T.C.
KIRSEHIR AHI EVRAN UNIVERSITY
INSTITUTE OF NATURAL AND APPLIED SCIENCES
DEPARTMENT OF CHEMISTRY

ASSESSMENT OF CERTAIN BIOCHEMICAL RISK FACTORS FOR CARDIOVASCULAR DISEASES IN DIABETIC PATIENTS IN BAGHDAD

MAYTHAM QASIM HUMADI

MASTER THESIS

KIRŞEHİR / 2022

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ADVISOR
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KIRŞEHİR / 2022

THESIS STATEMENT

I hereby certify that all information in this thesis was obtained and presented in accordance with academic rules and ethical conduct. I also declare that I have fully quoted and referenced all non-original materials and results in this work as required by these rules and conduct.

Maytham Qasim HUMADI

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PREFACE

I would like to express my sincere gratitude to my dear professors, especially Doç. Dr. Aslihan GÜNEL and Co-Advisor: Dr. Ahmed Abdul Hussein and Iwould like to thank them for their knowledge and interest in this process. I would also like to express my sincere gratitude to my father and mother and my family who have supported me throughout my educational life.

December, 2023 **Maytham Qasim HUMADI**

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LIST OF ABBREVATION

Symbols	Description
%	:Percent
a	:Significant
b	:Significant
<	:Greater than
=	:Equel
>	:Less than
±	:Plus-minus
≤	:Greater or equal to
≥	:Less or equal to
dL	:Deciliter
g	:Gram
gm	:Gram
kg	:Kilogram
L	:Liter
m^2	:Square meter
mg	:Milli gram
mIU	:Milli-international units
mL	:Milliliter
mmol	:Milli mole
mol	:Mole
N	:Number of patients
ng	:Nanogram
nm	:Nanometer
µL	:Microliter
SD	:Standard deviation

Abbreviations	Description
ACE	:Acute coronary syndrome
ADA	:American Diabetes Association
BMI	:Body mass index
CCU	:Critical care unit
CAD	:Coronary artery disease
CHD	:Coronary heart disease
CRP	:C-reactive protein
CVD	:Cardiovascular disease
ELISA	:Enzyme linked immunosorbant assay
FSG	:Fasting serum glucose
GDM	:Gestational diabetes mellitus

HbA1c :Hemoglobin A1c
HF :Heart failure
HDL :High-density lipoprotein
IDDM :Insulin-dependent diabetes mellitus
IGT :Impaired glucose tolerance
IHD :Ischemic heart disease
IR :Insulin resistance
LDL :Low-density lipoprotein
MI :Mycardial ınfraction
NIDDM :Non-insulin-dependent diabetes mellitus
NO :Nitric oxide
OGTT :Oral glucose tolerance test
Oxid-LDL :Oxidized LDL
PKC :Protein kinase-C.
RAGE :Receptor of advanced glycation end-products
T1D :Type1 daibetes milletus
T2D :Type2 daibetes milletus
T-Ch :Total Cholesterol
TG :Triglyceride
TyG-index :Triglyceride-glucose-index
WHO :World Health Organization

ABSTRACT

MASTER THESIS

ASSESSMENT of CERTAIN BIOCHEMICAL RISK FACTORS for CARDIOVASCULAR DISEASES in DIABETIC PATIENTSin BAGHDAD

Maytham Qasim HUMADI

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Diabetes mellitus is expected to rise from 371 million people in 2013 to 552 million by 2030. Type 2 diabetes mellitus (T2D), which accounts for 90–95 percent of all cases, is to blame for the majority of the epidemic. Cardiovascular disease (CVD) is the primary cause of mortality in diabetics, accounting for more than half of all fatalities. High levels of hyperlipidemia and blood pressure, as well as classic risk factors such as smoking and obesity, are all risk factors for major cardiovascular events. Finally, insulin resistance (IR) and hyperglycemia raise the risk of CVD complications. This study was planned to investigate the relationship between T2D and CVD in the Iraqi population, as well as how these parameters relate to CVD risk factors, because there is little research on Iraqis in this field. To carry out the research, a total of 150 participants over the age of 40 were recruited for this research (88 men and 62 women). There were 50 "healthy" adults (29 males and 21 females) in the control group who had no history of CVD, T2D , or other disease, 50 T2D patients (22 males and 28 females) in the T2D group, and 50 T2D with CVD patients (37 males and 13 females) in the T2D & CVD From April to November 2021, these individuals will be recorded at Baghdad Teaching Hospital and all participants completed an interview-administered questionnaire. Serum fasting serum glucose (FBG), hemoglobin A1c (HbA1c), serum C-reactive protein (CRP), total cholesterol (T-Ch), triglycerides (TG), Triglyceride-glucose index (TyG-index), low-density lipoprotein cholesterol (LDL), high-density

lipoprotein cholesterol (HDL), and levels were measured by photometric transmission method using the Cobas c111 system measurement using kit (Roche/Germany), while Oxidized LDL (Oxid-LDL) was quantified using the competitive-ELISA approach (Cusabio/United States) and compared to the control group as well as across patient groups. T2D patients group had a greater prevalence of age (over 50 years), obesity, and high blood pressure. T2D & CVD patients group had significantly higher glucose and HbA1c levels, as well as Oxid-LDL, TyG-index, CRP, TG, LDL, and lower levels of HDL when compared to T2D group and control group. The increased risk of CVD in T2D patients is a result of these findings. The current study also discovered that individuals with poor HbA1c control are more likely to experience macrovascular problems than those with adequate HbA1c management. As a conclusion in individuals with T2D and CVD, higher levels of both Oxid-LDL and TyG-index may be a valuable marker for risk and prognosis in T2D and CVD patients. Obesity and dyslipidemia were both linked to an elevated risk of CVD in T2D individuals with high HbA1C levels.

December 2022, 55 Pages

Keywords: T2D, CVD, HbA1c, Oxid-LDL, TyG-index.

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ÖZET

YÜKSEK LİSANS TEZİ

BAĞDAT'TA DİYABETİK HASTALARDA KARDİYOVASKÜLER HASTALIKLAR için BAZI BİYOKİMYASAL RİSK FAKTÖRLERİNİN DEĞERLENDİRİLMESİ

Maytham Qasim HUMADI

Kırşehir Ahi Evran Üniversitesi
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Danışman: Prof. Dr. Aslıhan GÜNEL
Eş-Danışman: Dr. Ahmed Abdul HUSSEIN

2013'te 371 milyon olan Diabetes mellitus'lu kişi sayısının 2030'a kadar 552 milyona çıkması beklenmektedir. Tüm vakaların yüzde 90 ila 95'ini oluşturan Tip 2 diyabet (T2D), Hastalığın büyük kısmından sorumludur. Kardiyovasküler hastalıklar (KVH) tüm ölümlerin yarısından fazlasını oluşturmakla beraber diyabetli kişilerde birincil ölüm nedenlerindendir. Yüksek düzeylerde hiperlipidemi ve kan basıncı gibi geleneksel risk faktörlerinin yanı sıra sigara ve obezite de ciddi kardiyovasküler olaylar için risk faktörleridir. Sonuç olarak, insülin direnci (IR) ve hiperglisemi, KVH komplikasyonları riskini artırmaktadır. Bu çalışmanın amacı, Irak popülasyonunda T2D ve KVH arasındaki bağlantıya ve bu parametrelerin KVH risk faktörleriyle ilişkisini araştırmaktır. Bu alanda gerçekleştirilen çalışmalar incelendiğinde Iraklılar hakkında çok az araştırma söz konusudur. Toplamda 40 yaş üstü 150 kişi (88 erkek ve 62 kadın) araştırmaya dahil edilmiştir. Kontrol grubunda KVH, T2D veya başka hastalık öyküsü olmayan 50 "sağlıklı" kişi (29 erkek ve 21 kadın), T2D grubunda 50 T2D hastası (22 erkek ve 28 kadın) ve T2D ve KVH grubu için KVH'li 50 T2D hastası (37 erkek ve 13 kadın) yer almıştır. Nisan- Kasım 2021 arasında, bu kişiler Bağdat Eğitim Hastanesinde kayıt altına alınmış ve tüm katılımcılar görüşme yoluyla uygulanan bir anketi doldurmuşlardır. Açlık glukoz, hemoglobin A1c (HbA1c), serum C-reaktif protein (CRP), toplam kolesterol (T-Ch), trigliseritler (TG), trigliserit-glikoz indeksi

(TyG-index), düşük yoğunluklu lipoprotein kolesterol (LDL-Ch)), yüksek yoğunluklu lipoprotein kolesterol (HDL-Ch) düzeyleri, bir kit (Roche/Almanya) yardımıyla Cobas c111 sistem ölçümü kullanılarak fotometrik iletim yöntemiyle ölçülürken, oksitlenmiş LDL (Oxid-LDL) rekabetçi-ELISA yaklaşımı (Cusabio/Amerika Birleşik Devletleri) kullanılarak ölçülmüş ve kontrol grubu ile hasta grupları arasında karşılaştırma yapılmıştır. T2D hasta grubunda yaş (50 yaş üstü), obezite ve yüksek tansiyon prevalansı daha yüksek çıkmıştır. (T2D ve CVD) hasta grubu, T2D grubu ve kontrol grubu ile karşılaştırıldığında, glukoz ve HbA1c düzeylerinin yanı sıra Oxid-LDL, TyG-index, CRP, TG, LDL ve daha düşük HDL düzeylerine sahip olmuştur. T2D hastalarında KVH riskinin artması bu bulguların bir sonucudur. Mevcut çalışma ayrıca, yetersiz HbA1c kontrolüne sahip bireylerin, yeterli HbA1c yönetimine sahip olanlara göre makrovasküler problemler yaşama olasılığının daha yüksek olduğunu ortaya koymuştur. T2D ve CVD'li bireylerde, hem Oxid-LDL hem de TyG-indeksinin daha yüksek düzeyleri, diyabetik ve kardiyovasküler hastalarda risk ve prognoz için değerli bir belirteç olabilir. Obezite ve dislipidemi, yüksek HbA1C düzeyleri olan T2D bireylerde yüksek KVH riski ile bağlantılı olmuştur.

Aralık 2022, 55 sayfa

Anahtar Kelimeler: T2D, CVD, HbA1c, Oksit-LDL, TyG in

1. INTRODUCTION

Diabetes Mellitus is a chronic metabolic condition that affects a large percentage of the global population. Due to recent lifestyle changes, the global prevalence of diabetes has risen fast. Diabetesis considered to cause microangiopathy and macroangiopathy, as well as being a major risk factor for both (Menini, S. *et al.*, 2020). CVD is a common comorbidity in T2D. Overall, 32.2 percent of T2D patients developed CVD. CVD was defined as coronary heart disease (CHD), stroke, myocardial infarction (MI), atherosclerosis, angina pectoris, heart failure (HF), ischemic heart disease (IHD), and cardiovascular mortality (Einarson, T. R. *et al.*, 2018).

Diabetes patients are two to four times more likely to develop CVD and have a threefold greater overall mortality risk than individuals who do not have diabetes. It has also been established that diabetics are more likely to develop HF (Adler, A. *et al.*, 2021; Kayama, Y. *et al.*, 2015). T2D is usually linked to metabolic syndrome, which includes obesity, hypertension, and high T-Ch levels in the blood. Patients with T2D are more likely to develop CVD and associated complications (Shah, A. D. *et al.*, 2015).

One of the major causes of increased cardiovascular disease risk is the abnormal lipid metabolism seen in T2D. The change to a more atherogenic lipid profile is the result of a combination of qualitative, and quantitative abnormalities that constitute diabetic dyslipidemia. Although the pathogenesis of diabetic dyslipidemia is not fully understood (Vergès, B., 2015). Dyslipidemia is a key risk factor for the development of atherosclerosis, according to several epidemiological and clinical researches (Miura, Y. and Suzuki, H., 2019). One of the goals of this study was to investigate the function of lipid abnormalities in patients with T2D and how these lipid abnormalities are related to the development of cardiovascular disease.

Insulin resistance and relative insulin deficiency seen in patients with T2D have been linked to insulin's critical role in regulating lipid metabolism. , may contribute to these lipid abnormalities (Vergès, B., 2005).

In clinical practice, the TyG-index, which is computed from FSG and TG, has been recommended as a reliable marker of IR. Furthermore, new evidence suggests that the TyG-index is linked to not only the development of T2D but also the incidence of CVD. These findings show that using the TyG-index as a predictor of future CVD risk in people with T2D and CVD may be feasible (Wang, L. *et al.*, 2020). T2D patients with hypertension account for more than 60% of all cases. Controlling blood pressure has been shown to improve CVD outcomes in a number of trials. According to a recent meta-analysis, lowering systolic blood pressure to less than 130 mmHg is linked to lower CVD events and all-cause mortality (Valensi, P. *et al.*, 2021).

1.1. Objectives of the Study

The overall goals of this thesis are to examine the primary and possible development CVD risk factors in T2D patients, as well as to comprehend the fundamentals of a prediction tool to avoid the disease's severe vascular consequences**.**

The objectives of this research were to **:**

1. Determine which patients are at a high risk of future CVD events. The discovery of readily available and accurate markers might have huge therapeutic implications in terms of improving recurrent CVD risk classification**.**
2. Determine whether the TyG-index has predictive value for CVD in T2D patients**.**
3. Determine the serum Oxid-LDL as a predictive value in patients groups**.**
4. Determine the impact of obesity on patients groups.
5. Determine the role of the main risk factors for CVD in patient groups, such as blood pressure**.**
6. Determine the role of serum CRP levels in patient groups.
7. Determine the function of dyslipidemia in patient groups.
8. Determine the factors associated with poor and good HbA1c management in Iraqi patients with CVD and/or T2D patients**.**

2. GENERAL PARTS

2.1. Cardiovascular disease

Coronary heart disease (CHD), coronary artery disease (CAD), and acute coronary syndrome (ACS) are examples of CVD that affect the heart and blood arteries. CAD, on the other hand, is defined by atherosclerosis in the coronary arteries and can be asymptomatic, but ACS almost always includes a symptom, such as unstable angina, and is frequently associated with MI, regardless of whether or not CAD exists (Sanchis-Gomar, F. *et al.*, 2016).

CVD is one of the top causes of death in diabetic persons (de Vries, T. I., 2020), although standard CVD risk factors cannot account for a significant portion of the disease burden (Strain, W. D. and Paldánius, P. M., 2018).

2.1.1. Causes and risk factors

Cardiovascular disease, which is caused by a mix of inherited and lifestyle factors, is the leading cause of mortality and morbidity in the world. Previous research has found that modifiable health behaviors and variables, such as smoking, physical activity, food, and body mass index (BMI), are strongly linked to the risk of developing CVD and other long-term diseases and death (Eckel, R. H. *et al.*, 2021).

The significance of lipids and other risk variables cannot be underestimated, however, the reasons underlying this connection remain uncertain. When it comes to health habits and variables, smoking, BMI, physical activity and food consumed are at the forefront. Non-behavioral variables include T-Ch, blood pressure and FBG levels (Said, M. A. *et al.*, 2018).

2.1.2. Pathophysiology

The most important pathophysiological pathways of atherothrombosis in diabetic individuals are depicted in Figure 2.1.

