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Association of *APOA5* and *APOC3* gene polymorphisms with plasma apolipoprotein A5 level in patients with metabolic syndrome

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ABSTRACT

Apolipoprotein A5 gene (*APOA5*) variants are associated with increased plasma triglycerides, a risk factor for the metabolic syndrome (MS), but a correlation with apolipoprotein C3 (*APOC3*) genotypes is controversial. We investigated the correlation of *APOA5* genotypes with plasma apoA5 levels and *APOC3* genotypes in MS patients from a Romanian population. *APOA5* (*−1131T>C*, *c.56C>G*) and *APOC3* (*−482C>T*, *−455T>C*) genotypes and plasma apoA5 concentration were determined in MS patients and healthy subjects. Results showed higher apoA5 levels in plasma and high density lipoproteins (HDL) from MS patients, carriers of the *APOA5 c.56G* allele, as compared to MS carriers of *APOA5 −1131C* allele or the common genotype. ApoA5 levels in plasma and HDL fraction from MS carriers of *−1131C* and *c.56G* alleles correlated positively with plasma triglycerides levels and negatively with HDL-cholesterol in MS carriers of *c.56G* allele. Higher frequencies of *APOC3 −482T* and *−455C* alleles were detected in MS patients compared with healthy subjects. We demonstrated the association of *APOC3 −482T* and *−455C* with *APOA5 −1131C* allele, but not with c.56G allele in MS patients. We propose *APOA5 c.56C>G* as a functional polymorphism, whereas *APOA5 −1131T>C* is not an independent risk factor, being effective only when associated with *APOC3 −482T* or *−455C* alleles.

Introduction

The metabolic syndrome (MS) is characterized by the clustering of several factors: abdominal obesity, insulin resistance and/or glucose intolerance, atherogenic dyslipidemia, increased systolic blood pressure and a pro-inflammatory/pro-thrombotic state [1,2]. The etiology of MS is complex, being determined by the interplay of both genetic and environmental factors.

Apolipoprotein A5 gene (*APOA5*) was identified 30 kb upstream of the well-characterized *APOA1/C3/A4* gene cluster on 11q23 and published data suggest a strong correlation with plasma triglycerides levels [3]. Human *APOA5* gene consists of four exons and encodes a 366-amino acid protein – apolipoprotein A5 (apoA5), which is produced only by the liver and is found associated with high (HDL) and very low (VLDL) density lipoproteins particles [3,4]. The plasma apoA5 concentration in human plasma is low compared to other apolipoproteins, but it remains to be explained why it is sufficient to regulate the turnover of VLDL by stimulating lipoprotein lipase-mediated triglycerides hydrolysis [4,5].

The relevance of apoA5 in the regulation of plasma triglycerides concentration has been deduced from studies of its common genetic variants. Five common *APOA5* haplotypes have been defined by several single nucleotide polymorphisms (SNP) in the *APOA5* gene [6]. Two haplotype-tagged *APOA5* gene variants (*−1131T>C* and *c.56C>G*) have been associated with hypertriglyceridemia and with increased remnant-like particle triglycerides concentrations in many studies [7–9]. The chromosomal cluster that contains the *APOA5* gene has not been clearly associated with the metabolic syndrome. Recently, it was reported that in Caucasians both *−c.56G* [10] and *−1131C* alleles are associated with MS [11,12].

Another important apolipoprotein, the liver and intestine-secreted apolipoprotein C3 (apoC3), plays a key role in controlling the plasma triglycerides levels by a well known mechanism, being present in VLDL and HDL particles [13]. ApoC3 inhibits apolipoprotein E-mediated liver uptake of triglycerides-rich lipoproteins and their remnants, and inhibits hepatic and lipoprotein lipase activity

Abbreviations: *APOA5*, apolipoprotein A5 gene; apoA5, apolipoprotein A5; *APOC3*, apolipoprotein C3 gene; ATP-III, Adult Treatment Panel III; BMI, body mass index; CI, confidence interval; ELISA, enzyme-linked immunosorbent sandwich assay; FPLC, fast-protein liquid chromatography; HDL, high density lipoproteins; LDL, low density lipoproteins; MS, metabolic syndrome; NCEP, National Cholesterol Education Program; OR, odds ratio; RFLP, restriction fragment length polymorphism; SD, standard deviation; SNP, single nucleotide polymorphism; VLDL, very low density lipoproteins

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[14]. Published data reveal that apoC3 gene (*APOC3*) is transcriptionally inhibited by insulin and promoter sequences with high affinity for nuclear transcription factors that mediate this insulin response are highly polymorphic [15,16]. Variants of *APOC3 −455* and *−482* promoter sites have reduced affinity for these transcription factors, being associated with insulin resistance. *APOC3 −455T>C* SNP was associated with increased triglycerides levels and represents an independent risk factor for cardiovascular disease [17,18], while *APOC3 −482C>T* has been associated with dyslipidemia and insulin resistance [19].

