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Effect of the rs1862513 variant of resistin gene on insulin resistance and resistin levels after two hypocaloric diets with different fat distribution in subjects with obesity

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Abstract. – OBJECTIVE: Polymorphisms of a single nucleotide in resistin gene (RETN) have been associated with insulin resistance. We decide to investigate the role of this polymorphism on insulin resistance and resistin levels after two hypocaloric diets.

PATIENTS AND METHODS: A sample of 361 obese non-diabetic Caucasian was enrolled. Biochemical evaluation and anthropometric data were measured at the start of the trial and repeated after 3 months of both diets (Diet P, Polyunsaturated *vs.* diet M, Monounsaturated).

RESULTS: With both diets and in both genotype groups, BMI, weight, fat mass, waist circumference, systolic blood pressure, and diastolic blood pressure decreased. After diet P, insulin levels (GG *vs.* GC+CC genotypes) (-1.2±3.8 UI/L *vs.* -0.7±2.1 UI/L; $p<0.05$), HOMA-IR (-0.6±1.0 units *vs.* -0.4±0.9 units; $p<0.05$), total cholesterol (-10.5±20.1 mg/dl *vs.* -6.1±15.1 mg/dl; $p<0.05$) and LDL-total cholesterol (-8.6±10.1 mg/dl *vs.* -2.2±9.1 mg/dl; $p<0.05$) decreased in subjects with GG genotype. After diet M, insulin levels (-1.8±2.1 UI/L *vs.* -0.6±3.0 UI/L: $p>0.05$), HOMA-IR (-0.5±1.0 units *vs.* -0.3±1.1 units: $p>0.05$), total cholesterol (-9.5±13.1 mg/dl *vs.* -4.4±8.1 mg/dl; $p<0.05$) and LDL-total cholesterol (-8.1±6.1 mg/dl *vs.* -2.9±9.1 mg/dl; $p<0.05$) decreased, too.

CONCLUSIONS: We suggest that GG genotype of RETN rs1862513 could be a predictor of the reduction of HOMA-IR, insulin, and LDL cholesterol secondary to two hypocaloric diet in obese subjects.

Key Words:
Resistin, rs1862513 gene variant, Dietary fat.

Abbreviations
(BMI) = body mass index, (RETN) Resistin gene, HOMA-IR = Homeostasis model assessment-insulin resistance, CRP: C reactive protein.

Introduction

The clinical and biological significance of the adipokine resistin in the area of obesity remains uncertain. Resistin was identified as a gene whose expression is inhibited by peroxisome proliferator-activated receptor ligands and induced by adipocyte differentiation in 3T3-L1 cells[1]. In animal models, current evidence indicates that a reduction in functional resistin can improve insulin sensitivity and lower blood glucose levels[2]. In humans, circulating resistin levels are higher among patients with type 2 diabetes mellitus and obese subjects than healthy subjects[3,4], while other studies failed to observe such correlations[5]. The gene encoding resistin (*RETN*) is located on chromosome 19p13. Some single nucleotide polymorphisms (SNPs) described in the resistin gene have been associated with resistin levels[6,7], it is estimated that up to 70% of the variation in circulating resistin levels can be explained by genetic factors[8]. The association of these SNPs with body mass index (BMI) or other measures of adiposity has shown very inconsistent results[9,10]. In spite of everything previously mentioned, (SNPs) in *RETN* have been associated with indexes of insulin resistance[11]. One of these SNPs has been recently reported, showing that the GG genotype of RETN promoter SNP at -420 (rs1862513) is associated with diabetes mellitus type 2 susceptibility[12]. The relationship between changes in circulating levels of resistin after weight loss and this polymorphism has been poorly studied. In a recent design[13], it was reported that the mutant genotype of the variant (rs1862513) of the RETN gene was associated with a lack of change in resistin secondary to biliopancreatic diversion. In another interventional study[14], it was showed that