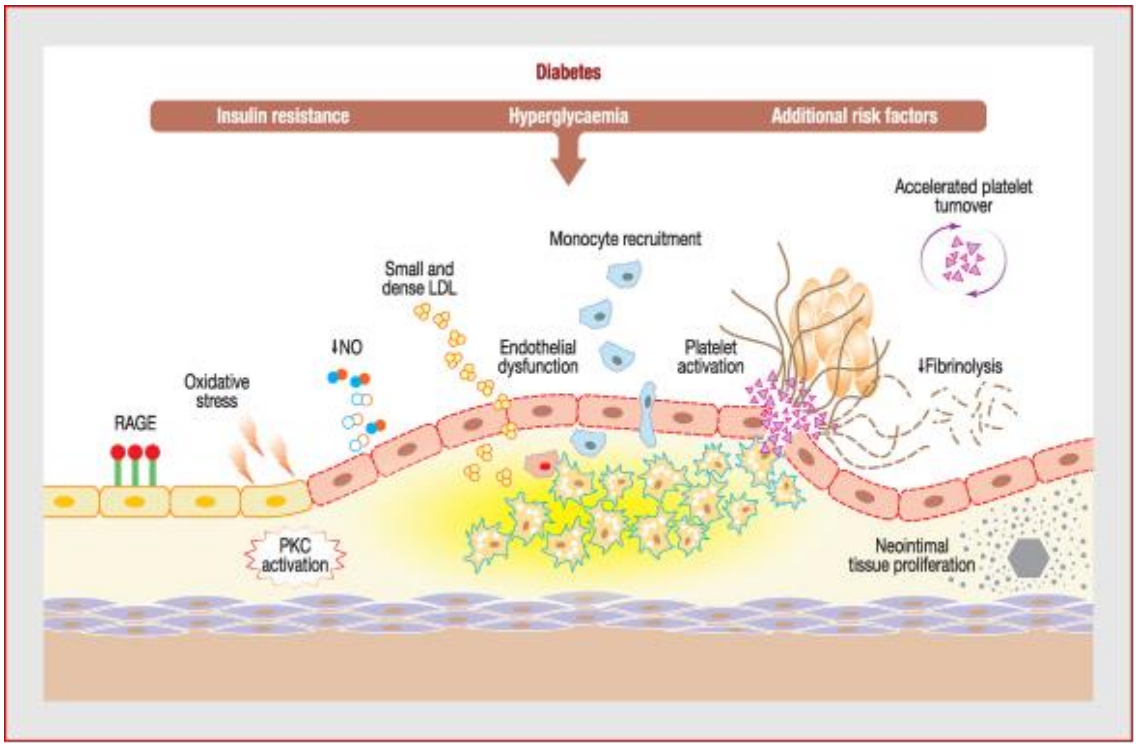

Figure 2.1. Atherothrombosis pathophysiology in diabetic people (Valensi, P. *et al.*, 2021).

Chronic inflammation has been associated to increased oxidative stress and abnormal macrophage activity in diabetes-related CAD. Diabetic macroangiopathy is caused by a variety of biochemical pathways, including reactive oxygen species (ROS) overproduction, activation of protein kinase-C (PKC), increased formation of advanced glycation end-products (AGE) and activation of the receptor for advanced glycation end-products (RAGE), and chronic vascular inflammation (Valensi, P. *et al.*, 2021).

2.1.3. The classification of cardiovascular risk in T2D

Diabetic people are more prone to develop CVD. Table 2.1 illustrates how to classify risk levels.

Table 2.1. The different types of CVD risk in diabetic people (Cosentino, F. *et al.*, 2020).

Extremely high risk	Patients with diabetes who have a history of CVD or other target organ damage* or a combination of three or more main risk factors** or long-term T1D (>20 years) with early onset
High risk	Patients with T2D for more than ten years who do not have target organ impairment or any other risk factor
Moderate risk	Young patients (T1D less than 35 years old or T2D less than 50 years old) with a duration of T2D less than 10 years and no additional risk factors
* renal impairment (eGFR 30 mL/min/1.73 m^2), left ventricular hypertrophy, or retinopathy are among conditions that can cause proteinuria. **Age, smoking, dyslipidemia, hypertension, and obesity are all factors to consider.	

2.2. Diabetes Mellitus

2.2.1. Definition

Diabetes mellitus is a collection of metabolic illnesses characterized by hyperglycemia and carbohydrate, lipid, and protein metabolism abnormalities caused by deficiencies in insulin production, insulin action, or both (Szatko, A. *et al.*, 2020, Saraswati, P. A. I. *et al.*, 2021).

Diabetes mortality rates are increasing as a result of a variety of causes including poor nutrition, lack of physical exercise, obesity, and smoking (Menini, S. *et al.*, 2020, Szatko, A. *et al.*, 2020).

2.2.2. Aetiology

The dysfunction or loss of pancreatic cells is now well recognized as a typical characteristic of all types of T2D . Many pathways can cause cells to lose their function or even die. These pathways include IR, genetic vulnerability and abnormalities, autoimmunity, inflammation, epigenetic mechanisms, concurrent disorders,and environmental factors (Adler, A. *et al.*, 2021, WHO, 2019).

2.2.3. Complications

Diabetes is expected to impact 693 million individuals by 2045 (Cole, J. B. and Florez, J. C., 2020), making it one of the world's fastest rising illnesses. In people with diabetes, devastating macrovascular consequences (like heart disease) and microvascular problems (including diabetic kidney disease, diabetic retinopathy, and neuropathy) result in increased mortality, blindness, kidney failure, and a lower overall quality of life. Numerous genetic studies have revealed a clear hereditary component to both diabetes and its complications. Clinical risk factors and glycemic control cannot predict the development of vascular issues on their own (Cole, J. B. and Florez, J. C., 2020).

2.2.4. Diagnosis

Fasting serum glucose or HbA1c should be used to study diabetes. In order to identify impaired glucose tolerance (IGT), oral glucose tolerance test (OGTT) is required. Individuals with established CVD should be tested with HbA1c and/or FSG; if FSG and HbA1c are equivocal, an OGTT can be performed (Cosentino, F. *et al.*, 2020).

Diabetes is defined as FSG of more than 7.0 mmol/L, HbA1c of more than 6.5 percent, or a random blood glucose of (11.11 mmol/L). If high levels are seen in patients, repeat testing, ideally with the same test, the next day to confirm the diagnosis (Adler, A. *et al.*, 2021, Keutmann, S. *et al.*, 2020).

2.2.5. Classification

The World Health Organization's (WHO) and the American Diabetes Association's (ADA, 2003)guidelines are used to classify diabetes (Cosentino, F. *et al.*, 2020). In 1999, the WHO divided diabetes into aetiological types and clinical stages. T1D and T2D have superseded the earlier diabetes classification and nomenclature (insulin-dependent diabetues mellitus (IDDM) and non-insulin-dependent diabetues mellitus (NIDDM). Individuals who require exogenous insulin were diagnosed using the clinical staging system. Pathology of the pancreatic-cell, its malfunction or destruction, is a frequent hallmark of all types of diabetes. Genetic predisposition, epigenetic changes, IR, immunization, inflammation, and environmental factors are some of the pathways that contribute to these factors (Szatko, A. *et al.*, 2020).

Glucose intolerance that initially appears during pregnancy is known as gestational diabetes mellitus (GDM). GDM is a serious public health issue that affects one out of every six pregnancies throughout the world (Goyal, A. *et al.*, 2020). Maternal hyperglycemia that is less severe than that seen in diabetes is linked to higher birth weight, a higher likelihood of cesarean delivery, and other negative effects (Bruns, D. E. *et al.*, 2020).

2.2.6. Type2 diabetes; Epidemiology

Type 2 diabetes is the most common, accounting for 90-95 percent of all cases. In 2019, diabetes claimed the lives of 4.2 million people. Diabetes impacted 463 million individuals between the ages of 20 and 79, with the amount anticipated to rise to 693 million by 2045. The greatest age group exists between the ages of 40 and 59. T2D is more widespread in low-to-middle-income countries, where more than 80% of patients live (Galicia-Garcia, U. *et al.*, 2020).

2.2.7. Type2 diabetes; Symptoms and Signs

Diabetes is characterized by symptoms such as thirst, polyuria, blurred eyesight, and weight loss. Ketoacidosis or a non-ketotic hyperosmolar condition is the most serious clinical signs, which can lead to dehydration, unconsciousness, and in the absence of adequate treatment, death (Adler, A. *et al.*, 2021).

2.2.8. Type2 diabetes; Pathophysiology

T2D risk factors and the pathophysiological changes that enable insulin dysfunction to persist are complex mixes of genetic, metabolic, and environmental variables that interact with one another (obesity, low physical activity and an unhealthy diet). These factors affect cell function, resulting in a complex network of pathogenic changes that interact and sustain insulin failure (Figure 2.2) (Galicia-Garcia, U. *et al.*, 2020).

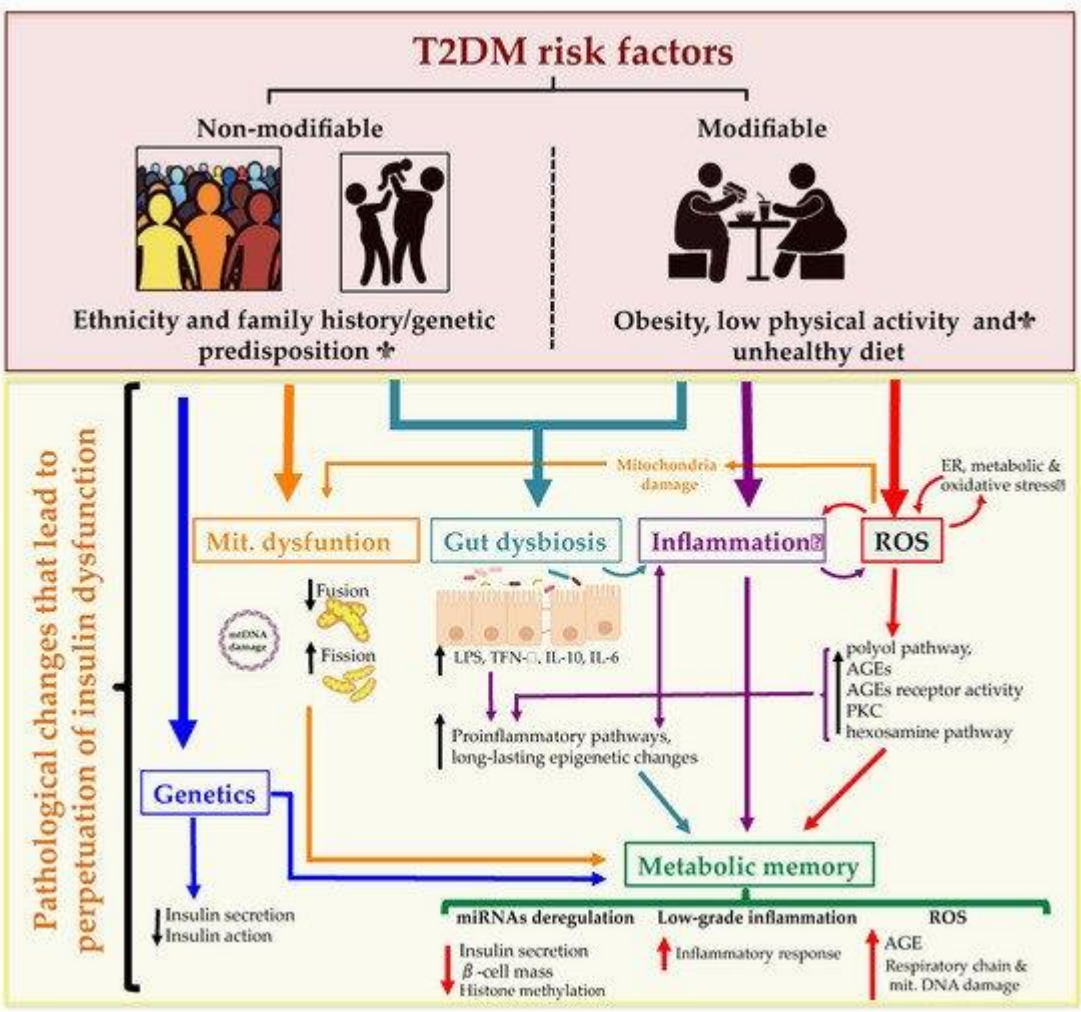

Figure 2.2. Risk factors for T2D and pathological alterations (Galicia-Garcia, U. *et al.*, 2020)

2.2.9. Type2 diabetes: Mechanisms (β-Cell Dysfunction)

Type 2 diabetes might be caused by a more intricate network of interactions between the environment and many metabolic processes involved in cell biology. Overeating disorders, such as obesity, are associated with hyperglycemia and hyperlipidemia, both of which induce IR and chronic inflammation. Cells are vulnerable to toxic stimuli such as inflammation, inflammatory stress, amyloid stress, and metabolic/oxidative stress under these circumstances, all of which have the potential to induce islet integrity loss due to genetic vulnerabilities (Christensen, A. A. and Gannon, M., 2019).

Sterile, systemic, low-grade chronic inflammation has long been recognized as a major biological feature of diabetes, and emerging data shows that inflammation plays a key role in diabetes complications (Figure 2.3) (Menini, S. *et al.*, 2020).

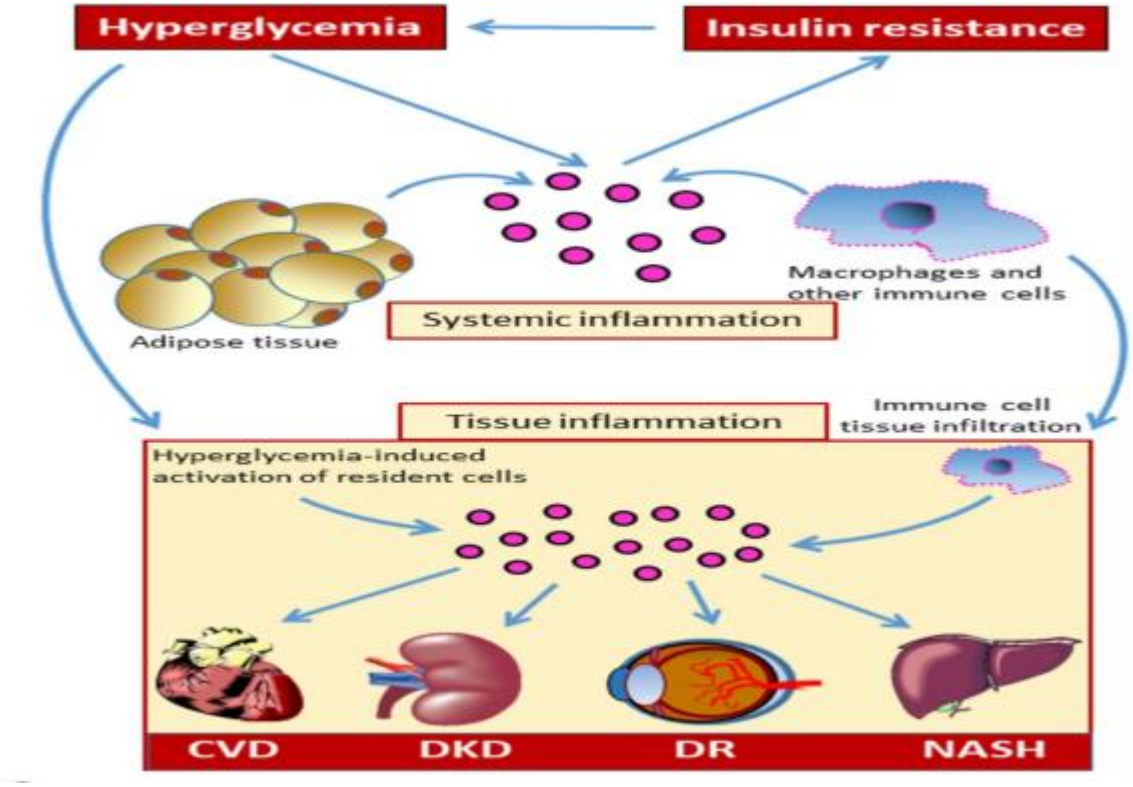

Figure 2.3. Diabetic problems cause systemic tissue inflammation (Menini, S. *et al.*, 2020). DKD: Diabetic kidney disease. DR:Diabetic retinopathy. NASH: Non-alcoholic steatohepatitis.

