood lipid content was insignificant in all patients (*n*=28, Table 1). Subdivision of the patients into 4 groups according to triglyceride and/or total cholesterol concentration showed that the hypolipidemic effect of the preparation increased with increasing the initial plasma cholesterol and/or triglyceride content (Fig. 1). The mechanisms underlying hypolipidemic activity of biologically active compounds of garlic preparations are poorly understood. Aqueous garlic extract modulates various stages of cholesterol synthesis and activity of 3-hydroxy-3-methylglutaryl coenzyme A reductase (HMG-CoA reductase) in primary cultures of rat hepatocytes [4]. There is little likelihood that Kwai *in vivo* modulates cholesterol biosynthesis in human liver, because of high lability of compounds *in vitro* inhibiting cholesterol biosynthesis and their rapid transformation into unidentified substances during the first contact with the blood.

Plasma level of lipids is determined by the rates of migration of apoB-48- and apoB-100-containing particles into the circulation and elimination of apolipoprotein-lipid complexes from the blood. Large lipoprotein particles containing 98% lipids and 2% proteins (chylomicrons) are synthesized in human small intestine after cleavage of dietary lipids by gastrointestinal lipoprotein lipase. It should be noted that essential polyunsaturated fatty acids are used in a native form. Fatty acids secreted with chylomicrons are resynthesizing from acetyl CoA after degradation of carbohydrates and lipids, which requires considerable amounts of NADPH. In the circulation apolipoprotein-phospholipid complexes rapidly dissociate from the surface of chylomicrons. The remaining apoB-48-lipid complexes are transported into the liver and used

TABLE 2. Plasma Lipid Concentration in Patients of Various Groups before Therapy (*M*±*m*)

Group	1 (*n*=5)	2 (*n*=6)	3 (*n*=9)	4 (*n*=8)
Total cholesterol, mmol/liter				
concentration	5.70±0.31	6.54±0.28	6.44±0.35	7.89±0.49
range	5.20-6.20	6.21-6.99	6.21-6.99	7.00-8.80
Triglycerides, mmol/liter				
concentration	1.36±0.42	1.47±0.38	2.84±0.70	2.24±0.92
range	<1.92	<1.92	1.93-4.00	1.93-4.00
LDL cholesterol, mmol/liter	3.90±0.39	4.60±0.73	4.19±0.58	5.66±0.77
HDL cholesterol, mmol/liter	1.19±0.21	1.04±0.18	0.97±0.11	0.97±0.13

for the synthesis and secretion of apoB-100-containing lipoproteins into the circulation. The decrease in plasma lipid level produced by Kwai is probably associated with inhibition of gastrointestinal lipase cleaving dietary lipids caused by Kwai components (*e.g.*, allicin). These changes reduce the availability of fatty acids for triglyceride resynthesis [3,5]. Triglyceride content decreased more significantly than cholesterol concentration, which agrees with our hypothesis.

Another possible mechanism underlying the influence of biologically active compounds of Kwai on plasma lipid content is stimulation of neutral steroid excretion with bile, which can suppress utilization of cholesterol for resynthesis of lipoproteins particles. The maximum decrease of total and LDL cholesterol (by 13.4±6.7 and 16± 10%, respectively) and increase in HDL cholesterol content (by 9.5±8.5%) were found in group 4 patients with the highest initial level of cholesterol. The observed changes probably resulted from normalization of steroid excretion, which was initially impaired in these patients. Our observations are consistent with the hypothesis that the impaired secretion of cholesterol with bile can contribute to the development of hypercholesterolemia in old people. The absence of correlations between the contents of cholesterol and fibrinogen agrees with the data on increased fibrinogen (but not cholesterol) content in patients with a history of acute myocardial infarction [6]. Plasma fibrinogen concentration depends on the individual genetic status and development of inflammatory processes. Fibrinogen is an acute-phase protein. Activation of fibrinogen synthesis in the liver can be determined by increased production of IL-6 by macrophages and monocytes in response to phagocytosis of fibrinogen degradation products [1]. Kwai decreased fibrinogen level in patients of groups 1, 2, and 4 to a similar extent (0.42, 0.45, and 0.54 g/liter, respectively). Fibrinogen content in group 3 patients receiving Kwai decreased insignificantly (0.04 g/liter). It can be hypothesized that this decrease in plasma fibrinogen level is related to the fact that allicin characterized high antibiotic activity decreased the content of proinflammatory mediators. The relationship between high content of blood leukocytes (within normal limits, *i.e.* <9000/μl or 9×10^{-9}/liter) and probability of repeated coronary events is determined by their im-

