Caveolin-1 Genetic Polymorphism Interact with Polyunsaturated Fatty Acids to Modulate Metabolic Syndrome Risk

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Number of Tables: 6

Number of Figures: 1

Running Title: Interactions of dietary fatty acid and CAV-1

This peer-reviewed article has been accepted for publication but not yet copyedited or typeset, and so may be subject to change during the production process. The article is considered published and may be cited using its DOI 10.1017/S0007114521002221

The British Journal of Nutrition is published by Cambridge University Press on behalf of The Nutrition Society
Abstract

Several studies have reported a significant association between metabolic syndrome (MetS) and mortality around the world. Caveolin-1 (CAV-1) has been widely studied in dyslipidemia, and several studies have indicated that CAV-1 genetic variations may correlate with dietary intake of fatty acids. This study aimed to investigate the interaction of CAV-1 rs3807992 with types of dietary fatty acid in MetS risk. This cross-sectional study was carried out on 404 overweight and obese females. Dietary intake was obtained from a 147-item FFQ. The CAV-1 genotype was measured using the PCR-RFLP method. Anthropometric values and serum levels (TC, LDL, HDL, TG, FBS) were measured by standard methods. It was observed that the (AA+AG) group had significantly higher BMI, WC, and DBP (P=0.02, P=0.02, and P=0.01, respectively) and lower serum LDL, HDL, and TC (P < 0.05) than the GG group. It was found that A allele carriers were at higher odds of MetS (P= 0.01), abdominal obesity (P=0.06), increased TG concentration (P=0.01), elevated blood pressure (BP) (P=0.01), increased glucose concentration (P=0.45), and decreased HDL-cholesterol concentration (P=0.03). Moreover, the interaction of CAV-1 and SFA intake was significant in terms of MetS (P=0.03), LDL (P=0.03), and BP (P=0.01). Additionally, the (AA+AG) group was significantly related to PUFA intake in terms of MetS (P=0.04), TG (P=0.02), glucose (P=0.02), and HOMA-IR (P=0.01). Higher PUFA consumption might attenuate the CAV-1 rs3807992 associations with MetS, and individuals with greater genetic predisposition appeared to have a higher risk of MetS, associated with higher SFA consumption.

Keywords: Caveolin-1, Dietary fat intakes, Gene-diet interaction, Metabolic syndrome, Obesity
Introduction

Metabolic syndrome (MetS) is related to metabolic abnormalities including obesity, hyperglycemia, dyslipidemia, and hypertension, and is prevalent worldwide (1). Compared to people without MetS, people with metabolic syndrome had a higher overall number of deaths for all causes (2). Particularly, the evidence demonstrates that genetic variations play a main part in the prevention and treatment of various chronic diseases, particularly in MetS (3; 4; 5). MetS and cardiovascular diseases (CVD) also demonstrate the role of genetic and environmental factors in diet-related disorders. The genetic background of metabolic disorders has been shown to relate to death rate over the last few decades (6). There is therefore a need to identify the genes that derive MetS and to develop new therapies. CAV-1 is a key protein component of caveolae, and has been widely studied in dyslipidemia and CVD due to signal transduction, trafficking in cholesterol hemostasis, and triacylglycerol metabolism (7). Mice treated with CAV-1 are resistant to high-fat diets, and they show lipodystrophy, hypertension, insulin resistance, and abnormal glucose metabolism (8). Moreover, several studies have indicated that CAV-1 genetic variations might interact with other risk factors, including dietary intake of fatty acids, suggesting a positive association between CAV-1 and hypercholesterolemia (9). In studies conducted among Caucasian and Hispanic cohorts, the prevalence of CAV-1 gene variant rs926198 is related with higher odds of MetS risk and low HDL (10; 11). Also, CAV-1 overexpression has been observed to relate to higher odds of atherosclerosis in experimental models (12). The exact mechanisms are unclear, but it seems that CAV-1 is able to regulate several key enzymes in lipid metabolism, such as cholesterol ester transfer protein and phospholipid transfer protein (13). While the association between CAV-1 polymorphisms and type 2 diabetes risk has been widely reported in various populations (14), these relationships with MetS have been inconsistent, despite several publications on the association between CAV-1 gene variants and serum lipid profiles (14; 15; 16; 17). To the authors’ knowledge, there has been no study evaluating CAV-1 rs3807992 variant, metabolic risk factor, and the interaction of fatty acid intake levels with this SNP. Hence, the aim of the current study was to evaluate the interaction of CAV-1 genetic polymorphism with the types of dietary fatty acids, in terms of MetS risk factor status.
Material and Methods

