CETP TaqIB polymorphism in Turkish adults: association with dyslipidemia and metabolic syndrome

Türk yetişkinlerinde CETP TaqIB polimorfizmi: Dislipidemi ve metabolik sendrom ilişkisi

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ABSTRACT

Objective: In this study, our aim was to investigate the association of cholesterol ester transfer protein (CETP) TaqIB polymorphism with the likelihood of metabolic syndrome (MetS).

Methods: Study was designed as a cross-sectional analysis of the Turkish Adult Risk Factor follow-up study. Randomly selected sample of 1585 persons were included in the analyses. Genomic DNAs were isolated and the genotyping was performed using TaqMan system. ANOVA, Chi-square, univariate analyses and logistic regression models were used to investigate the association of genotypes with clinical and biochemical measurements.

Results: The frequencies of the B1B1, B1B2 and the B2B2 genotypes were 33.3%, 46.4% and 20.3%, respectively. The B2B2 genotype was associated with elevated high-density lipoprotein -cholesterol (HDL-C) levels ($p<0.0001$). After adjusting for sex and age B2B2 individuals had 15.9% higher HDL-C levels than B1B1 individuals. Furthermore, the likelihood of dyslipidemia was lower in the presence of the B2B2 genotype (30.9% non-dyslipidemic vs. 69.1% dyslipidemic, $p=0.001$) when compared to the other genotypes. Moreover, in a logistic regression model comprising age and environmental factors, B1 allele carriers showed higher odds ratios of 1.49 (OR=1.49, 95% CI; 1.03-2.14, $p=0.032$) for MetS only in females.

Conclusions: These results suggest that B1 allele is associated with MetS in adult females. However, TaqIB polymorphism appears not associated with the components of MetS other than atherogenic dyslipidemia in adult Turkish population.

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Key words: CETP TaqIB, metabolic syndrome, dyslipidemia, HDL cholesterol, Turkish adults, logistic regression analysis

ÖZET

Amaç: Bu çalışmada, kolesteril ester transfer proteini (CETP) TaqIB polimorfizmi ile metabolik sendrom (MetS) gözlenme riski arasındaki ilişkinin araştırılması amaçlanmıştır.

Yöntemler: TEKHARF takip çalışması dahilinde kesitsel olarak dizayn edilen bu çalışmada, rastgele seçilen 1585 kişinin genomik DNA'sı ayrıştırıldı ve TaqMan sistemi kullanılarak genotipleme yapıldı. ANOVA, Ki-kare, tek-yönlü analiz ve lojistik regresyon modelleri kullanılarak genotiplerin klinik ve biyokimyasal ölçütler ile ilişkisi incelendi.

Bulgular: B1B1, B1B2 ve B2B2 genotiplerinin frekansları sırası ile %33.3, %46.4 ve %20.3 olarak tespit edildi. B2B2 genotipinin yüksek HDL-K (yüksek dansiteli lipoprotein-kolesterol) seviyeleri ile ilişkili olduğu gözlendi ($p<0.0001$). Çalışma grubu, cinsiyet ve yaş için ayarlandıktan sonra, B2B2 genotipindeki bireylerin, HDL-K seviyelerinin B1B1 genotipindeki bireylere göre, %15.9 daha yüksek olduğu tespit edildi. Ayrıca, diğer genotiplerle karşılaştırıldığında, B2B2 genotipi varlığında dislipidemi gözlenme riskinin, daha düşük olduğu belirlendi (%30.9 non-dislipidemik ve %69.1 dislipidemik, $p=0.001$). Yaş, çevresel faktörler ve genotiplerin dahil edildiği bir lojistik regresyon modelinde, B1 allel taşıyıcısı kadınlarda, MetS için 1.49 kat (OR=1.49, %95 GA; 1.03-2.14, $p=0.032$) daha yüksek oranda risk olduğu tespit edildi.

