Original Research Article

# **EVALUATION OF SERUM IL-10,** TNF-α **AND ALBUMIN LEVELS AMONG MALE TYPE 2 DIABETES MELLITUS PATIENTS IN A TERTIARY HOSPITAL IN NIGERIA**

**ABSTRACT**

Type 2 diabetes (T2DM) is associated with a chronic inflammatory state and a disturbance in cytokine balance. This study aimed to evaluate the levels of TNF-α, IL-10, and albumin in the serum of males with type 2 diabetes compared to healthy individuals. A case-control study was conducted involving 60 participants (30 patients and 30 healthy individuals), and the samples were analyzed using ELISA techniques and the BCG method. The results showed a significant increase in TNF-α (53.97 ± 10.22 Vs 15.11 ± 2.82) and IL-10 (17.07 ± 1.42 Vs 4.12 ± 0.88) levels among the patients (P = <0.001), with no significant difference in albumin levels (p>0.05). The results indicate the presence of a simultaneous inflammatory and anti-inflammatory response, which underscores the importance of these cytokines as biomarkers in diabetes monitoring.

**KEY WORDS:** Diabetes mellitus, Inflammation, Biomarkers, TNF-α, IL-10, Serum albumin, male T2DM patients.

**INTRODUCTION**

Type 2 Diabetes Mellitus (T2DM) formerly known as non-insulin-dependent Diabetes Mellitus is the most common form of Diabetes Mellitus characterized by hyperglycemia, insulin resistance and relative insulin deficiency (Agarwal *et al*., 2024; Ogbodo *et al*., 2019). Individuals living with T2DM are more susceptible to various forms of both short- and long-term complications, which often lead to their premature death (Chijioke *et al*., 2010). This tendency of increased morbidity and mortality seen in patients with T2DM especially in resource-poor developing countries like Nigeria has been linked to rapid lifestyle changes, population growth, and increased longevity due to increasing urbanization and industrialization (Flood *et al*., 2021; Hill-Briggs *et al*., 2020). Globally, the prevalence of T2DM has reached alarming proportions, with significant regional variations influenced by socioeconomic factors, healthcare access, and urbanization (Pinchevsky *et al*., 2020). Type 2 Diabetes Mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia and associated with systemic inflammation (Młynarska *et al*., 2025). Elevated levels of pro-inflammatory cytokines, such as Tumor Necrosis Factor-alpha (TNF-α), and altered levels of anti-inflammatory cytokines like Interleukin-10 (IL-10), have been implicated in the pathogenesis of T2DM. Studies have demonstrated that T2DM patients exhibit significantly higher serum TNF-α concentrations compared to healthy individuals, suggesting a state of heightened inflammation (Chen *et al*., 2017; Akash *et al*., 2018). Tumour necrosis factor alpha is a cytokine that has pleiotropic effects on various cell types. It has been identified as a major regulator of inflammatory responses and is known to be involved in the pathogenesis of some inflammatory and autoimmune diseases (Jang *et al.,* 2021).

Conversely, IL-10, an anti-inflammatory cytokine, plays a crucial role in modulating immune responses and maintaining metabolic homeostasis (Saraiva *et al*., 2020). Dysregulation of IL-10 has been associated with the progression of T2DM and its complications (Islam *et al*., 2025; Novianti and Nur'aeny, 2024; Ayelign *et al*., 2021). According to Iyer & Cheng (2012), interleukin 10 is a cytokine with potent anti-inflammatory properties that plays a central role in limiting host immune response to pathogens, thereby preventing damage to the host and maintaining normal tissue homeostasis. Dysregulation of IL-10 is associated with enhanced immunopathology in response to infection as well as an increased risk for the development of many autoimmune diseases.

Albumin, a major serum protein, is essential for maintaining oncotic pressure and serves as a marker of nutritional and inflammatory status. Alterations in serum albumin levels have been observed in T2DM patients, with studies indicating a negative correlation between albumin levels and the presence of diabetic complications, such as retinopathy (Alhalwani *et al*., 2023; Hui *et al*., 2023; Wang *et al*., 2022; Zhang *et al*., 2022).