Study Population

For this cross-sectional study, 404 women in the range of 18-55 years old were recruited from health referral centers. Participants provided written informed consent. The inclusion criteria were: obese or overweight, no alcohol consumption, and no smoking. CVDs, hypertension, type 2 diabetes (T2D), polycystic ovary syndrome (PCO), kidney failure, stroke, thyroid disease, liver disease, cancer, inflammatory diseases, and those who were on certain therapeutic drugs, weight loss programs, or supplements during the study time were all excluded. Each participant was interviewed in order to obtain demographic data, then referred to the laboratory for blood sampling. Anthropometric measures were taken, including: height (m), weight (kg), waist circumference (WC, cm) measured at the narrowest part of the abdomen, and body mass index (BMI, kg/m², calculated by dividing weight by height squared). BP was measured with a sphygmomanometer (BP) after 5 minutes’ rest. The study was approved by the Ethics Committee at the Tehran University of Medical Sciences (TUMS) (97-03-161-41017).

MetS Definition

MetS cases were required to meet three or more of the following criteria according to the Adult Treatment Panel III (ATP III) criteria:

1) Elevated fasting blood glucose FPG≥100 mg/dl.

2) Hypertriglyceridemia TG≥150 mg/dl

3) Elevated BP (≥130/85 mmHg)

4) Low (HDL-c) <50 mg/dl in women

5) WC cut-off 80 cm [women] was considered as an indicator of abdominal obesity\(^{(18)}\).

Dietary Assessment

The Food Frequency Questionnaire (FFQ) was a useful tool for evaluating dietary intake, and included 147 items\(^{(19)}\). This assessment was carried out by interviewing the occurrence of food items consumed on the basis of a predetermined list of foods. The extracted FFQ values were then changed to grams/day. For the evaluation of macro- and micronutrient content, N4 software was used; all measurements were then entered into IBM SPPS.
**Physical activity measurement:**

Physical activity was measured using the International Physical Activity Questionnaire (IPAQ) short form. Women’s physical activity was divided into three categories: low (under 600), mild (600-3500), and extreme (over 3500) (MET-h/wk). IPAQ's validity and reliability were tested in Iranian people. 

**Genotyping**

For genotyping the CAV-1 polymorphisms, DNA was extracted from whole blood via a Mini Columns kit (Type G; Genall; Exgene). The polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) technique was employed to investigate CAV-1 polymorphisms (rs3807992) in gene fragments (major allele G and minor allele A). PCR was carried out using the following primers: F:3’AGTATTGACCTGATTTGCCATG5’ R:5’GTCTTCTGGAAAAAGCACATGA-3’. PCR reactions were performed in a volume of 20 µl, containing 50 ng extracted DNA, 10 µM Forward primers, 10 µM, Reverse primers .7 µl distilled water and Taq DNA Polymerase Master Mix (2X) in a DNA thermocycler. The DNA templates were denatured at 94 °C for 3 min and 40 cycles, including a min denaturation at 94 °C, a min annealing at 42-50 °C and elongation at 72 °C for 2 min. Amplified DNA was digested with HinIII (NiaIII) (2.5 U) restriction enzyme at 37 °C overnight, then separated by electrophoresis on an agarose gel (2.5%). Fragments concluding three genotypes of the CAV-1 rs3807992 variant were detected: uncut homozygous AA (213bp), cut heterozygous GA (3 bands: 118 & 95 & 213 bp) and cut homozygous GG (2 bands: 118 & 95 bp).

**Biochemical analysis:**

All blood samples were drawn, after 12-14 hours of fasting, at the nutrition laboratory of TUMS. Blood samples were centrifuged for 10 minutes at 3000 rpm to extract serums, then aliquoted into 1 ml tubes and stored at 70 C before analysis. Auto-analyzer BT 1500 (Selectra 2; Vital Scientific, Spankeren, Netherlands) was used to test the samples. Fasting plasma glucose (FPG) was determined using the Glucose Oxidase Phenol 4-Aminoantipyrine Peroxidase (GOD/ PAP) process. Triacylglycerol kits (Pars Azmoon Inc, Tehran, Iran) were used to measure serum triglycerides (TG) by colorimetric method tests with Glycerol-3-phosphate oxidase Phenol 4-Aminoantipyrine Peroxidase (GPO-PAP). The cholesterol oxidase Phenol 4-Aminoantipyrine Peroxidase (CHOD-PAP) was used to calculate total cholesterol, and the direct method and immunoinhibition were used to measure low-density lipoprotein cholesterol.
lipoprotein (LDL) and high-density lipoprotein (HDL). Pars Azmoon (Pars Azmoon Inc., Tehran, Iran) provided all of the kits.

