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Research Article:

Evaluation of Immunoinflammatory and Metabolic Markers in Obesity-Related Disorders

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Abstract

Background: Obesity induces alterations in the immune system, liver function tests, inflammation, glucose metabolism, and insulin resistance, all of which may culminate in diabetes.

Aim: The study aimed to identify immune-inflammatory markers and differences insulin resistance in the obese and normal participants.

Methodology: The results of a laboratory study comparing the obese individuals with the control participants were analyzed in this quantitative cross-sectional study. The body mass index (BMI) of thirty-two participants (18.2-25) was chosen as the controls. Sixty-eight participants whose BMI exceeded 30 kg/m² were assigned in the " obese" (case) participants. Markers of inflammation and immunity, such as C-reactive protein (CRP), white blood cell (WBC) count, and lymphocytes, were analyzed. Enzyme-linked immunosorbent assay (ELISA) tests for Interleukin -1 beta (IL-1β) and interleukin-10 (IL-10) were used as markers for pro-inflammatory and anti-inflammatory response. Hemoglobin A1c (HbA-1c), glucose, insulin, and adiponectin tests for diabetes and insulin resistance. Thyroid-stimulating hormone (TSH), liver function tests such as aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were used.

Results: The result showed that the obese participants had higher levels of body weight, fat percentage, CRP, WBC, and lymphocytes compared to the control participants. Also, IL-1 β levels was higher, and IL-10 levels was lower in the obese participants compared to the control participants. Adiponectin, insulin, blood glucose, HbA-1c%, were higher in the obese participants compared to the control participants. In addition, TSH, AST and ALT were higher in the obese participants compared to the control participants.

Conclusion: Our research determined that immune-inflammatory markers, obesity and liver function tests were higher in the obese participants, which leads to the development of insulin resistance and type-2 diabetes mellitus.

Keywords: *Obesity, immune-inflammatory markers, metabolic markers.*

1. Introduction

Obesity has grown to be a significant health issue around the world. It is therefore one of the main causes of poor health and reduced quality of life around the world. The prevalence of participants globally living with obesity has almost tripled from 1975 to 2021 and in 2022, it is estimated that 1.9 billion adults experienced being overweight and 650 million of them were identified as the obese, WHO (2023) reported. These figures show that obesity is becoming a more serious threat to public health. New scientific research has introduced meta-inflammation which refers to chronic low-grade inflammation, as a main trait of obesity [1]. Such inflammation is not a random part of the disease but an essential part of what happens. Meta-inflammation is not an acute inflammatory response to an injury or infection; instead, it comes from problems within the fat tissue and sticks around. Adipose tissue was previously regarded

as only a place to store energy, but it is known that it has several bioactive substances called adipokines [2]. Once adipocytes increase in weight, they start to release larger amounts of the pro-inflammatory cytokines, tumor necrosis alpha (TNF-α) and IL-6, in the obese individuals. Cytokines initiate a sequence of immune actions such as summoning macrophages that help increase local inflammation in fat deposits [3]. This constant inflammation upsets insulin's normal function, leading to generalized resistance to insulin in the body, one of the main symptoms of metabolic syndrome including diabetes. In addition, obesity brings about important changes in the cells found in adipose tissue. In addition to adipocytes, the tissue is invaded by immune cells such as macrophages, which help create more inflammation. When immune cells are reprogrammed, they disrupt the normal way adipose tissue functions and the effects of this inflammation are also seen in the liver, muscles and blood vessels, making obesity-related inflammation systemic [4, 5]. This study aimed to investigate inflammation markers, obesity-related disease markers that lead to development of diabetes.

2. Materials and Methods

2.1. Study Design and Materials

This study employed a quantitative cross-sectional design comparing obesity participants to the control participant. Participating was determined by BMI; participants with a BMI above 30 kg/m² were categorized into the obese (case) participants, while those with a BMI ranging from 18.2 to 25 were assigned to the control participants. A total of 68 cases were selected from individuals with obesity, while thirty-two controls were selected from those with a normal body weight (BMI 18.2-25). This shows that the control sample is a good representation of the healthy population and lowers the chance of sampling effect or estimate bias.

For our study, male and female subjects were assessed separately, and statistical analyses were conducted accordingly. The serum samples were collected from the obese and the control participants; the study participants were recruited based on the controlled factors such as sex and age (20-60 years); the mean was 40 years. Participants with smoking history, recent infection, autoimmune disease, cancer patients, current medication use and chronic systematic disease were excluded. In addition, every participant provided written consent.

2.2. Ethics of the Study

This study adhered to the ethical principles outlined in the Declaration of Helsinki (2013). Approval for the longitudinal study was granted by the scientific and ethics committee of the College of Science, University of Raparin (Approval No. 25, dated September 7, 2024).

