EasyChair Preprint

№ 14239

Association of Plasma Adiponectin Levels and ADIPOQ Rs266729 Gene Variants with the Risk of Obesity in a Vietnamese Population

Phat Tung Ma, Nam Quang Tran, Linh Le Hoang Gia and Nien Vinh Lam

August 1, 2024

Association of Plasma Adiponectin Levels and ADIPOQ rs266729 Gene Variants with the Risk of Obesity in a Vietnamese Population

Phat Tung Ma[1,2][0000-0003-1715-0323], Nam Quang Tran[1,2] , Linh Le Hoang Gia[1], Nien Vinh Lam[1,2]

[1] University of Medicine and Pharmacy, Ho Chi Minh City, Vietnam
[2] University Medical Center, Ho Chi Minh City, Vietnam
matungphat@ump.edu.vn

Abstract. Obesity is a growing metabolic health issue in Vietnam. Lower adiponectin levels are reported in obesity. Variants of the adiponectin gene are promising candidates in the study of obesity-related conditions due to their possible impact on insulin production. Therefore, we investigated the associations of rs266729 gene variants with obesity risk and adiponectin levels. We collected anthropometric and biochemical data from 100 obese adults and 100 age- and sex-matched control subjects. Obesity was defined as having both a high body mass index ($\geq$ 25 kg/m^2) and central obesity. Adiponectin levels were measured using a latex-enhanced immunoturbidimetric method. The genotyping of rs266729 was analyzed using tetra-primer ARMS-PCR methods. In this study, the prevalence of rs266729 genotypes CC, GC, and GG was 68%, 28%, and 5%, respectively. Compared to genotype CC, subjects with genotypes GG and GC presented significantly higher obesity risk with odds of 1.89. The median adiponectin level in the obese subjects was 4.35 µg/ml, significantly lower compared to 9.31 µg/ml in the controls. Adiponectin levels were independently associated with obesity in multivariate analysis (OR=0.75, $p<0.001$). Nevertheless, adiponectin levels did not differ between rs266729 genotypes. In conclusion, the association of plasma adiponectin and ADIPOQ rs266729 gene variants was determined. However, the link between adiponectin and rs266729 gene variants was not confirmed. Further investigations, including extensive and diverse populations in Vietnam, are needed to clarify these findings.

Keywords: Adiponectin, ADIPOQ, obesity.

1 Introduction

Obesity is a complex disease characterized by excessive fat deposits, which increases the risk of cardiometabolic diseases worldwide. In Vietnam, obesity is becoming an emerging public health issue, contributing to a progressive rise in non-communicable diseases [1]. Data from 2009 to 2015 shows an increasing prevalence of obesity, from 10.1% to 17.6% [2], using the criteria of body mass index (BMI) $\geq$ 25 kg/m^2, following the guidance of World Health Organization specific for the Asian populations [3].

Despite ongoing preventive strategies in Vietnam, the prevalence of obesity is going to escalate as a result of a Westernized diet and sedentary lifestyles [1]. While BMI correlates well with total body fat, its interpretation in Asian and elderly individuals requires consideration [4, 5]. Normal BMI may conceal excess fat, and subjects with normal BMI can suffer from cardiometabolic complications [6–9]. Central obesity, defined as having an increased waist circumference (WC), is an anthropometric indicator of health risk [7]. People with increased WC show a stronger correlation with visceral fat, insulin resistance, and inflammation compared to high BMI [10]. In a recent report, central obesity, in reference to the International Diabetes Federation (IDF) 2006 criteria, was found to have consistently link with several metabolic-related traits in an Asian population [2]. The co-occurrence of central obesity and high BMI can also exacerbate the risk of cardiovascular disease [11, 12].

Adiponectin, secreted from adipose tissue, has several metabolically protective effects, including enhancing insulin sensitivity, anti-atherogenic effects, and anti-inflammation [13]. Therefore, it holds significant potential against the development of cardiometabolic diseases. Adiponectin can have weight-reducing effects by promoting lipid oxidation within skeletal muscle and other metabolically active organs, including the pancreas and liver [14]. Notably, plasma adiponectin levels manifest a negative correlation with BMI [15], suggesting its inverse correlation with adiposity.

Hara [16] discovered 13 single nucleotide polymorphisms (SNP) of the adiponectin gene (ADIPOQ) in French and Japanese populations. Of these SNPs, rs266729, with a G>C substitution at position -11377 in the promoter region, has attracted considerable attention in research on obesity and metabolic-related traits. This genetic variant is likely to possess a significant impact on adiponectin production, thereby contributing to metabolic risk. Several studies have found that individuals carrying at least one G allele of rs266729 have reduced plasma adiponectin levels, as well as an elevated risk of obesity compared to those without the G allele [17–21]. However, some studies have found no statistically significant association [22–24]. The effects on plasma adiponectin levels and the occurrence of obesity associated with ADIPOQ SNP levels appear to conflict because of differences in age, ethnicity, environmental influences, and sample populations [25, 26]. Our previous study highlights an association between rs266729 and type 2 diabetes [27], suggesting that rs266729 could be linked to other metabolic disturbances in the Vietnamese population. Given the growing burden of obesity in Vietnam, the Ministry of Health has, for the first time, published guidelines for the diagnosis and management of obesity [28]. Studying the underlying mechanisms and genetic background could help identify strategies for future individualized obesity management. Therefore, we set out to assess the potential associations between variants of the ADIPOQ gene and plasma adiponectin levels and obesity in Vietnamese people.

2 Methods

2.1 Study population and enrollment

We enrolled unrelated participants in this case-control study at the University Medical Center, Ho Chi Minh City (HCMC), Vietnam, from September 2023 to December

2023. The study protocol received agreement from the Ethical Committee of the University of Medicine and Pharmacy, HCMC (Number 381/HĐĐĐ-ĐHYD on March 22nd, 2023). Before participating in our study, every eligible subject had to understand the study procedure, risks, and benefits. Consequently, they were required to provide informed written consent.