Recent studies demonstrate a correlation between polymorphism and specific haplotypes in the *APOA1/C3/A4/A5* gene cluster with increased plasma triglycerides and reduced HDL-cholesterol levels, as well as with diabetic dyslipidemia [20,21]. Talmud and her team showed that the association of *APOA5 −1131T>C* SNP-defined haplotype with the triglycerides levels is not due to individual effects and suggested an association with other mutations that belong to the same *APOA5* haplotype or a strong linkage with a functional polymorphism of the *APOC3* gene (*−482C>T*) [9].

Starting from the previous reported genotype information on the *APOA5 −1131T>C* and *c.56C>G* SNPs [15], in the present study we explored the relation between the *APOA5* gene variants, apoA5 levels and some biochemical parameters in the plasma from patients with MS in a Romanian urban population. Asking whether *APOA5* genotypes effects are correlated with *APOC3* genotypes in MS patients, we investigated the possible association between the *APOA5* and *APOC3* gene variants in the studied subjects.

Materials and methods

Study design and subjects. The study covered 279 subjects, 127 women and 152 men, aged 23–64 years, selected from the population of Bucharest, Romania, and divided into two groups (1) 91 healthy subjects as controls and (2) 188 metabolic syndrome patients recruited from the "N. Paulescu" Institute of Diabetes, Nutrition and Metabolic Diseases, following to the criteria of the National Cholesterol Education Program (NCEP) – Adult Treatment Panel III (ATP-III) [2]. According to the World Health Organization recommendations, the subjects were classified in four categories according to their anthropometric parameters: normal (body mass index, BMI < 25 kg/m^2), overweight (25 < BMI < 30 kg/m^2), obese (30 < BMI < 40 kg/m^2) and very obese (BMI > 40 kg/m^2). From each subject, fasting blood samples were taken on EDTA for biochemical and DNA analysis. None of the subjects underwent lipid lowering therapy (fibrates or statins). The study was carried out according to the principles of the Declaration of Helsinki; all participants gave informed consent; the institutional review board of the Institute of Cellular Biology and Pathology "N. Simionescu" has approved the study.

Plasma biochemical parameters. Plasma was separated by centrifugation and biochemical parameters assayed with commercially available kits (Dialab, Wiener Neudorf, Austria) for total cholesterol, triglycerides, LDL-cholesterol (LDL-C), HDL-cholesterol (HDL-C) and fasting glucose.

ApoA5 protein measurements. An ELISA sandwich assay was used to measure apoA5 levels in human plasma and in the HDL fraction (obtained after precipitation of apoB-containing lipoproteins with phosphotungstic acid), as was previously described [22].

Lipoprotein analysis. Lipoproteins from pooled human plasma were fractionated by fast-protein liquid chromatography (FPLC) (see Appendix 1).

APOA5 and APOC3 gene polymorphisms analysis. Genomic DNA was isolated from peripheral blood leukocytes using a commercially available kit (Wizard Genomic DNA purification kit, Promega Corp., Madison, WI, USA). Two *APOA5* and two *APOC3* gene polymorphisms were assayed by the restriction fragment length polymorphism (RFLP) technique (see Appendix 1).

Statistical analysis. Statistical analysis was performed using SPSS for Windows 15.0 (Chicago, IL, USA) (see Appendix 1).

Results

Anthropometric and biochemical parameters

Table S1 (see Appendix 2) presents the mean biochemical, metabolic and anthropometric parameters for the 279 studied subjects divided in the two groups: 188 MS patients and 91 controls, consisting of healthy subjects. In general, patients presented NCEP-ATP-III-defined risk factors for MS: higher waist circumference, BMI, systolic blood pressure, plasma total cholesterol, triglycerides, LDL-C and fasting glucose levels, while having lower HDL-C concentrations compared to healthy subjects.

