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Original article

Implication of the rs670 variant of *APOA1* gene with lipid profile, serum adipokine levels and components of metabolic syndrome in adult obese subjects

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SUMMARY

Background & aims: A G-to-A transition located 75 base pairs upstream (rs670) from transcription start site of the *APOA1* gene is related with metabolic parameters. The aim of the present investigation was to describe the association of rs670 with metabolic syndrome and metabolic parameters.

Methods: The study involved a population of 1000 obese subjects. Measurements of anthropometric parameters, arterial blood pressure, fasting blood glucose, C-reactive protein (CRP), insulin concentration, insulin resistance (HOMA-IR), lipid profile, adipokines levels and prevalence of MetS was recorded. Genotype of *ApoA1* gene polymorphism (rs670) was evaluated.

Results: A sample of 1000 obese subjects with a mean BMI of 36.5 ± 5.0 kg/m^2 was enrolled. In males, weight (delta: 3.3 ± 1.2 kg; p = 0.01), fat mass (delta: 2.7 ± 1.1 kg; p = 0.01), waist circumference (delta: 2.8 ± 1.1 cm; p = 0.02), fasting glucose (delta: 8.9 ± 2.2 mg/dl; p = 0.01), insulin levels (delta: 3.7 ± 1.2 UI/L; p = 0.04) and HOMA-IR (delta: 1.2 ± 1.1 units; p = 0.02) were higher in non-A allele carriers than A allele carriers. In males without A allele, an increased risk of hyperglycemia (OR = 1.40, 95% CI = 1.09−2.09, p = 0.04), percentage of central obesity (OR = 4.55, 95% CI = 1.36−15.39, p = 0.01), percentage of low HDL-C (OR = 2.02, 95% CI = 1.02−4.03, p = 0.03) and prevalence of diabetes mellitus (OR = 2.14, 95% CI = 1.03−5.04, p = 0.03) were reported.

Conclusions: rs670 of *APOA1* gene has a gender specific influence on serum glucose, insulin, HOMA-IR, fat mass, weight and waist circumference. Males without A allele showed high rates of central obesity, low levels of HDL, hyperglycemia and diabetes mellitus.

1. Introduction

Metabolic syndrome (MetS) is defined by the clustering of several factors; insulin resistance and/or glucose intolerance, abdominal obesity, dyslipemia, increased blood pressure and pro-inflammatory state. Dyslipemia related to MetS consisted of two alterations, one is the elevation of plasma triglyceride and the other the decrease of high-density lipoprotein cholesterol concentration (HDL-C), both changes are associated with increased risk of coronary heart event [1].

Apolipoprotein ApoA1 is the most important protein constituent of HDL-C and plays a role in lipid metabolism and transport. Genetics studies have shown that gene encoding ApoA1 is highly polymorphic and common single nucleotide polymorphisms (SNPs) of this gene, that maps to the long arm of chromosome 11, have been described in relation to plasma lipoproteins [2]. In vitro studies have shown that overexpression of the human *APOA1* gene in mice increased HDL-C levels and protect mice from the atherosclerosis secondary to a high fat diet [3].

The *APOA1* gene polymorphisms have been found to be strongly associated with variation in MetS components such as HDL-C and glucose levels [4,5]. A G-to-A transition located 75 base pairs upstream (rs670) from transcription start site of the *APOA1* gene has been evaluated in an important way [6]. In one study [7] a high transcription rate was observed in −75A allele carriers in comparison with non A allele carriers. However in the literature there

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are conflicting data, with a few studies reporting either negative association [8] or lack of association [9] between rs670 polymorphism and lipid levels. Finally, other studies reported that A allele carriers have been shown to have elevated concentrations of ApoA1 and HDL-C [10–12]. Also, G allele has been reported to be related with hypertension [13], metabolic syndrome components [14]. In other designs, A allele carriers have shown an increased risk of type 2 diabetes mellitus [15].

Finally, adipose tissue is expressed as an endocrine organ. Many proteins, produced by adipose tissue, named adipokines, have been described (leptin, adiponectin and resistin). Besides that, adipose tissue plays a major role in the presence of metabolic syndrome (MetS) [16]. In the literature there is no studies evaluating the relationship of this SNP of *APOA1* gene with serum adipokine levels.

The aim of the present investigation was to describe the association of rs670 with metabolic syndrome, adipokines and metabolic parameters.