We defined obesity as having as having both a BMI ≥ 25 kg/m^2 and central obesity. Our study excluded the obese subjects having significant renal or hepatic disease and those having any conditions or using any substance affecting weight. We selected participants with BMI < 25 kg/m^2 and no central obesity to be controls. Subjects with confirmed diabetes, having any conditions or using any substances affecting weight were excluded. Most of the study subjects were patients or regular visitors for health checkups at the study hospital. The study subjects in each group were recruited to be sex- and age-matched with those in the control group.

2.2 Clinical and Laboratory Measurements

Standardized questionnaires were utilized to collect subjects' demographic data and medical histories. The process involved trained personnel administering the questionnaires. Anthropometric measurements, including height, weight, and WC were obtained following a standard technique. Subjects' weights and heights were measured in the morning while they were standing without shoes and dressing in lightweight clothing. The measurement of WC was performed at the midpoint between the lower ribs and the top of the iliac crest. Following the IDF criteria for East Asian populations, the cutoff of WC for central obesity was ≥ 90 cm in males and ≥ 80 cm in females [29]. BMI was computed by dividing weight in kilograms by height in meters squared. The obesity status of study subjects was classified based on specific guidelines established for the Vietnamese population with a cutoff of 25 kg/m^2 [28]. Trained nurses performed systolic and diastolic blood pressure measurements after the study subjects had rested for a minimum of 5-10 minutes.

We collected two blood samples from each study subject: one for biochemical measurements and the other for genomic DNA extraction and later genotyping. Analyses of biochemical parameters were performed from overnight fasting blood samples. Measurements of glucose and lipid panels were immediately performed at the study hospital. In addition, the remaining plasma samples of study subjects were preserved at -20°C to maintain the integrity for additional future measurements of key metabolic parameters involving fasting plasma insulin and adiponectin levels in Hoa Hao Medic Center, HCMC. The measurements of adiponectin levels were performed using a latex-enhanced immunoturbidimetric assay with a Diazyme kit on a Beckman Coulter AU5800 autoanalyzer (Beckman Coulter Diagnostics, Brea, California). Plasma insulin levels were measured using a Roche Elecsys® kit on a Roche cobas e 801 autoanalyzer (Roche Diagnostics, Rotkreuz, Switzerland). All the biochemical measurements followed manufacturer protocols. The insulin resistance index was computed using the homeostasis model assessment-insulin resistance equation as described in reference [30]

$$\text{HOMA-IR} = \text{fasting glucose (mmol/L)} \times \text{fasting insulin (}\mu\text{U/mL)} / 22.5 \quad (1)$$

2.3 Genotyping

Following manufacturer instructions, we extracted genomic DNA within 24 hours using a GeneJETTM whole-blood genomic DNA purification kit (Thermo Fisher Scientific, MA, USA). The genotyping of rs266729 was performed using a tetra-primer amplification refractory mutation system-polymerase chain reaction (ARMS-PCR). A set of four specific primers (Table 1) was designed using the CLC Main Workbench software by downloading the human ADIPOQ gene sequence from NCBI (NG_021140) and then using the software primer design tool to design the PCR primers for SNP ADIPOQ genotyping.

Table 1. Primers and PCR product sizes for genotyping rs266729 variants

	Primers sequences	PCR product sizes
Forward outer primer	5'- TTGTTGAAGTTGGTGCTGGC-3'	447 bp
Reverse outer primer	5'- GAACCGGCTCAGATCCTGCC-3'	
Forward inner primer	5'- CACGCTCATGTTTTGTTTTTGAGGC-3'	372 bp
Reverse inner primer	5'- GCTTGTGGCCTCGAATCGTA-3'	119 bp

PCR, polymerase chain reaction

Each PCR tube contained the following components: 1.5µl of 10x PCR buffer; 1.5µl of 2.5mM dNTPs; 1.5µl of each forward and reverse primer pair (10nM/µl) both inside and outside; 0.1µl of TaKaRa Taq^{TM} HotStart Polymerase (Takara Bio Inc., Japan); 2µl of genomic DNA (10-50ng/µl); and 8.4µl of twice-ion-exchanged distilled water. Negative controls without DNA were included in the reactions to monitor contamination. The thermal cycling procedure for PCR was performed using a Mastercycler® Pro S machine (Eppendorf, Germany). PCR products were stored at 16°C and were then electrophoresed on a 2% agarose gel. The lengths of the PCR products were 447 bp, 119 bp, and 372 bp (Table 1). Samples with the GG genotype displayed bands at 447 bp and 19 bp; CC samples showed bands at 447 bp and 372 bp; and GC samples exhibited bands at 447 bp, 372 bp and 119 bp on the electrophoresis results (Fig. 1).

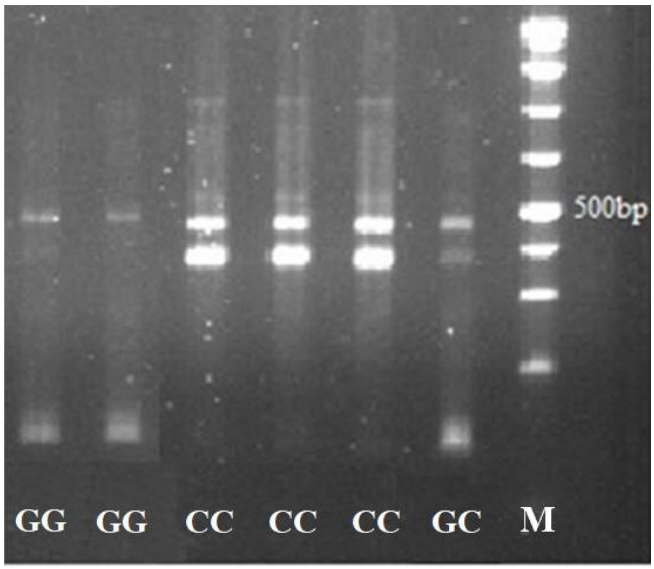

Fig. 1. Polymerase chain reaction product sizes by agarose gel electrophoresis for genotyping rs266729. M: DNA marker, Genotype CC: bands at 447 bp and 372 bp, GC genotype: bands at 447 bp, 372 bp and 119 bp, GG genotype: bands at 447 bp and 119 bp.

