



Assessment of some Cytokines in Obese patients with Diabetic

Samar A.Z, Mutashar.J.J

Department of Biology, Collage of Science, Wasit University, Iraq

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Abstract

Background: Obesity and diabetes mellitus are closely interrelated metabolic disorders characterized by chronic low-grade inflammation mediated by pro-inflammatory cytokines. Cytokines such as tumor necrosis factor-alpha (TNF- α) and interleukin-6 (IL-6) play a crucial role in the development of insulin resistance and progression of type 2 diabetes mellitus (T2DM). Aim: This study aimed to evaluate the levels of TNF- α and IL-6 in obese patients with and without diabetes and to investigate their association with HbA1c levels and lipid profile parameters. Methods: A case-control study was conducted including 90 participants divided into three groups: 30 obese patients with diabetes, 30 obese patients without diabetes, and 30 healthy controls. Serum levels of TNF- α and IL-6 were measured using enzyme-linked immunosorbent assay (ELISA). HbA1c and lipid profile parameters were assessed using standard biochemical methods. Statistical analysis was performed using SPSS version 25 with ANOVA, chi-square, correlation analysis, and ROC curve analysis. Results: The findings demonstrated significantly elevated levels of TNF- α and IL-6 in both obese groups compared with healthy controls ($P \leq 0.001$). HbA1c levels were significantly higher in obese patients with diabetes compared to other groups ($P < 0.001$). Dyslipidemia was evident in obese individuals, with increased cholesterol, triglycerides, LDL, and VLDL levels, and decreased HDL levels compared with controls. ROC curve analysis indicated that TNF- α and IL-6 possess good diagnostic performance in distinguishing obese patients from healthy individuals, with AUC values of 0.869 and 0.847, respectively. Significant positive correlations were observed between HbA1c and TNF- α in obese patients, and between HbA1c and IL-6 in obese diabetic patients. Conclusion: The study confirms that elevated TNF- α and IL-6 levels are strongly associated with obesity and diabetes and may serve as valuable biomarkers for inflammation-related metabolic dysfunction.

Keyword: Obesity, Diabetes, IL-6, TNF- α , cytokines

1. **Background: Obesity and type 2 diabetes mellitus (T2DM) share a complex pathophysiological relationship, with chronic inflammation playing a key role. Pro-inflammatory cytokines like tumor necrosis factor-alpha (TNF- α) and interleukin-6 (IL-6) have been implicated in driving insulin resistance. Aim: We sought to measure TNF- α and IL-6 in obese patients—some with diabetes, some without—and examine how these markers relate to HbA1c and lipid abnormalities. Methods: Ninety subjects participated: 30 obese with T2DM, 30 obese without diabetes, and 30 healthy controls matched for age and sex. TNF- α and IL-6 were quantified by ELISA. Lipid profiles and HbA1c were measured by standard methods. We used ANOVA, chi-square, correlation, and ROC analyses. Results: Both TNF- α and IL-6 were significantly higher in obese groups versus controls ($P \leq 0.001$). HbA1c was markedly elevated in the obese diabetic group ($P < 0.001$). Lipid profiles showed clear dyslipidemia—raised cholesterol, triglycerides, LDL, and VLDL, with lower HDL—in obese individuals. ROC curves showed that TNF- α (AUC = 0.869) and IL-6 (AUC = 0.847) can effectively distinguish obese patients from controls. TNF- α correlated with HbA1c in obese patients, while IL-6 correlated with HbA1c in the diabetic subgroup. Conclusion: TNF- α and IL-6 appear to be reliable indicators of the inflammatory burden associated with obesity and diabetes, and they may serve as useful clinical markers.**

The two most vital systems for survival are the immune and metabolic systems. They also exhibit a significant interconnection in the regulation of metabolism and immune responses. The metabolic systems of organisms involve the cells and the biochemical reactions they carry out. Metabolic disorders, including the closely related conditions of obesity, insulin resistance, and T2DM, are associated with chronic inflammation. This results from abnormal cytokine production and the activation of inflammatory signaling pathways. Increased plasma levels of leptin, a protein secreted by adipose tissue that functions to regulate body weight by suppressing appetite while simultaneously enhancing metabolism and energy expenditure, are associated with obesity. In individuals with obesity, adipose tissue acts as a metabolically active organ. It can contribute to low-grade chronic inflammation. The inflammatory process is crucial in the pathogenesis of diabetes mellitus, with chronic inflammation occurring prior to the disease's onset.

The first proinflammatory cytokine described as involved in the pathogenesis of insulin resistance and type 2 diabetes mellitus (T2DM) was TNF- α . It has been reported that TNF- α reduces the expression of the insulin-regulated glucose transporter type 4 (GLUT4), which is mainly in adipocytes and skeletal and cardiac muscles. In addition, TNF- α can inhibit the action of insulin in the periphery by causing serine phosphorylation of insulin receptor substrate-1, thus contributing to insulin resistance (Akash et al., 2018). Recent meta-analyses have shown that both type 1 and type 2 diabetics have significantly higher serum TNF- α levels compared to healthy individuals; these levels positively correlate with insulin resistance (Qiao et al., 2017; Liu et al., 2016). In obese patients with T2DM, plasma TNF- α levels are associated with quantity of visceral fat and do not respond acutely to changes in blood glucose levels in individuals with poorly controlled diabetes (Berlin et al., 2000). The question of whether TNF- α causes obesity and diabetes is still open and is actively investigated.