2. Materials and methods

2.1. Subjects and clinical investigation

For the study one thousand unrelated Caucasian obese subjects (body mass index $\geq$30 kg/m^2) were consecutively recruited from Primary Care Physicians of an urban area of Castilla y Leon (Norwest of Spain). All participants underwent a medical evaluation including physical examination and complete medical history. Data on blood pressure, anthropometric parameters (weight, height, body mass index (BMI), fat mass by impedance and central obesity by waist circumference) were collected. Venous blood samples were collected in EDTA-treated and plain tubes after a 10 h overnight fast for analysis of serum adipokine levels (leptin, total adiponectin and resistin), insulin, total cholesterol, LDL-cholesterol, HDL-cholesterol and triglycerides. To estimate the prevalence of diabetes mellitus was considered American Diabetes Association criteria [17] and the prevalence of Metabolic Syndrome, the definitions of the ATPIII was considered [1]. Subjects need to fulfill at least 3 of the following 5 criteria: elevated fasting glucose or treatment for diabetes, elevated triglycerides (>150 mg/dl) or treatment for dyslipemia, low HDL cholesterol <40 mg/dl (males) or <50 mg/dl (females), elevated systolic or diastolic blood pressure (>130/85 mmHg or antihypertensive treatment) and increased waist circumference (>94 cm (males) or >80 cm (females)).

The recruited subjects fulfilled the following criteria; body mass index $\geq$30 kg/m^2, had no a history of coronary events, thyroid disease, renal or hepatic disorders, had no history of alcoholism, malignant tumor, and within the 6 months before the study were not receiving medications known to influence lipid levels (hormonal therapy, glucocorticoids and anti-inflammatory drugs). The Ethics Committee (HCUVA Committee) approved the study and was in accordance with the guidelines laid down in the Declaration of Helsinki. An ethical approval for genetic research was obtained, too. All participants provided written informed consent.

2.2. Blood pressure and anthropometric parameters

Mean systolic and diastolic blood pressures were calculated by averaging three measurements (Omrom, LA, CA), after the subjects sat for 10 min. Body weight were determined in the morning while the subjects were minimally unclothed and not wearing shoes. They were measured using digital scales (Omrom, LA, CA) and recorded to the nearest 50 g. Height was measured with a tape measure (Omrom, LA, CA) while patients were standing with shoulders in normal alignment and no wearing shoes. Body mass index (BMI) was calculated as body weight (in kg) divided by height (in m^2). Waist circumferences (WC) was measured at the umbilical level with the use of an upstretched tape measure. Bio impedance was used to determine body composition with an accuracy of 5 g [18] (EFG BIA 101 Anniversary, Akern, It). This equation was used (0.756Height^2/Resistance) + (0.110Body mass) + (0.107Reactance) − 5.463.

2.3. Biochemical procedures

Fasting venous blood samples were taken for measurements of fasting glucose, total cholesterol, triglyceride, low density lipoprotein cholesterol (LDL) and high density lipoprotein cholesterol (HDL) using a biochemical auto-analyzer (Hitachi 7060, Tokyo, Japan). LDL cholesterol was determined using Friedewald formula [19]. Fasting glucose was determined by the enzymatic colorimetric method using glucose oxidase. Fasting insulin was measured by radioimmunoassay method (RIA) (RIA Diagnostic Corporation, Los Angeles, CA) with a sensitivity of 0.5 mUI/L (normal range 0.5–30 mUI/L) [20]. We calculate the homeostasis model assessment of insulin resistance (HOMA-IR) (HOMAIR = (insulin × glucose)/22.5) [21]. C-reactive protein (CRP) was determined by immunoturbimetry (Roche Diagnostics GmbH, Mannheim, Germany), with a normal range of (0–7 mg/dl) and analytical sensitivity 0.5 mg/dl.

Leptin was determined by ELISA (Diagnostic Systems Laboratories, Inc., Texas, USA) (DSL1023100); sensitivity of 0.05 ng/ml, a normal range of 10–100 ng/ml and a CV% 3.5% [22]. Adiponectin was determined by ELISA (R&D systems, Inc., Minneapolis, USA) (DRP300); sensitivity of 0.246 ng/ml, a normal range of 8.65–21.43 ng/ml and a CV% 3.8% [23]. Resistin was determined by ELISA (Biovendor Laboratory, Inc., Brno, Czech Republic) (RD191016100); sensitivity of 0.2 ng/ml, a normal range of 4–12 ng/ml [24] and a CV% 3.2%.