The interplay between these biomarkers reflects underlying pathophysiological processes in T2DM, including insulin resistance, beta-cell dysfunction, and systemic inflammation. Elevated TNF-α contributes to insulin resistance by interfering with insulin signaling pathways, while reduced IL-10 levels may exacerbate inflammatory responses, further impairing insulin sensitivity. Changes in albumin levels can indicate systemic inflammation and are associated with the severity of diabetic complications. Despite the global prevalence of T2DM, there is a paucity of data regarding the behavior of these biomarkers in male T2DM patients in Nigeria. This present study aims to unearth the relationship between the serum levels of the cytokines (interleukin-10, TNF-alpha) and albumin in correlation with type 2 diabetes mellitus in male subjects.

This study employed a case and control study design involving male subjects with Type 2 Diabetes Mellitus in Federal Medical Centre, Asaba. Thus, understanding the specific patterns of TNF-α, IL-10, and albumin levels in this population is crucial for early diagnosis, risk stratification, and monitoring of therapeutic responses. This study aims to fill this research gap, providing insights that could lead to personalized treatment strategies, ultimately improving clinical outcomes and informing public health policies in Nigeria.

MATERIALS AND METHODS

## **Study Site**

The research was conducted at the Federal Medical Centre (FMC) Asaba, Delta State, Nigeria, a tertiary healthcare facility with a well-equipped laboratory capable of conducting biomarker assays relevant to Type 2 Diabetes Mellitus.

## **Study Design**

This study adopted a case-control design to assess the serum levels of TNF-α, IL-10, and albumin in male T2DM patients. A total of sixty (60) participants were randomly recruited, with 30 individuals diagnosed with T2DM serving as test subjects and 30 healthy individuals serving as controls. Informed consent was obtained from all participants, and each subject completed a structured questionnaire capturing demographic data (such as age, gender), and clinical information.

## **Sample Size Determination**

The sample size for this study was calculated using the documented prevalence of diabetes mellitus in Nigeria which was found to be 1.7% (International Diabetes Federation*,* 2017). The sample size for this study was calculated using the formula described by Charan and Biswas (2013);

N = $\frac{Z^{2}p(1-p)}{d^{2}}$

Where:

N = Required sample size when population of the facility is large

Z = The standard normal deviate, set at 1.96 which corresponds to the 95% confidence level

p = Estimated prevalence of diabetes mellitus in Nigeria which was found to be 1.7% (International Diabetes Federation*,* 2017)

d = Degree of accuracy required which set at 0.05 (margin of error at 5%)

Applying the method,

N = $Z^{2}$ x p x (1-p)/$d^{2}$

N = $\frac{1.96^{2} x 0.017(1-0.017)}{(0.05)^{2}}$

N = $\frac{0.0653072(1-0.017)}{0.0025}$

N = 25.7 ̴ N = 26

N = 26 minimum sample sizes

60 subjects were used for this study; 30 subjects served as control subjects while the other 30 type 2 diabetic male patients served as the test group.

**Inclusion Criteria**

The study included male subjects who were up to the age of 18 and above (those with and without diabetes mellitus). Those with T2DM served as the test group while subjects without diabetes mellitus which served as controls.

**Exclusion Criteria**

1. The study excluded subjects that were below the age of 18, male subjects with or without diabetes mellitus and other forms of diabetes mellitus.
2. Individuals with chronic inflammatory diseases, autoimmune disorders (e.g., rheumatoid arthritis), chronic infections (e.g., HIV, hepatitis), active malignancies, obesity, etc as well as active smokers were excluded from the study.
3. Use of corticosteroids or nonsteroidal anti-inflammatory drugs (NSAIDs) within the past six months was also an exclusion criterion due to their potential immunomodulatory effects.

## **Ethical Considerations**

The study adhered to ethical guidelines outlined in the Declaration of Helsinki (Puri *et al*., 2009). Ethical approval was obtained from the Ethics Committee of the Federal Medical Centre Asaba, Delta State, Nigeria. All participants provided informed consent before enrollment. Confidentiality was strictly maintained, and participants were assigned unique identification numbers to anonymize data. No financial compensation was provided to participants to minimize coercion. Participants had the right to withdraw at any stage without consequences.

## **Informed Consent**

Prior to participation, informed consent was obtained from all subjects. Each participant completed a questionnaire designed to capture relevant demographic and clinical information.

## **Sample Collection and Handling**

Fasting venous blood samples (5 mL) were collected using standard venepuncture techniques and dispensed into plain tubes. Proper sample handling procedures were implemented to prevent biomarker degradation. Samples were immediately placed on ice and centrifuged at 3000 rpm for 10 minutes within one hour of collection to separate serum. Aliquots were stored at −20°C until analysis to preserve biomarker stability and prevent degradation. Quality control measures included the use of positive and negative controls, adherence to manufacturer protocols, and strict compliance with standard operating procedures.