Distribution of APOA5 gene polymorphism

The *APOA5 −1131T>C* and *c.56C>G* (*S19W*) genotypes frequencies did not deviate from Hardy–Weinberg equilibrium in the entire population, in controls as well as in MS patients separately. For the *APOA5 −1131TC/CC* genotypes we observed similar frequencies in MS patients (15.7%) as in control subjects (18.3%, χ^2 test $p = 0.760$). The other studied *APOA5* gene variant, *c.56C>G*, presented a higher frequency in MS patients for the *c.56CG/GG* genotype (19.7%) than in healthy subjects (12.7%, χ^2 test $p = 0.014$). An additional observation was that *APOA5 −1131C* and *c.56G* alleles had similar frequencies in MS patients (14.8% and 14.6%, respectively).

Analysis of the *APOA5* genotypes distribution according to the weight status showed that the obese and very obese patients had higher incidence of *APOA5 −1131TC/CC* genotype (81.8%) than the carriers of the common *APOA5* genotype (70.4%, $p = 0.027$). In MS patients, the weight status was not correlated with the presence of the *APOA5 c.56G* allele, as compared with subjects having the common *c.56CC* genotype (71.8% vs. 75.0%, NS). For the controls, no difference was observed in the *APOA5* genotypes distribution according to the weight categories.

APOA5 genotypes association with plasma biochemical parameters

We analyzed the group distribution of *APOA5* genotypes according to the plasma biochemical parameters and we found that the presence of the *−1131C* allele in MS patients was associated with hypertension ($\geqslant$130/85 mmHg, $p = 0.027$), high plasma triglycerides ($\geqslant$1.7 mmol/L, $p = 0.026$) and low HDL-C ($\leqslant$1.15 mmol/L, $p = 0.033$) levels, as compared with the controls. We observed that high plasma triglycerides ($p = 0.021$) and low HDL-C ($p = 0.032$) levels, but not hypertension, were associated with the increased incidence of the *c.56G* allele in MS patients. No correlation between the incidence of high plasma cholesterol ($\geqslant$6.15 mmol/L), LDL-C ($\geqslant$3.84 mmol/L) and fasting glucose ($\geqslant$6.11 mmol/L) and the presence of *APOA5 −1131C* or *c.56G* alleles in MS patients was observed (data not shown).

Plasma apoA5 levels and biochemical parameters

We analyzed the apoA5 concentration in plasma from healthy subjects and MS patients. The measured plasma apoA5 level ranged between 73 and 482 ng/mL, with an overall mean of 223.2 ± 116.2 ng/mL for men (from 73 to 435 ng/mL) and 242.8 ± 99.8 ng/mL for women (from 73 to 482 ng/mL), these mean values being not significantly different. There were few subjects, 6

men and 16 women, out of which 7 were controls and 15 MS patients, with extremely high values of apoA5 plasma concentration (between 584 and 1756 ng/mL) and they were excluded from the statistical analysis. Mean values of apoA5 levels in plasma of MS patients did not differ statistically significant in comparison with those in healthy subjects (see Table S1, Appendix 2). In addition, plasma apoA5 levels in MS patients were positively correlated with plasma triglycerides levels ($r = 0.308$, $p = 0.002$), while in the healthy subjects no correlation could be established (data not shown). FPLC analysis revealed a relocation of apoA5 from the HDL to the VLDL fraction in plasma isolated from both MS men (Fig. 1A) and women (Fig. 1B) (see Fig. 1, Appendix 2).

APOA5 genotypes and plasma apoA5 levels

The presence of *APOA5 −1131C* allele in MS patients was associated with lower apoA5 concentrations in plasma and HDL fraction, as compared with those measured in MS patients having the common genotype (Table 1). In contrast, MS carriers of the *c.56G* allele had higher apoA5 levels measured in plasma and HDL fraction, as compared with the levels determined in MS patients with the common genotype (Table 1). Moreover, higher plasma and HDL-associated apoA5 levels were measured in the plasma of MS carriers of *APOA5 c.56G* allele, as compared with MS patients having the *−1131C* allele.

Correlation of plasma apoA5 levels with APOA5 genotypes and biochemical parameters

A positive correlation between plasma and HDL-associated apoA5 levels with triglycerides levels was obtained for MS patients, but not for healthy subjects (Table 2).

We further analyzed the association of apoA5 levels with the plasma biochemical parameters in MS patients and healthy subjects carriers of *APOA5 −1131T>C* and *c.56C>G* genotypes. In MS patients, a positive correlation between the plasma level of apoA5 and triglycerides was observed in carriers of *−1131C* allele (see Table S2 from Appendix 2). In addition, apoA5 levels in the HDL fraction were positively correlated with plasma triglycerides concentrations in MS carriers of *APOA5 −1131TC + CC* genotypes. No association of the apoA5 concentrations (plasma and HDL fraction) with BMI, total cholesterol, HDL-C, LDL-C and fasting glucose levels was established in MS carriers of *APOA5 −1131C* allele (Table S2 in Appendix 2).

