Are IVS4 SNPs of *OLR1* gene associated with coronary artery disease: Is there a linkage between IVS4 SNPs?

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Abstract

Background. The *OLR1* gene has been identified as a candidate gene for coronary artery disease (CAD). Six single-nucleotide polymorphisms (SNPs) of the *OLR1* gene located within intron 4 (IVS4-27G>C, IVS4-73C>T, IVS4-14A>G), intron 5 (IVS5-70A>G, IVS5-27G>T) and 3'UTR (188C>T) comprise a linkage disequilibrium (LD) block, which is strongly associated with the elevated risk of CAD.

Objectives. We aimed to investigate the effects of the *OLR1* IVS4-14A>G and -73C>T SNPs on metabolic parameters in Turkish CAD patients, and the linkage between these 2 genetic variants.

Material and methods. The present study was carried out in 97 CAD patients and 78 healthy individuals. The OLR1 IVS4 genotypings were performed by polymerase chain reaction – restriction fragment length polymorphism (PCR-RFLP) method.

Results. Serum high-density lipoprotein (HDL) cholesterol levels and body mass index (BMI) were higher in control subjects with IVS4-73CC genotype than in T allele carriers (CT+TT) (respectively, $p = 0.002$ and $p = 0.024$), while BMI values were lower in patients with CC genotype ($p = 0.046$). Patients with IVS4-14G allele (AG+GG) had a statistically higher low-density lipoprotein (LDL) cholesterol level ($p = 0.027$) than patients with -14AA genotype. Also the systolic blood pressure (SBP) levels were statistically higher in IVS4-73C allele carriers (CT+CC) than in non-carriers (TT) ($p = 0.045$). A strong linkage between IVS4-14A>G and -73C>T SNPs of the *OLR1* gene was detected in patients ($D' = 0.76$).

Conclusions. Our results indicated that the intron 4-14A>G and -73C>T SNPs of the *OLR1* gene can be inherited together. The present data also suggests that the *OLR1* gene may contribute to the development of hypercholesterolemia in patients with CAD.

Key words: single nucleotide polymorphism, coronary artery disease, serum lipids, linkage disequilibrium, *OLR1* gene

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Introduction

Elevated plasma and tissue levels of oxidized low-density lipoprotein (ox-LDL), as well as traditional risk factors, including age, sex, diabetes mellitus, hypercholesterolemia, high blood pressure, obesity, and smoking, were shown to have contributory effects on the development of atherosclerotic lesions.[1,2] Under some pathological conditions, such as acute myocardial infarction (AMI) and coronary artery disease (CAD), increased levels of ox-LDL have been reported. Ox-LDL, which also regulates LOX-1 activity, shows pro-atherogenic effects through oxidized low density lipoprotein (lectin-like) receptor 1 (*OLR1, LOX-1*).[3–6] The *OLR1* gene has been identified as a candidate gene which may be associated with AMI and CAD.[3,4] *OLR1* is induced by pro-atherogenic stimuli and by inflammatory cytokines, and it is upregulated in ischemia reperfusion injury in the rat.[2,5,7]

The *OLR1* gene is mapped on 12p13.1-p12.3 constituting 6 exons and 5 introns. First 3 exons show functional consistency with different functional domains of protein (cytoplasmic, transmembrane and neck domain), and the other 3 exons encode the carbohydrate recognition domain and are similar to those in other C-type lectin genes.[8] Seven polymorphisms have been identified on the *OLR1* gene. Six of them (in intron 4, 5 and 3'UTR region) are in linkage disequilibrium (LD).[9]

Association studies have shown a role for the *OLR1* gene variants in AMI susceptibility. In particular, 6 of the 7 single-nucleotide polymorphisms (SNPs) of the *OLR1* gene located within intron 4 (IVS4-27G>C, IVS4-73C>T [rs3736234], IVS4-14A>G [rs3736235]), intron 5 (IVS5-70A>G, IVS5-27G>T) and 3'UTR (188C>T) comprise a linkage disequilibrium block, which is strongly associated with the elevated risk of CAD.[10,11]

