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INVESTIGATION OF THE USE OF SERUM FGF-23 AS AN INDICATOR OF EARLY DIYABETCI NEPHROPATHY IN PATIENTS WITH TYPE 2 DIABETES

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By Ola Hussein Ali ALGHURAIRI

February 2023

We certify that we have read this thesis and that in our opinion it is fully adequate, in scope and in quality, as a thesis for the degree of Master of Science

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ABSTRACT

INVESTIGATION OF THE USE OF SERUM FGF-23 AS AN INDICATOR OF EARLY DIYABETCI NEPHROPATHY IN PATIENTS WITH TYPE 2 DIABETES

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Master of Science in Chemistry

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February 2023

Diabetes mellitus (DM) is a fatal metabolic disorder The prevalence of this disease has significantly increased in the past couple of decades. It is caused by the continuous increase in the severity and incidence of the disease. Despite the increasing prevalence of diabetes, the number of people with diabetic kidney disease (DKD) has not decreased over the past 30 years. One of the key factors contributing to a shorter lifetime among diabetics is this illness. to determine those who are most at risk of contracting this illness. This study aims To investigate the use of serum FGF-23 as an indicator of early diabetic nephropathy in patients with type 2 diabetes mellitus. 150 participants in all were chosen from those participating at 4 significant clinical laboratories. 100 individuals with type 2 diabetes mellitus were included in the patient group, 50 of them had microalbuminuria and the remaining 50 did not. The average patient age was (minimum=45,maximum=75) year and another 50 participants were healthy once for control. the patients used to attend Al-Yarmouk hospital and Al-Numaan hospital the samples of the patient has been collected from 5th April to 20th august of 2022 all the participants were assessed for whole blood (CBC,hba1c) and serum glucose, renal function test, and liver function test, FGF-23 and ACR (albumin creatinine ratio in urine). The SPSS program version 23 was used to statistically analyze the results. The present study's findings showed a significant difference in the mean serum FGF-23 levels across the groups during investigation. In all groups examined, there was a connection between ACR and serum FGF-23 levels. FGF-23 can be used as an alternative to ACR as an indicator of early diabetic nephropathy. FGF-23 can be useful as an indicator of the progression of diabetic nephropathy.

2023, 44 pages

Keywords: Fibroblast growth factor 23 (FGF23), Type 2 diabetes mellitus (T2DM)

ÖZET

TİP 2 DİYABETLİ HASTALARDA ERKEN DİYABETÇİ NEFROPATISİNİN GÖSTERGESİ OLARAK SERUM FGF-23 KULLANIMININ ARAŞTIRILMASI

Ola Hussein Ali ALGHURAIRI

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Diabetes mellitus (DM) ölümcül bir metabolik bozukluktur Bu hastalığın prevalansı son birkaç on yılda önemli ölçüde artmıştır. Hastalığın şiddetinin ve görülme sıklığının sürekli artmasından kaynaklanır. Diyabetin artan prevalansına rağmen, diyabetik böbrek hastalığı (DKD) olan kişilerin sayısı son 30 yılda azalmadı. Şeker hastalarının ömrünün kısalmasına katkıda bulunan en önemli faktörlerden biri bu hastalıktır. Bu hastalığa yakalanma riski en yüksek olanları belirlemek. Bu çalışma, tip 2 diabetes mellituslu hastalarda erken diyabetik nefropatinin bir göstergesi olarak serum FGF-23'ün kullanımını araştırmayı amaçlamaktadır. Toplamda 150 katılımcı, 4 önemli klinik laboratuvara katılanlar arasından seçildi. Hasta grubuna 100 tip 2 diabetes mellituslu birey dahil edildi, bunların 50'sinde mikroalbuminüri vardı ve geri kalan 50'sinde yoktu. Ortalama hasta yaşı (minimum=45,maksimum=75) yıldı ve diğer 50 katılımcı bir kez kontrol için sağlıklıydı. Al-Yarmouk hastanesine ve Al-Numaan hastanesine başvuran hastalardan alınan numuneler 5 Nisan - 20 Ağustos 2022 tarihleri arasında tüm katılımcılar tam kan (CBC,hba1c) ve serum glukozu, böbrek fonksiyon testi için değerlendirildi. ve karaciğer fonksiyon testi, FGF-23 ve ACR (idrarda albümin kreatinin oranı). Sonuçları istatistiksel olarak analiz etmek için SPSS program versiyonu 23 kullanıldı. Mevcut çalışmanın bulguları, araştırma sırasında gruplar arasında ortalama serum FGF-23 seviyelerinde anlamlı bir farklılık gösterdi. İncelenen tüm gruplarda ACR ile serum FGF-23 seviyeleri arasında bağlantı vardı. FGF-23, erken diyabetik nefropatinin bir göstergesi olarak ACR'ye alternatif olarak kullanılabilir. FGF-23, diyabetik nefropatinin ilerlemesinin bir göstergesi olarak faydalı olabilir.

2023, 44 sayfa

Anahtar Kelimeler: Fibroblast büyüme faktörü 23 (FGF23), Tip 2 diabetes mellitus (T2DM)

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Ola Hussein Ali ALGHURAIRI

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CONTENTS

LIST OF SYMBOLS

%	Percent
<	Less than
=	Equal
>	Greater than
±	Plus-minus
≤	Greater or equal to
≥	Less or equal to
°C	Degrees Celsius
µg/mg	Microgram per milligram
cm	Centimeters
dL	Deciliter
g	Gram
L	Liter
m mol/L	Millimole per liter
mg	Milligram
mg/dL	Milligram per deciliter
mg/g	Milligram per gram
mL	Milliliters
mmol	Millimoles
pg/ml	Picograms per millilitre

LIST OF ABBREVIATIONS

ACR	Albumin creatinine ratio
CBC	Complete blood count
CKD	Chronic kidney disease
CVD	Cardiovascular disease
DKD	Diabetes kidney disease
DM	Diabetes mellitus
DN	Diabetes nephropathy
eGFR	Estimated glomerular filtration rate
ESRD	End-stage renal disease
FBG	Fasting plasma glucose
FGF-23	Fibroblast growth factor
GDM	Gestational diabetes mellitus
GFR	Glomerular filtration rate
IDF	International diabetes federation
IDM	International diabetes federation
OGTT	Oral glucose tolerance test
PTH	Parathyroid hormone
T1DM	Type 1 diabetes mellitus
T2DM	Type 2 diabetes mellitus
T3CDM	Type-3c diabetes mellitus
UACR	Urine albumin-to-creatinine ratio
WHO	World health organization

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1. INTRODUCTION

Diabetes mellitus (DM) a type of metabolic disorder known as diabetes mellitus is characterized by a chronic high-glycemic condition that can be caused by a lack of insulin secretion. Insufficient glucose-insulin signaling is another factor contributing to it (Conroy *et al*. 2003). The number of people with diabetes is expected to double by 2030 as the disease continues to rise globally. According to the WHO, 350 million people worldwide will have diabetes at that time. By 2025, experts project a 64% increase in the disease's occurrence. (Rowley and Bezold 2012). In 2010, An estimated 285 million individuals worldwide have diabetes. By 2030, this number is expected to rise to around 439 million (Shaw *et al.* 2010). Diabetes comes in various forms, but type 1 diabetes is one that develops when the body does not create enough insulin. It is sometimes referred to as an insulin-dependent wager. On the other hand, type 2 diabetes, which is non-insulin-dependent, affects the tissues that are sensitive to insulin. In type 2 diabetes, the reduction in the sensitivity to insulin causes the metabolism of various food components to be altered. This condition is referred to as insulin resistance. The majority of the body's cells are unable to use glucose effectively as a result of this kind of diabetes (Hall and Guyton 2006) . The effects of diabetes mellitus can be severe. These include various macro-vascular diseases such as kidney disease and heart disease, as well as micro-vascular disorders such as neuropathy and nephropathy. In addition, these conditions can lead to high mortality rates and reduced life expectancy. (Bhattacharjee *et al*. 2018) The morbidity and mortality of diabetes are caused by both its chronic and acute consequences. These include diabetic ketoacidosis, hypoglycemia, and hyperosmolar hyperglycemic non-ketotic coma. In addition, chronic hyperglycemia is a central factor in the pathophysiology of multiple chronic conditions such as kidney disease, retinopathy, and neuropathy.(Loewen and Haas 1991) the complications of diabetes, such as microvascular damage, are known to induce renal damage, which is the most common type of kidney disease that occurs in people with type 2 diabetes. (Long and Dagogo-Jack 2011) It is a leading cause of death and morbidity in individuals with this condition. (Maggiore *et al*. 2017). About 40% of patients with diabetes develop diabetic nephropathy after 10 years over-diagnosed

(Alicic *et al*. 2017). If your albuminuria excretion rate is above 300 mg/d or 200 g/min more than twice in a three-to-six-month period, you may have a form of kidney disease known as chronic albuminuria. Increased blood pressure is a common cause of this illness. It can lead to the end stage of the disease. It is important to diagnose patients with diabetes who are at risk of developing kidney disease (DN) to improve their control of the disease. Various factors can affect the development and outcome of this condition. One of these is albuminuria. It can be used as a screening tool to identify individuals with kidney disease. However, it does not always pose possible to detect the early stages. (Thipsawat 2021) Although albuminuria can be used as a screening tool, it cannot predict the outcome of kidney disease. It has various limitations that prevent it from being able to identify paper patients' kidney multiple disease biomarkers cans that can be used to detect kidney damage. a biomarker is a characteristic factor that can be used to identify or measure the normal functioning of a physiological or pathological process. Some examples of these are proteins, lipids, and genes. In addition, certain types of electrical signals can also be used to identify or measure the presence of these markers. (Bonventre 2014) Early detection and treatment of diabetes can help lower the incidence of diabetes and improve the life expectancy of people with the condition. In addition, it can help prevent the development of kidney disease(Thornton Snider *et al*. 2019). Several biological markers have been identified that can be used to predict the development and severity of diabetes. This review aims to identify the most sensitive and specific biomarkers that can be used to predict the development and severity of diabetes. These are those that are earlier in their development and can be used to detect the condition at an earlier stage there are a lot of studies showing early detection of diabetic nephropathy by many biomarkers. Some of these studies by Amer et al.2018(Amer and Haridas 2018) where (Cystatin C) is used to detect diabetic nephropathy, (Ye et al. 2021) used Homocysteine to detect DN, (El-Dawla *et al*. 2019) used E-cadherin, (Motawi *et al*. 2018) used NGAL.

1.1 Aim of Study

Among those who have type 2 diabetes mellitus, blood FGF-23 may be used as an early predictor of diabetic nephropathy, thus researchers are looking into this possibility.

2. LITERATURE REVIEW

2.1 Diabetes Mellitus (DM)

Diabetic mellitus is a metabolic condition that hinders your body's natural capacity to make good utilization of insulin. Environmental factors and genetic mutations are two potential causes (Alegre-Díaz *et al.* 2016). The rise in the number of diabetics as a result of fast urbanization and rising population growth has become a major global health concern. Studies predict that the percentage of people with diabetes will rise from from 4% in 1995 to about 5.4% in 2025 (King *et al.* 1998). The annual cost of treating these individuals is estimated to be around 850 billion US dollars (Cho *et al.* 2018). In recent years, it has become one of the most common non-infectious diseases(Wang *et al.* 2005). Despite the progress that has been made in the treatment of this condition, the mortality rates for these individuals continue to increase (Sommese *et al.* 2017) According to studies, half of all diabetics worldwide are unaware that they have the disease, and as complications rise, new research problems and opportunities arise (Rufo *et al.* 2021).

2.2 Types of Diabetes Mellitus

Type 1: Type 1 diabetes is thought to be caused by B cell degeneration in the pancreatic islets. This condition is caused by the actions of the immune system, which mistakenly destroys healthy B cells. About 95% of cases of diabetes are caused by autoimmunity. Less than 5% of cases of pancreatic cancer are affected by this condition. The rate at which pancreatic cells are destroyed varies depending on the individual. Although it can occur at any age, it usually affects kids with the peak of infection during their preschool years (Jwad and AL-Fatlawi 2022).