We observed that plasma triglycerides levels were positively correlated with total and HDL-associated apoA5 levels in plasma from MS patients that are carriers of *APOA5 c.56G* allele (Table S3, Appendix 2). In addition, we demonstrated positive correlation between fasting glucose levels with total apoA5 and HDL-apoA5 levels in MS carriers of *APOA5 c56G* allele. In contrast, HDL-C levels showed negative correlation with total and HDL-associated apoA5 concentrations in plasma of MS carriers of *APOA5 c.56G* allele. No correlation between apoA5 levels (total and HDL-associated fraction) with BMI, total cholesterol and LDL-C levels was shown in MS patients with *APOA5 c.56G* allele (Table S3, Appendix 2).

Table 2
Correlation coefficients (r, and associated p-values) between the total and HDL-associated apoA5 plasma levels with the plasma biochemical parameters of control subjects and MS patients.

	Control		MS	
	r	p Value	r	p Value
Total apoA5				
BMI	0.168	NS	0.175	NS
Total cholesterol	0.143	NS	0.008	NS
Triglycerides	0.237	NS	0.512	<0.001
LDL-C	0.159	NS	−0.130	NS
HDL-C	0.156	NS	−0.014	NS
Fasting glucose	0.105	NS	0.144	NS
HDL fraction apoA5				
BMI	0.116	NS	0.192	NS
Total cholesterol	0.197	NS	0.008	NS
Triglycerides	0.116	NS	0.516	<0.001
LDL-C	0.172	NS	−0.138	NS
HDL-C	0.155	NS	0.024	NS
Fasting glucose	0.062	NS	0.154	NS

Abbreviations: BMI, body mass index; LDL-C, LDL-cholesterol; HDL-C, HDL-cholesterol.

Association of APOA5 and APOC3 gene polymorphisms

In order to investigate the role of the studied *APOA5* minor alleles in the prediction of the metabolic syndrome complications, we assessed the correlation between these SNPs and the two *APOC3* gene polymorphisms that occur in the insulin responsive element of the *APOC3* gene promoter [19]. To this purpose, we analyzed the distribution of *APOC3 −482C>T* and *−455T>C* genotypes in the studied subjects and their correlation with the occurrence of MS. The two studied *APOC3* genotypes did not deviate from Hardy–Weinberg equilibrium in the entire population, in healthy subjects, as well as in MS patients.

APOC3 −482C>T gene polymorphism

The overall frequencies obtained for *APOC3 −482C>T* genotype in the studied subjects are presented in Table 3. The allele frequencies were 70.1% for the *APOC3* common allele *−482C* and 29.9% for the minor allele *−482T*.

The analysis of the genotype frequency for *APOC3 −482C>T* in all the studied subjects revealed a statistically significant different

Table 1
Mean apoA5 levels in plasma and HDL fraction of control subjects and MS patients; correlation with *APOA5 −1131T>C* and *c.56C>G* genotypes. p-Values were obtained after comparison with *APOA5* common allele homozygote in each group.

Plasma apoA5 level	*APOA5* genotype	Control		MS	
		Mean	p Value	Mean	p Value
	−1131T>C				
Total (ng/mL)	TT	253.7 ± 99.8	NS	236.4 ± 110.0	0.033
	TC + CC	254.6 ± 123.0		168.8 ± 62.4	
HDL fraction (ng/mL)	TT	150.4 ± 77.3	NS	138.0 ± 84.1	0.041
	TC + CC	145.5 ± 93.7		89.1 ± 34.7	
	c.56C>G				
Total (ng/mL)	CC	249.6 ± 98.5	NS	220.3 ± 103.3	0.028
	CG + GG	276.7 ± 131.0		340.4 ± 111.1	
HDL fraction (ng/mL)	CC	144.9 ± 74.5	NS	123.9 ± 72.8	0.025
	CG + GG	174.4 ± 105.6		248.3 ± 110.9	

Abbreviations: Control, healthy subjects; MS, metabolic syndrome patients.

Table 3
Distribution of *APOC3* genotypes defined by the *−482C>T* and *−455T>C* SNP for control subjects and MS patients.

