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A biochemical study of fibroblast growth factor -23 , PTH and phosphorus metabolism in adult patients with obstructive renal failure in Babylon-Iraq

A Thesis

Submitted to the Council of the College of Medicine University of Babylon in Partial Fulfillment of the Requirements for the Degree of Master of Science in Clinical Biochemistry

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بِسْمِ اللَّهِ الرَّحْمَٰنِ الرَّحِيمِ

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صَدَقَ اللّٰهُ العلـيّ العَظيم

سوره يوسف الايه:٧٦

Supervisor Certificates

We certify that this thesis entitled **(A biochemical study of fibroblast growth factor -23, PTH and phosphorus metabolism in adult patients with obstructive renal failure in Babylon-Iraq)** has been prepared under our supervision at the Department of Biochemistry, College of Medicine, University of Babylon, in partial requirements for the degree of master in Clinical Biochemistry.

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Dedication

Foremost, I thank Allah (subhanahu wa ta`ala) for letting me live till this moment to see this thesis through My great teacher and messenger Mohammed (God bless him and grant him salvation) who Lead us in this life from the darkness to the light Of hope my dear All patients Especially those who took part in the research, My parents For their endless support and they were always a source of strength during moments of despair and discouragement, My wife Who taught me that even the largest task can be accomplished if I do it one step at a time, to all my family, my friends, I dedicate this research.

Mohammed 2021

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Summary

Obstructive uropathy is the obstruction to the flow of urine, which may be complete or partial, and may cause gradual and progressive damage to the kidneys. The obstruction could be caused to one or both ureters and may occur at the level of the bladder neck or distal to that (such as in the urethra). Obstruction is one of the major causes of renal failure and several studies have focussed at understanding the etiology and consequences of obstructive uropathy.

The study's objective is to investigate the relationship between serum levels of Fibroblast growth factor-23 (FGF-23), PTH, and phosphorus in patients with obstructive renal failure and healthy controls.

The study involved collecting blood samples from 100 volunteers, 50 healthy subjects (38 men and 12 women) and (34 men and 16 women) suffering from obstructive renal failure.age was (25 –75) years BMI with (24-27) Kg/m^2. Patients were subjected to Urology department at Hillah Hospital. The levels of FGF23&PTH& PO4 was measured by ELISA technique whereas the levels of other parameters were measured by colorimetric method according to the manufacturer manual.

As the results of the tests that were conducted showed that the levels of Hemoglobin, Sodium, GFR, Calcium, in the people with obstructive renal disease were significantly lower than healthy people, as the value of $P < 0.05$. The levels of Fibroblast growth factor 23, Parathyroid hormone, Phosphorus, Potassium, in people with obstructive renal disease were significantly higher than healthy people, as the value of $P < 0.05$.Also, the relationship between the levels of Fibroblast growth factor 23 and GFR was studied, where the value of $r = 0.185$, P 0.199 was found and there was anon significant positive correlation between them&also non-significant

positive correlation between Fibroblast growth factor 23& Parathyroid hormone r = 0.029, P 0.843.

It can be concluded from the current study that we carried out that the level of occurrence of The Fibroblast growth factore23 , parathyroid hormone & phosphors is higher in patient with obstructive renal failure than those healthy control ,The FGF23 could be served as a prognostic marker in obstructive renal failure patients to predict the possibility to develop chronic kidney disease, The occurrence of obstructive renal disease at a large rate in old age and in men more than women, The current study showed that the BMI is not significant in patient compared with healthy control ,There was a decrease in sodium and calcium and GFR levels in patients compared to healthy people , The presence of a positive relationship between the fibroblast growth factor& parathyroid hormone, The presence of a positive relationship between the GFR& fibroblast growth factor, parathyroid hormone, phosphorus & hemoglobin that were performed and inverse relationship between GFR & creatinine, urea ,calcium, potassium ,We worked on studying the inverse relationship between hemoglobin levels and creatinine ,urea & in present study we worked on studying the relationship between phosphorus ,potassium ,sodium &calcium levels and parameters, and a strong inverse and positive relationship existed between them.

Contents

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List of Abbreviation

Abbreviation	Details
AKI	Acute Kidney Injury
AKD	Acute kidney disease
ADHR	Autosomal dominant hypophosphatemic rickets
BMI	Body mass index
BPH	Benign prostatic hyperplasia
β-Klotho	Beta Klotho
CKD	Chronic Kidney Disease
Cl	Chloride
eGFR	Estimated Glomerular Filtration Rate
ESRD	End-Stage Renal Disease
FGF	Fibroblast growth factors
FGFR	fibroblast growth factor receptor
FBS	Fasting Blood Sugar
GFR	Glomerular Filtration Rate
GOD	Glucose oxidase
HRP	Horseradish Peroxidase
iFGFs	intracellular FGFs
KL	Klotho
n-;oxPTH	Non-oxidized PTH
NF -κ B	nuclear factor kappa B
kPTH	Parathyroid hormone
POD	Peroxidase
Rpm	Revolution per minute'
SKl	Soluble Klotho

SCr	Serum creatinine
TIO	tumor-induced rickets/osteomalacia
UTIs	Urinary tract infections
UN	United Nations
NaPi-2a	Sodium-dependent phosphate transport protein 2A
Na	Sodium
NCC	Sodium chloride cotransporter
OU	Obstructive uropathy
ON	Obstructive nephropathy
OxPTH	oxidized PTH
TMB	Tetramethylbenzidine
TMB	Tetramethylbenzidine
TGF-β	transforming growth factor beta
VDR	vitamin D receptor
WHO	World Health Organization
XLH	X-linked hypophosphatemic
Symbols	
O.D.	Optical density
Da	Dalton
ng/mL	Nanograms per milliliter

1. Obstructive Renal Failure

1.1. Overview

Obstructive uropathy is the blockage to the flow of urine which may be complete or partial, and may cause gradual and progressive damage to the kidneys. The blockage could be caused to one or both ureters and may occur at the level of the bladder neck or distal to that (such as in the urethra). Obstruction is one of the major causes of renal failure and several studies have focussed at understanding the etiology and consequences of obstructive uropathy [1]. Kidney obstructions are common medical problems that affect many people. Most renal obstructions are the result of nephrolithiasis or kidney stones[2]. However, polycystic kidney disease and renal artery stenosis also contribute to renal obstructions in differing pathologic ways. Other less common causes of kidney obstruction include renal polyps or tumors[3].

Obstructive uropathy (OU) is a common clinical condition in the elderly, particularly in men who frequently suffer from benign prostate hyperplasia and from prostate cancer. It refers to either structural and/or functional abnormalities of the urinary tract that obstruct urine flow. Obstructive nephropathy (ON) refers to renal injury and damage of the renal parenchyma that results from this impaired urine flow. The clinical manifestation of OU varies in the elderly, ranging from an asymptomatic clinical condition to a severe acute kidney injury needing dialysis. Hydronephrosis, defined as a distension of renal pelvis and the calyces, is an indicative hallmark of OU[4]. In the elderly (>64 years), the incidence of hydronephrosis is higher than in younger age (5.1% vs. 3.1%) especially in elderly males (6.2% in men and 2.9% in women) [3]. If inadequately treated (or if it remains untreated), OU results in Obstructive nephropathy (ON). Obstructive nephropathy (ON) may lead to chronic kidney disease and may progress to end-stage renal disease (ESRD). Prolonged

obstruction (for more than 6 weeks) leads to hydronephrosis and a significant loss of functional kidney parenchyma of the obstructed kidney. However, renal function may be also altered even by a short-term obstruction. To note, 57.4% of patients with ESRD due to ON are older than 64 years and 73.8% are males[5]. In the elderly, ON is a very important factor influencing morbidity and mortality and a major cause of ESRD.

1.2 Etiology

Prostate disease including hyperplasia or cancer, retroperitoneal, cirrhosis or pelvic neoplasms and lithiasis are the primary causes of OU in the elderly[6]. Urinary tract obstruction can occur at any point in the urinary tract and is classified by the level of obstruction and whether the cause is intrinsic or extrinsic to the urinary tract (Table 1-1)[7]. Renal obstruction in the elderly can be due to benign conditions such as renal cystic or calculous disease or malignant conditions such transitional cell carcinoma of the renal pelvis[8]. Along the course of the ureters, intrinsic causes for obstruction include calculi, ureteral strictures, and neoplasms of the transitional cell epithelium[9][10]. Extrinsic compression can be due to vascular lesions such as aortic or iliac artery aneurysms, retroperitoneal malignancies such as colon cancer or metastatic bladder cancer, or inflammatory conditions such as retroperitoneal fibrosis. Although less common in elderly women, gynecologic malignancies may also be a source of urinary tract obstruction[7]. In the lower urinary tract, the most common reason for obstruction is Benign prostatic hyperplasia (BPH)[11]. Other causes include bladder calculi, urethral stricture, and neoplasms of the bladder, prostate, or urethra[12]. In women, prolapse of pelvic organs such as the bladder, rectum, or small bowel through the vagina can also lead to functional outlet obstruction through kinking or compression of the urethra[13]. Iatrogenic injuries such as bladder neck contractures

secondary to radical prostatectomy and urethral strictures secondary to urethral instrumentation are also possibilities[14]. Finally, indwelling urethral catheters, suprapubic catheters, or percutaneous nephrostomy tubes may become dislodged or kinked resulting in obstruction to urinary outflow[15]that rarely cause obstructive failure.

Table (1-1): Causes of urinary tract obstruction [7]

	Intrinsic	Extrinsic
Kidney	• Calculous disease • Cystic disease • Renal cell carcinoma • Transitional cell carcinoma of the renal pelvis • Obstructive pyelonephritis • Congenital fibrous ureteropelvic ,junction obstruction	• Ureteropelvic junction obstruction as a result of a crossing vessel
Ureter	• Calculous disease • Stricture • Transitional cell carcinoma • Congenital megaureter	• Aortic or iliac artery aneurysm • Compression because of vascular graft • Retroperitoneal malignancy • Retroperitoneal fibrosis • Pelvic lipomatosis • Gynecologic malignancy
Bladder	• Neurogenic bladder • Bladder neck contracture • Malignancy of the bladder neck or prostate • Calculous disease	• BPH • Prostatitis • Pelvic organ prolapse
Urethra	• Stricture • Phimosis • Meatal stenosis • Malignancy of the urethra	

1. 3 Clinical Manifestations

Symptoms and signs of OU/ON are usually not specific and depend largely on the extension (partial or complete), the duration (acute or chronic) and the site of the obstruction. In addition, the clinical presentation depends on whether the obstruction is unilateral or bilateral. Given that the clinical presentation depends on the degree of the obstruction, partial or/and unilateral obstructions can cause oliguria whereas bilateral obstruction results in acute kidney disease (AKI) with anuria[16]. Often, elderly patients remain asymptomatic even in severe obstruction if it occurs gradually and is chronic. To note, elderly patients with impaired cognitive function may lose the ability to describe their symptoms making the diagnosis of OU more difficult, resulting in treatment delay and permanent kidney damage[7]. Acute upper tract obstruction has a similar presentation to renal colic, Pain and discomfort of the flank, suprapubic or groin region are indicative of OU (complete bilateral or complete unilateral). The pain indicates an acute or rapidly developing obstruction of the urinary tract; it is of increased intensity radiating to the groin, the testicles or the labia[17]. Patients may present with anuria if they have complete obstruction of the ureter, both ureters, or have unilateral obstruction of a solitary kidney[18]. Extrinsic compression of the urinary system is more commonly a chronic process with a slow progression to renal insufficiency. Consequently, patients often present with vague symptoms, such as back pain, anorexia, lethargy, and/or mental status changes[19]. While no specific prevalence is known, in advanced prostate cancer, urinary tract obstruction is often insidious and silent. These patients can be relatively asymptomatic, with hydronephrosis being discovered as an incidental finding during a workup for renal insufficiency. In some cases, a urinary tract infection may be the heralding symptom[20][12].