Type 2: Type 1 diabetes (T1D) and type 2 diabetes (T2D) are difficult to identify from one another due to the intricacy of the disease. Both of these have the same clinical characteristics, but they share the same pathogenesis. It is believed that the first type of

diabetes will eventually develop insulin resistance. Diabetes that is not insulin-dependent is the term used to describe this condition. About 90% of those who have diabetes are affected. It is caused by the development of resistance to insulin. It can appear at a younger age due to the presence of genes that play a role in the development of diabetes II.

Gestational diabetes mellitus (GDM): Gestational diabetes is a type of diabetes that occurs when a woman's blood sugar levels are high during or after pregnancy. There are two types of this condition: A1GDM and A2GDM.

A1GDM is a diet-controlled condition that can be managed through diet and nutritional therapy, while A2GDM is a type of diabetes controlled through drugs. Beta cell abnormalities in the pancreas are thought to be a potential cause of gestational diabetes. It is possible for the illness to develop as a result of the beta cells' delayed reaction to blood sugar levels. One of the most common factors that can lead to the development of gestational diabetes is the lack of the proper secretion of the hormone lactogen Human. This hormone can stimulate the development of this condition and increase blood sugar levels during pregnancy. Other hormones that can stimulate the development of this condition include growth hormone, prolactin, and progesterone. (Jwad and AL-Fatlawi 2022).

Neonatal diabetes mellitus: NDM is a rare genetic disorder that affects about 1 in 90,000 individuals in Europe and about 1 in 21,000 individuals in the Middle East. It is characterized by the onset of diabetes within six mon months topes of diabetes can that can be inherited: permanent and transient(Habeb *et al*. 2012). In neonates, intensive treatment is required to maintain a healthy balance between their glucose levels and their neurological development. This condition can be hypoglycemia, which can affect their cognitive and physical geneticnetic ananAnandsis can also help in the diagnosis of diabetes. A comprehensive genetic review has revealed that about 80% of the genetic mutations in children with type 1 diabetes (NDM) are found to be harmless(De Franco *et al*. 2015). This means that infants with these conditions can be treated with oral

glibenclamide instead of insulin injections. Besides this, genetic testing can also help identify the possible need for a stem cell transplant in these patients.

2.3 The MODY Syndromes (Monogenic Diabetes Mellitus In Youth)

MODY is a group of diabetes disorders that are caused by pancreatic dysfunction onset of diabetes, which does not have features of autoimmunity or insulin resistance, this is one of the main reasons why MODY is considered a subtype of diabetes (Rubio-Cabezas *et al*. 2014) The first cases of MODY were identified in the 1970s. They are present in every ethnicity and race. It is estimated that the population of this disorder has a prevalence of around 1.1 to 1,000.(Froguel *et al*. 1992) Patients with rare mutations in the b-cell dysfunction that result in the deletion of certain genes are considered o have MODY variants. These include the genes HNF1B, MODY4, and MODY5.

MODY5 is a subtype of diabetes that affects 5% of all patients. It is also known as "renal cysts and diabetes syndrome". This condition typically includes other urological disorders and pancreatic atrophy. (Bishay and Greenfield 2017).

Latent autoimmune diabetes in adults (LADA): Islet autoantibody is the defining feature of the subtype of diabetes known as LADA, which has many traits with type 1 diabetes. Additionally, it is well documented to exhibit varying degrees of insulin resistance and cell dysfunction. As the most prevalent form of autoimmune diabetes, it. Age at diagnosis, the presence of islet autoantibodies, and insulin independence are the three key features of this illness.

Type 3c diabetes: Type 3c diabetes, sometimes referred to as pancreatogenic diabetes, is a kind of diabetes that can be brought on by pancreatic illness. (Conlon and Duggan 2017) It can be caused by the presence of pancreatic cancer. Aside from pancreatic cancer, chronic pancreatitis can also be caused by other conditions such as cystic fibrosis, hemochromatosis, and fibrocalculous pancreatopathy(Cui and Andersen 2011).

Although the most common underlying cause of this condition is chronic pancreatitis, other conditions such as pancreatic cancer can additionally cause this condition. About 77.5% of type 3 diabetes mellitus (T3cDM) patients have chronic pancreatitis. Pancreatic cancer, on the other hand, is regarded as the second most common cause of pancreatic ductal adenocarcinoma (PDAC). A recent study revealed that about 85% of type 3c diabetes cases were caused by benign conditions. These conditions could be expected to cause similar results in other studies (Price *et al*. 2010).

2.4 Mechanisms of Action and Resistance of Insulin

Insulin controls blood glucose levels as one of its functions. It is a peptide hormone that has a role in preserving a balanced and healthy metabolic system. It is also known to have an impact on the growth and upkeep of numerous tissues, including the liver, skeletal muscle, and adipocytes. In addition, it can additionally affect the development and maintenance of multiple tissues, such as the liver, skeletal muscle, and adipocytes. In the liver, insulin activates the process of glycogen synthesis and increases the expression of a protein known as lipogenic genes. It can also prevent the decomposition of fat tissue and increase lipid formation. In addition to being directly related to insulin, it has indirect effects on the target tissues. Inhibition of the decomposition of fat tissue by insulin leads to a decrease in the activity of the acetyl-CoA in the liver. The reduction in the lipolysis and formation of hepatic sugar by glycone-genesis is also beneficial for the development of obesity. It increases the tissue resistance to insulin functions and lowers the blood sugar level through the reduction in the sensitivity of insulin in various organs of metabolism. The action of insulin on the skeletal muscle cells prevents glucose from being absorbed by these tissues. This leads to the transfer of glucose to the liver. As a result, the formation of fat cells is not affected, which increases insulin resistance.

2.5 Etiology of Diabetes Mellitus

Multiple etiologies have been identified for diabetes mellitus. Environment and genetic factors play a role in the development of this condition. Some of the factors that can

affect the development of this condition include medication use, physical inactivity, viral infections, and obesity (Adeghate *et al*. 2006).

2.6 Symptoms of Diabetes Mellitus

Some of the symptoms of diabetes that are commonly seen are blurred vision, polyphagia, thirst, and weight reduction. Depression is also a sign of diabetes. Other symptoms that can be triggered by the condition include frequent abdominal pain, constipation, and fecal incontinence. If the hyperglycemic state is not controlled, these symptoms can additionally raise (Eker 2018).

Although these symptoms can be triggered by diabetes, they are usually not considered to be serious. For instance, children with diabetes are more prone to experiencing these symptoms. In addition, high blood glucose levels and ketonuria are also common (Yamamoto-Honda and Akanuma 2002)

2.7 Risk Factors

There are two types of risk factors for diabetes: irreversible risk factors and modifiable risk factors. These include genetics, race, sexual orientation, and age. Aside from being overweight, physical inactivity, and dietary intake, these factors can also affect the quality of fats and carbohydrates.

2.8 Complications of Diabetes

The various factors that can affect the development and maintenance of diabetes are known to be categorized into their complications. These include age, gender, education, income, and family history. In addition to these, other factors such as blood pressure and cholesterol levels can also affect the development and maintenance of diabetes. Unclassified complications such as skin disorders, gum disease, and bone disease are also known to be caused by diabetes. One of the most common types of diabetes is

peripheral neuropathy. This condition affects the nerves in the peripheral area and can lead to different problems such as diabetes and sexual dysfunction. Other common types of diabetes are known to be caused by the presence of diseases in the small vessels of the blood. These include a type of diabetes known as diabetic nephropathy.

2.9 Current Treatments for Diabetes Mellitus

Diabetes is treated with two primary groups of medications. Insulin is one of these, and oral medications are another. These two medicines are used to control the symptoms of diabetes and lower blood glucose levels. However, different people have different responses to different types of medicines. These drugs include various types of insulins, such as insulin secretagogues, thiazolidinediones, and meglitinide derivatives. They are also used to treat children with diabetes. (Modi 2007)

2.10 Global Burden of Diabetes Mellitus

The number of people with diabetes and impaired glucose tolerance has increased significantly over the past couple of decades. Through the years, the IDF has been monitoring the incidence of diabetes in different regions and countries. In 2017, almost half of all adults worldwide were diagnosed with diabetes. By 2045, more than 690 million people are expected to have diabetes. About half of all people with diabetes were not diagnosed. Also, over 300 million people have impaired glucose tolerance. During pregnancy, almost 20 million women were affected by hyperglycemia. Five million people died due to diabetes between the ages of 20 and 99 (Biswas 2022).

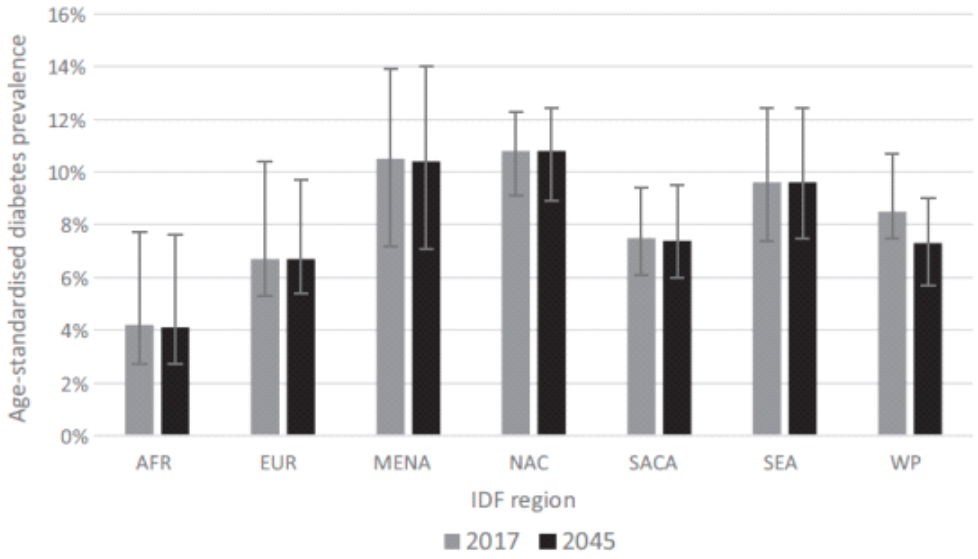

Figure 2.1 Diabetes Prevalence by IDF Region between 2017 and 2045, Age-standardized (Age Range:18-99) (Cho *et al*. 2018)

2.11 Nephropathy

Nephropathy is one of the main causes of end-stage renal disease and is associated with a high mortality rate and poor quality of life. Early detection and intervention are important to prevent this condition from developing. Although it is possible to identify individuals at risk for developing this condition, it is also important to treat them at an early stage(Adler *et al*. 2003) through the use of validated prediction models, it is possible to identify individuals at risk for developing nephropathy and make informed decisions regarding their treatment. These models should be based on the most cost-effective and routine clinical markers.

2.12 Diabetic Nephropathy

Diabetes nephropathy is one of the most common types of complications of this disease. It can be caused by factors such as fibrosis, microalbuminuria, and mesangial matrix expansion. In the initial stages, this condition can lead to renal failure and progressive kidney damage. The urinary albumin removal rate is an important indicator of the early

onset of the disease. It can also help identify individuals who are at risk of developing other health conditions (Cai *et al*. 2021).

2.13 Glomerular Filtration Rate (GFR)

The kidney is responsible for various functions, such as maintaining a healthy body and preventing diseases. Administering GFR is used to evaluate the kidney's functioning and manage chronic kidney disease. It can also determine drug dosages and predict the severity of the disease.

The rate at which the kidney produces an ultrafiltrate is known as GFR, which is the level of clearance that can be assessed from the presence of various levels of filtration markers. In most clinical practices, the clearance of serum levels of these markers is not performed routinely. However, GFR can be estimated using a combination of equations and serum concentrations. Although GFR estimates are generally accurate, they may not be appropriate for every patient. To improve the accuracy of the assessment, a confirmatory test is required (Inker and Titan 2021).

