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# Polymorphisms in the leptin gene and their role in adolescent obesity and metabolic health

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## ABSTRACT

The leptin (LEP) gene polymorphism is one of the most common polymorphisms and is believed to be associated with high circulating LEP levels that can occur in obese individuals. Thus, this study purposed to determine the relationship of LEP1 and LEP2 gene polymorphisms with overweight/obese and non-obese adolescents in Palembang and their correlation with various metabolic and anthropometric parameters. This research was an analytical and observational study. A total of 50 blood samples from adolescents (aged 10-18 years) were analyzed in this study. Standard laboratory protocols for serum lipid measurements, deoxyribonucleic acid (DNA) extraction, allele analysis by polymerase chain reaction (PCR), and LEP and insulin analysis by enzyme-linked immunosorbent assay (ELISA) were employed. The study revealed the allelic frequencies for gene LEP1: non-obese (G = 0.45, A = 0.55) and obese (G = 0.87, A = 0.13), and gene LEP2: non-obese (G = 0.54, A = 0.45) and obese (G = 0.67, A = 0.33). The frequencies of alleles in the LEP1 gene showed a statistically significant association with nutritional status ($p = 0.011$), while there was no significant association between genotypes in the LEP gene and nutritional status. The level of LEP was 6.39 + 1.12 in non-obese and 11.57 + 1.68 in obese, with p 0.000; and the triglyceride level was 82.56 ± 43.22 in non-obese and 113.72 ± 54.03 in obese, with p 0.029. In conclusion, this study shows that LEP levels and the polymorphism in the LEP1 gene are associated with obese adolescents.

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## INTRODUCTION

Obesity is becoming a worldwide problem. According to the World Obesity Atlas 2023, the current obesity rate is 38% of the population, and estimates suggest that prevalence might double to 10% to 20% between 2020 and 2035 [1]. Obesity is a significant health issue in the population, which increases the likelihood of developing numerous diseases [2]. Besides the influence of diet and physical activity, genetics is also one of the causes of obesity. Genetic risk factors can cause the incidence of obesity, which is approximately 20–25% [3]. Besides, various genes may contribute to abnormalities in food regulation, energy expenditure, and fat storage [4].

A Pakistani family with severe early obesity found two new mutations [5]. Leptin (LEP) resistance is observed in people who are obese [6]. The hypothalamic-melanocortin-LEP signaling pathway mostly comprises genes linked to obesity, which influences people

to overeat and become severely obese very quickly [7, 8]. LEP is a cytokine that maintains energy balance in the hypothalamus and increases satiety, secreted by adipocytes [6]. The secreted LEP will bind to and activate its receptor called the LEP receptor (LEP-R) [9]. This receptor location allows LEP to exert different effects, and control body weight through feedback between adipose tissue and the hypothalamus [10, 11]. Loss of feelings, excessive nutrient consumption, and overall weight gain are signs of LEP resistance. LEP resistance in some populations has a significant correlation with the incidence of obesity [9, 12, 13].

Multiple studies have demonstrated that variations in the LEP gene can impact the LEP level in the bloodstream, which is linked to obesity [14-16]. Research conducted on obese women has revealed a significant correlation between LEP polymorphisms, specifically rs7799039 and rs2167270, and the levels of LEP [17]. These genetic variations were discovered to account for a significant proportion of the variability in LEP levels [17]. Moreover, several studies have examined the variations in LEP genes and the receptors for LEP in various populations to understand their influence on obesity. The researchers additionally examined the presence of single nucleotide polymorphisms in the LEP and LEP-R genes and their correlation with LEP levels and obesity. These findings highlighted the significance of genetic differences in LEP and LEP-R in the development of obesity [16, 18]. Various ethnic groups have studied the association between LEP gene polymorphisms and obesity. For example, there was a link between the G-2548A polymorphism in the LEP gene and extreme obesity in Taiwanese Aborigines that showed no difference in the genotype frequency of the LEP-R polymorphism between the extremely obese and control groups [19]. However, there was an association between the LEP G-2548A gene polymorphism, LEP levels, and lipid profiles in obese Turkish Cypriots [20]. This suggests how ethnicity influences gene polymorphism and how genetic differences can affect metabolic biomarkers related to obesity [20].