1.4 The Clinical Examination and Laboratory Findings associated with Obstructive Uropathy

Similarly, to the symptoms of urinary obstruction, the physical examination signs are often related to the cause and location of the obstruction. These same signs can also be absent completely and often the diagnosis can only be made on laboratory testing and diagnostic imaging. Lower tract obstruction signs that may be present are an distended caused by over distension or palpable masses. Physical examination therefore should include an abdominal examination to evaluate for abdominal tumor or a palpable distended bladder; if male, a prostate examination; or, if female, a full pelvic examination looking for obstruction[21]. Upper urinary tract obstruction signs are even less obvious on physical examination, although in some instances costovertebral angle tenderness may be present. All patients for whom obstruction is a concern should have basic serum studies in addition to a urinalysis. Serum findings are often nonspecific but can range from a mild increase in the white blood cell counts on complete blood count to a severe increase of creatinine and potassium levels on the basic metabolic panel related to complete obstruction[22]. Microscopic hematuria is an abnormal laboratory finding that is sometimes seen in the urinalysis with obstructive uropathy. The American Urologic Association guidelines define microscopic hematuria as 3 or more red blood cells per high-power field from 2 of 3 properly collected urinalysis specimens, Red blood cell count on urinalysis can be an indicator of calculi, infection, or malignancy as the source of obstruction and should be worked up accordingly[23] .

1. 5. Complications of Obstructive Uropathy

1.5.1 Acute Kidney Injury

As mentioned, bilateral obstruction or complete obstruction of a solitary kidney lead to Acute Kidney Injury (AKI)[24]. AKI is the rapid loss of renal function occurring over hours or days. The risk of AKI in the elderly is high, due to the several anatomic and physiological changes of the aging kidney. Post renal AKI or obstructive AKI is very common in the elderly. Prostate processes (benign hypertrophy/carcinoma), retroperitoneal adenopathy, malignancies, and neurogenic bladder are the most common causes of obstructive AKI in men, whereas pelvic and retroperitoneal carcinomas in the women. Nephrolithiasis and calculi are common causes of post-renal AKI in the elderly. In post renal AKI, anuria or oligoanuria suggests complete obstruction, whereas flank or abdominal pain with or without suprapubic fullness may also present. To note, elderly patients with partial obstruction may remain asymptomatic, and usually they report macroscopic hematuria, hesitancy, urgency and nocturia. Urine outflow varies from oliguria to polyuria. Given that these symptoms are not specific for post renal AKI, the aged patients are being undervalued, which delays the diagnosis. Laboratory findings are nonspecific and they do not differ from the other types of AKI. Obstruction of the urinary tract should be considered in elderly patients with uremia and no previous history of CKD or cardiovascular disease with relatively benign urine sediment (absence of hematuria of kidney origin and massive proteinuria)[25].

1.5.2 Hypertension

Hypertension is common in acute or chronic obstruction and concomitant ON. The pathogenesis of hypertension in ON is multifactorial. Volume overload with increased extracellular fluid volume, decreased sodium excretion and increased sodium absorption with an activation of

the renin-angiotensin system are implicated in the pathogenesis of hypertension. In bilateral obstruction, it seems that hypertension is due to water and sodium retention. In support of this, the use of diuretics leads also to blood pressure control, suggesting that hypertension is volume dependent. Interestingly, the concentration of rennin in renal and peripheral venous is normal in patients with bilateral obstruction. On the contrary, there is evidence that hypertension in patients with unilateral obstruction is renin-dependent[26]. Data from animal studies with unilateral ureteral obstruction indicate that hypertension in this clinical condition is renin dependent [27]. In support of this, high renin blood levels have been reported both in the renal and peripheral veins. Furthermore, in chronic unilateral ureteral obstruction the venous levels of renin are normal suggesting that the pathogenesis of hypertension in OU is more complex. It should be emphasized that relief of obstruction reverses the high blood pressure. Hypertension may also contribute to renal fibrosis, which characterizes ON. In addition, hypertension is a common finding in the elderly partially due to structural and functional changes of the aged kidney. Arterial stiffness, neurohormonal, and autonomic dysregulation are implicated in the pathogenesis[28].In this regard, OU may aggravate persistent hypertension due to aging and in return may aggravate ON and develop CKD.

1.5. 3 Urinary Tract Infection

The urinary tract is one of the most common sites of bacterial infection, UTI is a disease of worldwide importance and in some instances it threatens renal function and may be life. However a considerable proportion of the population may acquire asymptomatic infection. It may involve just the lower tract or both lower and upper tracts[29]. Factors that

predispose to urinary tract infections (UTIs) in the elderly are the high postvoid residual urine volume in the bladder that enables bacterial growth and some alterations of the urinary tract epithelial cells especially in the glycoprotein composition of the bladder epithelium that facilitate bacterial adhesion. The most common organism responsible for UTI is Escherichia coli. To note, the obstruction of the upper urinary tract usually is not complicated with an infection[30].

1.5.4 Hyperkalemic Hyperchloremic Acidosis

Hyperkalemic hyperchloremic acidosis due to renal tubular acidosis type IV has been reported in patients with unilateral obstruction. Interestingly, the unilateral ureteral obstruction in animal models leads to a defect of urine acidification[31].

1. 6. Pathologic Changes of Obstructive Nephropathy

Hydronephrosis is the result of inadequate drainage of urine relative to the amount of urine produced by the kidney. Inadequate drainage of the collecting system causes increased pressure within the collecting system reflected into the renal tubules. In the acute setting (6–48 h), increased pressure causes increased intraparenchymal vascular resistance, but this phenomenon often normalizes after 48 h[34]. Long-standing hydronephrosis can result in renal injury, fibrosis, and atrophy of the affected kidney[33][34].When hydronephrosis is unilateral, the contralateral kidney hypertrophies to compensate for the increased urine excretion. However, when both kidneys are affected, urine will not be adequately excreted and can result in electrolyte abnormalities and uremia, necessitating dialysis. If the cause of the hydronephrosis is treated when the kidney is still functioning with a GFR > 10 mL/min/1.73 m2 , the renal injury can be partially or completely reversible[35]. As such, determining acuity of the hydronephrosis and residual renal function is

important for guiding management and preserving renal function. Causes of hydronephrosis can be classified into functional and obstructive etiologies. Functional hydroureteronephrosis (kidneys + ureter dilation) is normally seen in 80–90% of pregnancies, is more pronounced in primigravid patients, and will resolve by 6 weeks postpartum[36]. This is thought to be due to ureteral compression at the pelvic brim with some evidence suggesting hormone alterations, mainly progesterone, can enhance the collecting system dilation. The right ureter is usually dilated more than the left due its acute angle as it courses over the iliac vessels [37]. Obstructive hydronephrosis will have varying etiology based on the patient's age. In neonates and young children, anatomic abnormalities including congenital stenosis at the ureteropelvic/ureterovesical junctions, vesicoureteral reflux, or posterior urethral valves are the most likely etiology. In adults, the most common cause is urolithiasis; approximately 8.8% of adults in the United States have a history of at least one urinary calculus with the highest incidence in obese white males with a history of prior renal calculi [38]. In older adults, benign prostatic hyperplasia and urothelial carcinomas should also be considered, particularly in patients with hematuria. Other causes include urinary tract injury (including iatrogenic), strictures, obstructing infections (nephritis, cystitis, prostatitis), external compressing masses, and neurogenic bladder with inadequate/ineffective voiding[30]**.**

1.7. Pathophysiology of Obstructive Nephropathy

Acquired obstructive nephropathy in humans results from partial urinary obstruction in most cases and tends to be prolonged in its clinical course. But most physiologic studies of renal function in obstruction are based on models of acute complete obstruction for 24 hours[39].

1. 7.1 Hemodynamic Effects

Urinary obstruction significantly alters renal blood flow, glomerular filtration rate (GFR), and tubular function even before anatomic changes occur in the kidney[40]. Within the first 2 to 3 hours of obstruction, there is an early vasodilator response; termed the 'hyperemic phase.' The rise in hydrostatic pressure in the proximal tubule initially results in reduced resistance of the afferent arteriole and increased glomerular hydrostatic pressure to counteract the proximal tubular pressure[39]. The reduced distal tubular flow contributes to the initial rise in single nephron GFR as part of tubuloglomerular feedback. Thus in the initial phase of obstruction, single nephron GFR is maintained at approximately 80% of the pre-obstruction values, despite the marked increase in proximal tubular pressure[41]. As obstruction persists in the next 12 to 24 hours, there is a late vasoconstrictor phase, which is characterized by a drop in renal blood flow to about 40% of normal and poor renal perfusion[30]. Two major vasoconstrictors, angiotensin II and thromboxane A2, play an important role in the markedly reduced renal blood flow and reduction in single nephron GFR in obstruction[42]. After the obstruction is relieved, there is further vasoconstrictor response in the kidney caused by the release of angiotensin II, as the macula dense senses the change in tubular flow. In animal experiments, simultaneous inhibition of thromboxane A2 and angiotensin production normalized GFR in the post obstructed kidney (figure (1-1))[43]. The administration of atrial natriuretic peptide after release of obstruction in rats also resulted in an increase of GFR, urine flow, and sodium excretion, suggesting a role of atrial natriuretic peptide in the hemodynamic changes of the post obstructed kidney[44].

