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*Article*

# Estimation Of Some Biochemical Parameters In Type -2 Diabetic Patients After Pfizer Vaccination Among Patients From Iraqi

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**Abstract:** This study aimed to assess the changes in biochemical markers in type 2 diabetic patients after receiving the Pfizer COVID-19 vaccine. The research focused on measuring cardiac biochemical markers, specifically myoglobin and lipid profiles, as well as routine kidney function tests (blood urea and serum creatinine) following the second dose of the Pfizer vaccine. Myoglobin levels were estimated using an ELISA kit, while lipid profiles and renal function were determined using the Cobas analyzer. The results indicated higher myoglobin levels in both vaccinated and non-vaccinated diabetic groups compared to healthy controls. There were significant differences in lipid profiles between diabetic groups, although no significant differences were found between vaccinated and non-vaccinated groups. HDL levels were lower in both diabetic groups compared to healthy controls. The study found no significant adverse effects of the Pfizer vaccine on kidney function tests, with T2DM patients showing higher levels of myoglobin and lipid profiles but no significant difference between vaccinated and non-vaccinated individuals.

**Keywords:** Type 2 Diabetes Mellitus (T2DM), Myoglobin, Pfizer COVID-19 Vaccine, Lipid Profile, Kidney Function, Biochemical Markers..

## 1. Introduction

Diabetes mellitus is in increasing incidence and often results in significant metabolic problems and dire outcomes. The new COVID-19 is a highly infectious diseases, caused by (SARS-CoV-2) (1). T2DM appears to be a risk factor for acquiring the novel coronavirus infection. Diabetes is correlated with a (two-fold) increase in severity and mortality of COVID-19 in individuals with COVID-19 as compared to non-diabetic(2). The Pfizer vaccines are mRNA-based and are approved for emergency use(3). Use of the Pfizer vaccine and other vaccinations against the

COVID-19 resulted in a significant decrease in coronavirus disease mortality of (COVID-19). The Pfizer vaccine was not linked to a higher risk of the most of the adverse events analyzed. The vaccine was linked to an increased risk of myocarditis (1 to 5 events per 100,000 persons) (4). After COVID-19 infection, the risk of this potentially serious adverse event as well as many other potentially serious adverse events was significantly increased. Myoglobin is an iron and oxygen-binding protein that is essential for the backup of oxygen in skeletal and cardiac muscles. Myoglobin is released in response to

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inflammation signals and can be rapidly excreted into the bloodstream (5).The aim of this study to see change in biochemical marker after second dose of Pfizer vaccination .

## 2. Materials and Methods

### Material and Methods

Ethical approval

Written consent in writing had been obtained from all participants before getting blood for this investigation.

Topics and methods of gathering

Participants in the research were individuals with T2DM who had been diagnosed by an endocrine disease expert. The patient got two doses of the Pfizer vaccination.

Criteria for inclusion

1. One hundred people with diabetes classified as type 2 mellitus, groups 20 to 70, received Pfizer vaccines.21 days following the second dose of immunization to assess the Pfizer vaccine's effectiveness
2. 50 Type -2 diabetes mellitus with no vaccine age ranged (20-70 years).
3. 50 normal control, vaccinated with p-fizer vaccine age ranged (25-50 years). to examine efficacy of p-fizer vaccine The whole time of study was 4 months from (October 2022 to December 2023).

## 3. Results and Discussion

Criteria for exclusion

Patients with autoimmune disorders, liver and kidney disorders, those who have had just one shot of the Pfizer vaccine, those who have received a Sinopharum or AstraZeneca vaccination, and those who have already contracted COVID-19 are all ineligible.

Preparation specimens and testing of products

Two components were recovered from 4 milliliters of venous blood: whole blood (measured by HPLC), the HbA1C test. Serum for detection of lipid profile and kidney function test. Kidney function test and Lipid profile were estimated by Cobas c.