Several studies reported the correlation of polymorphisms in the LEP gene with obesity, but there is a lack of studies explicitly targeting the Indonesian population, specifically the adolescents residing in Palembang. This study aims to investigate the association between LEP1 and LEP2 gene polymorphisms in both obese and non-obese adolescents and to examine their interaction with several metabolic and anthropometric characteristics in Palembang. Through the examination of LEP gene polymorphisms and metabolic health in this particular demography, a comprehension of the genetic inclination towards obesity in this community may be enhanced. Ultimately, this will aid in the development of more efficient and customized obesity prevention and management approaches that may take into account local variations.

## MATERIALS AND METHODS

### Ethical approval

The ethical approval was provided by the Medical and Health Research Ethics Committee, Faculty of Medicine University of Sriwijaya (No. 043-2024) and the Committee on Bioethics, Humanities and Islamic Medicine, Faculty of Medicine-Muhammadiyah University of Palembang (No. 006/EC/KBHKI/FK-UMP/I/2024).

### Participants of the study

The study was conducted between January 2024 and March 2024. The study participants were adolescents who volunteered to participate in this study, as well as senior high school students in Palembang, Indonesia. A total of 50 adolescents, comprising 30 girls and 20 boys, were used in this study. All participants and parents provided informed consent, and the inclusion criteria included individuals aged 10 to 18 who were willing to give blood samples. The exclusion criteria included experiencing or having a history of diabetes mellitus, hypothyroidism, Cushing's disease, polycystic ovary syndrome, Prader-Willy syndrome, acute inflammatory processes, treatment with any anti-obesity agent, or experiences of weight loss during the previous 6 months. Additionally, participants who had not agreed to allow necessary measurements or who had decided to withdraw from the study were excluded. Those who met the inclusion criteria were asked if they were willing to participate in the research. Furthermore, after receiving a complete and accurate explanation of the study's purpose, work, and benefits, the respondents and parents were asked to sign an informed consent sheet.

The data collected are primarily in the form of interviews (subject characteristics and disease history), anthropometric measurements (height, weight, waist, hip, and middle arm circumferences), and laboratory examinations (blood sampling). An automatic height-weight scale was used to measure heights and weights to the nearest 0.1 cm and 0.1 kg, respectively, and calculated body mass index (BMI) values by dividing weight (kg) by height squared (m2). A soft plastic measuring tape was used to measure the waist, hip, and arm circumferences to the nearest millimeter. A fat monitor analyzer (one med 872H, Jakarta, Indonesia) was used to determine the body fat percentage. The analyzer with the necessary data was programmed, which included weight, height, age, and sex. The participant then positioned their foot on the equipment's electrodes until the screen displayed the recorded results. The Centers for Disease Control and Prevention (CDC) 2000 classified adolescents based on their BMI (kg/m2), using this value to plot a curve; a curve exceeding 85 indicates overweight/obesity, while a curve below 85 indicates non-obesity. Then, researchers took blood samples from certified medical personnel for PCR, biochemical, and enzyme-linked immunosorbent assay (ELISA) analysis.

### Blood sampling

A total of 50 blood samples (6 ml) were collected from the median cubital vein of individuals and divided into two tubes, each containing 3 ml. One tube with EDTA was used as an anticoagulant for the PCR assay, and another tube was used without an anticoagulant for the biochemistry and ELISA assays. The Research Laboratory, Faculty of Medicine, Universitas Muhammadiyah Palembang immediately received the collected samples and preserved them at 4–60 °C until DNA extraction. For LEP and insulin level assayed samples, after clotting for 2 hours at room temperature, the samples were centrifuged at 1000 x g for 20 minutes and then, stored the sample at -200 °C before being transferred to the biotech laboratory at Universitas Sriwijaya for assaying. Mohammad Hosein Hospital received fresh blood samples for lipid profile and fasting glucose analysis.