The metabolic and immune systems are two of the most critical physiological networks, and their interaction is being understood more comprehensively. It is now firmly established that chronic low-grade inflammation, which results from excessive cytokine production and activation of inflammatory signaling pathways, plays a significant role in the development of metabolic disorders.

These include obesity, insulin resistance, and type 2 diabetes mellitus. Adipose tissue was once considered a simple fat depot but is now known to be a dynamic endocrine organ. Among its products are several bioactive molecules, including leptin, which acts in the brain to reduce food intake and increase energy expenditure. When fat depots are expanded in obesity, this tissue undergoes remodeling. Oxygen supply cannot keep up with the growth of the tissue, leading to hypoxia at a localized level. This, in turn, results in a persistent low-grade inflammatory environment that, in turn, is associated with an increased risk for diabetes to develop. (Hotamisligil, 2009).

TNF- α was among the first cytokines linked to insulin resistance. It works, at least in part, by reducing the expression of GLUT4—the main glucose transporter in muscle and fat cells—thereby limiting how much glucose enters these tissues. TNF- α also interferes with insulin receptor substrate-1 (IRS-1) through serine phosphorylation, effectively blocking the normal insulin signaling cascade (Akash et al., 2018). Multiple meta-analyses have shown that both type 1 and type 2 diabetic patients carry higher serum TNF- α concentrations, and these levels tend to track with the severity of insulin resistance (Qiao et al., 2017; Liu et al., 2016). Interestingly, in obese T2DM patients, the amount of visceral fat seems to be the main determinant of circulating TNF- α , and acutely lowering blood glucose does not appear to change TNF- α levels right away (Berlin et al., 2000). The precise mechanisms remain under investigation.

Interleukin-6 is a pleiotropic cytokine that affects a wide range of cell types and tissues, both within and outside the immune system (Hamza and Shakir, 2025). Its sometimes contradictory effects—pro-inflammatory in some settings, anti-inflammatory in others—stem from a uniquely complex signaling system (Schaper, 2015). In the context of metabolic disease, IL-6 promotes tissue-specific insulin resistance and impairs insulin secretion from pancreatic β -cells, both of which accelerate the development of T2DM (Rehman et al., 2017). Obese individuals with elevated IL-6 face a greater risk of cardiovascular disease and metabolic syndrome (El-Mikkawy et al., 2020). Given these associations, the present study was undertaken to explore how TNF- α and IL-6 relate to HbA1c levels and to determine whether these cytokines are differentially altered in obese patients with and without diabetes.

In this case-control study, 60 patients divided into two groups: 30 obesity patients with diabetic disease and 30 obesity patients without diabetic disease were recruited from Al-Zahra Teaching Hospital in Al-Kut-Wasit Governorate / Iraq, during the period from October 2024- to April 2025. Both obesity and diabetes significantly impacts immune responses and cytokine levels. Diabetes patients were clinically diagnosed by a specialized physician. In addition to patients; 30 healthy individual who attended for .laboratory for routine checkups were considered as a control group for comparison with the patient group All subjects aged from 15 to 73 years. None of the both patient participants had received a medication ,that effect in metabolic process or inflammation markers. Patients with cardiovascular diseases, cancer osteoarthritis, history of other inflammatory diseases, autoimmune diseases, patients with smoking, or .drinking were excluded from the study

Laboratory parameters affect the disease

This was a case-control study involving 60 obese patients: 30 with confirmed T2DM and 30 without diabetes. All patients were recruited from Al-Zahra Teaching Hospital, Al-Kut, Wasit Governorate, Iraq, over the period from October 2024 to April 2025. Diabetes was diagnosed clinically by specialist physicians. We also enrolled 30 healthy, non-obese individuals as controls; these were people who came to the hospital laboratory for routine checkups. Ages ranged from 15 to 73 years. Anyone taking medication that could alter metabolic or inflammatory markers was excluded, as were individuals with cardiovascular disease, cancer, osteoarthritis, autoimmune conditions, smokers, and those with a history of alcohol use.

The Lipid profile parameters included in this study were: the total cholesterol, Triglyceride, LDL, VLDL and HDL these parameters were measured by spinreact kit

Immunological parameters

The lipid panel measured in this study included total cholesterol, triglycerides, LDL cholesterol, VLDL cholesterol, and HDL cholesterol. All measurements were performed using commercial diagnostic kits from Spinreact (Spain).

Briefly, TNF- α and IL-6 assay was conducted by 50 μ L of standard and 40 μ L of sample were added along with the 10 μ L of either (Human TNF- α antibody or Human IL-6 antibody) respectively and 50 μ L of streptavidin-HRP. The contents were mixed well. The plate was covered with a sealer and incubated for 60 minutes at 37°C. The plate was then washed five times by 300 μ L wash buffer to ensure removal of unbound materials. 50 μ L of both substrate solutions A and B were added and incubated in the dark for 10 minutes at 37°C before stopping the reaction by 50 μ L of stop solution, which produced a color change from blue to yellow. Finally, the optical density was measured at 450 nm within ten minutes to ensure accurate and reliable results.

