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Scavenger receptor class B type I polymorphisms and peripheral arterial disease

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Abstract

Genetic variations of the scavenger receptor class B type I (SR-BI) have been demonstrated to be associated with plasma lipid parameters, anthropomorphic parameters, and coronary artery disease. We determined the frequency of 3 single-nucleotide polymorphisms within the SR-BI gene (*SCARB1*) in 354 patients with peripheral arterial disease (PAD) and 354 controls matched for age, sex, and diabetes and related to lipids and disease state, that is, PAD. *SCARB1* combined genotype *exon 1/intron 5/exon 8* were found to be associated with plasma total and low-density lipoprotein cholesterol levels, respectively. In terms of disease, a significant risk for PAD was observed in female subjects carrying the common allele of *exon 8* (odds ratio, 2.623; 95% confidence interval, 1.321-5.208; $P = .003$). The variant allele of *intron 5* was found to be a risk factor for PAD in men (odds ratio, 2.182; 95% confidence interval, 1.288-3.698; $P = .005$). Furthermore, the *SCARB1* combined genotype *intron 5/exon 8* proved predictive for PAD in the whole population ($P = .006$), which remained significant after correction for traditional risk factors. In conclusion, in the present study population, *SCARB1* polymorphisms not only show associations with plasma levels of total and low-density lipoprotein cholesterol, respectively, but also with the risk for PAD.

1. Introduction

The scavenger receptor class B type I (SR-BI) is a high-density lipoprotein (HDL) receptor that mediates "selective lipid uptake" of HDL-derived cholesteryl esters into cells without internalization of the major apolipoproteins of HDL, that is, apolipoprotein A-I and apolipoprotein A-II. The SR-BI complementary DNA (cDNA) was first identified in mice by expression cloning [1] followed by cloning of SR-BI homologues of several species. The human homologue of SR-BI originally identified as *CD36 and LIMPII analogous-1* was mapped to chromosome 12 [2,3]. In rodents, SR-BI was shown in in vitro and in vivo studies to function as physiologic and pathophysiologic germane receptor for HDL metabolism [4-9] and to play a role in the development of atherosclerosis [10-13]. In humans, the role of SR-BI on plasma HDL cholesterol and other lipids and lipoproteins is not well known as studies concerning SR-BI expression and function in humans are limited by the availability of samples from SR-BI–expressing tissues. However, on the genetic level, several polymorphisms have been identified in various populations, and associations were found with plasma lipid levels and anthropomorphic parameters [14-22]. Some of these studies are suggesting that *SCARB1* genotypes may play a role in the development of coronary artery disease (CAD); in female patients with CAD, combined genotypes at *exon 8* (C→T transition at cDNA 1050 base position) and *intron 5* (C→T transition) were associated with abnormal lipids [16]. The common allele of the *exon 8* polymorphism was higher in Korean patients with CAD [22]. In addition, carriers of the *exon 8* common allele showed an elevated risk for CAD [21].

In this study, we were interested whether these polymorphisms are also associated with atherogenic effects in other vascular territories, that is, with peripheral arterial disease (PAD).

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Many factors related to the development, outcome, and prevention of atherosclerosis in peripheral arterial territories are shared with coronary atherosclerosis. However, there are some differences regarding traditional risk factors, pathophysiology, genetic risk factors, and therapy [23]. Within the classic risk factors for CAD, cigarette smoking and diabetes convey the greatest risks for patients with PAD. In addition, the combination of elevated triglycerides and low HDL cholesterol, as well as sex and ethnicity are associated with peripheral artery disease [23]. As an example for pathophysiologic differences, thrombosis may have a more important role in PAD as inhibition of the platelet ADP receptor is more effective in reducing cardiovascular events in patients with PAD than with CAD [24]. Moreover, once disease is apparent in one vascular territory, there is increased risk for adverse events in other territories. Patients with PAD have a 4-fold greater risk of myocardial infarction and a 2- to 3-fold greater risk of stroke than patients without PAD.

In the present study, we further addressed the question regarding potential differences between coronary and peripheral artery diseases. Accordingly, we determined genotype frequency of 3 common *SCARB1* polymorphisms in *exon 1*, *exon 8*, and *intron 5* cross-sectionally in 354 patients with chronic atherosclerotic arterial disease of the lower extremities and 354 controls matched for age, sex, and diabetes.