Sonuç: Bu sonuçlar B1 allelinin kadınlarda MetS ile ilişkili olduğunu göstermektedir. Ancak, yetişkin Türk toplumunda TaqIB polimorfizminin aterojenik dislipidemi haricinde MetS'un diğer bileşenleri ile ilişkili olmadığı gözlenmiştir. *(Anadolu Kardiyol Derg 2008; 8: 324-30)*

Anahtar kelimeler: CETP TaqIB, metabolik sendrom, dislipidemi, HDL kolesterol, Türk yetişkinleri, lojistik regresyon analizi

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Introduction

Cholesteryl ester transfer protein (CETP) is an enzyme that facilitates the exchange of triglyceride and cholesterol between lipoproteins and involves in the reverse transport of cholesterol (1). Cholesteryl ester transfer protein plays an important role in high-density lipoprotein cholesterol (HDL-C) catabolism and also in the determination of HDL size and subclass distribution (2).

The human CETP gene consists of 16 exons encompassing 25 kbp on chromosome 16q21 (3, 4). This protein is primarily expressed in liver, spleen, and adipose tissue, and lower levels have been detected in small intestine, adrenal gland, heart, kidney, and skeletal muscle (5). Several polymorphisms of the CETP gene are found to be responsible for the differences in CETP activity and HDL-C level (6). Patients with CETP deficiency have elevated HDL-C levels and decreased low-density lipoprotein cholesterol (LDL-C) plasma levels (2). One of the most studied polymorphisms at CETP locus is TaqIB polymorphism which occurs as a silent base change by guanine to adenine nucleotide substitution at the 279th nucleotide position in the first intron of the gene (7). The less common allele, B2, is associated with decreased CETP activity and mass that mimics a mild form of CETP deficiency. In normolipidemic subjects, this allele is observed to associate with increased levels of serum HDL-C concentration as a result of CETP activity reduction (8-10). However, this association is observed as a population specific characteristic (11, 12) and is found to be highly influenced by environmental factors, such as alcohol consumption and tobacco smoking (10, 13, 14).

The aim of this study was to examine a possible correlation of the CETP TaqIB polymorphism with metabolic syndrome (MetS) occurrence in a sample of the Turkish Adult Risk Factor (TARF) Study (15), representative of Turkish adults. The interaction between environmental risk factors such as exercise, smoking and alcohol consumption were included in the analyses.

Methods

Study population

The study was designed as a cross-sectional analysis of the Turkish Adult Risk Factor (TARF) follow-up study. Design and methodology of the TARF Study have been previously described (15). Briefly, participants were randomly selected from residents of all 7 different regions of Turkey, and attended the 4 surveys 2000 through 2006. Data were obtained for history of the past years via a questionnaire, physical examination of the cardiovascular system and recording of a resting electrocardiogram. Unselected 1585 subjects (773 male and 812 female) were examined for their CETP TaqIB genotype.

Study subjects were unrelated and they gave their written consent to participate in the study after being informed of its nature. The study protocol was approved by the Ethics Committee of the Istanbul Medical Faculty, Istanbul University.

Definitions

Individuals with metabolic syndrome (MetS) were identified when 3 out of the 5 criteria of the National Cholesterol Education Program (ATP III) (16) were met, modified for prediabetes (fasting glucose 100-125 mg/dl (17)) and further for abdominal obesity using as cutpoint ≥95 cm in men, as recently assessed in the Turkish Adult Risk Factor study (18, 19). Atherogenic dyslipidemia (or simply dyslipidemia) referred to combined presence of high triglyceride (≥150mg/dl) and low HDL-C (<40 mg/dl for men and <50 mg/dl for women) values as defined by the ATP III.

Measurement of risk factors

Never- and former smokers combined (as non-smokers) and smokers formed the categories in cigarette smoking. Anyone consuming alcohol once a week or more was considered as an alcohol user. Weight was measured without shoes in light indoor clothes using a scale. Body mass index (BMI) was calculated as weight divided by height squared (kg/m^2). Waist circumference was measured with a tape (Roche LI95 63B 00), the subject standing and wearing only underwear, at the level midway between the lower rib margin and the iliac crest.

Blood samples were collected after an 11-hour or longer fasting. Samples were shipped within a few hours on cooled gel packs to Istanbul to be stored at -75°C, until analyzed at the Yıldız Technical University. Serum concentrations of total cholesterol, fasting triglycerides, glucose, and HDL-C (directly without precipitation) were determined using enzymatic kits from Roche Diagnostics with a Hitachi 902 autoanalyzer. Concentrations of apolipoprotein B and A-I were measured by Behring kits and nephelometry (BN Prospec, Behring Diagnostics, Westwood, MA).