### Biochemical parameters analysis

Biochemical parameter analysis was conducted, including measuring glucose metabolism using fasting glucose, fasting insulin, and the homeostasis model assessment for insulin resistance (HOMA-IR). The fasting glucose levels and lipid profile consisting of total cholesterol, triglycerides, high-density lipoproteins (HDL), and low-density lipoprotein (LDL) from the participant's fresh blood sample were analyzed using an automated molecular diagnostics analyzer (Alinity, Abbot Germany).

### Determination of LEP

The LEP and insulin parameters were analyzed using ELISA. The LEP level was measured using the immunoturbidimetric method and the ELISA kit (Biotinylated and Avidin-Horseradish Peroxidase-HRP) 96/T from ELKbio, United States. The optical density (OD) was measured spectrophotometrically at a wavelength of 450 nm to 10 nm. Also, the insulin levels were measured using the immunoturbidimetric method and an ELISA kit (Biotinylated for Human and Avidin Horseradish-HRP) 96/T from Elabscience, United States. Similarly, the OD was measured spectrophotometrically at a wavelength ranging from 450 nm to 2 nm. The ELISA kit used the sandwich ELISA principle, using an iMark microplate reader system (BIO-RAD, California, USA) that automatically calculates.

### DNA extraction and PCR amplification of LEP gene

DNA extraction was performed using TIAamp Genomic DNA Kit (TIAGEN Biotech (Beijing) Co., Ltd.), and the LEP gene in 50 blood samples was amplified, following the manufacturer's instructions. A 25 µl reaction volume with 4 µl of DNA, 10µl of master mix (2x Taq Plus PCR Mix (with dye) TIAGEN Biotech (Beijing) Co., Ltd), 10µl of ddH$_2$O, and 1 µl of primer Standard-Oligo Synthesis, and 0.05 µmole of MOPC (Macrogen Oligonucleotide Purification Cartridge) were used for the PCR amplification. The LEP gene was detected using the PCR amplification method. A PCR reaction was conducted under conventional PCR conditions to identify the LEP gene. The reaction consisted of an initial denaturation step followed by 35 cycles, each consisting of denaturation, annealing, and final extension. The details of these stages are found in Tables 1 and 2. Following the amplification of the polymorphic fragment using PCR, the subsequent step involved determining the genotype.

**Table 1**. Primers used in the present study.

| Gene identified | Primer sequence | References | Product length (bp) |
|---|---|---|---|
| LEP1 | 5′-GCCAGAGCAGAAAGCAAA-3′ | [18] | 397 |
| | 5′-TCAGGAGGCGTTCAATAA-3′ | [18] | |
| LEP2 | 5-GAGCACTTGTTCTCCCTCTT-3′ | [20] | 435 |
| | 5′-TTCCCTTAACGTAGTCCTTG-3′ | [20] | |

bp: base pair; LEP: leptin

**Table 2**. Conditions of PCR for LEP1 and LEP2.

| Steps | Temperature and duration | Cycle |
|---|---|---|
| Initial denaturation | 95$^o$C for 5 minutes | 1 cycle |
| Denaturation | 94$^o$C for 15 seconds | |
| Annealing | 51$^o$C for 30 seconds | 35 cycles |
| Extension | 72$^o$C for 30 seconds | |
| Final Extension | 72$^o$C for 5 minutes | 1 cycle |
| Holding | 4$^o$C ∞ until use | - |

By adjusting the threshold during the geometric phase of amplification, a cycle time (Ct) value was collected, and it was then scored in relation to the delta cycle time (DCt) produced between the matched and mismatched primer pairs.