2. Materials and methods

2.1. Study population

Patients included in this study were participants of the Linz Peripheral Arterial Disease (LIPAD) study [25]. Peripheral arterial disease was defined as chronic atherosclerotic disease of the lower extremities associated with typical symptoms, such as claudication or leg pain upon exertion, rest pain, or minor or major tissue loss, and was verified by interview, physical examination, sonography, and angiography. All cases with acute ischemia, defined as described previously [26], were excluded. Further exclusion criteria were PAD caused by nonatherosclerotic causes (eg, cardioembolic disease, vasculitis) and the history or presence of any malignancy. Diabetes mellitus was defined by use of any glucose-lowering medication or by fasting glucose levels of 126 mg/dL or higher. Control subjects were matched to the patients with PAD in a 1:1 design by sex, age (±2 years), and diabetes. All control subjects were patients of the Konventhospital (Linz, Austria), were generally in good health, and were admitted for treatment of minor health problems, such as cataract surgery, vertebrogenic pain, or nonvascular surgery (eg, herniotomia, varicose vein extirpation). All of them had an ankle-brachial pressure index of 1.0 or higher (median ankle brachial index, 1.17; interquartile range, 1.09-1.29; range, 1.00-2.00), no pathologic pattern of pulse waves in lower limbs by continuous-wave spectral analysis, no ischemic heart disease, and no stenosis of the internal carotid artery of 50% or greater on color duplex sonography. Smoking was classified according to recommended standards as any amount of tobacco use including abstinence of less than 1 year [26]. Arterial hypertension was defined as the use of any antihypertensive medication, or systolic blood pressure of 145 mm Hg or higher, or diastolic blood pressure of 90 mm Hg or higher as recommended by Rutherford et al [26]. Blood samples were taken from all subjects under standardized conditions after an overnight fasting period. We measured conventional cardiovascular risk factors, such as lipids, lipoproteins, fasting glucose, glycosylated hemoglobin A_{1c} (HbA_{1c}), total homocysteine, and high sensitivity C-reactive protein. Eighty-seven cases and none of the controls were on lipid-lowering medication. All subjects provided informed consent, and the protocol was approved by the review committee.

In this series of patients with PAD, the frequencies of concomitant CAD and cerebrovascular disease were 109 (31%) and 72 (20%), respectively. Furthermore, 96 patients with PAD exhibited internal carotid stenosis of 50% or greater. At enrollment, 96 patients with PAD reported a history of remote percutaneous transluminal angioplasty with or without stenting; 74 patients, a history of vascular surgery; and 20 patients, one of minor amputations, respectively. Per definition, none of the 354 control subjects had CAD, cerebrovascular disease, or an internal carotid stenosis of 50% or more. The median ankle-brachial pressure index in patients with PAD was 0.65 ± 0.29 and 1.21 ± 0.16 in controls. In addition, all examination techniques used in the LIPAD study have been described in more detail recently [25]. In addition to the present study, 2 polymorphism-based studies concerning the LIPAD population have been performed so far [25,27].

2.2. DNA analysis

Genomic DNA was isolated from buffy coat prepared from EDTA whole blood samples using the Purgene DNA isolation kit (Gentra Systems, Lund, Sweden) and was frozen at −80°C for further assessment. *SCARB1* genotyping was carried out by digestion of polymerase chain reaction (PCR) products for *exon 1* (G→A at cDNA position 4, Gly→Ser change at amino acid position 2), *intron 5* (C→T, at intron position 55), and *exon 8* (C→T at cDNA position 1050, silent mutation at amino acid position 350) [14], and/or by allelic discrimination using the 5′ nuclease assay with fluorogenic probes [28] on a Mx4000 Multiplex Quantitative PCR System (Stratagene, La Jolla, CA) as described.

2.3. Statistical analysis

Statistical analysis was performed using SPSS version 11.5 (SPSS, Chicago, IL). Differences in allele frequencies and assumption of Hardy-Weinberg equilibrium were estimated by gene counting followed by χ^2 test. Differences in mean values between cases and controls were determined using 2-tailed Student *t* test. Normality distribution for all continuous variables was checked using Kolmogorov-