We decided to use the tetra-primer ARMS-PCR method for our SNP study due to its rapid results and cost-effectiveness. DNA sequencing was performed at the rs266729 locus in 20 randomly selected control samples to verify the compatibility of the genotyping methods. The direct sequencing protocol was described previously [31].

2.4 Statistical Analyses

The data for age, height, weight, BMI, WC, systolic and diastolic blood pressure, glucose, and HDL cholesterol were summarized using means and standard deviation as these variables followed a normal distribution. On the contrary, non-normal distribution variables including LDL cholesterol, triglycerides, insulin, adiponectin and HOMA-IR were described as medians and interquartile ranges. Depending on data properties, comparisons between the groups of subjects with obesity and the group of control subjects were performed using Student's t-test or the Mann-Whitney U test.

Sex and genotype frequencies were described in numbers and percentages. The distribution of genotype frequencies was examined to ensure that they adhered to the principles of Hardy-Weinberg equilibrium (HWE) using exact tests. In this study, we employed SNPStats, a designed web-based tool specialized in assessing SNP association studies, to test for the relationship of rs266729 polymorphisms with the likelihood of developing obesity under multiple inheritance models [32]. Age and sex were used as covariates for statistical adjustment in all genetic model analyses. Multivariate logistic regression analysis was employed to test the associations of obesity with adiponectin

levels, and anthropometric and metabolic variables. Odds ratios (OR) were calculated with a 95% confidence interval (CI).

Data analyses were performed using the software Stata v.14.0 (StataCorp LLC, TX, USA). P-value <0.05 was considered statistically significant.

3 Results

3.1 Anthropometrics and Metabolic Features of All Study Subjects

200 unrelated subjects (100 obese subjects and 100 control subjects), aged from 25 to 66, were recruited for data collection and genotyping. All anthropometric and metabolic features of the obese subjects and the controls were described and compared in Table 2. The sex distribution and mean age were similar between the two study groups. The mean age was 46,33 ± 8,67 years in the obese subjects and was 47.1 ± 8,79 years in the control subjects. Compared to the controls, subjects with obesity had higher blood pressures, along with increased glucose and lipid levels, except for HDL cholesterol levels, which showed an inverse tendency. These differences were statistical differences between the subjects with obesity and the controls. Moreover, the computed HOMA-IR was observed to be greater in the obese subject. Adiponectin levels were significantly reduced in the group of obese subjects (p<0.001). Specifically, the median adiponectin levels in the obese subjects and the control subjects were 4.35(2.44-6.87) µg/ml and 9.31(6.59-13.66) µg/ml, respectively.

Table 2. Comparison of anthropometric and metabolic features between obese subjects and control subjects

Variables	Obese subjects (n=100)	Control subjects (n=100)	P-value
Sex			
Male	50 (50%)	50 (50%)	1
Female	50 (50%)	50 (50%)	
Age* (year)	46.33 ± 8,67	47.1 ± 8,79	0.536
Height* (cm)	161.18 ± 7,83	160.45 ± 7,94	0.329
Weight* (kg)	73.01 ± 11,18	53.53 ± 5,88	<0.001
Body mass index* (kg/m^2)	28.01 ± 2.96	20.78 ± 1.54	<0.001
Waist circumference* (cm)	95.42 ± 9.40	76.43 ± 5.58	<0.001
Blood pressure (mmHg)			
Systolic*	133.52 ± 15.39	121.06 ± 13.68	<0.001
Diastolic*	85.46 ± 11.93	77.86 ± 9.75	<0.001
Lipid panel (mmol/L)			
Total cholesterol*	5.34 ± 1.28	4.87 ± 0.8	0.002
LDL cholesterol**	3.58 (2.75-4.14)	3.25 (2.75-3.63)	0.012
HDL cholesterol*	1.14 ± 0.24	1.39 ± 0.31	<0.001
Triglycerides**	2.41 (1.89-3.42)	1.18 (0.89-1.59)	<0.001
Insulin** (µU/mL)	9.25 (5.96-13.65)	4.06 (2.75-5.71)	<0.001

Glucose* (mmol/l)	6.72 ± 1.50	5.1 ± 0.38	<0.001
HOMA-IR**	2.62 (1.67-4.07)	0.90 (0.60-1.3)	<0.001
Adiponectin** (µg/ml)	4.35 (2.44-6.87)	9.31 (6.59-13.66)	<0.001

Data are : *mean ± standard deviation; **median (interquartile range). LDL, low-density lipoprotein; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment - insulin resistance.

3.2 Associations of Clinical and Biochemical Parameters with Obesity Risk

The associations of obesity risk with anthropometric and metabolic features are shown in a multivariate analysis, as detailed in Table 3. The parameters, including age, sex, total cholesterol, and LDL cholesterol levels, were not statistically linked to obesity ($p>0.05$). However, our analysis revealed a decreased risk of obesity in subjects with higher HDL cholesterol levels (OR = 0.88, 95% CI = 0.81-0.95, $p<0.001$) and adiponectin levels (OR = 0.75, 95% CI = 0.62-0.89, p=0.002). On the other hand, we found positive associations between obesity and several other metabolic parameters. There were relatively weak associations between triglyceride levels (OR = 1.01, 95% CI = 1-1.02, p=0.021), systolic blood pressure (OR =1.07, 95% CI = 1.01-1.12, p=0.014) and obesity. Notably, HOMA-IR showed a particularly strong positive association with obesity (OR = 5.1, 95% CI = 2.27-11.48, p< 0,001).