Serum TNF- α and IL-6 concentrations were measured by ELISA using human TNF- α (BT LAB, Catalog No.: E0082Hu) and IL-6 (BT LAB, Catalog No.: E0090Hu) kits. Assays were carried out following the manufacturer's instructions.

In brief, 50 μ L of standard and 40 μ L of sample were pipetted into wells, along with 10 μ L of the appropriate antibody (anti-TNF- α or anti-IL-6) and 50 μ L of streptavidin-HRP. After mixing, plates were sealed and incubated at 37°C for 60 minutes. Wells were washed five times with 300 μ L of wash buffer. Then 50 μ L each of substrate A and substrate B were added, and plates sat in the dark at 37°C for 10—minutes. The reaction was stopped with 50 μ L of stop solution—the color changed from blue to yellow and absorbance was read at 450 nm within 10 minutes

Ethics Approval

Statistical analyses were done using SPSS version 25.0 (IBM Corp., Armonk, NY, USA). One-way ANOVA was used to compare means among the three groups when data were normally distributed. The chi-square test was applied for comparing proportions. Statistical significance was set at $P \leq 0.05$

RESULTS

Ethical approval was obtained from the Ethics Committee of the Biology Department, College of Science, University of Wasit, as well as from the Iraqi Ministry of Health. Written informed consent was obtained from each participant before inclusion in the study.

Sixty patients of Obese were studied in contrast to 30 sex and age-matched control group of healthy individuals. The demographic characteristics of patients and control subjects are shown in table (1). According to age, the mean age of Obese patients was 32.70 ± 8.11 years old, 50.43 ± 9.55 years old for obese patients with DM, and that of control subjects was 33.93 ± 7.34 years old and there was significant difference between patients and control subjects in mean age ($P = 0.001$). Regarding to gender, Obese patients included 12 (40.0%) cases were male gender and 18 (60.0%) cases were female, Obese patients with DM included 12 (40.0%) cases were male gender and 18 (60.0%) cases were female, while control group included 15 (50.0%) cases were male gender and 15 (50.0%) cases were female and there was non-significant difference in the frequency distribution of patients and control subjects according to gender ($P = 0.665$).

Table (1): Characteristics of patients and healthy control

Characteristic	Obese patients (n=30)	Obese with DM (n=30)	Healthy Control (n=30)	P
Age (years)	32.70 ± 8.11	50.43 ± 9.55	33.93 ± 7.34	0.001
Gender				
Male	12 (40.0%)	12 (40.0%)	15 (50.0%)	0.665
Female	18 (60.0%)	18 (60.0%)	15 (50.0%)	

Sixty obese patients and 30 healthy controls were included. Their baseline characteristics are shown in Table (1). The obese non-diabetic group had a mean age of 32.70 ± 8.11 years, the obese diabetic group 50.43 ± 9.55 years, and healthy controls 33.93 ± 7.34 years. The age difference was statistically significant ($P = 0.001$). Males constituted 40.0% (n=12) and females 60.0% (n=18) in both obese groups. Among controls, 50.0% were male and 50.0% female. There was no significant gender difference across groups ($P = 0.665$).

The comparison of some metabolic factors (HbA1c) between patients and control group has been carried out and the results were demonstrated in table (2). The mean of HbA1c were 4.89 ± 0.41 , 8.72 ± 1.22 and 4.44 ± 0.51 , in Obese patients, Obese patients with DM and healthy control group respectively; the mean was higher in Obese patients with DM in compared to other both groups (Obese patients and healthy control) and the difference was highly significant ($P < 0.001$). But there was non-significant difference between Obese patients and healthy control themselves.

Table (2): Comparison of some metabolic factors (HbA1c) in patients and healthy controls

Groups		HbA1c (%)
Obese patients	Mean $\pm$ SD	4.89 ± 0.41^A
Obese with DM	Mean $\pm$ SD	8.72 ± 1.22^B
	Mean $\pm$ SD	4.44 ± 0.51^A
p-value		0.001**
Different latters denote to the significant differences at $p < 0.05$		

HbA1c levels across the three groups are presented in Table (2). Mean values were $4.89 \pm 0.41\%$ for obese non-diabetics, $8.72 \pm 1.22\%$ for obese diabetics, and $4.44 \pm 0.51\%$ for controls. The obese

diabetic group had significantly higher HbA1c than both other groups ($P < 0.001$). There was no significant difference between obese non-diabetics and controls.

Determine the lipid profile parameters of patients and controls

The comparison of lipid profile between patients and control group has been carried out and the results were demonstrated in table (3). Mean levels of serum cholesterol were 182.71 ± 18.72 mg/dl, 205.69 ± 18.9 mg/dl and 101.73 ± 11.26 mg/dl, in Obese patients, Obese patients with DM and healthy control group respectively; the mean levels was higher in both group of patients (Obese patients and Obese patients with DM) in compared to control group and the difference was significant ($P > 0.05$). But the mean levels was non-significant difference between both group of patients (Obese patients and Obese patients with DM) themselves ($P < 0.05$). The mean levels of serum triglycerides were 145.91 ± 23.4 mg/dl, 180.71 ± 24.2 mg/dl and 78.3 ± 8.71 mg/dl, in Obese patients, Obese patients with DM and healthy control group respectively; the mean levels was higher in both group of patients (Obese patients and Obese patients with DM) in compared to control group and the difference was significant ($P > 0.05$). Also the mean levels was significant difference between both group of patients (Obese patients and Obese patients with DM) themselves ($P > 0.05$).