Table 1
Clinical characteristics of cases and controls

Characteristic	Controls (n = 354)	Patients with PAD (n = 354)	*P*
Sex, M/F (n [%])	248 (70.1)/106 (29.9)	248 (70.1)/106 (29.9)	1.000 [a, b]
Age (y)	67.1 ± 10.6	66.8 ± 10.6	.700 [b]
Body mass index (kg/m^2)	27.0 ± 4.0	26.7 ± 4.1	.267
HbA_{1c} (%)	6.42 ± 1.54	6.70 ± 1.69	.011 [c]
Fasting glucose (mg/dL)	120.3 ± 54.5	123.1 ± 56.3	.500
Total cholesterol (mg/dL)	212 ± 44	228 ± 53	<.001
LDL cholesterol (mg/dL)	134 ± 34	152 ± 46	<.001
HDL cholesterol (mg/dL)	53 ± 17	51 ± 18	.020 [c]
Triglycerides (mg/dL)	134 ± 76	166 ±103	<.001 [c]
Total homocysteine (μmol/L)	15.6 ± 7.2	17.7 ± 7.9	<.001 [c]
hs C-reactive protein (mg/L)	7.9 ± 19.5	15.8 ± 40.8	.001
Current smoking [d] (n [%])	40 (11.3)	154 (43.5)	<.001 [a]
Arterial hypertension (n [%])	159 (44.9)	207 (58.5)	<.001 [a]
Ankle brachial index	1.21 ± 0.16	0.65 ± 0.29	<.001 [c]

hs indicates high sensitivity.
[a] Statistical test was performed with χ^2 test for categorical values.
[b] Matched variables.
[c] Statistical test was performed with Student *t* test, where logarithmic transformations were applied to nonnormally distributed parameters.
[d] Current smoking was defined as any amount of tobacco use including abstinence for less than 1 year.

Smirnov statistic, and logarithmic transformations were applied for ankle brachial index, HDL cholesterol, HbA_{1c}, homocysteine, and triglyceride plasma levels. χ^2 test was used to compare categorical variables. Differences between genotypes concerning lipid parameters were examined with analysis of variance. Relationship between *SCARB1* genotypes and PAD was tested using multivariate logistic regression with PAD as dependent variable. Current smoking, arterial hypertension, low-density lipoprotein (LDL) cholesterol, triglycerides, HbA_{1c}, and total homocysteine were included as covariates into the final model. Covariate selection was based on significant univariate differences and clinical relevance. *P* values of less than .01 were considered to indicate statistical significance to correct for multiple testing.

3. Results

3.1. Main characteristics of study population

Patients included in this study were participants of the LIPAD study [25]. Clinical characteristics of our study sample consisting of 354 patients with PAD and 354 controls matched for age, sex, and diabetes are shown in Table 1. The prevalence of current smoking and arterial hypertension was significantly lower in the control group. In addition, there was also a statistically significant difference between cases and controls with respect to their HbA_{1c}, total and LDL cholesterol, triglycerides, high-sensitivity C-reactive protein, and total homocysteine.

3.2. Genotype distribution

Frequencies of *SCARB1* polymorphisms in *exon 1*, *intron 5*, and *exon 8* were analyzed by allele-specific Taqman PCR in all study samples. Results from comparison of these measurements with data obtained using restriction digestion of amplified fragments in 20 patients were in total agreement (data not shown). The genotype distribution of all 3 polymorphisms did not differ from that expected in Hardy-Weinberg equilibrium (data not shown) and are listed in Table 2. Regarding the total population, allele frequencies of *SCARB1* polymorphisms were not different between patients with PAD and controls, respectively. However, post hoc analyses of men and women showed significant different frequencies of *intron 5* variants exclusively in men and those of *exon 8* variants exclusively in women, respectively (Fig. 1). Accordingly, a significant risk for PAD was observed in subjects carrying the common allele of *exon 8* in women (odds ratio [OR], 2.623; 95% confidence interval [CI], 1.321-5.208; $P = .003$) but not in men (OR, 1.000; 95% CI, 0.767-1.879; $P = .494$). Concerning subjects carrying the variant allele of *intron 5*, a borderline significant risk for

Table 2
Genotype frequencies of cases and controls

Genotype	Controls (n [%])	Cases (n [%])	*P*-value *
Exon 1			
GG	283 (79.9)	284 (80.2)	
GA	68 (19.2)	65 (18.4)	.752
AA	3 (0.8)	5 (1.4)	
Intron 5			
CC	311 (87.9)	288 (81.4)	
CT	39 (11.0)	63 (17.8)	.036
TT	4 (1.1)	3 (0.8)	
Exon 8			
CC	101 (28.5)	107 (30.2)	
CT	170 (48.0)	188 (53.1)	.077
TT	83 (23.4)	59 (16.7)	

* As compared by χ^2-test (Pearson).