2.2.10. Type2 diabetes: Insulin Resistance

Insulin resistance is characterized as a decreased metabolic response to insulin by insulin-responsive cells. The IR refers to significant metabolic cross talk between the skeletal muscle, liver, pancreas, and adipose tissue (Czech, M. P., 2017). Other chemicals, such as Growth Hormone and Insulin-like Growth Factor 1, interact with insulin function in the case of feeding. In fasting, glucagon, catecholamines and glucocorticoids decrease the insulin response to avoid insulin-induced hypoglycemia. Therefore, IR might be caused by an increase in the secretion of these hormones. IR in various organs often occurs before systemic IR, eventually, T2D develops (Galicia-Garcia, U. *et al.*, 2020).

2.2.11. Type2 diabetes: Obesity

Obesity is recognized as a severe public health problem in both developed and developing countries, and as a disease that affects people of all ages. Obesity has been identified as one

of the leading causes of death since it is linked to the development of a number of various noncommunicable disorders. It has been designated as one of the main causes of mortality. According to the WHO, 44 % of people with obesity were diagnosed with T2D, 23 % with CVD, and 7-14 % with malignancies. Fatter persons are more likely to acquire T2D, according to the findings of several clinical research investigations (Bansal, C. *et al.*, 2021; Olaogun, I. *et al.*, 2020). Obesity is currently the leading cause of the rising prevalence of T2D. The link between obesity and T2D is believed to be a sustained positive lipid influx that triggers beta-cell dysfunction. Fatty acids (FA), a major part of lipids, have important implications for beta cells. Through cell surface receptors and intracellular mechanisms (Figure 2.4). FA rapidly enhance glucose-induced insulin production (Imai, Y. *et al.*, 2020; Yahya *et al.*, 2021).

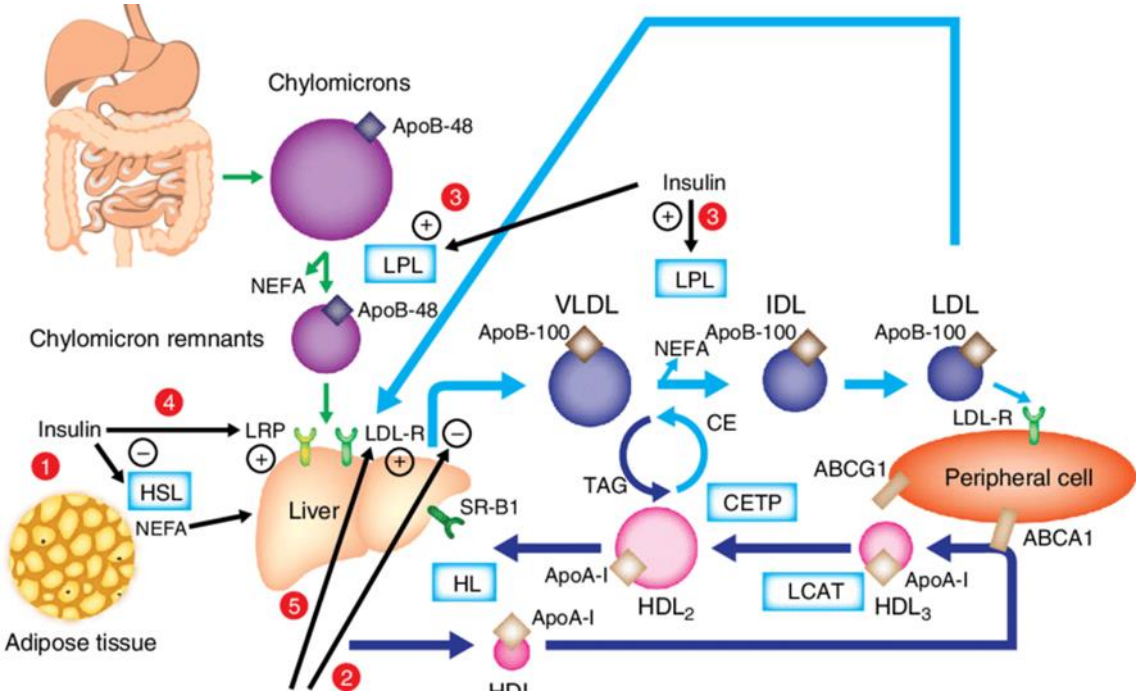

Figure 2.4. An overview of Insulin affects lipoprotein metabolism in humans (Vergès, B., 2015).

2.2.12. Type2 diabetes: Complications/ Cardiovascular Risk

Type2 diabetes is a multisystem illness that has a significant link to the development of CVD. After an acute MI, glycemic variability has been demonstrated to be a major predictive factor for worse cardiac outcomes in people with T2D, displacing other recognized glycemic markers such as HbA1c and FBG. Strain, W. D. and Paldánius, P. M. (2018) found that elevated glycemia is a substantial factor of both arterial stiffness and carotid intimal media

thickness. T2D is regarded a substantial risk factor for CVD as a result of these variables, which are likely due to the involvement of various molecular processes and pathological pathways. The roles of IR in atherosclerosis are showen in Figure 2.5 (Galicia-Garcia, U. *et al.*, 2020).

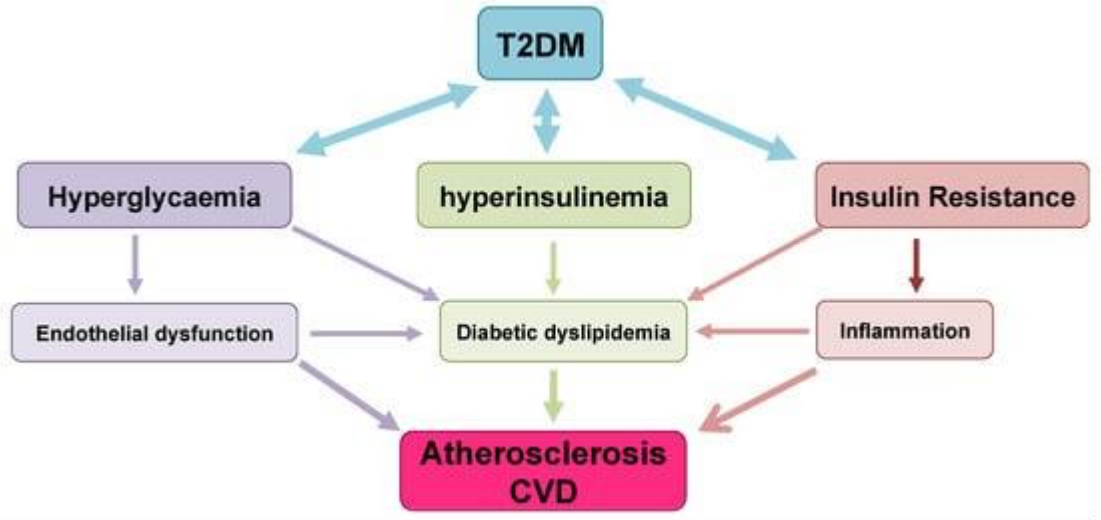

Figure 2.5. T2D-related factors and their interactions (Galicia-Garcia, U. *et al.*, 2020)

2.2.13. Type 2 diabetes: Dyslipidaemia/ Cardiovascular Development

Type 2 diabetes is distinguished by dyslipidaemia, which increases the risk of atherosclerosis and mortality in diabetics. Diabetic dyslipidemia is distinguished by a dyslipidaemic profile that includes elevated TG, TG-rich lipoproteins, LDL, and low HDL values. Epidemiological evidence suggests that TG-rich lipoproteins and their remnants have a role in atherogenesis and CVD risk (Varbo, A. *et al.*, 2018).

T2D is characterized by dyslipidemia (Figure 2.6), which raises the risk of atherosclerosis and death in diabetics. Diabetic dyslipidemia is characterized by increased TG, TG-rich lipoproteins, LDL, and low HDL levels. According to epidemiological research, TG-rich lipoproteins and their remnants have a role in atherogenesis and CVD risk (Varbo, A. *et al.*, 2018).

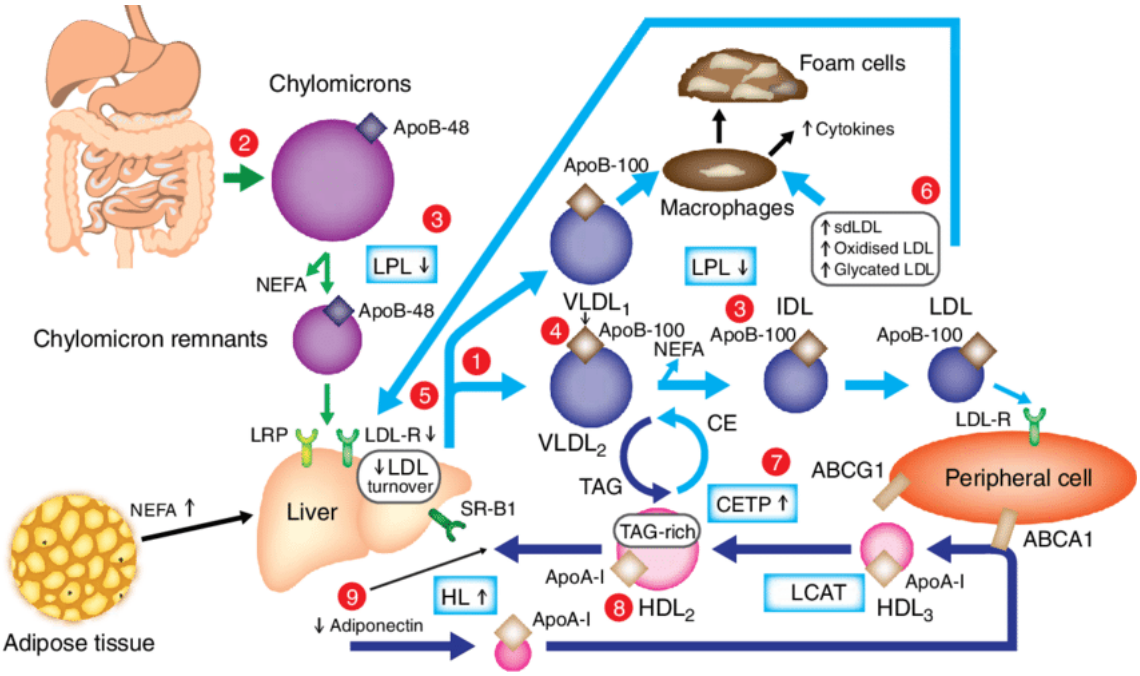

Figure 2.6. Lipid abnormalities in T2D (Varbo, A. *et al.*, 2018).

3. MATERIALS AND METHODS

3.1. The Subjects

This study involved 150 (male and female) volunteers, ranging in age from 40 to 70 years old, who were seen at the Medical City in Baghdad Teaching Hospital in Baghdad, Iraq, between April and November 2021.

All patients in this research were evaluated in the hospital by their physician to confirm the diagnosis of T2D and CVD based on clinical history, physical examination, laboratory, and echocardiographic features. The groups were divided into the following categories:

A- Healthy Subjects (Control Group):

Fifty seemingly healthy persons (29 males, 21 females) were chosen at random to act as controls, ranging in age from 40 to 61 years. None of them had diabetes; test results of FSG less than 6 mmol/L and HbA1c less than 6.0 percent were judged normal. They all had a negative family history of diabetes and/or CVD, as well as a negative medication history for both.

B- Diabetic Patients (T2D Group):

Fifty patients (22 males, 28 females) with T2D were treated (antidiabetic medications) and ranged in age from 45 to 66 years. In the selection questionnaire, the patient's medical history was taken into account first (Appendix 1).

Clinical types were assigned to each patient. T1D was ruled out, while T2D was characterized as FSG equal or more than 7.0 mmol/L or current hypoglycemic medication use.

C- Diabetic Patients with Cardiovascular Diseases (T2D &CVD Group):

Fifty diabetic individuals (37 males, 13 females) and a range of 48-70 years with T2D and cardiac problems such as atherosclerosis, cardiomyopathy, congenital heart disease, and heart failure/attack were studied.

3.2. Criteria for Exclusion

1. Gestational diabetes (GDM).

2. Patients with T1D or diabetes of unknown type.

3.3. Collection of Blood

After fasting for at least 10-12 hours, blood samples were taken from each patient's capital vein to assess serum FSG, HbA1c, T-Ch, TG, LDL, HDL, Oxidized LDL, TyG-index, and CRP.

3.4. Collection of Data and Definitions

Clinical information was gathered for all patients. The data obtained included age, gender, height, weight, diabetes history, diabetes duration, blood pressure, and a family history of CVD. The average of systolic and diastolic blood pressure (SBP and DBP) measures were taken after reviewing the CVD (coronary heart disease, cerebrovascular disease, and peripheral arterial disease) clinical history and key information about the illness's duration.

Other conditions were investigated, regardless of whether the patient was taking medication (for heart failure, lipid-lowering medications, or another ailment).

Direct interviews with all groups in this study were used to fill up the questionnaire (Appendix 1) form. Diabetes was defined as having an FSG level of more than 7.0 mmol/l or a HbA1c level of more than 6.5 percent, or having diabetes diagnosis and/or treatment. Hypertension was described as an SBP of more than 140 mmHg and/or a DBP of more than 90 mmHg, as well as hypertension medication. Weight divided by height squared was used to compute BMI. Dyslipidemia was defined as T-Ch > 5.18 mmol/L, serum TG > 1.72 mmol/L, HDL <1.0 mmol/L, LDL > 3.37 mmol/L, or dyslipidemia diagnosis and therapy. The ln [fasting TG level (mg/dL) × FSG level (mg/dL)/2] was used to calculate the TyG-index. The Fridewald formula was used to

compute LDL (Pradhan, S. *et al.*, 2020). An ELISA test was used to calculate Oxid-LDL. If HbA1c was less than 7.0 percent, glycemic control was considered good, and if HbA1c was

greater than or equal to 7.0 percent, glycemic control was considered poor.

3.5. Matrials

No.	Equipment or Instruments	Company	Country
1	Micropipettes (different volumes1000+ 200+ 10 µl)	Eppendorf	Germany
2	Multichannelpipette(50-300µL)	Slamid	Germany
3	ELISA reader	Biotek	USA
4	Capped plastic tubes	Afco-Dispo	Jordan
5	Centrifuge	Kokusan	Germany
6	Ependroff tubes	Sigma	England
7	ELISA washer	biotek	USA
9	Gel Tubes	Afco	Jordan
10	Hot plat stirrer	LabTech®	Korea
11	Incubator	Memmert	Germany
16	Cobas c111	Roche	Germany
20	Sensitive balance	Sartorius	Germany
21	Spectrophotometer	Shimadzu	Japan
23	Vortex mixer	CYAN	Belgium
25	Refrigerator	hitachi	Japan

3.6. Method

3.6.1. Enzyme Linked Immunosorbent Assay (ELISA) For the determination of human Oxid-LDL concentrations

Principle of the Test

In this test, the competitive inhibition enzyme immunoassay technique was employed. The microtiter plate supplied in this kit has been pre-coated with Oxid-LDL. Horseradish peroxidase (HRP) conjugated anti-Oxid-LDL antibody preparations were added to the appropriate microtiter plate wells with standards or samples.

A competitive inhibitory response occurs in samples containing pre coated Oxid-LDL and Oxid-LDL. When a substrate solution was applied, the color developed in the wells in the opposite direction of the amount of Oxid-LDL in the sample.