2.3. Methods

BMI was calculated as weight (kg) divided by the square of height (m²). Measurements acquired for all subjects comprised BMI, body fat percentage (BF%), and lean body mass. Prior to the examination, the participants fasted for 12 hours. After blood collection, the following markers were measured: serum insulin, fasting blood sugar (FBS), HbA1c, ALT, AST, and adiponectin. Serum IL-1β and IL-10 concentrations were analyzed by ELISA using commercially available kits (ELK Biotechnology Co., Ltd., China, Cat. No. ELK1227 for IL-1β and ELK1142 for IL-10) following the manufactures' instructions, the sandwich ELISA method was used to measure the amount of adiponectin in accordance with the manufacturer's instructions. Fasting serum insulin was conducted by electrochemiluminescence immunoassay (ECLIA) on a Roche Cobas e 411 analyzer using the manufacturer's reagents and calibration protocols. Routine biochemical assays including ALT and AST were assessed on the Roche Cobas C111 analyzer using the IFCC-recommended optimized kinetic methods; while fasting blood glucose was performed enzymatically via the hexokinase method on the same platform. Glycated hemoglobin (HbA1c) was performed in whole blood using the Roche Cobas C111 immunoassay standardized to IFCC/DCCT criteria to assure comparability. CRP concentrations were evaluated by particle-enhanced immunoturbidimetric testing on the Roche Cobas C111 analyzer. For all tests, internal and external quality the controls and calibrators were conducted according to laboratory policy; laboratory workers were blinded to clinical participants assignments and data failing quality-the control criteria were re-analyzed.

2.4. Statistical Analysis

Statistical analysis was performed using GraphPad Prism (GraphPad Software, San Diego, California, USA). Data were expressed as mean ± standard error mean (SEM). Normality of data distribution was assessed using the Shapiro–Wilk test. For comparisons between two independent groups, the unpaired Student's t-test was applied. For comparisons involving more than two groups (four groups: males and females), one-way analysis of variance (ANOVA) was used. When ANOVA showed significant differences, appropriate post hoc multiple comparison tests (e.g., Tukey's test) were performed. Parametric tests were applied because the data were normally distributed. A p-value < 0.05 was considered statistically significant.

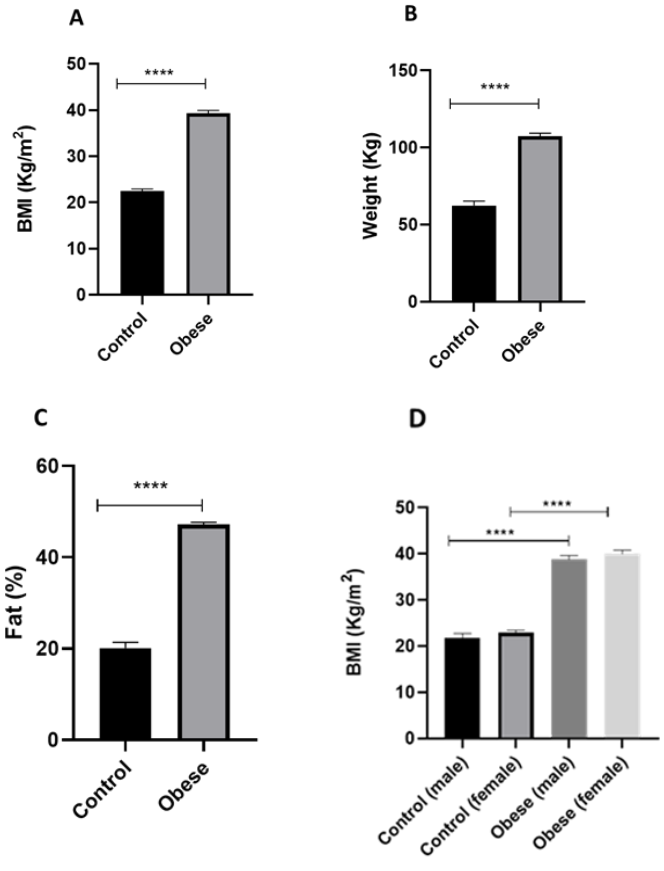
3. Result and Discussion

3.1. Obesity Investigation Markers

To indicate the obese participants, BMI, body weight and fat percentage were investigated. Theses markers are linked with obesity features, in the obese participants (regardless of gender) body mass index (BMI) levels had higher (39.37±0.57 kg/m^2) levels compared to the control (22.44±0.51 kg/m^2) (p value= <0.0001) (figure 1, panel-A).

In addition, BMI was measured in male and the female obese participants (gender based), the result showed that BMI levels was higher in male participants (38.44±0.82) compared to the male control participants (21.44±0.93 kg/m2) (p value= 0.0007) and the female obese participants (40.02±0.78 kg/m^2) had higher levels compared with the female (22.90±0.58 kg/m^2) control participants (p value <0.0001) (figure 1, panel-D).

After that, body weigh was measured, the result showed that the obese participants (regardless of gender) had higher body weight (107.4±1.99 Kg) compared to the control participants (62.39±2.91 Kg) (P value<0.0001) (figure 1, panel-B). To investigate the differences between male and females, weight was measured (gender based), the result showed that body weigh was higher in the male obese participants (115.1±2.36) compared to the male control participants (67.18±3.7 Kg) (p value <0.0001) and the female obese participants (98.76±2.61 Kg) had higher weight levels compared with the female (54.86±3.07 Kg) control participants (p value <0.0001) (figure 1, panel-E). To investigate obesity markers, fat percentage was measured, our result showed higher fat percentage in the obese participants compared to the control participants (regardless of gender) (20.11±1.25) (figure 1, panel-C) (p value <0.0001). Moreover, fat percentage was measured in the male and female obese participants (gender based), our result indicated that fat percentage was higher in male (46.31±0.70 %) compared to the male control participants (20.71±1.28 %) (p value <0.0001) and the female obese participants (48.02±0.70 %) had higher fat percentage compared to the female (15.50±1.34 %) control participants (p value <0.0001) (figure 1, panel-F).