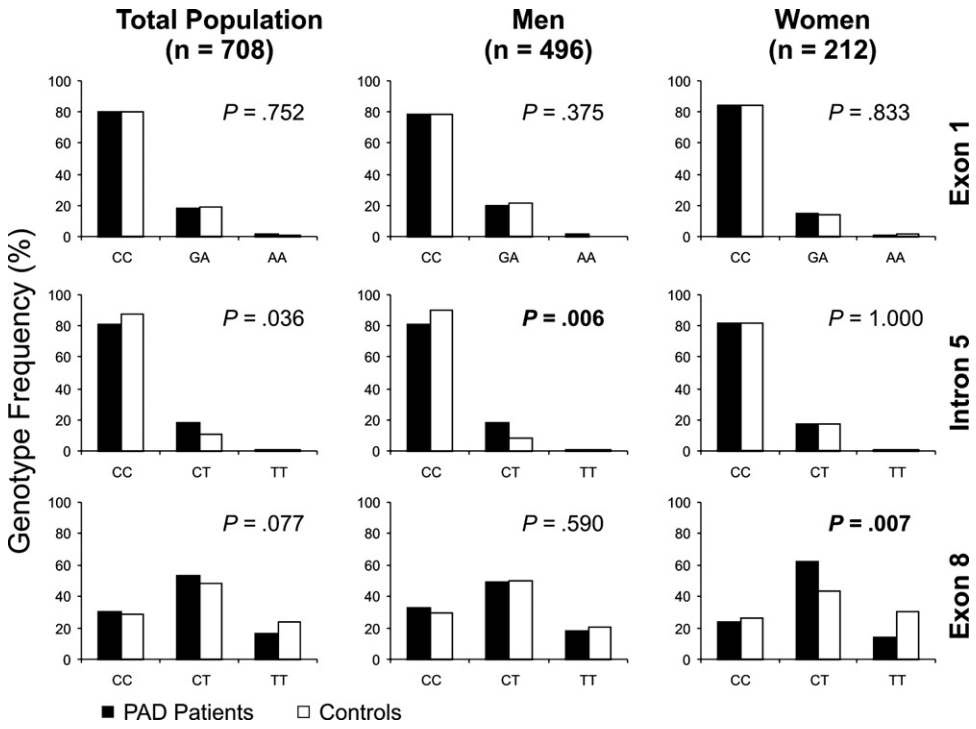

Fig. 1. Genotype frequencies of polymorphisms *exon 8* and *intron 5* shown for the total population, for men, and for women, respectively. *P* values of χ^2 tests (Pearson) comparing genotype frequencies between patients with PAD (black bars) and controls (white bars) are given.

PAD was found in the total population (OR, 1.657; 95% CI, 1.093-2.513; $P = .022$). In subgroup analysis, this association turned out to be significant in men (OR, 2.182; 95% CI, 1.288-3.698; $P = .005$) but not in women (OR, 1.000; 95% CI, 0.496-2.018; $P = 1.000$).

In agreement with previous studies, polymorphisms in *exon 8* and *intron 5* were in strong linkage disequilibrium ($P < .0001$). The linkage between these polymorphisms was found not only in the total population but also in separate analyses of men and women. Further examination of combined genotype *exon 8/intron 5* showed an association of this combined genotype with case/control status ($P = .006$, Fig. 2).

Next to univariate analysis, we performed a multivariate logistic regression analysis adjusting for classic risk factors of atherosclerosis including current smoking, arterial hypertension, LDL cholesterol, triglycerides, HbA_{1c}, and total homocysteine. Concerning individual polymorphisms, a borderline significant increased OR was found for *intron 5* (OR, 1.736; 95% CI, 1.123-2.684; $P = .013$) but not for polymorphism *exon 8* (OR, 1.536; 95% CI, 0.958-2.464; $P = .075$). However, the combined *SCARB1* genotype *intron 5/exon 8* proved predictive for PAD (OR, 2.321; 95% CI, 1.280-4.208; $P = .006$) (Table 3).

3.3. Genotype-phenotype associations

Concerning individual polymorphisms, a borderline significant association could be found between *SCARB1 exon 1* and total cholesterol (222 ± 49 vs 211 ± 50 mg/dL for genotypes GG and GA + AA, $P = .012$), as well as with LDL cholesterol (145 ± 42 vs 135 ± 41 mg/dL for genotypes GG and GA + AA, $P = .013$), respectively. In addition, we found significant associations between *SCARB1* combined genotype *exon 1/intron 5/exon 8* with total and LDL cholesterol, respectively (Table 4).