DNA isolation and analysis of the CETP TaqIB polymorphism genotypes

Genomic DNA was extracted from peripheral blood leucocytes using a QIAmp^R DNA Maxi KIT (Qiagen, Hilden, Germany). Genotyping was performed using the TaqMan technology (ABI 7900HT, Applied Biosystems, UK). DNA amplification was set up in 384 well plates (ABGENE Ltd.) Typical 5µl PCR reaction consisted of 5ng dried DNA, 2.5µl Jumpstart TAQ ready mix (Sigma, #D6442), 0.125µl Assay mix [TaqMan probes (VIC-CCCTAACTCGAACCC, FAM-CCCTAACTTGAACCC) and primers (5'-GCCAGGTATAGGGATTTGTGTTTGT-3', 5'-CCCCTA-ACCTGGCTCAGATC-3')], and 2.375µl distilled water. PCR was carried out on a MBS 384 thermal cycler (Thermo Electron, UK) using the following conditions: 95^{o}C for 5min, 95^{o}C for 15 sec, 60^{o}C for 1 min (40 cycles). Allelic discrimination was assessed using the TaqMan software.

Statistical analysis

All statistical analyses were performed using Windows SPSS version 10.0 software (Chicago, IL, USA). Genotypic and allelic distributions were compared using the Chi-square test. Hardy-Weinberg equilibrium was computed for the expected genotype distribution. One-way ANOVA analyses were done and two-tailed t-tests were used to compare continuous variables-expressed as means and standard deviation (SD)- while categorical variables were compared using the Chi-square test. Furthermore, post-hoc Tukey analyses were done for the comparison of the groups for the analyzed variables. A two-tailed P value of <0.05 was considered statistically significant. Due to skewness, logarithmic value of triglyceride level was used in all statistical analyses. Univariate analyses were used to test the association of the covariates with HDL-C levels.

Logistic regression analyses were performed to investigate the association of the environmental factors and CETP TaqIB genotypes (independent variables) with low HDL-C levels

(dependent variable, Model 1) and with MetS (dependent variable in Model 2). Logistic regression models were used to derive maximum likelihood estimates of odds ratios (OR) and associated 95% confidence intervals (CI). The 95% CI not overlapping 1 was considered statistically significant.

Results

Subject characteristics

To investigate the association of CETP TaqIB polymorphism and MetS components in Turkish population, we analyzed a total of 1585 subjects (773 males and 812 females) from TARF Study population and genotyped for CETP TaqIB polymorphism. A summary of demographic, and biochemical characteristics is provided in Table 1. Allele frequency of the less common B2 allele was 43% (Table 2). The distribution of the alleles among males and females was consistent with Hardy-Weinberg equilibrium (Table 2).

Association of TaqIB polymorphism with plasma HDL-C levels and lipoproteins

Regardless of sex, TaqIB genotype was significantly associated with HDL-C plasma levels (Table 3). In the univariate analysis where age, sex and genotypes were included in the model, B2B2 genotype was strongly associated with increased HDL-C levels ($p<0.0001$). After physical activity, smoking status, alcohol consumption and BMI were also included in the model, B2B2 genotype was strongly and independently associated with increased HDL-C levels ($p<0.0001$). After adjusting for sex and age, the mean HDL-C levels exhibited an increase of 6.12% in B1B2 individuals ($p<0.0001$) and 15.9% in B2B2 individuals ($p<0.0001$) compared to HDL-C levels in B1B1 individuals in all population (41.3 mg/dl, 43.8 mg/dl, and 47.8 mg/dl for B1B1, B1B2 and B2B2 genotypes respectively).