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the G/G genotype of RETN rs1862513 could be a predictor of the reduction of HOMA-IR, insulin, fasting glucose, and LDL cholesterol secondary to a standard hypocaloric diet in obese subjects. Recently, Cabrera et al[15] showed that serum resistin level is associated with fat intake. This relationship showed a positive association with saturated fat intake and inversely with a monounsaturated fat intake. At last, the actual view of adipose tissue is that of an active secretor organ, sending out and responding to signals that modulate lipid metabolism, chronic inflammation, immunity, appetite, insulin resistance, and energy expenditure. In attempting to understand the role of rs1862513 DNA variant in obese patients, we decide to investigate the role of this polymorphism on insulin resistance, resistin levels, and weight loss secondary to different hypocaloric diets.

Patients and Methods

Patients

A sample of 361 obese non-diabetic Caucasian outpatients was enrolled in a prospective way in Valladolid Spain. This study was conducted according to the guidelines laid down in the Declaration of Helsinki. These patients were recruited in a Nutrition Clinic Unit. All participants provided informed consent to a protocol approved by the local Ethical Review Boards. Inclusion criteria were: body mass index (BMI) ≥30 kg/m^2 and absence of a diet during the 6 months previously to the study. Exclusion criteria included history of cardiovascular disease or stroke during the previous 24 months, total cholesterol ≥200 mg/dl, triglycerides ≥200 mg/dl, blood pressure ≥140/90 mmHg, fasting plasma glucose ≥126 mg/dl, as well as the use of metformin, sulfonylurea, type IV inhibitors dipeptidyl drugs, thiazolidinediones, insulin, glucocorticoids, antineoplastic agents, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, psychoactive drugs, statins, and other antidyslipidemic drugs.

Procedure

Basal fasting glucose, insulin, homeostasis model assessment-insulin resistance (HOMA-IR), total cholesterol, low-density lipoprotein (LDL-cholesterol), high-density lipoprotein (HDL-cholesterol), plasma triglycerides concentration, adipokines (leptin, adiponectin, resistin), and C reactive protein were measured. These measurements were realized at the start of the trial and repeated after 3 months of both diets. Weight, height, BMI, fat mass by bioimpedance and blood pressure measures were measured at both times, too. These determinations were realized at the same time of the day (morning) at overnight fasting state for 10 hours. Rs 1862513 was evaluated in RETN gene.

Genotyping of rs1862513 RETN Polymorphism

Oligonucleotide primers and probes were designed with the Beacon Designer 5.0 (Premier Biosoft International®, Los Angeles, CA, USA). The polymerase chain reaction (PCR) was carried out with 50 ng of genomic DNA, 0.5 uL of each oligonucleotide primer (primer forward: 5'-ACGTTGGATGGAGCCTTCCCACTTCCAACA-3' and reverse 5'-ACGTTGGATGCACAGCCCCTGGCATTATC-3' in a 2 uL final volume (Thermal Cycler Life Tecnologies, Los Angeles, CA, USA). DNA was denaturated at 95ºC for 3 min; this was followed by 45 cycles of denaturation at 95ºC for 15 s, and annealing at 59.3º C for 45 s. The PCR was run in a 25 µL final volume containing 12.5 uL of IQTM Supermix (Bio-Rad®, Hercules, CA, USA) with hot start Taq DNA polymerase. Hardy-Weinberg equilibrium was verified (p=0.23).

Dietary Intervention

A total of 361 obese subjects were randomly allocated to one of two diets for a period of three months. Diet P (high polyunsaturated (PUFAs) fat hypocaloric diet) consisted of a diet of 1459 kcal per day, 45.7% of carbohydrates, 34.4% of lipids, and 19.9% of proteins). The distribution of fats was 21.8% of saturated fats, 55.5% of monounsaturated fats, and 22.7% of polyunsaturated fats (7 g per day of w-6 fatty acids, 2 g per day of w-3 fatty acids, and a ratio w6/w3 of 3.5). The second diet was Diet M (high monounsaturated fat hypocaloric diet). This diet had 1342 kcal per day with the next distribution of the percentage of macronutrients: 46.6% of carbohydrates, 34.1% of lipids, and 19.2% of proteins. The distribution of fats was 21.7% of saturated fats, 67.5% of monounsaturated fats, and 10.8% of polyunsaturated fats. The exercise program consisted of an aerobic exercise at least 3 times per week (60 min each). All enrolled subjects received instruction to record their daily dietary intake for three days including a weekend day. Records were reviewed by a dietitian and analysed with a computer-based data evaluation system