4. Discussion

Results from recent studies suggest a role of *SCARB1* genotypes for the development of CAD in humans; in female patients with CAD, combined genotypes at *exon 8*

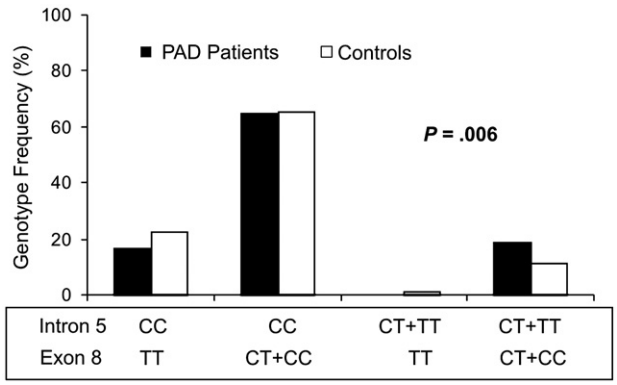

Fig. 2. Genotype frequencies of combined *exon 8/intron 5* genotypes. *P* value of χ^2 test (Pearson) comparing genotype frequencies between patients with PAD (black bars) and controls (white bars) is given.

Table 3
Multivariate logistic regression analysis for the presence of PAD including *SCARB1 exon 8* (A) and *intron 5* (B) polymorphisms, as well as combined genotype of *SCARB1 exon 8* and intron 5 polymorphisms (C), respectively

	OR of PAD	95% CI	P [a]
A.			
Current smoking [b]	7.588	4.892-11.770	<.001
Arterial hypertension	2.485	1.736-3.557	<.001
HbA_{1c} (per 0.1%)	1.014	1.003-1.025	.015
Total homocysteine (per 1 μmol/L)	1.047	1.019-1.075	.001
LDL cholesterol (per 10 mg/dL)	1.119	1.070-1.171	<.001
Triglycerides (per 10 mg/dL)	1.009	0.988-1.030	.406
Exon 8 C-allele (CC + CT vs TT)	1.736	1.123-2.684	.013
B.			
Current smoking [b]	7.370	4.760-11.411	<.001
Arterial hypertension	2.531	1.769-3.623	<.001
HbA_{1c} (per 0.1%)	1.013	1.002-1.024	.020
Total homocysteine (per 1 μmol/L)	1.045	1.107-1.073	.001
LDL cholesterol (per 10 mg/dL)	1.116	1.067-1.167	<.001
Triglycerides (per 10 mg/dL)	1.010	0.989-1.031	.346
Intron 5 T-allele (TT + CT vs CC)	1.536	0958-2.464	.075
C.			
Current smoking [b]	7.436	4.791-11.542	<.001
Arterial hypertension	2.492	1.738-3.574	<.001
HbA_{1c} (per 0.1%)	1.013	1.002-1.025	.019
Total homocysteine (per 1 μmol/L)	1.046	1.018-1.075	.001
LDL cholesterol (per 10 mg/dL)	1.117	1.068-1.169	<.001
Triglycerides (per 10 mg/dL)	1.009	0.989-1.030	.388
Exon 8/intron 5	2.321	1.280-4.208	.006

[a] Multivariate odds ratios were calculated with logistic regression analysis without variable selection technique (all variables were included simultaneously into the model). Age and sex were not included into the analysis because patients and controls were matched for these variables.
[b] Current smoking was defined as any amount of tobacco use including abstinence for less than 1 year.

and *intron 5* were found associated with an abnormal lipid profile [16]; in Korean patients with CAD, the frequency of the common allele of *exon 8* polymorphism was elevated [22]; and in the Prospective Cardiac Gene study, carriers of the *exon 8* common allele showed an elevated risk for CAD [21]. In this report we present the first study to extend these findings to patients with PAD; in women carrying the common allele of *exon 8* and in men carrying the variant allele of *intron 5*, the risk for PAD was found significantly elevated. These relationships may be explained by the known associations between *SCARB1* genotypes and plasma lipid parameters. We indeed found significant associations between *SCARB1* combined genotype *exon 1/intron 5/exon 8* with total and LDL cholesterol in our study. Further explanations may be envisioned looking at additional functions of SR-BI without considerable influence on plasma levels of lipoproteins, for example, efflux of cholesterol from foam cells within atherosclerotic plaques, which is not believed to result in significant changes of plasma lipid levels. However, data concerning the relative contribution of SR-BI to cholesterol efflux from macrophages are still controversial [29-31].