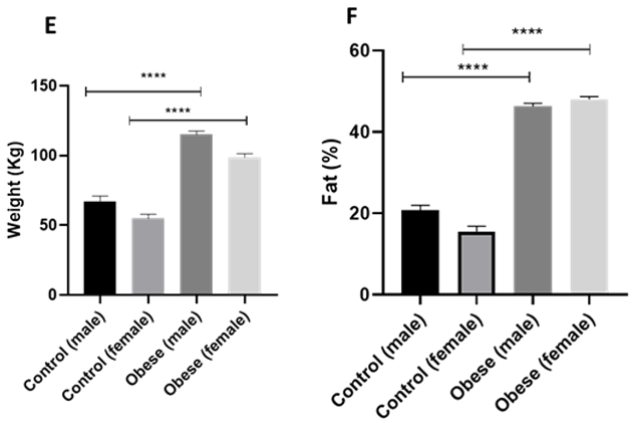


Figure 1: The impact of obesity on BMI, body weight, and fat percentage. Panel A: A comparison of BMI between the obese and control groups (regardless of gender). Panel B: A comparison of body weight between obese and control groups (regardless of gender). Panel C: A comparison of the percentage of fat in obese and control groups (regardless of gender). Panel D: A comparison of BMI between male and female participants (gender based). Panel E: A comparison of the body weight of male and female participants (gender based). Panel F: Comparison of fat percentage between male and female participants (gender based). Results are presented as averages ±SEM from duplicate determinations. ****p < 0.0001 (adjusted p values).

Individuals with obesity, irrespective of age, had elevated BMI levels compared to the control participants. Moreover, individuals with obesity exhibited elevated body weighs in comparison to the control participants. Moreover, fat percentages were elevated in the obese individuals compared to the control participants. This coincides with earlier studies, which demonstrates that percentage of fat, fat mass, body weight, and BMI are correlated with obesity associated with type 2 diabetes[6-8].

3.2. Inflammatory Markers in the Obese Participants

Our results showed that there was significantly higher of CRP levels in the obese participants (5.86±0.60 mg/L) (regardless of gender) compared to the control participants (1.23±0.26 mg/L) (p value < 0.0001). (figure 2, panel-A). To determine the differences between males and females, CRP levels was measured (gender based), our result showed a tendency to be higher CRP levels in the male obese participants (4.76±0.65 mg/L) compared to the male control participants (0.64±0.18 mg/L) (p value =0.05) and CRP levels in the female obese participants (8.87 ±1.057 mg/L) were higher compared to the female (0.81±0.28 mg/L) control participants (p value = 0.0004) (figure 2, panel-D). Notably, females had higher CRP levels (8.87 ±1.057) compared to the male obese participants (4.76±0.65 mg/L) (p value=0.002)) (figure 2, panel-D). In addition, there was a significant lower of WBC% in the control samples (7.04±0.41 %) compared to the obese participants (9.61±0.31 %) (p value <0.0001) (figure 2, panel-B). To investigate males and female's differences in WBC%, the result showed that the male obese participants (9.571±0.53 %) had higher WBC% compared to the male control participants (6.78±0.54 %) (p value=0.0074). However, WBC% in the female obese participants (9.66±0.30 %) were not significantly different compared to the female (54.86±3.07 %) control participants (p value =0.050) (figure 2, panel-E). To

further investigation of inflammation markers, lymphocytes were measured. Our result showed that lymphocyte levels were higher in participants who had obesity (37.97±2.03) compared to the control (32.25±1.97) (p value =0.550). (figure 2, panel-C). However, there was no significant differences in males and female's lymphocyte percentage (gender based) (p value =0.271) (figure 2, panel-F).

