The Levels of Lipids, Lipoproteins and Apolipoproteins in Healthy People in the Central Region of the Black Sea

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SUMMARY

Lipids, lipoproteins and apolipoproteins are among the risk factors for the most serious health problem of the age — coronary artery disease (CAD). They vary from country to country, from area to area within a country, depending on genetic, environmental, dietary and many other factors. Our aim was to determine the levels of lipids, lipoproteins and apolipoproteins in healthy people in the central Black Sea region of Turkey.

Subjects included 1348 volunteers (682 women, 666 men) referred to the Medical Faculty hospital from the study area. The population consisted of healthy people or those whose disease was not affecting the metabolism of lipids. Cholesterol, triglyceride and HDL-cholesterol levels in the obtained serum samples were measured spectrophotometrically, while apolipoprotein A-I, apolipoprotein B, apolipoprotein E and lipoprotein(a) levels were measured nephelometrically.

The levels of lipid parameters were as follows: total cholesterol for men was 4.22 ± 1.00 mmol/l (mean arithmetic $\pm$ SD), triglyceride 1.20 mmol/l (0.30–4.44) [geometric mean (range)], HDL-cholesterol 0.88 ± 0.22 mmol/l, LDL-cholesterol 2.69 ± 0.85 mmol/l, apolipoprotein A-I 1.26 ± 0.22 mmol/l, apolipoprotein B 1.12 ± 0.32 mmol/l, apolipoprotein E 0.037 ± 0.012 mmol/l and lipoprotein(a) 0.25 g/l (0.03–2.75); total cholesterol for women was 4.53 ± 1.00 mmol/l, triglyceride 1.05 mmol/l (0.28–4.50), HDL-cholesterol 1.08 ± 0.26 mmol/l, LDL-cholesterol 2.87 ± 0.88 mmol/l, apolipoprotein A-I 1.45 ± 0.25 mmol/l, apolipoprotein B 1.11 ± 0.31 mmol/l, apolipoprotein E 0.039 ± 0.011 mmol/l and lipoprotein(a) 0.22 g/l (0.03–2.16).

In conclusion, our study in four different regions in Turkey reflected that the people living in the central Black Sea region are less vulnerable to the risk of CAD, although at a relatively higher risk compared to some other countries. (Jpn Heart J 1999; 40: 427–434)

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NUMEROUS studies have established the predictive role of serum total cholesterol (TC) levels as a major risk factor for CAD in various populations.¹⁻⁵⁾ When lipoprotein subfractions have been determined, cross-sectional and prospective epidemiological studies have shown that elevated low-density lipoprotein cholesterol (LDL-C) is also associated with increased risk of CAD,⁶⁻⁸⁾ whereas high high-density lipoprotein cholesterol (HDL-C) levels appear to be protective.^{1,8-13)} High levels of lipoprotein (a) (Lp[a]) also have been reported as a risk factor for CAD in most¹⁴⁻¹⁷⁾ but not all studies.^{13,18)} Severe coronary heart disease has been related with lowered serum apolipoprotein A-I (Apo A-I) concentrations, as have high concentrations of apolipoprotein B100 (Apo B).^{17,19,20)} It has been proposed that apolipoprotein E (Apo E) plays an important role in regulating the metabolism of tryglyceride (TG) and TC.²¹⁾

Our study has been undertaken to characterize the lipid, lipoprotein and apolipoprotein variables of Turkish people, who live in the central region of the Black Sea area. Turkey has a high incidence of CAD (37%²²⁾) and is unique in having a diversity of diets consumed in different regions of the country. Lipid and lipoprotein have not been studied, in general in Turkey, except for two important recent reports.^{22,23)}

Our study identified important environmental factors that impact serum lipids, lipoproteins and apolipoproteins, highlights individuals with increased risk of heart disease in the study area (3 million people) and provides baseline data for future studies.