**Statistical Analysis**

Statistical analysis was performed using SPSS v.25 software (SPSS Inc., IL, USA) and significance level was considered $P < 0.05$. The Kolmogorov-Smirnov was utilized to test the normality of the data, and all data were expressed as means ± SD. The Pearson’s chi-square test was used to determine the Hardy-Weinberg Equilibrium deviation among allele frequency of CAV-1 G32124A (rs3807992). We used the dominant model for genetic analysis, risk allele carriers (AA, AG) compare to homozygotes major allele (GG). Medians and interquartile ranges (IQR) were calculated for dietary intakes between CAV-1 genotypes. Mann-Whitney U-test and Quade’s ANCOVA were used for associations between dietary and genotypes in crude and adjusted models respectively. Independent-sample t-test was used to evaluate the differences between the two groups. Binary logistic regression was used to estimate interactions between rs3807992 and high and low dietary fat intake on the Odds Ratio (OR) of the MetS component. Interaction analyze were adjusted for variables proven to be related to MetS components, (such as age, physical activity, energy intake, BMI, age at onset of obesity, and total SFA and PUFA intake).

**Results**

**Clinical Characteristics According to CAV-1 rs3807992 Genotypes**

The overall prevalence of rs3807992 genotypes was 38.34% and 61.66 % for the A, and G alleles. The genotype distributions had a deviation from HWE ($P < 0.05$). Table 1 expresses the mean clinical characteristics of all women according to CAV-1 genotypes. It was observed that the (AA+AG) group had significantly higher BMI, WC and DBP ($P=0.02$, $P=0.02$ and $P=0.01$ respectively). Furthermore, women with (AA+AG) allele had significantly lower serum LDL ($P=0.006$) and HDL ($P<0.0001$) than the GG group. Additionally, no significant differences were detected for age, height, weight, FBG, TG and SBP.

**Genotype Frequencies between MetS and Control Groups**

Distributions of allele frequencies and the effect size of rs3807992 on various genetic models (codominant, dominant, recessive) had significant differences between MetS and control groups. In the codominant model, the A homozygous allele for the CAV-1 (rs3807992) gene was 52 % higher in the control group than the MetS group (OR 2.52 [1.11, 5.70], $P=0.02$). In
the dominant model, the A homozygous allele for the CAV-1 (rs3807992) gene was 31% higher in the control group than the MetS group (OR 2.31[1.16,4.61], P=0.01) (Table 2).

**Associations between CAV-1 rs3807992 and the Risk of MetS and Its Components**

It was found that (rs3807992) A allele carriers were at higher odds of MetS (OR 2.31 [1.16, 4.16], P=0.01), abdominal obesity (OR 1.42 [0.98,2.06], P=0.06), increased TG concentration (OR 2.12 [1.13,3.95], P=0.01), elevated BP (OR 7.03 [1.43,34.44], P=0.01), increased glucose concentration (OR 0.7[0.27,1.78], P =0.45), and decreased HDL-cholesterol concentration (OR -1.4 [1.02,1.93] P=0.03) (Table 3).

**Dietary Intake Relationships between CAV-1 rs3807992 Gene Polymorphisms, and their Interaction on MetS and Its Components**

The results of dietary nutritional intake are described according to genotype groups, which show no significant differences were found in nutrient consumption values (Table 4).

It was observed that the interaction of CAV-1 rs3807992 gene polymorphism and SFA intake was significant on MetS (OR 5.60 [1.14,27.40], P=0.03) and its components, including LDL (OR 12.95 [1.21,24.69], P=0.03), BP (OR 6.52 [1.59,11.45], P=0.01) in multi-adjusted model (adjusted for age, energy intake, BMI, age at onset of obesity, and total PUFA intake). There was no further significant interaction between the (AA+AG) group and SFA intake on other biochemical parameters, including HDL and TG in both the crude and adjusted models (P >0.05) (Table 5).