To investigate the association of the environmental factors on low HDL-C levels a logistic regression model was used comprising BMI, age, smoking, physical activity grade and alcohol consumption (Table 4A). Males and females were analyzed separately and cut-off values for low HDL-C concentrations were used such as <50 mg/dl for women and <40 mg/dl for men. It was observed that age and BMI were important variables predicting low HDL-C levels in both sex. However, smoking was a strong risk factor for low-HDL only in men (OR= 1.64, 95% CI; 2.72-1.19, $p=0.003$) (Table 4A) and low physical activity had no lowering effect on HDL-C levels in both sex. Interestingly, no alcohol consumption was also a risk factor for low HDL-C in men (OR= 2.55, 95% CI; 1.70-3.83, $p<0.0001$). To investigate the effect of CETP TaqIB on HDL-C, genotypes were included to the previous model involving the environmental factors. Compared with the B2B2 genotype, B1B2 and B1B1 genotypes showed significantly higher odds ratio for low HDL-C levels in men (OR=1.72, 95% CI; 1.15-2.58, $p=0.009$ and OR=2.56, 95% CI; 1.65-3.96 $p<0.0001$, respectively) and women (OR=2.08,

Table 1. Demographic and biochemical characteristics of the participants

Parameters	Total (n)	Males (n)	Females (n)
Number of participants	1585	773	812
Age, years	54.1±11.7 (1585)	54.2±11.7 (773)	54.0±11.7 (812)
BMI, kg/m^2	29.3±8.4 (1551)	28.1±10.4 (757)	30.5±5.7 (794)
TC, mg/dl	194.4±40.7 (1581)	188.7±40.2 (771)	199.9±40.4 (810)
HDL-C, mg/dl	43.8±11.9 (1581)	39.7±10.7 (771)	47.7±11.7 (810)
LDL-C, mg/dl	119.0±34.3 (1428)	114.8±33.7 (680)	122.8±34.4 (748)
logTG, mg/dl	2.1±0.2 (1428)	2.2±0.3 (680)	2.1±0.2 (748)
TC/HDL	4.7±1.4 (1581)	5.0±1.6 (771)	4.38±1.2 (810)
ApoAI, mg/dl	139.3±27.6 (750)	131.8±24.9 (358)	146.3±28.1 (392)
ApoB, mg/dl	103.7±27.9 (755)	100.6±25.9 (359)	106.6±29.3 (396)
Fasting glucose, mg/dl	97.6±35.5 (1428)	98.6±36.5 (680)	96.6±34.6 (748)
SBP, mmHg	128.1±24.0 (1567)	125.0±21.6 (766)	131.1±25.7 (801)
DBP, mmHg	80.3±12.9 (1567)	79.3±12.3 (766)	81.4±13.4 (801)
WHR	0.9±0.1 (1564)	0.9±0.1 (350)	0.9±0.1 (320)
MS, % (n)	47.4 (752)	45.4 (351)	49.3 (400)
Dyslipidemia, % (n)	30.9 (441)	34.6 (235)	27.5 (206)
Alcohol Consumption, % (n)	8 (126)	15.4 (118)	1 (8)
Smoking*, % (n)	28.6 (447)	41.8 (319)	16 (128)
Physical Activity**, % (n)	28.3 (439)	42.3 (321)	14.9 (118)

Continuous variables are represented as mean±SD, categorical variables are displayed as percentages/proportions. *Percentage of current smokers. **Percentage of individuals performing high physical activity

ApoA1 - apolipoprotein A1, apoB - apolipoprotein B, BMI - body mass index, DBP- diastolic blood pressure, HDL-C- high density lipoprotein cholesterol, LDL-C- low density lipoprotein cholesterol, logTG- logarithmic value of triglyceride, MS- metabolic syndrome, SBP- systolic blood pressure, TC- total cholesterol, TC/HDL- total cholesterol HDL ratio, WHR- waist hip ratio

95% CI; 1.41-3.07, p<0.0001 and OR=3.70, 95% CI; 2.40-5.67, p<0.0001, respectively). This genotype effect was more pronounced in women than men (Table 4B).

Association of TaqIB polymorphism with other lipids and lipoproteins

We found that higher total cholesterol (TC) / HDL-C ratios were significantly associated with B1B1 genotype in both sex (p<0.0001, Table 3). However, we observed striking sex-specific differences among lipid parameters. B2B2 females had significantly higher TC (206.8±41.3mg/dl, p=0.045) levels than B1B1 females while no significant difference existed in males. This sex-specific difference was not accompanied by any significant LDL-C level alterations among genotypes observed in females (Table 3). Additionally, B2 allele carriage (B1B2+B2B2 genotypes) was associated with higher apoA1 level in females (p=0.01). No associations were found in triglyceride levels.