Regarding the mean levels of serum LDL and serum VLDL, the present results show the mean levels of LDL and VLDL were higher in both group of patients (Obese patients and Obese patients with DM) in compared to control group and the difference was significant ($P > 0.05$). But the mean level of serum HDL was lower in both group of patients (Obese patients and Obese patients with DM) in compared to control group and the difference was significant ($P > 0.05$).

Table (3): Results of lipid profile in patients and healthy controls.

Characteristic	Obese patients <i>n</i> = 30	Obese with DM <i>n</i> = 30	Healthy control <i>n</i> = 30	P value
Cholesterol (mg/dl)				
Mean± SD	182.71 ± 18.72^A	205.69 ± 18.9^A	101.73 ± 11.26^B	0.001
Triglycerides (mg/dl)				
Mean± SD	145.91 ± 23.4^A	180.71 ± 24.2^B	78.3 ± 8.71^C	0.001
low-density lipoprotein (LDL) (mg/dl)				
Mean± SD	117.65 ± 17.8^A	115.16 ± 19.2^A	41.86 ± 9.11^B	0.001
Very low-density lipoprotein (VLDL) (mg/dl)				
Mean± SD	28.9 ± 8.8^A	37.5 ± 7.6^B	15.6 ± 2.77^C	0.001
High-density lipoprotein (HDL) (mg/dl)				
Mean± SD	36.3 ± 6.6^A	37.3 ± 6.3^A	44.3 ± 2.91^B	0.001
Different latters denote to the significant differences at $p < 0.05$				

LDL and VLDL were also significantly elevated in both obese groups relative to controls ($P = 0.001$). On the other hand, HDL was significantly lower: 36.3 ± 6.6 mg/dL in obese non-diabetics, 37.3 ± 6.3 mg/dL in obese diabetics, compared with 44.3 ± 2.91 mg/dL in controls ($P = 0.001$).

Immunological parameters of patients and controls

Comparison of immunological data between patients and controls as Table 4. Mean levels of TNF- α were 170.6 ± 10.31 , 160.37 ± 18.5 and 136.9 ± 15.7 , in Obese patients, Obese patients with DM and healthy control group respectively; the mean levels was higher in both group of patients (Obese patients and Obese patients with DM) in compared to control group and the difference was highly significant ($P > 0.001$). But the mean levels was non-significant difference between both group of patients (Obese patients and Obese patients with DM) themselves ($P < 0.05$). Likewise, IL-6 levels were markedly increased in group of patients (170.6 ± 10.31 and 160.37 ± 18.5) compared to controls (136.9 ± 15.7 ; $P = 0.001$).

Immunological findings are detailed in Table (4). TNF- α levels averaged 170.6 ± 10.31 pg/mL in obese patients, 160.37 ± 18.50 pg/mL in obese diabetics, and 136.9 ± 15.70 pg/mL in controls. Both obese groups had significantly elevated TNF- α compared with controls ($P = 0.001$), though the two obese groups did not differ from each other. IL-6 showed a similar pattern: 149.52 ± 16.41 pg/mL in obese patients, 128.57 ± 11.20 pg/mL in obese diabetics, and 102.58 ± 11.40 pg/mL in controls ($P = 0.001$).

Table (4): Comparison of Immunological parameters (TNF- α and IL-6) in patients and healthy controls

Groups	TNF- α	IL-6
Obese patients	170.6 ± 10.31^A	149.52 ± 16.41^A
Obese with DM	160.37 ± 18.5^A	128.57 ± 11.2^B
Control	136.9 ± 15.7^B	102.58 ± 11.4^C
p-value	0.001**	0.001**
Means followed by different letters are significantly different according to Duncan's multiple range comparisons (DMRTs), Means followed by the same letter are not significantly different.		

SD: standard deviation; †: one way ANOVA; **: significant at $P < 0.05$

Diagnostic performance of Immunological parameter (TNF- α)

Receiver operating characteristic (ROC) analysis was performed to reveal the prognostic accuracy of using Tumor necrosis factor alpha (TNF- α) concentrations to distinguish Obese patients from healthy control subjects. An optimal TNF- α cut-off value more than of 158.21 resulted in an AUC value of 0.869 (95% confidence interval [CI], 0.770- 0.963, $P = 0.001$), sensitivity of 86.7%, specificity of 86.7%, PPV of 86.7%, and NPV of 86.7%. The present results indicates TNF- α is considered as good prognostic marker to distinguish Obese patients from healthy control.