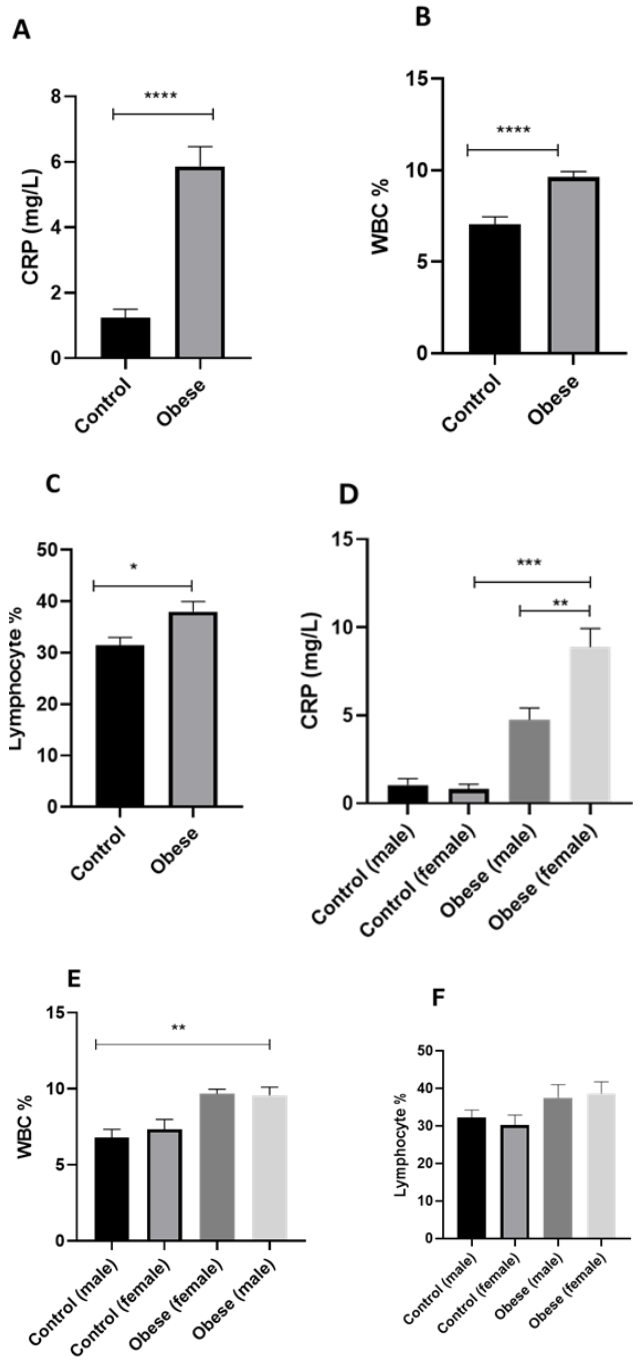


Figure 2: Effect of obesity on inflammatory markers (CRP, WBC% and Lymphocytes%). Panel A: A comparison of CRP level between the obese and control groups (regardless of gender). Panel B: A comparison of WBC percentage level between obese and control groups (regardless of gender). Panel C: A comparison of the percentage of lymphocyte in obese and control groups (regardless of gender). Panel D: A comparison of CRP level between male and female participants (gender based). Panel E: A comparison of the WBC percentage of male and female participants (gender based). Panel F: Comparison of lymphocyte percentage between male and female participants (gender based). Results are presented as Mean±SEM from duplicate determinations. *p < 0.05, **p < 0.01, ***p < 0.001 ****p < 0.0001 (adjusted p values).

Rumińska et *al.* (2019)[9] reported that CRP levels and WBC percentages were elevated in the obese individuals and diminished during weight reduction. This agrees with our findings, that demonstrated high levels of CRP, WBC and percentage of lymphocytes in the obese individuals compared to the control participants. It was demonstrated that the obese participants had higher levels of inflammatory markers, including CRP, WBC count, neutrophils, and lymphocytes compared to the controls[10, 11].

Females frequently demonstrate elevated baseline CRP levels compared to males, indicative of sex-specific variations in adiposity and hormonal profiles [12]. Females possess a naturally elevated percentage of body fat, with white adipose tissue recognized as an endocrine organ that synthesizes pro-inflammatory cytokines, which subsequently drive hepatic CRP generation in the liver. Adiposity and elevated CRP levels exhibit a significantly stronger correlation in women than to men [12]. Moreover, both endogenous estrogens directly upregulate production of CRP [13]. Therefore, it is important to consider these intrinsic physiological sex differences when interpreting changes in CRP.

3.3. Pro-Inflammatory and Anti-Inflammatory Features in Obesity

To investigate the pro-inflammatory cytokine in the obese participants serum, estimation of IL-1β was conducted. Our result showed that there were higher levels of IL-1β in the obese participants (77.94±7.26 pg/mL) compared to the control participants (40.02±2.31 pg/mL) (p value=0.0002), (regardless of gender) (figure 3, panel-A). To determine the differences between males and females, IL-1β levels was measured (gender based), our result had higher IL-1β levels in the male obese participants (94.91±10.36 pg/mL) compared to the male control participants (40.02±2.3 pg/mL) (p value=0.03) (figure 3, panel-C). However, IL-1β levels in the female obese participants (62.36±3.31 pg/mL) were a tendency to be higher compared to the female control participants (42.00±0.5 pg/mL) (p value =0.98) (figure 3, panel-C). To determine the anti-inflammatory feature of macrophages in the obese participants, IL-10 levels was investigated. Our result showed that there was a lower of IL-10 levels in the obese participants (12.87±1.28 pg/mL) compared to the control participants (9.62±0.40 pg/mL) (regardless of gender) (p value=0.013) (figure 3, panel-B). Noteworthy, no significant differences were detected between males and females when compared with the control participants (gender based) (figure 3, panel-D).

Our results indicated that the obese participants showed high levels of IL-1β (which is a feature of pro-inflammatory cytokine) compared to the control participants. Obese individuals possess a high amount of adipose tissue, which leads to an increase in the NOD-like receptor family pyrin domain containing 3 (NLRP3) inflammasome, resulting in the release of IL-1β. Nevertheless, lean individuals, regarded as the control participants, exhibit lower levels of IL-1 β compared to the control participants [14].

A sexual dimorphism marked by increased circulating

Interleukin-1 beta (IL-1β) levels in males is shaped by essential biological organizational levels linked to neuroendocrine and mRNA expression molecule that direct systemic immunometabolic interactions. The male innate immune system is inherently predisposed to reactivity in the absence of the robust immunosuppressive effects of estrogens, which actively inhibit pro-inflammatory signaling and diminish adipocyte-mediated metaflammation. At the cellular levels, male macrophages and microglia show more basal activation of IL-1β transactivation when the body is under stress from immune or metabolic factors. This primed transcriptome state elucidates the significant IL-1β expression in males, a revelation that integrates into a molecular framework marked by changed androgen profiles that promote systemic inflammation [15].