MATERIALS AND METHODS

Study design: 1348 volunteers were selected and recruited for the study in the above mentioned region according to the following criteria: a) healthy participants or participants with illnesses not affecting their lipid metabolism, b) 12-hour fasting required of the participants over 15 years of age, and c) people with abnormal levels of blood urea nitrogen, creatinine, glucose, gamma-glutamyl transferase, aspartate amino transferase and alanine amino transferase were excluded. Blood samples were taken and centrifuged immediately. The serum samples were stored at -70°C.

Region characteristics: The study area of the central Black Sea region included the districts of Samsun (population 1,000,000), Amasya (population 350,000), Ordu (population 800,000), and Sinop (population 250,000). This region is noted for its consumption of fish and vegetable oil, especially sunflower oil. Participants from towns and villages were primarily involved in agricultural

work, whereas people living in cities were factory workers, business people and professionals.

Personal and clinical data obtained at the time of the interview: Personal data (age, sex, weight, height, and blood pressure) were obtained from all participants. Blood pressure measurements (mmHg) were determined while the participants were seated. In addition, a questionnaire with approximately 33 questions was designed to obtain relevant social information (marital status, profession, salary, smoking, alcohol consumption, physical activity), medical history (general health, cardiovascular health, physician visits) and family history (general and cardiovascular health of parents and siblings). Many of these variables will be evaluated later.

The following variables were ascertained by the bio-data questionnaire, and were found to be associated with variations in the different lipid levels determined in this study. The participants were asked to estimate the total monthly income for their families. The levels of educational background were as follows: none, primary school, middle school, high school/university. Asking how many cigarettes were smoked per day assessed smoking. The amount of alcohol consumed (beer, wine, or hard liquor) was recorded in one of three categories: none, less than or more than 5 glasses per week. The type of work, as a measure of physical activity, was determined on the basis of three general categories: physical/active work, office/shop work, and administrative/management work.

Biochemical analyses: A diagnostic lipid laboratory was established at the Ondokuzmayis University Hospital, Samsun, to measure TG, TC, HDL-C, Apo A-I, Apo B, Apo E and Lp[a]. The laboratory is affiliated with Bio-Rad Laboratories of the United States. The LDL-C was calculated using Friedwald's formula²⁴⁾ for participants with triglyceride levels < 400 mg/dl. The reagent kits for TG, TC, HDL-C were provided by Boehringer-Mannheim, Mannheim, (Germany). All samples were measured within 4 hours. Serum TC and TG levels were determined by a Hitachi multianalyzer using an enzymatic colorimetric method, while Apo A-I, Apo B, Apo E and Lp[a] levels were determined by a Behring Nephelometer 100.

Data entry and statistical analysis: Questionnaire development and data collection were accomplished with *Epi Info Version 6.0* software. The prevalence of selected categorical variables and mean levels of continuous variables were calculated utilizing the same software. Risk factors analyzed as continuous variables included age, body mass index, total cholesterol, LDL-C, HDL-C, triglycerides, Lp[a], apoprotein A-I, apoprotein B and apoprotein E. Statistical analysis was performed using the computer statistical package of SPSS/PC + V6.0 (SPSS Inc, Chicago). Descriptive statistics were calculated for the various lipid, lipoprotein and apolipoprotein levels, and stratified according to sex and age groups. Since

the distribution of TG and Lp[a] in the study population did not exhibit a gaussian distribution, their results are presented as geometric means (range) and the other results as means $\pm$ standard deviation.

RESULTS

Characterization of study populations: The number and general characteristics of the participants are summarized in Table I. The numbers of male and female participants were 666 and 682, respectively, 78% of whom were from urban areas. The ages ranged from 15–78 (36.2 ± 13.9) for males and 15–82 (36.7 ± 12.6) for females. The average weight of the males was 74.5 kg and the BMI (Body Mass Index) was 25.2, while for females they were 65.6 kg and 26.3, respectively.