Percentage of HbA1c (%) in Studied Group

The results summarized in table (1-1) showed a higher percentage of HbA1C in T2DM (8.027 ± 0.18) and (8.321±0.2) (%),(Vaccinated and non vaccinated respectively, higher than the healthy control mean (4.647±0.09). Significant difference in p-value is shown between T2DM (vaccinated and Non vaccinated) compared to healthy control p-value (0.0001 ,0.0001) repetitively.Non –Significant differences are shown between T2DM vaccinated and T2DM non vaccinated in P-Value(0.720).

Table (1-1): Comparison mean of HbA1c (%) According to studied Group patients T2DM and control

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DM Vaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| HbA1C (%) | Controls | 50 | 4.647 | 0.6500 | 4.5 | 6.1 | | 0.720 |
| | T2DM non vaccinated | 50 | 8.321 | 1.748 | 6.5 | 13.5 | <0.0001 | |
| | T2DM vaccinated | 100 | 8.027 | 1.758 | 6.5 | 14.5 | <0.0001 | |

These findings match those of Ahmed *et al.,* who discovered that glycated hemoglobin levels in diabetes patients were significantly higher than in healthy control (6). Another study represents that there was a higher level of FSG and HbA1C in T2DM when compared with healthy controls (7). The results agree with Hasanato, who investigated controls with HbA1c less than 6.5 % and diabetics with HbA1c greaterthan 6.5 %(8)

**Level of Myoglobin (ng/ml) among Studied Group**

Data presented in table (1-2) were of myoglobin in T2DM (78.63±14.12) (75.57±8.703) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (70.57±5.69). Significant difference in p-value was shown between T2DM (vaccinated compared to healthy control p-value (0.0002) and Non-Significant differences were shown between T2DM T2DM non vaccinated and healthy control in P-Value (0.0677).Non-Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.2704).

| Variables | Study Groups | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DMVaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| Myoglobin (60-85) ng/mL | Controls | 50 | 70.57 | 5.69 | 60 | 82.68 | | 0.2704 |
| | T2DM non vaccinated | 50 | 75.57 | 8.703 | 60.30 | 108.7 | 0.0677 | |
| | T2DM vaccinated | 100 | 78.63 | 14.12 | 71 | 128.1 | 0.0002 | |

The current results represented normal mean for all groups' normal values of myoglobin (60-85) ng/mL. Myoglobin levels in T2DM revealed higher levels of myoglobin in comparison with healthy controls. A study by Sen et al mentioned that higher level of myoglobin due to esterase activity being found in heme proteins such as hemoglobin and

myoglobin. The esterase activity assessed from the hydrolysis of p-nitro phenylacetate (p-NPA) in pure hemoglobin from normal people and diabetic patients was greater in the diabetic condition and increased gradually with the degree of the diseases. HbA1c, the predominant glycated hemoglobin that rises in proportion to blood glucose levels in diabetes mellitus, displayed more esterase activity than the non-glycated hemoglobin fraction. In vitro-glycated myoglobin (GMb) furthermore, had stronger esterase activity than its non-glycated counterpart (Mb). The increased esterase activity of hemoglobin and myoglobin may be related to glycation-induced structural changes in the proteins (9). Myoglobin is nephrotoxic and can precipitate in the renal tubules, causing acute kidney damage, which can occur in up to one-third of patients(10). The results of our study showed no effect of myoglobin because all vaccinated individuals in this study showed normal kidney tests (B. urea and S. creatinine) after the second dose of vaccination.

Level of Serum Total Cholesterol (mg/dL) among Studied Group

Consideration to the mean serum, total cholesterol in table (1-3) was higher level of total cholesterol in T2DM (181.07±36.92) (178.84±41.922) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (124.61± 28.128). Significant difference was shown in p-value between T2DM (vaccinated and Non vaccinated) compared to healthy control p-value (0.0001, 0.0001) repetitively. Non –Significant differences between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.372).