(Dietosource®, Gen, Gen, Sw). National composition food tables were used as reference[16]. In order to improve compliment of the calorie restriction and macronutrient distribution, the adherence of these diets was assessed every 7 days with a phone call by a dietitian.

Assays

Glucose metabolism: Fasting plasma glucose levels were determined by using an automated glucose oxidase method (Glucose analyser 2, Beckman Instruments, Brea, CA, USA). Insulin was measured by RIA (RIA Diagnostic Corporation, Los Angeles, CA, USA) with a sensitivity of 0.5 mUI/L (normal range 0.5-30 mUI/L)[17], and HOMA-IR was calculated using these values[18]. Lipid metabolism: Serum total cholesterol and triglyceride concentrations were determined by enzymatic colorimetric assay (Technicon Instruments, Ltd., New York, NY, USA), while HDL cholesterol was determined enzymatically in the supernatant after precipitation of other lipoproteins with dextran sulphate-magnesium. LDL cholesterol was calculated using the Friedewald formula[19]. Adipokines and inflammation: Resistin was measured by ELISA (Biovendor Laboratory, Inc., Brno, Czech Republic) with a sensitivity of 0.2 ng/ml with a normal range of 4-12 ng/ml[20]. Leptin was measured by ELISA (Diagnostic Systems Laboratories, Inc., Houston, TX, USA) with a sensitivity of 0.05 ng/ml and a normal range of 10-100 ng/ml[21]. Adiponectin was measured by ELISA (R&D Systems, Inc., Minneapolis, MN, USA) with a sensitivity of 0.246 ng/ml and a normal range of 8.65-21.43 ng/ml[22]. CRP was measured by immunoturbidimetry (Roche Diagnostics GmbH, Mannheim, Germany), with a normal range of (0-7 mg/dl) and analytical sensitivity 0.5 mg/dl.

Blood Pressure and Anthropometric Measurements

Body weight was measured to an accuracy of 0.1 kg and body mass index computed as body weight/(height^2). Waist (narrowest diameter between xiphoid process and iliac crest) and hip (widest diameter over greater trochanters) circumferences to derive waist-to-hip ratio (WHR) were measured, too. Tetrapolar body electrical bioimpedance was used to determine body composition with an accuracy of 50 g[19] (Akern EFG, Florence, FI, Italy). Blood pressure was measured twice after a 10 minutes rest with a random-zero mercury sphygmomanometer, and averaged.

Statistical Analysis

The sample size was calculated to detect differences over 3 kg in body weight after dietary intervention with 90% power and 5% significance (n=175, in each diet group). The Kolmogorov-Smirnov test was used to determine variable distribution. The distribution of variables was analyzed with Kolmogorov-Smirnov test. Quantitative variables with normal distribution were analyzed with a two-tailed Student's *t*-test. Non-parametric variables were analyzed with the Wilcoxon test. To retain a prescribed family-wise error rate α in our analysis involving more than one comparison, Bonferroni correction method was used. Qualitative variables were analyzed with the chi-square test, with Yates correction as necessary, and Fisher's test. The X^2-test was used to evaluate the Hardy-Weinberg equilibrium. The statistical analysis was performed for the combined GC and CC as a group (mutant group) and wild-type GG as second group, with a dominant model. Adjustment for potential confounder factors (BMI, age, and sex) was realized in statistical analysis. *p*-value <0.05 was considered statistically significant. SPSS Software version 15.0 (SPSS Inc., Chicago, IL, USA) was used.