The color development process was paused, and the intensity of the color was measured.

Detection range: 1.56 mU/mL- 100 mU/mL

Intra-assay Precision: CV%<8%

Inter-assay Precision: CV%<10%

Assay procedure

All samples and reagents should be at room temperature before use. Samples should be thawed before testing and then centrifuged again. It was recommend running all samples and standards in duplicate.

1. As instructed in the sections above, prepare all reagents, working standards, and samples.
2. Determine the number of wells to be used by consulting the assay layout sheet. Place any extra wells and desiccant back into the pouch, close the bag, and store at 4°C.
3. Create a blank well with no fillings.
4. Add 50 µl of standards and samples per well. Immediately add 50 µL of HRP conjugate (1x) to each well (do not add to blank wells). Mix thoroughly by pipetting or gently shake the plate for 60 seconds. Plate placement was provided to keep track of standards and sample analyses.
5. Cover using the included adhesive strips. Incubate at 37 °C for 1 hour.
6. Aspirate each well, then repeat the wash 4 times for a total of 5 washes. Complete removal of liquid at each stage was critical for effective performance. To wash, fill each well with 200 µl wash buffer using a squirt bottle, multichannel pipette, manifold dispenser, or autowasher and let sit for 2 minutes. After the last wash, aspirate or decant any remaining wash buffer. Invert the plate and wipe it with a new paper towel.
7. Fill each well with 90 µl of TMB substrate. Incubate at 37 °C for 20 minutes. block the light.
8. Add 50 µl of stop solution to each well and tap the plate to mix everything well.
9. Measure the optical density of each well within 5 minutes using a microplate reader set at 450 nm. Set the wavelength adjustment to 540 or 570 nm, if available. Readings at 540 or 570 nm should be subtracted from readings at 450 nm. This subtraction was used to correct for optical defects in the plate. A direct measurement at 450 nm without correction was more accurate, but can be higher.

Calculation of results

Take the mean of duplicate measurements for each standard and sample and take the mean optical density of the blank from the equation. Utilize computer software capable of creating a four-parameter logistic (4 PL) curve fit to reduce the data to create a standard curve. Alternatively, plot the average absorbance of each standard on the x-axis and the concentration on the y-axis to create a standard curve and trace the line of best fit across the points on the graph. The data can be linearized by graphing the log of OxLDL concentration versus the log of O.D., and regression analysis cought be used to find the best fit line for the data. This approach produces a good but less accurate fit of the data. It was necessary to multiply the concentration determined from the standard curve by the dilution factor if samples have been diluted.

3.6.2. The Cobas c111 system was used to measure photometric transmission

The Roche diagnostics corp's Cobas c111 is an in system for in vitro diagnostic testing for determining FSG, HbA1c, T-Ch, HDL, and TG in whole blood or plasma quantitatively.

3.6.2.1. Serum Glucose Determination

Summary

Glucose was the main carbohydrate in peripheral blood. The body's main source of cellular energy was the oxidation of glucose. Dietary glucose was converted to fatty acids or glycogen for storage in adipose tissue or liver. The most important hormones were produced by the pancreas and work together to regulate blood glucose levels within specific limits. Diabetes mellitus, caused by lack of insulin production or activity, was the most common cause of hyperglycemia. Elevated glucose levels were also influenced by various auxiliary variables. Pancreatitis, thyroid problems, kidney failure, liver disease were some of them. Less commonly, hypoglycemia was seen. Low blood sugar levels can result from various diseases, including insulinoma, hypopituitarism, and insulin-induced hypoglycemia. Urinary glucose analysis was used as a screening method for diabetes to assess glycosuria, detect renal tubular abnormalities, and help treat diabetes mellitus. Glucose levels were measured in the cerebrospinal fluid for evaluation of meningitis, meningeal lesions due to neoplastic disease, and other neurological disorders.

Principle of the Test

To detect glucose in serum, the cobas c111 uses a UV test. Hexokinase (HK) catalyzes the conversion of glucose to glucose 6 phosphate (G6P) and adenosine diphosphate (ADP). G6P is converted to 6 phosphogluconate and reduced nicotinamide-adenine-dinucleotide (NADH) in the presence of NAD by the enzyme G6P- dehydrogenase (G6PD). NADH concentration rises in lockstep with glucose amount and may be detected spectrophotometrically at 340 nm.

Assay procedure

To ensure that tests run as smoothly as possible, please follow the instructions provided in this document for the relevant analyzers. Please refer to the relevant operator's manual for instructions on how to run the assay using a particular analyzer. Applications not validated by Roche are not guaranteed to work as expected. Instead, the user was responsible for defining this Application for plasma, serum, urine and CSF.

Assay type	2-Point End		
Reaction time / Assay points	10 / 6-32 (STAT 7 / 6-32)		
Wavelength (sub/main)	700/340 nm		
Reaction direction	Increase		
Units	mmol/L (mg/dL, g/L)		
Reagent pipetting	Diluent (H2O)		
R1	28 µL	141 µL	
R2	10 µL	20 µL	
Sample volumes	Sample	Sample dilution	
		Sample	Sample Diluent (NaCl)
Normal	2 µL	—	—
Decreased	10 µL	15 µL	135 µL
Increased	2 µL	—	—

Expected values

Glucose=74- 106 mg/dl

Conversion factors (mmol/L x 18) = mg/dl

(mg/dl / 18) = mmol/L

3.6.2.2. HbA1c Determination

Summary

A red-colored iron-containing protein known as hemoglobin (Hb) was found in red blood cells. Its main job was to move carbon dioxide and oxygen through the blood. Hb consists of several derivatives and variations, such as adult HbA and fetal HbF. (e.g. acetylation, saccharification). In adults, HbA, composed of four protein chains, makes up the majority (>95%) of Hb. (2 alpha, 2 beta chains).Glycated hemoglobin, or one of its subfractions, is HbA1c. This was due to the different sugars attached to the HbA molecule. By coupling the N-terminal amino group of the beta chain of normal adult Hb with glucose, HbA1c was made nonenzymatically in two processes (HbA). Unstable HbA1c was produced in the first reversible step. In the next step of the reaction, it rearranges to produce stable HbA1c. As average blood glucose levels rise, so does the percentage of HbA that was converted to stable HbA1c in red blood cells. Red blood cells had a life expectancy of approximately 100-120 days, limiting their conversion to stable HbA1c. Therefore, HbA1c represents the average blood glucose level over the past 2-3 months, not the peak blood glucose level.

Principle of the Test

To release hemoglobin from the erythrocytes, a blood sample was diluted and combined with TRIS buffer. In a reaction chamber, a portion of the material was combined with sodium lauryl sulfate (SLS). The SLS-hemoglobin complex was created with SLS. The concentration of SLS-hemoglobin complex was measured with a 525 nm wavelength to determine total hemoglobin amount. Potassium ferricyanide and sucrose laurate were used to denaturize HbA1c in another part of the sample. The latex agglutination inhibition response was then initiated by reacting the agglutinator with a synthetic antigen capable of bonding with the HbA1c antibody. By measuring the latex agglutination inhibition response at a wavelength of 625 nm, the concentration of HbA1c may be determined. The HbA1c percent value was calculated as a ratio of HbA1c to total hemoglobin concentrations.

Measuring range 4- 14 %

Expected values HbA1c ≥ 6.5 %, to diagnose diabetes

3.6.2.3. Lipid Panel Determination

Summary

Cholesterol levels are used to assess the risk of developing atherosclerosis. It also aids in the diagnosis and treatment of diseases associated with high cholesterol levels and abnormal lipid and lipoprotein metabolism. Measurement of HDL was of great importance for clinical purposes because of the negative relationship between HDL levels and the risk of atherosclerosis. Elevated HDL levels reduce the risk of coronary heart disease, whereas low HDL levels increase the risk of CVD, especially when combined with high TG. Methods have been developed to raise HDL levels to treat CVD. TG measurementwere used to diagnose and treat individuals with T2D , metabolic syndrome, dyslipidemia, liver obstruction, and various other endocrine disorders. It was common practice to calculate LDL concentrations using the Friedewald method to determine a comprehensive lipid panel.

LDL plays an important role in the development of coronary artery disease, especially atherosclerosis. LDL makes up the majority of cholesterol found in atherosclerotic plaques. Of all the single features that can be used to predict coronary atherosclerosis, LDL was the most effective clinical predictor. As a result, reduction of LDL was a major goal of pharmaceuticals aimed at lowering lipid levels.

Principle of the Test

In this study, blood plasma was used and the plasmaCentrifugation is was used to separated by centrifugation from the erythrocytes. Phosphate buffer was used to dilute the plasma sample after that. The HDL test employs a precipitation technique using the precipitant method (with Mg^{2+}) and the precipitant reagent (phosphotungstic acid). The components were precipitated and eliminated, with the exception of HDL. Using an enzymatic technique, Cobas c111 system calculates T-Ch and HDL. The sample's cholesterol esters were hydrolyzed to produce T-Ch and fatty acids (FA). In the presence of cholesterol dehydrogenase, T-Ch and NAD^{+} were formed cholestenone and NADH. Through an oxidation-reduction process, WST8 was converted to formazan dye by diaphorase and NADH. The content of HDL and T-Ch in the sample was exactly related to the color intensity of formazan, which was evaluated in a particular wave length (460 nm).

Cholesterol ester $\xrightarrow{\text{cholesterol esterase}}$ Cholesterol + Fatty acids

Cholesterol + NAD^+ $\xrightarrow{\text{Cholesterol dehydrogenase}}$ Cholestenone + NADH

NADH + WST8 $\xrightarrow{\text{Cholesterol dehydrogenase}}$ NAD+ + Formazan dye

The TG test was an enzyme-based procedure. Lipoprotein lipase (LPL) hydrolyzes TGs in the sample to glycerol and FA. In the presence of glycerol dehydrogenase, glycerol and NAD+ produce dihydroxyacetone and NADH. Through an oxidation-reduction process, diaphorase and NADH were converted WST8 to formazan dye. The wavelength of 460 nm was used to compute the intensity of color of the formazan, which was proportional to the TG content.

Triglycerides $\xrightarrow{\text{Lipoprotein Lipase}}$ Glycerol + Fatty acids

Glycerol + NAD^+ $\xrightarrow{\text{Glycerol dehydrogenase}}$ Dihydroxyacetone + NADH

NADH + WST8 $\xrightarrow{\text{Diaphorase}}$ Formazan dye+ NAD^+

Low density lipoprotein (LDL-Ch determination)

The Friedewald method was used to calculate LDL when the TG content was less than 4.52 mmol/L (400 mg/dL).

LDL = TC - HDL – TG/5

The measured LDL was not given when the TG value was more than 400 mg/dL (4.52 mmol/L). Non-fasting individuals and those with type III hyperlipoproteinemia were also excluded from using the formula.

Measuring range:

T-Ch: 50-500 mg/dL or 1.28-12.95 mmol/L

TG: 45-650 mg/dL or 0.50-7.35 mmol/L

HDL: 15-100 mg/dL or 0.38-2.60 mmol/L

Conversion factors: mmol/L x 38.66 = mg/dL

mg/dL x 0.0259 = mmol/L

Analyte	Concentration mg/dl (mmol/l)	Classification

LDL	< 100 (< 2.59) 100-129 (2.59-3.34) 130-159 (3.37-4.12) 160-189 (4.14-4.90) > 190 (> 4.92)	Normal Above optimal Borderline high High Extremely high
HDL	< 40 (< 1.04) > 60 (> 1.55)	Low High
T-Ch	< 200 (< 5.18) 200-239 (5.18-6.19) > 240 (> 6.20)	
TG	< 150 (< 1.70) 150-199 (1.70-2.25) 200-499 (2.26-5.64) > 500 (> 5.65)	Normal Borderline high High Extremely high

3.6.2.4. Serum CRP Determination

Principle of the Test

Centrifugation separates the erythrocytes from the plasma. The plasma sample is then diluted with buffer (HEPES) and transported to a reaction chamber to be combined with CRP-antibody-latex reagent. The CRP-antibody on the latex particle binds to the CRP in the diluted plasma. The quantity of agglutination is related to measure the change in absorbance at 525 nm and 625 nm, which is used to compute CRP concentration.

Measuring range

3.0-400 mg/l or 0.30-40.0 mg/dl

Expected values

Adults: < 5.0 mg/l (< 0.5 mg/dl)

Assay procedure

Instructions for use

~ Use soap to wash hands, warm water promote blood circulation.

~ Wash the fingers thoroughly. dry hands

~ The fingertip can be disinfected by wiping the puncture site three times with a cotton swab or sterile gauze soaked in 70%-100% isopropanol emollient-free or 70%-100% ethanol

emollient-free. Then repeat the process using a second swab or sterile gauze pad soaked with the same 70% to 100% isopropanol emollient.

~ Prick the patient's finger with a disposable lancing device, such as the Accu-Chek Safe-T-Pro Plus. Blood samples should be taken according to the instructions of the lancing device

~ Remove the first drop of blood with a cotton swab.

~ With the imprinted side of the disc facing up, place the suction point of the disc over the blood drop. The disk will fill itself up.

~ Apply blood and confirm that there was blood in the designated area. Turn the CD over and listen to the volume of the sample. Blood should completely fill the space indicated in blue. Avoid getting blood on the disc.

~ Press down firmly on the hinge cover to close the disc.

~ Ensure that the area where the sample is applied and the outer disc of the hinged cover are free of blood.

~ Place the disc in the Cobas device.

~ Measurement starts automatically when the lid is closed.

3.6.3. TyG-index

The TyG index was a practical and approachable tool that strongly correlates with both HbA1c and IR for assessing long-term blood glucose in patients with T2D. This indicator has the advantage of being used in a clinical setting because TG and glucose levels are easily and frequently monitored (Hameed, E. K., 2019).

The ln [fasting TG level (mg/dL) × FSG level (mg/dL)/2] was used to calculate the TyG-index (Hameed, E. K., 2019).

3.7. Statistical Analysis

The Microsoft Excel 2013 and SPS version 20.0 were used. The data was presented as numbers with a mean and standard deviation. A paired t-test for two dependent means was

used to determine the significance of the difference. Pearson's correlation coefficient was used to determine the correlation of parameters, using $p<0.05$ being the lowest level of significance.

4. RESULTS

In Baghdad/ Iraq, a cross-sectional hospital-based study was conducted to investigate the biochemical risk variables for CVD in diabetes patients.Acomparative analysis was carried out to discover if there were any differences between the study groups. The findings of this study included 150 Iraqi patients separated into three groups: 50 diabetic patients without CVD (T2D group) and 50 diabetic patients with CVD (T2D & CVD group), with the third group serving as a healthy control group (50 healthy subjects). All of the research participants came from Baghdad's Medical City Center. FSG, HbA1c, T-Ch, TG, LDL, HDL, Oxid-LDL, TyG-index, and CRP were all measured in this research.

All parameters' mean and SD were calculated and compared between the T2D and T2D&CVD patient groups, as well as to the control group.

4.1. Age Distribution

The study age group was separated into two groups (Age <50 and Age ≥50). The majorityof the research patients in the T2D & CVD group were more than 50 years (n= 43, 86%) and in the same age group were (N= 31, 62%) in the T2D group, more than control group (N= 21, 42%) while the majority of the individuals in the control group were less than 50 years (N= 29, 58%) (Table 4.1, Figure 4.1).

Table 4.1. Age distribution for patients with Control, T2D , and T2D & CVD groups.