2.4. Genotyping of APOA1 gene polymorphism

For genotyping the AOA1 polymorphism, buffy coats were separated from non-coagulated whole blood samples and stores at −70 °C until processing. Genomic DNA was extracted by using commercial kit extraction (Biorad, LA, CA) according to the manufacturer's protocol. Primers were realized with the Sequenom Assay Design v4 (SEQUENOM, Inc. San Diego, California CA). Genotyping for the rs670 polymorphism was performed by polymerase chain reaction real time analysis. This polymerase chain reaction (PCR) was carried out with 30 ng of genomic DNA, 0.1–0.15 μl each of oligonucleotide primer for rs670 (primer forward: 5′- ACGTTGGATGAAGTTCCACATTGCCAGGAC -3′ and reverse 5′- ACGTTGGATGCAGGGCCTATTTATGTCTGC -3′ in a 2.5-μl final volume (Termociclador Lifetechnologies, LA, CA)). DNA was denatured at 85 °C for 5 min; this was followed by 45 cycles at 65 °C for 15 s, and annealing at 58.1 °C for 45 s. The PCRs were run in a 2-μl final volume containing 0.1 μl of iPLEx Termination mix (Bio-Rad®, San Diego, CA) with hot start Taq DNA polymerase. Hardy Weinberg equilibrium was calculated with a statistical test (Chi-square). The variant of *ApoA1* gene was in Hardy Weinberg equilibrium ($p = 0.36$).

2.5. Statistical analysis

All the data were analyzed using SPSS for Windows, version 19.0 software package (SPSS Inc. Chicago, IL). Sample size was calculated to detect differences over 3 mg/dl of HDL-C levels with 90% power and 5% significance. All analysis were performed under a dominant genetic model with rs670 A-allele as the risk allele (AA + AG vs. GG). The results were expressed as average ± standard deviation. Variables were analyzed with ANOVA test (for normally-distributed variable) or Kruskal–Wallis test (for non-normally-distributed variable). Logistic regression analyses adjusted by age, gender and BMI were used to calculated odds ratio (OR) and 95% confidence

Table 1
Anthropometric variables and blood pressure.

Parameters	Total group (n = 1000)	
	Male (n = 267)	Female (n = 733)
BMI	36.3 ± 5.1	36.1 ± 5.0
Weight (kg)	107.4 ± 10.1	90.1 ± 10.2*
Height (m)	1.72 ± 0.3	1.57 ± 0.2*
Fat mass (kg)	34.8 ± 5.0	41.6 ± 4.1*
WC (cm)	119.4 ± 7.1	109.1 ± 7.1*
SBP (mmHg)	132.8 ± 9.1	126.1 ± 8.0
DBP (mmHg)	83.5 ± 5.1	81.8 ± 4.2
Fasting glucose (mg/dl)	109.4 ± 10.1	100.9 ± 6.1
Total cholesterol (mg/dl)	205.5 ± 21.8	196.9 ± 12.1
LDL-cholesterol (mg/dl)	132.4 ± 10.9	125.2 ± 21.1
HDL-cholesterol (mg/dl)	49.2 ± 5.5	52.1 ± 8.1
Triglycerides (mg/dl)	147.1 ± 31.1	119.0 ± 32.1*
Insulin (mUI/l)	18.8 ± 7.9	12.7 ± 8.2*
HOMA-IR	5.3 ± 1.1	3.1 ± 0.9*

BMI: body mass index DBP, diastolic blood pressure; SBP, systolic blood pressure; WC, waist circumference; HOMA-IR (homeostasis model assessment of insulin resistance). *$p < 0.05$ between genders.

interval (CI) to estimate the association of the rs1670 SNP with the risk of Metabolic syndrome, components of MetS and diabetes mellitus. A p-value under 0.05 was considered statistically significant.

3. Results

The sample comprised of 1000 obese subjects. The mean age was 46.3 ± 9.1 years (range: 25–65) and the mean body mass index (BMI) 36.3 ± 5.0 kg/m^2 (range: 31.3–41.4). Gender distribution was 733 women (73.3%) and 267 men (26.7%). Table 1 shows the clinical and biochemical characteristic of the sample by gender. The distribution of the rs670 polymorphism in this adult population was 67.1% (n = 671) (GG), 29.9% (n = 299) (GA) and 3% (n = 30) (AA). The allele frequency was G (0.82) and A (0.18). The subjects were grouped into two groups (GG vs GA + AA). Age was similar in both genotype groups (GG; 46.3 ± 9.1 years vs GA + AA; 46.2 ± 10.1 years: ns). Gender distribution was similar in both genotype groups (GG 25.9% males vs 74.1% females vs GA + AA; 28.8% males vs 71.2% females).