### Agarose gel electrophoresis and gel documentation

After performing PCR, agarose gel electrophoresis and gel documentation were used to confirm the amplification of the LEP gene sequence fragment. A volume of 25 µl of each PCR product for gel electrophoresis in a 2% agarose gel, stained with 5 µl of ethidium bromide. The amplified PCR product was run for 90 minutes using a Wide Mini-Sub Cell GT Horizontal Electrophoresis System and PowerPac Basic Power Supply BIO-RAD. DNA fragments were visualized using a gel documentation machine (GelDoc Go Gel Imaging System, BIO-RAD, California, USA), which emitted UV light after the dye line had approximately run through the gel. LEP1 genotypes were G/G: 416 bp, G/A: 416, 363, 242 bp, and A/A: 416, 242 bp. LEP2 genotypes were G/G: 229 bp, G/A: 229, 187, 61 bp, and A/A: 187, 61 bp.

### Statistical analysis

The statistical analysis was done by using SPSS, v. 24.0 (IBM Corporation, Armonk, New York). The means and standard deviation (SD) of continuous variables such as age, weight, height, body mass index, waist circumferences, hip circumferences, arm circumferences, systolic pressure, diastolic pressure, body fat, fasting blood glucose, serum level of LEP, and insulin were used. The genotype distribution for deviation from Hardy-Weinberg equilibrium using chi-squared was evaluated. The LEP gene variant was in Hardy-Weinberg equilibrium. All parameters for normality using the Kolmogorov-Smirnov test were analyzed. The necessary statistical tests, namely the T-test and Chi-square were utilized. A p-value below 0.05 signifies a significant difference between the groups, allowing for rejecting the null hypothesis.

## RESULTS

### Demographic, and anthropometric characteristics of the participants

Based on the results of the study, it was found that body weight, body mass index, waist, hip, and arm circumferences, systolic and diastolic pressure, and body fat (except age and height) showed a significant difference ($p < 0.05$) between non-obese and obese individuals (Table 3).

**Table 3**. Demographic and anthropometric characteristics of the study participants.

| Variables | Non-Obese (Mean ± SD) | Obese (Mean ± SD) | $p^{\dagger}$ |
|---|---|---|---|
| Age (years) | 19.65 ± 1.23 | 19.88 ± 1.15 | 0.535 |
| Weight (kg) | 75.71 ± 8.69 | 99.09 ± 1.05 | 0.000* |
| Height (cm) | 169.97 ± 5.91 | 171.50 ± 7.04 | 0.426 |
| Body mass index (kg/m$^2$) | 26.57 ± 1.96 | 34.34 ± 3.81 | 0.000* |
| Waist circumferences (cm) | 89.26 ± 7.52 | 103.81 ± 10.40 | 0.000* |
| Hip circumferences (cm) | 103.88 ± 5.65 | 113.38 ± 7.92 | 0.000* |
| Arm circumferences (cm) | 30.24 ± 3.23 | 35.00 ± 3.29 | 0.000* |
| Systolic pressure (mmHg) | 118.82 ± 9.78 | 130.63 ± 12.89 | 0.001* |
| Diastolic pressure (mmHg) | 75.88 ± 6.57 | 81.25 ± 5.00 | 0.006* |
| Body fat (%) | 26.79 ± 4.37 | 32.83 ± 4.57 | 0.000* |

*Reached statistical significance ($p < 0.05$); $^{\dagger}$Independent T-Test; SD: standard deviation

### Biochemical characteristics of the study participants

Based on the results of the study, it was found that triglycerides and serum levels of LEP showed a significant difference (p <0.05) between non-obese and obese individuals (Table 4).