A new splicing isoform (lacking the exon 5) of the *OLR1* gene with a new function is named LOXIN.[9] In humans, the incidence of myocardial infarction was negatively associated with the LOXIN mRNA and protein expression levels. LOXIN lacks the ligand-binding site, but interacts with the full-length *OLR1* receptors by blocking their cellular expression, ox-LDL binding activity, and uptake.[12] The expression of LOXIN mRNA is dramatically related to the *OLR1* LD polymorphism. Macrophages of subjects with the "no risk" polymorphism have higher levels of mRNA as well as protein expression than macrophages of subjects carrying the risk haplotype.[12] The *OLR1* gene IVS4-14A>G (rs3736235) polymorphism influences the transcription of 2 isoforms OLR1/LOXIN, whose ratio could allow the identification of subjects who are at cardiovascular disease risk. Subjects carrying the mutant IVS4-14G allele express less LOXIN than those with the wild type IVS4-14A allele.[12]

The aim of the present work was to investigate the effects of the *OLR1* IVS4-14A>G (rs3736235) and -73C>T (rs3736234) polymorphisms on lipid parameters in Turkish CAD patients and to show the linkage disequilibrium between these 2 polymorphisms.

Material and methods

Patient selection and clinical investigation

The study protocol was approved by both the Ethical Committee of the Faculty of Medicine and the Research Fund of Istanbul University, Turkey. All the procedures were in accordance with the Helsinki Declaration laid down in 1964 and its later amendments. All participants in study signed informed consent forms in accordance with ethics guidelines regarding the study. *OLR1* IVS4-73C>T and -14A>G gene polymorphisms were studied in 97 patients with CAD (31 women, 66 men). The presence of CAD was documented by an angiography in patients with acute coronary syndrome. Angiographic inclusion criteria were: ≥50% stenosis of at least 1 major coronary vessel because of atherosclerosis, and a vascular event, defined as myocardial infarction, percutaneous transluminal coronary angioplasty or coronary artery by-pass grafting.[13] Patients were included irrespective of concomitant risk factors for atherosclerosis such as smoking, arterial hypertension and diabetes mellitus.

To identify normal distribution of the *OLR1* IVS4-73C>T and -14A>G genotypes, we enrolled a control population of 78 healthy unrelated individuals (35 women, 43 men). This group primarily included the spouses of CAD patients and volunteers. A coronary angiography was not performed on these individuals, and therefore the presence of atherosclerotic coronary arteries could not be excluded. However, none of these individuals had any history of vascular event. Before the subjects were admitted into the study, their medical history was taken with special emphasis on coronary risk factors, including smoking, family history of CAD, hypertension, diabetes mellitus, and hyperlipidemia. The study was approved by the Ethics Committee of the Faculty of Medicine, Istanbul University, and written informed consent was obtained from each participant.

Genotyping

Genomic DNA was extracted from human leukocyte nuclei isolated from whole blood by standard methods.[14] IVS4-73C>T and -14A>G genotypes were performed by the method described by Trabetti et al.[7]

Statistical analysis

All statistical analyses were performed by SPSS for Windows v. 20.0 (SPSS Inc., Chicago, USA). To evaluate the difference in the occurrence of the *OLR1* IVS4-73C>T and -14A>G genotypes in the study groups, the χ^2 test was used. Differences in the distributions of genotypes according to clinical phenotypes (presence or absence of left ventricular hypertrophy – LVH) were assessed by using the χ^2 test in 2 × 2 tables. In order to determine the relative

risks, odds ratios (ORs) and 95% confidence intervals (CIs) were used. Lipid and the other parametric analyses were compared by the Student's t and ANOVA tests. The linkage between the -73C>T and -14A>G polymorphisms was assessed by using D' and r^2 values obtained through the Haploview Program (Broad Institute, Cambridge, USA) and p-values of <0.05 were considered as statistically significant.