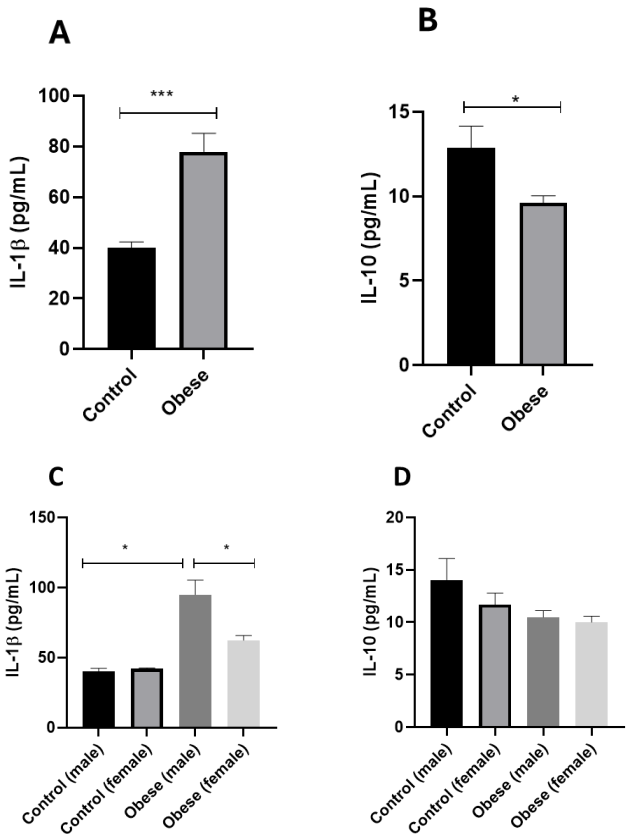


Figure 3: IL-1β and IL-10 levels in the obese participants compared with the control participants. Panel A: A comparison of IL-1β level between the obese and control groups (regardless of gender). Panel B: A comparison of IL-10 between obese and control groups (regardless of gender). Panel C: A comparison of the IL-1β level in obese and control groups (gender based). Panel D: A comparison of the IL-10 level of male and female participants (gender based). Results are presented as Mean±SEM from duplicate determinations. *p < 0.05, ***p < 0.001 (adjusted p values).

Interleukin 10, considered as anti-inflammatory cytokine, was lower in obesity status and insulin resistance condition [16]. Our results indicated that IL-10 levels were lower in the obese individuals compared to the control participants. In patients with obesity, adipose tissue caused the polarization of macrophages from pro-inflammatory to anti-inflammatory states. IL-10 in muscle tissue alleviates insulin resistance induced by a high-fat diet by diminishing macrophage infiltration and the synthesis of proinflammatory cytokines [16].

3.4. Liver Function Tests in Obesity

The increase of liver function tests is a feature of obesity. Our result showed that there was a significant increase in liver function tests, including AST, and ALT levels, in the serum of the obese participants compared to the control participants (figure 4, panel A and B). AST levels were significantly higher in the serum of the obese participants (22.93±0.89), compared to the control participants (17.60±0.91), (p value =0.0015) (figure 4, panel-A). To determine males and female's differences in AST levels, there were not significant differences in male the obese participants (24.21±1.23 U/L) compared to the male control participants (21.35±1.59 U/L) (p value=0.478) (figure 4, panel-C). In addition, AST levels in the female obese participants (17.27±0.73 U/L) were not significantly different compared to the female control participants (15.58±1.074 U/L) (p value =0.912) (figure 4, panel-C). Surprisingly, females had lower AST levels (17.27 ±0.73 U/L) compared to the male obese participants (24.21±1.23 U/L) (p value=0.002) (figure 4, panel-C). To further investigation of liver function, ALT levels was measured. Our result showed that ALT levels were higher in the obese participants (33.11±1.23), compared to the control participants (15.50±2.26 U/L) (p value <0.0001) (figure 4, panel-B). ALT levels in the male obese participants were higher (37.99±2.43 U/L) compared to the male control participants (24.64±3.97 U/L). However, the female obese participants (19.41±1.45 U/L) were not significantly differences compared to the female control participants (24.64±3.97 U/L) (p value =0.912) (figure 4, panel-C). Noteworthy, males had higher ALT levels (37.99±2.43 U/L) compared to the female obese participants (19.41±1.45 U/L) (p value=0.002) (figure 4, panel-D).

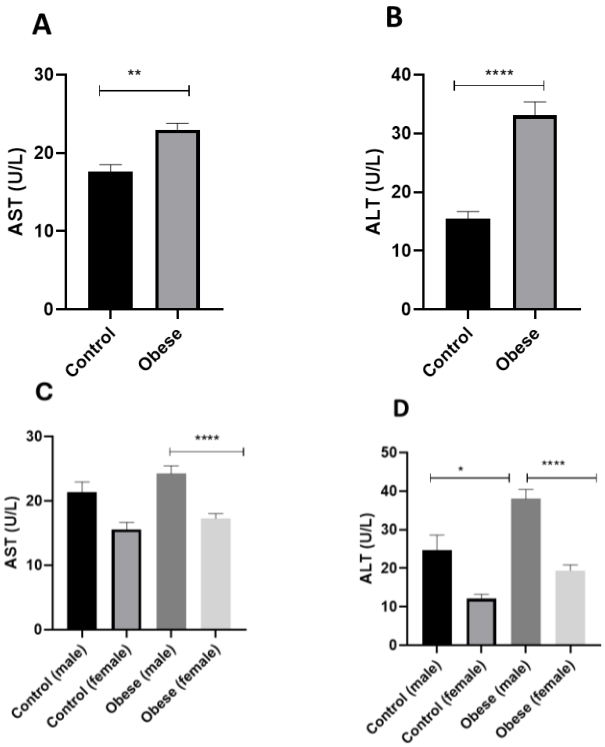


Figure 4: AST and ALT in serum of the obese participants compared to the control participants. Panel A: A comparison of AST level between the obese and control groups (regardless of gender). Panel B: A comparison of ALT between obese and control groups (regardless of gender). Panel C: A

comparison of the AST level in obese and control groups (gender based). Panel D: A comparison of the ALT level of male and female participants (gender based). Results are presented as Mean ±SEM from duplicate determinations. *p < 0.05, **p < 0.01, ****p < 0.0001 (adjusted p values).