**Table 4**. Biochemical characteristics of the study participants.

| Variables | Non-Obese (Mean ± SD) | Obese (Mean ± SD) | p† |
|---|---|---|---|
| Total cholesterol (mg/dl) | 155.13 ± 37.92 | 161.15 ± 32.56 | 0.565 |
| LDL (mg/dl) | 105.60 ± 36.30 | 110.60 ± 31.63 | 0.606 |
| HDL (mg/dl) | 49.32 ± 7.80 | 46.80 ± 7.97 | 0.264 |
| Triglycerides (mg/dl) | 82.56 ± 43.22 | 113.72 ± 54.03 | 0.029* |
| Fasting blood glucose (mg/dl) | 67.60 ± 5.59 | 75.08 ± 21.70 | 0.102 |
| Serum level of leptin (ng/mL) | 6.39 ± 1.12 | 11.57 ± 1.68 | 0.000* |
| Serum level of insulin (µIU/ml) | 7.91 ± 6.96 | 10.31 ± 7.78 | 0.278 |
| HOMA-IR | 1.33 ± 1.33 | 2.02 ± 1.41 | 0.081 |

*Reached statistical significance (p < 0.05); †Independent T-Test; LDL: low-density lipoprotein; HDL: high-density of lipoproteins; HOMA-IR: homeostasis model assessment for insulin resistance

### Relationship of polymorphisms in the LEP1 and LEP2 gene in nutritional status

As shown in Table 5, there was a significant relationship between LEP1 gene polymorphism and allele frequencies in obese individuals where the p value= 0.011 with RR = 0.123, and IC 95% = 0.027-0.557.

As shown in Table 6, there was no relationship between polymorphism of the LEP genes with genotype frequencies in obese individuals.

**Table 5**. Allele frequencies of the LEP1 and LEP2 gene polymorphism.

| Gene | Allele | Non-Obese | | Obese | | p† | RR (95% CI) |
|---|---|---|---|---|---|---|---|
| | | n | % | n | % | | |
| LEP1 | G | 5 | 12.8 | 34 | 87.2 | 0.011* | 0.123 |
| | A | 6 | 54.5 | 5 | 45.5 | | (0.027-0.557) |
| LEP2 | G | 6 | 18.8 | 26 | 81.3 | 0.701 | 0.600 |
| | A | 5 | 27.8 | 13 | 72.8 | | (0.154-2.340) |

*Reached statistical significance (p < 0.05); †Chi-square; LEP: leptin; RR: relative risk; CI: confidence interval

**Table 6**. Genotype frequencies of the LEP1 and LEP2 gene polymorphism.

| Gene | Allele | Non-Obese | | Obese | | p† |
|---|---|---|---|---|---|---|
| | | N | % | n | % | |
| LEP1 | G/G | 2 | 12.5 | 14 | 87.5 | 0.320 |
| | G/A | 7 | 31.8 | 15 | 68.2 | |
| | A/A | 2 | 16.7 | 10 | 83.3 | |
| LEP2 | G/G | 3 | 50.0 | 3 | 50 | 0.210 |
| | G/A | 5 | 17.9 | 23 | 82.1 | |
| | A/A | 3 | 18.8 | 13 | 81.3 | |

*Reached statistical significance (p < 0.05); †Chi-square

Based on genotype frequency (G/G; G/A; A/A), no relationship was obtained between LEP1 and LEP2 gene polymorphisms and nutritional status (Figures 1-3). However, the LEP1 gene was found to have the highest percentage in the G/G allele (87.5%) for obese status (Table 6). The LEP2 gene showed the largest percentage of the G/A allele (82.1%) in individuals with obese nutritional status (Table 6).