Interestingly, when CAV-1 (AA+AG) group interaction was analyzed by PUFA intake, the relationship was significant in terms of MetS (OR -0.207 [0.04, 0.95], P=0.04) and its components, including TG (OR -0.27 [0.05, 0.8], P=0.02), FPG (OR -0.06 [0.005, 0.75], P=0.02) and HOMA-IR (OR -0.22 [0.06, 0.78], P= 0.01) in the multi-adjusted model (adjusted for age, energy intake, BMI, age at onset of obesity, and SFA intake) (Table 6).
Discussion

According to these findings, female A-allele carriers had significantly higher BMI, WC and DBP, and had lower serum LDL and HDL, compared to GG genotypes. This demonstrates that their clinical parameters are predisposed to MetS. A-allele carriers were at higher odds of MetS. To the authors’ knowledge, there is little evidence describing an association between CAV-1 and metabolic syndrome. It was observed that a genetic variant of CAV-1 (rs3807992) was associated with increased MetS risk, which is consistent with previous studies that revealed a significant association between the minor allele in CAV-1 variations and the odds of metabolic diseases (10; 11; 15; 17). Previous studies by our research team have shown association between CAV-1 rs3807992 and metabolic disease (21; 22). Also, compared with other candidate gene studies, no studies had evaluated SNP rs3807992 and MetS risk.

It is proposed that CAV-1 polymorphisms increase MetS risk through altered CAV-1 gene expression, attenuating dyslipidemia and hypertension, while impairing glucose and insulin homeostasis (12; 13; 14; 23). CAV-1 regulates signaling molecules, such as IRS1, that have a key role in appropriate insulin responses, protein kinase A (PKA), angiotensin II receptors, active blood pressure molecules, binding sites for calcium ions, insulin, lipids, and hormone metabolisms; all of these may affect various clinical traits of MetS (14; 24; 25; 26). Apart from genetic mutations, increasing evidence indicates that epigenetic modifications can provide an upstream regulatory switch to regulate the expression of CAV-1 thus leading to disease conditions (27). Most of the recent research focuses on the effects of methylation on CAV1 expression. MiRNA-103 and 107 are up-regulated in obese mice, which can lead to impaired glucose homeostasis, insulin receptor stabilization, and a receptive insulin signaling system (28; 29). In lipid metabolism, docosahexaenoic acid (DHA) has been shown to modify the transcriptome of miRNAs. The expression of miRNA-192 in endothelial cells is greatly increased by DHA, and CAV1 is estimated to be a target for miRNA-192. CAV1 expression is decreased when miRNA-192 is overexpressed (30).

Most previous intervention studies have shown that environmental factors, in particular dietary fat composition may alter the risk of MetS. Based on previous studies higher intake of SFA was detrimental to maintaining insulin sensitivity, whereas PUFA showed beneficial effects (31). Additionally, nutrigenetic research has indicated that dietary fat background can influence genotype-phenotype relations (32). In our study, the genetic association between CAV-1
polymorphisms and MetS by dietary fat intake was reported. High dietary SFA intake (≥25gr) especially accentuated the negative effects of rs3807992 in terms of MetS risk.

The evidence for the CAV1 gene and nutrient is limited so far, and it is mostly based on animal studies. In mice models, the role of dietary fat in the relationship between the CAV1 gene and lipid and glucose metabolism has been investigated previously. CAV-1 KO mice have demonstrated resistance to obesity-diet, as well as increased plasma levels of chylomicron/VLDL particles and triglycerides caused by a defect in lipid droplet formation and hydrolysis. (33) Under fat diet conditions, CAV-1 null mice have shown white adipose tissue atrophy and triglyceride deposition in plasma. Besides, CAV-1 knockout mice were fed a western-type diet represent a significant rise in VLDL and IDL/LDL (34).

Besides, caveolin-1 has also been observed moving from the plasma membrane to lipid droplets in response to free fatty acids (35). Caveolin-1’s function in lipid transport had also been confirmed by CAV-1 deficient mice’s lean phenotype, which had significantly higher serum levels of free fatty acids and triglycerides in the postprandial state (36; 37; 38). Accordingly, caveolae is a main center for several nutrient metabolisms through the cell membrane and plays a unique function in the uptake of various lipid and glucose metabolites (39). Caveolae is able to uptake fatty acid, triacylglycerol, and cholesterol in many tissues, which leads to an elevation in caveolae density in obese rats (40; 41). In line with previous studies, experimental studies have reported that sphingomyelin is a key phospholipid of caveolae. SFA intake may lead to increases in sphingolipids levels in the cardiac cell membranes, thus disrupting the caveolae contents (42; 43). Furthermore, CAV-1 mRNA levels are up-regulated by free cholesterol in human cells (44). An appropriate level of cholesterol in the cell membrane under dietary fat conditions causes the caveolae activity are disrupted. (45). Experimental studies suggest that lipid abnormalities can influence caveolae formation and function, thereby disrupting fatty acid metabolism and contributing to the development of metabolic syndrome and obesity (46).