Results

Patient characteristics

Demographic, biochemical and clinical data is summarized in Table 1. There were significant differences in the frequencies of the total cholesterol (TC) levels (p = 0.023) and smoking (p = 0.001) between patients with CAD and the control subjects. However, no significant differences were detected in systolic and diastolic blood pressures, sex, BMI, concentrations of serum TG, LDL-cholesterol, HDL-cholesterol, and very low-density lipoprotein (VLDL) cholesterol between patients with CAD and the control subjects (p > 0.05).

Table 1. Characteristics of the study population

Baseline characteristics	Control (n = 78)	CAD (n = 97)
Age [years] (X ±SD)	58.05 ±10.43	59.93 ±9.70
Sex (women/men) (n)	35/43	31/66
BMI [kg/m^2] (X ±SD)	25.19 ±3.63	26.08 ±3.10
SBP [mm Hg] (X ±SD)	121.53 ±13.46	123.56 ±27.02
DBP [mm Hg] (X ±SD)	73.14 ±8.97	77.02 ±16.62
TC [mmol/L] (X ±SD)	4.87 ±1.37	5.40 ±1.36*
TG [mmol/L] (X ±SD)	1.56 ±0.71	1.64 ±0.98
HDL-cholesterol [mmol/L] (X ±SD)	1.01 ±0.35	0.98 ±0.19
LDL-cholesterol [mmol/L] (X ±SD)	3.19 ±1.24	3.41 ±1.15
VLDL-cholesterol [mmol/L] (X ±SD)	0.73 ±0.40	0.74 ±0.54
Smoking (%)	46.3	69.1†
Type 2 DM (%)	–	24.2
Hypertension (%)	–	38.2
LVH	–	25.6

The results are shown as X (mean) ±SD (standard deviation).
CAD – patients with coronary artery disease; BMI – body mass index; SBP – systolic blood pressure; DBP – diastolic blood pressure; TC – total cholesterol; TG – triglyceride; HDL – high-density lipoprotein; LDL – low-density lipoprotein; VLDL – very low-density lipoprotein; LVH – left ventricular hypertrophy; n – number of individuals; * p = 0.023; † p = 0.001.

Distributions of the *OLR1* IVS4 -14A>G and IVS4 -73C>T genotypes

The distributions of genotypes and alleles of *OLR1* IVS4-73C>T and -14A>G are shown in Table 2. No significant deviation from Hardy-Weinberg equilibrium (HWE) was observed for *OLR1* IVS4 polymorphisms in the study groups (p > 0.05). In addition, statistical analysis revealed no significant difference in the genotype and allele frequencies of *OLR1* IVS4 in the study groups (p > 0.05).

Table 2. The distributions of *OLR1* IVS4-73C>T and IVS4-14A>G genotypes and alleles in the study groups

Distributions of studied *OLR1* SNPs		Control group	CAD group
IVS4-73C>T		n = 78	n = 91
Genotypes n(%)	CC	4 (5.1)	8 (8.8)
	TT	25 (32.1)	28 (30.8)
	CT	49 (62.8)	55 (60.4)
C allele n(%)		57 (36.53)	71 (39.01)
T allele n(%)		99 (63.46)	111 (60.98)
IVS4-14A>G		n = 76	n = 97
Genotypes n(%)	AA	26 (34.2)	22 (22.7)
	GG	11 (14.5)	15 (15.5)
	AG	39 (51.3)	60 (61.9)
A allele n(%)		91 (59.86)	104 (53.60)
G allele n(%)		61 (40.13)	90 (46.36)

X^2 test was used to compare genotypes in the study group. For determining allele frequencies gene count method was used.
CAD – patients with coronary artery disease; n – number of individuals.