Table 3. Multivariate logistic regression analysis for the risk of obesity

Variables	OR (95% CI)	P-value
Male sex	0.26 (0.05-1.13)	0.072
Age (year)	1.06 (0.98-1.15)	0.120
Blood pressure (mmHg)		
Systolic	1.07 (1.01-1.12)	0.014
Diastolic	1.09 (1.01-1.17)	0.19
Lipid panel (mmol/L)		
Total cholesterol	1.04 (0.98-1.11)	0.194
LDL cholesterol	0.97 (0.9-1.04)	0.340
HDL cholesterol	0.88 (0.81-0.95)	0.001
Triglycerides	1.01 (1-1.02)	0.021
HOMA-IR	5.1 (2.27-11.48)	<0.001
Adiponectin (µg/ml)	0.75 (0.62-0.89)	0.002

OR, odds ratio; CI, confidence interval; HDL, high-density lipoprotein; LDL, low-density lipoprotein; HOMA-IR, homeostasis model assessment - insulin resistance

3.3 Association between rs266729 Genotypes and Obesity Risk

For all subjects, the frequencies of rs266729 genotype CC, GC, and GG were 68%, 28%, and 5%, respectively. HWE was tested and maintained in the entire study subjects (p=0.17), obese subjects (p=0.42) and control subjects (p=0,39), indicating no genotyping bias. The association between rs266729 genotypes and obesity was analyzed in multiple inheritance models. All analyses were performed with age and sex adjustment.

While the overall genotype distribution differed between obese subjects and control subjects (details in Table 4), only the dominant model presented a statistically significant association. In the dominant model, individuals carrying at least one G allele (GG and GC genotypes) were associated with a higher likelihood of obesity compared to subjects carrying only the C allele or CC genotype (OR = 1.89, 95% CI = 0.02-3.48, p = 0.04). However, the recessive and co-dominant models did not show significant associations despite the higher prevalences of the GC and GC genotypes in the group of obese subjects.

Table 4. The associations of rs266729 gene variants with obesity risk

Models	Genotypes	Obese subjects (n=100)	Control subjects (n=100)	OR (95% CI)*	P-value
Co-dominant	CC	61	74	1	0.094
	GC	32	23	1.75 (0.92-3.34)	
	GG	7	3	2.91 (0.72-11.77)	
Dominant	CC	61	74	1	0.04
	GC + GG	39	26	1.89 (1.02-3.48)	
Recessive	CC + GC	93	97	1	0.18
	GG	7	3	2.46 (0.62-9.82)	

* Adjusted for age and sex. OR, odds ratio; CI, confidence interval

3.4 Association between rs266729 Genotypes and Adiponectin Levels

Given the established relationship between GC + GG genotypes of rs266729 and obesity risk, we further investigated adiponectin levels between the genotypes across the study groups. However, our study revealed that adiponectin levels did not differ significantly between genotypes in either the group of obese subjects or the control subjects, as detailed in Table 5. Specifically, median adiponectin levels remained comparable across genotypes (CC vs GC + GG) in both obese subjects (4.47 vs. 4.00 μg/ml, p=0.356) and the control subjects (9.18 vs. 9.88 μg/ml, p=0.512).

Table 5. Comparisons of plasma adiponectin levels between rs266729 genotypes

Genotypes	n	Adiponectin levels	P-value
Total			
CC	135	7.13 (4.22-10.17)	0.25
GG+GC	65	6.52 (2.92-9.49)	
Obese subjects			
CC	61	4.47 (2.64-7.26)	0.356
GG+GC	39	4 (2.03-6.64)	
Control subjects			
CC	75	9.18 (6.42-14.2)	0.12
GG+GC	26	9.88 (7.21-13.12)	

Data are median and interquartile range

4 Discussion

The ADIPOQ rs266729, located on the promoter region, was assessed for obesity risk in this Vietnamese population, as well as its relationship with adiponectin levels. The C allele has been found to be the major allele of rs266729 in several studies involving different ethnicities. Genotypes containing at least one allele G (GG and GC) of rs266729 are reported to link with hypoadiponectinemia, increased body weight, and central obesity [17–21, 33]. Both higher BMI and increased WC are associated with insulin resistance, metabolic-related traits, and, ultimately, severe health risks, e.g., metabolic syndromes and cardiovascular events. Hence, in this study, we defined obesity as having both a high BMI and increased WC, to test for the association with rs266729 polymorphism, thereby strengthening our preliminary investigation. The BMI cutoff followed Vietnamese guidelines (BMI ≥25 kg/m^2) [28], while the WC cutoff followed the IDF criteria for central obesity specific to the East Asian population [29].

In the 200 subjects included in this analysis, the testing of HWE for rs266729 genotypes' frequencies was confirmed, demonstrating the validity of our study population. The G allele genotypes had lower frequencies; however, they were associated with a higher likelihood of obesity in the dominant inheritance model analysis. This finding in a Vietnamese population is consistent with findings in Korean [20], Chinese [34], Finnish [35], Nigerian [32], and Croatian adults [19]. However, this association has not been seen in Caucasian Americans [22], African Americans [23], or Italians [36]. A meta-analysis including 18 association studies evaluating various ADIPOQ SNPs and the risk of obesity, Lu [26] showed that the SNP rs266729 presents a significant association with obesity in Asian ethnicities, while it did not demonstrate an increased risk with Caucasian ethnicities. A subgroup analysis based on BMI criteria (≥25 kg/m²) further supported this association of rs266729 in Asian populations, reinforcing the ethnicity-specific impact of this SNP on obesity. Moreover, in a cross-sectional study of

obesity measures, Ogundele [33] demonstrated that rs266729 polymorphism is related to several anthropometric parameters of obesity in a linear regression analysis.