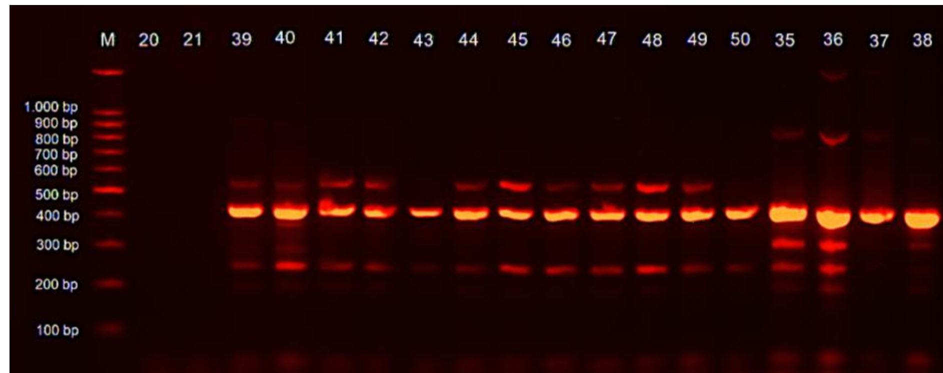


**Figure 1**. PCR products of LEP1 gene on 2% agarose gel; genotypes were G/G: 414 bp, G/A: 414, 362, 242 bp, and A/A: 362, 242 bp. Line M: Ladder, Line 20, 21: Non-digest PCR product, Line 39-50: A/A, Line 35-36: G/A, Line 37-38: G/G. Respondents no. 20, 21, 35 and 36 are classified as non-obese, while respondents no. 37-50 are classified as obese.

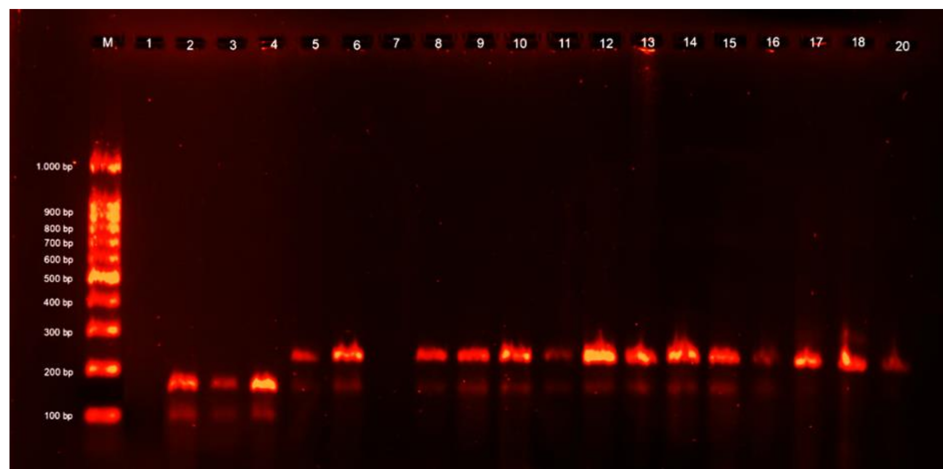


**Figure 2**. PCR products of LEP2 gene on 2% agarose gel; genotypes were G/G: 229 bp, G/A: 229, 187, 61 bp, and A/A: 187, 61 bp. Line M: Ladder, Line 1, 7: Non-digest PCR product, Line 2-4: A/A, Line 5-6, 8-16: G/A, Line 17-20: G/G. Respondents no. 1, 7, 8, 9, 11, 15-18 are classified as non-obese, while respondents no. 2-6, 10, 12-14, 20 are classified as obese.