In addition, an optimal TNF- α cut-off value of more than 141.89 could be used to distinguish obese patient with DM from healthy control subjects with a sensitivity of 73.3%, specificity of 70.0%, PPV of 71.0%, and NPV of 72.4% and 0.710 (0.581- 0.839). The present results indicates TNF- α is considered as acceptable prognostic marker to distinguish obese patient with DM from healthy control.

ROC analysis for TNF- α yielded an AUC of 0.869 (95% CI: 0.770–0.963, $P = 0.001$) when comparing obese patients with controls. At a cutoff of 158.21 pg/mL, the sensitivity was 86.7%, specificity 86.7%,

PPV 86.7%, and NPV 86.7%. This suggests that TNF- α is a good marker for distinguishing obese individuals from healthy ones.

When comparing obese diabetic patients to controls, the TNF- α cutoff of 141.89 pg/mL gave an AUC of 0.710 (95% CI: 0.581–0.839), with sensitivity 73.3%, specificity 70.0%, PPV 71.0%, and NPV 72.4%. This indicates an acceptable, though more modest, diagnostic performance in the diabetic subgroup.

Table (5): Roc curve of Tumor necrosis factor alpha (TNF- α) level

Characteristic	Obese patients / control	Obese with DM /control
Cutoff value	> 158.21	> 141.89
P value	0.001	0.001
Sensitivity %	86.7 %	73.3 %
Specificity %	86.7 %	70.0 %
PPV %	86.7 %	71.0 %
NPV %	86.7 %	72.4 %
AUC (95% CI)	0.869 (0.770- 0.963)	0.710 (0.581- 0.839)

CI: Confidence interval, AUC: Area under curve.

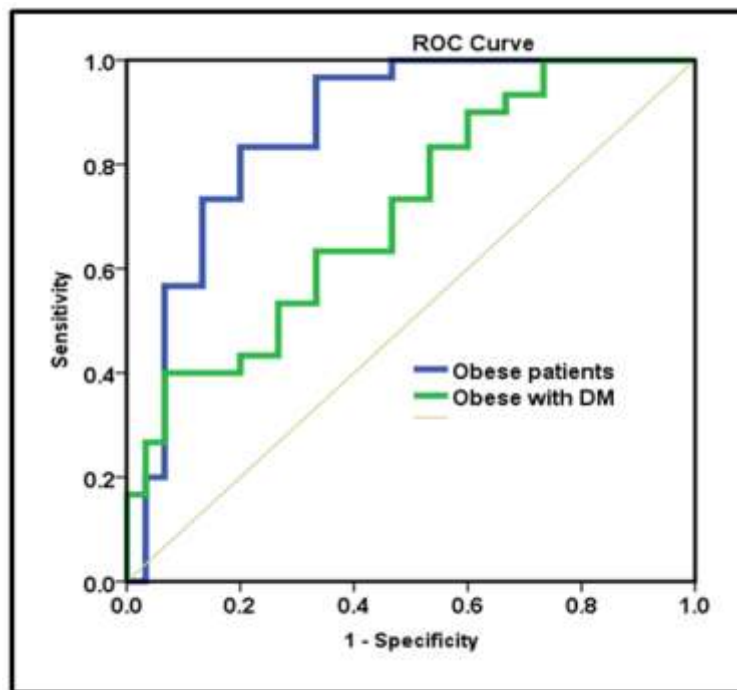


Figure (1): (Blue curve) Receiver operating characteristic curve for TNF- α levels to distinguish Obese patients from healthy control subjects. (Green curve) Receiver operating characteristic curve for TNF- α levels to distinguish Obese patients with DM from healthy control subjects.

Diagnostic performance of Immunological parameter (IL-6)

Receiver operating characteristic (ROC) analysis was performed to reveal the prognostic accuracy of using Interleukin-6 (IL-6) concentrations to distinguish Obese patients from healthy control subjects. An optimal IL-6 cut-off value more than of 114.40 resulted in an AUC value of 0.847 (95% confidence interval [CI], 0.751- 0.943, $P= 0.001$), sensitivity of 86.7%, specificity of 83.3%, PPV of 83.9%, and NPV of 86.2%. The present results indicates IL-6 is considered as good prognostic marker to distinguish Obese patients from healthy control.

In addition, an optimal IL-6 cut-off value of more than 14.00 could be used to distinguish obese patient with DM from healthy control subjects with a sensitivity of 76.7%, specificity of 80.0%, PPV of 79.3%, and NPV of 77.4% and 0.775 (0.644- 0.905). The present results indicates IL-6 is considered as acceptable prognostic marker to distinguish obese patient with DM from healthy control.

Table (6): Roc curve of Interleukin-6 (IL-6) level

Characteristic	Obese patients / control	Obese with DM /control
Cutoff value	> 114.40	> 114.00
P value	0.001	0.001
Sensitivity %	86.7 %	76.7 %
Specificity %	83.3 %	80.0 %
PPV %	83.9 %	79.3 %
NPV %	86.2 %	77.4 %
AUC (95% CI)	0.847 (0.751- 0.943)	0.775 (0.644- 0.905)

For the obese diabetic versus control comparison, the IL-6 cutoff was 114.00 pg/mL, with an AUC of 0.775 (95% CI: 0.644–0.905), sensitivity 76.7%, specificity 80.0%, PPV 79.3%, and NPV 77.4%.