2.14 Microalbuminuria

In patients with diabetes, a microalbuminuria test is often performed to check for signs of kidney disease. Diabetic nephropathy is a type of condition that can involve various pathological processes, such as hyperfunction and kidney hypertrophy. If one has diabetes and other chronic conditions, then they are at risk of developing a chronic condition known as glomerulosclerosis. The process of this disease continues for several years, and it eventually ends with an increase in the membrane's permeability. This condition is referred to as microalbuminuria. The early detection of diabetic nephropathy is regarded as the gold standard. However, a study revealed that changes in the histopathologic structure could occur in patients with this condition without any other symptoms. Although albumin can be used as a potential early detection test for

diabetes, it is not enough to identify patients with this condition. To provide a Tote diagnosis, the albumin albuminuria must be nephropathy (Amelia *et al*. 2021).

2.15 Albumin Creatinine Ratio (ACR)

The albuminuria screening involves taking the urine sample for 24 hours. The ACR is the preferred method, although measurement is not always feasible. Since albumin concentration can vary, taking the sample continuously is important. The ACR is a test that can be used to determine albuminuria, especially in patients with diabetic nephropathy. Taking a urine sample is a standard method for diagnosing albuminuria (Amelia *et al*. 2021). It is different from 24-hour retrieval because it takes into account the excretion of protein. A urine sample can be used to detect protein excretion. (Kouri *et al*. 1991) .The initial signs of diabetes are usually a rise in albuminuria and a decrease in the eGFR, which is a measure of the filtration rate of blood. This condition can lead to end-stage renal failure. Some of the risk factors that can affect this condition include diabetes, high blood pressure, and obesity (McFarlane *et al*. 2018).

2.16 Serum Albumin Level and Renal Outcome

Human serum albumin is a plasma protein that is produced in the liver. It is a vital component of the maintenance of homeostasis, and it helps maintain a balance between the colloid and hydrostatic pressure within vessels. In addition to being a vital component of the maintenance of homeostasis, serum albumin also has various physiological functions. Some of these include its anti-inflammatory and antioxidant properties. It has been known that hypoalbuminemia is caused by a combination of factors, such as insufficient energy intake, poor liver synthesis, and increased tissue catabolism. This condition is also known to increase the risk of experiencing morbidity and mortality. The study revealed that the level of albumin, which is a type of blood substance, is associated with the prognosis of kidney disease. It also found that the presence of proteinuria and other kidney lesions was associated with the development of hypoalbuminemia. This implies that the absence of exercise and the existence of these disorders are the two risk factors that can raise the likelihood of getting this condition.

2.17 The Fibroblast Growth Factors (FGF)

The FGF family is composed of seven members that are known to promote the growth and regeneration of soft tissues. These members have a similar mitogenic spectrum and are angiogenic. They also promote the proliferation of various cell types in the mesodermal and neuroectodermal regions (Basilico and Moscatelli 1992).

2.18 The Fibroblast Growth Factor 23 (FGF-23)

One of the most important factors that regulate the metabolism of calcium-phosphate is the FGF-23. This factor, which is also known to enhance the excretion of kidney phosphate, is a cofactor of Klotho. This combination helps maintain normal levels of serum phosphate. In people with chronic kidney disease, the levels of FGF-23 increase as the disease progresses. However, these levels are not always observed to be associated with a significant increase in serum phosphate concentration. In cross-sectional studies, the effects of higher FGF-23 levels on the development of secondary hyperparathyroidism were also observed. These result suggests that the effects of this factor on the skeletal and cardiovascular systems are also independent of the conventional calcium-phosphate metabolism markers. According to research, people with chronic kidney disease (CKD) who were not receiving dialysis had faster disease progression. They had a higher mortality rate as well. These results imply that FGF-23 may be a useful therapeutic target for the management of this illness (Seiler *et al*. 2009).

Chronic kidney disease has been linked to high levels of the phosphate-regulated fibroblast growth factor 23 (FGF-23) (CKLD). However, it is unclear exactly how they affect different elements of health such as all-cause mortality, cardiovascular events, or the start of chronic dialysisThe lowest quartile of serum phosphorus levels was 4.3 mg/dl, while the highest was 392 RU/ml. In the 2.9 years of follow-up, 453 individuals died from any reason, 215 experienced a cardiovascular incident, and 613 (56%), among the greatest numbers of patients, began chronic dialysisThe following quartiles have a greater risk of death when compared to the lowest quartile. The top two quartiles of FGF-23 were shown to be substantially related with a greater risk of cardiovascular

events and advanced chronic kidney disease (CKD) in patients. This finding demonstrates an association between FGF-23 and cardiovascular events and all-cause mortality in this situation (Kendrick *et al*. 2011).

2.18.1 Structure of FGF-23

There are about 120 conserved amino-acid residues in the family of FGFs' shared core region. A -trefoil structure with several folding strands and loops distinguishes this location. (Goetz *et al*. 2007) The three main subfamilies of human FGFs are composed of the following proteins: FGF-19, FGF-21, and FGF-23The blood levels of phosphate and calcitriol are controlled in large part by the FGF-23. A member of the FGF family, FGF-19, prevents the production of an enzyme necessary for the synthesis of bile acids. (Crestani *et al*. 1998) The three FGF subfamilies, namely FGF-19, FGF-21, and FGF-23, have a disulfide bond that is not found in other subfamilies. This allows them to maintain their unique foil-the structure. This peculiar bond may also help explain the unusual structure of the core region of the FGF family. A change in the structure of the FGF-19 and FGF-23 subfamilies may explain why these two molecules have a low affinity for the blood-borne protein heparin(Goetz *et al*. 2007). Members of these subfamilies also bind to the cell surface using a combination of factors, which results in their capture and explanation of their paracrine function. (Yamashita 2005) The 251-amino-acid protein known as FGF-23 is mainly produced by bone cells. It is a member of the FGF family and has a unique carboxyl- and amino-terminal sequence. It can also be cleaved using a proprotein convertase. The half-life of intact FGF-23 is approximately 58 minutes (Khosravi *et al*. 2007).

One of the two methods used to measure human FGF-23 is the sandwich immunosorbent assay. This method combines two types of antibodies to find the presence of the N- and C-terminal sections of the protein. The conventional method only recognizes the full-length fragments (Yamashita 2005).

2.18.2 Physiological functions of FGF-23

The presence of Na/Pi Type II cotransporters in the apical membrane of proximal tubular cells controls phosphate reabsorption in the kidney primarily. The release of phosphate ions is regulated by this area. About one-third of the reabsorbed phosphate is carried via the Na/Pi IIa cotransporter, and the FGF-23 inhibitor lowers the level of this cotransporter in the proximal tubular cells. (Baum *et al*. 2005)

2.19 FGF-23 and CKD

Renal function is known to diminish in people with chronic kidney disease (CKD) due to rising levels of FGF-23(Gutierrez *et al*. 2005). However, among those with advanced kidney disease, this is not the case. In a MrOS investigation,(Marsell *et al*. 2008) The estimated glomerular filtration rates and the amount of FGF-23 were found to be significantly correlated. When the serum phosphate levels are within the normal range, the increase in FGF-23 levels is shown in individuals with advanced kidney disease in cross-sectional investigations (Larsson *et al*. 2003). However, when the levels of FGF-23 are higher in these patients, the estimated filtration rate is impaired (Larsson *et al*. 2003).

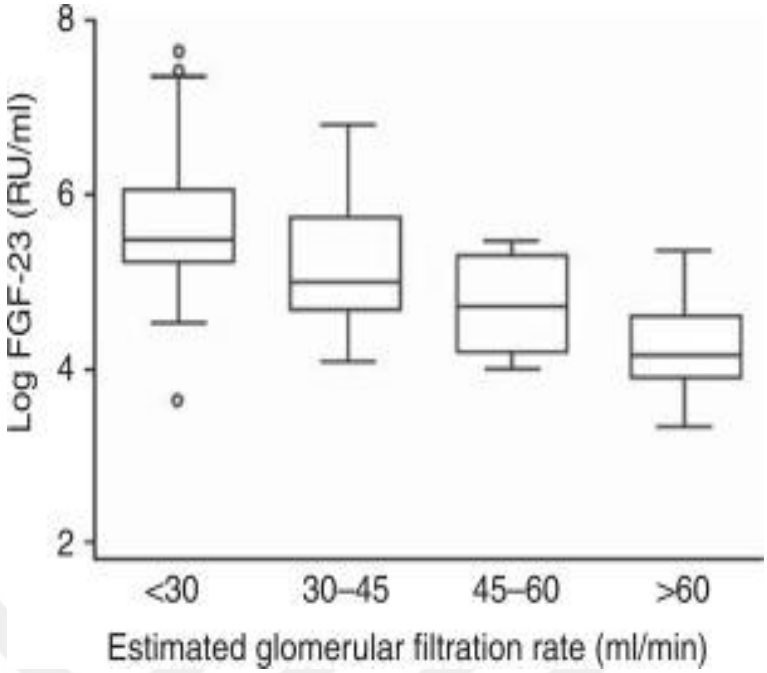

Figure 2.2 Rising levels of fibroblast growth factor 23 (FGF-23) are associated with the decline in kidney function in patients with chronic kidney disease

2.19.1 Increased FGF-23 secretion in CKD

FGF-23 levels were originally thought to be elevated in chronic kidney disease (CKD) because of impaired renal clearance. This is not, however, the situation. The research instead contends that the rise in FGF-23 secretion is what is responsible for the elevated levels of this protein. Furthermore, it implies that the end-organ resistance to the effects of phosphaturic stimuli may be caused by a deficiency of the essential Klotho co-factor.In a study conducted on the effects of Klotho on the expression of mRNA in kidney biopsy samples, the researchers noted that the reduction in the Klotho expression significantly decreased the expression of this protein in the kidney(Koh *et al*. 2001). As the number of intact nephrons decreases, higher FGF-23 levels may cause serum phosphate levels to stabilize. The breakdown of phosphate may also be hampered by this situation. FGF-23 can also impair the body's capacity to keep phosphate levels within a physiological range by inhibiting 1-hydroxylase and lowering circulating phosphate levels. There is a study by SEEK4 revealed that normal phosphate levels were observed in most patients with chronic kidney disease (CKD) stages 1–4.

However, this increase in phosphate levels did not occur until stage 5 when the phosphaturic capacity was exhausted. (Levin *et al*. 2007)

2.19.2 FGF-23 as a prognostic factor in CKD

Following the discovery that FGF-23 was linked to worse outcomes in individuals with chronic renal disease in two prospective cohort studies, interest in this protein rose (CKD). Additionally, the study discovered that patients with chronic renal disease who had high levels of FGF-23 had a noticeably increased risk of dying. The researchers came to the conclusion that those with the highest levels of FGF-23 were six times more likely to die after correcting for numerous confounders, such as serum phosphate levels (Gutiérrez *et al*. 2008). At the 2008 American Society of Nephrology meeting, the first clinical evidence indicating high levels of FGF-23 are related to end-organ damage in chronic kidney disease (CKD) was presented. In this study, a multivariate analysis showed a correlation between high FGF-23 levels and left ventricular hypertrophy.

3. MATERIALS AND METHODS

3.1 Study Design

We enrolled 145 participants in this study, who were chosen among those working at 4 important clinical laboratories. In 2022, the study was conducted in the labs of the Al-Yarmouk teaching hospital and the Medical City Complex Teaching Laboratories.

3.2 Patient Group

Patients comprised 95 people with type 2 diabetes mellitus:

(51 patients) with microalbuminuria.

(44 patients) without microalbuminuria.

Patients' ages ranged from 45 to 75, on average.

3.2.1 Inclusion criteria

Patients aged 18 years and older with microalbuminuria or longer-standing type 2 diabetes (at least 5 years).

3.2.2 Exclusion criteria

- Diabetic patients have a different subtype of the disease.
- Macroalbuminuric patients.
- Patients suffering from any other chronic condition that may compromise their kidneys.
- Kidney patients, whether they have acute or chronic disease.