Applying a dominant genetic model, we did not found a significant association between rs670 A-allele and fat mass, weight, waist circumference, blood pressure and BMI in the total and female groups. In males, weight (delta: 3.3 ± 1.2 kg; p = 0.01), fat mass (delta: 2.7 ± 1.1 kg; p = 0.01) and waist circumference (delta: 2.8 ± 1.1 cm; p = 0.02) were higher in non-A allele carriers than A allele carriers (Table 2). BMI, weight, and waist circumferences were higher in males than females in both genotypes. Fat mass was higher in females than males.

Biochemical characteristics according to genotype are shown in Table 3. HDL-c levels was higher in A allele carriers (delta total group: 5.4 ± 1.1 mg/dl: p = 0.02), (delta male group: 7.2 ± 1.2 mg/dl: p = 0.01) and (delta female group: 6.1 ± 1.3 mg/dl: p = 0.02) than non A allele carriers. In males, fasting glucose (delta: 8.9 ± 2.2 mg/dl; p = 0.01), insulin levels (delta: 3.7 ± 1.2 UI/L; p = 0.04) and HOMA-IR (delta: 1.2 ± 1.1 units; p = 0.02) were higher in non-A allele carriers than A allele carriers (Table 2). These differences were not detected in female subjects. In both genotypes, HOMA-IR and insulin levels were higher in males than females. All previous statistical analysis were realized after exclusion of patients using lipid lowering drugs (n = 31), antidiabetic drugs (n = 37) and antihypertensive drugs (n = 98).

Serum adipokine levels and C reactive protein are shown in Table 4. No differences were detected between genotype groups. In both genotypes, leptin levels were higher in females than males.

According to the results of demographic and metabolic characteristics, the percentage of individuals who had metabolic syndrome (MetS) was 53.8% (n = 540) and 46.2% patients without MetS (n = 462). The percentage of subjects with metabolic syndrome, some components of MetS and diabetes mellitus (central obesity, hypertriglyceridemia, hypertension or hyperglycemia) are shown in Table 5. In both genotypes, metabolic syndrome rate,

Table 2
Anthropometric variables and blood pressure.

Parameters	Total group (n = 1000)		Male (n = 267)		Female (n = 733)	
	GA + AA n = 329	GG n = 671	GA + AA n = 93	GA + AA n = 329	GA + AA n = 236	GG n = 497
BMI	36.1 ± 5.8	36.5 ± 5.3	36.2 ± 5.1	36.6 ± 5.0*	35.6 ± 5.8$	36.1 ± 5.1$
Weight (kg)	94.3 ± 11.1	95.1 ± 10.7	105.3 ± 13.1	108.8 ± 12.7*	90.6 ± 9.1$	89.9 ± 10.7$
Height (m)	1.61 ± 0.3	1.62 ± 0.2	1.72 ± 0.2	1.73 ± 0.3	1.58 ± 0.1$	1.57 ± 0.2$
Fat mass (kg)	39.3 ± 5.1	39.4 ± 4.0	32.6 ± 8.0	35.3 ± 7.0*	41.7 ± 8.1$	41.1 ± 9.0$
WC (cm)	111.4 ± 8.1	111.1 ± 9.0	117.6 ± 8.1	120.4 ± 9.1*	109.0 ± 8.0$	108.9 ± 9.1$
SBP (mmHg)	127.8 ± 9.8	129.7 ± 8.1	131.2 ± 7.8	133.3 ± 8.1	126.7 ± 9.8	125.8 ± 8.1
DBP (mmHg)	82.1 ± 5.0	82.4 ± 4.8	83.7 ± 5.0	82.9 ± 4.7	81.4 ± 5.0	82.3 ± 4.2

BMI: body mass index DBP, diastolic blood pressure; SBP, systolic blood pressure; WC, waist circumference; *$p < 0.05$, in GG vs GA + AA genotypes. $$p < 0.05$ between genders.

Table 3
Biochemical parameters (mean ± SD).