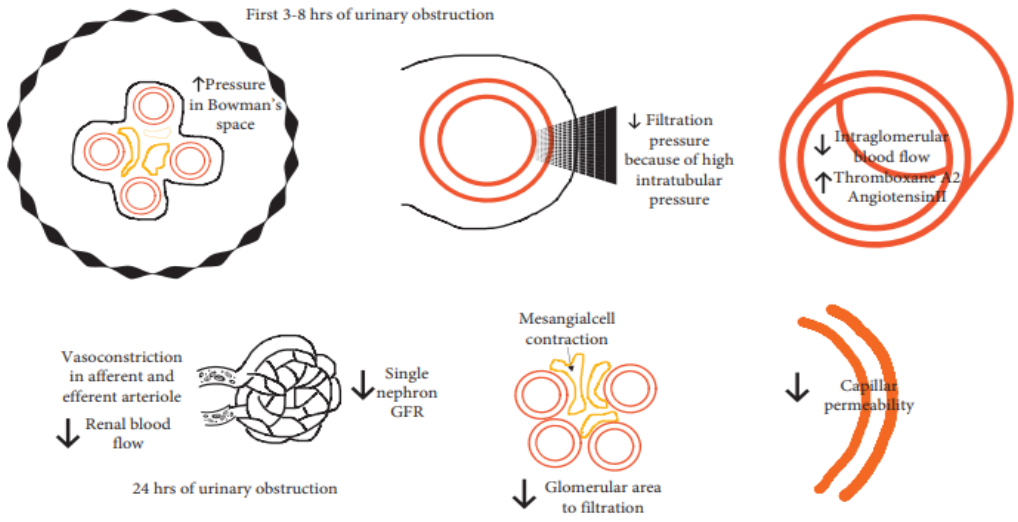
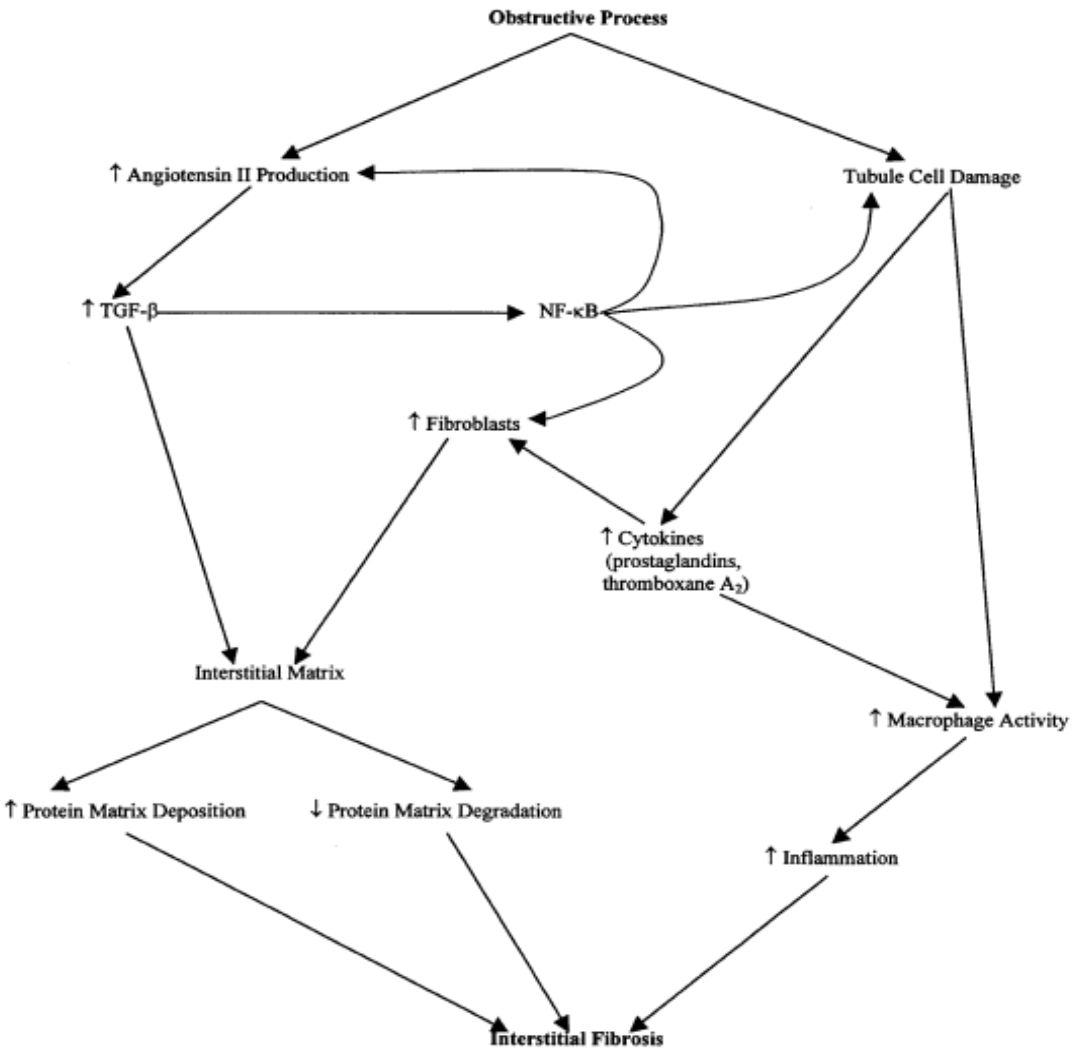


Figure (1-1): Glomerular changes during Obstructive Nephropathy and Acute kidney injury. Glomerular changes during obstructive nephropathy described according to time in hours.[43]

1.7.2 Tubular Alterations

Obstruction affects tubular function by reducing the ability of renal tubules to transport sodium (Na1), potassium (K1), and hydrogen (H1), and reduces the ability to concentrate and dilute the urine[45]. This contributes to post obstructive diuresis[39]. Severe downregulation of aquaporin 2 expression contributes to the impaired urinary concentrating ability[46]. The local increase in prostaglandin E2 synthesis in post obstructed kidney is thought to play a role in aquaporin 2 downregulation[47]. Significant downregulation of apical membrane expression of the distal convoluted tubule Na1 Cl- cotransporter also occurs from obstruction[46]. The defect in H1 and K1 secretion in the distal nephron in obstructive uropathy has been shown to be independent of aldosterone. In the first few days after onset of obstruction, there is interstitial edema and an influx of leukocytes, predominantly macrophages, into the kidneys. If the obstruction is persistent and not relieved, glomerular size decreases, tubular cells lose apical microvilli and basolateral interdigitations and have fewer mitochondria. Nephrons atrophy from reduced renal blood flow and

inflammatory responses[48]. If obstruction is maintained for a longer period of time, hydronephrosis eventually develops and there is tissue loss with tubular atrophy, interstitial fibrosis, and interstitial inflammation(Figure 1-2)[42]. In murine experimental models, interstitial fibrosis has been shown to develop within days in the obstructed kidney with increased renal synthesis of extracellular matrix proteins and transforming growth factor-b[49]. The interstitial fibrosis is also mediated by angiotensin II[50]. Proliferation of interstitial fibroblasts with myofibroblast transformation leads to extracellular matrix deposition. Phenotypic transition of renal tubular cells, endothelial cells, and pericytes has been implicated in this process[51]. The compression of medullary and cortical tissue from the renal calyceal distension results in widespread apoptosis and tubular atrophy as early as 3 days post obstruction. The interstitial response to urinary tract obstruction further intensifies the injury, resulting in renal dysfunction[52]. In patients with malignant urinary obstruction, these changes happen over time and may not manifest as clinically evident renal failure for several days to weeks[39].



Figure(1-2) A simplified version of the mechanisms contributing to interstitial fibrosis resulting from obstructive nephropathy[42]

(TGF-β = trans;forming growth factor beta; NF -κ B = nuclear factor kappa B)

1.8. Fibroblast Growth Factor (FGF)

Fibroblast Growth Factor (FGF) family is comprised of secreted signaling proteins (secreted FGFs) that signal to receptor tyrosine kinases and intracellular non-signaling proteins (intracellular FGFs (iFGFs)) that serve as cofactors for voltage gated sodium channels and other molecules. Additionally, secreted FGFs and iFGFs may have direct functions in the nucleus and functional interactions with other cellular proteins. Members

of both branches of the FGF family are related by core sequence conservation and structure and are found in vertebrates and invertebrates[53].Secreted FGFs are expressed in nearly all tissues and they serve essential roles in the earliest stages of embryonic development, during organogenesis, and in the adult, where they function as homeostatic factors that are important for tissue maintenance, repair, regeneration, and metabolism in general, secreted FGFs function as autocrine or paracrine factors (canonical FGFs; also called paracrine FGFs), however, three members of the secreted FGFs have evolved to function as endocrine factors (endocrine FGFs) with essential roles in the adult where they regulate phosphate, bile acid, carbohydrate and lipid metabolism in addition to the canonical FGF functions that control cell proliferation, differentiation and survival[54].

1.8.1 Families

Fibroblast growth factors (FGF) constitute a large family of proteins with pleiotropic effects on development, organogenesis, and metabolism. Although the name, not all FGF stimulate fibroblast activity. Inclusion in the FGF family is based on structural similarity. The FGF family includes 22 agents that have been classified into seven subclasses based on phylogenetic similarity. The FGF1 subfamily contains two typical growth factors: FGF1 (also referred to as acidic fibroblast growth factor) and FGF2 (basic fibroblast growth factor). Factors from the FGF1, FGF4, FGF7, FGF8, and FGF9 subclasses are characterized by paracrine and/or autocrine activities. The FGF11 subclass consists of so-called nuclear FGFs that operate intracellularly and do not bind to the fibroblast growth factor receptor (FGFR). In turn, the FGF19 subclass includes growth factors circulating with the blood referred to as endocrine FGF (Figure 1-3)[55]. FGFs can have paracrine, endocrine, or intracrine functions[56].

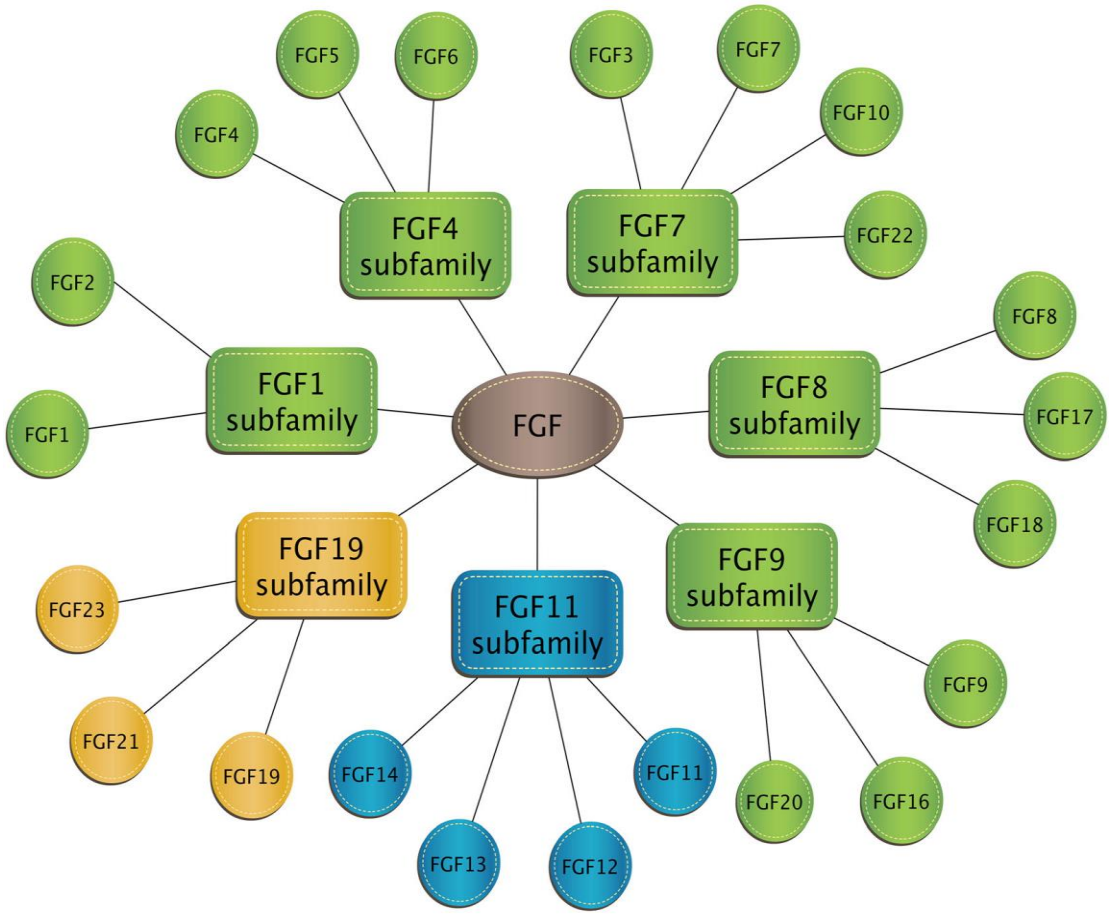


Figure 1-3 Classification of human fibroblast growth factors into seven subclasses. Green color indicates subclasses containing paracrine and/or autocrine factors: blue/ nuclear FGF, yellow/endocrine FGF[55]