Elevated liver function tests, especially AST and ALT, are associated with liver steatosis, which leads to insulin resistance and subsequently the onset of diabetes[17] [18] which is identified an enhancement in liver function associated with diabetes. A meta-analysis including twenty-five studies showed that liver function tests were increased in the obese individuals compared to the control participants [19]. This coincidences with our results that showed higher AST and ALT values in the obese participants compared to the control participants. Our result showed the high AST and ALT levels in males compared to the female obese people. The sustained suppression of postprandial serum AST and ALT levels in lean females relative to their male counterparts is primarily influenced by sexually dimorphic adipose tissue distribution and divergent immunometabolic profiles. Estrogen sends extra fat to subcutaneous depots instead of the liver, which protects it from direct portal delivery of lipotoxic free fatty acids that cause male visceral enlargement. Additionally, female endocrine profiles directly inhibit local complement-mediated inflammatory cascades, particularly macrophage polarization induced by C5a and the consequent elevation of IL-1β, thereby averting systemic metabolic dysfunctions and maintaining hepatoprotective leptin levels. At the hepatocellular levels, ERα signaling enhances the clearance of hepatic mitochondrial lipids to reduce oxidative stress and the intracellular release of enzymes. In contrast, the diminished absolute mass of skeletal muscle in females reduces the postural contribution to enzymatic output into circulation at rest [20-22].

3.5. Effect of Obesity on Diabetes

Diabetes is a feature of obesity. To investigate diabetes features in obesity, glucose levels insulin and HbA-1c were performed. Our result showed that there were significantly higher fasting glucose levels in the obese participants (118.3±4.2 mg/dL) (regardless of gender), compared to the control participants (100.6±0.86 mg/dL) (p value =0.0005) (figure 5, panel-A). Males and females fasting blood glucose in the obese participants were not significantly different compared to the control participants (figure 5, panel-D) (gender based). To investigate diabetes features, insulin ELISA test was performed. The result showed higher insulin levels in the obese participants (25.83±2.00 µU/mL) compared to the control participants (8.20±1.27 µU/mL) (p value <0.0001) (regardless of gender) (figure 5, panel-B). In addition, Insulin levels was measured in the male and female obese participants (gender based), the result showed that insulin levels was higher in the male participants (25.59±1.76 µU/mL) compared to the male control participants (7.36±1.28 µU/mL) (p value <0.0001) and the female obese participants (22.75±1.53 µU/mL) had higher insulin levels compared with the female (7.273±1.46 µU/mL) control participants (p value =0.024) (figure 5, panel-E).

To determine glucose binding with hemoglobin, HbA-1c % was performed resulted in higher HbA-1c levels in the obese participants (5.800±0.11 %) compared to the control participants (4.94±0.100 %) (figure 5, panel-C) (p value<0.0001). HbA-1c percentage in males and females' the obese participants were measured. Male the obese participants (5.784±0.13 %) had higher HbA-1c percentage compared to male normal participants (4.75±0.11 %) (p value=0.0012) (figure 5, panel-F). No significantly differences were detected of HbA-1c percentage in the female obese participants compared to female normal participants (figure 5, panel-F).