Results

Three hundred and sixty-one patients were enrolled in the study. All patients completed the 3-month follow-up period without drop-outs in both branches (diet P *vs.* diet M). The mean age was 44.1±11.9 years and the mean BMI 37.5±5.1, with 101 males (28.0%) and 260 females (72.0%). One hundred and forty-six patients (40.5%) had the genotype GG (wild group) and 215 (59.5%) patients had the next genotypes; GC (105 patients, 29.1%) or CC (110 patients, 30.4%) (Mutant group). Mean ages were similar in both genotype groups (wild-type: 44.4±9.1 years *vs*. mutant group: 44.0±11.3 years: ns). Sex distribution was similar in both genotype groups, males (26.7% *vs.* 28.8%) and females (73.3% *vs.* 71.2%). In the group of 185 subjects (75 GG genotype and 110 C allele carriers) treated with diet P basal, basal assessment of nutritional intake with a 3 days written food record showed a calorie intake of 2016.1±691.6 kcal/day. The macronutrient distribution was: carbohydrate intake of 219.9±61.9 g/day (43.2% of calories), fat intake of 80.1±32.3 g/day (36.4% of calories) and protein intake of 88.9±37.1 g/day (20.4% of calories). During the

Table I. Changes in anthropometric variables (mean ± SD).

	rs1862513							
	Diet P				Diet M			
	GG		GC+CC		GG		GC+CC	
Characteristics	Basal	3 months	Basal	3 months	Basal	3 months	Basal	3 months
BMI	37.3±5.1	35.6±5.5*	37.9±6. 1	36.2±5.7*	37.5+4.6	35.9±4.2*	37.8±5.1	35.9±4.1*
Weight (kg)	96.7±19.6	92.2±18.1*	96.8±16.1	92.4±10.1*	95.1±12.6	92.3±9.1*	95.9±12.0	91.8±9.1*
Fat mass (kg)	40.2±11.1	36.6±10.1*	39.9±11.2	36.7±10.0*	39.5±9.4	36.5±9.0*	38.8±8.1	35.2±8.0*
WC (cm)	112.7±8.1	108.8±7.1*	113.4±10.1	109.2±10.2*	111.2±9.1	108.8±7.1*	114.7±8.0	110.8±6.2*
Waist to hip ratio	1.01±0.3	1.00±0.2	0.99±0.2	0.98±0.1	1.00±0.2	0.99±0.1	0.99±0.1	0.97±0.2
SBP (mmHg)	127.3±10.1	121.8±6.1*	126.6±12.6	124.2±10.1*	125.3±7.0	122.1±5.1*	128.7±7.2	123.6±9.1*
DBP (mmHg)	79.9±7.2	77.1±4.2*	80.9±6.1	78.4±6.2*	82.5±5.0	79.2±4.0*	82.2±5.0	80.0±5.1*

BMI: body mass index DBP, diastolic blood pressure; SBP, systolic blood pressure; WC, waist circumference; *$p<0.05$, in each genotype group. No statistical differences between genotype groups.