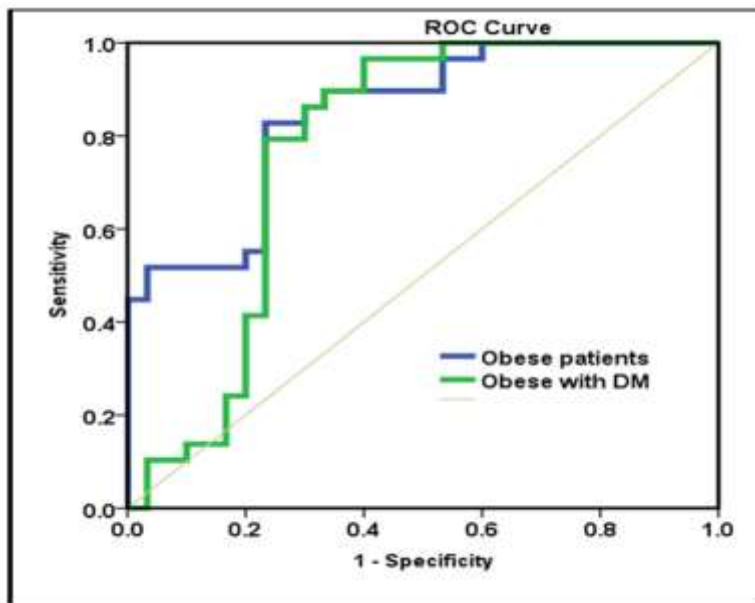


Figure (2): (Blue curve) Receiver operating characteristic curve for IL-6 levels to distinguish Obese patients from healthy control subjects. (Green curve) Receiver operating characteristic curve for IL-6 levels to distinguish Obese patients with DM from healthy control subjects.

Logestic regression correlations between immunological parameters levels in patient groups.

The Logistic regression model show that the correlation immunological parameter such as IL-6 in which have directly correlate with TNF- α among obese patients as in figure (3-15) this result might be refer to that obese condition enhances production of IL-6 in suppression the expression of TNF- α .

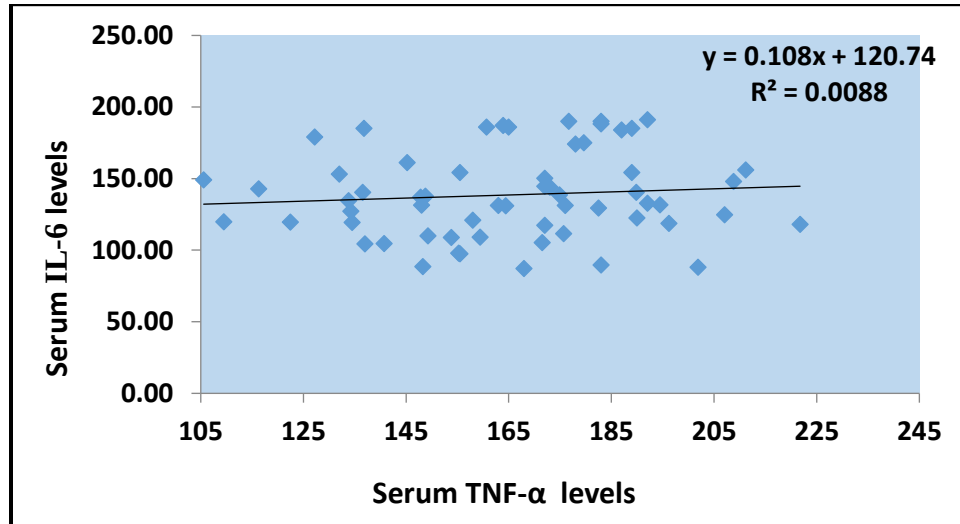


Figure (3): The Logistic scatter IL-6 and TNF- α levels among patients.

Correlation between HbA1c and immunological parameters in both group patients.

The correlation between HbA1c level and immunological parameters in both group patients were shown in tables (Berlin et al., 2000). There are positive correlation between HbA1c levels and TNF- α ($r=0.457$ and $p=0.011$) in Obese patients. Also there are positive correlation between HbA1c and IL-6 levels ($r=0.349$ and $p=0.039$), in obese patients with DM.

Table (7): The correlation between HbA1c and immunological parameters in both group patients

Other parameters	HbA1c			
	Obese patients		Obese with DM patients	
	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>
TNF- α	0.457	0.011*	0.084	0.657
IL-6	0.049	0.799	0.349	0.039*

The correlations between HbA1c and cytokine levels are shown in Table (7). In the obese non-diabetic group, TNF- α correlated positively with HbA1c ($r = 0.457$, $P = 0.011$). In the obese diabetic group, IL-6 was positively correlated with HbA1c ($r = 0.349$, $P = 0.039$).