Groups	Age (Year)	N (%)	Mean ± SD
T2D	< 50	19 (38 %)	46.62±1.40
	≥ 50	31(62%)	58.67±6.68
	Total	50 (100 %)	53.72±7.07
T2D & CVD	< 50	7 (14%)	49.86±3.2
	≥ 50	43 (86%)	61.40±5.99
	Total	50 (100 %)	60.12±5.01
Control	< 50	29 (58%)	44.45±3.38
	≥50	21 (42%)	56.11±7.37
	Total	50 (100 %)	49.32 ± 6.99
P-value	Chi-square =20.89, DF= 2.0 , P-value <0.001		

SD: Standard deviation. N: Number of patients. %: Percentage.

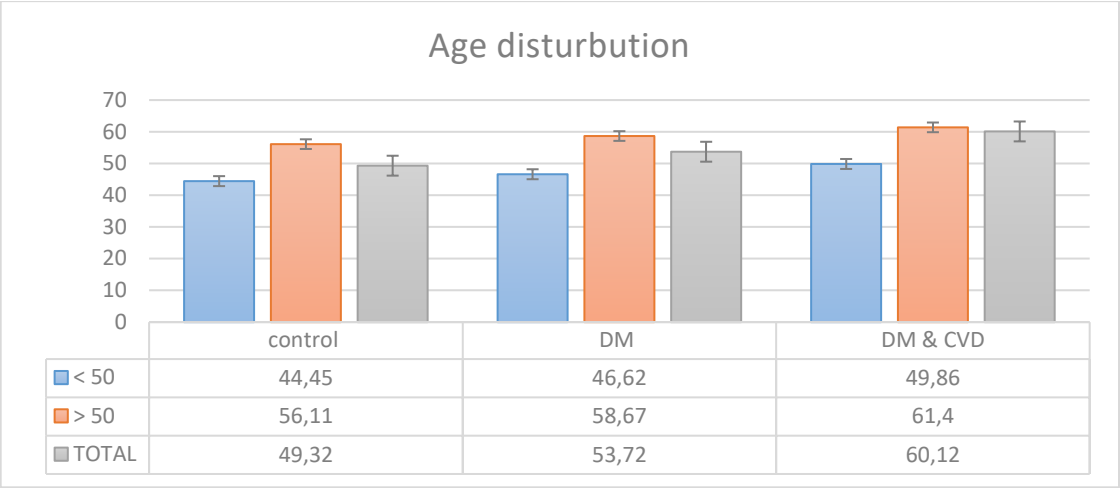

Figure 4.1. Age distribution for patients with Control, T2D , and (T2D & CVD) groups.

The findings revealed that there were significantly increase of age in T2D group (53.14±7.07 years; P= 0.02) and T2D &CVD group (60.12±5.01 years; P= 0.001) as compared to the control group (49.32 ± 6.99 years). In addition, statistical examination of the findings revealed that T2D & CVD group was significantly greater than the values seen in the T2D group (P = 0.012) (Table 4.2, Figure 4.2).

Table 4.2. The mean of Age for patients with Control, T2D , and T2D & CVD groups.

Groups Parameter	**Control (N= 50)**	**T2D (N= 50)**	**T2D &CVD (N= 50)**
Age (Year) (mean ± SD)	49.32 ± 6.99	53.14±7.07[a]	60.12±5.01[a,b]

a: statistically significant when compared to control. b: statistically significant when compared to T2D group. SD: standard deviation. N: Number of patients.

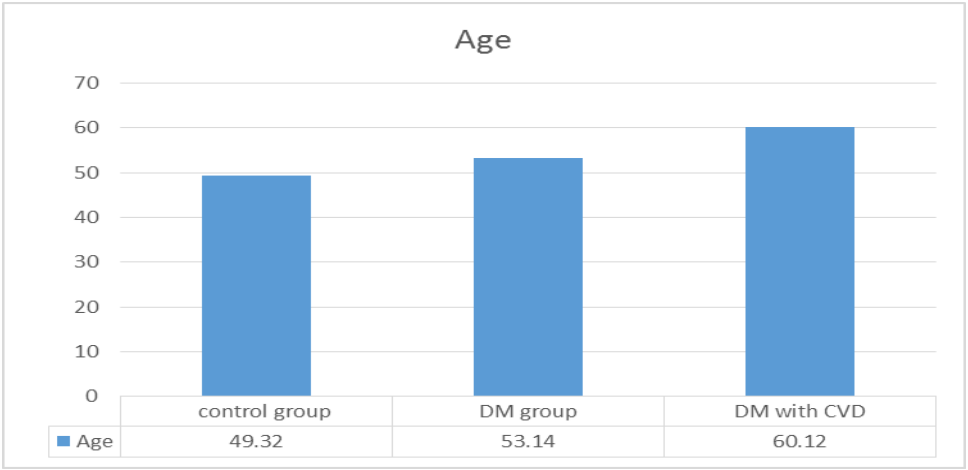

Table 4.2. The mean of Age for Participants with Control, T2D , and (T2D & CVD) groups.

4.2. Gender Distribution

Total of 150 patients; 88 male (59%) and 62 females (41%) were studied. Gender distribution for patients with T2D , T2D & CVD and control groups was shown in Table 4.3.

Table 4.3. Gender distribution for patients with Control, T2D , and T2D & CVD groups.

Groups / **Gender**	**Control (N= 50)**	**T2D (N= 50)**	**T2D & CVD (N= 50)**	**Total**
Male	29 (58%)	22 (44%)	37 (74%)	88 (100%)
Female	21 (42%)	28 (56%)	13 (26%)	62 (100%)
Total	50 (100%)	50 (100%)	50 (100%)	150 (100%)
p-value	Chi-square =9.23, DF= 2.0 , P-value =0.0099			

SD: Standard deviation. N: Number of patients. %: Percentage.

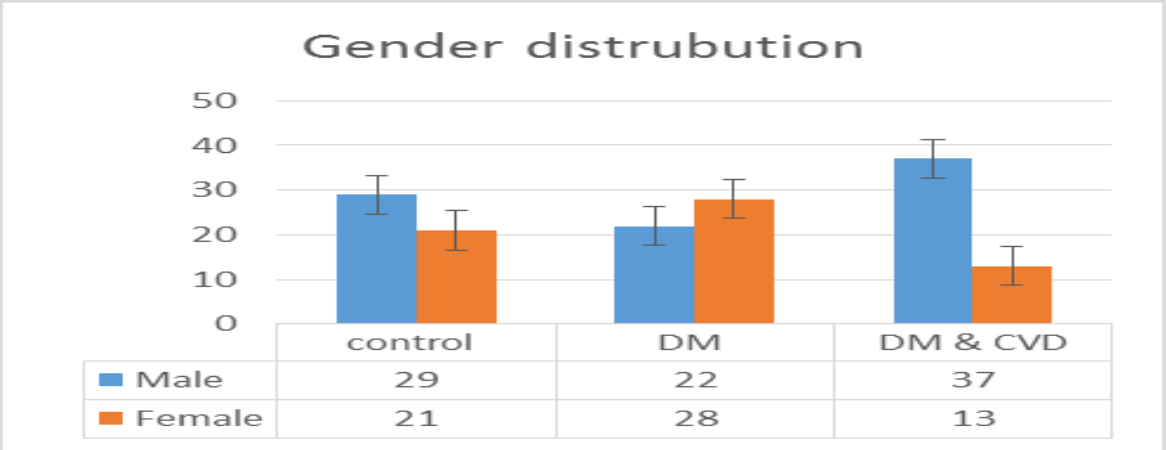

Figure 4.3. Gender distribution for patients with Control, T2D , and (T2D & CVD) groups.

4.3. BMI Distribution

The findings showed the number of obese patients were 30 (60 %) in T2D group, 21 (42 %) in the T2D & CVD and 18 (36 %) in the control group and the number of non-obese was 32 (64 %) in control group, 29 (58%) in the T2D & CVD and 20 (40 %) in the T2D group. Table 4.4 and Figure 4.4.

Table 4.4. BMI distribution for patients with Control, T2D , and T2D & CVD groups.

Groups	BMI (kg/m2)	N (%)	Mean ± SD
T2D	Obese	30 (60 %)	31.45±4.1
	None –obese	20 (40 %)	25.83±1.46
	Total	50 (100 %)	31.14± 3.22
T2D & CVD	Obese	21(42 %)	32.49±4.50
	Non-obese	29 (58%)	27.40±1.99
	Total	50 (100 %)	29.12±2.94
Control	Obese	18 (36 %)	30.88± 3.49
	Non-obese	32 (64 %)	26.52± 1.82
	Total	50 (100 %)	28.78±2.90
p-value	Chi-square =6.23, DF= 2 , P-value =0.0433		

SD: Standard deviation. N: Number of patients. %: Percentage.

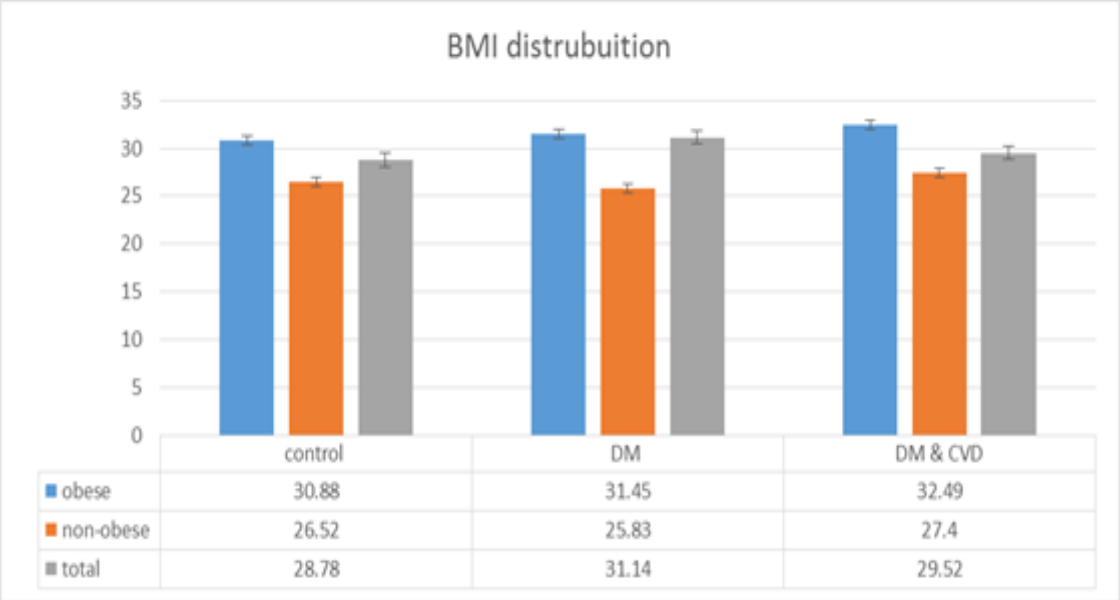

Figure 4.4. BMI distribution for patients with Control, T2D , and (T2D wih CVD) groups.

The results in Table 4.5 and Figure 4.5 showed a significant (P= 0.033) increase of BMI in T2D group (31.14± 3.22 kg/m^2) in comparison with control (28.78±2.90 kg/m^2) and a non-significant (P= 0.10) increase of BMI in T2D &CVD group (29.52±2.94 kg/m^2) in comparison with control group.

The results also showed that there was a significant (P= 0.012) decrease in T2D & CVD group when compared with that of T2D group.

Table 4.5. The mean BMI for patients with Control, T2D , and T2D &CVD groups.

Groups Parameter	Control (N= 50)	T2D (N= 50)	T2D &CVD (N= 50)
BMI (kg/m^2) (mean ± SD)	28.78±2.90	31.14± 3.22 [a]	29.12±2.94 [b]

a: Statistically significant when compared to control. b: statistically significant when compared to T2D group. SD: standard deviation. N: Number of patients.

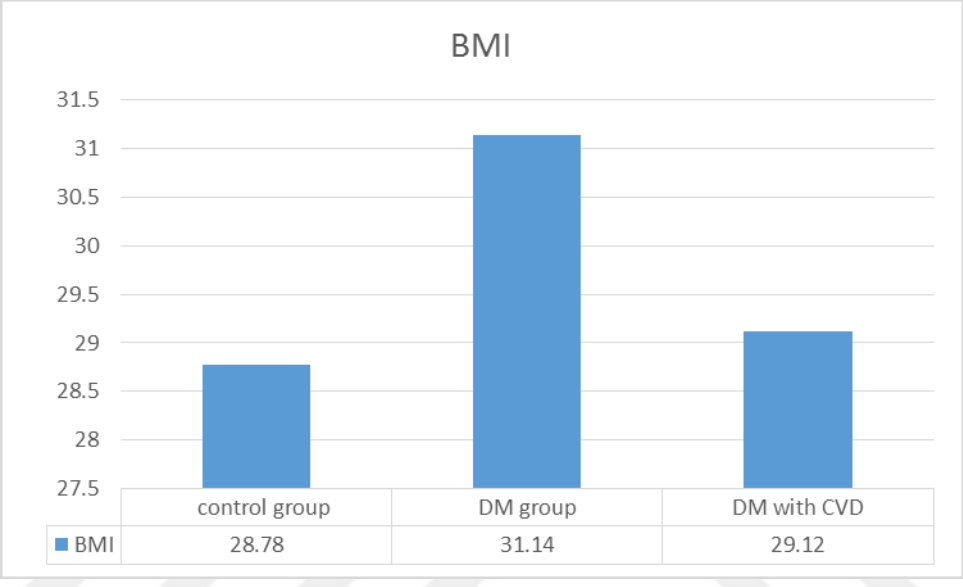

Figure 4.5. The mean of BMI for patients with Control, T2D , and (T2D & CVD) groups.

4.4. Blood Pressure

Blood pressure systolic (sBP) and diastolic (dBP) were calculated in all groups. The mean of sBP and dBP for these groups were shown in Table 4.6 and Figure 4.6. Our findings show that Hypertension with sBP of 140 mmHg and/or dBP of 90 mmHg was shown to be substantially more common in patients with T2D & CVD group (40%) than among of T2D group (32%) and control group (24%).

The mean of blood pressure is higher in both T2D (sBP= 134.1±17.19 mmHg; P=0.118 and dBP= 87.5±11.12 mmHg; P=0.033) and T2D & CVD groups (sBP= 142.7±21.9 mmHg; P<0.001 and dBP=94.3±16.03 mmHg; P=0.010) in comparison with control group (sBP= 128.8±14.05 mmHg and dBP=82.4±11.3 mmHg). The results also revealed that there were a significant (P<0.001) increase in sBP and dBP in T2D & CVD group when it was compared with that of T2D group.

Table 4.6. The blood pressure parameters for patients with Control, T2D , and T2D & CVD groups.

Groups Parameter	Control Group (N=50)	T2D Group (N=50)	T2D & CVD Group (N=50)
sBP mmHg mean±SD	128.8±14.05	134.1±17.19[a]	142.7±21.9[a,b]
dBP mmHg mean±SD	82.4±11.3	87.5±11.12[a]	94.3±16.03[a,b]

a: Statistically significant when compared to control. b: Statistically significant when compared to T2D group. SD: standard deviation. n= Number of patients.