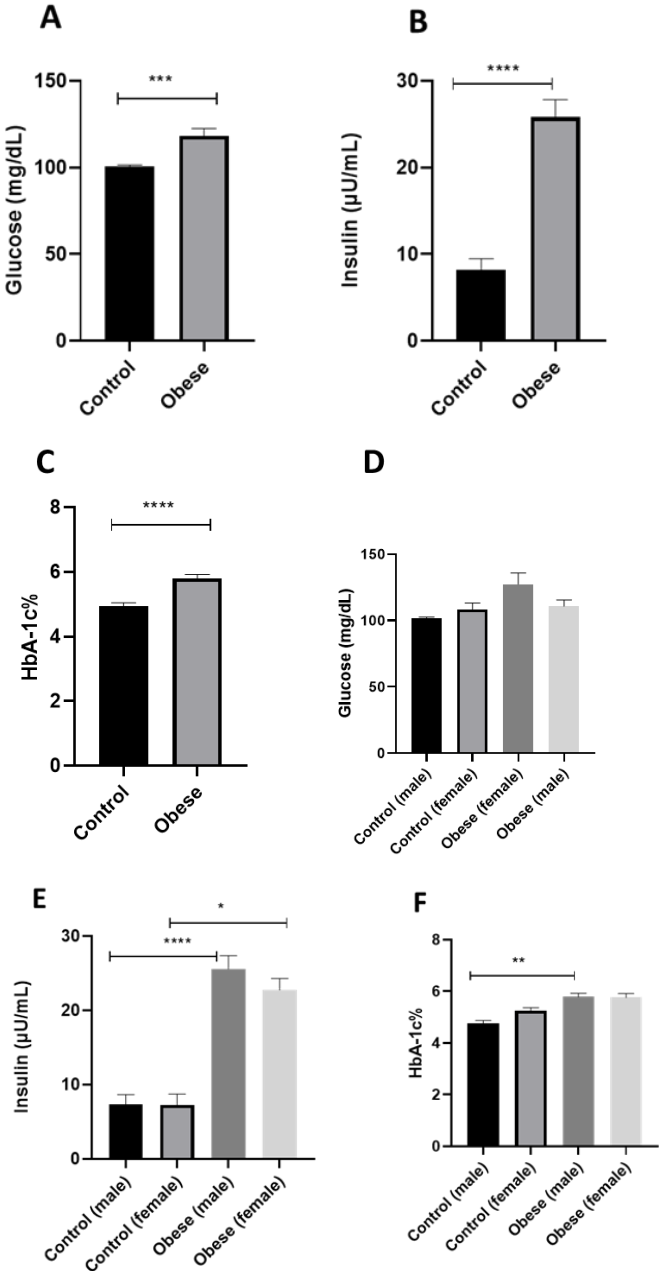


Figure 5: Glucose, insulin and HbA-1c% in the obese participants compared to the control participants. Panel A: A comparison of glucose level between the obese and control groups (regardless of gender). Panel B: A comparison of insulin level between obese and control groups (regardless of gender). Panel C: A comparison of the HbA-1c % in obese and control groups (regardless of gender). Panel D: A comparison of glucose between male and female participants (gender based). Panel E: A comparison of the insulin level of male and female participants (gender based). Panel F: Comparison of HbA-1c % between male and female participants (gender based). Results are presented as Mean ±SEM from duplicate determinations. *p < 0.05, **p < 0.01, ***p < 0.001, ****p < 0.0001 (adjusted p values).

Our results indicated that the obese individuals showed significantly elevated glucose levels compared to the control participants. To examine glucose binding with hemoglobin, HbA-1c testing revealed elevated HbA-1c levels in the obese individuals compared to the control participants. Our findings coincidence with those of [23], who showed in a cohort study conducted in China and England that glucose dysregulation and elevated HbA1c levels in the obese individuals over the years contribute to the development of diabetes. A study performed in Saudi Arabia found that BMI and HbA-1c levels were higher in the obese individuals compared to their non-the obese participants. Our findings align with those results[24], which showed that, elevated HbA1c levels, insulin resistance, and obesity are all associated with elevated insulin levels. Elevated HbA1c and glucose levels are linked to higher insulin levels, which also exacerbate insulin resistance[20, 25, 26]. This appears to suggest that elevated insulin levels are associated with glucose buildup and increased HbA1c, leading to the development of diabetes.

3.6. Effect of Obesity on HOMA-IR (Homeostatic Model Assessment of Insulin Resistance)

Insulin resistance, which is associated with diseases like diabetes that are related to obesity, was examined using the Homeostatic Model Assessment of Insulin Resistance (HOMA-IR). This is how it was calculated: Quick Insulin [µU/mL] × Quick Glucose [mg/dL]) / 405 is HOMA-IR. The following are the HOMA-IR interpretations: Early/Mild Insulin Resistance: 1.0–1.9; Normal: < 1.0. Significant/Moderate Insulin Resistance: > 2. Regardless of gender, the obese participants' HOMA-IR score was higher (6.18±0.61) than that of the control participants (1.64±0.25) (p value=<0.0001) (figure 6, panel-A). Additionally, the HOMA-IR score was determined for both the male and the female obese participants based on gender; the male participants had a higher HOMA-IR score (7.77±1.25) than the male control participants (1.69±0.35) (p value = 0.0007), while the female obese participants had a higher HOMA-IR score (6.20±0.59) than the female control participants (1.54±0.29) (p value <0.0001) (figure 6, panel-B). It is appeared that the presence of insulin resistance in the obese people due to the high glucose and insulin secretion.

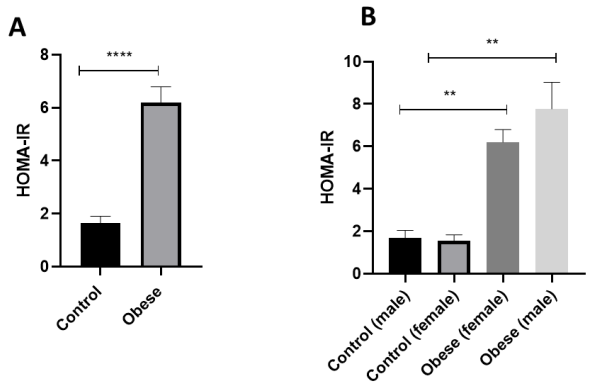


Figure 6: HOMA-IR score in the obese participants compared to the control participants. Panel A: A comparison of HOMA-IR between the obese and control groups (regardless of gender). Panel B: A comparison of HOMA-IR between obese and control groups (gender based). Results are presented as Mean ±SEM from duplicate determinations. **p < 0.01, ****p < 0.0001 (adjusted p values).

The obese people have a higher HOMA-IR because of increased pro-inflammatory cytokines (IL-1β, TNFα), lipid toxicity, dysfunctional adipocytes, and direct impairment of insulin receptor substrate-1 (IRS-1) phosphorylation in peripheral tissues, which leads to hyperinsulinemia and elevated HOMA-IR [27, 28].