Table (1-3): Comparison of S.Cholesterol in T2DM and Healthy Control

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DM Vaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| **Total Cholesterol (mg/dL)** | Controls | 50 | 124.61 | 28.128 | 84.8 | 189 | | 0.372 |
| | T2DM non vaccinated | 50 | 178.84 | 41.922 | 102 | 300 | <0.0001 | |
| | T2DM vaccinated | 100 | 181.07 | 36.92 | 101 | 290 | <0.0001 | |

The current study's findings revealed that higher level of cholesterol in both groups of T2DM but T2DM vaccinated slightly higher than T2DM non vaccinated in cmparison with healthy control . The Current study agree with Simonen et al study that demonstrate Cholesterol biogenesis was high in T2DM patients, cholesterol metabolism different from that of nondiabetic (11). Present findings are consistent with Parcero-Valdés who mentioned that Many patients with T2DM have dyslipidemia, which is essential in the rising of cardiovascular (CV) risk. Lipids and glucose play a crucial role in energy metabolism. It is well known that patients with diabetes often have dyslipidemia, characterized by increased triglycerides, low high-density lipoprotein cholesterol (HDL-c), a predominance of small-dense low-density lipoprotein (LDL) particles (12).A higher level of lipid in T2DM may

be due to a number of variables, including insulin resistance and relative insulin deficit, adipocytokines (e.g., adiponectin) ,and hyperglycemia may contribute to alterations in lipid metabolism in T2DM patients(13). Pfizer vaccine contains the excipients (4-hydroxybutyl)(azanediyl) (hexane6,1diyl) (2-hexyl decanoate), (2- cholesterol)(14). According to the public health of canada the chemical components of the lipid nanoparticle that supplies mRNA to cells : 2 [(polyethylene glycol)-2000], N-ditetradecylacet-amide It forms a protective layer around the nanoparticle, improving storage stability and reducing non-specific protein binding, 1,2-distearoyl-sn-3-phos-phocholine (DSPC) Part of the lipid bilayer (double layer of lipids) that forms the nanoparticle .Cholesterol provides structural support for the nanoparticle's lipid bilayer and promotes lipid component mobility, (4-hydroxybutyl) azanediyl) bis(hexane-6,1-diyl) bis(2-hexyldecanoate). The primary component of the lipid-nanoparticle delivers mRNA into the cell. Cationic (charged positively) lipid during the manufacturing process, it encourages the nanoparticle to assemble into a virus-sized particle with the negatively charged mRNA in the center, and it facilitates mRNA release from the nanoparticle once inside the cell. All lipid component safe to the cell except For the polyethylene glycol (PEG) which may lead to allery . The present data showed no effect of Pfizer vaccine on T2DM and healthy control after the second dose of vaccination .

Level of Serum Triglyceride(mg/dL) among Studied Group

Data demonstrated in table (4-11) represented ahigher level of triglyceride in T2DM (170.34±39.342) (176.50±54.017) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (78.20± 30.175). Significant difference in p-value was shown between T2DM (vaccinated and Non vaccinated) compared to healthy control p-value (0.0001, 0.0001) repetitively.Non –Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0. 721).

Table (1-4): Comparison of S.Triglyceride in T2DM and Healthy Control

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DM Vaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| **Triglyceride (mg/dL)** | **Controls** | 50 | 108.60 | 30.175 | 72 | 301.0 | | 0.721 |
| | **T2DM non vaccinated** | 50 | 176.50 | 54.017 | 90 | 368.5 | <0.0001 | |
| | **T2DM vaccinated** | 100 | 170.34 | 39.342 | 94 | 270 | <0.0001 | |

Thapa et al documented elevated triglyceride levels to diabetes and obesity's poor glycemic control. Improvement in glycemic management had been linked to a decrease in TG levels. The increase in triglycerides in poorly regulated individuals was linked to a decrease in activities (15). In normal individuals, insulin

allows glucose to be acquired by peripheral tissues while restricting glucose production through the kidney and liver (16).