3.2.3 Control group

Fifty people (both sexes) were randomly picked from the general public to serve as a control group. The median age of a healthy population was 60 years old (range: 40-69).

3.3 Materials

3.3.1 Kits and chemicals

Table 3.1 displays the research kits utilized here.

Table 3.1 Kits and manufacturer

No	Kits and Chemicals	Manufacturer	Lot number
1	Glucose	ROCH c311 (Germany)	00574944
2	UREA	ROCH c311(Germany)	00577330
3	CREATININE	ROCH c311 (Germany)	00556887
4	ALT	ROCH c311 (Germany)	00567727
5	AST	ROCH c311 (Germany)	00567186
6	FGF-23 (Elisa kit china)	Sandwich, Human	0263043
7	HBA1C	ROCH c111(Germany)	00629598
8	CBC	Beckman coulter (USA)	

3.3.2 Equipment

Table 3.2 lists the instruments and gadgets that were used over the course of this investigation.

Table 3.2 Equipment

No	Equipment	Manufacturer
1	Incubator(37c)	Memmert (Japan)
2	Fully automated analyzer	ROCH C311 ROCH C111 (Germany) ROCH C601
3	CBC analyzer	horiba (japan)
4	Automated urine chemistry analyzer	AVE-752(CHINA)
5	Mechanical pipette single-channel and disposable tips	DRAGON LAB (CHINA)
6	ELISA	HUMAREADER HS (Germany)
7	Centrifuge	Fastgene (japan)

3.4 Methods

Several patients' venous blood samples totaling five milliliters were taken. They were divided into two tubes, one containing an EDTA tube and the other a gel tube. Ten minutes later, the serum was centrifuged and aliquoted. Then, to prevent any errors caused by varying analysis conditions, the aliquots were maintained at -20 C. A cup of each person's urine was then collected in order to calculate the albumin-creatinine ratio.

3.4.1 Determination of FGF-23

Serum FGF-23 will be measured in all participating patients together with

other routine investigations including:

- CBC
- Urea
- Creatinen
- Alt
- Ast
- Random blood suger
- Hba1c
- Albumin-creatinine ratio (ACR)

Test principle: To measure the immune system's reaction to this protein, the ELISA kit combines the dual antibody sandwich method with human fibroblast growth factor-23. A pre-coated antibody that binds to the FGF- 23 protein was used to construct the wells. The anti-FGF-23 antibodies were combined with streptavidin-HRP to create an immunological complex that binds to the FGF-23 protein. After incubation and cleaning, the solution took on an acidic hue, turning yellow and blue. Human fibroblast growth factor-23 concentration and solution color are linked.

Types of samples: Different types of samples were collected from the participants in this study and these samples have been collected very accurately each in their appropriate conditions.

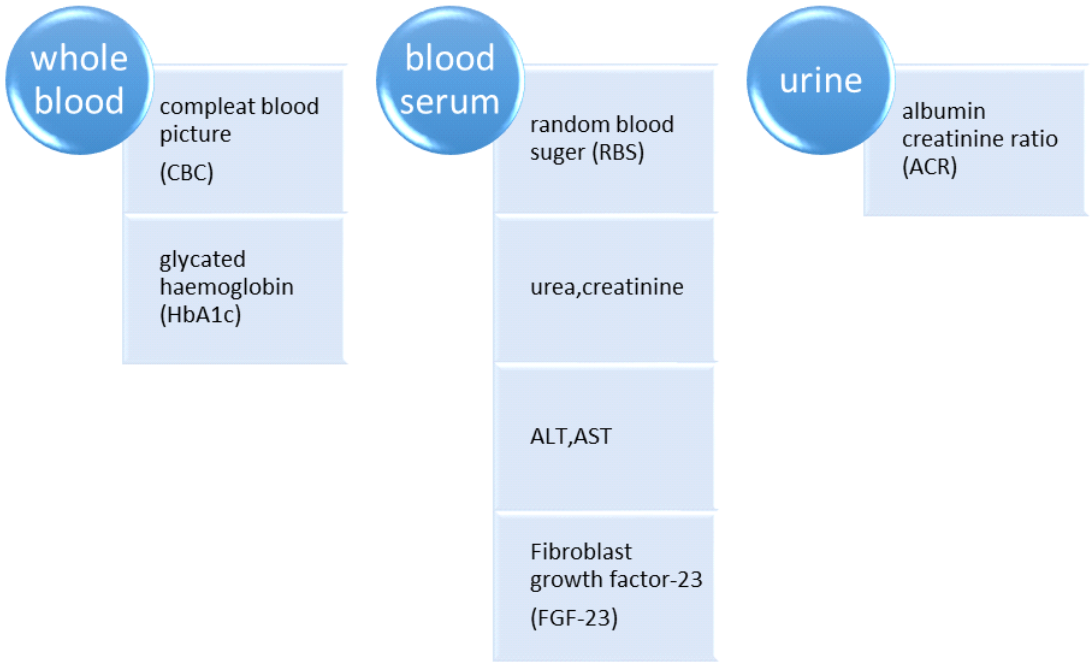

Figure 3.1 The types of samples and the tests

3.5 Statistical Analysis

Statistics are often summarized using the mean, median, mode, and standard deviation. To compare the groups, we used the ANOVA technique. The Mann-Whitney test and

the Bonferroni correction were used to analyze the data. The P0.05, P0.015, and P0.001 levels of statistical significance were discovered during data analysis.

4. RESULTS AND DISCUSSION

4.1 Gender

Table 4.1 and Figure 4.1 show the breakdown of the 145 patients who participated in the present research into the three groups: Control (50 participants), Diabetic without Microalbuminuria (44 patients), and Diabetic with Microalbuminuria (51 patients).

Table 4.1 Different groups are separated into subsets based on their gender

control and diabetic group	Sex		Total
	female	male	
Control group	23	27	50
Diabetes patients without microalbuminuria	20	24	44
Diabetes patients with microalbuminuria	11	40	51
Total	54	91	145

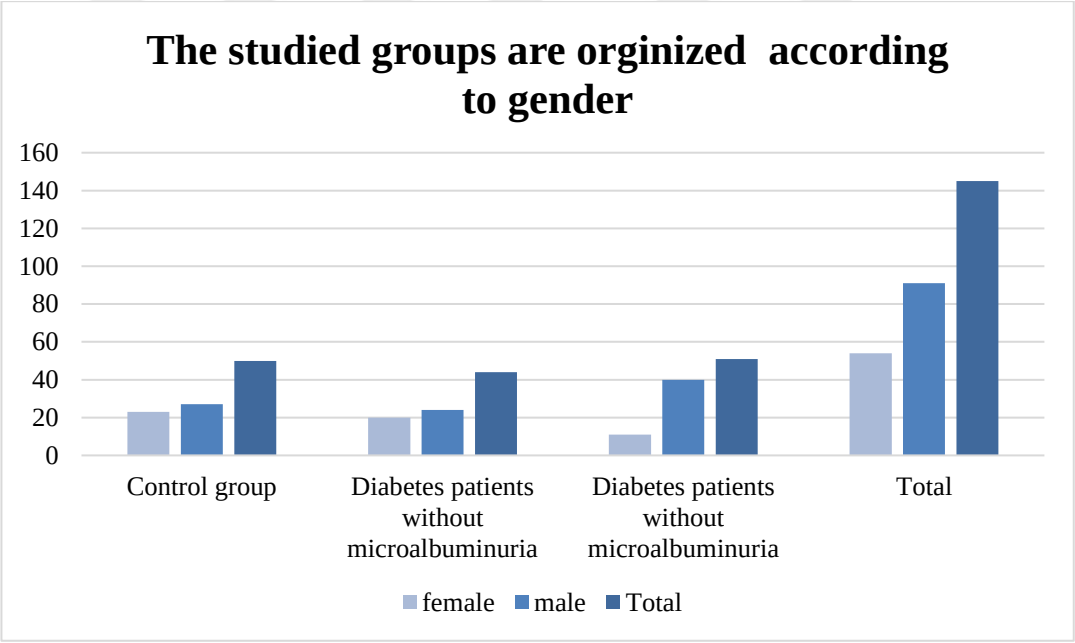

Figure 4.1 The groups under study are broken down by gender

4.2 Age

Table 4.2 and Figure 4.2 demonstrate that the average age of the participants was (36.22±14.006) in the control group, (48.01±12.297) in the Diabetic group without microalbuminuria, and (49.01±13.701) in the Diabetic group with microalbuminuria.

Table 4.2 Participants' average age

Parameter	Control group Mean±SD	Diabetes patients without microalbuminuria	Diabetes patients with microalbuminuria
Age	36.22±14.006	48.01±12.297	49.01±13.701
	Std.error (1.810)	Std.error (1.788)	Std.error (1.671)

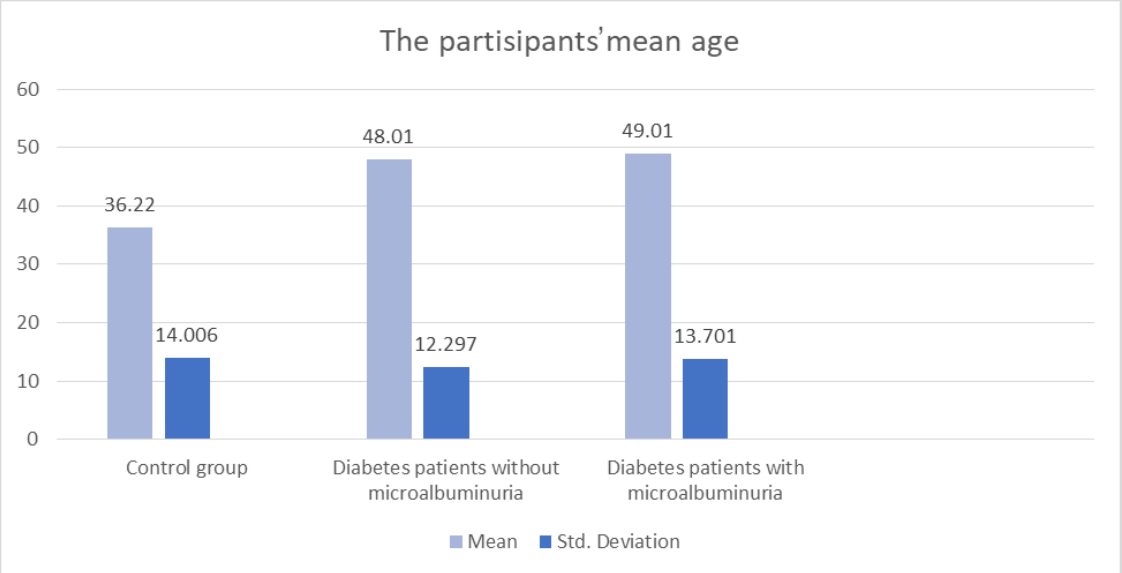

Figure 4.2 Average age of study subjects

4.3 Tests

Participants were divided into four groups based on their performance on the following tests:

When compared to the test group, the average glucose level in the control group was (102.564±17.6236), the diabetic group without microalbuminuria was (185.308±53.564) and the diabetic group with microalbuminuria was (279.826±90.1108).

HbA1c ranged from (6.0841±0.81491) in the control group to (9.0524±1.00974) in the diabetics without microalbuminuria group and (11.657±3.20034) in the diabetics with microalbuminuria group. as shown in table microalbuminuria was (10.6716 ±2.10041), as shown in Table 4.3, Figure 4.3 and Figure 4.4.