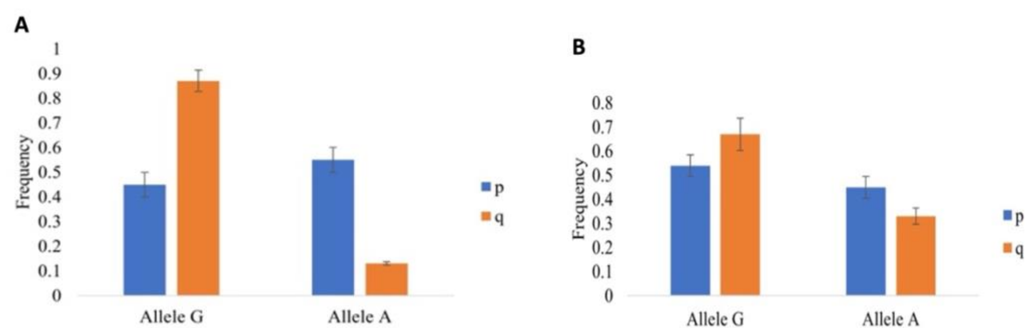


**Figure 3**. Allelic frequencies for gene LEP1 and LEP2; Hardy–Weinberg Equilibrium. p: dominant allele; q: recessive allele. The population of non-obese individuals has more alleles G than the obese population, which tends to have more alleles A for the variation in the LEP1 and LEP2 genes. (A) Allele G: p=0.54, with a small margin of error. q=0.67, also with a slightly larger error bar than p. This suggests that in the q setting, allele G is more prevalent; allele A: p=0.45, slightly lower compared to q=0.33. Both the p and q frequency error bars are small and similar. In general, Allele G is more prevalent than allele A in both situations. This can point to a pattern in the population under study, requiring more research into the environmental or biological variables affecting these frequencies. (B) Allele G: p=0.54, with a small margin of error. q=0.67, also with a slightly larger error bar than p. Allele G is more frequent in the q context. Allele A: p=0.45, slightly lower compared to q=0.33. The error bars for both p and q frequencies are modest and comparable. Overall, allele G has higher frequencies in both contexts compared to allele A. This could suggest a trend in the population being studied, warranting further investigation into the biological or environmental factors influencing these frequencies.

## DISCUSSION

Obesity has become a significant public health concern, particularly among adolescents [21]. This study found biochemical differences between non-obese and obese adolescent individuals and the role of LEP gene polymorphisms. Significant differences were found in parameters such as weight, BMI, waist, hip, and arm circumferences, diastolic pressure, body fat percentage, triglycerides, and serum LEP levels between the non-obese and obese individuals. These parameters are crucial indicators of obesity-related health risks and metabolic syndrome. Obesity-related metabolic syndrome is characterized by the presence of visceral adipose tissue that is metabolically active and has a higher macrophage count [22].

Recent studies have explored the complex interplay between these factors, where there is a prevalence of hypertriglyceridemic waist, a phenotype characterized by elevated waist circumference and triglyceride levels, observed in school children [23]. This condition is associated with an increased risk of cardiovascular disease and metabolic abnormalities, underscoring the importance of understanding its underlying factors [23].

Adolescent obesity is a significant health concern in Palembang, Indonesia, with genetic factors playing a role in this complex issue. Obesity has been associated with polymorphisms in genes related to LEP, a hormone that regulates energy balance and body weight [24-26]. Specifically, variants in the LEP gene and LEP-R gene have been linked to increased plasma LEP levels and BMI in different populations [24, 26, 27]. These genetic variations can influence the levels of adipokines like LEP and adiponectin, altering the predisposition to obesity in children and adolescents [26].

The LEP gene, responsible for the production of the LEP hormone, has been implicated in the regulation of body weight and fat distribution [3]. LEP is secreted by adipose tissue and acts on the hypothalamus to suppress appetite and increase energy expenditure, making it a key player in energy homeostasis [6]. Alterations in LEP signaling, whether due to LEP resistance or genetic variations in the LEP gene, have been linked to obesity and related metabolic disorders [4,6]. LEP secretion is directly linked to an individual's body mass and nutritional state [6]. LEP secretion is regulated by factors such as food consumption, total body fat, and several hormones like insulin, glucagon, and pancreatic polypeptide. Insulin was observed to boost the production of LEP through glucose metabolism, as indicated by *in vitro* studies [8-9]. Changes in glucose metabolism due to implementing a high-fat diet for 24 hours cause a decrease in LEP levels, thereby encouraging weight gain and obesity [12]. Studies have shown that LEP levels are associated with insulin resistance, a common feature of obesity, in adolescents [28].

Furthermore, studies have found a positive correlation between LEP concentrations, BMI, and weight in children and adolescents [29]. Moreover, researchers have discovered LEP as a potential indicator for metabolic syndrome and insulin resistance in obese adolescents [30]. Furthermore, studies have linked LEP levels to cardiovascular risk factors and subclinical cardiac dysfunction in obese adolescents [31-32].