Adipose tissue insulin resistance is defined by insufficient lipolysis in adipocytes, whereas hepatic insulin resistance is defined by a diminished capacity to limit hepatic glucose synthesis in the context of active lipogenesis (17). As a result, dysfunctional signaling in adipose tissue is the primary mechanism implicated in the progression of insulin resistance and diabetics.Chronic hyperinsulinemia is caused by impaired glucose uptake, which activates complicated lipogenic metabolic pathways known as de novo lipogenesis, resulting in the buildup of intracellular lipids in skeletal muscle (18).There is little information available about the potential effects of the new Pfizer vaccine on serum lipoprotein levels. in patients suffering from family history (A case study of hypertriglyceridemia caused by the Pfizer vaccine in a patient with familial hypercholesterolemia)Heterozygous FH (HeFH) male with a history control of elevated TG with medication and LDL apheresis who had very high levels of TG one day following his second Vaccination against COVID-19 (19).Our results represent no effect of the Pfizer vaccine on triglyceride levels after vaccination .

LDL Level (mg/dL) in Studied Group

Data present in table (4-12) revealed a higher level of LDL in T2DM (113.56±38.409) (106±48.944) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (78.20± 30.175). Significant difference in p-value was shown between T2DM (vaccinated and Non vaccinated) compare to healthy control p-value (0.0001, 0.0001) repetitively.Non –Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.7433)

Table (1-5): Comparison of LDL in T2DM and Healthy Control

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DMVaccinated &T2DMnon vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| LDL (mg/dL) | Controls | 50 | 51.508 | 62.48 | 15.2 | 113.4 | | 0.7433 |
| | T2DM non vaccinated | 50 | 106 | 48.944 | 22 | 232 | <0.0001 | |
| | T2DM vaccinated | 100 | 113.56 | 38.409 | 35.7 | 229.4 | <0.0001 | |

The current study's findings showed higher level of LDL in two group of T2DM vaccinated and non vaccinated in compared with healthy controls . These findings are consistent with those of (Artha, *et al*., 2019), who found that lipid profile findings such as total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), and triglycerides (TG) were higher in patients in the poor glycemic control group (p 0.05), and concluded that lipid profiles (LDL-C) and lipid ratios show potential

markers that can be used in predicting glycemic. According to the Framingham Study and the United States National Health and Nutritional Examination Survey, the prevalence of elevated Low density lipoproteins in diabetes patients appeared to be comparable to that of the general population (20).The pathway of LDL production is well understood, and both cholesteryl ester transfer protein (CETP) and hepatic lipase are involved: (1) CETP promotes triglyceride transfer from VLDL1 to LDL; (2) triglyceride-rich LDL is a preferential substrate for hepatic lipase; and (3) enhanced lipolysis of triglyceride-rich LDL leads in the creation of LD(21)

**HDL Level(mg/dL) in Studied Group**

Data presented in table (4-13) revealed a lower level of HDL in T2DM (33.4±8.4536) (31.3 ± 9.3534) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (49 ±6.925). Significant difference in p-value was shown between T2DM (vaccinated and Non vaccinated) compared to healthy control p-value (0.0001, 0.0001) repetitively. Non –Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.654).

**Table (1-6): Comparison of HDL in T2DM and Healthy Control**

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DM Vaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| HDL (mg/dL) | Controls | 50 | 49 | 6.925 | 25 | 63 | | 0.654 |
| | T2DM non vaccinated | 50 | 31.3 | 9.3534 | 20 | 54 | <0.0001 | |
| | T2DM vaccinated | 100 | 33.4 | 8.4536 | 19 | 53 | <0.0001 | |

The study is consistent with Srivastava who found metabolic environment in diabetes mellitus condition such as hyperglycemia-induced advanced glycation end products, oxidative stress, and inflammation promote HDL dysfunction leading to greater risks of CVD. (22).According to study done by Kaysen, various mechanisms were thought to be involved in diabetic aberrant lipid and lipoprotein synthesis, transport, and clearance. Thus, diabetic patients have lower lipoprotein lipase expression, impaired reverse cholesterol transportation, and a lower number of receptors regulating lipid absorption(23).Diabetes worsens lipid profile (blood cholesterol levels) by lowering "good" cholesterol levels in the blood (high-density lipoprotein or HDL),increasing "bad" cholesterol levels in the blood LDL. ,and increasing fat particle levels in the blood

(triglycerides)(24). HDL was not affected by vaccine.