Haplotype analysis

IVS4-14G allele carriers in the control group also carried IVS4-73T allele (p = 0.008). Moreover, it was determined that most patients with IVS4-14 G allele carry IVS4-73T allele (p < 0.001) (Table 3). Therefore, we assessed haplotype analysis and found an observed LD with D'= 0.741 between IVS4-14A>G and -73C>T polymorphisms (p > 0.05) (Fig. 1). On the other hand, haplotype frequencies of IVS4-14A>G and IVS4-73C>T were estimated, and the

Fig. 1. The linkage disequilibrium (LD) analysis of IVS4-14A>G and IVS4-73C>T

LD plot was generated by HaploView software v. 4.2. The pairwise LD value (D^1 = 0–100) is given in the colored pink square with "74", which indicates D^1 = 0.74. A value of 100 (D^1 = 1) represents maximum possible linkage disequilibrium.

Table 3. Interactions between *OLR1* IVS4-14A>G and IVS4-73C>T variants

OLR1 IVS4-14AG	Control group		CAD group	
	OLR1 IVS4-73CT		*OLR1* IVS4-73CT	
	CC	CT+TT	CC	CT+TT
AA	4 (18.2%)	18 (81.8%)	7 (33.3%)	14 (66.7%)
AG+GG	–	47(100%)	1 (5.5%)	66 (98.5%)
p-value	p = 0.008*		p < 0.001*	

X^2 test was used to compare genotypes in the study group. For determining allele frequencies gene count method was used.
*p < 0.05 indicates statistical significance (Fisher's exact test).

Table 4. Haplotype associations of *OLR1* IVS4-14A>G and -73C>T polymorphisms

Haplotype association	Frequencies			X^2	p-value
	overall	patients	controls		
GT	0.399	0.422	0.369	0.795	0.3726
AC	0.338	0.339	0.335	0.001	0.9695
AT	0.221	0.193	0.258	1.524	0.217
GC	0.043	0.046	0.038	0.216	0.6425

To evaluate the combined effect of *OLR1* IVS4-14A>G and -73C>T polymorphisms on CAD, the haplotype frequencies for significant loci and the standardized disequilibrium coefficient (D') for pairwise linkage disequilibrium (LD) were calculated using r^2 and LOD values (LOD is the log of the likelihood odds ratio, r^2 is the correlation coefficient between the 2 loci). The 1st allele indicates *OLR1* IVS4-14A>G; the 2nd allele indicates *OLR1* IVS4-73C>T polymorphism.
D' = 0.741; r^2 = 0.274; LOD = 9.04; LD = 74.

following 4 haplotypes with frequency were observed: G-T (39.9%); A-C (33.8%); A-T (22.1%); G-C (4.3%) (p > 0.05) (Table 4).

Association of the *OLR1* IVS4-14A>G and IVS4-73C>T SNPs with lipid and metabolic parameters

Patients with IVS4-14 G allele (AG+GG genotypes) have significantly higher LDL-cholesterol levels (p = 0.027) (Fig. 2) and higher TC, as close to statistical significance (p = 0.063) (Fig. 3) than patients with IVS4-14AA genotypes (Table 5). Also, we found that the controls carrying the -14G allele were prone to high levels of LDL-cholesterol (12.5%) and TC (10.08%) without any statistical significance (Table 5).

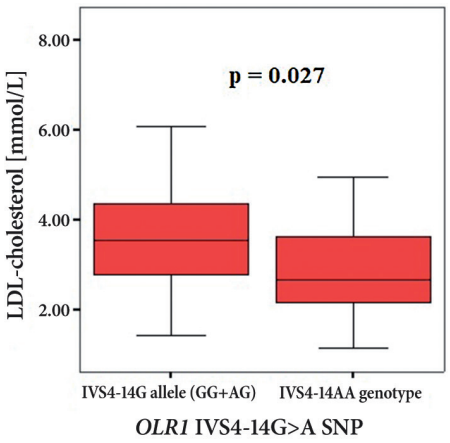

Fig. 2. The distribution of LDL-cholesterol levels between *OLR1*-14G>A polymorphism