Surprisingly, we found a greater incidence of PAD in women that are heterozygous for the *exon 8* polymorphism than in women homozygous for either allele. This phenomenon may de due to a conceptual bias of this study due to use of exclusion criteria leading to diminished representation of female patients with PAD homozygous for the common allele of *exon 8*. Alternatively, this phenomenon may be due to a gene-dose effect. Accordingly, different levels of SR-BI expression in mice led to controversial effects regarding the development of atherosclerotic lesions. A moderate expression of SR-BI was shown to be antiatherogenic, but this effect was reversed by highly elevated expression levels of SR-BI in these animals [13].

Most of the associations found in this work appear to act in a sex-dependent way. Subgroup analysis of men and women showed significant different frequencies of *intron 5* variants exclusively in men and those of *exon 8* variants exclusively in women, respectively. These results were not unexpected as sex-dependent associations between *SCARB1* genotypes and various clinical parameters including lipid parameter, BMI, and risk for CAD were found in most investigations dealing with *SCARB1* polymorphisms. One explanation for this phenomenon may be a possible interaction between *SCARB1* genotype and estrogen-dependent regulation of SR-BI expression; estrogens have been shown to influence plasma HDL cholesterol levels on one hand and expression of SR-BI on the other [32,33]. In addition, the response of HDL cholesterol to hormone-

Table 4
Plasma lipoproteins by *SCARB1* combined phenotypes

Combined phenotype [a] (*exon 1/intron 5/exon 8*)	n	T-CHOL (mg/dL)	HDL (mg/dL)	LDL (mg/dL)	Triglycerides (mg/dL)
GG/CC/TT	104	226 ± 47 [b]	53 ± 21	147 ± 39	150 ± 96
GG/CC/(CT + CC)	379	218 ± 49	52 ± 17	141 ± 42	147 ± 83
GG/(CT + TT)/(CT + CC)	82	236 ± 48	54 ± 16	157 ± 42	151 ± 78
(GA + AA)/CC/TT	35	207 ± 45	53 ± 19	129 ± 41	142 ± 98
(GA + AA)/CC/(CT + CC)	81	217 ± 52	49 ± 15	140 ± 39	166 ± 126
(GA + AA)/(CT + TT)/(CT+CC)	25	192 ± 48	46 ± 15	123 ± 43	145 ± 95
		$P = .002$ [c]	$P = .106$	$P = .002$	$P = .684$

[a] Data of combined genotype 121 (n = 2) are not shown.
[b] Data are presented as mean ± SD.
[c] One-way analysis of variance.

replacement therapy in postmenopausal women has been shown to be dependent on the presence of a genetic variant in the estrogen receptor gene [34]. It has been proposed that polymorphisms within the *SCARB1* gene could, in an analogous manner, modulate the influence of estrogen on both SR-BI expression and lipoprotein parameters [16,21]. Sex-specific effects may also be explained by the fact that SR-BI is capable of exerting different functions depending on the site of expression (steroidogenic tissues vs liver and macrophages) and on parameters influencing its function, for example, cholesteryl gradient in cells (liver cell vs foam cell within atherosclerotic plaques), plasma concentration and composition of HDL particles, as well as plasma triglyceride levels influencing HDL metabolism via cholesterol ester transfer protein. Major differences are observed concerning these parameters between males and females.

Despite numerous studies having shown associations between *SCARB1* genotypes in *intron 5* and *exon 8* with various clinical parameters, the causative role of these variants is still unclear. As both variants do not lead to any change in amino acid sequence of SR-BI, possible effects must be limited to regulatory mechanisms. The *intron 5* polymorphism is not known to interfere with splicing or gene regulation. *Exon 8* polymorphism has been claimed to constitute a marker of other functional polymorphisms. However, no linkage has been found between a known functional promoter variant with both *exon 8* and *intron 5* polymorphisms [21]. In addition, this promoter variant seems to be extremely rare in white populations. As far as the functionalities of the *exon 8* and *intron 5* polymorphisms are concerned, Rodriguez et al have indeed demonstrated an association between *SCARB1* messenger RNA levels and *SCARB1 exon 8/intron 5* haplotypes in isolated peripheral blood mononuclear cells with highest levels of expression in cells carrying the variant allele of *exon 8* and the common allele in *intron 5*. Taken together with data demonstrating associations of these variants with CAD shown by others and with PAD reported in this study, these experiments suggest an atheroprotective role of SR-BI in humans.

A limitation of the present study may be the fact that the study design was cross-sectional and that the study population was a selected subgroup of the overall population with PAD as described in the Materials and methods section (white patients admitted for inpatient diagnostics and treatment of atherosclerotic PAD). Thus, the findings cannot be generalized to patients who are not of Caucasian origin, asymptomatic patients with PAD, or patients who do not meet the criteria for hospitalization. Another limitation may be due to the limited number of patients, especially in subgroup analysis. This could be the cause for the rather high ORs concerning PAD observed in our population, as studies including rather small number of patients have been shown to overestimate OR values [35].