1.8.2. Fibroblast Growth Factor 23(FGF23)

1.8.2.1 Overview of Fibroblast Growth Factor 23(FGF23)

Fibroblast Growth Factor 23 (FGF23) plays an important role as a hormone whose principal function is increasing phosphate excretion via kidney[57] .It is secreted by osteocytes in bone in response to active vitamin D, parathyroid hormone (PTH) and elevations in serum phosphorus levels[58] (figure1-4)Vitamin D metabolism[59].FGF23 belongs to the subfamily of FGF19 which are ligands for four FGF receptors (FGFR1-FGFR4). FGFR1 isoform is the main receptor of FGF23 and is expressed in kidney, parathyroid gland, pituitary and choroid

plexus[60].The binding affinity of FGF23 to FGFR1 is enhanced by its co-receptor, klotho[61].Klotho plays an important role in regulating mineral metabolism homeostasis. Specifically, klotho decreases renal phosphate reabsorption by acting as a co-receptor for FGF23 binding to FGFR1[61].Klotho also directly can promote the internalization and degradation of the NaPi2a cotransporter in the renal proximal tubules[62].Klotho also may be a suppressor of vitamin D signaling[63]Klotho protein is a co-receptor which is localized in the tissues targeted by FGF action and stabilize FGF/FGFR binding[64]. There are different forms of klotho protein; transmembrane and soluble. A soluble form of Klotho, produced by cleavage of the membrane bound Klotho ectodomain, is released into the circulation and exerts both paracrine and endocrine functions independent of the FGF23 signaling pathway[65].In the case of FGF23, only α-klotho, which is soluble, The FGFR-binding site is located in the N-terminus while the α-klotho binding site is located in the C-terminus [66].α -Klotho expression is limited to certain tissues, such as the proximal and distal renal tubules, parathyroid, pituitary, heart, and testis. Here, the membrane-bound α -Klotho interacts with several FGFRs, such as FGFR1c, FGFR3c, and FGFR4 therefore it determines tissue specificity for FGF23[64].For a long time, it was believed that klotho protein is essential for FGF23 action. Few years ago, FGFR4, a specific isoform localized on heart and great vessels, has been shown to mediate FGF23 action without the need of α-klotho[56].FGF23 is described as a phosphatonin[67]and its physiological role is to control hyperphosphatemia which can be harmful because it causes ectopic calcification and alters the function of several organs[68].Pathological plasma FGF23 concentrations were firstly described in patients with renal phosphate wasting diseases such as autosomal dominant hypophosphatemic rickets (ADHR), autosomal recessive (ARHR), X-linked hypophosphatemic rickets/osteomalacia (XLH) and tumor-induced

rickets/osteomalacia (TIO)[69].Disturbances in FGF23 function cause several hypophosphatemic and hyperphosphatemic diseases, However, the functions of FGF23 beyond its role as a phosphotropic hormone remain controversial[57].The most experimental and clinical data about interactions of FGF23 with iron metabolism and erythropoiesis, inflammation, insulin resistance, proteinuria, acute kidney injury and left ventricular hypertrophy[70],Figure(1-5)[71].

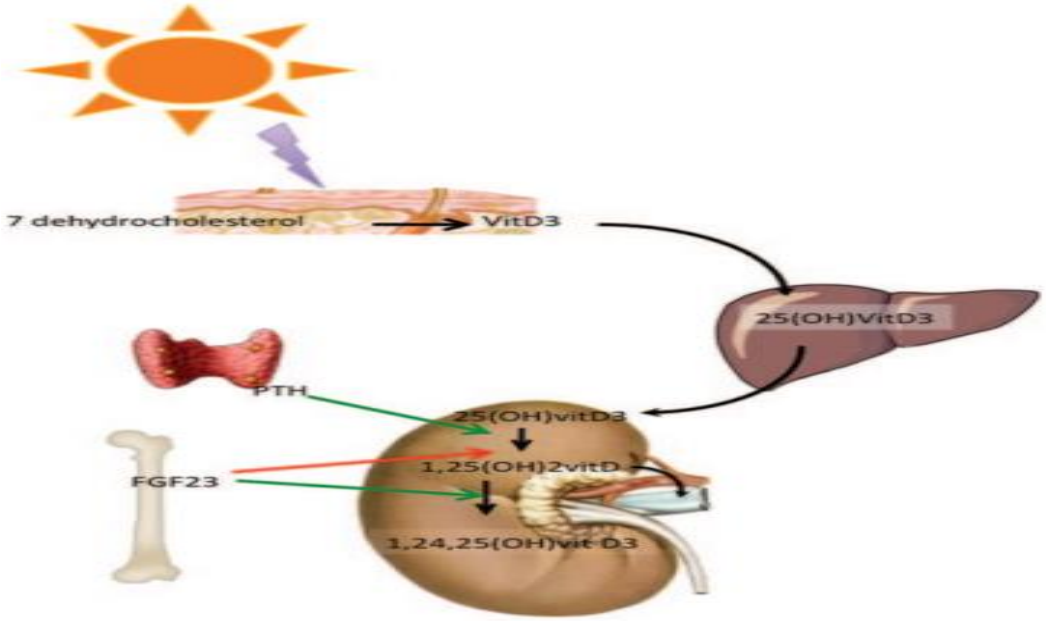


Figure (1-4) Vitamin D metabolism.[59]Vitamin D3 is produced in the skin from a UVB-mediated conversion of 7-dehydrocholesterol. In the liver, vitamin D is converted to 25(OH)vitD3, which is released in blood. In the kidney, 25(OH)vitD3 is converted to 1,25-dihydroxyvitamin D [1,25(OH)2vitD]. Two hormones regulate this step: PTH and FGF23, a hormone that is synthesized by osteocytes and osteoblasts. In the kidney, PTH stimulates whereas FGF23 represses 25(OH)D-1-hydroxylation. In the kidney, FGF23 increases the expression of the 24-hydroxylase an enzyme that inactivates calcitriol.

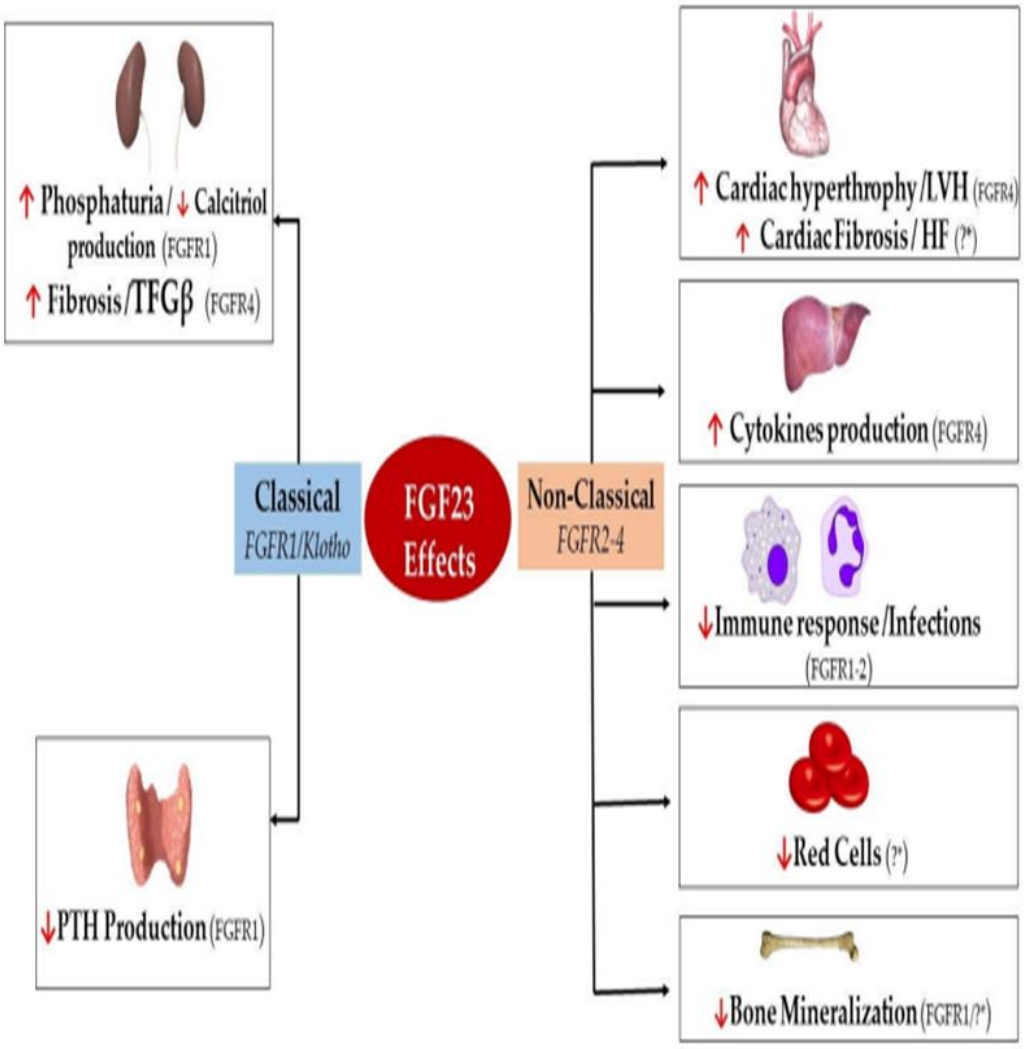


Figure (1-5) Final classical and non-classical clinical effects of FGF23 on different organs. TFGβ, transforming growth factor βeta; PTH, parathyroid hormone; LVF, left ventricle hypertrophy; HF, heart failure. ↓ Decrease; ↑ Increase; ?* Presumably Klotho-independent[71].