Adiponectin levels are inversely related to LEP levels, and the LEP-to-adiponectin ratio has been linked to weight gain in non-obese children over time [33-34]. Also, the concentrations of LEP are higher in obese children and teens compared to those who are a healthy weight [35]. It is interesting to note that LEP levels can predict future weight gain in obese teens, which highlights the role of this hormone in the development of obesity [36].

Analysis of the LEP gene GG genotype results indicates no statistically significant correlation between the existence of the LEP gene GG allele and a lower risk of obesity. The LEP-2548 GG genotype and obesity have not shown a significant correlation in the population, indicating possible modulation through additional genetic, environmental, or demographic factors [14]. This highlights the variability and complexity of genetic influences on obesity across different demographic groups. Some studies have found a significant association of the GG homozygote genotype with obesity in the American population, suggesting potential ethnic or geographic variances in the gene's obesity link [37]. The genetic determinants of obesity have been a subject of interest, with studies highlighting the role of gene pleiotropy, adaptations to diverse environments, and the impact of genetic drift on allele frequencies across generations [38-39].

This research showed that the LEP2 gene has the largest percentage of the G/A allele. This finding aligns with previous literature that has reported a higher prevalence of the G/A genotype in overweight and obese patients [40]. Research has shown that genetic variations in genes such as beta-2 adrenoceptor and uncoupling proteins are associated with altered adipocyte function and obesity susceptibility [40-41]. Additionally, studies have emphasized the importance of familial predisposition to obesity and its impact on childhood obesity and related metabolic complications [42]. Furthermore, the interaction between genome and nutrient intake has been explored, indicating that genetic factors play a significant role in determining an individual's predisposition to obesity [43]. Although lifestyle factors such as diet and physical activity also contribute to obesity, genetic predisposition has been recognized as a critical determinant of an individual's vulnerability to becoming obese [44-46].

Moreover, studies have demonstrated that physical fitness can attenuate the genetic predisposition to obesity in children and adolescents, highlighting the importance of lifestyle interventions in mitigating genetic risks [37]. However, the lack of significant differences in many characteristics based on genotype within each group suggests that genetic factors alone may not fully explain the variations in these traits [4]. Environmental factors, lifestyle choices, and interactions between multiple genes likely contribute to the observed differences [2-4]. Factors such as food consumption, total body fat, and hormones like insulin, glucagon, and pancreatic polypeptide regulate LEP secretion. Insulin was discovered to boost the production of LEP through glucose metabolism, according to an in vitro study [8-9]. The implementation of a high-fat diet over 24 hours leads to alterations in glucose metabolism, resulting in a reduction in LEP levels [12]. This reduction in LEP levels promotes weight gain and the development of obesity [2-3].

However, the limitations found in this study include that a) this study did not examine inter-individual variations in nutritional intake that might inhibit gene expression (nutrigenomics), and b) besides, this variation in gender and combining overweight and obese groups in one group may also influence the research results.

## CONCLUSIONS

LEP levels and LEP1 gene polymorphisms are significantly associated with overweight and obesity among the adolescents studied. These significant results can be found in the relationship between LEP1 gene polymorphism and nutritional status ($p = 0.011$). The study underscores the importance of genetic factors, particularly LEP1 gene polymorphisms, in influencing obesity and metabolic health in adolescents. These findings can contribute to more effective obesity prevention and management strategies tailored to the local context in Palembang. Additional investigation is required to

examine the fundamental mechanisms and potential therapies that focus on these genetic variations.

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## AUTHOR CONTRIBUTIONS

RAT, I, YK, and IAL planned, designed the study, and analyzed the data. MDH, NMEM, WA, LC, TP, IR, WA, and DL participated in the collection of the samples and performed tests. RAT, MDH, and NMEM were involved in drafting and revising the manuscript. RAT, MDH, NMEM, and WA performed data and critical review of the article. All authors read and approved of the final manuscript.

## CONFLICTS OF INTEREST

There is no conflict of interest among the authors.

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