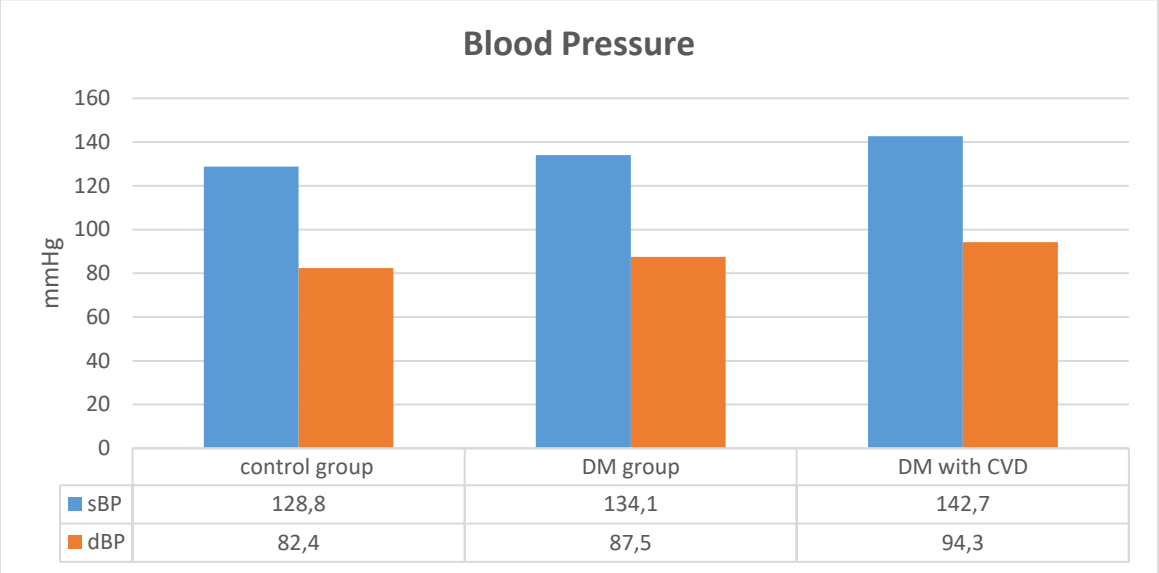

Figure 4.6. The blood pressure for patients with Control, T2D , and (T2D &CVD) groups.

4.5. Fasting Serum Glucose and HbA1c Levels

Fasting serum glucose and HbA1c levels were measured in all groups. The mean of FSG and HbA1c levels for these groups was shown in Table 4.7 and Figure 4.7. The statistical analysis of the results showed that there were a significant (P< 0.0001) increase of mean FSG and HbA1c level in T2D group (8.92±2.85 mmol/L and 8.06±1.03%) in comparison with control group (4.88±0.43 mmol/L and 5.11±0.50%) and a significant (P< 0.0001) increase of mean FSG and HbA1c level in (T2D & CVD) group (9.69±2.81 mmol/L and 8.23±1.43%) in comparison with control group. The results revealed that there were no significant difference in mean FSG (P= 0.692) and HbA1c level (P= 0.42) between the T2D and CVD group when it were compared with T2D group.

Table 4.7. The mean of Glucose level for patients with Control, T2D , and T2D & CVD groups.

Groups Parameter	Control Group (N=50)	T2D Group (N=50)	T2D & CVD Group (N=50)
FSG (mmol/L) mean±SD	4.88±0.43	8.92±2.85[a]	9.69±2.81[a]
HbA1c (%) mean±SD	5.11±0.50	8.06±1.03[a]	8.23±1.43[a]

a: Statistically significant when compared to control. SD:Standard deviation. N: Number of patients.

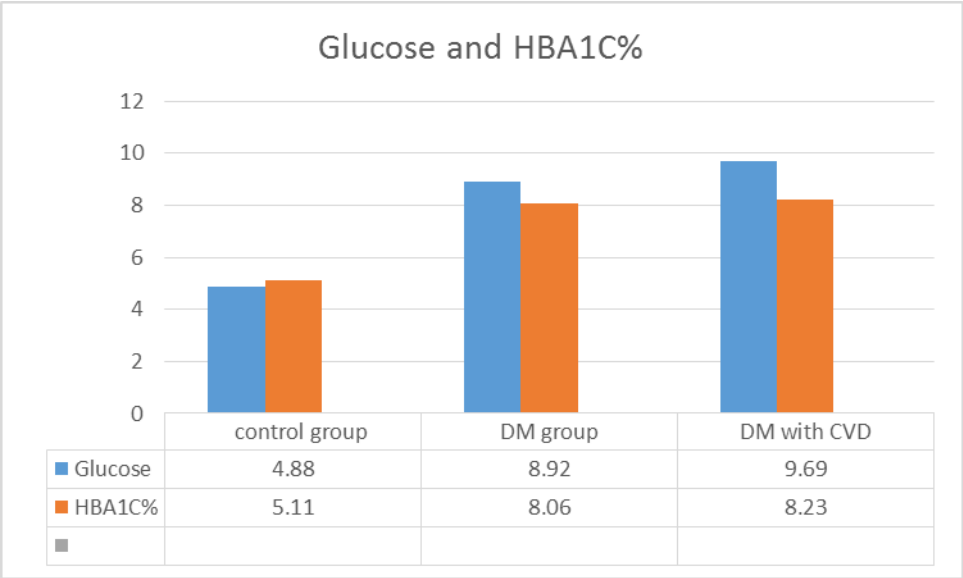

Figure 4.7. Serum glucose levels for patients with Control, T2D , and (T2D &CVD) groups.

4.6. Serum T-Ch, TG, HDL and LDL Levels

Dyslipidemia is frequent in diabetic people and has been linked to a higher risk of CVD. The results in Table 4.8 demonstrated the significantly increased in mean of serum T-Ch (P= 0.0031), TG (p= 0.0019), LDL (P=0.0029) levels accompanied with a low significant in the HDL (P= 0.002) level in the T2D group when compared to the control group.

The same results were founds in the T2D & CVD group (T-Ch; P= 0.001, TG; P= 0.001, LDL; P< 0.001 and HDL; P< 0.001) levels when compared to the control group.

Table 4.8. The mean of serum T-Ch, TG, HDL and LDL level for patients with Control, T2D , and T2D & CVD groups.

Parameter Groups	T-Ch (mmol/L) mean±SD	TG (mmol/L) mean±SD	HDL (mmol/L) mean±SD	LDL (mmol/L) mean±SD
Control	4.57±0.47	1.32±0.53	1.34±0.17	2.63±0.47
T2D	5.02±0.77*	1.84±0.67[a]	1.08±0.30[a]	3.05±0.81[a]
T2D & CVD	5.43±0.70*	2.03±0.81[a]	1.0±0.30[a]	3.48±0.72[a,b]

a: Statistically significant when compared to control. b: Statistically significant when compared to T2D group. SD: Standard deviation. N: Number of patients.

Moreover, in contrast to the T2D group, the results in the T2D & CVD group demonstrated a higher levels of serum T-Ch and TG and a lower HDL but statistically not significant (P= 0.056, P=0.186 and P=0.217). The findings also indicated that there were a substantial difference in LDL (P=0.0027) in T2D & CVD group when it were compared with that of T2D group Table 4.8 and Figure 4.8.

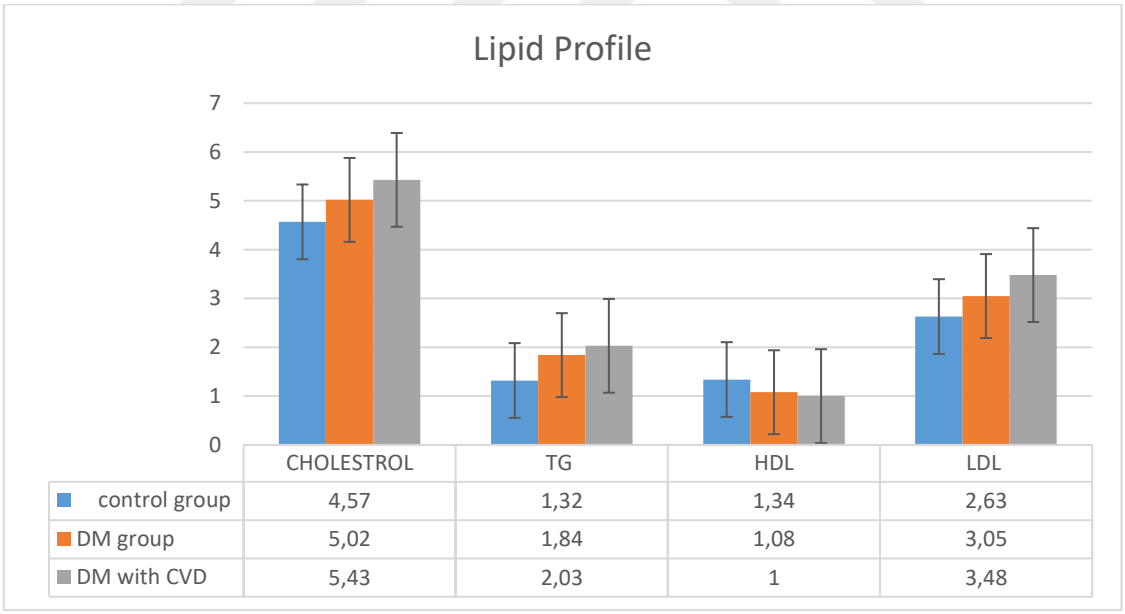

Figure 4.8. Serum TC, TG, HDL and LDL level for patients with Control, T2D , and (T2D & CVD) groups.

4.7. Serum Oxid-LDL Level

Oxid-LDL is higher in both T2D (67.88±17.65 mU/mL; P= 0.0020) and T2D & CVD (77.42±15.38 mU/mL; P<0.001) group in comparison with control group (55.08±12.01 mU/mL) as shown in Table 4.9. The results also indicated that there were a significant (P= 0.017) increase in T2D & CVD group (77.42±15.38 mU/mL) when it were compared with that of T2D group (67.88±17.65 mU/mL).

Table 4.9. The mean of Oxid-LDL level for patients with Control, T2D , and T2D & CVD groups.

Groups **Parameter**	**Control**	**T2D**	**T2D & CVD**
Oxid-LDL (mU/mL) mean±SD	55.08±12.01	67.88±17.65[a]	77.42±15.38[a,b]

a: Statistically significant when compared to control. b: Statistically significant when compared to T2D group. SD: Standard deviation. N: Number of patients.

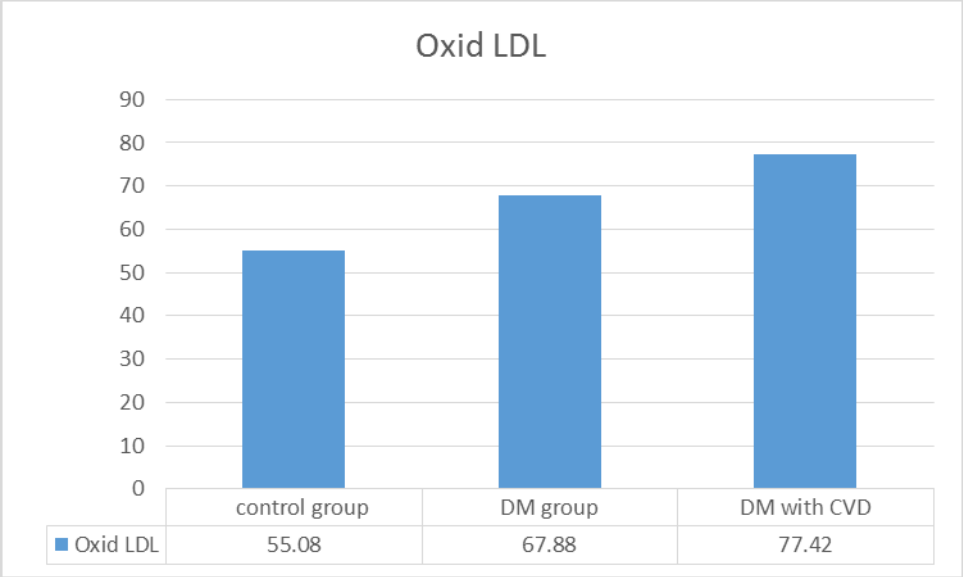

Figure 4.9. Serum Oxid LDL level for patients with Control, T2D , and (T2D & CVD) groups.

4.8. TyG-index

The mean of TyG-index for these groups was shown in Table 4.10 and Figure 4.10. TyG-indexwas higher in both T2D (5.08±0.28; P<0.001) and T2D & CVD (5.14±0.29; P<0.001) groups in comparison with control group (4.58±0.21).

The mean TyG-index level in T2D & CVD patients (5.14±0.29) was higher than the mean of T2D group (5.08±0.28) but statistically not significant (P= 0.364).

Table 4.10. The mean of TyG-index for patients with Control, T2D , and T2D & CVD groups.

Groups Parameter	Control	T2D	T2D & CVD
TyG-index mean±SD	4.58±0.21	5.08±0.28[a]	5.14±0.29[a]

a: Statistically significant when compared to control. SD: Standard deviation. N: Number of patients.

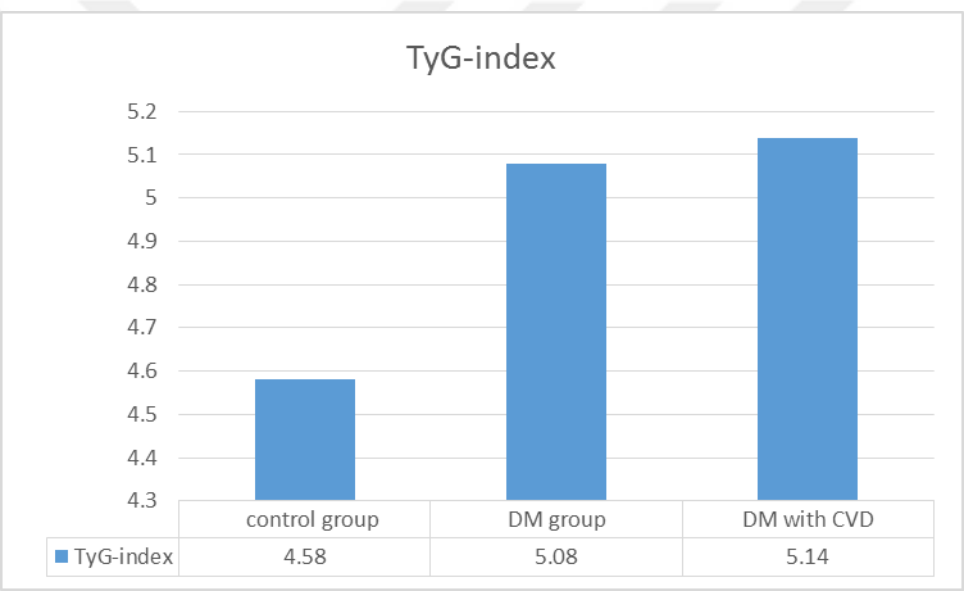

Figure 4.10. TyG index for patients with Control, T2D , and (T2D & CVD) groups.

4.9. Serum CRP Level

Serum CRP test was measured in all groups. The mean±SD of CRP for these groups was shown in Table 4.11. CRP is significantly (P<0.0001) higher in both T2D (11.25±5.28 mg/L) and T2D & CVD (17.39±9.69 mg/L) group in comparison with that of control group (3.55±1.92 mg/L).

The findings also indicated that there was a significant (P< 0.001) increase in T2D & CVD group (5.43±0.70 mg/L) when compared with T2D group.

Table 4.11. The mean of CRP level for patients with Control, T2D , and T2D & CVD groups.

Groups Parameter	Control	T2D	T2D & CVD
CRP (mg/L) mean±SD	3.55±1.92	11.25±5.28[a]	17.39±9.69[a,b]

a: Statistically significant when compared to control. b: Statistically significant when compared to T2D group. SD: Standard deviation. N: Number of patients.