1.8.2.2 Fibroblast Growth Factor 23 Function

FGF23 increases urinary phosphate excretion by reducing expression of the sodium-phosphate co-transporters, NaPi2a and NaPi2c, expressed on the apical surface of the renal proximal tubular cells, and it decreases circulating 1,25-dihydroxyvitamin D (1,25D) levels by reducing expression of the 1α-hydroxylase cytochrome P450 (CYP) enzyme, CYP27B1, which is also expressed in renal proximal tubular cells[72]**.**FGF23 may also decrease circulating 1,25D levels by increasing the renal expression of the catabolic 24-hydroxylase enzyme, CYP24A1(Fig1-6)[57].In addition, FGF23 can increase distal sodium reabsorption by increasing the expression of the sodium chloride cotransporter(NCC) and it increases calcium absorption by increasing the expression of transient receptor potential cation channel subfamily V member 5 channels in the distal tubule[73].In states of excess circulating FGF23 levels and normal kidney function, humans and animals develop hyperphosphaturia, hypophosphatemia, and rickets,[74]whereas in states of diminished FGF23 levels or function, humans and animals develop increased circulating levels of phosphate and 1,25D, and ectopic soft-tissue and vascular calcification[75],[76].

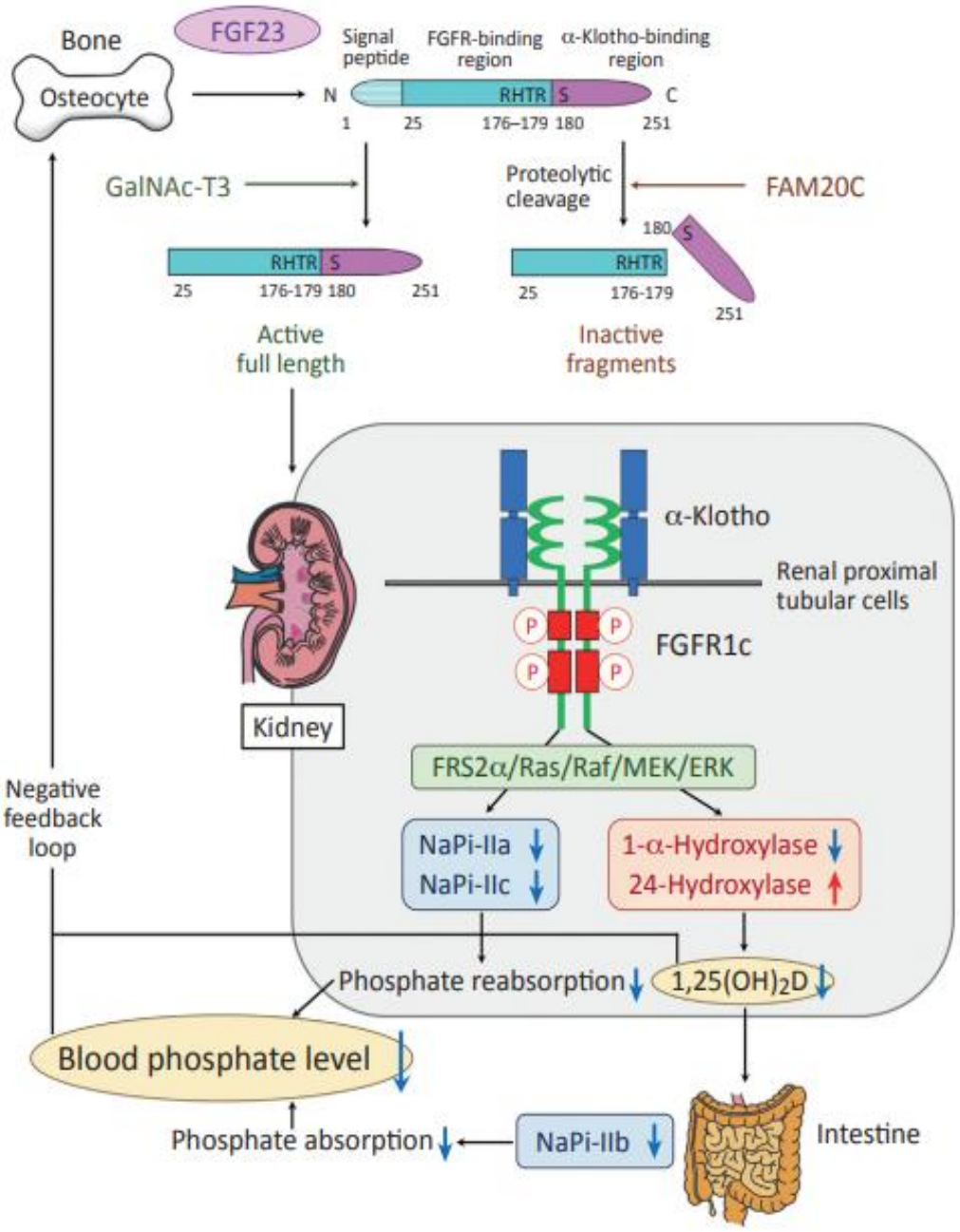


Figure (1-6) The Function of Fibroblast Growth Factor 23 (FGF23) as Phosphotropic Hormone and Its Post Translational Regulation and Signal Transduction Pathway[57].

FAM20C**:** Family with sequence similarity 20, membrane C

1.9. Parathyroid hormone (PTH)

Parathyroid hormone (PTH) is a major endocrine regulator of extracellular calcium and phosphate levels [77]. In the blood, the sensitive process of calcium and phosphate homeostasis is maintained primarily by an appropriately functioning parathyroid gland. The parathyroid gland is comprised of 4 small glands located posteriorly to the thyroid in the middle aspect of the anterior neck. The parathyroid gland secretes parathyroid hormone (PTH), a polypeptide, in response to low calcium levels detected in the blood[78][79]. PTH facilitates the synthesis of active vitamin D, calcitriol (1,25-dihydroxycholecalciferol, or vitamin D3) in the kidneys[80][81]. In conjunction with calcitriol, PTH regulates calcium and phosphate. PTH effects are present in the bones, kidneys, and small intestines. As serum calcium levels drop, the secretion of PTH by the parathyroid gland increases. Increased calcium levels in the serum serve as a negative-feedback loop signaling the parathyroid glands to stop the release of PTH. The mechanism of PTH in the body is intricate, and the clinical ramifications of irregularities are significant. The understanding of PTH is of paramount relevance and importance[82][83].

1. Development

Parathyroid hormone is a polypeptide that is synthesized and cleaved into an active form within the parathyroid gland. The initial structure formed is a pre-pro-PTH, a 115 amino acid polypeptide that is cleaved to form pro-PTH comprised of 90 amino acids. It is then cleaved a second time, again at the amino-terminal portion, to form active parathyroid hormone comprised of 84 amino acids. This is the primary hormone that is stored, secreted, and functions in the body. The process of synthesis, cleavage, and storage is estimated to take less than an hour. Active PTH secretion can occur as quickly as a few seconds when low serum calcium

is detected. The mechanism of secretion is via exocytosis, a process where the hormone is released through a membrane vesicle carried to the cell membrane, releasing the hormone after the vesicle fuses with the outer membrane. The serum half-life of activated PTH is a few minutes and is removed from the serum quickly by the kidney and liver[84].

2. Organ Systems Involved

Parathyroid hormone is directly involved in the bones, kidneys, and small intestine. At the kidneys, parathyroid hormone has 3 functions in increasing serum calcium levels. Most of the physiologic calcium rekabsorption in the nephron takes place in the proximal convoluted tubule and additionally at the ascending loop of Henle. Circulating parathyroid hormone targets the distal convoluted tubule and collecting duct, directly increasing calcium reabsorption. Parathyroid hormone decreases phosphate reabsorption at the proximal convoluted tubule. Phosphate ions in the serum form salts with calcium that are insoluble, resulting in a decreased plasma calcium. The reduction of phosphate ions, therefore, results in more ionized calcium in the blood[85].

1.10 Aim of the study

To Measurement serum level of Fibroblast growth factor- 23(FGF-23), PTH and phosphorus in patients with obstructive renal failure and healthy control subject and to assess the relation of each of them.

2. Materials and Methods

2.1. Materials:

2.1.1. Chemical (kits)

Enzyme-linked immunosorbent assay (ELISA Kits) used in this study were pass by the Quality Control in a manufacturer company in USA were valid for laboratory work. All kits used in the study are listed in table(2-1).

Table (2-1): List of chemical (Kits) used in the study

No	The Item	Name of the Company	Manfacuture Country
1	Fibroblast Growth Factor 23 FGF23	Mybiosource	USA
2	parathyroid hormone	Mybiosource	USA
3	Urea	BIOLABO	France
4	Creatinin	Biolmaghreb	Tunisia
5	Glucose	BIOLABO	France
6	Calicum	Pionte	USA
7	Phosphorus	Mybiosource	USA
8	Potassium	Spectrum	Egypt
9	Sodium	CHEMPAK	India

2.1.2. Instruments and Equipments

The instruments and Equipment's used in this study are listed in the Table (2-2).

Table (2-2): The instruments and equipment that used in the study.

No	Instrument and Equipment	Name of the company	Manufactured country
1	Automated plate washer	BioTek	**USAk**
2	Centrifuge	Labnet	**USA**
3	Cobas C111	Roche	**Germany**
4	Cotton	Local company	**Iraq**
5	Disposable syringe	Medeco	**Germany**
6	ELISA system	BioTek	**USA**
7	elyte 4A electrolyte analyzer serice	Medonic	**Sweden**
8	Eppendorf Tube 5ml	AFCO	**Jordan**
9	Examination Gloves	WRP Asia	**Malaysia**
10	Gel Tube	ARZERGRANDE	**Italian**
11	Incubator	Blutdruckpass	**Germany**
12	microtubes or conical flasks, Bottles	Cusabio	**China**
13	Multichannel micropipette reservoir	Mybiosource	**USA**
14	Plain Tube.	AFCO	**Jordan**
15	Refrigerator	Hitachi	**Japan**
16	Spectrophotometer	Jenway	**Italian**
17	Water Bath	HH-2	**Chain**
18	(10-1000) µL micropipettes with disposable tips.	Mybiosource	**USA**
19	(50-300)µL micropipette with disposable tips	Mybiosource	**USA**

2.2. Subjects

2.2.1. Study Protocol

This study is case-controlled study included one hundred. The samples were collected from Urology department at Hillah Teaching Hospital, with age ranged (25-75) years and BMI with (24-27) Kg/m^2. The period from1st of December, 2020 to 20 of June, 2021. Written informed consent was obtained from all participants and the study was approved by the ethical committees of the institute of Review Board of collage of medicine**.** The data was collected from the study participants directly and filled in prepared questionnaire.

The samples were divided in two groups:

***Group(I)**:Consist of fifty patients (males & female) which have an obstructive renal failure(13 females and 37 males (Females 26% - Males 74%) were included in the study.