Parameters	Total group (n = 902)		Male (n = 239)		Female (n = 663)	
	GA + AA n = 297	GG n = 605	GA + AA n = 84	GG n = 155	GA + AA n = 213	GG n = 450
Fasting glucose (mg/dl)	101.4 ± 12.1	103.9 ± 9.1	103.5 ± 11.1	112.4 ± 10.1*	99.8 ± 9.1	101.0 ± 10.1
Total cholesterol (mg/dl)	199.6 ± 22.8	196.8 ± 19.1	208.8 ± 30.6	204.8 ± 28.1	197.8 ± 30.8	192.6 ± 28.1
LDL-cholesterol (mg/dl)	126.4 ± 11.9	126.1 ± 22.1	134.4 ± 16.1	129.4 ± 20.1	124.4 ± 20.1	125.4 ± 22.1
HDL-cholesterol (mg/dl)	52.3 ± 7.5	46.9 ± 8.0*	50.6 ± 8.1	43.4 ± 8.1*	54.1 ± 8.1	48.1 ± 7.1*
Triglycerides (mg/dl)	125.1 ± 60.1	125.5 ± 39.1	143.1 ± 50.1	149.9 ± 37.1	119.1 ± 40.1$	118.4 ± 36.1$
Insulin (mUI/l)	13.8 ± 7.0	14.4 ± 9.0	17.0 ± 7.1	20.7 ± 9.2*	12.7 ± 7.0$	12.7 ± 8.0$
HOMA-IR	3.4 ± 1.0	3.5 ± 1.2	4.5 ± 1.1	5.8 ± 1.1*	3.1 ± 0.2$	3.2 ± 1.1$

HOMA-IR (homeostasis model assessment of insulin resistance). *$p < 0.05$, in GG vs GA + AA genotypes. $$p < 0.05$ between genders.

Table 4
Serum adipokine levels and C reactive protein (mean ± SD).

Parameters	Total group (n = 902)		Male (n = 239)		Female (n = 663)	
	GA + AA n = 297	GG n = 605	GA + AA n = 84	GG n = 155	GA + AA n = 213	GG n = 450
Resistin (ng/dl)	5.2 ± 1.1	5.3 ± 1.0	4.7 ± 1.1	4.8 ± 1.4	5.3 ± 1.1	5.6 ± 1.2
Adiponectin (ng/dl)	20.7 ± 8.0	22.5 ± 5.0	10.8 ± 8.9	13.8 ± 5.5	23.6 ± 1.9	26.4 ± 5.0
Leptin (ng/dl)	63.1 ± 11.4	62.8 ± 12.2	28.1 ± 10.4	32.8 ± 9.4	74.1 ± 8.2$	75.8 ± 12.4$
CRP (ng/dl)	5.4 ± 1.4	5.6 ± 1.3	4.9 ± 1.8	5.0 ± 1.9	5.6 ± 1.4	5.8 ± 1.2

CRP: C reactive protein. No statistical differences between genotypes. $\$p < 0.05$ between genders.

Table 5
Metabolic syndrome, components of MetS and diabetes mellitus.

Parameters	Total group (n = 1000)		Male (n = 267)		Female (n = 733)	
	GA + AA n = 329	GG n = 671	GA + AA n = 93	GG n = 174	GA + AA n = 236	GG n = 497
Percentage of MetS	53.6%	53.9%	57.1%	59.2%	46.6%$	46.5%$
Percentage of central obesity	80.9%	80.3%	90.0%	97.7%*	73.6%$	75.2%$
Percentage of hypertriglyceridemia	10.6%	12.5%	12.0%	14.0%	10.1%	12.0%
Low HDL cholesterol	10.5%	12.1%	9.9%	19.3%*	11.6%	11.9%
Percentage of hypertension	43.2%	45.8%	40.1%	52.1%*	46.1%	45.5%
Percentage of hyperglycemia	24.2%	24.1%	24.9%	36.3%*	19.5%$	21.1%$
Diabetes mellitus	6.2%	7.0%	6.9%	13.8%*	6.0%	6.6%

The cutoff points for the criteria of; central obesity (waist circumference >88 cm in female and >102 in male), hypertension (systolic BP >130 mmHg or diastolic BP >85 mmHg or specific treatment), hypertriglyceridemia (triglycerides >150 mg/dl or specific treatment) or hyperglycemia (fasting plasma glucose >110 mg/dl or drug treatment for elevated blood glucose). Diabetes mellitus by American diabetes Association (Ref. [10])*$p < 0.05$, in GG vs GA + AA genotypes. $\$p < 0.05$ between genders.

percentage of central obesity, percentage of hyperglycemia and diabetes mellitus prevalence were higher in males than females.