Thirty-three % of the males and 25% of the females were smokers. Alcohol consumption rates were 6% and 0.2% in males and females, respectively. The diet of 16% of the study group included only unsaturated vegetable oil, while 2% consumed only saturated oil, and 82% had a diet including mixed oils. 20% of

Table I. The General Characteristics of the Participants

	Male	Female
N	666	682
Range and mean of age (yr)	15–78 (36.2 ± 13.9)*	15–82 (36.7 ± 12.6)*
Weight (kg)	74.5 ± 10.8 *	65.6 ± 11.4 *
BMI (kg/m^2)	25.2 ± 3.7 *	26.3 ± 5.1 *
Systolic Blood Pressure (mmHg)	117 ± 15 *	118 ± 19 *
Diastolic Blood Pressure (mmHg)	76 ± 9 *	76 ± 11 *
Percent of urban people (%)	75	80
Percent of Smokers (%)	33	25
Percent of Alcohol Intake (%)	6	0.2

* Mean (SD)

Table II. The Levels of Serum Lipids in the Study Population

	Male	Female
N	666	682
TC (mmol/L)*	4.22 ± 1.00 (2.20–6.24)	4.53 ± 1.00 (2.48–6.57)
TG (mmol/L)**	1.20 (0.30–4.44)	1.05 (0.28–4.50)
HDL-C (mmol/L)*	0.88 ± 0.22 (0.43–1.33)	1.08 ± 0.26 (0.53–1.61)
LDL-C (mmol/L)*	2.69 ± 0.85 (1.01–4.40)	2.87 ± 0.88 (1.08–4.66)
Apo A1 (mmol/L)*	1.26 ± 0.22 (0.82–1.7)	1.45 ± 0.25 (0.96–1.95)
Apo B (mmol/L)*	1.12 ± 0.32 (0.47–1.77)	1.11 ± 0.31 (0.50–1.72)
Apo E (mmol/L)*	0.037 ± 0.012 (0.013–0.060)	0.039 ± 0.011 (0.016–0.062)
Lp[a] (g/L)**	0.25 (0.03–2.75)	0.22 (0.03–2.16)

* arithmetic means $\pm$ SD and range, ** geometric means (range).

the females in the study were in menopause, and menstruation was irregular in 19% of these women. The educational backgrounds of the males were as follows: 3% none, 14% primary school, 12% middle school, 24% high school and 47% higher education, while for the females they were: 10% none, 28% primary school, 10% middle school 21% high school and 31% higher education. 17% of the males and 2% of the females engaged in daily exercise.

Lipid data: Serum lipid, lipoprotein and apolipoprotein levels in the study population are shown in Table II.

DISCUSSION

When we compared this study with respect to lipid and lipoprotein levels with the Turkish Heart Study and foreign countries (Table III), the following results were obtained; The TC levels in males and females (M/F) were lower than those in the Istanbul district (a large, modern, metropolitan city with a population of approximately 12 million) and in the other populations, except for Ayvalik (a small city on the Aegean Sea in a region noted for the growing of olives and the production of olive oil) and Turkish females in Germany; TG levels were lower for males than in the all studies except Chinese and Finns. In females there were no marked differences among Germans, Turks in Germany, Chinese and Finns, however the TG values of other studies were higher than those in our study population. The levels of HDL-C in M/F were lower than in all study groups. However, in females there was no difference between Istanbul and Ayvalik. The LDL-C levels in males and females were lower than in all populations except Ayvalik. The ratio of TC/HDL-C in males was lower than in

Table III. Comparison of Serum Lipid and Lipoprotein Levels (mmol/L) among Turkish Heart Study and Different Populations