Table 4.3 Glucose and HbA1c findings

Parameter	Control group Mean±SD	Diabetes patients without microalbuminuria	Diabetes patients with microalbuminuria	p-value
Glucose	102.564±17.6236	185.308±53.564	279.826±90.1108	Less than 0.05
	Std.error (2.5862)	Std.error (8.1274)	Std.error (11.974)	
Hba1c	6.0841±0.81491	9.0524±1.00974	11.657±3.20034	Less than 0.05
	Std.error (0.12083)	Std.error (0.14285)	Std.error (0.38321)	

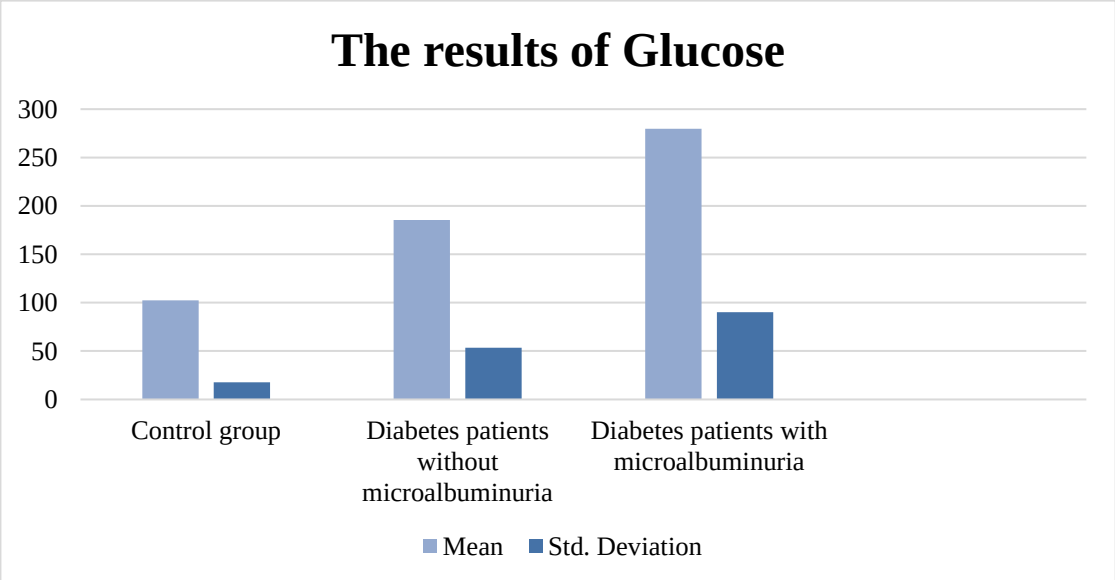

Figure 4.3 Impact of glucose

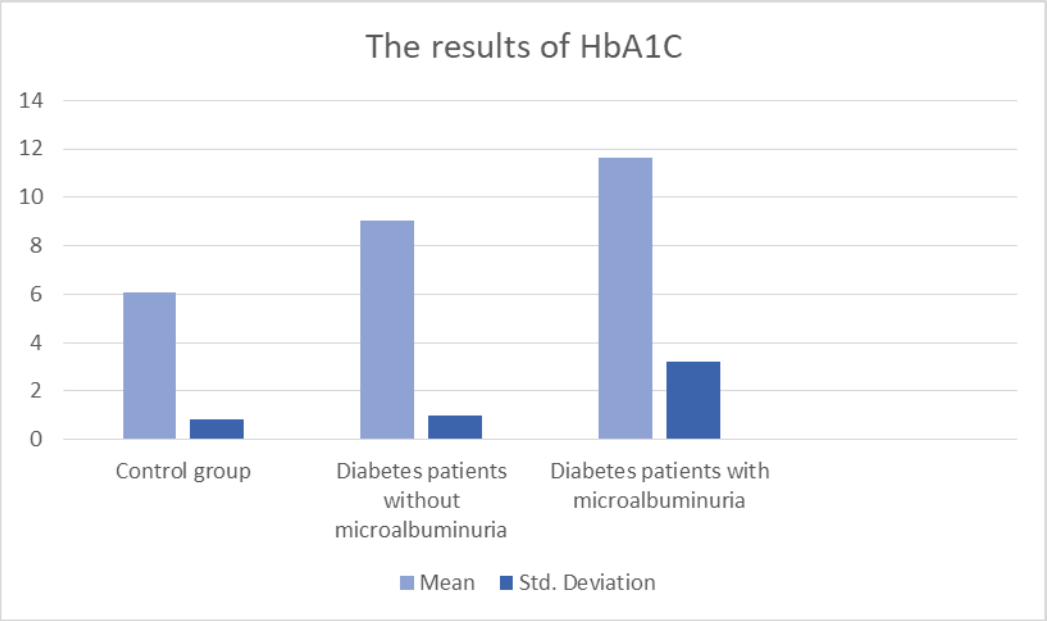

Figure 4.4 Evidence from the HbA1c

The mean of Urea in the control group was (31.2510±6.10582), The diabetic group without microalbuminuria was (39.2101±8.21803) and the diabetic group with microalbuminuria was (41.2717±89.17321). Figure 4.5.

Table 4.4 and Figure 4.6 reveal that the average levels of creatinine in the three groups were as follows: (0.7214±0.31150) in the control group; (1.234±1.32511) in the diabetic group without microalbuminuria; and (1.254±2.57174) in the diabetic group with microalbuminuria.

Table 4.4 Tests for uric acid and creatinine

Parameter	Control group Mean±SD	Diabetes patients without microalbuminuria	Diabetes patients with microalbuminuria	p-value
Urea	31.2510±6.10582	39.2101±8.21803	41.2717±89.17321	more than 0.05
	Std.error (0.83537)	Std.error (1.16369)	Std.error (12.40578)	
Creatinine	0.7214±0.31150	1.2343±1.32511	1.2547±2.57174	more than 0.05
	Std.error (0.04749)	Std.error (0.29452)	Std.error (0.32404)	

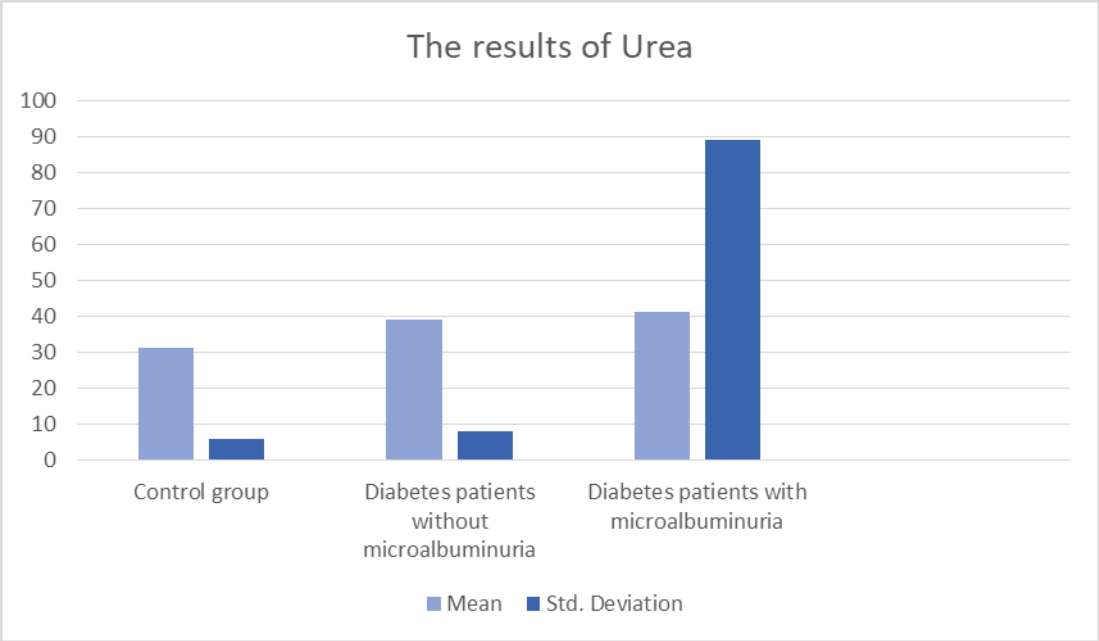

Figure 4.5 Ulcerogenic activity of urea

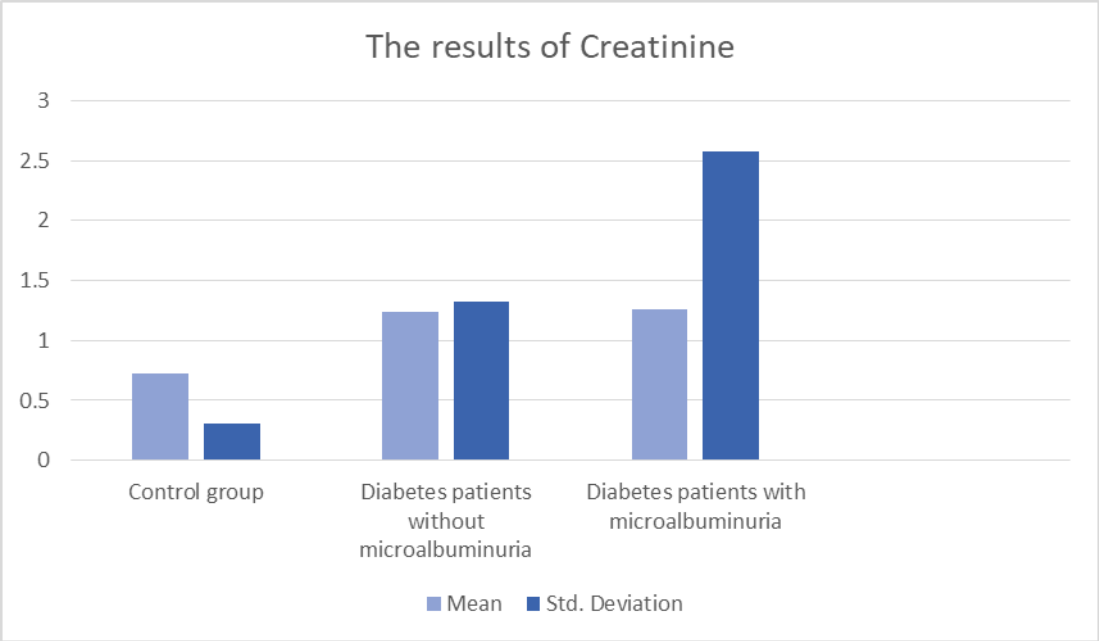

Figure 4.6 When analyzing creatinine levels

The average levels of ALT in the control group were (30.260±6.1073), those in the diabetic group without microalbuminuria were (30.160±7.5315), and those with microalbuminuria were (36.427±1.125). Alanine aminotransferase (AST) ranged from (35.0710±6.37230) in the control group, (36.0566±7.0332) in the diabetics without

microalbuminuria, and (44.1034±14.52288) in the diabetics with microalbuminuria, as shown in Table 4.5, Figure 4.7 and Figure 4.8.