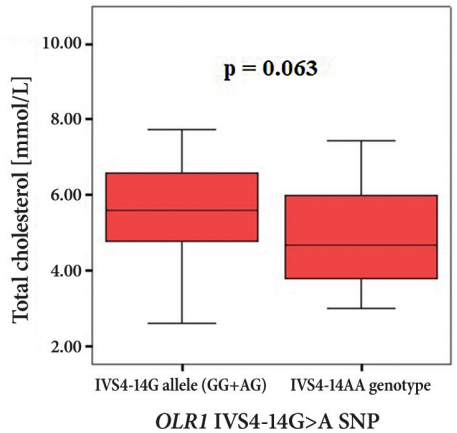

Fig. 3. The distribution of TC levels between *OLR1*-14G>A polymorphism

As shown in Table 5, in patients with IVS4-73 T allele (CT+TT), the BMI values increased compared to those with IVS4-73 CC genotype (p = 0.046), while they decreased in the controls with T allele compared to those with CC genotype (p = 0.024). When we analyzed the effects of IVS4 -73 variant on the serum lipid profile, it was observed that the HDL-cholesterol levels were statistically lower in the control subjects with IVS4-73 T allele than in those with IVS4-73 CC genotype (p = 0.002) (Table 5). Also, SBP levels were statistically higher in the IVS4-73C allele carriers (CT+CC) than in the IVS4-73TT genotype carriers (C allele: 127.17 ±30.09 vs TT genotype: 115.83 ±19.76; p = 0.045) in the group of patients (Table 5).

Discussion

Genetic risk factors are considered to be responsible for about half of CAD events. In recent years, several functional SNPs of the *OLR1* gene have been associated with CAD in humans. One of these functional SNPs is the IVS4-14A>G.[15] IVS4-14A allele encodes a truncated *OLR1* splice isoform, LOXIN, which lacks a part of the extracellular domain resulting in reduced binding capacity for ox-LDLs, and the cellular expression of the full-length *OLR1* receptors and their ox-LDL binding activity.[15,16] LOXIN expression provides increased resistance to ox-LDL-induced macrophage apoptosis and atherogenesis in vitro. It was shown that IVS4-14A allele carriers are protected from cardiovascular disease, whereas homozygous IVS4-14G allele carriers are predisposed to cardiovascular disease in vivo.[3,15] However, the effect of the *OLR1* IVS4-14A>G SNP on lipoprotein metabolism has not yet been investigated.

Chen et al. reported that the intron 4/G allele frequencies of the *OLR1* gene were higher in the white population than in the black population in Women's Ischemia Syndrome Evaluation (WISE) Study (49.2% vs 18.8%; p < 0.001).[10] In the WISE study, it was found an association between the common genetic 3'UTR 188C>T variation in the *OLR1*

Table 5. The effects of *OLR1* IVS4-73C>T and IVS4-14A>G genotypes and alleles on serum lipoprotein levels, BMI and blood pressure in patients group