Association of TaqIB polymorphism with metabolic syndrome

The prevalences of B2B2 and B1B1 genotypes in individuals with MetS (n=751) were 18.9% and 33.6% respectively. The overall population was sub-grouped considering the presence of MetS. In MetS group, we observed no difference in CETP genotype distribution for the conventional risk factors of MetS other than atherogenic dyslipidemia. In the presence of B2B2 genotype, the likelihood of atherogenic dyslipidemia was lower for MetS patients than other two genotypes (38.4%, 46.2% and 15.4% for B1B1 and B1B2 and B2B2 genotypes respectively, p=0.003). Furthermore, the MetS subjects with B2B2 genotype had higher HDL-C levels (43.5±12.1 mg/dL) than the heterozygotes (40±9.3 mg/dL) and B1B1 subjects (37.7±8.5 mg/dL, p<0.001). This was similar in non-MetS subjects, however HDL-C levels were obviously higher in all genotypes (data not shown). Moreover, the B2B2 genotype was observed to be associated with increased apoA1 levels in non-MetS subjects (136.9±27.4 mg/dL in B1B1 vs 146.3±28.2 mg/dL in B2B2, p=0.03).

To investigate the effect of age, current smoking, low physical activity, alcohol consumption and Taq1B genotypes on the MetS risk, a logistic regression model was used. In this model we included B1 allele carriage (B1B2+B1B1 genotype carriers) as a variable since B1 allele is associated with low HDL-C levels. We observed that only female B1 allele carriers had 1.49 fold higher risk for MetS than the females in B2B2 genotype (OR=1.49, 95% CI; 1.03-2.14, p=0.032) (Table 5).

Table 2. Genotype and allele frequencies of CETP Taq1B polymorphism in the studied group

Variables	Study population	Males	Females
Genotype frequency*			
B1B1, % (n)	33.3 (528)	33.1 (256)	33.5 (272)
B1B2, % (n)	46.4 (736)	47 (363)	45.9 (373)
B2B2, % (n)	20.3 (321)	19.9 (154)	20.6 (167)
Allele frequency			
B1, %	56	56	56
B2, %	43	43	43

Data are represented as percentages/proportions

*Frequencies are computed using Chi-square test

Discussion

In the study population which is a representative of Turkish adults, the frequency of the less common allele, B2, (%43, Table 2) was similar to other two studies on the Turkish population (the large Turkish Heart Study (20) and a small-sized study on coronary artery disease patients (21)) and to those in other populations (5, 22, 23). We found that likelihood of low HDL-C/high triglyceride dyslipidemia was significantly lower in the presence of B2B2 genotype than of other two genotypes. This

Table 3. Lipid profiles of males and females according to Taq1B genotypes

Variables	Males (n=773)			F*	p*	Females (n= 812)			F*	p*
	B1B1 (n=256)	B1B2 (n=363)	B2B2 (n=154)			B1B1 (n=272)	B1B2 (n=373)	B2B2 (n=167)		
Age, years	53.1±11.5	54.4±11.8	55.5±11.7	2.005	NS	54.3±11.9	53.8±11.5	54.2±11.8	0.161	NS
BMI, kg/m^2	27.9±4.1	27.5±4.3	27.7±4.2	0.854	NS	30.3±6.1	30.7±5.5	30.1±5.6	0.403	NS
TC, mg/dL	186.8±38.6	191.0±42.4	186.5±37.6	1.103	NS	198.5±40.2	197.9±39.9	206.8±41.3	3.104	0.045
HDL-C, mg/dL	37.2±8.6**	40.0±10.9**	43.3±12.2**	16.669	<0.0001	45.1±11.4#	47.5±10.7#	52.3±12.9#	20.379	<0.0001
LDL-C, mg/dL	114.4±32.8	115.4±35.5	114.2±30.9	0.081	NS	123.3±35.2	121.7±34.4	124.4±33.5	0.376	NS
logTG, mg/dL	2.2±0.2	2.2±0.3	2.1±0.2	1.567	NS	2.1±0.2	2.1±0.2	2.0±0.2	0.923	NS
TC/HDL	5.28±1.7***	5.05±1.5***	4.56±1.3***	10.405	<0.0001	4.61±1.3	4.31±1.1	4.15±1.2	8.483	<0.0001
ApoAI, mg/dL	128.5±26.0	132.3±23.2	135.1±26.3	1.770	NS	140.6±23.7	148.1±30.6	151.8±27.7	4.660	0.01
ApoB, mg/dL	99.7±23.0	102.8±26.7	97.4±28.2	1.303	NS	109.5±35.1	104.7±24.9	105.9±27.5	1.092	NS