***Group(II)**: Consist of fifty apparently healthy subject as control(12 females and 38 males (Females 24% - Males 76%) were included in the study.

***Inclusion criteria**

The criteria of inclusion were patients with obstructive renal failure and no other medical disorders.

*** exclusion criteria**

The criteria of exclusion were patients with

1. History of prolonged use of nephrotoxic drugs.
2. Nephritis.
3. Bone disease.
4. Immune disease (DM).
5. Parathyroid disease.

2.3. Methods

2.3.1. Collection of Samples

The study samples were serum.

2.3.1.1. Blood Samples

About Five ml of blood sample were taken from all participants, the overnight fasting patients and controls in plain tubes at room temperature for 10-15 minutes and allowed to clot. The tube then were centrifuged (3000 rpm) for 15min. The clear serum was pipetted into Two clear dry Eppendorf Tube and stored at (-20ºC) until used for the various investigations. The serum was thawed at (20-25ºC) temperature for two hours then submitted to the centrifuge for five minute at 3600 rpm.

2.3.2. Body Mass Index (BMI):

BMI is diagnostic tool used widely to know the problem related with the obesity within population. BMI was measured by following formula:

BMI = the weight (Kilogram)/the height (meter) ^{2}

1- BMI < (18.5) represents less than normal.

2- BMI = (18.5 - 24.9) represents normal.

3- BMI = (25- 29.9) more than normal or overweight.

4- BMI > 30 obesity.

2.3.3. Assay of Human Fibroblast Growth Factor 23 (FGF23)

A. Principle

This experiment use double-sandwich ELISA technique and the ELISA Kit provided is typical. The pre-coated antibody is human FGF23 monoclonal antibody and the detecting antibody is polyclonal antibody with biotin labeled. Samples and biotin labeling antibody are added into ELISA plate wells and washed out with PBS or TBS. Then Avidin-peroxidase conjugates are added to ELISA wells in order; TMB substrate for coloring after reactant thoroughly washed out by PBS or TBS. TMB turns into blue in peroxidase catalytic and finally turns into yellow under the action of acid. The color depth and the testing factors in samples are positively correlated. The concentration of FGF23 in each serum sample was expressed in ng/ml for comparison of results with those of controls concentration.

Detection range15.6 -1000 pg/ml*

Sensitivity the minimum detectable human FGF23 Up to 5 pg/ml.*

B. Preparation of reagents &Procedure

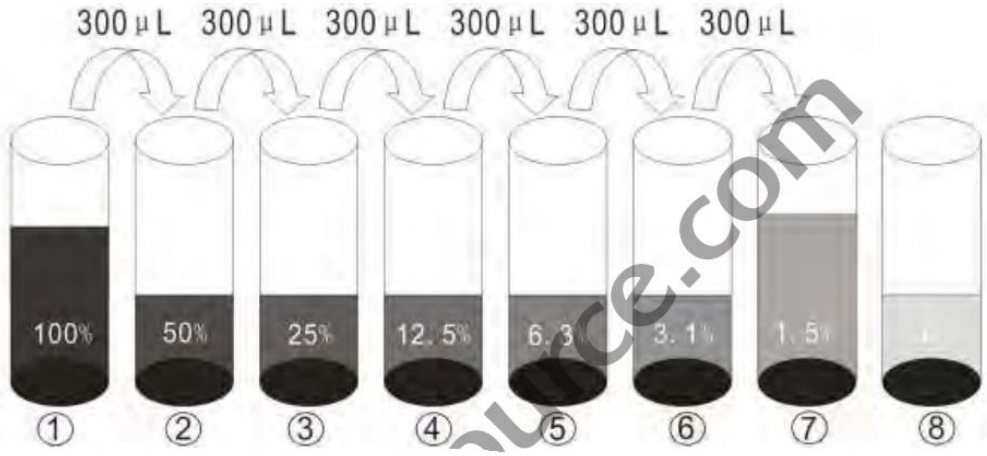
1. The Elisa Kit has been taken out of refrigerator 20 minutes in advance and took test after it had balanced to room temperature.

2. The concentrated washing solution has been diluted with double distilled water (1:25).Put the unused back.

3. Human FGF23 standard sample: diluent 1.0ml has been added into human FGF23 lyophilized standard sample and kept it still for 30 min. After the sample completely dissolved, mixed slightly and marked label on the tube①, then took dilution as needed

4. Legend of standard sample dilution method: 7 clean tubes has been taken and labeled them with②,③,④,⑤,⑥,⑦,⑧ respectively. Added 300μl standard sample diluent into each tube.

the Pipette has been used out 300μl diluent from tube ① to tube ② and mixed well. Further Pipette has been used out 300μl diluent from tube ② to tube ③, and mixed well. Repeated steps above up to tube ⑦. Standard sample dilution in tube ⑧ is negative control.



5. Biotinylated human FGF23 antibody liquid: Referring to need amount, it has been employed antibody diluent to dilute the concentrated biotinylated antibody (1:100) to form biotinylated antibody liquid.

6. Enzyme-conjugate liquid: Referring to need amount, the concentrated enzyme-conjugate has been diluted by enzyme-conjugate diluent (1:100) to form enzyme-conjugate liquid.

7. Color Reagent liquid: Color Reagent liquid has been prepared 30 min in advance with Color Reagent A and Color Reagent B by the proportion of 9:1.

***. Washing method**

1. Automatic plate-washing machine: The required amount of lotion is 350μl and the injection and extraction interval should be 20－30secs.

2. Manual plate-washing machine: 350μl lotion had been added to each well and kept it still for 30secs. Individual wells has been shacked as dry as could and cleaned them with absorbent paper

*** .Steps**

1. the needed strips had been taken out from zip lock bag which balanced to room temperature. The unused strips and desiccant had been put back into the sealed aluminum foil bag at 2-8℃ for storage.

2. the blank wells had been set aside (when dual-wavelength reading plate is used, the blank wells has been ignored) .

3. samples or different concentration of human FGF23 standard samples have been added to corresponding wells (100μl for each well), 0pg/ml well

should be filled with standard \ diluent. it has been sealed the reaction wells with adhesive tapes, hatching in incubator at 37℃ for 90 min.

4. Biotinylated human FGF23 antibody liquid had been prepared 30min in advance.

5. The Elisa plate had been watched 2 times

6. The biotinylated human FGF23 antibody liquid had been added to each well (100μl for each). Reaction wells had been sealed with adhesive tapes, hatching in incubator at 37℃ for 60 min.

7. enzyme-conjugate liquid had been prepared 30min in advance.

8. The Elisa plate had been watched 3 times

9. enzyme-conjugate liquid had been added to each well except blank wells (100μl for each). the reaction wells had been Sealed with adhesive tapes, hatching in incubator at 37℃ for 30 min.

10. The Elisa plate had been Washed 5 times.

11. 100μl Color had been Added Reagent liquid to individual well (also into blank well), hatching in dark incubator at 37℃. When color for high concentration of standard curve become darker and color gradient appears, the hatching can be stopped. The chromogenic reaction should be controlled within 30 min.

12. 100μl Color Reagent C had been Added to individual well (also into blank well). Mixed well. Readed OD（450nm）within 10 min.

C. Result determination

1. OD value of each sample and specimen should minus that of blank well (if not, the standard curve of zero well should intersect at Y axis)

2. Standard curve was Drawn manually. The concentration value of samples took as abscissa and OD readings as vertical coordinate. Used smooth line to connect each coordinate point of standard sample. The concentration of samples can be found by checking sample OD reading. It is recommended to employ the professional curve software (e.g. curve expert 1.3) to analyze and compute the result.

3. when the sample OD is higher than the upper limit of standard curve, the sample re-diluted and the experiment rerun. Multiplied the result by dilution factor when calculating the unknown.

***. Reference curve**

* Reference curve see Figure (2-1).

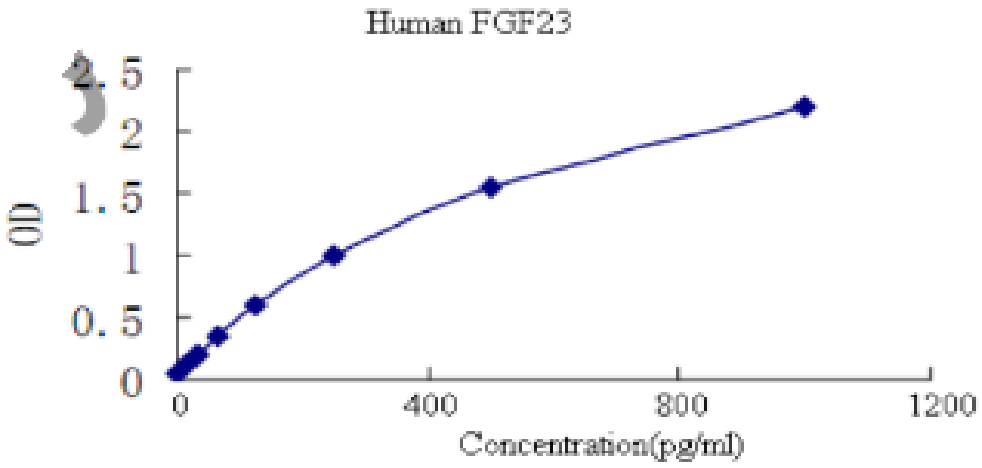


Figure (2-1): Standard curve of FGF23 concentration.

2.3.4 Determination of Human PTH (Parathyroid Hormone)

A. Principle

This ELISA kit uses the Sandwich-ELISA principle. The micro ELISA plate provided in this kit has been pre-coated with an antibody specific to HumanI-PTH. Samples (or Standards) are added to the micro ELISA plate wells and combined with the specific antibody. Then a biotinylated detection antibody specific for HumanI-PTH and Avidin-Horseradish Peroxidase (HRP) conjugate are added successively to each micro plate well and incubated. Free components are washed away. The substrate solution is added to each well. Only those wells that contain HumanI-PTH, biotinylated detection antibody and Avidin-HRP conjugate will appear blue in color. The enzyme-substrate reaction is terminated by the addition of stop solution and the color turns yellow. The optical density (OD) is measured spectrophotometrically at a wavelength of 450 ± 2 nm. The OD value is proportional to the concentration of HumanI-PTH. it can calculate the concentration of HumanI-PTH in the samples by comparing the OD of the samples to the standard curve.

B. Preparation of reagents

1. Brought all reagents to room temperature (18-25℃) before use. when the kit will was not used up in one assay, It has only taken out the necessary strips and reagents for present experiment, and stored the remaining strips and reagents at required condition.