3.7. Effect of Obesity on Adiponectin

Besides the insulin ELISA test, adiponectin levels were measured. This is because adiponectin is reciprocal to insulin secretion. Surprisingly, the adiponectin levels were higher in the obese participants (5.34±0.20 ng/mL) (regardless of gender) compared to the control participants (3.89±0.11 ng/mL) (p value <0.0001) (figure 7, panel-A). Notably, no significant differences were observed between the obese males and females when compared with the male and female control participants (gender based) (figure 7, panel-B).

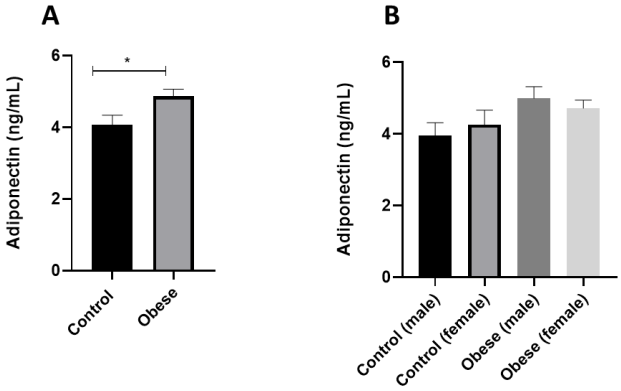


Figure 7: Adiponectin in the obese participants compared to the control participants. Panel A: A comparison of adiponectin between the obese and control groups (regardless of gender). Panel B: A comparison of adiponectin between obese and control groups (gender based). Results are presented as averages ±SEM from duplicate determinations. *p < 0.05 (adjusted p values).

The obese individuals exhibit elevated adiponectin levels compared to lean individuals as a result of the inhibition of adiponectin gene expression. Classically activated macrophages and the vigorous release of pro-inflammatory cytokines, such as TNFα and IL-6, transcriptionally inhibit the ADIPOQ gene. In these patients, there is a significantly higher levels of adiponectin among those who are morbidly the obese, have hepatic impairment, or suffer from nephropathy. Furthermore, particular polymorphisms in the adiponectin receptor gene induce adiponectin resistance, leading to hyperadiponectinemia in unhealthy fat individuals. [29, 30].

3.8. Effect of Obesity on Thyroid Disorder

Because obesity has a positive association with TSH. This is due to the increase metabolism and fat percentage. Interestingly, the TSH levels increased in the obese participants (2.56±0.22 µIU/ml) (regardless of gender) compared

to normal participants (1.51±0.18 µIU/ml) (p value =0.0316) (figure 8, panel-A). Noteworthy, no significant differences were detected between the obese male and female participants when compared with the male and female control participants (gender based) (figure 8, panel-B).

In our study, increased TSH levels were noted in individuals with obesity. This occurrence is believed to indicate a compensatory upregulation of the hypothalamic–pituitary–thyroid axis, partially attributed to adiposity-related leptin signaling affecting the hypothalamus, which stimulates hypothalamic TRH and enhances pituitary TSH secretion. Moreover, obesity-related inflammatory signaling causes relative central thyroid hormone resistance, elevates the HPT-axis set-point, and alters deiodinase activity, resulting in increased conversion of T4 to T3 [31, 32].

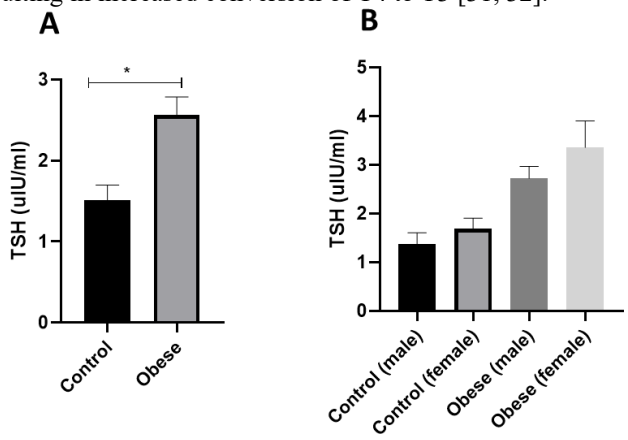


Figure 8: TSH in the obese participants compared to the control participants. Panel A: A comparison of TSH between the obese and control groups (regardless of gender). Panel B: A comparison of TSH between obese and control groups (gender based). Results are presented as Mean ±SEM from duplicate determinations. *p < 0.05 (adjusted p values).

4. Conclusion

It can be concluded that pro-inflammatory cytokine such as IL-1β was higher in the obese participants compared to the control participants. However, an inflammatory marker such as IL-10 was lower in the obese participants compared to the control participants. This indicated macrophage polarization from anti-inflammatory property to anti-inflammatory property. The increase of pro-inflammatory cytokines led to the have more CRP-levels in the obese participants resulting in the development of insulin resistance. The fatty liver disease markers such as AST and ALT were higher in the obese participants which play a significant role to enhance insulin resistance. Central obesity can cause insulin disruption due to the high levels of free fatty acid. The presence of insulin resistance in the obese participants caused the development of Type 2 diabetes mellitus.

Conflict of Interest

There are no financial and non-financial interests competing.

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Author Contributions

Baiz Hamad Abdalla performed experiments, data analysis and preparing the draft. Ramiar Kamal Kheder: supervised the project and revised the manuscript.

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