intervention, these subjects reached the recommendations of diet P: 1426.3 calories (45.2% of carbohydrates, 34.0% of lipids, and 20.8% of proteins). The distribution of dietary fats was 20.7% of saturated fats, 54.0% of monounsaturated fats, and 23.3% of polyunsaturated fats (6.5 g per day of w-6 fatty acids, 2.0 g per day of w-3 fatty acids, and a ratio w6/w3 of 3.2). In the group of 176 subjects (71 GG genotype and 105 C allele carriers) treated with diet M, basal assessment of nutritional intake with a 3 days written food record showed a calorie intake of 2012.8±339.1 kcal/day. Macronutrient distribution was carbohydrate intake of 198.7±34.3 g/day (43.0% of calories), fat intake of 66.3±19.2 g/day (34.0% of calories), and protein intake of 80.1±20.1 g/day (24.0% of calories). During the intervention, these patients reached the recommendations of diet: 1476.1 calories (45.1% of carbohydrates, 33.9% of lipids, and 21.0% of proteins). The distribution of dietary fats was 20.5% of saturated fats, 67.6% of monounsaturated fats, and 11.9% of polyunsaturated fats. Anthropometric changes of participants after dietary intervention are reported in Table I. With the diet type P and in wild-type and mutant type groups, BMI (GG *vs*. GC+CC genotypes) (-0.7±1.1 kg/m^2 *vs*. -0.7±1.2 kg/m^2: $p>0.05$), weight (-4.5±3.2 kg *vs*. -4.4±3.1 kg: $p>0.05$), fat mass (-3.6±4.1 kg *vs*. -3.2±3.0 kg: $p>0.05$), waist circumference (-4.1±3.9 cm *v*s. - 4.2±4.1 kg: $p>0.05$), systolic blood pressure (-5.5±4.1 mmHg *vs*. -5.4±3.1 mmHg: $p>0.05$), and diastolic blood pressure (-2.8±2.1 mmHg *vs*. -2.5±2.1 mmHg: $p>0.05$) decreased significantly. With the diet type M and in both genotypes, BMI (-1.4±2.0 kg/m^2 *vs*. -1.8±2.0 kg/m^2: $p>0.05$), weight (-2.8±2.9 kg *vs*. -4.0±3.1 kg: $p>0.05$), fat mass (-3.0±3.1 kg *vs*. -3.6±3.0 kg: $p>0.05$), waist circumference (-2.4±3.1 cm *vs*. -3.1±3.0 cm: $p>0.05$), systolic blood pressure (-3.2±4.1 mmHg *vs*. -5.1±3.0 mmHg: $p>0.05$), and diastolic blood pressure (-3.3±2.0 mmHg *vs*. -2.2±1.9 mmHg: $p>0.05$) decreased, too. There were no statistically significant differences in the changes induced by both diets in anthropometry and blood pressure. There were also no differences between baseline and nutritional post-treatment values in both groups. Table II shows the classic cardiovascular risk factors. After weight loss with the diet type P and in wild-type group, insulin levels (GG *vs.* GC+CC genotypes) (-1.2±3.8 UI/L *vs.* -0.7±2.1 UI/L; $p<0.05$), HOMA-IR (-0.6±1.0 units *vs.* -0.4±0.9 units; $p<0.05$), total cholesterol (-10.5±20.1 mg/dl *vs.* -6.1±15.1 mg/dl; $p<0.05$), and LDL- total cholesterol (-11.1±8.1 mg/dl *vs.* -2.2±9.1 mg/dl; $p<0.05$) decreased. Secondary to weight loss with the diet M and in wild genotype, insulin levels (-1.8±2.1 UI/L *vs*. -0.6±3.0 UI/L: $p>0.05$), HOMA-IR (-0.5±1.0 units *vs*. -0.3±1.1 units: $p>0.05$), total cholesterol (-9.5±13.1 mg/dl *vs.* -4.4±8.1 mg/dl; $p<0.05$), and LDL-total cholesterol (-8.1±6.1 mg/dl *vs.* -2.9±9.1 mg/dl; $p<0.05$) decreased. There were no statistically significant differences in the changes induced by both diets on insulin levels, lipid profile, and HOMA-IR. There were also no differences between baseline and nutritional post-treatment values in both groups. Table III shows levels of adipocytokines, with the diet P leptin levels (-5.2±2.1 ng/ml *vs.* -8.2±10.2 ng/ml: $p<0.05$) decreased in both genotypes. With the diet M, leptin levels (-6.3±11.1 ng/ml *vs*. -8.2±9.8 ng/ml: $p<0.05$) decreased, too. Resistin and adiponectin levels didn't change after

weight loss. There were no statistically significant differences in the changes induced by both diets on leptin levels.