2. **Wash Buffer:** Dilute 30mL of Concentrated Wash Buffer with 720mL of deionized or distilled water to prepare 750mL of Wash Buffer. 3. **Standard working solution:** after Centrifuged the standard at 10,000×g for 1 min. Added 1.0 mL of Reference Standard &Sample Diluent, let it

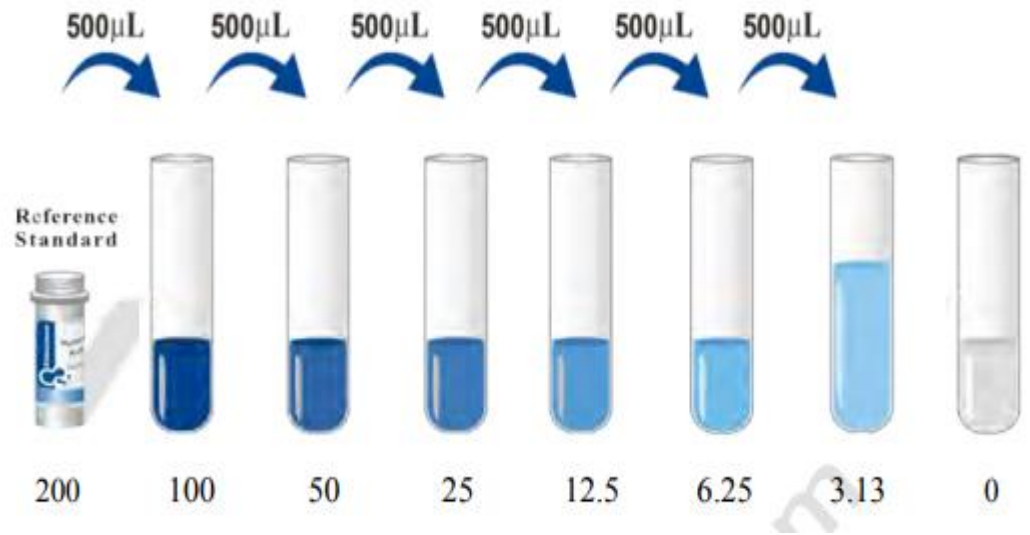
stand for 10 min and inverted it gently several times. After it dissolves fully, it has been mixed it thoroughly with a pipette.

This reconstitution produced working solution of 200pg/mL, let it stand for 1-2 min and then it has been mixed thoroughly with a vortex meter of low speed. Bubbles generated during vortex could be removed by centrifuging at a relatively low speed). Then make serial dilutions as needed. The recommended dilution gradient is as follows: 200、100、50、25、12.5、6.25、3.13、0 pg/mL.

Dilution method: 7 EP tubes has been Taken, 500μL added of Reference Standard & Sample Diluent to each tube. 500μL has been pipetted of the 200pg/mL working solution to the first tube and mixed up to produce a 100pg/mL working solution. Then pipetted 500μL of the solution from the former tube into the latter one according to this step.

4. **Biotinylated Detection Ab working solution:** by calculated the required amount before the experiment (100μL/well). In preparation, slightly more than calculated was prepared. it has been Centrifuged the Concentrated Biotinylated Detection Ab at 800×g for 1 min, then dilute the 100× Concentrated Biotinylated Detection Ab to 1× working solution with Biotinylated Detection Ab Diluent(Concentrated Biotinylated Detection Ab: Biotinylated Detection Ab Diluent= 1: 99).

5. **HRP Conjugate working solution:** it has been calculated the required amount before the experiment (100μL/well). In preparation, slightly more than calculated had been prepared. Centrifuge the Concentrated HRP Conjugate at 800×g for 1 min, then diluted the 100× Concentrated HRP Conjugate to 1× working solution with HRP Conjugate Diluent(Concentrated HRP Conjugate: HRP Conjugate Diluent= 1: 99).



C. Assay procedure

1. Determine wells for diluted standard, blank and sample. 100µL had been added each dilution of standard, blank and sample into the appropriate wells. the plate had been Covered with the sealer provided in the kit. it had been Incubated for 90 min at 37℃.

2. the liquid had been decanted from each well, do not wash. Immediately add 100 µL of Biotinylated Detection Ab working solution to each well. the plate had been Covered with anew sealer. Incubated for 1 hour at 37°C.

3. the solution had been Decanted from each well added 350µL of **wash buffer** to each well. Soaked for 1-2 min and aspirated or decanted the solution from each well and patted it dry against clean absorbent paper. this wash had been Repeated step 3 times.

4. 100µL of **HRP Conjugate working solution** had been Added to each well. Covered the plate with anew sealer. Incubated for 30 min at 37°C.
5. Decanted the solution from each well, repeated the wash process for 5 times as conducted in step 3.

6. 90μL of **Substrate Reagent** had been Added to each well. Covered the plate with anew sealer. Incubated for about 15 min at 37°C. Protected the plate from light.

7. 50μL of **Stop Solution** had been Added to each well.

8. the optical density (OD value) had been Determined of each well at once with a micro-plate reader set to 450 nm.

***. Calculation of results**

Average the duplicate readings for each standard and samples, then subtract the average zero standard optical density. Plot a four-parameter logistic curve on log-log graph paper, with standard concentration on the x-axis and OD values on the y-axis. If the OD of the sample surpasses the upper limit of the standard curve, it should re-test it with an appropriate dilution. The actual concentration is the calculated concentration multiplied by the dilution factor.

***. Typical data**

As the OD values of the standard curve may vary according to the conditions of the actual assay performance (e.g. operator, pipetting technique, washing technique or temperature effects), the operator should establish a standard curve for each test. Typical standard curve and data is provided below for reference only.

Concentration(pg/mL)	200	100	50	25	12.5	6.25	3.13	0
OD	2.478	1.629	0.954	0.475	0.266	0.171	0.122	0.062
Corrected OD	2.416	1.567	0.892	0.413	0.204	0.109	0.06	-

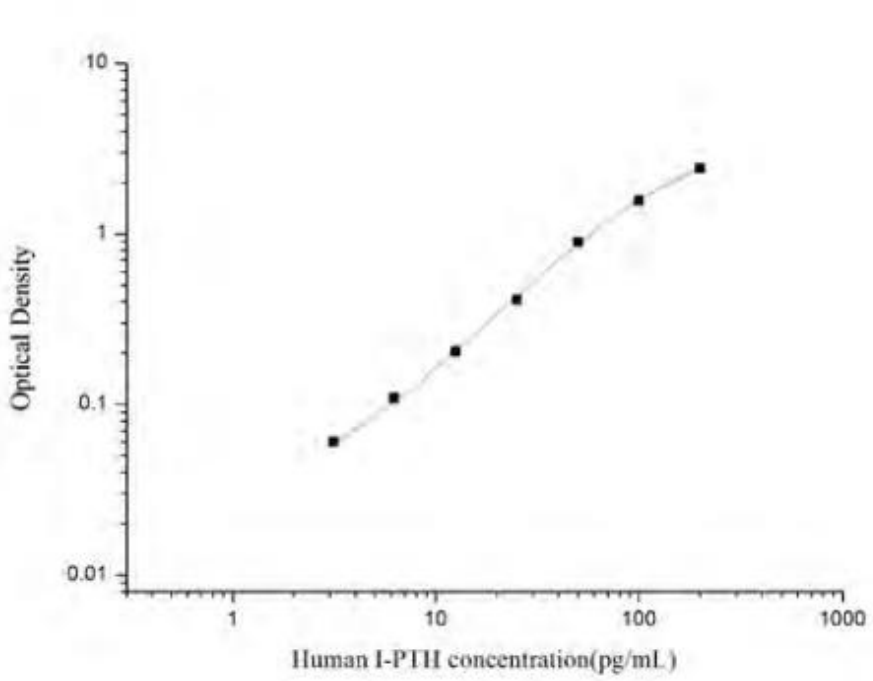


Figure (2-2): Standard curve of Human PTH concentration.

2.3.5 Determination of Urea

A. Principle[86]

Enzymatic and colorimetric method based on the specific action of urease which hydrolyses urea in ammonium ions and carbon dioxide Ammonium ions then form with chloride and salicylate a blue-green Complex.

B. Assay procedure:

	Blank	**Standard**	**Sample**
Working Reagent (R1+R2)	1 ml	1ml	1ml
Standard	-	5µl	-
Sample	-	-	5 µl
Demineralized water	5µl		
Mix and wait for 4 minutes at room temperature or 2 minutes at 37°C			
Base (vial R3) diluted ¼	1 ml	1ml	1ml
Mix. Let stands for 8 minutes at room temperature or 5 minutes at 37°C. Read absorbance at 600 nm against blank.			

C. Calculations

Urea (mg/dl) $= \frac{A\ Sample}{A\ Standard} * 40$

*The reference values of serum urea concentration according to this procedure were 13 - 43 mg/dL (2.1 - 7.1 mmol/L) .

2.3.6 Determination of Creatinine

A. Principle[87]

Creatinine in a basic picrate solution forms a colored complex. The Δ extinction at predetermined times during conversion is proportional to the concentration of creatinine in the sample.

B. Assay procedure

	Blank	**Standard**	**Sample**
Standard		100 μl	
Sample			100µl
Working Reagent (R_1+R_2)	1 ml	1 ml	1 ml

Mix well. After 30 seconds, record absorbance A_1 at 492 nm. Exactly 1 min after the first reading, record absorbance A_2.

C. Calculations

Creatinine (mg/dl) $=\frac{(A2-A1)\ Assay}{(A2-A1)\ Standard} * 2$

*The reference values of serum Creatinine concentration according to this Procedure were 0. 7-1.4 mg/dl (61.8– 132.6)mmol/L

2.3.7 Estimation of GFR [88]

The eGFR is a useful and precise indicator of renal function and is calculated using serum creatinine as well as age, race and gender variables. The most frequently used assessment equation is the abbreviated Renal Disease Modified Diet (MDRD) equation .

eGFR (mL/min/1.73 m^2) = 175× $(Scr)^{-1.154}$ × $(Age)^{-0.203}$ × (0.742 if female) 1.212 if African American).

Age is one factor affecting kidney function. Normal GFR in individuals aged 30 years or younger is about 125 mL/min/1.73 m2. After the age of 30 years, GFR decreases by 1 mL/min/1.73 m2 per year[89].

2.3.8 Determination of serum Glucose

A. Principle[90]

Trinder Method. Glucose is oxidised by Glucose oxidase (GOD) to gluconic acid and hydrogen peroxide which in conjunction with Peroxidase (POD) , reacts with chloro phenol and PAP to form a red quinoneimine. The absorbance of the colored complexes, proportional to the concentration of glucose in the specimen.

B. Assay procedure

	Blank	Standard	Sample
Standard		10 μl	
Sample			10 μl
Working Reagent	1ml	1ml	1ml

The content of tubes were mixed well and incubated for 10 minutes at 37°C. The absorbance (A) of standard and sample was read against the blank at 500 nm.

C. Calculations

Glucose (mg/dl) = $\frac{A\ Sample}{A\ Standard} * 100$

*The reference values of serum Glucose concentration according to this procedure were 82-115 mg/dl (4.6-6.4 mmoI/L).

2.3.9 Determination of serum Sodium

A. Principle[91]

The present method is based on reaction of sodium with a selective chromogen producing a chromophore whose absorbance varies directly as the concentration of sodium in the test specimen**.**

B. Assay procedure:

	Blank	Standard	Sample
Standard		$10\mu l$	
Sample			$10\mu l$
Working Regent	1 ml	1 ml	1 ml

The content of tubes were mixed well and incubated for 5 minutes at 37°C. The absorbance (A) of standard and sample was read against the blank at 630 nm.