	TC		TG		HDL-C		LDL-C		TC/HDL-C	
	M	F	M	F	M	F	M	F	M	F
Turks (in Central Black Sea) [#]	4.20	4.53	1.20	1.05	0.88	1.08	2.69	2.87	4.8	4.2
Turkish heart study ²²⁾										
Istanbul	5.23	4.68	1.60	1.01	0.98	1.16	3.52	3.03	5.5	4.2
Ayvalik (Aegean Sea)	4.14	4.19	1.40	1.26	0.98	1.08	2.59	2.56	4.3	3.9
Turks (in Germany) ²³⁾	4.99	4.50	1.52	0.99	0.98	1.19	3.21	2.89	5.1	3.8
Germans (in Germany) ²⁵⁾	5.64	5.46	1.34	0.99	1.21	1.55	3.72	3.41	4.6	3.5
Americans (in United States) ²⁶⁾	5.33	5.38	1.68	1.45	1.19	1.42	3.41	3.26	4.5	3.8
North Americans (White) ²⁷⁾	5.46	5.46	1.58	1.18	1.16	1.47	3.52	3.39	4.7	3.7
Canadian ⁹⁾	5.20	5.15	1.70	1.22	1.19	1.42	3.23	3.08	4.4	3.6
Chinese ²⁸⁾	5.49	5.46	1.22	1.00	1.24	1.42	3.57	3.52	4.4	3.8
Finns ²⁹⁾	5.62	5.62	1.26	1.00	1.16	1.42	3.80	3.70	4.8	3.9

[#] Our study.

Table IV. Comparison of Serum Apolipoprotein and Lipoprotein [a] Levels among Different Populations

	ApoAI		ApoB		ApoE		ApoB/ApoAI		Lp[a]	
	mmol/L						g/L			
	M	F	M	F	M	F	M	F	M	F
Turks (in Central Black Sea) [#]	1.26	1.45	1.12	1.11	0.037	0.040	0.88	0.76	0.25	0.22
Japanese ³⁰⁾	1.28	1.41	0.84	0.82	0.039	0.042	0.65	0.58	—	—
Americans ³¹⁾	—	—	1.03	0.96	—	—	—	—	—	—
Yugoslavians ³²⁾	1.48	1.51	1.21	1.08	—	—	0.81	0.71	—	—
Chinese ²⁸⁾	1.34	1.46	1.15	1.06	—	—	0.85	0.72	—	—
Finns ²⁹⁾	1.38	1.58	1.21	1.09	—	—	0.87	0.68	0.09	0.08
North Americans (White) ²⁷⁾	—	—	—	—	—	—	—	—	0.14	0.15

Our study.

Istanbul and Turks in Germany, but higher than in the other study groups. In females, the ratio of TC/HDL-C was higher than the reported studies, except for Istanbul.

As a result, the levels of lipids and lipoproteins of the study population were lower than those from the other regions of Turkey, except for Ayvalik. In the study region, due to the high consumption of fish and vegetable oils, especially sunflower oil, there is a positive effect on CAD risk compared to the other regions of Turkey. The levels of TC, TG, HDL-C and LDL-C of the study population tended to be lower than those in populations outside Turkey. The low levels of TC, TG and LDL-C in the Turkish population are an advantage, however, the low level of HDL-C is a disadvantage for the development of CAD since the ratio of TC/HDL-C, one of the important markers for determining the risk of CAD, is very high.

Table IV shows apolipoprotein and Lp[a] levels after comparing our study with those in foreign countries. The serum Apo AI levels demonstrated that females in the central Black Sea region tended to have lower mean levels than the other populations. However, Apo B levels in male Turks were similar to Chinese, higher than the Japanese and Americans, and lower than the Yugoslavians and Finns. The levels of Apo B in females were higher than the Japanese and similar to other populations. The Apo E level was relatively lower than that in Japanese. The ratio of Apo B/Apo AI, which is an important indicator for CAD, and the level of Lp[a], which is an independent risk factor, are higher than those in other populations. As can be seen from Table V, the levels of Apo B and Lp[a] and the ratio of Apo B/Apo AI were higher. Thus, it can be concluded that the Turkish population has a high potential risk for CAD.