In conclusion, showing associations between *SCARB1* polymorphisms *exon 8* and *intron 5* and PAD, we suggest that SR-BI not only is associated with the development of CAD as was shown by others, but also plays a role in the development of PAD in humans.

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References

[1] Acton SL, Scherer PE, Lodish HF, et al. Expression cloning of SR-BI, a CD36-related class B scavenger receptor. J Biol Chem 1994;269: 21003-9.
[2] Calvo D, Vega MA. Identification, primary structure, and distribution of CLA-1, a novel member of the CD36/LIMPII gene family. J Biol Chem 1993;268:18929-35.
[3] Cao G, Garcia CK, Wyne KL, et al. Structure and localization of the human gene encoding SR-BI/CLA-1. Evidence for transcriptional control by steroidogenic factor 1. J Biol Chem 1997;272:33068-76.
[4] Kozarsky KF, Donahee MH, Rigotti A, et al. Overexpression of the HDL receptor SR-BI alters plasma HDL and bile cholesterol levels. Nature 1997;387:414-7.
[5] Rigotti A, Trigatti BL, Penman M, et al. A targeted mutation in the murine gene encoding the high density lipoprotein (HDL) receptor scavenger receptor class B type I reveals its key role in HDL metabolism. Proc Natl Acad Sci U S A 1997;94:12610-5.
[6] Temel RE, Trigatti B, DeMattos RB, et al. Scavenger receptor class B, type I (SR-BI) is the major route for the delivery of high density lipoprotein cholesterol to the steroidogenic pathway in cultured mouse adrenocortical cells. Proc Natl Acad Sci U S A 1997;94:13600-5.
[7] Ueda Y, Royer L, Gong E, et al. Lower plasma levels and accelerated clearance of high density lipoprotein (HDL) and non–HDL cholesterol in scavenger receptor class B type I transgenic mice. J Biol Chem 1999;274:7165-71.
[8] Varban ML, Rinninger F, Wang N, et al. Targeted mutation reveals a central role for SR-BI in hepatic selective uptake of high density lipoprotein cholesterol. Proc Natl Acad Sci U S A 1998;95:4619-24.
[9] Wang N, Arai T, Ji Y, et al. Liver-specific overexpression of scavenger receptor BI decreases levels of very low density lipoprotein ApoB, low density lipoprotein ApoB, and high density lipoprotein in transgenic mice. J Biol Chem 1998;273:32920-6.
[10] Arai T, Wang N, Bezouevski M, et al. Decreased atherosclerosis in heterozygous low density lipoprotein receptor–deficient mice expressing the scavenger receptor BI transgene. J Biol Chem 1999;274: 2366-71.
[11] Kozarsky KF, Donahee MH, Glick JM, et al. Gene transfer and hepatic overexpression of the HDL receptor SR-BI reduces atherosclerosis in the cholesterol-fed LDL receptor–deficient mouse. Arterioscler Thromb Vasc Biol 2000;20:721-7.
[12] Trigatti B, Rayburn H, Vinals M, et al. Influence of the high density lipoprotein receptor SR-BI on reproductive and cardiovascular pathophysiology. Proc Natl Acad Sci U S A 1999;96:9322-7.
[13] Ueda Y, Gong E, Royer L, et al. Relationship between expression levels and atherogenesis in scavenger receptor class B, type I transgenics. J Biol Chem 2000;275:20368-73.
[14] Acton S, Osgood D, Donoghue M, et al. Association of polymorphisms at the SR-BI gene locus with plasma lipid levels and body mass index in a white population. Arterioscler Thromb Vasc Biol 1999;19: 1734-43.

[15] Hsu LA, Ko YL, Wu S, et al. Association between a novel 11-base pair deletion mutation in the promoter region of the scavenger receptor class B type I gene and plasma HDL cholesterol levels in Taiwanese Chinese. Arterioscler Thromb Vasc Biol 2003;23:1869-74.

[16] Knott TJ, Pease RJ, Powell LM, et al. Complete protein sequence and identification of structural domains of human apolipoprotein B. Nature 1986;323:734-8.

[17] Morabia A, Ross BM, Costanza MC, et al. Population-based study of SR-BI genetic variation and lipid profile. Atherosclerosis 2004;175: 159-68.