Discussion

The present study suggests that the GG genotype of the RETN variant rs1862513 may be an independent predictor to the improvement induced by weight loss of lipid profile, insulin levels and insulin resistance secondary to two different hypocaloric diets in obese subjects. In the literature, there is a lack of information about the influence of lifestyle factors on this association in intervention studies. In a previous study[14], we reported the effects of a standard hypocaloric diet during 3 months (1520 calories per day with the next distribution of macronutrient; 52% of carbohydrates, 25% of lipids, and 23% of proteins) and the *RETN* variant rs1862513 on body weight loss and subsequent changes of metabolic parameters. GG genotype induces a better metabolic response than C allele carriers. In morbidly obese patients, 12 months after a biliopancreatic surgery[13], C allele carriers showed a lower response of insulin and HOMA-IR after massive weight loss (average more than 20 kg per year). Finally, Makino et al[23] showed after 12 weeks of treatment with pioglitazone a lack of response to insulin and HOMA-IR in diabetic subjects with C allele. In the literature, the effect of weight loss on resistin levels is a topic area with contradictory results. Some dietary interventions in obese subjects have demonstrated an increase of resistin levels after weight loss[24], and other studies have reported a decrease of this adipokine[25]. The presence of diabetes mellitus in the patients studied may be an independent variable that marks the response of resistin levels after weight loss. In one study[26], morbid obese subjects without diabetes mellitus showed an increase in the postoperative resistin levels and resistin remained unchanged in diabetic patients. The type of bariatric surgery could be another independent parameter in this unclear relationship. For example, Santoro et al[27] described after a sleeve gastrectomy, omentectomy, and enterectomy, a decrease in resistin levels. After laparoscopic gastric bypass[28] or a sleeve gastrectomy[29], resistin levels decreased, too. Moreover, Moschen et al[26] have demonstrated that resistin levels increased after laparoscopic adjustable gastric banding (LABG). Some drugs such as metformin have shown a decreased of resistin levels[30].

The reasons for the contradictories resistin results are unknown but may be due to overlapping feedback control mechanisms, unknown physiological pathways or interaction with the type of intervention to induce weight loss. Moreover, genetic background of obese patients could influence in this relationship as an uncontrolled bias. For example, Makino et al[23] suggest that PPAR-gamma ac-

Table II. Biochemical parameters (mean±SD). CRP: C reactive protein. HOMA-IR (homeostasis model assessment); $^{*}p<0.05$, in each genotype group. No statistical differences between genotype groups.

	rs1862513							
	Diet P				Diet M			
	GG		GC+CC		GG		GC+CC	
Characteristics	Basal	3 months	Basal	3 months	Basal	3 months	Basal	3 months
Glucose (mg/dl)	99.4±13.1	99.8±10.1	100.1±13.1	98.8±10.1	108.1±8.0	101.4±9.0	100.1±10.1	97.7±10.2
Total cholesterol (mg/dl)	211.9±22.7	201.4±20.2*	205.9±44.8	199.7±35.1	204.6±10.7	195.4±11.2*	198.9±18.1	194.4±20.1
LDL-cholesterol (mg/dl)	133.0±13.1	125.6±12.5*	124.7±18.1	122.6±31.1	128.2±22.1	120.1±10.4*	123.4±28.1	120.5±17.1
HDL-cholesterol (mg/dl)	53.8±10.1	51.2±11.4	53.3±11.2	52.8±11.1	49.9±7.1	49.8±8.1	50.5±11.0	51.7±11.2
Triglycerides (mg/dl)	131.9±60.1	131.6±50.2	118.3±30.9	117.9±33.1	136.2±31.1	136.8±19.4	125.4±31.9	123.6±23.1
CRP (ng/dl)	6.1±3.3	5.5±2.6	7.6±3.9	6.9±3.2	6.5±3.0	6.2±2.8	6.3±3.2	6.1±3.1
Insulin (mUI/l)	12.8±10.1	11.2±6.2*	12.7±6.8	11.5±7.6	13.4±7.1	11.6±5.2*	12.9±7.3	12.3±5.1
HOMA-IR	3.4±2.2	2.8±1.7*	3.2±2.7	2.8±2.7	3.4±1.8	2.9±1. 6*	3.2±2.1	2.9±2.0

CRP: C reactive protein. HOMA-IR (homeostasis model assessment); $^{*}p<0.05$, in each genotype group. No statistical differences between genotype groups

Table III. Adipokines and cytokine levels (mean6±SD).