VLDL Level (mg/dL) in Studied Group

Data presented in table (4-14) revealed a higher level of VLDL in T2DM (33.9±7.922) (35.5±11.85) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (21.6±6.94).Significant difference in p-value was shown between T2DM (vaccinated and Non vaccinated) compare to healthy control p-value (0.0001, 0.0001) repetitively Non –Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.3497).

Table (1-7): Comparison of VLDL in T2DM and Healthy Control

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DM Vaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| VLDL (mg/dL) | Controls | 50 | 21.6 | 6.94 | 10 | 50 | | 0.3497 |
| | T2DM non vaccinated | 50 | 35.5 | 11.85 | 16 | 73 | <0.0001 | |
| | T2DM vaccinated | 100 | 33.9 | 7.922 | 18 | 54 | <0.0001 | |

The study agreed with Hirano who demonstrated that elevated serum concentrations of TG-rich lipoproteins, a high prevalence of small (LDL), and low concentrations of cholesterol-rich high-density lipoprotein (HDL) -C characterize atherogenic dyslipidemia in diabetes. An increase in large TG-rich (VLDL) is a central lipoprotein abnormality, and other lipoprotein abnormalities are metabolically linked to increased Triglyceride. Insulin regulates serum VLDL concentrations by suppressing hepatic VLDL production and stimulating VLDL removal via lipoprotein lipase activation. It is still unclear whether hyperinsulinemia, which is used to compensate for insulin resistance, is causally linked to VLDL overproduction

**Blood Urea level among Studied Group**

Blood urea in table (4-18) showed a level of Blood urea in T2DM (26.04±13.64) (25.40± 8.091) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (25.52±7.58) .Non-Significant difference in p-value was shown between T2DM (vaccinated and Non vaccinated) compare to healthy control p-value (0.7335, 0.643) repetitively.Non –Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.135).

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DM Vaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| B.Urea (mg/dL) | Controls | 50 | 24.52 | 7.58 | 16 | 26 | | 0.135 |
| | T2DM non vaccinated | 50 | 25.40 | 8.091 | 15 | 25 | 0.643 | |
| | T2DM vaccinated | 100 | 25.04 | 13.64 | 13 | 23 | 0.7335 | |

Table (1-8) :Level of Blood Urea among Studied Group

The current study showe there is no effect of Pfizer vaccine on blood urea.

S. Creatinine Level among Study Group

Serum Creatinine level in table (4-19) showed a level of AST in T2DM (0.7944±0.468) (0.6806±0.196) (vaccinated and non-vaccinated) respectively, in comparison with the mean healthy control (0.6582±0.268). Non-Significant difference in p-value was shown between T2DM (vaccinated and Non vaccinated) compared to healthy control p-value (0.1, 0.2) repetitively. Non –Significant differences were shown between T2DM vaccinated with Pfizer and T2DM non vaccinated in P-Value (0.120).

Table (1-9) : Serum Creatinine among Studied Group

| Variables | Study Group | N | Mean | Std. Deviation | Min | Max | Compared With control P. value | Compared T2DMVaccinated &T2DM non vaccinated P. value |
|---|---|---|---|---|---|---|---|---|
| S. Creatinine (mg/dL) | Controls | 50 | 0.6582 | 0.268 | 0.30 | 1.1 | | 0.120 |
| | T2DM non vaccinated | 50 | 0.6806 | 0.196 | 0.9 | 1.2 | 0.1 | |
| | T2DM vaccinated | 100 | 0.7944 | 0.468 | 0.8 | 1.35 | 0.2 | |

The present study revealed there is no effect of Pfizer vaccine on serum creatinine.

## 4. Conclusion

In the light of the results obtained, the study concluded the following:Cardiac function test means with normal range (T2DM show a higher level of Myoglobin and lipid profile in comparison with healthy control). The research reveals no adverse effects of the Pfizer vaccine on kidney function tests after the second dose of Pfizer vaccinations.

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