Table 4.5 The results of ALT and AST

Parameter	Control group Mean±SD	Diabetes patients without microalbuminuria	Diabetes patients with microalbuminuria	p-value
ALT	30.260±6.1073	30.160±7.5315	36.427±11.125	more than 0.05
	Std.error (0.7861)	Std.error (1.0316)	Std.error (1.4404)	
AST	35.0710±6.37230	36.0566±7.03321	44.1034±14.52288	more than 0.05
	Std.error (0.82241)	Std.error (0.87219)	Std.error (2.21217)	

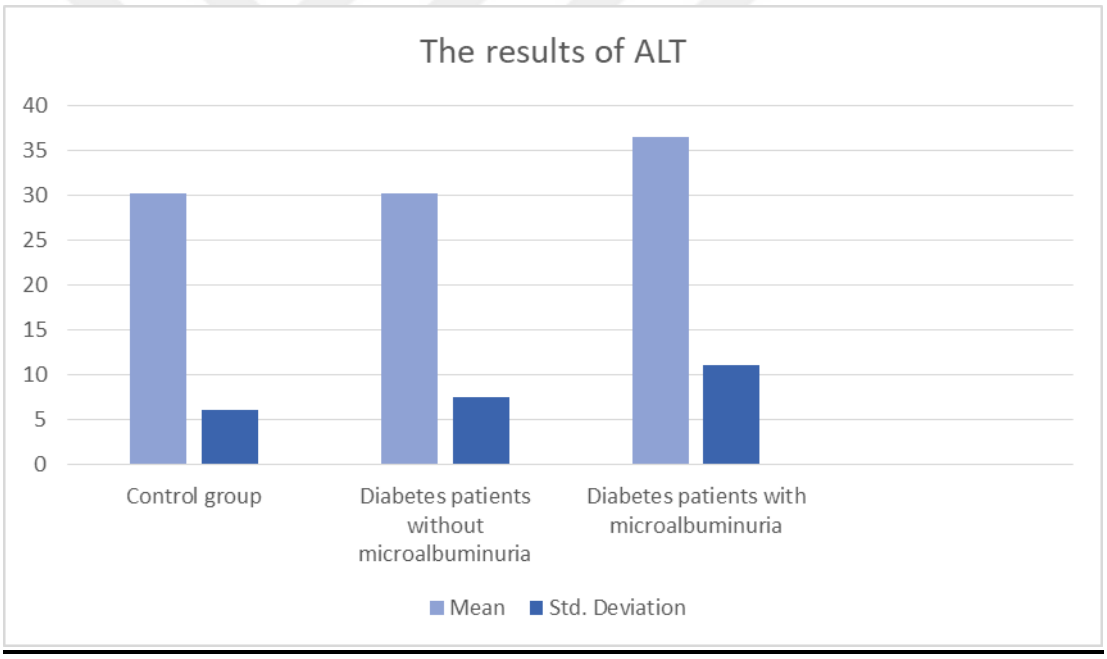

Figure 4.7 Analysis of ALT outcomes

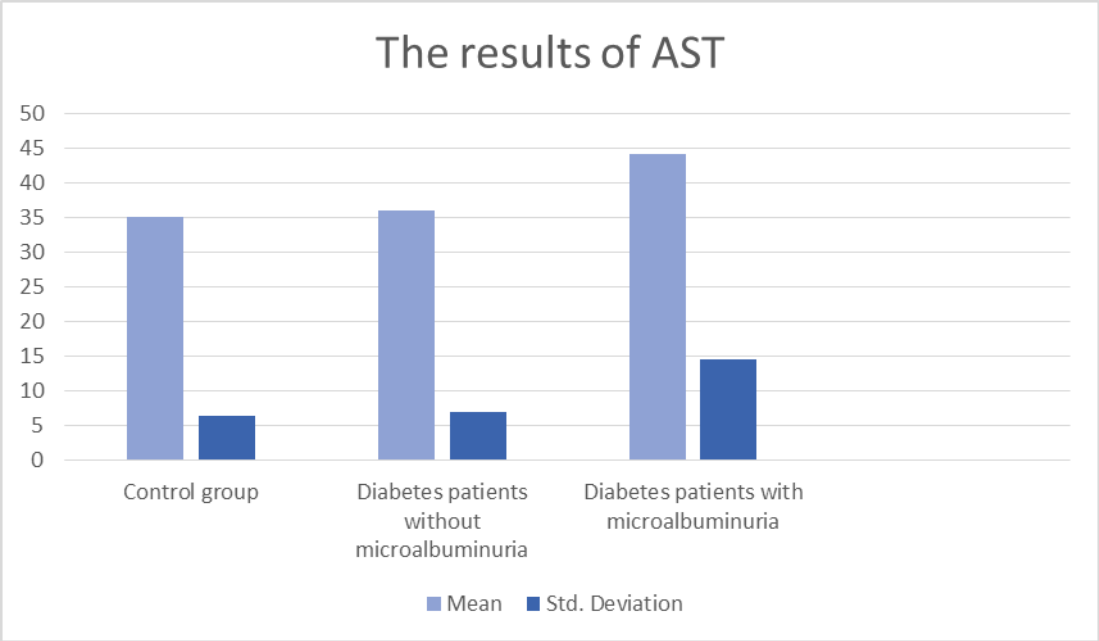

Figure 4.8 Analysis of AST outcomes

Hemoglobin ranged from (14.5630±1.68282) in the control group to (13.9375±1.84353) in the diabetics without microalbuminuria and (14.1662±1.9988) in the diabetics with microalbuminuria.

The average white blood cell count was (4.9154±1.23874) in the healthy controls, (5.9575±1.47616) in the diabetics without microalbuminuria, and (5.9847±2.91094) in the diabetics with microalbuminuria.

As can be seen in Table 4.6, Figure 4.9, Figure 4.10, and Figure 4.11, the average number of platelets in the control group was (206.534±41.1383), in the group without diabetes (213.755±52.6125), and in the group with diabetes (186.564±61.9586).

Table 4.6 The results of Hemoglobin, WBC count, and platelets

Parameter	Control group Mean±SD	Diabetes patients without microalbuminuria	Diabetes patients with microalbuminuria	p-value
Hemoglobin	14.5630±1.68282	13.9375±1.84353	14.1662±1.9988	more than 0.05
	Std.error (0.32511)	Std.error (0.38420)	Std.error (0.36450)	
Wbc count	4.9154±1.23874	5.9575±1.47616	5.9847±2.91094	more than 0.05
	Std.error (0.24564)	Std.error (0.30910)	Std.error (0.52022)	
Platelet	206.534±41.1383	213.755± 52.6125	186.564±61.9586	more than 0.05
	Std.error (6.8764)	Std.error (8.6960)	Std.error (9.6359)	

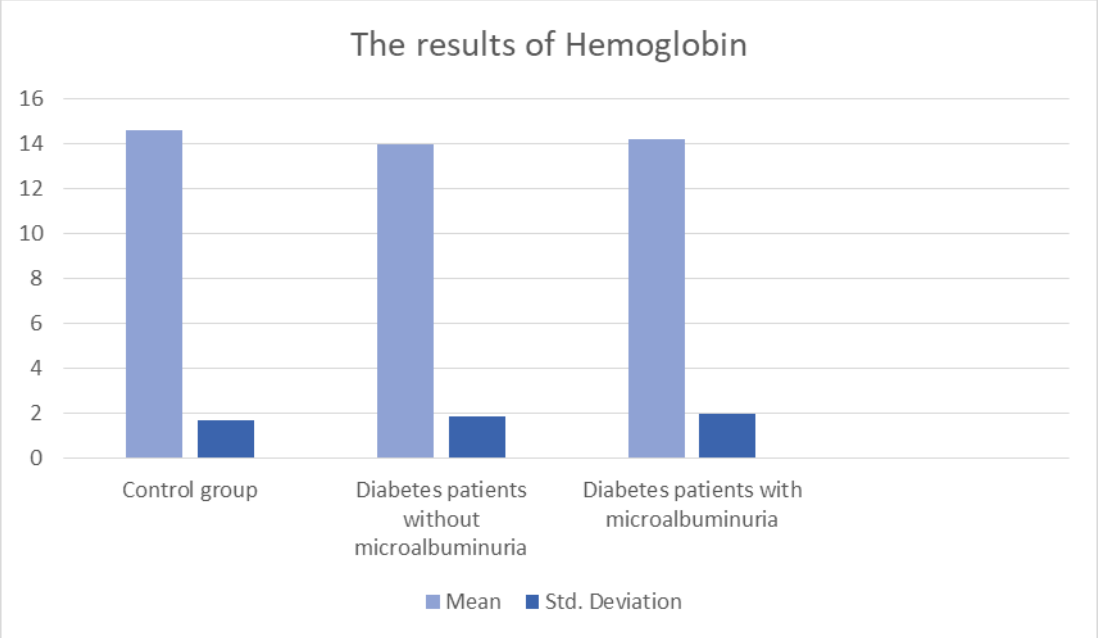

Figure 4.9 Hemoglobin results

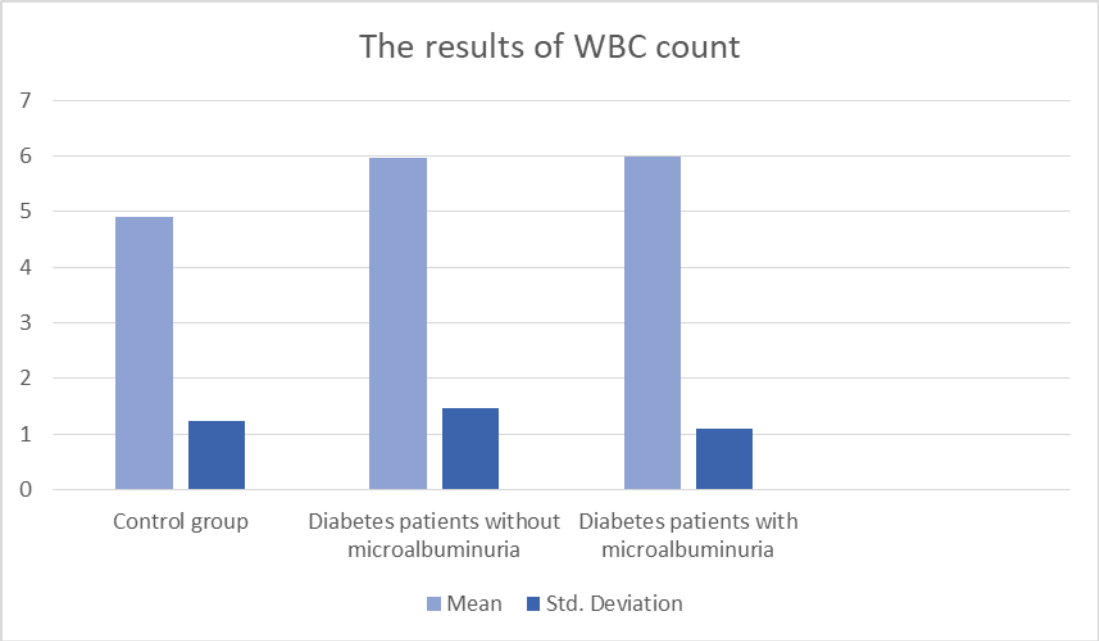

Figure 4.10 The WBC count results

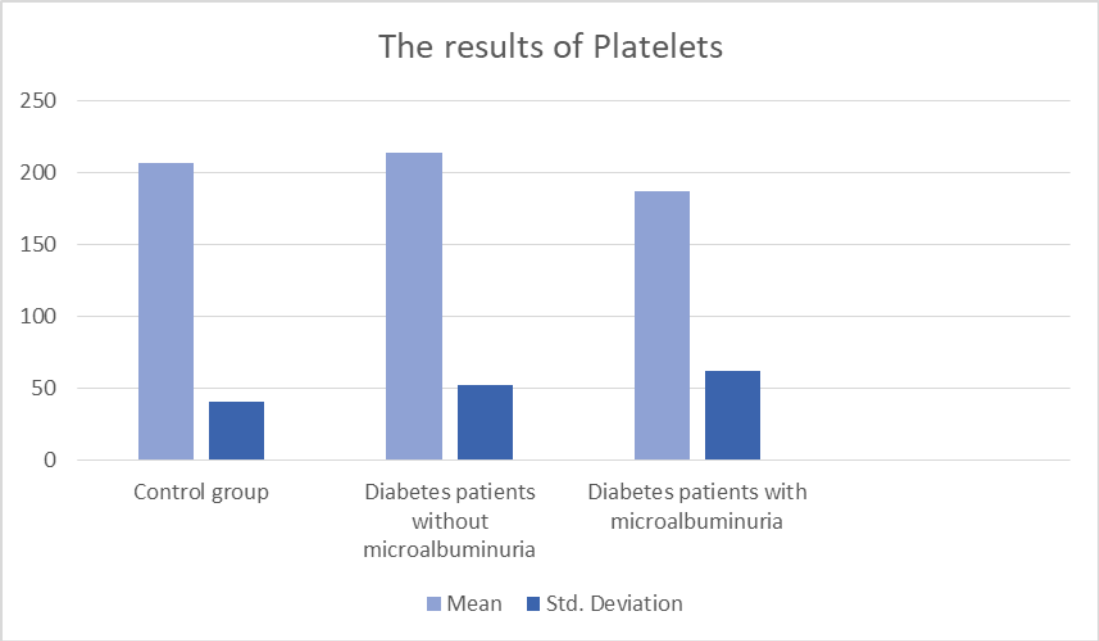

Figure 4.11 Platelets' test results

Within the population analyzed, there was no statistically significant variation in the primary variable.