**Determination of Cytokine Levels Tumor necrosis factor-alpha (TNF-α)**

The sandwich enzyme linked immunosorbent assay (sandwich-ELISA) method was used for the determination of tumor necrosis factor-alpha and interleukin-10 levels in the participants’ sera.

**Estimation of Serum Albumin**

This was determined using the Bromocresol green method (BCG), as described by Doumas and Watson (1971) and cited by Okpogba *et al*. (2021).

**Statistical Analysis**

Data were analyzed using IBM SPSS Statistics (version 25.0) and presented using mean $\pm $ standard deviation. For comparison of continuous variables and testing associations of biomarkers with others, we have used t-tests and Pearson analysis respectively. Significance was assumed at p < 0.05.

**RESULTS**

The analysis of the cytokine levels (TNF-α and IL-10) revealed significant differences between the test and control groups. The mean concentration of tumor necrosis factor-alpha was significantly elevated in the male T2DM patients test group (53.97 ± 10.22 Vs 15.11 ± 2.82; p < 0.001), suggesting a heightened inflammatory response in individuals diagnosed with T2DM. Similarly, the mean serum level of IL-10 was significantly higher in the test group than in the control group (17.07 ± 1.42 Vs 4.12 ± 0.88; p < 0.001), indicating a potential compensatory anti-inflammatory mechanism in the male T2DM patients. Conversely, no significant difference in serum albumin level was observed between the test and control groups (p = 0.267). Furthermore, the test group exhibited no significant differences in the mean age compared to the control group (p = 0.705). (See table 1).

**Table 1: Comparison of TNF-α, IL-10, Albumin, and Mean Age Between Test (male T2DM patients) and Control Groups**

| Variables | Test Group (Mean ± SD) | Control Group (Mean ± SD) | t-test | p-value |
| --- | --- | --- | --- | --- |
| TNF-α (pg/ml) | 53.97 ± 10.22 | 15.11 ± 2.82 | 17.216 | <0.001 |
| IL-10 (pg/ml) | 17.07 ± 1.42 | 4.12 ± 0.88 | 36.662 | <0.001 |
| Albumin (g/L) | 34.52 ± 5.43 | 36.14 ± 4.07 | -1.124 | 0.267 |
| Age (years) | 56.09 ± 4.32 | 54.86 ± 3.25 | 0.968 | 0.705 |

\* *Statistically significant at p < 0.05; SD = Standard deviation.*

Correlation analysis among TNF-α, IL-10, albumin, and age in the test group (male T2DM patients) revealed no statistically significant associations among the measured variables (p > 0.05). Specifically, TNF-α did not exhibit a significant correlation with IL-10 (r = -0.124, p = 0.574), albumin (r = -0.130, p = 0.554), or age (r = 0.030, p = 0.890). Similarly, IL-10 was not significantly associated with albumin (r = 0.332, p = 0.122) or age (r = 0.224, p = 0.304). See table 2.

**Table 2: Correlation Analysis in Male T2DM Patients**

| Variables | Statistics | TNF-α | IL-10 | Albumin | Age |
| --- | --- | --- | --- | --- | --- |
| TNF-α | r  | - | -0.124 | -0.130 | 0.030 |
|  | p-value | - | 0.574 | 0.554 | 0.890 |
| IL-10 | r  | -0.124 | - | 0.332 | 0.224 |
|  | p-value | 0.574 | - | 0.122 | 0.304 |
| Albumin | r  | -0.130 | 0.332 | - | 0.244 |
|  | p-value | 0.554 | 0.122 | - | 0.262 |
| Age | r  | 0.030 | 0.224 | 0.244 | - |
|  | p-value | 0.890 | 0.304 | 0.262 | - |

\* *Statistically significant at p < 0.05*

In contrast to the test group, correlation analysis among control subjects revealed a significant negative correlation between IL-10 and age (r = -0.521, p = 0.013), suggesting that IL-10 levels declined with increasing age in healthy individuals. However, no significant correlations were found between TNF-α and IL-10 (r = 0.201, p = 0.370), albumin (r = 0.245, p = 0.273), or age (r = 0.204, p = 0.362). Likewise, albumin did not exhibit a significant correlation with IL-10 (r = -0.259, p = 0.245) or age (r = 0.194, p = 0.387). These findings suggest that, unlike T2DM patients, the regulatory mechanisms influencing inflammatory cytokine levels in healthy individuals may be influenced by age-related physiological changes rather than metabolic dysregulation.