The obese subjects of our study had substantially lower adiponectin levels than the controls. Even after multivariate adjustment, the lower adiponectin levels remained independently associated with obesity risk. This kind of association aligns with most previous reports, indicating that adiponectin may have a beneficial impact on obesity. Specifically, increased levels of circulating adiponectin are linked to lower obesity [37, 38]. In rats, transgenic overexpression of adiponectin prevents the onset of diet-induced obesity [39]. Additionally, adiponectin administration is proven to reduce insulin resistance and lower glycemic levels in experimental studies [40–42]. The precise mechanisms by which adiponectin achieves these favorable metabolisms are incompletely understood. Nonetheless, adiponectin possesses insulin-mimetic effects, as well as enhances insulin sensitivity[43]. Furthermore, Liu [44] showed that adiponectin can promote fatty acid oxidation, resulting in lower body triglyceride levels. Conversely, some studies could not find any links between adiponectin levels and body weight [45, 46].

The SNP rs266729 has been demonstrated to be linked with plasma adiponectin levels in particular studies. Specifically, genotypes containing the G allele have lower adiponectin levels compared to the CC genotype [17, 18, 20]. However, this relationship was not observed in our study. Similarly, a study including young Croatians found statistical associations between the SNP rs266729, as well as various metabolic parameters, but could not show a direct relation between the study SNP and circulating adiponectin levels [19]. In another association study including the SNP rs266729 in South Asia, adiponectin levels did not differ between genotypes [47]. Several factors may account for this lack of association. First, the restricted number of subjects might reduce the statistical power to detect significant associations. Second, we measured total adiponectin levels without distinguishing between the various isoforms of adiponectin; therefore, interpretation of adiponectin levels requires caution, as the multimeric form of adiponectin has primary biological effects significantly contributing to metabolic syndrome, compared to the trimeric and hexametric forms [48–50]. Additionally, our study utilized a case-control design to evaluate the link between the studying SNP and obesity. Thus, the ability to detect differences in adiponectin levels across different genotypes is relatively weak. Finally, the study included only one SNP of the ADIPOQ gene for analysis. Studies have shown that other SNPs located in the promotor region [51], exon [52], or even in an intron of the ADIPOQ gene [53] impact circulating adiponectin levels. Furthermore, interactions with other genes associated with obesity may have influenced the results. Haplotype analysis of SNPs rs266729 and rs17300539 demonstrated an association with adiponectin levels in a population of French Caucasians [54].

Our study may be considered the first Vietnamese study to provide preliminary findings on the relationship between ADIPOQ gene variants and obesity. Compared with our previous study, this present study collected data on plasma adiponectin levels along with ADIPQ SNP genotyping. Nevertheless, we could not show variations in adiponectin levels between genotypes. Data analysis did not include covariates, such as eating patterns, smoking, alcohol status, levels of physical exercise, or other environmental factors, despite their effects on weight and plasma adiponectin levels. These variables

should be accounted for in future studies examining gene-environment interactions. Additionally, only SNP rs266729 was analyzed; other ADIPOQ SNPs and related gene variants could have a considerable impact on adiponectin levels, as well as the risk of obesity. Future studies should also include haplotype analysis involving other SNPs for profound analyses of their impacts on obesity. Lastly, all participants were recruited from our hospital in HCMC with a small sample size, so it is uncertain whether our findings could be generalized nationally.

From the present study's results, we conclude that the GC and GG genotypes of rs266729 are associated with high BMI and increased WC in a population of Vietnamese adults. This reinforces our comprehension of how ADIPOQ SNPs may potentially influence body composition and their role in metabolism. However, our findings are preliminary and may require reassessment for various regions and ethnic groups of Vietnam for better confirmation.

Acknowledgments.
We acknowledge the University Medical Center, HCMC, for providing support with subject enrollment and blood sample storage, and Hoa Hao Medic Center for performing insulin and adiponectin measurements.

References

1. Nguyen, T.T., Hoang, M.V.: Non-communicable diseases, food and nutrition in Vietnam from 1975 to 2015: the burden and national response. Asia Pac J Clin Nutr. 27, 19–28 (2018). https://doi.org/10.6133/apjcn.032017.13
2. Pham, T., Bui, L., Giovannucci, E., Hoang, M., Tran, B., Chavarro, J., Willett, W.: Prevalence of obesity and abdominal obesity and their association with metabolic-related conditions in Vietnamese adults: an analysis of Vietnam STEPS survey 2009 and 2015. Lancet Reg Health West Pac. 39, 100859 (2023). https://doi.org/10.1016/j.lanwpc.2023.100859
3. IASO: The Asia-Pacific Perspective: redefining obesity and its treatment, https://iris.who.int/bitstream/handle/10665/206936/0957708211_eng.pdf
4. Wellens, R.I., Roche, A.F., Khamis, H.J., Jackson, A.S., Pollock, M.L., Siervogel, R.M.: Relationships between the Body Mass Index and body composition. Obes Res. 4, 35–44 (1996). https://doi.org/10.1002/j.1550-8528.1996.tb00510.x
5. Jeong, S.-M., Lee, D.H., Rezende, L.F.M., Giovannucci, E.L.: Different correlation of body mass index with body fatness and obesity-related biomarker according to age, sex and race-ethnicity. Sci Rep. 13, 3472 (2023). https://doi.org/10.1038/s41598-023-30527-w
6. Choi, D., Choi, S., Son, J.S., Oh, S.W., Park, S.M.: Impact of Discrepancies in General and Abdominal Obesity on Major Adverse Cardiac Events. J Am Heart Assoc. 8, e013471 (2019). https://doi.org/10.1161/JAHA.119.013471
7. Janssen, I., Katzmarzyk, P.T., Ross, R.: Waist circumference and not body mass index explains obesity-related health risk. Am J Clin Nutr. 79, 379–384 (2004). https://doi.org/10.1093/ajcn/79.3.379
8. Sun, Y., Liu, B., Snetselaar, L.G., Wallace, R.B., Caan, B.J., Rohan, T.E., Neuhouser, M.L., Shadyab, A.H., Chlebowski, R.T., Manson, J.E., Bao, W.: Association of Normal-Weight Central Obesity With All-Cause and Cause-Specific Mortality Among Postmenopausal