In males non carriers of A allele, logistic regression analysis showed an increased risk of hyperglycemia (OR = 1.40, 95% CI = 1.09–2.09, p = 0.04), percentage of central obesity (OR = 4.55, 95% CI = 1.36–15.39, p = 0.01), percentage of low HDL-C (OR = 2.02, 95% CI = 1.02–4.03, p = 0.03) and prevalence of diabetes mellitus (OR = 2.14, 95% CI = 1.03–5.04, p = 0.03), after adjusting by BMI and age.

4. Discussion

The main finding of this cross sectional study was the fact that males with the GG genotype of SNP (rs670) of the *APOA1* gene showed high values of fasting glucose levels, insulin, HOMA-IR, waist circumference, weight, fat mass, and low values of HDL-C than A allele carriers. Obese males without A allele showed a high risk of diabetes mellitus, hyperglycemia and central obesity.

The relationship of A allele with HDL-C concentrations, confirmed in most of studies with different populations [10–12,25,26], was also reported in our sample of Caucasian. However, some studies [8] have reported an inverse association of this allele and HDL-C levels. A possible explanation for these contradictory results in the literature is due to the influence of other genes (ApoA4, ApoA5, ApoC3), which could be in linkage disequilibrium [26]. Other potential hypothesis to explain this fact may be secondary to differences in study design, sample size, genetic background, and level of BMI, gender or dietary environment.

A allele frequency of the ApoA1 polymorphism in our sample (0.18) was similar to that previously showed in Caucasian population (0.11–0.15) [10–12] and Asiatic population (0.13–0.15) [27,28]. Moreover, the frequency of A allele has been higher in some populations such as Taiwanese (0.32) [29]. These differences in allele's frequencies have been recognized in different populations.

Our study is the second to show a relationship between gender and rs670 of *APOA1* gene with respect to criteria of Metabolic Syndrome or metabolic variables such as glucose, insulin and insulin resistance (HOMA-IR) in as much as only men displayed this association. Coban et al. [27] reported an interaction between this SNP and risk of atherogenic dyslipemia in males. Moreover, other authors have been reported significant associations of rs670 in both gender with hypertension [13], body mass index [14], diabetes mellitus [15] and metabolic syndrome [16]. We explain this gender interaction as a gene environment interaction, the environmental factor could be the diet or the hormonal status. For example, in the Philips et al. [30] study, the risk of developing MetS appeared to be modified by dietary fat intake, so the metabolic negative effect conferred by rs670 were worsened among individuals taking high-fat diet. And genotype did not affect phenotypes among subjects who habitually consumed a low-fat diet.

An important finding in our study is the relationship of rs670 with hyperglycemia and diabetes in males. The A allele has been associated with phenotypes related to reduce risk of diabetes such as higher HDL-C concentrations [31]. In other study, increased risk of impaired diabetes mellitus type 2 and impaired glucose tolerance have been reported in subjects with AA genotype, showing allele A as a metabolic risk factor [15]. *APOA1* gene is hypothesized to be stimulated by insulin through SP-1 binding elements [32] and ApoA1 rs670 as lying in a sequence homologous to the binding site for this nuclear factor SP-1. Such differences among studies may reflect genetic heterogeneity, differences in study designs, and other uncontrolled factors. Diabetes mellitus has a multifactorial polygenic origin, so that an abnormality in the promotor region of ApoA1 such as rs670 may act or interact with other environmental or genetic factors and induce to development diabetes. The potential effect of adipokines can be ruled out because we did not detect any relationship between this polymorphism and circulating adipokine levels. Only we reported the well-known gender interaction with leptin levels [33].

Limitations of our study are; one is, that the study has been realized in obese subjects, so the data are not generalizable to the entire population. The second might be the lack of determination of subclasses of HDL-C. The third, the design as a cross-sectional design does not allow to extract causality. The fourth, the lack of

assessing the combined effect of other genetic factors on lipid levels and metabolic parameters.

In conclusion, rs670 of *APOA1* gene has a gender specific influence on serum glucose, insulin, HOMA-IR, fat mass, weight and waist circumference. Males without A allele carriers showed high rates of central obesity, low levels of HDL, hyperglycemia and diabetes mellitus. Further designs are needed to study the role of these associations with possible diagnostic and therapeutic measures in obese patients at risk of metabolic syndrome.

Conflict of interest

The authors declare no conflicts of interest.

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