A further novel finding is that high PUFA intake (≥6% energy) reduced the negative effects of rs3807992 in terms of MetS risk, with the greatest protection achieved by A-allele carriers. Caveolae membrane fatty acids (in the internal and external leaflet) is also significantly altered by n-3 PUFA intake, and is even able to change the function of the caveolae. In this regard, Chapkin et al. reported that in animal models, n-3 PUFA intake may modulate the function of caveolae proteins/lipid, affecting membrane fusion and cell-cell signaling, and improving insulin signaling (47). Previous studies have indicated that H-Ras and endothelial nitric-oxide
synthase (eNOS) are moved from caveolae in n-3 PUFA supplemented rats, which suppressed the Ras-dependent signaling and reduced BP, thereby lowering the MetS risk \(^{(48; 49)}\). CAV-1 prevents the production of nitric oxide. These findings demonstrate that PUFA intake affects CAV-1 abundance, which in turn affects eNOS activity and, as a result, vascular function \(^{(50; 51)}\).

Besides, CAV-1 can regulate fatty acid translocase (FAT/CD36) surface accessibility by acting as an intracellular shuttle for long-chain fatty acids (LCFAs) to lipid droplets, and thus indirectly control long fatty acid uptake \(^{(33; 37; 38)}\). FAT/CD36 is required for LCFA uptake. CAV-1 deficiency has previously been linked to a total loss of caveolae, the lack of FAT/CD36 cellular membrane expression, and a decrease in fatty acid uptake \(^{(52)}\).

In this research, we observed that a PUFA can be effective on glucose profile in risk allele carriers. Changes in insulin receptor (IR) activity are one of the proposed pathways for the effect of dietary fat and CAV-1 on metabolic markers. According to strong evidence, caveolae are implicated in the pathogenesis of IR and MetS \(^{(14)}\). In animals were fed a high-fat diet, Insulin receptor substrate 1 (IRS-1) and Protein Kinase B (Akt) showed reduced activity, indicating insulin resistance. These results point to a mechanism by which a high-cholesterol diet affected CAV-1 expression in vivo, as well as IR localization and function \(^{(14; 53; 54)}\). It is suggested that a correlation between hyperlipidemia and IR through caveolae under dietary fat conditions.

However, another mechanism by which fatty acids can alter the genetic risk posed by CAV-1 polymorphisms may also contribute to the Apolipoprotein A-1 (apoA1) \(^{(13)}\). Consistent with our results CAV-1–deficient animals fed a western-style diet have significantly lower apoA-I plasma levels. Loss of CAV-1 contributes to the development of metabolic syndrome, according to apoA-I levels, the key protein marker of HDL \(^{(55)}\).

The mechanism observed in the present study, by which fatty acids are able to modify the genetic risk posed by CAV-1 polymorphisms, remains unknown; further studies are needed to indicate such gene-diet interventions. Caveolae and its components may become useful sites for further investigation into treating MetS.
Limitations:

Several limitations can also be identified in the current study. Dietary intake was assessed by a FFQ, which is self-reported and thus dependent on patient memory. Due to financial limitations, it was not possible to perform western blot analysis to determine whether rs-3807992 SNP alters the expression of CAV-1. The focus of the current study was on dietary fat composition, but other nutrient components, including carbohydrates or fiber, can also play a role in the progression of MetS. Furthermore, lipid parameter measurements were taken while the subjects were fasted, which may have obscured differences in CAV-1 under fed conditions. Given the observational nature of the study, it is not possible to tell whether the associations which were identified in women (but not men) are of a causal nature. Finally, we did not include a normal-weight participant due to financial constraints, and instead focused on overweight and obese women as high-risk categories for metabolic syndrome. However, as the first study in this regard, we recommend that future research concentrate on normal-weight people both men and women.

Conclusion

To the authors’ knowledge, this is the first study presenting the association of a genetic variant of CAV-1 rs3807992 with the risk of MetS and its components, including TG, BP, and HDL level. However, further studies are needed to determine the strength of this association in a larger population; the contribution of this study is the novel finding that rs3807992 clearly predicts MetS among obese women. Analyses of the individual components of MetS confirmed that the rs3807992 variant is related to elevated BP, dyslipidemia, low HDL cholesterol, and high TG levels. Also, CAV-1 rs3807992 genotypes are sensitive to dietary SFA and PUFA, which allows individuals to monitor and adjust SFA and PUFA consumption accordingly. Finally, these results can be used in combination with a patient’s genetic history in order to provide more applicable and tailored nutritional advice for preventing or attenuating MetS in overweight and obese women.