Characteristics	rs1862513							
	Diet P				Diet M			
	GG		GC+CC		GG		GC+CC	
	Basal	3 months	Basal	3 months	Basal	3 months	Basal	3 months
Resistin (ng/dl)	6.8±4.1	6.4±3.2	6.7±4.5	6.2±3.9	7.1±4.0	7.0±3.1	6.9±1.9	7.1±2.2
Adiponectin (ng/dl)	11.5±7.1	10.4±6.5	9.3±5.0	8.7±4.3*	10.2±6.0	9.7±5.1	11.5±9.0	10.7±8.1
Leptin (ng/dl)	37.2±21.6	32.4±23.5*	43.4±22.4	35.2±16.0*	44.1±22.1	37.8±20.5*	42.5±11.8	34.2±9.0*

HOMA-IR (homeostasis model assessment); *$p<0.05$, in each genotype group. No statistical differences between genotype groups.

tivation with pioglitazone represses the expression of the resistin gene by modulating Sp1 activity and this fact induce a lack of response in subjects with minor allele. In our present study, in obese patients without diabetes mellitus, the improvement of insulin levels, HOMA-IR, total cholesterol, and LDL cholesterol levels were higher in wild-type group than mutant-type group with both hypocaloric diets. This better metabolic response is independent of dietary fat. Diet P and M produces the same improvement in anthropometric and biochemical parameters. However, Cabrera et al[15] showed that, in general population, resistin level is positively associated with saturated fat intake and inversely associated with monounsaturated fat intake. This study showed a linear relation of resistin levels with fat intake (changes of serum resistin levels of 1 mg/dl with fat dietary intake of 1 g per day). In some studies, it has been described an inverse association between resistin levels and cholesterol. This inverse relationship can be attributed to the ability of resistin to sequester serum lipids and store them in macrophages[31]. This relationship could be secondary to an unknown molecular interaction of transcriptors of RETN and metabolic or lipid metabolism. Other potential explanation is the effect of the type of hypocaloric diet. For example, duration of dietary intervention, macronutrient distribution, type of fat, and amount of protein could be parameters that influenced in these different metabolic responses. As far as we know this is the second study to explore the interaction of this RETN variant and modifications of serum adipokines concentrations secondary to weight loss due to a dietary intervention. Our report confirmed the lack of association of other previous study[14]. However, some researches have confirmed the relationship between rs1862513 SNP and expression of inflammatory molecules or adipokines[12,32,33].

Our work has some limitations. First, we only analysed one SNP of *RET* gene, so other genetic variants could be related to metabolic syndrome and comorbidities. Second, the self-reported dietary intake is also a limitation with potential bias in the design. Third, there are many uncontrolled factors that could influence our results (epigenetic, hormonal status, level of physical activity, etc.). Fourth, the possibility of racial-ethnic differences is not evaluated in our study, and only Caucasian subjects were enrolled. Finally, our interventional study is a short-term study of 3 months.

Conclusions

We suggest that G/G genotype of *RETN* (rs1862513) could be a predictor of the reduction of HOMA-IR, insulin, total cholesterol, and LDL cholesterol secondary to two different hypocaloric diets, one reached in polyunsaturated fatty acids and other in monounsaturated fatty acids.

Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the Institutional and/or National Research Committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. This article does not contain any studies with animals performed by any of the authors.

Informed consent

Informed consent was obtained from all individual participants included in the study.

Conflict of Interest

The Authors declare that they have no conflict of interest.

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