In summary, our study has characterized serum lipid, lipoprotein and apolipoprotein levels in people living in the central Black Sea region in Turkey.

Thus, our study of four different regions in Turkey indicate that the central Black Sea region is less vulnerable to the risk of CAD. However, our study group had high risk factors for CAD when compared to foreign populations. The low level of HDL-C in Turks and Turks in Germany suggests that it is likely to have a genetic basis. In addition, the reasons for an increase in the lipid levels are as follows: (1) They tend to lead a sedentary life style, (2) increases in body weight, (3) negative dietary habits and (4) menopause in women. We believe that reducing calorie intake, using monounsaturated or polyunsaturated vegetable oils instead of saturated fatty acids, increasing physical activity (by sport or exercise), and women in menopause having hormone replacement therapy, will decrease the risk of CAD.

REFERENCES

1. National Cholesterol Education Program. Second Report of the Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel II). *Circulation* 1994; 89: 1329.
2. Stamler J, Wentworth D, Neaton JD. Is the relationship between serum cholesterol and risk of premature death from coronary heart disease continuous and graded? Findings in 356, 222 primary screens of the Multiple Risk Factor Intervention Trial (MRFIT). *JAMA* 1986; 256: 2823.
3. Anderson KM, Castelli WP, Levy D. Cholesterol and mortality: 30 years of follow-up from the Framingham Study. *JAMA* 1987; 257: 2176.
4. Connelly PW, Maclean DR, Horlick L, O'Connor B, Petrasovits A, Little A, Canadian Heart Health Surveys Research Group. Plasma lipids and lipoproteins and the prevalence of risk for coronary heart disease in Canadian adults. *CMAJ* 1992; 146: 1977.
5. Ettinger WH, Wahl PW, Kuller LH, *et al.* Lipoprotein lipids in older people: result from the Cardiovascular Health Study. *Circulation* 1992; 86: 858.
6. Gordon T, Kannel WB, Castelli WP, Dawber TR. Lipoproteins, cardiovascular disease and death: the Framingham study. *Arch Intern Med* 1981; 141: 1128.
7. Wilson PWF, Garrison RJ, Castelli WP, Feinleib M, McNamara PM, Kannel WB. Prevalence of coronary heart disease in the Framingham Offspring Study: role of lipoprotein cholesterol. *Am J Cardiol* 1980; 46: 649.
8. Grundy SM. Role of low-density lipoproteins in atherogenesis and development of coronary heart disease. *Clin Chem* 1995; 41: 139.
9. Willems JP, Saunders JT, Hunt DE, Schorling JB. Prevalence of coronary heart disease risk factors among rural blacks: a community-based study. *South Med J* 1997; 90: 814.
10. Barter PJ, Rye K-A. High-density lipoproteins and coronary heart disease. *Atherosclerosis* 1996; 121: 1.
11. Watkins LO, Neaton JD, Kuller LH. Racial differences in high-density lipoprotein cholesterol and coronary heart disease incidence in the usual-care group of the Multiple Risk Factor Intervention Trial. *Am J Cardiol* 1986; 57: 538.
12. Gordon DJ, Probstfield JL, Garrison RJ, *et al.* High-density lipoprotein cholesterol and cardiovascular disease: four prospective American studies. *Circulation* 1989; 79: 8.
13. Tervahauta M, Pekkanen J, Kivinen P, *et al.* Prevalence of coronary heart disease and associated risk factors among elderly Finnish men in the Seven Countries Study. *Atherosclerosis* 1993; 104: 47.
14. Kostner GM, Avogaro P, Cazzolato G, Marth E, Bittolo Bon G, Quici GB. Lipoprotein (a) and the risk for myocardial infarction. *Atherosclerosis* 1981; 38: 51.
15. Dahlen GH, Guyton JR, Attar M, Farmer JA, Kautz JA, Gotto AM. Association of levels of lipoprotein (a), plasma lipids and other lipoprotein with coronary artery disease documented by angiography.