Groups	*OLR1* IVS4-14A>G genotypes				*OLR1* IVS4-73C>T genotypes			
	AA	AG+GG	GG	AG+AA	CC	CT+TT	TT	CT+CC
Control								
TC [mmol/L]	4.66 ±1.10	5.23 ±1.35	5.31 ±1.15	4.99 ±1.33	5.02 ±0.91	4.90 ±1.40	4.88 ±1.29	4.92 ±1.43
TG [mmol/L]	1.70 ±1.29	1.79 ±0.75	1.77 ±0.90	1.77 ±0.99	2.41 ±0.97	1.69 ±0.94	1.90 ±0.87	1.66 ±0.99
HDL-C [mmol/L]	0.97 ±0.27	0.98 ±0.37	1.05 ±0.52	0.96 ±0.30	1.02 ±0.35	0.89 ±0.08§	1.06 ±0.45	0.97 ±0.36
LDL-C [mmol/L]	3.03 ±0.97	3.41 ±1.38	3.35 ±0.81	3.27 ±1.34	3.22 ±0.72	3.23 ±1.28	3.03 ±1.11	3.33 ±1.31
VLDL-C [mmol/L]	0.77 ±0.33	0.80 ±0.60	0.81 ±0,41	0.78 ±0.47	0.80 ±0.46	0.74 ±0.43	0.76 ±0.40	077 ±0.45
BMI [kg/m^2]	25.46 ±3.44	24.93 ±3.21	23.53 ±3.06	25.40 ±3.25	29.26 ±3.92	25.03 ±3.55¤	25.51 ±4.82	25.17 ±3.14
SBP [mm Hg]	122.72 ±16.08	120.42 ±7.43	119.09 ±7.00	121.55 ±11.48	117.50 ±5.00	120.97 ±11.58	121.19 ±16.27	120.60 ±8.67
DBP [mm Hg]	70.0 ±9.25	73.51 ±7.21	70.98 ±9.44	72.67 ±7.99	67.50 ±5.00	73.58 ±8.60	75.71 ±14.54	72.20 ±6.79
Patients								
TC [mmol/L]	4.96 ±1.32	5.58 ±1.35*	5.67 ±151	5.39 ±1.34	5.03 ±1.41	5.42 ±1.37	5.27 ±1.55	5.43 ±1.31
TG [mmol/L]	1.46 ±0.73	1.76 ±1.15	1.81 ±0.85	1.67 ±1.11	1.45 ±0.77	1.71 ±1.12	1.65 ±0.83	1.71 ±1.19
HDL-C [mmol/L]	1.05 ±0.31	1.09 ±0.30	0.94 ±0.21	1.02 ±0.31	1.00 ±0.45	1.10 ±0.29	1.01 ±0.32	0.98 ±0.28
LDL-C [mmol/L]	2.94 ±1.04	3.58 ±1.15†	3.57 ±1.32	3.40 ±1.12	2.78 ±1.06	3.45 ±1.16	3.38 ±1.20	3.39 ±1.17
VLDL-C [mmol/L]	0.66 ±0.31	0.78 ±0.30	0.83 ±0.42	0.74 ±058	0.64 ±0.30	0.77 ±0.58	0.74 ±0.41	0.78 ±0.42
BMI [kg/m^2]	26.25 ±3.35	26.10 ±3.07	26.33 ±3.04	26.09 ±3.16	24.50 ±1.76	26.41 ±3.20¥	26.38 ±3.12	26.22 ±3.19
SBP [mm Hg]	120.0 ±19.76	124.13 ±29.11	122.0 ±25.69	123.36 ±27.54	124.37 ±19.89	123.97 ±28.75	115.83 ±19.76	127.17 ±30.09Ψ
DBP [mm Hg]	75.45±11.00	76.95 ±18.15	74.33 ±18.98	77.04 ±16.27	78.12 ±14.12	77.37 ±17.57	72.71 ±15.25	79.27 ±17.69

The results are shown as mean ±standard deviation. TC – total cholesterol; TG – triglyceride; HDL-C – high-density lipoprotein cholesterol; LDL-C – low-density lipoprotein cholesterol; VLDL-C – very low-density lipoprotein cholesterol; BMI – body mass index; SBP – systolic blood pressure; DBP – diastolic blood pressure. Statistical analyses were performed by using the Student's t-test. * p = 0.063; † p = 0.027; § p = 0.002; ¤ p = 0.024; ¥ p = 0.046; Ψ p = 0.045.

gene and stenosis severity of CAD. They also suggested that the IVS4 -14A>G, the intron 5 T>G and the 3'UTR 188C>T polymorphisms of the *OLR1* gene were in significant linkage disequilibrium, and therefore exhibited similar genotype/allele frequencies. They asserted that all 3 polymorphisms could be considered a single marker for discussion purpose. They also found no significant association between *OLR1* polymorphisms (in the intron 4 G>A the intron 5 T>G, and the 3'UTR T>C) and plasma lipid levels (TC, LDL-cholesterol, HDL-cholesterol, and TG).[10]