Data are represented as mean ±SD. * F and P values for One-way ANOVA analysis

Post-hoc Tukey analyses were done for the comparison of groups: **p=0.003 (B2B2 vs. B1B2) and p<0.0001 (B2B2 vs. B1B1);***p=0.003 (B2B2 vs. B1B2) and p<0.0001 (B2B2 vs. B1B1); # p<0.0001, (B2B2 vs. B1B2) and p<0.0001 (B2B2 vs. B1B1)

ApoA1- apolipoprotein A1, apoB- apolipoprotein B, BMI- body mass index, HDL-C- high density lipoprotein cholesterol, LDL-C- low density lipoprotein cholesterol, logTG- logarithmic value of triglyceride, N- number of cases, NS- not significant, TC- total cholesterol, TC/HDL- total cholesterol HDL ratio

protective mechanism is probably due to the increased levels of HDL-C rather than decreased levels of triglyceride (latter had no significance). Additionally, we found that female B1 allele carriers had 1.49 fold higher odds ratio for MetS.

There are several well-known environmental factors influencing HDL-C levels that act through the activities of lipases or lipid transfer proteins. It is known that alcohol consumption, exercise, and female sex have a correlation with an increase HDL-C levels. On the other hand, smoking, obesity, male sex, and diet with high in polyunsaturated fat decrease HDL-C levels (6, 24). It was reported that the effects of TaqIB on the above parameters are sex-dependent and also influenced by alcohol usage, body mass index, and insulin levels (5, 24, 25). However, the confounding effects of the environmental factors and BMI did not reach to significant level in our study group. On the other hand, our finding that B2B2 genotype was associated with an increase in plasma HDL-C levels is consistent with other studies (20, 23, 26). In previous studies, significant association with carrying TaqIB B2 allele and with having lower CETP activity had been described (5, 23). This association clearly explains the HDL level increase in B2B2 individuals.

Furthermore, in this analyzed group of TARF Study population, the influence of B2B2 genotype on HDL-C levels was higher than observed in other populations. We observed that after adjusting for sex and age, the mean HDL-C levels were 15.9% higher in B2B2 individuals compared to HDL-C levels in B1B1 individuals. This percentage was approximately two-fold compared with previously stated proportion in both the Turkish population (20) and other populations (23). The difference might probably result due to characteristics of the study populations. The basic differences of these two large sized Turkish population studies involve patient characteristics (age, BMI, HDL-C levels and triglyceride levels), geographical distributions of the study groups and probably the statistical analysis (i.e., adjusting for sex and age) and also the protocol used for the measurement of HDL-C (precipitation vs. direct method).

Recently, it was reported that CETP mass was significantly increased in men with MetS and this increase might be responsible for the reduced HDL-C and reduced LDL particle diameter observed in MetS (27). However, we found that B1 allele of CETP TaqIB polymorphism had an independent effect on MetS risk in females (Table 5). The association of B1 allele with lower HDL-C levels and B2 allele with lower dyslipidemia risk are the probable explanations to this mechanism. However, the additional effects of the other polymorphisms in determination of the HDL level and metabolic syndrome should be evaluated.