**Table 3: Correlation Analysis in Male Control Subjects**

| Variables | Statistics | TNF-α | IL-10 | Albumin | Age |
| --- | --- | --- | --- | --- | --- |
| TNF-α | r  | - | 0.201 | 0.245 | 0.204 |
|  | p-value | - | 0.370 | 0.273 | 0.362 |
| IL-10 | r  | 0.201 | - | -0.259 | -0.521\* |
|  | p-value | 0.370 | - | 0.245 | 0.013\* |
| Albumin | r  | 0.245 | -0.259 | - | 0.194 |
|  | p-value | 0.273 | 0.245 | - | 0.387 |
| Age | r  | 0.204 | -0.521\* | 0.194 | - |
|  | p-value | 0.362 | 0.013\* | 0.387 | - |

\**Statistically significant at p<0.05.*

## **DISCUSSION**

In the current study, we found significantly elevated levels of tumor necrosis factor-alpha and interleukin-10 in the patients compared to the control group, indicating a dysregulated inflammatory response in individuals with Type 2 Diabetes Mellitus. This finding aligns with previous research demonstrating increased pro-inflammatory cytokines in T2DM patients (Bashir *et al*., 2022; Akash *et al*., 2018). The significantly elevated TNF-α levels in patients reinforce the cytokine's role in insulin resistance as TNF-α is known to impair insulin and contribute to chronic low-grade inflammation (Bashir *et al*., 2020). TNF-α has been implicated in the disruption of insulin signaling via serine phosphorylation of insulin receptor substrate-1 (IRS-1), thus exacerbating hyperglycaemia (Lee *et al*., 2022).

Furthermore, the increased IL-10 levels observed in the male test subjects in this study contradict earlier studies suggesting an anti-inflammatory role of IL-10 (Islam *et al*., 2022). This paradoxical increase may indicate a compensatory response to heightened inflammatory burden in T2DM (dos Santos Haber *et al*., 2023).

Notably, albumin levels remained comparable between groups (p > 0.05). This is in keeping with the reports of Analike *et al*. (2019) that documented no significant alterations in diabetic patients compared to control subjects. Nevertheless, some other studies showed lower serum albumin levels in diabetic patients than in control group (Ubani *et al*., 2024; Junaid *et al*., 2022; Chang *et al*., 2019). The disparity in the results by these various studies may be due to variations in the durations of diabetes mellitus patients recruited in the different studies as well as the differences in the sample sizes employed.

The lack of significant correlations in this study may be attributed to sample size limitations. In male control subjects, a significant negative correlation between IL-10 and age was observed, indicating that IL-10 levels declined with increasing age in healthy individuals. This aligns with studies suggesting that aging is associated with a decrease in anti-inflammatory cytokines, potentially contributing to a pro-inflammatory state (Navarro-González *et al*., 2011). However, no significant correlations were found between TNF-α and IL-10, albumin, or age, suggesting that in the absence of metabolic dysregulation, these biomarkers may not be interrelated. Notably, the study did not assess correlations in female subjects, limiting the ability to assess sex-based differences in these associations. Previous research has indicated that inflammatory responses and cytokine profiles may differ between sexes, potentially influencing T2DM pathogenesis and progression (Panagi *et al*., 2019). Future studies should include sex-specific analyses to elucidate these potential differences.

Potential limitations included the small sample size, which may limit generalizability. Additionally, only male participants were included, precluding sex-based comparisons. Biomarker levels were assessed at a single time point, limiting insights into temporal changes in inflammation and albumin levels. Future studies should incorporate longitudinal designs with larger, more diverse populations to enhance generalizability and causal inference.

**CONCLUSION**

The study highlights the significant role of inflammatory markers in Type 2 Diabetes Mellitus, with elevated TNF-α and IL-10 levels observed in male T2DM patients, indicating a complex interplay between pro- and anti-inflammatory responses. While albumin levels did not differ significantly between groups, the negative correlation between IL-10 and age in male controls suggests age-related declines in anti-inflammatory activity.

Disclaimer (Artificial intelligence)

Option 1:

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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