- Circulation 1986; 74: 758.
16. Rosengren A, Wilhelmsen L, Eriksson E, Risberg B, Wedel H. Lipoprotein (a) and coronary heart disease: a prospective case-control study in a general population sample of middle-aged men. *BJM* 1990; 301: 1248.
 17. Genest J Jr, McNamara JR, Ordovas JM, *et al.* Lipoprotein cholesterol, apolipoprotein A-I and B and lipoprotein (a) abnormalities in men with premature coronary artery disease. *J Am Coll Cardiol* 1992; 19: 792.
 18. Jauhiainen M, Koskinen P, Ehnholm C, *et al.* Lipoprotein[a] and coronary heart disease risk: a nested case-control study of the Helsinki Heart Study participants. *Atherosclerosis* 1991; 89: 59.
 19. Maciejko JJ, Holmes RD, Kottke AB, *et al.* Apolipoprotein A-I as a marker of angiographically assessed coronary-artery disease. *N Engl J Med.* 1983; 309: 385.
 20. Vergani C, Trovato G, Dioguardi N. Serum total lipid, lipoprotein cholesterol, apoprotein A and B in cardiovascular disease. *Clin Chim Acta* 1978; 87: 127.
 21. Eto M, Watanabe K, Iwashima Y, *et al.* Apolipoprotein E polymorphism, and hyperlipidemia in type II diabetics. *Diabetes* 1986; 35: 1374.
 22. Mahley RW, Palaoğlu EK, Atak Z, *et al.* Turkish heart study: lipids, lipoproteins and apolipoproteins. *J Lipid Res* 1995; 36: 839.
 23. Onat A, Şurdum-Avcı G, Şenocak M, Örnek E, Gözükar Y. Plasma lipids and their interrelationship in Turkish adults. *J Epidemiology Community Health* 1992; 46: 470.
 24. Friedwald WT, Levy RI and Frederickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972; 18: 499.
 25. Assmann G, Schulte H. Relation of high-density lipoprotein cholesterol and triglycerides to incidence of atherosclerotic coronary artery disease. (the PROCAM experience) *Am J Cardiol* 1992; 70: 733.
 26. Johnson CL, Rifkind BM, Sempos CT, *et al.* Declining serum total cholesterol levels among US adults. The National Health and Nutrition Examination Surveys. *JAMA* 1993; 269: 3002.
 27. Jenner JL, Ordovas JM, Lamon-Fava S, *et al.* Effects of age, sex and menopausal status on plasma lipoprotein(a) levels: The Framingham Offspring Study. *Circulation* 1993; 87: 1135.
 28. Fong PC, Tam SC, Tai Y, Lau C, Lee J, Sha Y. Serum lipid and apolipoprotein distributions in Hong Kong Chinese. *J Epidemiol Community Health* 1994; 48: 356.
 29. Leino A, Impivaara O, Kaitsaari M, Jarvisalo J. Serum concentrations of apolipoprotein A-I, apolipoprotein B and lipoprotein(a) in a population sample. *Clin Chem* 1995; 41: 1633.
 30. Noma A, Hata Y, Goto Y. Quantitation of serum apolipoprotein A-I, A-II, B, C-II, C-III ve E in healthy Japanese by turbidimetric immunoassay: reference values and age- and sex-related differences. *Clinica Chimica Acta* 1991; 199: 147.
 31. Contois JH, McNamara JR, Lammi-Keefe CJ, *et al.* Reference intervals for plasma apolipoprotein B determined with a standardised commercial immunoturbidimetric assay: result from the Framingham Offspring Study. *Clin Chem* 1996; 42: 515.
 32. Zunic G, Jelic-Ivanovic Z, Spasic S, Stojiljkovic A, Majkic-Singh N. Reference values for apolipoproteins A-I and B in healthy subjects by age. *Clin Chem* 1992; 38: 566.