[18] Osgood D, Corella D, Demissie S, et al. Genetic variation at the scavenger receptor class B type I gene locus determines plasma lipoprotein concentrations and particle size and interacts with type 2 diabetes: the Framingham study. J Clin Endocrinol Metab 2003;88: 2869-79.

[19] Perez-Martinez P, Ordovas JM, Lopez-Miranda J, et al. Polymorphism exon 1 variant at the locus of the scavenger receptor class B type I gene: influence on plasma LDL cholesterol in healthy subjects during the consumption of diets with different fat contents. Am J Clin Nutr 2003;77:809-13.

[20] Plat J, Mensink RP. Relationship of genetic variation in genes encoding apolipoprotein A-IV, scavenger receptor BI, HMG-CoA reductase, CETP and apolipoprotein E with cholesterol metabolism and the response to plant stanol ester consumption. Eur J Clin Invest 2002; 32:242-50.

[21] Rodriguez-Esparragon F, Rodriguez-Perez JC, Hernandez-Trujillo Y, et al. Allelic variants of the human scavenger receptor class B type 1 and paraoxonase 1 on coronary heart disease: genotype-phenotype correlations. Arterioscler Thromb Vasc Biol 2005;25:854-60.

[22] Zhong S, Sharp DS, Grove JS, et al. Increased coronary heart disease in Japanese-American men with mutation in the cholesteryl ester transfer protein gene despite increased HDL levels. J Clin Invest 1996;97:2917-23.

[23] Faxon DP, Creager MA, SmithJr SC, et al. Atherosclerotic vascular disease conference: executive summary: atherosclerotic vascular disease conference proceeding for healthcare professionals from a special writing group of the American Heart Association. Circulation 2004;109:2595-604.

[24] Aronow WS. Antiplatelet therapy in peripheral arterial disease. Curr Drug Targets Cardiovasc Haematol Disord 2004;4:265-7.

[25] Eller P, Schgoer W, Mueller T, et al. Hepatic lipase polymorphism and increased risk of peripheral arterial disease. J Intern Med 2005;258: 344-8.

[26] Rutherford RB, Baker JD, Ernst C, et al. Recommended standards for reports dealing with lower extremity ischemia: revised version. J Vasc Surg 1997;26:517-38.

[27] Mueller T, Marschon R, Dieplinger B, et al. Factor V Leiden, prothrombin G20210A, and methylenetetrahydrofolate reductase C677T mutations are not associated with chronic limb ischemia: the Linz Peripheral Arterial Disease (LIPAD) study. J Vasc Surg 2005;41: 808-15.

[28] Osgood-McWeeney D, Galluzzi JR, Ordovas JM. Allelic discrimination for single nucleotide polymorphisms in the human scavenger receptor class B type 1 gene locus using fluorescent probes. Clin Chem 2000;46:118-9.

[29] Brundert M, Heeren J, Bahar-Bayansar M, et al. Selective uptake of HDL cholesteryl esters and cholesterol efflux from mouse peritoneal macrophages independent of SR-BI. J Lipid Res 2006;47:2408-21.

[30] Duong M, Collins HL, Jin W, et al. Relative contributions of ABCA1 and SR-BI to cholesterol efflux to serum from fibroblasts and macrophages. Arterioscler Thromb Vasc Biol 2006;26:541-7.

[31] Ji Y, Jian B, Wang N, et al. Scavenger receptor BI promotes high density lipoprotein–mediated cellular cholesterol efflux. J Biol Chem 1997;272:20982-5.

[32] Fluiter K, van der Westhuijzen DR, van Berkel TJ. In vivo regulation of scavenger receptor BI and the selective uptake of high density lipoprotein cholesteryl esters in rat liver parenchymal and Kupffer cells. J Biol Chem 1998;273:8434-8.

[33] Landschulz KT, Pathak RK, Rigotti A, et al. Regulation of scavenger receptor, class B, type I, a high density lipoprotein receptor, in liver and steroidogenic tissues of the rat. J Clin Invest 1996;98:984-95.

[34] Hulley S, Grady D, Bush T, et al. Randomized trial of estrogen plus progestin for secondary prevention of coronary heart disease in postmenopausal women. Heart and Estrogen/progestin Replacement Study (HERS) Research Group. JAMA 1998;280:605-13.

[35] Keavney B, McKenzie C, Parish S, et al. Large-scale test of hypothesised associations between the angiotensin-converting-enzyme insertion/deletion polymorphism and myocardial infarction in about 5000 cases and 6000 controls. International Studies of Infarct Survival (ISIS) Collaborators. Lancet 2000;355:434-42.