Acknowledgments: We would like to thank all the individuals who participated in this project. This study was supported by a grant from TUMS (41017).

Conflict of Interest: The authors declare no conflict of interest.
Statement of authorship: FA contributed to conception, design, data analyses, data interpretation, and manuscript drafting. KHM supervised the study. All authors approved the final manuscript for submission.

List of Abbreviations: BMI: Body mass index, CVD: Cardiovascular disease, DBP: Diastolic blood pressure, DHA: Docosahexaenoic acid, EPA: Eicosapentaenoic acid, FBS: Fasting blood sugar, FFQ: Food frequency questionnaire, FA: Fatty acid, GWAS: Genome-wide association studies, LDL: Low-density lipoprotein, MA: Minor Allele, MetS: Metabolic syndrome, PCR: Polymerase chain reaction, RFLP: Restriction fragment length polymorphism, SNP: Single nucleotide polymorphism, TC: Total cholesterol, TG: Triglyceride, WC: Waist circumference
References

Accepted manuscript


36. A Caveolin Dominant Negative Mutant Associates with Lipid Bodies and Induces Intracellular Cholesterol Imbalance


44. <Caveolin mRNA levels are up-regulated by free cholesterol.pdf>.
### Table 1. Clinical characteristics of all subjects based on CAV-1 rs3807992 genotypes

<table>
<thead>
<tr>
<th></th>
<th>(GG)</th>
<th>(AG/AA)</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Mean ±SD</strong></td>
<td><strong>Mean ±SD</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age (year)</td>
<td>37.56± 9.49</td>
<td>35.75± 7.8</td>
<td>0.05</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>161.3±6.08</td>
<td>160.9±5.87</td>
<td>0.58</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>79.7±10.91</td>
<td>82.1±13.23</td>
<td>0.07</td>
</tr>
<tr>
<td>BMI (kg/m2)</td>
<td>30.6±4.01</td>
<td>31.6±4.46</td>
<td>0.02</td>
</tr>
<tr>
<td>WC (cm)</td>
<td>98.2±9.30</td>
<td>100.4±10.38</td>
<td>0.02</td>
</tr>
<tr>
<td>FPG (mg/dl)</td>
<td>87.9±8.6</td>
<td>86.9±5.75</td>
<td>0.36</td>
</tr>
<tr>
<td>LDL-C (mg/dl)</td>
<td>98.8±22.66</td>
<td>91.2±25.07</td>
<td>0.006</td>
</tr>
<tr>
<td>HDL-C (mg/dl)</td>
<td>49.7±1.16</td>
<td>44.0±1.16</td>
<td>&lt;0.0001</td>
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<tr>
<td>TC (mg/dl)</td>
<td>186.7±33.74</td>
<td>182.7±37.36</td>
<td>0.30</td>
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<tr>
<td>TG (mg/dl)</td>
<td>113.1±15.20</td>
<td>133.3±14.14</td>
<td>0.13</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>109.6±15.05</td>
<td>112.9±14.75</td>
<td>0.08</td>
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<tr>
<td>DBP (mmHg)</td>
<td>75.8±10.77</td>
<td>79.3±10.06</td>
<td>0.01</td>
</tr>
</tbody>
</table>

Values are presented as mean ± SD;

BMI: Body mass index; WC: Waist circumference; FPG: Fasting plasma glucose; LDL: Low density lipoprotein; HDL: High density lipoprotein; TC: Total cholesterol; TG: Triglyceride; SBP: Systolic blood pressure; DBP: Diastolic blood pressure.

Comparisons between groups were determined based on independent-samples t test.