4.4 Fibroblast Growth Factor-23 (FGF-23)

The mean levels of FGF-23 were (5.2885±6.28774) in the control group and (18.6180±24.40915) in the diabetes group without microalbuminuria and the diabetic group with microalbuminuria was (61.3294±138.20142), as shown in Table 4.7, Figure 4.12 and Table 4.8.

Table 4.7 The results of FGF-23

Parameter	Control group Mean±SD	Diabetes patients without microalbuminuria	Diabetes patients with microalbuminuria	p-value
FGF-23	5.2885±6.28774	18.6180±24.40915	61.3294 ±138.20142	0.003
	Std.error (0.88922)	Std.error (2.94731)	Std.error (19.35207)	

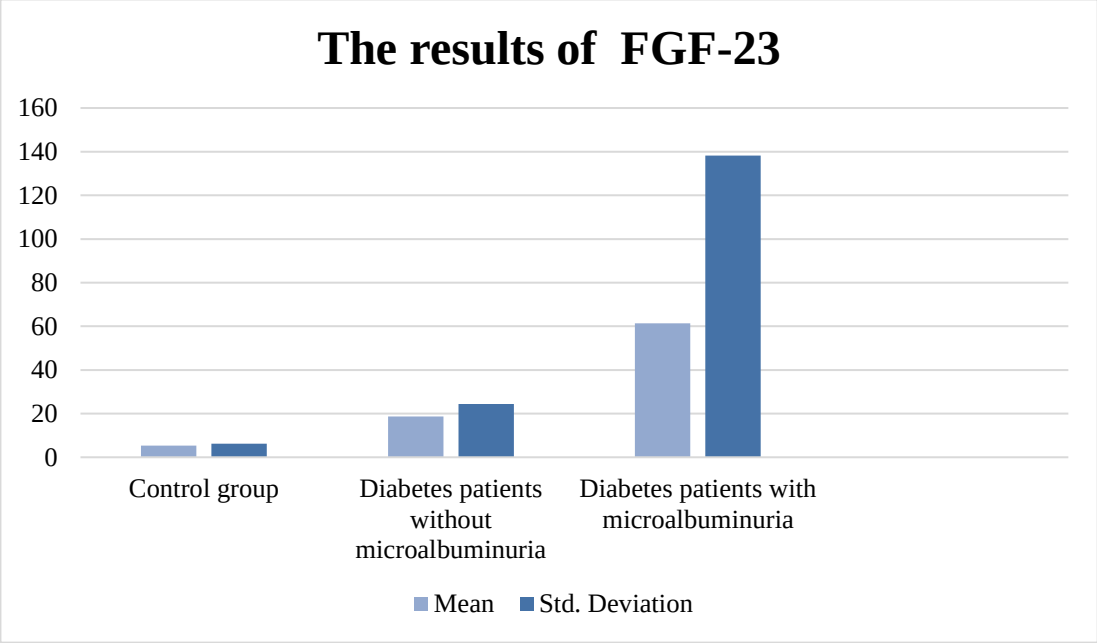

Figure 4.12 The effects of FGF-23

Table 4.8 Positive correlations between FGF-23 and study outcomes

FGF-23					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	86158.684	2	43079.342	6.226	0.003
Within Groups	982538.545	142	6919.286		
Total	1068697.229	144			

According to Table 4.8, the present research shows that there is a statistically significant difference in the mean serum FGF-23 concentration between the groups.

4.5 Fibroblast Growth Factor-23 (FGF-23) and Albumin Creatinine Ratio (ACR)

Table 4.9 shows that in all groups investigated, ACR was related to serum FGF-23 levels.

Table 4.9 Comparing ACR with FGF-23 outcomes

		ACR	FGF-23
ACR	Pearson Correlation	1	.431**
	Sig. (2-tailed)		.000
	N	145	145
FGF-23	Pearson Correlation	.431**	1
	Sig. (2-tailed)	.000	
	N	145	145

4.6 Discussion

Complications from diabetes mellitus may be severe and even fatal. Among the most prevalent kidney diseases, diabetic nephropathy. complication. of DM with high morbidity and mortality rate. Its early detection can improve the prognosis and general health of the patients. Microalbuminuria till now, is the best indicator of early Diabetic nephropathy that still be reversible when detected early. many other novel biomarkers are studied to find out their usefulness in the detection of nephropathy in comparison to

ACR, FGF-23 is one of the possible markers of nephropathy and has been studied in the current study for any possible role in the detection of early nephropathy.

FGF-23 was significantly higher in diabetic patients with microalbuminuria who participated in the current study this increase in FGF-23 in the group might be a clue to its usefulness in the detection of early nephropathy in replacement to ACR.

The correlation between FGF-23 and renal impairment has been explained by the associated deregulated phosphate homeostasis. it is established that FGF-23 levels progressively increase with the progressive impairment of renal function due to progressive PO4 relation.

The key factor that regulates the phosphate levels in the body is the fibroblast growth factor 23, or FGF23. It has been known that this hormone can be associated with adverse effects in patients with CKD. Emerging studies suggest that it plays a role in type 2 diabetes as well (Yeung *et al*. 2020). It is plausible that the role of FGF23 in the regulation of phosphate homeostasis is related to the development of chronic kidney disease. In addition to promoting the excretion of phosphate, it has been shown that high levels of FGF23 are also associated with the reduction of the renal capacity to remove phosphate. (Baia *et al*. 2015) Following kidney transplantation, the levels of FGF23 may decline, and this can lead to hypophosphatemia. In addition, deregulated levels of this important factor can affect other aspects of health, such as cardiovascular and neurodegeneration. moreover high levels of deregulated FGF23 are known to have an impact on various aspects of health, such as cardiovascular disease and mortality (van Londen *et al*. 2017).

Recently, it has been suggested that elevated FGF23 levels in patients with type 2 diabetes are also associated with adverse outcomes (Yeung *et al*. 2020).

In the current study, it seems that high FGF-23 increased at early stages (stage of microalbuminuria with normal GFR) making it a possible alternative to ACR. A similar

condition has been suggested by Ivana Pavik Philippe Jaeger and Lena Ebner. In their study which enrolled 87 patients with different stages of CKD.they have found that FGF-23 has been increased at early stages of CKD (Pavik *et al*. 2013).

In addition, there is study was made in 2017 by (alsaeed)and the study was conducted on 30 T2D patients with normal albumin excretion and matched them to a control group composed of 30 healthy individuals. The levels of FGF 23 were measured using an ELISA. In patients with diabetic nephropathy, the presence of microalbuminuria is not enough to identify them as at risk of developing kidney disease. Studies have also suggested that the growth factors of the fibroblasts could play a role in the development of this condition. The goal of this study was to determine the relationship between the levels of FGF23 in patients with type 2 diabetes and the other biochemical markers of this condition. In patients, the levels of FGF23 were significantly higher compared to those of the controls. There was also a positive correlation between this biochemical parameter and various other biochemical measures. It suggested that elevated FGF23 and serum levels could be used as a potential marker of kidney disease progression (El-Saeed and El-Mohasseb 2017).

This finding is very interesting as it suggests that FGF23 plays a role in the development and progression of chronic kidney disease (CKD).which is very important to find such a significant biomarker that can detect and monitor the progression of CKD and other studies confirm this statement, one of these studies is by Danilo Fliser that include the FGF23 is a novel independent predictor of progression of renal disease in patients with CKD (Fliser *et al*. 2007).

5. CONCLUSIONS AND RECOMMENDATION

5.1 Conclusions

- FGF-23 can be used as an alternative to ACR as an indicator of early diabetic nephropathy.

- FGF-23 can be useful as an indicator of the progression of diabetic nephropathy.

5.2 Recommendations

- Further studies are recommended to confirm the results of the current study hoping to include FGF-23 as an established biomarker of early diabetic nephropathy.

- Researches about new biomarkers of diabetic nephropathy are advised trying to minimize the catastrophic complications on patient's health.

- It is recommended to study any predictive role of the severity of increase FGF-23 in the progression of chronic kidney disease.

REFERENCES

Adeghate, E., Schattner, P. and Dunn, E. 2006. An update on the etiology and epidemiology of diabetes mellitus. Annals of the New York academy of sciences, 1084(1): 1-29.

Adler, A. I., Stevens, R. J., Manley, S. E., Bilous, R. W., Cull, C. A. and Holman, R. R. 2003. Development and progression of nephropathy in type 2 diabetes: the United Kingdom Prospective Diabetes Study (UKPDS 64). Kidney Int, 63(1): 225-232.

Alegre-Díaz, J., Herrington, W., López-Cervantes, M., Gnatiuc, L., Ramirez, R., Hill, M. and Collins, R. 2016. Diabetes and cause-specific mortality in Mexico City. New England Journal of Medicine, 375(20): 1961-1971.

Alicic, R. Z., Rooney, M. T. and Tuttle, K. R. 2017. Diabetic kidney disease: challenges, progress, and possibilities. Clinical journal of the American Society of Nephrology, 12(12): 2032-2045.

Amelia, R., Sari, D., Muzasti, R., Fujiati, I. and Wijaya, H. 2021. Early detection of diabetic nephropathy based on albumin creatinine ratio (acr) in type 2 diabetes mellitus patients in Medan. Indonesia. Family Medicine andamp; Primary Care Review, 23(2): 135-138.

Amer, A. H. and Haridas, N. 2018. Early Diagnostic Markers in Diabetic Nephropathy Patients. Journal of Clinical and Diagnostic Research, 1(2): 11.

Baia, L. C., Heilberg, I. P., Navis, G. and De Borst, M. H. 2015. Phosphate and FGF-23 homeostasis after kidney transplantation. Nature Reviews Nephrology, 11(11): 656-666.

Basilico, C. and Moscatelli, D. 1992. The FGF family of growth factors and oncogenes. Advances in cancer research, 59, 115-165.

Baum, M., Schiavi, S., Dwarakanath, V. and Quigley, R. 2005. Effect of fibroblast growth factor-23 on phosphate transport in proximal tubules. Kidney International, 68(3): 1148-1153.

Bhattacharjee, A., Easwaran, A., Leow, M. K.-S. and Cho, N. 2018. Evaluation of an artificial pancreas in silico patients with online-tuned internal model control. Biomedical Signal Processing and Control, 41: 198-209.

Bishay, R. H. and Greenfield, J. R. 2017. A review of maturity onset diabetes of the young (MODY) and challenges in the management of glucokinase-MODY. Med J Aust, 207(5): 223. doi:10.5694/mja16.01467

Biswas, S. 2022. A review on the progress of stem cell therapy as a treatment for Diabetes mellitus. Brac University,

Bonventre, J. V. 2014. Current biomarkers in kidney disease: Dawning of a new era. ASN Kidney News, 1: 7-8.

Cai, P., Wu, Z., Huang, W., Niu, Q., Zhu, Y. and Yin, D. 2021. Suoquan pill for the treatment of diabetic nephropathy: A protocol for systematic review and meta-analysis. Medicine (Baltimore): 100(17): e25613.

Cho, N. H., Shaw, J., Karuranga, S., Huang, Y., da Rocha Fernandes, J., Ohlrogge, A. and Malanda, B. 2018. IDF Diabetes Atlas: Global estimates of diabetes prevalence for 2017 and projections for 2045. Diabetes research and clinical practice, 138: 271-281.

Conlon, K. C. and Duggan, S. N. 2017. Pancreatogenic type 3c diabetes: underestimated, underappreciated and poorly managed. Practical gastroenterology, 1: 5.

Conroy, R. M., Pyörälä, K., Fitzgerald, A. e., Sans, S., Menotti, A., De Backer, G. and Keil, U. 2003. Estimation of ten-year risk of fatal cardiovascular disease in Europe: the SCORE project. European heart journal, 24(11): 987-1003.