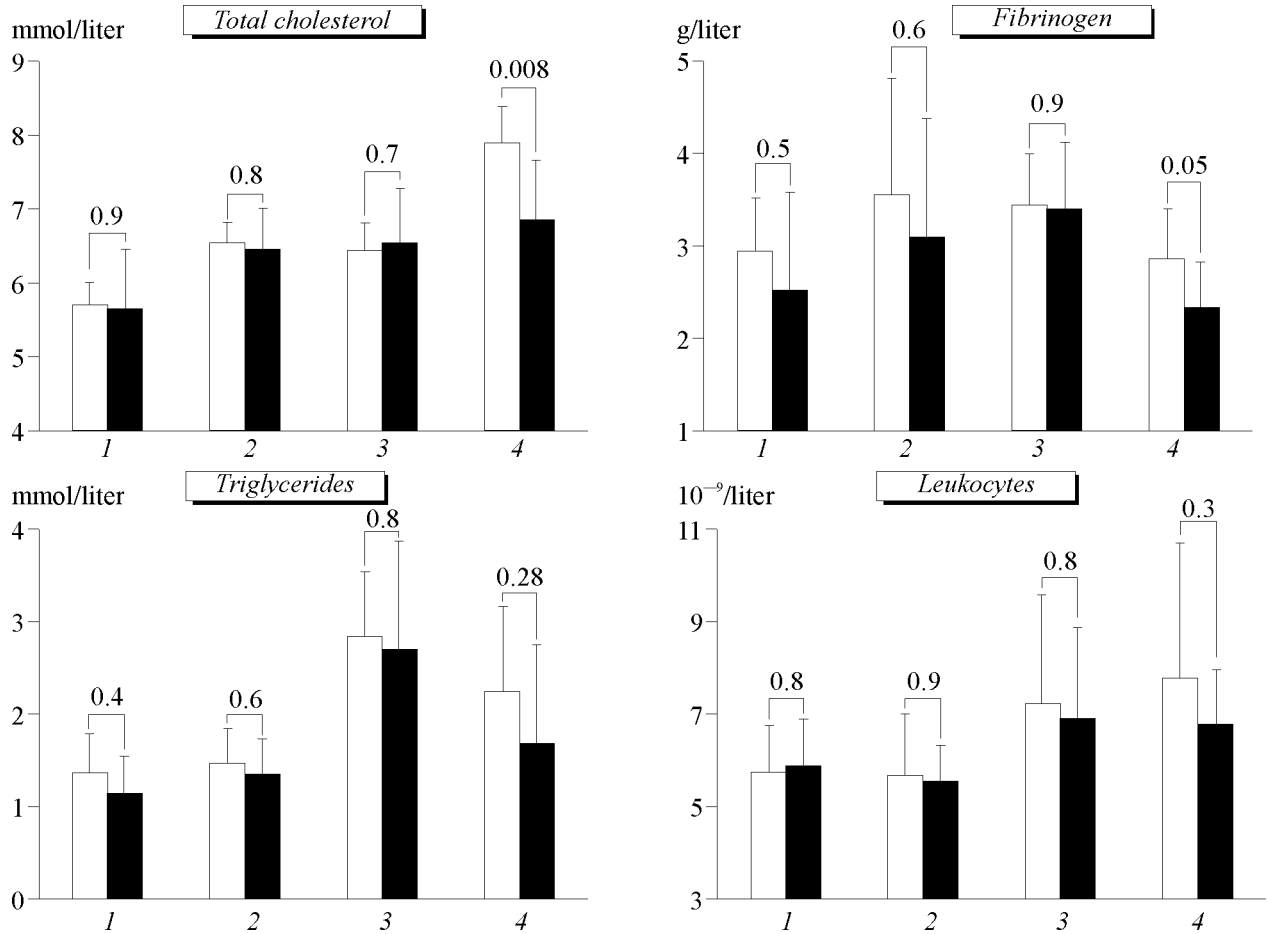

Fig. 1. Contents of lipids and fibrinogen and leukocyte count in patients differing in the initial concentrations of cholesterol and triglycerides before and after Kwai therapy. Light bars: before therapy. Dark bars: after therapy. Abscissa: groups (*1-4*).

portance for blood rheology, their increased adhesion during ischemia (particularly in the microcirculatory bed), and their role in acute and chronic damage to the endothelium via secretion of reactive oxygen species and proteolytic enzymes after adhesion to the endothelial surface.

Unfavorable effects of high levels of ApoB-containing lipoproteins on the cardiovascular system are primarily related to excessive accumulation of native LDL in the vascular wall. This leads to initiation of lipid peroxidation and/or activation of the complement system, generation of chemoattractant compounds, migration of blood monocytes and their transformation into macrophages, and synthesis of inflammatory mediators stimulating circulating blood cells (*e.g.*, leukocytes and platelets). As differentiated from LDL, binding of triglyceride-rich VLDL to scavenger receptors on macrophages does not require their pre-modification, which explains the relationship between triglyceride-rich particles and inflammation. It should be emphasized that the coefficients of correlation between cholesterol content and leukocyte count differed before and after therapy. However, the correlation coefficients for triglyceride concentration and leukocyte count remained unchanged (Table 3). No correlation was found between the contents of fibrinogen and cholesterol and between fibrinogen content and leukocyte count. It was probably related to the differences in the regulation of their synthesis.

TABLE 3. Correlation Coefficients for the Contents of Lipids and Fibrinogen and Leukocyte Count before and after Kwai Therapy

Parameter	Before therapy	After therapy
Cholesterol—triglycerides	0.21	0.07
Cholesterol—fibrinogen	-0.15	0.09
Cholesterol—leukocytes	0.34	0.06
Triglycerides—leukocytes	0.32	0.30
Triglycerides—fibrinogen	0.12	0.42
Fibrinogen—leukocytes	0.07	0.04

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