Bold values indicate statistical significance (P < 0.05).
Table 2. *CAV-1 rs3807992* frequencies between metabolic syndrome (MetS) and control groups

<table>
<thead>
<tr>
<th>Models</th>
<th>SNP rs3807992</th>
<th>Frequencies for the <em>CAV-1</em> SNP</th>
<th>Odds Ratio(95% CI)</th>
<th>P-value</th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Control</td>
<td>MetS</td>
<td></td>
</tr>
<tr>
<td>Codominant</td>
<td>GG</td>
<td>31(18.4%)</td>
<td>13(19.7%)</td>
<td>2.04(0.79,5.26)</td>
</tr>
<tr>
<td></td>
<td>AG</td>
<td>41(24.7%)</td>
<td>23(34.8%)</td>
<td>2.52(1.11,5.70)</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>94(55.2%)</td>
<td>30(45.5%)</td>
<td>0.06</td>
</tr>
<tr>
<td>Dominant</td>
<td>GG</td>
<td>94(56.6%)</td>
<td>30(45.5%)</td>
<td>2.31(1.16,4.61)</td>
</tr>
<tr>
<td></td>
<td>AG/AA</td>
<td>72(43.4%)</td>
<td>36(54.5)</td>
<td>2.04(0.97,4.31)</td>
</tr>
<tr>
<td></td>
<td>AG/GG</td>
<td>125(75.3%)</td>
<td>43(65.2%)</td>
<td></td>
</tr>
<tr>
<td>Recessive</td>
<td>AA</td>
<td>41(24.7%)</td>
<td>23(34.8%)</td>
<td></td>
</tr>
</tbody>
</table>

MetS: metabolic syndrome

Comparisons between groups were determined based on logistic regression analysis

Bold values indicate statistical significance (P < 0.05).
### Table 3. Associations between CAV-1 rs3807992 and the risk of MetS and its components

<table>
<thead>
<tr>
<th></th>
<th>OR (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>MetS</strong></td>
<td>2.31(1.16,4.61)</td>
<td>0.01</td>
</tr>
<tr>
<td>Abdominal obesity ≥80 cm</td>
<td>1.42(0.98,2.06)</td>
<td>0.06</td>
</tr>
<tr>
<td><strong>BP</strong> ≥130/85 mmHg</td>
<td>7.03(1.43,34.44)</td>
<td>0.01</td>
</tr>
<tr>
<td>FPG ≥100 mg/dl</td>
<td>0.7(0.27,1.78)</td>
<td>0.45</td>
</tr>
<tr>
<td>HDL-C &lt;50 mg/dl</td>
<td>-1.4(1.02,1.93)</td>
<td>0.03</td>
</tr>
<tr>
<td><strong>TG</strong> ≥150 mg/dl</td>
<td>2.12(1.13,3.95)</td>
<td>0.01</td>
</tr>
</tbody>
</table>


OR (95% CI): odds ratio (95% confidence interval)

Comparisons between groups were determined based on binary logistic regression analysis adjusted by age.

Bold values indicate statistical significance (P < 0.05).
Table 4. Dietary fat intakes of all subjects based on *CAV-1 rs3807992* genotypes

<table>
<thead>
<tr>
<th></th>
<th>GG Median</th>
<th>GG IQR</th>
<th>AG/AA Median</th>
<th>AG/AA IQR</th>
<th>Pvalue*</th>
<th>Pvalue†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Energy (Kcal)</td>
<td>2490.89</td>
<td>1137.99</td>
<td>2567.98</td>
<td>1237.64</td>
<td>0.5</td>
<td></td>
</tr>
<tr>
<td>Total Fat (gr)</td>
<td>89.24</td>
<td>46.04</td>
<td>90.50</td>
<td>51.60</td>
<td>0.64</td>
<td>0.46</td>
</tr>
<tr>
<td>Protein (gr)</td>
<td>88.23</td>
<td>41.65</td>
<td>87.59</td>
<td>43.77</td>
<td>0.73</td>
<td>0.79</td>
</tr>
<tr>
<td>Carbohydrate (gr)</td>
<td>351.45</td>
<td>158</td>
<td>355.30</td>
<td>166</td>
<td>0.51</td>
<td>0.45</td>
</tr>
<tr>
<td>SFA (gr)</td>
<td>25.59</td>
<td>14.61</td>
<td>26.38</td>
<td>14.44</td>
<td>0.91</td>
<td>0.45</td>
</tr>
<tr>
<td>Cholesterol (gr)</td>
<td>245.47</td>
<td>146.83</td>
<td>237.49</td>
<td>140.89</td>
<td>0.23</td>
<td>0.42</td>
</tr>
<tr>
<td>Total fiber (gr)</td>
<td>43.75</td>
<td>25.19</td>
<td>44.31</td>
<td>28.10</td>
<td>0.56</td>
<td>0.54</td>
</tr>
<tr>
<td>PUFA intake, %E</td>
<td>4.75</td>
<td>3.01</td>
<td>5.95</td>
<td>2.5</td>
<td>0.47</td>
<td>0.75</td>
</tr>
<tr>
<td>MUFA intake, %E</td>
<td>10.56</td>
<td>3.37</td>
<td>10.55</td>
<td>3.63</td>
<td>0.69</td>
<td>0.31</td>
</tr>
<tr>
<td>n-6 PUFA intake, %E</td>
<td>5.30</td>
<td>3</td>
<td>5.38</td>
<td>2.47</td>
<td>0.51</td>
<td>0.54</td>
</tr>
<tr>
<td>n-3 PUFA intake, %E</td>
<td>0.41</td>
<td>0.23</td>
<td>0.43</td>
<td>0.26</td>
<td>0.46</td>
<td>0.51</td>
</tr>
</tbody>
</table>