Crestani, M., Sadeghpour, A., Stroup, D., Galli, G. and Chiang, J. Y. 1998. Transcriptional activation of the cholesterol 7α-hydroxylase gene (CYP7A) by nuclear hormone receptors. Journal of lipid research, 39(11): 2192-2200.

Cui, Y. and Andersen, D. K. 2011. Pancreatogenic diabetes: special considerations for management. Pancreatology, 11(3): 279-294.

De Franco, E., Flanagan, S. E., Houghton, J. A., Allen, H. L., Mackay, D. J., Temple, I. K. and Hattersley, A. T. 2015. The effect of early, comprehensive genomic testing on clinical care in neonatal diabetes: an international cohort study. The Lancet, 386(9997): 957-963.

Eker, S. 2018. Prevalence of depression symptoms in diabetes mellitus. Open access Macedonian journal of medical sciences, 6(2): 340.

El-Dawla, N. M. Q., Sallam, A.-A. M., El-Hefnawy, M. H. and El-Mesallamy, H. O. 2019. E-cadherin and periostin in early detection and progression of diabetic nephropathy: epithelial-to-mesenchymal transition. Clinical and experimental nephrology, 23(8): 1050-1057.

El-Saeed, A. M. and El-Mohasseb, G. F. 2017. Circulating Fibroblast Growth Factors 21 and 23 as Biomarkers of Progression in Diabetic Nephropathy in Type 2 Diabetes with Normoalbuminuria. The Egyptian journal of immunology, 24(2): 93-99.

Fliser, D., Kollerits, B., Neyer, U., Ankerst, D. P., Lhotta, K., Lingenhel, A. and Kronenberg, F. 2007. Fibroblast growth factor 23 (FGF23) predicts progression of chronic kidney disease: the Mild to Moderate Kidney Disease (MMKD) Study. Journal of the American Society of Nephrology, 18(9): 2600-2608.

Froguel, P., Vaxillaire, M., Sun, F., Velho, G., Zouali, H., Butel, M. O. and Cohen, D. 1992. Close linkage of glucokinase locus on chromosome 7p to early-onset non-insulin-dependent diabetes mellitus. Nature, 356(6365): 162-164.

Goetz, R., Beenken, A., Ibrahimi, O. A., Kalinina, J., Olsen, S. K., Eliseenkova, A. V. and Linhardt, R. J. 2007. Molecular insights into the klotho-dependent, endocrine mode of action of fibroblast growth factor 19 subfamily members. Molecular and cellular biology, 27(9): 3417-3428.

Gupta, K., Nayyar, S., Sachdeva, J. and Kumar, P. 2017. Cystatin C in the early diagnosis of diabetic nephropathy and its correlation with albuminuria. International Journal of Advances in Medicine, 4(1): 56-59.

Gutiérrez, O. M., Mannstadt, M., Isakova, T., Rauh-Hain, J. A., Tamez, H., Shah, A. and Jüppner, H. 2008. Fibroblast growth factor 23 and mortality among patients undergoing hemodialysis. New England Journal of Medicine, 359(6): 584-592.

Gutierrez, O., Isakova, T., Rhee, E., Shah, A., Holmes, J., Collerone, G. and Wolf, M. 2005. Fibroblast growth factor-23 mitigates hyperphosphatemia but accentuates calcitriol deficiency in chronic kidney disease. Journal of the American Society of Nephrology, 16(7): 2205-2215.

Habeb, A. M., Al-Magamsi, M. S., Eid, I. M., Ali, M. I., Hattersley, A. T., Hussain, K. and Ellard, S. 2012. Incidence, genetics, and clinical phenotype of permanent

neonatal diabetes mellitus in northwest Saudi Arabia. Pediatric Diabetes, 13(6): 499-505.

Hall, J. E. and Guyton, A. C. 2006. Pocket companion to Guyton and Hall textbook of medical physiology: Elsevier Health Sciences TW.

Inker, L. A. and Titan, S. 2021. Measurement and estimation of GFR for use in clinical practice: core curriculum 2021. American Journal of Kidney Diseases, 78(5): 736-749.

Jwad, S. M. and AL-Fatlawi, H. Y. 2022. Types of Diabetes and their Effect on the Immune System. Journal of Advances in Pharmacy Practices (e-ISSN: 2582-4465): 21-30.

Kendrick, J., Cheung, A. K., Kaufman, J. S., Greene, T., Roberts, W. L., Smits, G. and Investigators, t. H. 2011. FGF-23 Associates with Death, Cardiovascular Events, and Initiation of Chronic Dialysis. Journal of the American Society of Nephrology, 22(10): 1913-1922.

Khosravi, A., Cutler, C. M., Kelly, M. H., Chang, R., Royal, R. E., Sherry, R. M. and Collins, M. T. 2007. Determination of the elimination half-life of fibroblast growth factor-23. The Journal of Clinical Endocrinology and Metabolism, 92(6): 2374-2377.

King, H., Aubert, R. E. and Herman, W. H. 1998. Global burden of diabetes, 1995–2025: prevalence, numerical estimates, and projections. Diabetes care, 21(9): 1414-1431.

Koh, N., Fujimori, T., Nishiguchi, S., Tamori, A., Shiomi, S., Nakatani, T. and Kuroki, T. 2001. Severely reduced production of klotho in human chronic renal failure kidney. Biochemical and biophysical research communications, 280(4): 1015-1020.

Kouri, T. T., Viikari, J. S., Mattila, K. S. and Irjala, K. M. 1991. Microalbuminuria: invalidity of simple concentration-based screening tests for early nephropathy due to urinary volumes of diabetic patients. Diabetes care, 14(7): 591-593.

Larsson, T., Nisbeth, U., Ljunggren, Ö., Jüppner, H. and Jonsson, K. B. 2003. Circulating concentration of FGF-23 increases as renal function declines in patients with chronic kidney disease, but does not change in response to variation in phosphate intake in healthy volunteers. Kidney International, 64(6): 2272-2279.

Levin, A., Bakris, G., Molitch, M., Smulders, M., Tian, J., Williams, L. and Andress, D. 2007. Prevalence of abnormal serum vitamin D, PTH, calcium, and phosphorus in patients with chronic kidney disease: results of the study to evaluate early kidney disease. Kidney International, 71(1): 31-38.

Loewen, S. L. and Haas, L. B. 1991. Complications of diabetes: acute and chronic. Nurse Pract Forum, 2(3): 181-187.

Long, A. N. and Dagogo-Jack, S. 2011. Comorbidities of diabetes and hypertension: mechanisms and approach to target organ protection. The journal of clinical hypertension, 13(4): 244-251.

Maggiore, U., Budde, K., Heemann, U., Hilbrands, L., Oberbauer, R., Oniscu, G. C. and Abramowicz, D. 2017. Long-term risks of kidney living donation: review and position paper by the ERA-EDTA DESCARTES working group. Nephrology Dialysis Transplantation, 32(2): 216-223.

Marsell, R., Grundberg, E., Krajisnik, T., Mallmin, H., Karlsson, M., Mellstrom, D. and Ljunggren, O. 2008. Fibroblast growth factor-23 is associated with parathyroid hormone and renal function in a population-based cohort of elderly men. European journal of endocrinology, 158(1): 125-130.

McFarlane, P., Cherney, D., Gilbert, R. E. and Senior, P. 2018. Chronic kidney disease in diabetes. Canadian journal of diabetes, 42: S201-S209.

Modi, P. 2007. Diabetes beyond insulin: review of new drugs for treatment of diabetes mellitus. Current drug discovery technologies, 4(1): 39-47.

Motawi, T. K., Shehata, N. I., ElNokeety, M. M. and El-Emady, Y. F. 2018. Potential serum biomarkers for early detection of diabetic nephropathy. Diabetes research and clinical practice, 136: 150-158.

Pavik, I., Jaeger, P., Ebner, L., Wagner, C. A., Petzold, K., Spichtig, D. and Serra, A. L. 2013. Secreted Klotho and FGF23 in chronic kidney disease Stage 1 to 5: a sequence suggested from a cross-sectional study. Nephrology Dialysis Transplantation, 28(2): 352-359.

Price, S., Cole, D. and Alcolado, J. 2010. Diabetes due to exocrine pancreatic disease—a review of patients attending a hospital-based diabetes clinic. QJM: An International Journal of Medicine, 103(10): 759-763.

Rowley, W. R. and Bezold, C. 2012. Creating public awareness: state 2025 diabetes forecasts. Population health management, 15(4): 194-200.

Rubio-Cabezas, O., Hattersley, A. T., Njølstad, P. R., Mlynarski, W., Ellard, S., White, N., . . . Craig, M. E. 2014. ISPAD Clinical Practice Consensus Guidelines 2014. The diagnosis and management of monogenic diabetes in children and adolescents. Pediatr Diabetes, 15(20): 47-64.

Rufo, D. D., Debelee, T. G., Ibenthal, A. and Negera, W. G. 2021. Diagnosis of diabetes mellitus using gradient boosting machine (LightGBM). Diagnostics, 11(9): 1714.

Seiler, S., Heine, G. H. and Fliser, D. 2009. Clinical relevance of FGF-23 in chronic kidney disease. Kidney International, 76: S34-S42.

Sharifi, A. M., Zare, B., Keshavarz, M., Rahmani, M., Zaeefy, B. and Larijani, B. 2015. Urinary N-acetyl-β-D-glucosaminidase (NAG) activity in the early detection of diabetic nephropathy. International Journal of Diabetes in Developing Countries, 35(3): 369-374.

Shaw, J. E., Sicree, R. A. and Zimmet, P. Z. 2010. Global estimates of the prevalence of diabetes for 2010 and 2030. Diabetes research and clinical practice, 87(1): 4-14.

Sommese, L., Zullo, A., Mancini, F. P., Fabbricini, R., Soricelli, A. and Napoli, C. 2017. Clinical relevance of epigenetics in the onset and management of type 2 diabetes mellitus. Epigenetics, 12(6): 401-415.

Thipsawat, S. 2021. Early detection of diabetic nephropathy in patient with type 2 diabetes mellitus: A review of the literature. Diabetes and Vascular Disease Research, 18(6): 14791641211058856.

Thornton Snider, J., Sullivan, J., van Eijndhoven, E., Hansen, M. K., Bellosillo, N., Neslusan, C. and Kasiske, B. L. 2019. Lifetime benefits of early detection and treatment of diabetic kidney disease. PLOS ONE, 14(5): e0217487.

van Londen, M., Aarts, B. M., Deetman, P. E., van der Weijden, J., Eisenga, M. F., Navis, G. and de Borst, M. H. 2017. Post-transplant hypophosphatemia and the risk of death-censored graft failure and mortality after kidney transplantation. Clinical journal of the American Society of Nephrology, 12(8): 1301-1310.

Veiga, G., Alves, B., Perez, M., Alcantara, L. V., Raimundo, J., Zambrano, L. and Murad, N. 2020. NGAL and SMAD1 gene expression in the early detection of

diabetic nephropathy by liquid biopsy. Journal of Clinical Pathology, 73(11): 713-721.

Wang, L., Kong, L., Wu, F., Bai, Y. and Burton, R. 2005. Preventing chronic diseases in China. The lancet, 366(9499): 1821-1824.

Yamamoto-Honda, R. and Akanuma, Y. 2002. Classification of diabetes mellitus. Nihon rinsho. Japanese journal of clinical medicine, 60: 363-371.

Yamashita, T. 2005. Structural and biochemical properties of fibroblast growth factor 23. Therapeutic Apheresis and Dialysis, 9(4): 313-318.

Ye, B., Zhu, X., Zeng, Z., Ji, X. and Ji, M. 2021. Clinical significance of serum homocysteine as a biomarker for early diagnosis of diabetic nephropathy in type 2 diabetes mellitus patients. Pteridines, 32(1): 11-16.

Yeung, S. M., Bakker, S. J., Laverman, G. D. and De Borst, M. H. 2020. Fibroblast growth factor 23 and adverse clinical outcomes in type 2 diabetes: a bitter-sweet symphony. Current diabetes reports, 20(10): 1-9.

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