Table 4. Logistic regression analyses of association of low serum HDL-C levels with environmental factors (A) and TaqIB genotypes (B) in men and women

A) Variables	Males	p	Females	p
	OR (95% CI)		OR (95% CI)	
Age, years	0.98 (0.97-0.99)	0.002	0.98 (0.97-0.99)	0.001
Physical activity grade, (low)	0.93 (0.69-1.25)	0.625	1.30 (0.89-1.90)	0.170
Alcohol consumption, (no)	2.55 (1.70-3.83)	<0.0001	0.50 (0.10-2.50)	0.401
Smoking, (yes)	1.64 (2.72-1.19)	0.003	1.22 (1.85-0.81)	0.344
BMI, kg/m^2	1.10 (1.06-1.14)	<0.0001	1.05 (1.02 -1.07)	0.001

To define the association of low HDL cholesterol with environmental factors the following parameters were included in logistic regression analysis: dependent variable - HDL-C (normal=0, low=1), the independent variables were: age, BMI, physical activity (low=1, moderate+high=0), alcohol consumption (no=1, yes=0) and smoking (never+previous smokers=0, current smokers=1). Model comprised 854 men and 932 women with 348 and 339 cases of low HDL-C concentrations (<40 mg/dl in men and<50 mg/dl in women)

BMI- body mass index, CI- confidence interval, OR- odds ratio

B) Variables	Males	p	Females	p
	OR (95% CI)		OR (95% CI)	
Age, years	0.98 (0.97-0.99)	0.004	0.97 (0.96-0.99)	<0.0001
Physical activity grade, (low)	1.07 (0.78-1.48)	0.655	1.54 (1.00-2.35)	0.048
Alcohol consumption, (no)	2.37 (1.53-3.65)	<0.0001	0.64 (0.12-3.32)	0.593
Smoking, (yes)	1.42 (1.00-2.03)	0.006	1.25 (0.80-1.97)	0.316
BMI, kg/m^2	1.11 (1.06-1.15)	<0.0001	1.04 (1.01 -1.07)	0.012
B1B2 genotype	1.72 (1.15-2.58)	0.009	2.08 (1.41-3.07)	<0.0001
B1B1 genotype	2.56(1.65-3.96)	<0.0001	3.70 (2.40-5.67)	<0.0001

To define the association of low HDL cholesterol with environmental factors and CETP Taq1B genotypes the following parameters were included in logistic regression analysis: dependent variable - HDL-C (normal=0, low=1), the independent variables were: age, BMI, physical activity (low=1, moderate+high=0), alcohol consumption (no=1, yes=0), smoking (never+previous smokers=0, current smokers=1) and genotypes (B1B1= 2, B1B2=1, B2B2=0). Model comprised 738 men and 773 women with 307 and 293 cases of low HDL-C concentrations (<40 mg/dl in men and<50 mg/dl in women)

BMI- body mass index, CI- confidence interval, OR- odds ratio

Table 5. Multiple logistic regression analysis for association of metabolic syndrome risk with environmental factors and TaqIB genotypes in men and women

Variables	Males		Females	
	OR (95% CI)	p	OR (95% CI)	p
Age, years	1.01(0.99-1.02)	0.148	1.04 (1.02-1.05)	<0.0001
Smoking, (yes)	0.65 (0.47-0.88)	0.006	0.60 (0.40-0.92)	0.020
Physical activity grade, (low)	1.33 (0.98-1.80)	0.065	1.36 (0.90-2.06)	0.150
Alcohol consumption, (yes)	0.88 (0.58-1.33)	0.537	0.23 (0.03-1.92)	0.175
B1 allele carriers	0.94(0.65-1.36)	0.821	1.49 (1.03-2.14)	0.032

To define the association of metabolic syndrome risk the following parameters were included in logistic regression analysis: dependent variable - Metabolic Syndrome (no=0, yes=1), the independent variables were: age, physical activity (low=1, modarate+high=0), alcohol consumption (no=0, yes=1) and smoking (never+previous smokers=0, current smokers=1) and genotypes (B1B1+B1B2=1, B2B2=0). Model comprised 750 men and 784 women with 345 and 385 cases with metabolic syndrome
BMI- body mass index, CI- confidence interval, OR- odds ratio

Study limitations

The limitation of the study was the number individuals analyzed for their serum samples. The lipid and lipoprotein measurements were done in a non-selected subgroup of the genotyped individuals. However, this situation evolved from the longitudinal nature and organization of the TARF study.

Conclusion

In conclusion, CETP TaqIB polymorphism is a crucial determinant of the HDL-C concentration. Our results suggest that B1 allele of TaqIB polymorphism is associated with MetS in women. However, the polymorphism appears not associated with the components of MetS other than atherogenic dyslipidemia.

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