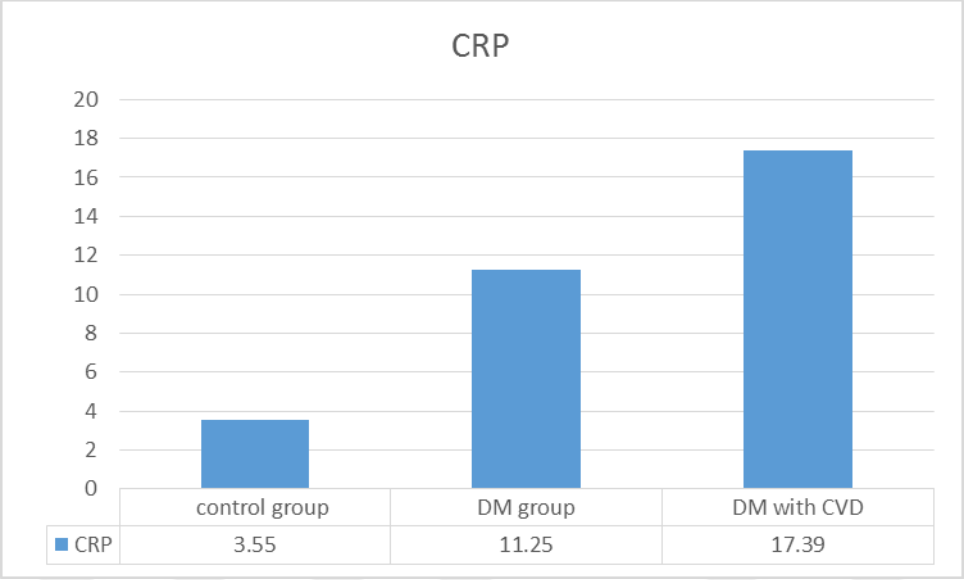

Figure 4.12. Serum CRP level for patients with Control, T2D , and (T2D & CVD) groups.

4.10. Comparison Between Patients with Good and Poor Glycemic Control

Glycemic control was classified as good (if HbA1c was less than 7.0 percent) and poor (if HbA1c was more than or equal to 7.0 percent). FSG, T-Ch, TG, LDL, HDL, Oxid-LDL, TyG-index, and CRP levels were tested in those who had poor glycemic control and those who had good glycemic control.

4.10.1. In T2D Group

The patients according to their glycemic control; FSG, TG, LDL, Oxid-LDL, TyG-index and CRP were significantly increased, and HDL significantly decreased in poor glycemic control when compared with good glycemic control as shown in Table 4.12.

Table 4.12, also shown that there were increased in T-Ch in poor glycemic control more than with good glycemic control but non-significant was found (P= 0.114).

Table 4.12. The mean of studied parameters level between patients with good and poor glycemic control in T2D group.

Groups / **Parameter**	**HbA1c %**		**T-test P- value**
	Good control (HbA1c < 7.0)	**Poor control (HbA1c ≥ 7.0)**	
FSG (mmol/L)	6.9156±1.01	10.5580 ±2.81	T-test =6.148 P< 0.001
CRP (mg/L)	5.51±0.56	11.82±5.51	T-test =3.362 P= 0.007
T-Ch (mmol/L)	4.7857± 0.712	5.2238± 0.750	T-test =1.65 P= 0.114
TG (mmol/L)	1.2400±0.26	2. 338±0.55	T-test =5.83 P < 0.001
HDL (mmol/L)	1.4337±0.177	0.9337±0.23	T-test =7.78 P< 0.001
LDL (mmol/L)	2.6373±458	3.4750±.61559	T-test =4.43 P= 0.002
Oxid-LDL (mU/ml)	54.55±10.94	75.14±13.53	T-test =6.418 P< 0.001
TyG-index	4.0780±0.26	5.1120±0.30	T-TEST =2.716 P= 0.0120

4.10.2. In T2D &CVD Group

The patients according to their glycemic control; Serum FSG, TG, Oxid-LDL, TyG-index and CRP were significantly increased in poor glycemic control more than with good glycemic control. While HDL significantly decreased in poor glycemic control than with good glycemic control.

There were increased in T-Ch and LDL in poor control more than with good control but non-significant as shown in Table 4.13.

Table 4.13. The mean of studied parameters level between patients with good and poor glycemic control in T2D &CVD group.

Groups / **Parameter**	**HbA1c %**		**T-test P- value**
	Good control (HbA1c < 7.0)	**Poor control (HbA1c ≥ 7.0)**	
FSG (mmol/L)	6.68±.59	10.11 ±1.65	T-test =6.54 P < 0.001
CRP (mg/L)	12.10 ±5.30	17.71 ±9.42	T-test =3.94 P =0.003
T-Ch (mmol/L)	5.35±.605	5.7333±.77	T-test =1.446 P =0.176
TG (mmol/L)	1.28±33	1.90±0.58	T-test =2.732 P =0.0189
HDL (mmol/L)	1.33±0.08	0.89±0.24	T-test =7.732 P <0.001
LDL (mmol/L)	3.2705±0.58	3.8044±0.81	T-test =2. 32 P =0.051
Oxid LDL (mU/ml)	69.70±5.99	81.43±12.91	T-test =4. 52 P =0.004
TyG-index	4.83±0.09	5.82±0.04	T-TEST =3. 55 P =0.009

5. DISCUSSION AND CONCLUSION

The study age group was separated into two groups (Age <50 and Age ≥50). The majority of the patients in the (T2D and CVD) group are more than 50 years (86%) and (62%) in the T2D group, while the majority of the individuals in the control group are less than 50 years (58%). The findings revealed that there were significantly increase of age in T2D group and T2D &CVD group as compared to the control group (Table 4.1 and Table 4.2). This result was consistent with earlier research (Leon, B. M. and Maddox, T. M., 2015) who reported that CVD mortality rates among people (> 18 years) with T2D were 1.7 times greater than those without diagnosed T2D in the United States as a result of a higher risk of stroke and MI. The Einarson, T. R. *et al.* (2018) study was reported that patients with T2D had a greater prevalence rate of CVD than adults without T2D . There are significant variations in the age-related rise in vascular-stiffness in elastic arteries of persons without diabetes compared to those in diabetic arteries, and their blood vessels appear to age at a quicker pace, beginning at a younger age and eventually reaching a functional plateau (Cameron, J. D. *et al.*, 2003).

Total of 150 patients; 88 male (59 %) and 62 females (41 %) were studied. Table 4.3. According to Einarson, T. R. *et al.* (2018), males had higher rates of prevalent disease than females, and CVD was responsible for 50.3% of all fatalities in T2D patients in a comprehensive review of 4,549,481 T2D patients, and CVD is associated with age and obesity, in addition to diabetes. In T2D individuals, both male and female have a higher risk of CVD death. When compared to individuals without T2D , the relative risk of CVD morbidity and death in persons with T2D varies from 1 to 3 for male and 2 to 5 for female (Leon, B. M. and Maddox, T. M., 2015).

According to our data, there were 60% of obese patients in the T2D group, 42% in the T2D & CVD group, and 36% in the control group. Obesity is a common comorbidity of T2D and has been linked to an increased risk of CVD morbidity and mortality (Leon, B. M. and Maddox, T. M., 2015). In 57 separate research, 4.5 million patients with T2D were evaluated, and heart failure was shown to be more prevalent in people with a BMI 30 kg/m^2 than in those with a BMI 30 kg/m^2, showing a 65% increase in prevalence due to obesity (Einarson, T. R. *et al.*, 2018).

Our findings in Table 4.5 also revealed a significant (P= 0.033) rise in BMI in the T2D group compared to the control group and a non-significant (P= 0.10) increase in BMI in the T2D &CVD group compared to the control group. The results also showed that there was a significant (P= 0.012) decrease in the T2D & CVD group when compared with that of T2D group. Similar results by Bhatti, G. K. *et al.* (2016) reported that obesity and higher CVD prevalence rates were found to have a beneficial association. Pre-diabetes is associated with IR and poor glucose tolerance, which raises the risk of CVD. Hyperglycemia was the primary predictor of T2D micro-vascular complications and plays a key role in CVD etiology. The pancreas normally produces adequate insulin, but the body cannot utilize it properly for unexplained reasons, resulting in IR (Adler, A. *et al.*, 2021). It was linked to obesity, particularly central obesity, although it can also be seen in healthy people with high blood pressure (Petrie, J. R. *et al.*, 2018).

Blood pressure systolic (sBP), diastolic (dBP) was measured in all groups. The mean of sBP and dBP for these groups was shown in Table 4.6. Our findings were showed that the mean of blood pressure was higher in both T2D and T2D & CVD groups in comparison with control group. Hypertension is a prevalent comorbidity in diabetic people and a major CVD risk factor (Leon, B. M. and Maddox, T. M., 2015). Furthermore, diabetes raises vascular tone, enhances salt retention, and contributes to nephropathy, all of which predispose persons to hypertension. Although both IR and hyperinsulinemia can contribute to hypertension in T2D patients, aortic stiffness as measured by aortic pulse wave velocity has been shown to predict future CVD (Kalofoutis, C. *et al.*, 2007). Atherosclerosis, endothelial dysfunction, arterial remodelling, vascular inflammation, obesity and dyslipidemia are all risk factors for both diabetes and hypertension. The cardiovascular consequences of diabetes and hypertension, which are predominantly linked to disease of microvascular and macrovascular, also overlap significantly. The strong association between diabetes and hypertension is likely due to common processes such as up regulation of the renin-angiotensin-aldosterone-system, inflammation, oxidative stress, and immune system activation (Petrie, J. R. *et al.*, 2018).

In the Table 4.7, the fasting serum glucose and HbA1c levels were measured in all groups. The statistical analysis of the results showed that there were significant (P< 0.0001) increase of mean FSG and HbA1c level in T2D group and the T2D & CVD group in comparison with the control group. The results also revealed that there was no significant difference in mean FSG (P= 0.692) and HbA1c level (P= 0.42) in the T2D & CVD group when it was

compared with T2D group. Cosentino, F. *et al.* (2020) discovered that the elevated risk of CAD begins at glucose levels below the T2D cut-off point (7 mmol/L) and increases as glucose levels rise. Similarly, Strain, W. D. and Paldánius, P. M. (2018) discovered that better glycemic control delays the onset, slows the progression, and (in some circumstances) partially reverses signs of microvascular abnormalities in diabetic individuals. Furthermore, the significance of glucose control in the prevention and treatment of macrovascular disease is currently being debated. While hyperglycemia was clearly associated with an increased risk of CVD (Valensi, P. *et al.*, 2021).

Einarson, T. R. *et al.* (2018) declared that people with T2D had a higher CVD prevalence rate than adults without T2D in the past, and that the risk of CVD increases continually as glucose levels rise. Hyperglycemia harms the vascular smooth muscle cells, endothelium, and macrophages, causing thrombosis and fibrinolysis, which contribute to plaque development.

Dyslipidemia is frequent in diabetic people and has been linked to a higher risk of CVD. The results in the Table 4.8 demonstrated the significantly increased in mean of serum T-Ch (P= 0.0031), TG (P= 0.0019), LD (P= 0.0029) levels accompanied with a low significant in the HDL (P= 0.002) level in the T2D group and the T2D & CVD group (T-Ch; P= 0.001, TG; P= 0.001, LDL; P< 0.001 and HDL; P< 0.001) levels when compared to the control group. Moreover, in contrast to the T2D group, the results in the T2D & CVD group demonstrated a higher levels of serum T-Ch and TG and a lower HDL but were statistically not significant (Table 4.8). Similar study was reported by Kalofoutis, C. *et al.*, (2007) that dyslipidemia is a known risk factor for CAD in both diabetic and non-diabetic individuals, and it was thought to play a major role in the increased CVD risk associated with T2D . Elevated TG levels, Low levels of HDL, and abnormalities in the size and distribution of lipoprotein particle subclasses. Atherogenic lipid abnormalities in T2D patients may be caused by a number of processes. The combination of fat accumulation and genetic susceptibility was hypothesized to cause adiposopathy, or dysfunctional adipose tissue. When compared to normal adipose tissue, evidence shows that this dysfunctional adipose tissue was less responsive to insulin and has lower hormone-sensitive-lipase activity. As a result, there was more intracellular TG breakdown and more FFA released into the circulation. In the end, this leads to IR in the liver and muscle, and may worsen it (Kalofoutis, C. *et al.*, 2007). Those with IR had significantly higher levels of TG, LDL, and HDL than those with normal insulin sensitivity (Olaogun, I. *et al.*, 2020). CVD is caused by a multitude of metabolic alterations that occur

during IR. For example, IR can cause an imbalance in glucose metabolism, leading in chronic hyperglycemia, which promotes oxidative stress and an inflammatory response, both of which cause cell damage. IR may also have an impact on systemic lipid metabolism, resulting in dyslipidemia and the lipid triad. The most prevalent symptoms were elevated TG levels, low HDL levels, and the development of small dense LDL. These triads, as well as endothelial dysfunction, which can be triggered by aberrant insulin signaling, all contribute to the formation of atherosclerotic plaques. In terms of the systemic effects of IR and metabolic cardiac changes, it can be inferred that IR in the myocardium causes harm through at least three mechanisms: (1) signal transduction disruption, (2) defective substrate metabolism control, and (3) changed substrate supply to the myocardial (Ormazabal, V. *et al.*, 2018; Petrie, J. R. *et al.*, 2018; Ma, X. *et al.*, 2020).

Oxid-LDL was higher in both T2D (P= 0.0020) and T2D & CVD (P< 0.001) groups in comparison with control group as shown in Table 4.9. The results also indicated a significant (P= 0.017) increase in T2D & CVD group when compared with that of T2D group. The finding results agree with Xie, L. *et al.* (2019) study were confirmed serum Oxid-LDL levels were considerably greater in patients with T2D with microvascular problems than in T2D patients without microvascular complications or controls, suggesting that elevated serum Oxid-LDL was linked to microvascular issues in T2D patients. Endothelial cells, platelets, macrophages, smooth muscle cells and fibroblasts, are all affected by Oxid-LDL, which plays a key role in atherosclerosis. Endothelial dysfunction was caused by Oxid-LDL, which activates apoptotic pathways, raises ROS, and causes endothelial dysfunction. They promote proliferation, migration, and collagen production in vascular smooth muscle cells and fibroblasts. They also cause plaque instability and thrombosis by stimulating the production of metalloproteinase (Kattoor, J. A. *et al.*, 2019).

The TyG-index was a new measure for metabolic disease that has recently been linked to the risk of CVD (Menini, S. *et al.*, 2020). In the Table 4.10, the TyG-index was higher (P< 0.001) in both T2D and T2D & CVD groups in comparison with control group. As well as, the mean TyG-index level in T2D & CVD patients was higher than the mean of T2D group, but statistically not significant (P= 0.364). The finding results agree with Ma, X. *et al.*, (2020). . It has been demonstrated that the TyG-index was positively linked with future CVD, suggesting that it might be a helpful marker for predicting clinical outcomes in individuals with T2D and ACS patients. According to a study by Wang, L. *et al.* (2020), the TyG-index was related with not only the CVD incidence, but also with the development of

T2D, these findings show that the TyG-index might be used to predict of risk CVD in people with diabetes and CAD. Finally, IR-induced metabolic alterations have been linked to the progress of CVD. IR may create an imbalance in glucose metabolism, leading to prolonged hyperglycemia, which produces oxidative stress, and an inflammatory response, resulting in vascular-endothelial damage of cell (Ma, X. *et al.*, 2020).

Serum CRP test was measured in all groups (Table 4.11). CRP was considerably ($P< 0.0001$) higher in the T2D and T2D &CVD groups than in the control group. The data also revealed a substantial ($P< 0.001$) rise in the T2D & CVD group when compared to the T2D group. Serum CRP was an inflammatory marker, and it has been associated to a range of ailments, including diabetes and cardiovascular disease. In the general population, CRP has been linked to an increased risk of T2D and dyslipidemia (Jeong, M.-J. *et al.*, 2019). In a cohort of diabetic individuals, mean hs-CRP was shown to have a substantial high correlation and was connected to an elevated CVD risk (Faradonbeh, N. A. *et al.*, 2018; V Valensi, P. *et al.*, 2021).