Women. JAMA Netw Open. 2, e197337 (2019). https://doi.org/10.1001/jamanetworkopen.2019.7337
9. Kim, H.Y., Kim, J.K., Shin, G.G., Han, J.A., Kim, J.W.: Association between Abdominal Obesity and Cardiovascular Risk Factors in Adults with Normal Body Mass Index: Based on the Sixth Korea National Health and Nutrition Examination Survey. J Obes Metab Syndr. 28, 262–270 (2019). https://doi.org/10.7570/jomes.2019.28.4.262
10. Tchernof, A., Després, J.-P.: Pathophysiology of human visceral obesity: an update. Physiol Rev. 93, 359–404 (2013). https://doi.org/10.1152/physrev.00033.2011
11. Cong, X., Liu, S., Wang, W., Ma, J., Li, J.: Combined consideration of body mass index and waist circumference identifies obesity patterns associated with risk of stroke in a Chinese prospective cohort study. BMC Public Health. 22, 347 (2022). https://doi.org/10.1186/s12889-022-12756-2
12. Zhu, S., Heshka, S., Wang, Z., Shen, W., Allison, D.B., Ross, R., Heymsfield, S.B.: Combination of BMI and Waist Circumference for Identifying Cardiovascular Risk Factors in Whites. Obes Res. 12, 633–645 (2004). https://doi.org/10.1038/oby.2004.73
13. Khoramipour, K., Chamari, K., Hekmatikar, A.A., Ziyaiyan, A., Taherkhani, S., Elguindy, N.M., Bragazzi, N.L.: Adiponectin: Structure, Physiological Functions, Role in Diseases, and Effects of Nutrition. Nutrients. 13, 1180 (2021). https://doi.org/10.3390/nu13041180
14. Yamauchi, T., Kamon, J., Waki, H., Terauchi, Y., Kubota, N., Hara, K., Mori, Y., Ide, T., Murakami, K., Tsuboyama-Kasaoka, N., Ezaki, O., Akanuma, Y., Gavrilova, O., Vinson, C., Reitman, M.L., Kagechika, H., Shudo, K., Yoda, M., Nakano, Y., Tobe, K., Nagai, R., Kimura, S., Tomita, M., Froguel, P., Kadowaki, T.: The fat-derived hormone adiponectin reverses insulin resistance associated with both lipoatrophy and obesity. Nat Med. 7, 941–946 (2001). https://doi.org/10.1038/90984
15. Yang, W.-S., Lee, W.-J., Funahashi, T., Tanaka, S., Matsuzawa, Y., Chao, C.-L., Chen, C.-L., Tai, T.-Y., Chuang, L.-M.: Plasma adiponectin levels in overweight and obese Asians. Obes Res. 10, 1104–1110 (2002). https://doi.org/10.1038/oby.2002.150
16. Hara, K., Boutin, P., Mori, Y., Tobe, K., Dina, C., Yasuda, K., Yamauchi, T., Otabe, S., Okada, T., Eto, K., Kadowaki, H., Hagura, R., Akanuma, Y., Yazaki, Y., Nagai, R., Taniyama, M., Matsubara, K., Yoda, M., Nakano, Y., Tomita, M., Kimura, S., Ito, C., Froguel, P., Kadowaki, T.: Genetic variation in the gene encoding adiponectin is associated with an increased risk of type 2 diabetes in the Japanese population. Diabetes. 51, 536–540 (2002). https://doi.org/10.2337/diabetes.51.2.536
17. de Luis, D.A., Izaola, O., Primo, D., Aller, R.: Relation of a variant in adiponectin gene (rs266729) with metabolic syndrome and diabetes mellitus type 2 in adult obese subjects. Eur Rev Med Pharmacol Sci. 24, 10646–10652 (2020). https://doi.org/10.26355/eurrev_202010_23422
18. Tankó, L.B., Siddiq, A., Lecoeur, C., Larsen, P.J., Christiansen, C., Walley, A., Froguel, P.: ACDC/adiponectin and PPAR-gamma gene polymorphisms: implications for features of obesity. Obes Res. 13, 2113–2121 (2005). https://doi.org/10.1038/oby.2005.262
19. Karmelić, I., Lovrić, J., Božina, T., Ljubić, H., Vogrinc, Ž., Božina, N., Sertić, J.: Adiponectin level and gene variability are obesity and metabolic syndrome markers in a young population. Arch Med Res. 43, 145–153 (2012). https://doi.org/10.1016/j.arcmed.2012.02.004
20. Park, J.W., Park, J., Jee, S.H.: ADIPOQ Gene Variants Associated with Susceptibility to Obesity and Low Serum Adiponectin Levels in Healthy Koreans. Epidemiol Health. 33, e2011003 (2011). https://doi.org/10.4178/epih/e2011003
21. Hammad, S.S., Eck, P., Sihag, J., Chen, X., Connelly, P.W., Lamarche, B., Couture, P., Guay, V., Maltais-Giguère, J., West, S.G., Kris-Etherton, P.M., Bowen, K.J., Jenkins, D.J.A., Taylor, C.G., Perera, D., Wilson, A., Castillo, S., Zahradka, P., Jones, P.J.H.:

Common Variants in Lipid Metabolism–Related Genes Associate with Fat Mass Changes in Response to Dietary Monounsaturated Fatty Acids in Adults with Abdominal Obesity. J Nutr. 149, 1749–1756 (2019). https://doi.org/10.1093/jn/nxz136
22. Warodomwichit, D., Shen, J., Arnett, D.K., Tsai, M.Y., Kabagambe, E.K., Peacock, J.M., Hixson, J.E., Straka, R.J., Province, M., An, P., Lai, C.-Q., Parnell, L.D., Borecki, I., Ordovas, J.M.: The monounsaturated fatty acid intake modulates the effect of ADIPOQ polymorphisms on obesity. Obesity (Silver Spring). 17, 510–517 (2009). https://doi.org/10.1038/oby.2008.583
23. Beebe-Dimmer, J.L., Zuhlke, K.A., Ray, A.M., Lange, E.M., Cooney, K.A.: Genetic Variation in Adiponectin (ADIPOQ) and the Type-1 Receptor (ADIPOR1), Obesity and Prostate Cancer in African Americans. Prostate Cancer Prostatic Dis. 13, 362–368 (2010). https://doi.org/10.1038/pcan.2010.27
24. Cieslak, J., Skorczyk, A., Stachowiak, M., Szydlowski, M., Grzes, M., Paczynska, P., Skowronska, B., Majewska, K., Stankiewicz, W., Fichna, P., Switonski, M.: Polymorphisms in 5'-flanking regions of genes encoding adiponectin, leptin, and resistin are not associated with obesity of Polish children and adolescents. Mol Biol Rep. 38, 1793–1798 (2011). https://doi.org/10.1007/s11033-010-0294-9
25. Yang, W.-S., Chuang, L.-M.: Human genetics of adiponectin in the metabolic syndrome. J Mol Med (Berl). 84, 112–121 (2006). https://doi.org/10.1007/s00109-005-0011-7
26. Lu, J., Zhou, Y., Huang, G., Jiang, H., Hu, B., Qin, S.: Association of ADIPOQ polymorphisms with obesity risk: a meta-analysis. Hum Immunol. 75, 1062–1068 (2014). https://doi.org/10.1016/j.humimm.2014.09.004
27. Truong, S., Tran, N.Q., Ma, P.T., Hoang, C.K., Le, B.H., Dinh, T., Tran, L., Tran, T.V., Gia Le, L.H., Vu, H.A., Mai, T.P., Do, M.D.: Association of ADIPOQ Single-Nucleotide Polymorphisms with the Two Clinical Phenotypes Type 2 Diabetes Mellitus and Metabolic Syndrome in a Kinh Vietnamese Population. Diabetes Metab Syndr Obes. 15, 307–319 (2022). https://doi.org/10.2147/DMSO.S347830
28. Về việc ban hành tài liệu chuyên môn "Hướng dẫn chẩn đoán và điều trị bệnh béo phì," https://emohbackup.moh.gov.vn/publish/home?documentId=8975
29. Alberti, K.G.M.M., Zimmet, P., Shaw, J.: Metabolic syndrome--a new world-wide definition. A Consensus Statement from the International Diabetes Federation. Diabet Med. 23, 469–480 (2006). https://doi.org/10.1111/j.1464-5491.2006.01858.x
30. Matthews, D.R., Hosker, J.P., Rudenski, A.S., Naylor, B.A., Treacher, D.F., Turner, R.C.: Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia. 28, 412–419 (1985). https://doi.org/10.1007/BF00280883
31. Kiet, N.C., Khuong, L.T., Minh, D.D., Nguyen The Vinh, Quan, N.H.M., Xinh, P.T., Trang, N.N.C., Luan, N.T., Khai, N.M., Vu, H.A.: Spectrum of mutations in the RB1 gene in Vietnamese patients with retinoblastoma. Mol Vis. 25, 215–221 (2019)
32. Solé, X., Guinó, E., Valls, J., Iniesta, R., Moreno, V.: SNPStats: a web tool for the analysis of association studies. Bioinformatics. 22, 1928–1929 (2006). https://doi.org/10.1093/bioinformatics/btl268
33. Ogundele, O.E., Adekoya, K.O., Osinubi, A.A.A., Awofala, A.A., Oboh, B.O.: Association of adiponectin gene (ADIPOQ) polymorphisms with measures of obesity in Nigerian young adults. Egyptian Journal of Medical Human Genetics. 19, 123–127 (2018). https://doi.org/10.1016/j.ejmhg.2017.08.005
34. Wang, X., Zhang, S., Chen, Y., Liu, H., Lan, C., Chen, X., Chi, S., Chen, S., Zhang, W.: APM1 gene variants −11377C/G and 4545G/C are associated respectively with obesity and