Group	*APOC3* genotype (%)			*p* Value
	−482 CC	*−482 CT*	*−482 TT*	
Overall	52.2	35.7	12.1	0.013
Control	61.7	34.6	3.7	
MS	45.3	38.9	15.8	
	−455 TT	*−455 TC*	*−455 CC*	
Overall	44.7	38.2	17.1	0.004
Control	62.0	24.6	13.4	
MS	39.2	39.7	21.1	

distribution between healthy subjects and MS patients (Table 3). The frequency of the minor allele *APOC3 −482T* was significantly increased in MS patients (about 40%) compared to control subjects (19.5%, χ^2 test: $p < 0.001$).

In analyzing the biochemical and anthropometric parameters for the subjects genotyped with the *APOC3 −482C>T*, we observed that the *−482T* carriers were having the characteristic features of the metabolic syndrome, namely high triglycerides and fasting glucose plasma levels, low HDL-C levels and obesity diagnosis (Table S4, Appendix 2).

We demonstrated a significant association between the frequency of *APOC3 −482T* and *APOA5 −1131C* alleles in MS patients (χ^2 test: $p = 0.005$; OR = 2.14, 95% CI: 1.068–4.288, $p = 0.032$), but not in controls (Table 4). No association was found between the presence of *APOC3 −482T* and *APOA5 c.56G* alleles, both for MS patients (χ^2 test: p = NS; OR = 0.505, 95% CI: 0.239–1.067, p = NS) and healthy subjects.

APOC3 −455C>T gene polymorphism

The overall and group-associated frequencies for *APOC3 −482T*>C genotypes in the studied subjects are presented in Table 3. The allele frequencies were 63.8% for the *APOC3* common allele *−455T* and 36.2% for the minor allele *−455C*.

We analyzed the distribution of *APOC3 −455T>C* genotypes for the two groups of subjects and we demonstrated a higher frequency of the *−455CC* genotype in the MS group as compared to healthy subjects (Table 3). In addition, the frequency of the minor allele *APOC3 −482T* was higher in MS patients (41%), as compared to healthy subjects (26%) (Table 3). However, no association was found between the presence of *APOC3* minor allele *−455C* and the main biochemical and anthropometric features of MS patients (Table S4, Appendix 2).

We analyzed the distribution of the minor alleles corresponding to *APOC3 −455T>C* and *APOA5 −1131T>C* or *c.56C>G* genotypes for the MS patients and controls (Table 4). Therefore, we demonstrated a significant association between the incidence of *APOC3 −455C* and *APOA5 −1131C* alleles in MS patients (χ^2 test: $p = 0.002$; OR = 3.032, 95% CI: 1.394–6.597, $p = 0.005$), but not in healthy subjects. No significant association between the incidence of *APOC3 −455C* and *APOA5 c.56G* alleles was demonstrated, both in MS patients (χ^2 test: $p = 0.662$; OR = 0.707, 95% CI: 0.350–1.426, p = NS) and in healthy subjects.

Table 4
Distribution of *APOC3 −482C>T* and *−455T>C* genotypes, respectively, for the carriers of *APOA5 −1131T>C* or *c.56C>G* genotypes in control subjects and MS patients.

APOA5 genotype	Group	*APOC3* genotype (%)		*p* Value
		−482 CC	*−482 CT + TT*	
−1131TT	Control	56.4	43.6	NS
	MS	52.6	47.4	
−1131TC + CC	Control	61.5	38.5	0.005
	MS	24.1	75.9	
c.56CC	Control	56.4	43.6	NS
	MS	45.3	54.7	
c.56CG + GG	Control	61.5	38.5	NS
	MS	68.2	31.8	
		−455 TT	*−455 TC + CC*	
−1131TT	Control	62.1	37.9	NS
	MS	43.9	56.1	
−1131TC + CC	Control	53.8	46.2	0.002
	MS	13.8	86.2	
c.56CC	Control	57.9	42.1	NS
	MS	38.5	61.5	
c.56CG + GG	Control	71.4	28.6	NS
	MS	44.0	56.0	

Discussion

In the present study we investigated the association of the frequencies distribution for *APOA5* and *APOC3* genotypes in a Romanian population with/without metabolic syndrome, and their correlation with plasma apoA5 levels. In a previous paper, we demonstrated the association of *APOA5 −1131C* allele frequency with some of the risk factors for the metabolic syndrome [12]. We report an average apoA5 plasma level of 236.5 ± 108.3 ng/mL, with no statistically significant difference between the mean values for healthy subjects and MS patients. The measured concentrations of plasma apoA5 protein were similar to those reported for other Caucasian populations [4,23–25].