Values are represented as Median (IQR).

(P value*): crude model.

(P value†): adjusted model by potential confounding factors (age, energy intake, educational level, DBP)

Mann-Whitney U-Test and Quade’s ANCOVA were performed to identify significant differences between *CAV-1 rs3807992* genotypes in crude and adjusted model respectively.
Table 5. Interactions between the *CAV-1 rs3807992* and SFA intake in relation to MetS and its components

<table>
<thead>
<tr>
<th>MetS</th>
<th>SFA (≥25 g/d)*rs3807992</th>
<th>OR (95% CI) (AG/ AA)</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>1(Ref) GG</td>
<td></td>
<td></td>
<td>0.03</td>
</tr>
<tr>
<td>MetS</td>
<td></td>
<td>5.60(1.14,27.40)</td>
<td></td>
</tr>
<tr>
<td>BP ≥130/85 mmHg</td>
<td></td>
<td>6.52(1.59,11.45)</td>
<td>0.01</td>
</tr>
<tr>
<td>HDL-C &lt;50 mg/dl</td>
<td></td>
<td>-0.87(-6.32,4.58)</td>
<td>0.75</td>
</tr>
<tr>
<td>LDL &gt;100 mg/dl</td>
<td></td>
<td>12.95(1.21,24.69)</td>
<td>0.03</td>
</tr>
<tr>
<td>TG ≥150 mg/dl</td>
<td></td>
<td>12.64(-17.89,43.19)</td>
<td>0.41</td>
</tr>
</tbody>
</table>

MetS: metabolic syndrome, BP: blood pressure, HDL: high density lipoprotein, LDL: Low density lipoprotein, TG: triglycerides.

P-value *: adjusted model by potential confounding factors (age, physical activity, energy intake, BMI, age at onset of obesity, and total PUFA intake)

Binary logistic was performed
Table 6. Interactions between the CAV-1 rs3807992 and PUFA intake in relation to MetS and its components

<table>
<thead>
<tr>
<th></th>
<th>PUFA (≥6% energy) *rs3807992</th>
<th>OR (95% CI) (AG/AA)</th>
<th>Pvalue*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1(Ref) GG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MetS</td>
<td>-0.2(0.04,0.95)</td>
<td>0.04</td>
<td></td>
</tr>
<tr>
<td>*TG ≥150 mg/dl</td>
<td>-0.2(0.05,0.8)</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>FPG ≥100 mg/dl</td>
<td>-0.06(0.005,0.75)</td>
<td>0.02</td>
<td></td>
</tr>
<tr>
<td>HOMA-IR ≥2.7</td>
<td>-0.22(0.06,0.78)</td>
<td>0.01</td>
<td></td>
</tr>
</tbody>
</table>

MetS: metabolic syndrome, TG: triglycerides, FPG: Fasting plasma glucose; Homeostasis model assessment insulin resistance (HOMA-IR)

P-value *: adjusted model by potential confounding factors (age, physical activity, energy intake, BMI, age at onset of obesity, and total SFA intake)

Binary logistic was performed
Figure 1. Adjusted ORs (95%CI) for MetS and its components according to median of dietary fat intake and CAV-1 rs3807992
MetS: metabolic syndrome, BP: blood pressure, HDL: high density lipoprotein, TG: triglycerides, LDL: Low density lipoprotein, HOMA-IR: The homeostatic model assessment, OR: odds ratio
P₁: High dietary PUFA intake (≥6% energy) adjusted for age, physical activity, energy intake, BMI, age at onset of obesity, and total SFA intake.
P₂: High dietary SFA intake (≥25gr) adjusted for age, physical activity, energy intake, BMI, age at onset of obesity, and total PUFA intake.
The lowest median of dietary fat intake and homozygote genotype of major allele (GG) was used as the reference group.