with non-obesity in Chinese type 2 diabetes. Diabetes Research and Clinical Practice. 84, 205–210 (2009). https://doi.org/10.1016/j.diabres.2009.03.004
35. Siitonen, N., Pulkkinen, L., Lindström, J., Kolehmainen, M., Eriksson, J.G., Venojärvi, M., Ilanne-Parikka, P., Keinänen-Kiukaanniemi, S., Tuomilehto, J., Uusitupa, M.: Association of ADIPOQ gene variants with body weight, type 2 diabetes and serum adiponectin concentrations: the Finnish Diabetes Prevention Study. BMC Med Genet. 12, 5 (2011). https://doi.org/10.1186/1471-2350-12-5
36. Daniele, A., Cammarata, R., Pasanisi, F., Finelli, C., Salvatori, G., Calcagno, G., Bracale, R., Labruna, G., Nardelli, C., Buono, P., Sacchetti, L., Contaldo, F., Oriani, G.: Molecular analysis of the adiponectin gene in severely obese patients from southern Italy. Ann Nutr Metab. 53, 155–161 (2008). https://doi.org/10.1159/000172976
37. Cohen, S.S., Gammon, M.D., North, K.E., Millikan, R.C., Lange, E.M., Williams, S.M., Zheng, W., Cai, Q., Long, J., Smith, J.R., Signorello, L.B., Blot, W.J., Matthews, C.E.: ADIPOQ, ADIPOR1, and ADIPOR2 polymorphisms in relation to serum adiponectin levels and BMI in black and white women. Obesity (Silver Spring). 19, 2053–2062 (2011). https://doi.org/10.1038/oby.2010.346
38. Khabour, O.F., Alomari, M.A., Abu Obaid, A.A.: The Relationship of adiponectin level and ADIPOQ gene variants with BMI among young adult women. Dermatoendocrinol. 10, e1477902 (2018). https://doi.org/10.1080/19381980.2018.1477902
39. Shklyaev, S., Aslanidi, G., Tennant, M., Prima, V., Kohlbrenner, E., Kroutov, V., Campbell-Thompson, M., Crawford, J., Shek, E.W., Scarpace, P.J., Zolotukhin, S.: Sustained peripheral expression of transgene adiponectin offsets the development of diet-induced obesity in rats. Proc Natl Acad Sci U S A. 100, 14217–14222 (2003). https://doi.org/10.1073/pnas.2333912100
40. Wang, Y., Li, Y., Qiao, J., Li, N., Qiao, S.: AMPK α1 mediates the protective effect of adiponectin against insulin resistance in INS-1 pancreatic β cells. Cell Biochem Funct. 37, 625–632 (2019). https://doi.org/10.1002/cbf.3440
41. Shi, W., Guo, Z., Ji, Y., Feng, J.: The protective effect of recombinant globular adiponectin on testis by modulating autophagy, endoplasmic reticulum stress and oxidative stress in streptozotocin-induced diabetic mice. Eur J Pharmacol. 879, 173132 (2020). https://doi.org/10.1016/j.ejphar.2020.173132
42. Liu, Y., Turdi, S., Park, T., Morris, N.J., Deshaies, Y., Xu, A., Sweeney, G.: Adiponectin corrects high-fat diet-induced disturbances in muscle metabolomic profile and whole-body glucose homeostasis. Diabetes. 62, 743–752 (2013). https://doi.org/10.2337/db12-0687
43. Fang, X., Sweeney, G.: Mechanisms regulating energy metabolism by adiponectin in obesity and diabetes. Biochem Soc Trans. 34, 798–801 (2006). https://doi.org/10.1042/BST0340798
44. Liu, Y., Sen, S., Wannaiampikul, S., Palanivel, R., Hoo, R.L.C., Isserlin, R., Bader, G.D., Tungtrongchitr, R., Deshaies, Y., Xu, A., Sweeney, G.: Metabolomic profiling in liver of adiponectin-knockout mice uncovers lysophospholipid metabolism as an important target of adiponectin action. Biochem J. 469, 71–82 (2015). https://doi.org/10.1042/BJ20141455
45. Kuo, S.-M., Halpern, M.M.: Lack of association between body mass index and plasma adiponectin levels in healthy adults. Int J Obes (Lond). 35, 1487–1494 (2011). https://doi.org/10.1038/ijo.2011.20
46. Pereira, R.I., Wang, C.C., Hosokawa, P., Dickinson, L.M., Chonchol, M., Krantz, M.J., Steiner, J.F., Bessesen, D.H., Havranek, E.P., Long, C.S.: Circulating adiponectin levels are lower in Latino versus non-Latino white patients at risk for cardiovascular disease, independent of adiposity measures. BMC Endocr Disord. 11, 13 (2011). https://doi.org/10.1186/1472-6823-11-13

47. Nadeem, A., Mumtaz, S., Naveed, A.K., Mansoor, Q., Aslam, M., Ismail, M.: Association of ADIPOQ C-11377G (rs266729), TNF-α αG-308A(rs1800629) and TNF-α αG-238A(rs361525) Single Nucleotide Polymorphisms with Type 2 Diabetes Mellitus in Pakistani Population. J Coll Physicians Surg Pak. 27, 625–630 (2017)
48. Fisher, F.F.M., Trujillo, M.E., Hanif, W., Barnett, A.H., McTernan, P.G., Scherer, P.E., Kumar, S.: Serum high molecular weight complex of adiponectin correlates better with glucose tolerance than total serum adiponectin in Indo-Asian males. Diabetologia. 48, 1084–1087 (2005). https://doi.org/10.1007/s00125-005-1758-7
49. Bobbert, T., Rochlitz, H., Wegewitz, U., Akpulat, S., Mai, K., Weickert, M.O., Möhlig, M., Pfeiffer, A.F.H., Spranger, J.: Changes of adiponectin oligomer composition by moderate weight reduction. Diabetes. 54, 2712–2719 (2005). https://doi.org/10.2337/diabetes.54.9.2712
50. Lara-Castro, C., Luo, N., Wallace, P., Klein, R.L., Garvey, W.T.: Adiponectin multimeric complexes and the metabolic syndrome trait cluster. Diabetes. 55, 249–259 (2006)
51. Schwarz, P.E.H., Towers, G.W., Fischer, S., Govindarajalu, S., Schulze, J., Bornstein, S.R., Hanefeld, M., Vasseur, F.: Hypoadiponectinemia Is Associated With Progression Toward Type 2 Diabetes and Genetic Variation in the ADIPOQ Gene Promoter. Diabetes Care. 29, 1645–1650 (2006). https://doi.org/10.2337/dc05-2123
52. Mackawy, A.M.H.: Association of the + 45T>G adiponectin gene polymorphism with insulin resistance in non-diabetic Saudi women. Gene. 530, 158–163 (2013). https://doi.org/10.1016/j.gene.2013.07.003
53. de Luis, D.A., Izaola, O., de la Fuente, B., Primo, D., Fernandez Ovalle, H., Romero, E.: rs1501299 Polymorphism in the Adiponectin Gene and Their Association with Total Adiponectin Levels, Insulin Resistance and Metabolic Syndrome in Obese Subjects. Ann Nutr Metab. 69, 226–231 (2016). https://doi.org/10.1159/000453401
54. Vasseur, F., Helbecque, N., Dina, C., Lobbens, S., Delannoy, V., Gaget, S., Boutin, P., Vaxillaire, M., Leprêtre, F., Dupont, S., Hara, K., Clément, K., Bihain, B., Kadowaki, T., Froguel, P.: Single-nucleotide polymorphism haplotypes in the both proximal promoter and exon 3 of the APM1 gene modulate adipocyte-secreted adiponectin hormone levels and contribute to the genetic risk for type 2 diabetes in French Caucasians. Hum Mol Genet. 11, 2607–2614 (2002). https://doi.org/10.1093/hmg/11.21.2607