Discussion

Obesity is widely regarded as one of the most important modifiable risk factors for T2DM, and it has been linked to roughly 80% of diabetes cases (Alnaqeeb et al., 2022). People who become obese at a young age seem to carry a particularly high risk of eventually developing T2DM (Strati et al., 2024). Part

of the reason is that excess adipose tissue releases large amounts of free fatty acids, glycerol, and inflammatory cytokines, all of which can interfere with insulin signaling (Wondmkun, 2020). In our study, a clear age gap separated the two patient groups. The obese non-diabetics tended to be younger (mean 32.70 ± 8.11 years), while obese diabetics were older (mean 50.43 ± 9.55 years). This fits well with the idea that prolonged exposure to obesity gradually wears down β -cell function and worsens insulin resistance over time (Ruze et al., 2023). Aging itself brings changes in body composition and mitochondrial function that compound the problem (Chao et al., 2020).

Our data confirmed that obese diabetic patients were significantly older than non-diabetic obese patients and controls ($P = 0.001$). Siddiqui et al. (2024) reported similar findings, with a mean age of 51.6 ± 12.4 years among their T2DM patients. We also found that 60.0% of the obese T2DM group were in their 40s or 50s, a pattern that mirrors the observations of Alhabbo et al. (2018), who noted that 63% of T2DM patients in Northern Iraq fell in the 40–59 age range. This probably reflects the cumulative toll of poor dietary habits and low physical activity—problems that are becoming more common in Iraqi society.

With respect to gender, our groups were reasonably well matched ($P = 0.665$ for gender distribution). We deliberately tried to balance male and female enrollment to minimize confounding. That said, women made up 60.0% of both obese groups. While this difference was not statistically significant, it is worth noting because higher obesity rates in women have been reported in several regional studies.

Several factors might explain the higher proportion of obese females. Menopause, for instance, leads to hormonal shifts that promote visceral fat deposition and worsen insulin sensitivity (Kodoth et al., 2022). Lifestyle factors like reduced physical activity and multiple pregnancies may also play a role.

Cultural attitudes matter too. In parts of Iraq and the broader Middle East, a heavier body weight in women is sometimes viewed positively—as a sign of good health or social standing. This can discourage weight management efforts. Alhabbo et al. (2018) described similar patterns in Iraqi populations. Estrogen changes during menopause also promote fat redistribution, further raising diabetes risk in women (Silitonga et al., 2019).

By contrast, Western studies tend to show more balanced diabetes rates between sexes, or even a slight male preponderance (Kautzky-Willer et al., 2023). The differences likely reflect varying lifestyles, occupational activity levels, and cultural norms.

HbA1c forms when glucose attaches to hemoglobin in a non-enzymatic process. Because red blood cells live for about two to three months, HbA1c provides a useful snapshot of average blood sugar over that period. It is widely used to diagnose diabetes and to track how well patients are managing their blood sugar (Majid et al., 2018).

We found that HbA1c was significantly higher in obese diabetics than in the other two groups ($P < 0.001$). This is in line with earlier work showing elevated HbA1c in diabetic populations (Sherwani et al., 2016). The fact that obese non-diabetics had HbA1c levels similar to healthy controls suggests that, at least early on, obesity alone does not necessarily disrupt glucose regulation—at least not in all individuals. This is consistent with the concept of "metabolically healthy" obesity (Jung and Choi, 2014).

HbA1c monitoring is central to clinical decision-making in diabetes care (Cefalu et al., 2022). A Pakistani study found a mean HbA1c of 8.9% in diabetic patients (Siddiqui et al., 2024), which is quite close to the 8.72% we observed in our obese diabetic group.

When adipose tissue expands beyond a certain threshold, it becomes stressed and inflamed, setting off a chain of metabolic disturbances that increase the risk for cardiovascular disease, fatty liver, and metabolic syndrome (Sakers et al., 2022). Dyslipidemia is one of the hallmarks of this process, typically featuring elevated triglycerides, high LDL, and low HDL.

In our study, cholesterol was significantly higher in both obese groups relative to controls ($P = 0.001$), consistent with the findings of Mohamed et al. (2019). Triglycerides were also elevated, particularly in the diabetic subgroup, similar to what Shahid and Sarwar (2020) reported. The mechanism is fairly straightforward: when insulin is deficient or ineffective, lipoprotein lipase activity drops, free fatty acid levels rise, and the liver ramps up VLDL production (Zhang et al., 2022). Reduced LDL receptor expression in the liver—another consequence of insulin deficiency—means that LDL is cleared less efficiently (Alhabbo et al., 2018).

The lipid data painted a consistent picture of dyslipidemia in both obese groups: elevated LDL and VLDL, suppressed HDL. This kind of lipid profile is a well-known risk factor for atherosclerosis. Excess visceral fat disrupts insulin signaling in the liver and muscle, feeding a vicious cycle of metabolic deterioration (Wang, 2025).

Both increased hepatic VLDL synthesis and reduced receptor-mediated clearance contribute to the elevated VLDL and LDL we observed (Abed and Hamid, 2022). The accelerated breakdown of HDL particles in the setting of hypertriglyceridemia explains the low HDL levels (Farhan et al., 2023). Saber et al. (2021) reported a somewhat different pattern in their study—LDL was not significantly different between groups, but VLDL was—underscoring the heterogeneity of lipid disturbances in metabolic disease.