Our data show a positive correlation between plasma apoA5 concentrations and plasma triglycerides levels in MS patients carriers of the *APOA5 −1131C* allele, in good agreement with recent data [26], but in contrast with other reports [4,5]. Interestingly, we observed significantly lower apoA5 levels in MS patients carriers of *APOA5 −1131C* allele compared with MS carriers of common *APOA5* genotype, but not in controls. A recent study shows that the low concentrations of apoA5 in plasma might be due in part to the *APOC3*-enhancer, which upregulates transcription of *APOA1/APOC3/APOA4* gene cluster and does not increase the expression of the *APOA5* gene, because some of distal *Alu* elements would block its action [27].

In the present study we demonstrate higher plasma apoA5 levels in MS patients carriers of *APOA5 c.56G* allele, as compared with those having *−1131C* allele or the common *APOA5* genotype, a positive correlation between plasma apoA5, triglycerides and fasting glucose levels, and a negative correlation with plasma HDL-C levels. These data are in good agreement with previous reports in Caucasians with hypertriglyceridemia [28]. However, Talmud et al. [29] demonstrate that *APOA5 c.56C>G* that occurs in the signal peptide for co-translational transport into the endoplasmic reticulum was predicted to result in a reduced secretion of apoA5 protein with Trp-19. As a consequence, the *c.56G* allele might result in lower plasma apoA5 levels and it remains to explore why the opposite effect was observed in hypertriglyceridemic subjects.

Recent studies demonstrate a correlation between the polymorphism and specific haplotypes from the *APOA1/C3/A4/A5* gene cluster, the increased plasma triglycerides and reduced HDL-C levels, as in diabetic dyslipidemia [20,21]. Our results demonstrate that the *APOC3 −482C>T* SNP in the promoter region is associated with MS, in agreement with Miller et al. [18]. Moreover, we provide evidence that the frequency of the *−482T* allele is associated with that of the *APOA5 −1131C* allele in MS patients, but not in the control group. We observed the same effect for *APOC3 −455C* allele, having even a higher frequency in MS patients.

Taken together, the previous demonstrated association between the *APOA5 −1131C* allele and the MS [12], along with the here

demonstrated correlation between the plasma apoA5 and triglycerides levels, suggest that this allele might not be an independent risk factor for MS, unless associated with the presence of at least two other minor alleles of *APOC3*, *−482T* and *−455C*. Similar results by Talmud et al. show that the association of *APOA5 −1131T>C* SNP-defined haplotype with the triglycerides level is not due to individual effects, and they suggested a connection with other mutations in this haplotype (*−3A>G*, *751G>T*), or a strong linkage with a functional polymorphism of *APOC3* gene (*−482C>T*) [9]. Following the association of the *APOA5 c.56G* allele with the *APOC3* gene variants, we demonstrate that the increase in plasma triglycerides is independent of the effect induced by the *APOC3* gene variants, similar to the results of Talmud et al. [9].

We observed a redistribution of the apoA5 from HDL to VLDL particles in the plasma from obese patients. In a previous paper, we reported the same tendency in plasma isolated from the double-transgenic *APOA5xAPOC3* mice during the postprandial state [22]. Pruneta-Deloche et al. reported the same effect, both in control subjects and type 2 diabetic patients, suggesting that this effect could be a result of the increased binding capacity of non-HDL lipoproteins, given their increase in the postprandial hypertriglyceridemia [25]. We propose that the increased plasma concentration of VLDL particles in MS patients, even in the fasting state, could induce the shift of apoA5 from the HDL reservoir to the triglyceride-rich particles, facilitating their LPL-mediated hydrolysis.

In conclusion, our data demonstrate (i) higher apoA5 levels in plasma and HDL from MS patients carriers of *APOA5 c.56G* allele as compared with those having the *APOA5 −1131C* allele or the common genotype; (ii) a positive correlation of plasma apoA5 levels with triglycerides and glucose levels, and a negative correlation with HDL-C in MS patients carriers of *APOA5 c56G* allele; (iii) a positive correlation between plasma apoA5 levels and triglycerides concentrations in MS patients carriers of *APOA5 −1131C* allele; (iv) a redistribution of the apoA5 protein from HDL to VLDL particles in plasma from MS patients and (v) an association between the presence of *APOA5 −1131C* with either *APOC3 −455C* or *−482T* alleles in MS patients. Thus, we propose that *APOA5 c.56C>G* is a functional polymorphism, while *APOA5 −1131T>C* function is still under debate.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bbrc.2009.11.103.

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