Trabetti et al. found a similar distribution frequency of the IVS4-73TT homozygous allele among AMI and non-AMI cases.[7] They observed the association between the IVS4-73C>T and CAD, as close to statistical significance (p = 0.065). Mango et al. examined 7 *OLR*1 polymorphisms (K167N, 3'UTR 188C>T, IVS4+27G>C, IVS4-73C>T, IVS4-14A>G, IVS5-70A>G, IVS5-27G>T) and found that 6 of them (except K167N) comprised a linkage disequilibrium block behaving as a single SNP.[11]

In the present study, no significant associations were observed between *OLR*1 IVS4-73C>T and -14A>G genotypes and alleles and the risk of CAD (p > 0.05). In general, serum lipid pattern was shown to indicate a predisposition to hyperlipidemic profile, as an independent CAD risk factor. In patients with CAD, IVS4-14G allele was associated with moderately higher cholesterol levels (in excess of 12%) in its carriers than in the IVS4-14AA genotype carriers (p = 0.063). When we investigated the effects of the IVS4-73C>T SNP on serum lipid levels and other characteristics in the controls, we observed that the HDL-cholesterol levels and BMI were lower in the IVS4 -73 T allele (CT+TT genotype) carriers than in non-carriers (CC genotype) (p = 0.002 and p = 0.024, respectively). In healthy controls with IVS4-73CC genotype, BMI was higher than in controls with TT and CT genotypes (p > 0.05). In contrast to detrimental effects of IVS4-73 T allele on lipids, it was related to a favorable effect on BMI. Although the distribution of *OLR1* IVS4-14A>G and IVS4-73C>T SNPs was similar in patient and control groups, it was observed that the 2 IVS4 polymorphisms of the *OLR1* gene were in a very high linkage disequilibrium (D' = 0.74; r^2 = 0.274). This finding indicates that both of the IVS4 SNPs of the *OLR1* gene can be inherited together. Furthermore, the *OLR1* IVS4-14A>G SNP has an unfavorable lipid profile (high total and LDL-cholesterol levels), though it is not associated with the risk of CAD.

Several studies reported that the *OLR*1 3'UTR 188C>T and IVS4-73C>T SNPs were in a linkage disequilibrium block.[10,11] In our previous study, 3'UTR 188TT genotype was associated with increased SBP levels in patients with CAD.[1] Moreover, we found that the SBP levels were statistically higher in the normal IVS4-73C allele carriers (CT+CC) than in the rare homozygote IVS4-73TT genotype carriers (115.83 ±19.76 vs 127.17 ±30.09; p = 0.045)

in the group of patients. When we analyzed these SNPs (-73C>T and 3'UTR 188C>T), our findings showed that most of the IVS4-73C allele carriers also have *OLR*1 3'UTR 188T allele (89.5%). This finding suggests the possibility of an interaction between these 2 SNPs (-73C>T and 3'UTR 188C>T) and hypertension in the presence of CAD. Although the definite mechanism requires further research, we think that the intron 4 variations of the *OLR*1 gene may result in an increased risk of CAD by increasing the SBP levels.

As a conclusion, our study is the first one to investigate the IVS4-14A>G and -73C>T variants of the *OLR1* gene in the Turkish population. It was shown that the IVS4-14A>G and -73C>T SNPs of the *OLR1* gene comprise a linkage disequilibrium block. Our results are in agreement with the hypothesis that the intron 4 SNPs of the *OLR1* gene are inherited together. The -14A>G SNP was associated with increased levels of TC and LDL-cholesterol in the patient group, while normal homozygote -73CC genotype was associated with increased levels of HDL-cholesterol in the control subjects. The present findings suggest that the *OLR1* gene IVS4 gene variants might play a role in hypercholesterolemia as an independent CAD risk factor in the Turkish population.

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