Glycemic control in our results were classified as good (if HbA1c was less than 7.0 %) and poor (if HbA1c was more than or equal to 7.0 %). The patients according to their glycemic control; FSG, T-Ch, TG, LDL, Oxid-LDL, TyG-index and CRP were increased (HDL decreased) in poor glycemic control when compared with good glycemic control as shown in T2D Group (Table 4.12) and T2D &CVD group in (Table 4.13). According to several studies, HbA1c was the gold standard for predicting long-term glycemic control and medical treatment response in diabetic patients. HbA1c, on the other hand, was ineffective in addressing glucose fluctuations. Glycemic variability refers to fluctuations in blood glucose levels. In diabetic patients, glycemic fluctuation has been associated with macrovascular and microvascular complications, as well as death (Martinez, M. *et al.*, 2021). Eckel, R. H. *et al.* (2021) reported that T2D has long been recognized as a substantial risk factor for CVD, but the mechanisms of this association remain unknown. Various pathophysiological pathways may be implicated. Lipids and other risk variables are definitely essential, but glucose was not, thus clinical research that failed to demonstrate that strict glycemic control lowers cardiovascular events were not worthwhile. Diabetes, particularly T2D, was linked to dyslipidemia, hypertension, and obesity. The poor glycemic control groups had higher T-Ch levels, higher serum LDL, higher TG, and lower serum HDL values than the excellent glycemic control groups, as indicated in Tables 4.12 and 4.13. A significant percentage of diabetic patients may be classified as being at high risk for CVD. Furthermore, by increasing

a person's lipid profile, glycemic control can help them avoid a high CVD risk. Poor glycemic control was linked to an increased risk of CVD and death, according to Li, F.-R. *et al.*, (2021).

Mullugeta, Y. *et al.*, (2012) found that HbA1c had a direct and significant relationship with T-Ch, TG, and LDL, as well as an inverse relationship with HDL, after analyzing laboratory data from T2D patients. Apart from its core purpose in monitoring long-term glycemic control, he believes HbA1c can give useful additional information regarding the degree of circulating lipids. The link between glycemic management and lipid levels emphasizes the need of optimizing dyslipidemia in addition to correcting hyperglycemia for cardiovascular health.

As demonstrated in Tables 4.12 and 4.13, elevated serum CRP, Oxid LDL, and TyG-index were all connected significantly to poor glycemic control in diabetic individuals with or without CVD. Based on Hermans, M. P. *et al.* (2019) study, patients with elevated CRP had increased HbA1c, BMI, and insulinemia. They showed higher levels of atherogenic dyslipidemia and non-HDL.

High amounts of lipids in diabetic individuals with or without CVD; might be the cause of increased Oxid-LDL levels in the poor glycemic group. Ganjifrockwala, F. *et al.*, (2016) found a significant positive connection of Oxid-LDL with TG, T-Ch, and LDL in diabetics. Our findings were also in line with those of Jiang, Z.-z. *et al.*, (2022), who found that a high TyG-index value was linked to a high carotid plaque load in those with pre-diabetes and new-onset T2D. The TyG-index was linked to not only the incidence of CVD, but also the development of T2D. According to the research by Wang, L. *et al.*, (2020), the TyG-index might be used to predict future CVD risk in diabetic individuals. Clinicians should pay close attention to the TyG index and Oxid-LDL in Iraqi populations, as they may be able to help these patients benefit from early therapy. Furthermore, dyslipdia and poor glycemic management increase the risk of CVD in T2D patients.

5.1. CONCLUSION

1. In T2D patients, older age (over 50 years) was associated with a higher risk of developing CVD.

2. In T2D patients, being male was related with a higher risk of developing CVD.

3. In T2D patients, high blood pressure are substantial risk factors for CVD.
4. In T2D patients, obesity and overweight were shown to be more strongly linked to an elevated risk of CVD.
5. In T2D patients, high blood glucose levels may raise the risk of CVD development.
6. In T2D patients, a high HbA1C level (>7.0%) increases the risk of CVD development.
7. Patients with T2D or/and CVD have higher levels of both Oxid-LDL and TyG-index. These data show that the Oxid-LDL and TyG-index may be valuable markers for risk and prognosis in diabetic and cardiovascular patients.
8. In T2D patients, high HbA1C levels are linked to dyslipidemia and can be utilized to predict CVD.
9. In T2D patients, high LDL-Ch cholesterol levels and low HDL-Ch cholesterol levels are also linked to an increased risk of CVD.
10. In T2D patients, high levels of CRP can be utilized as predictors of CVD.

To further understand the bidirectional association between diabetes and cardiovascular disease, particularly the pathogenesis of diabetic cardiomyopathy, more research was needed. As well as more investigation into the involvement of hypoglycemia in the incidence of CVD mortality was required.

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Appendixes

Appendix 1: Questionnaire

(1) Name

(2) No

(3) Weight

(4) Height

(5) BMI

(4) live

(5) Old

(6) History of Cardiovascular disease

Atherosclerosis

Heard failure

Cardiomyopathy

Congenital heart disease

Heart valve disease

Other:

(7) History of the following conditions?

-Diabetes	Yes	No
-Smoking history	Yes	No
-Family History of CVD	Yes	No
-Blood pressure	Yes	No
-Kidney disease	Yes	No
-Liver disease	Yes	No
-Thyroid disease	Yes	No
-Cancer	Yes	No

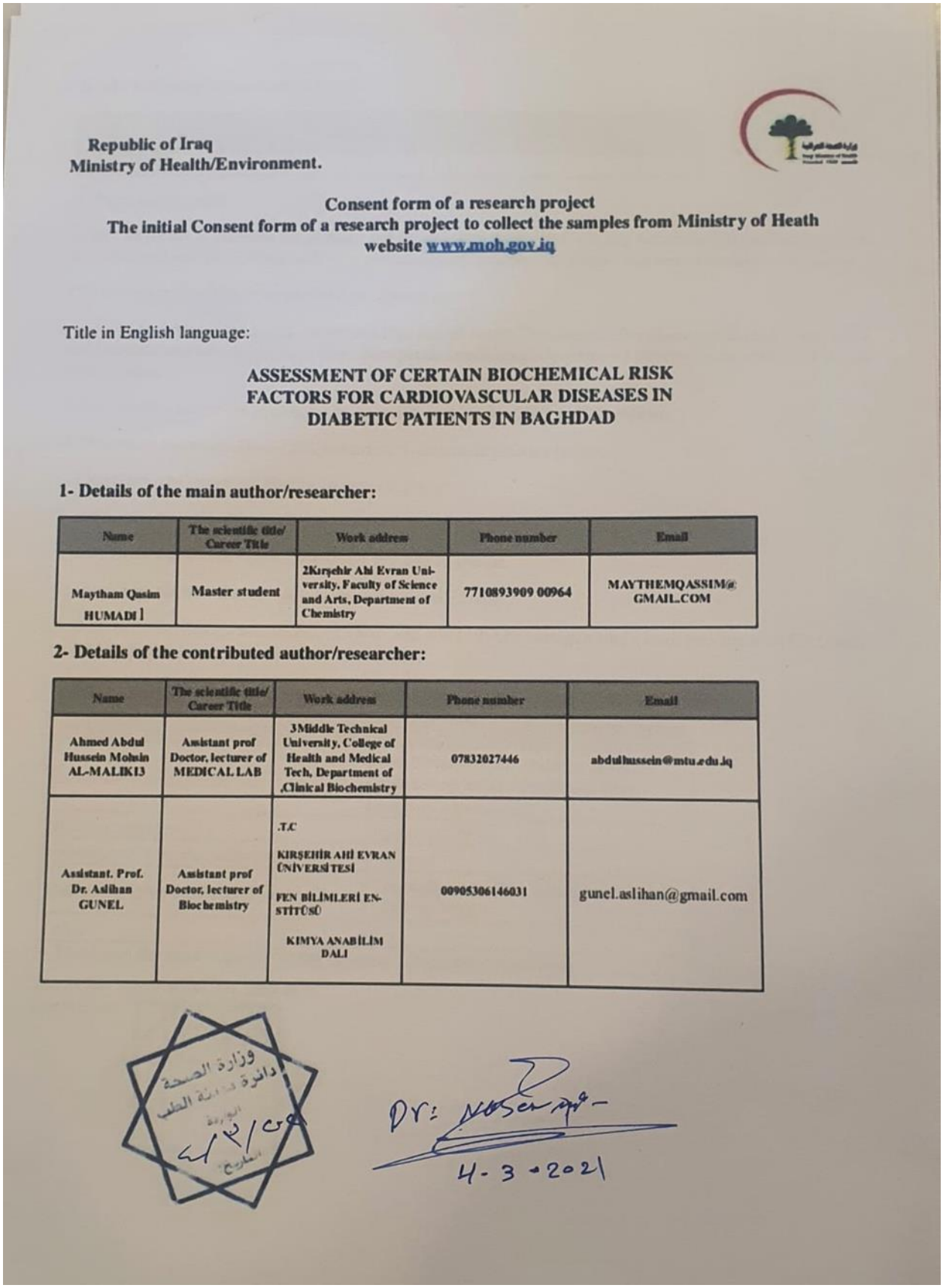
-Others

Appendix 2:

The tools and instruments that used in this study

Appendix 3:

Permission to collection of samples to complete a master's thesis



Republic of Iraq
Ministry of Health/Environment.

Consent form of a research project
The initial Consent form of a research project to collect the samples from Ministry of Heath website www.moh.gov.iq

Title in English language:

ASSESSMENT OF CERTAIN BIOCHEMICAL RISK FACTORS FOR CARDIOVASCULAR DISEASES IN DIABETIC PATIENTS IN BAGHDAD

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3- The Scientific Supervisor/ if found

Name	The scientific title/ Career Title	Work address	Phone number	Email
None				

A. Work background:

B. this thesis are to examine the primary and possible development CVD risk factors in T2D patients, as well as to comprehend the fundamentals of a prediction tool to avoid the disease's severe vascular consequences

C. The importance of the research and its objectives:

1. Determine which patients are at a high risk of future CVD events. The discovery of readily available and accurate markers might have huge therapeutic implications in terms of improving recurrent CVD risk classification.

2. Determine whether the TyG-index has predictive value for CVD in T2D patients.

3. Determine the serum Oxid-LDL as a predictive value in patients groups.

4. Determine the impact of obesity on patients groups.

5. Determine the role of main risk factors for CVD in patients groups, such as blood pressure.

6. Determine the role of serum CRP levels in patients groups.

7. Determine the function of dyslipidemia in patients groups.

8. Determine the factors associated with poor and good HbA1c management in Iraqi patients with CVD and/ or DM patients.

4- People or martials that are required for this research from the Ministry of Health.?

Martials	Type
Laboratories samples blood	Yes, blood samples, patients and con- . trols length, twits, weight and Blood pressure measurement
Martials/ equipment's	(No need (Privet
information from the patients Records	No need
Patients or other staff members	The study included a total of 150 male AND FEMALE
Others	No

5- Time and the date to perform the research: (suggested locations)

Time: 1st April 2021- 1st Jun 2022.
Locations:

وزارة الصحة
دائرة [illegible] الطب
٤/٣/٢٠٢١
التاريخ

DR: [signature]
4-3-2021

Name of institute	Approval
Medical city department	

6- Fund: None.
7- Methodology:
A. Study design: Blood samples and information were collected from 50 cardiac patients with 50 diabetic patients with 50 healthy controls
B. Studying the expansion of K. pneumonia in different age categories in both sexes..
C. The expected number of sampling: 100 patients and 50 controls
D. Statistical analysis: statistical descriptive tables and maths correlations.
E. Ethical consideration during research: patients names must not mention

F. Signed Commitment:
This is **Maytham Qasim HUMADI1** signed below to commit that I perform the research according to this protocol. Also, I commit that I will never change or modify it after it is being approved unless agreed with research committee in the health institute. Moreover, I commit following the laws, rules and instructions of Iraqi health ministry and any other official parties that follow the scientific and ethical commitment for research.

Name and the signature of the main researcher: **Maytham Qasim HUMADI1**
The name and the signature of the supervisor/ if the research is performed to obtain a BSc, MSc or PhD etc: Ahmed Abdul **Hussein Mohsin AL-MALIKI'**

Approval of the research committee at the health institution (or the body authorized to approve this form)

ASSESSMENT OF CERTAIN BIOCHEMICAL RISK FACTORS FOR CARDIOVASCULAR DISEASES IN DIABETIC PATIENTS IN BAGHDAD

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Abstract

Diabetes mellitus (DM) is expected to rise from 371 million people in 2013 to 552 million by 2030. Type 2 diabetes mellitus (T2D), which accounts for 90–95 percent of all cases, is to blame for the majority of the epidemic. Cardiovascular disease (CVD) is the primary cause of death in people with diabetes, with CVD accounting for more than half of all deaths. Traditional risk factors such as elevated levels of hyperlipidemia and blood pressure, as well as smoking and obesity, are all risk factors for severe cardiovascular events. In consequence, insulin resistance (IR) and hyperglycemia raise the risk of CVD complications. The goal of this study is to look at the link between T2D and CVD in the Iraqi population, as well as how these parameters relate to CVD risk factors. There is little research on Iraqis in this sector. A total of 150 people over the age of 40 were recruited (88 men and 62 women). There were 50 "healthy" people (29 males and 21 females) with no history of CVD, DM, or other illness as control group, 50 T2D patients (22 males and 28 females) as DM group, and 50 T2D with CVD patients (37 males and 13 females) as (DM & CVD) group. From April to November 2021, these individuals will be recorded at Baghdad Teaching Hospital and all participants completed an interview-administered questionnaire. Serum FBG, hemoglobin A1c (HbA1c), serum C-reactive protein (CRP), total cholesterol (T-Ch), triglycerides (TG), Triglyceride-glucose index (TyG-index), low-density lipoprotein cholesterol (LDL-Ch), high-density lipoprotein cholesterol (HDL-Ch), and levels were measured by photometric transmission method using the Cobas c111 system measurement using kit (Roche/Germany), while Oxidized LDL (Oxid-LDL) was quantified using the competitive-ELISA approach (Cusabio/United States) and compared to the control group as well as across patient groups. DM patients group had a greater prevalence of age (over 50 years), obesity, and high blood pressure. (DM & CVD) patients group had significantly higher glucose and HbA1c levels, as well as Oxid-LDL, TyG-index, CRP, TG, LDL, and lower levels of HDL when compared to DM group and control group. The increased risk of CVD in T2D patients is a result of these findings. The current study also discovered that individuals with poor HbA1c control are more likely to experience

((استثمار الطاقة النظيفة طريقنا نحو التنمية المستدامة))

الى / كلية التقنيات الصحية والطبية -بغداد

م/ اشراف مشترك

تحية طيبة...

حصلت الموافقة على ما جاء بمضمون كتابكم ذي العدد 1124/27/7 في 2022/3/7 والمتضمن الاشراف المشترك من قبل أ.م.د احمد عبد الحسين محسن على طالب الماجستير (ميثم قاسم حمادي) / جامعة Kirsehir hi Evran University وعلى ان لا يتجاوز سقف الاشراف (الداخلي والخارجي) وحسب التعليمات.

للتفضل بالاطلاع واتخاذ مايلزم مع التقدير ...

د. عمار عرب بداي

مدير قسم شؤون الدراسات العليا

2022/3/22

نسخة منه الى:

- قسم شؤون الدراسات العليا مع الاوليات.
- ملفة الكتب الصادرة.

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