TNF- α is mainly produced by adipocytes and tissue macrophages. It triggers inflammation through reactive oxygen species and activates inflammatory transcription pathways (Kunz et al., 2021). Its role in insulin resistance is well documented, and it is thought to be a key molecular bridge between obesity and diabetes (Kadhim and Shakir, 2020). One way TNF- α disrupts glucose metabolism is by suppressing GLUT4 expression in muscle and fat (Alzamil, 2020).

We found that TNF- α was elevated in both obese groups relative to controls, and that higher TNF- α levels went hand in hand with worse glycemic markers. This supports the notion that chronic inflammation contributes to diabetes pathogenesis (Alewi and Hatem, 2025). Mechanistically, persistent TNF- α exposure leads to IRS-1 serine phosphorylation in muscle and fat tissue, blunting insulin action (Alzamil, 2020). Similar results have been reported by several groups (Akash et al., 2018; Riaz et al., 2020).

The obese diabetic group had higher TNF- α than controls, which was expected and consistent with Rohm et al. (2022). TNF- α breaks down adipose tissue while also blocking insulin action—a dual effect that helps drive the transition from obesity to frank diabetes (Alzamil, 2020).

Not all studies agree, however. Al-Shukaili et al. (2013) actually found lower TNF- α in their T2DM patients. They attributed this to the fact that most of their patients had had diabetes for more than five years, their sample size was small, and the groups were not perfectly matched for age and sex.

The ROC analysis for TNF- α was encouraging. For the obese-versus-control comparison, the AUC was 0.869 (95% CI: 0.770–0.963), with both sensitivity and specificity at 86.7% using a cutoff of >158.21 pg/mL. These numbers suggest that TNF- α could be clinically useful for detecting obesity-related inflammation.

The picture was less clear-cut for the obese diabetic group, where the AUC dropped to 0.710 (95% CI: 0.581–0.839), with sensitivity of 73.3% and specificity of 70.0%. This is not surprising. In diabetes, many factors beyond TNF- α —hyperglycemia, oxidative stress, advanced glycation products—feed into the inflammatory milieu, which makes it harder for any single marker to stand out. Still, TNF- α retained acceptable diagnostic value even here, and elevated cytokines in early diabetes may help predict who will progress (Bashir et al., 2020).

IL-6 was also significantly elevated in both obese groups. The obese non-diabetics had the highest levels (149.52 ± 16.41 pg/mL), followed by the obese diabetics (128.57 ± 11.20 pg/mL), with controls lowest at 102.58 ± 11.40 pg/mL. The slightly lower IL-6 in diabetics might reflect ongoing treatment or differences in disease duration. Dutta et al. (2024) observed a similar pattern.

Abdulrhman et al. (2024) also found higher IL-6 in diabetic patients, and El-Mikkawy et al. (2020) showed a positive correlation between IL-6 and BMI in obese subjects.

Najim and Aboud (2022) reported that IL-6 correlated with BMI only in the morbidly obese, suggesting a dose–response relationship with fat mass. At the molecular level, high blood glucose pushes monocytes to produce more IL-6 through the PKC, p38 MAPK, and NF- κ B pathways (Nashtahosseini et al., 2025).

The IL-6 ROC analysis gave an AUC of 0.847 (95% CI: 0.751–0.943) at a cutoff of >114.40 pg/mL, with sensitivity of 86.7% and specificity of 83.3%. This makes IL-6 a reasonably strong diagnostic marker for obesity-related inflammation.

In the diabetic subgroup, the IL-6 AUC was 0.775, with sensitivity 76.7% and specificity 80.0% at a cutoff of >114.00 pg/mL. Like TNF- α , the performance was somewhat attenuated in diabetics, probably because diabetes introduces additional confounders.

Abdulrhman et al. (2024) reported that in a separate cohort of 70 diabetic patients, an IL-6 cutoff of 72.46 pg/mL predicted complications with 97.1% sensitivity, 87.1% specificity, and an AUC of 0.984. High IL-6 has been independently linked to T2DM risk and β -cell dysfunction (Akbari and Hassan-Zadeh, 2018).

Taken together, our IL-6 findings point to this cytokine as a useful marker of inflammatory burden in obese and diabetic patients, and possibly as a target for future therapeutic strategies.

Logistic regression revealed a direct relationship between IL-6 and TNF- α in both patient groups. This is consistent with the idea that these cytokines are co-regulated in inflamed adipose tissue and act together to amplify the metabolic damage (Varthaliti et al., 2025).

TNF- α was positively correlated with HbA1c in the obese non-diabetic group ($r = 0.457$, $P = 0.011$), meaning that higher TNF- α tracked with poorer glycemic control. Alewi and Hatem (2025) and Ullah et al. (2021) found the same thing.

In the diabetic group, IL-6 correlated with HbA1c ($r = 0.349$, $P = 0.039$). This suggests that chronic hyperglycemia and IL-6-driven inflammation feed into each other in an ongoing cycle.

CONCLUSION

This study shows that obesity—particularly when combined with T2DM—is accompanied by elevated serum TNF- α and IL-6. Both cytokines correlated with HbA1c, linking inflammation to glycemic dysregulation. These results suggest that TNF- α and IL-6 could serve as practical biomarkers for identifying patients at higher metabolic risk, and that anti-inflammatory approaches might have a place in the management of obesity-related diabetes.

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