C. Calculations

Sodium (mmoI/L) =

$$\frac{Abs\ of Blank - Abs\ of\ Sample}{Abs\ of Blank - Abs\ of\ Standard} \times 150$$

*The reference values of serum Sodium concentration according to this procedure were 135– 155 mmoI/L.

2.3.10 Determination of serum Potassium

A. Principle[92]

At an alkaline pH Potassium, ions and Turbid metric Tetraphenylborate form a turbid emulsion, the increase of which can be measured quantitatively in a photometer at 578 nm.

B. Assay procedure

	Blank	**Standard**	**Sample**
Standard		20 μl	
Sample			20 μl
Working Reagent	1 ml	1 ml	1 ml

The content of tubes were mixed well and incubated for 3 minutes at 37°C. The absorbance (A) of standard and sample was read against the blank at 578 nm.

C. Calculation

$$\text{Potassium (mmoI/L)} = \frac{A\ Sample}{A\ Standard} * 5$$

*The reference values of serum Potassium concentration according to this procedure were 3.6-5.5 mmoI/L.

2.3.11 Determination of serum Calcium

A-Principle

Alkaline Medium

Calcium + O-Cresolphthalein Complexone ----------→

Calcium-Cresolphthalein Complexone Complex (purple color)

Calcium reacts with CPC in an alkaline medium to form a purple-color that absorbs at 570nm .

B. Assay procedure:

	Blank	**Standard**	**Sample**
Standard		$20\mu l$	
Sample			$20\mu l$
Working Reagent (R_1+R_2)	1ml	1ml	1ml

Combine equal volumes of color and buffer reagent, mix and let stand for twenty minutes at room temperature before use and incubated for 1minute at room temperature. The absorbance (A) of standard and sample was read against the blank at 570 nm.

C .Calculations

Calcium (mg/ dl) = $\frac{A\ Sample}{A\ Standard} * 10$

*The reference values of serum Calcium concentration according to this procedure were 8.5-10.5mg/dl.

2.3.12 Determination of serum Phosphorous

A-Principle

Phosphorus Microplate Assay Kit provides a sensitive colorimetric means to directly measure phosphorus concentration in various samples. Phosphorus concentration is based on the reaction of phosphorus with ammonium molybdate to form a blue colored product. The color intensity at 620 nm is directly proportional to phosphorus concentration in the sample.

B. Sample Preparation

For serum and other biological fluids sample Add 100 µl sample and 900 µl Assay buffer into the microcentrifuge tube, mix, centrifuged at 8,000g 25 °C for 10 minutes, take the supernatant into a new centrifuge tube for detection.

C. Assay Procedure

Add following reagents into the microplate:

Reagent	Blank	Standard	Sample
Reaction Buffer	50 µl	50 µl	50 µl
Dye Reagent	50 µl	50 µl	50 µl
Distilled water	100 µl	-	-
Standard	-	100 µl	-
Sample	-	-	
Mix, wait for 10 minutes, measured at 620 nm and record the absorbance			

D .Calculations

According to the serum sample Phosphorus(mmol/L) = CStandard × (ODSample - ODBlank) / (ODStandard - ODBlank) × 10 = 4 × (OD Sample - ODBlank) / (ODStandard - ODBlank)

CStandard: the concentration of Standard, 0.4 mmol/L.

2.4 Statistical Analysis

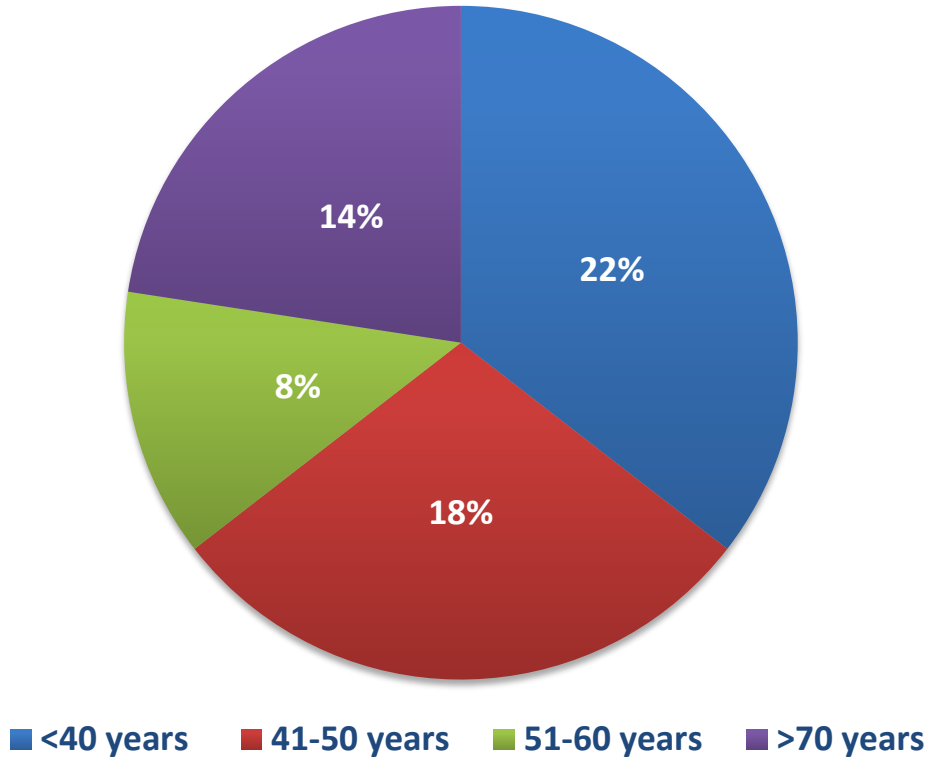
This study was a case -control research design. Statistical Package of Social Science (SPSS) version 26.0 (IBM SPSS Inc., Chicago, IL) and Microsoft excel software v. 2016 were used for the data analysis and graphs plotting. No missing values were observed for any variable. Normality and outliers presented by Skewness and Kurtosis values for each variable and group. Means ± Standard deviation or Median, range, and standard error of mean were used to describe parametric and non-parametric variables respectively. Bartlett's Test was used to examine for population variances homogeneity. ANOVA used to test parameters variances among the groups of the study. Whenever there were significant differences among the groups, a pairwise Tukey Analysis was used to test the differences in the means for each significantly different combination of groups. Being there any problems with normality or inequality of population variances, Kruskal-Wallis test was used instead of ANOVA. Scatterplots was used for visual demonstrating of relationships between independent variables. Pearson correlation conducted for measurement of correlation coefficients and their significance, Cohen's standard was used to evaluate the strength of the relationships, (Cohen, 1988). ROC survival test was used for evaluating the ability of study markers to discriminate disease from non-disease subjects. alpha level for statistical significance was set to $p \leq 0.05$.

3. Results and Discussion

3.1 General characteristics of studied groups

3.1.1. Age.

In this study, 100 participants were taken 50 patients and other healthy people and their ages ranged between (25 –75) years. Comparison between study groups regarding the age was performed using t-test. There was not statistically significant difference p value (0.340) High incidence of obstructive renal failure in the aged group (61-70 years) with (18) patients, representing (36.0%) of the total patients with obstructive renal failure. The aged group (<40 years) was in the second sequence with (11) patients representing (22.0%). And, The aged group (41-50 years) was in the third sequence with (9) patients representing (18.0%), The aged group (> 70 years) was in the fourth sequence with (8) patients representing (16.0%).And finally The aged group (51-60 years) was in the second sequence with (4) patients representing (8.0%) the dispersion of age the rate malady is appeared in table (3.1)Figure (3.1),The mean of obstructive renal failure (57.05±15.21) compared with healthy control (53.77±14.18) P value (0.304), Figure (3.2).



(Figure 3-1) Age of patients with obstractive renal failure

Acording to the results of this study were found more cases of obstructive renal failure in old age that agree with previous study The clinical manifestation of OU varies in the elderly, ranging from an asymptomatic clinical condition to a severe acute kidney injury needing dialysis [4]. In the elderly (>64 years), the incidence of hydronephrosis is higher than in younger age (5.1% vs. 3.1%) especially in elderly males (6.2% in men and 2.9% in women) [3].

3.1.2 Gender

In this study, 50 patients with obstractive renal faliure , there were 34 males and 16 females. The percentage of male 68.0% and female 32.0% as shown in figure (3.2):

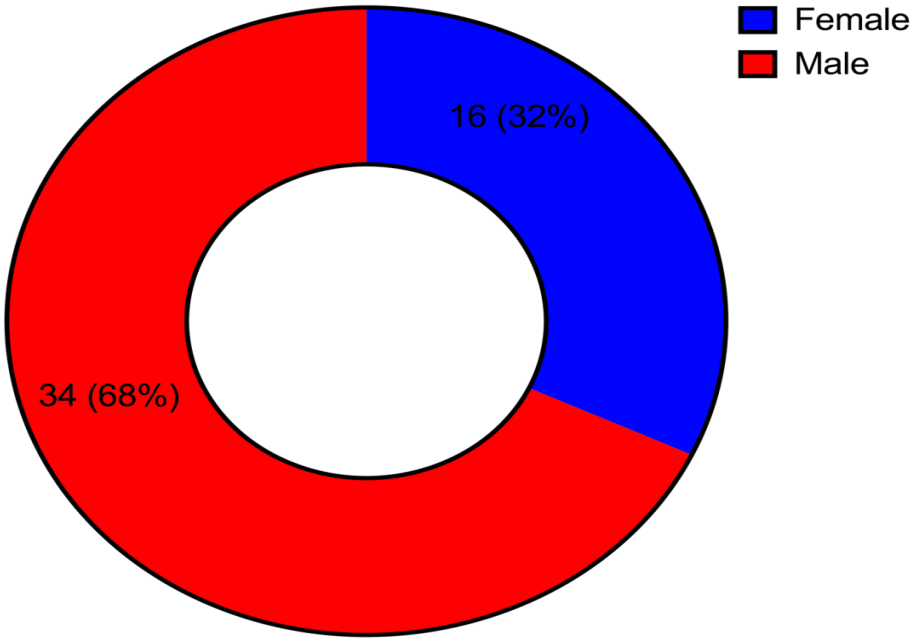


Figure (3.2): Gender of study groups**.**

In addition to the effect of age on the obstructive renal failure, as it was found that there were differences between the ratios of males and females, as estimated by him in our study in Fig (3. 2). We found the male was more than female because men are more prone to prostate diseases and stones.

Regularly, epidemiological studies of disease prevalence show that obstructive renal failure may occur more often in men, To note, 57.4% of patients with ESRD due to UN are older than 64 years and 73.8% are males[5].

3.1.3 BMI.

BMI they are not statistically significant P-value 0.171, BMI for case group in obstructive renal failure state (25.12±4.96) and control group (26.28±2.44) these statistics were summarized in figure (3-3)

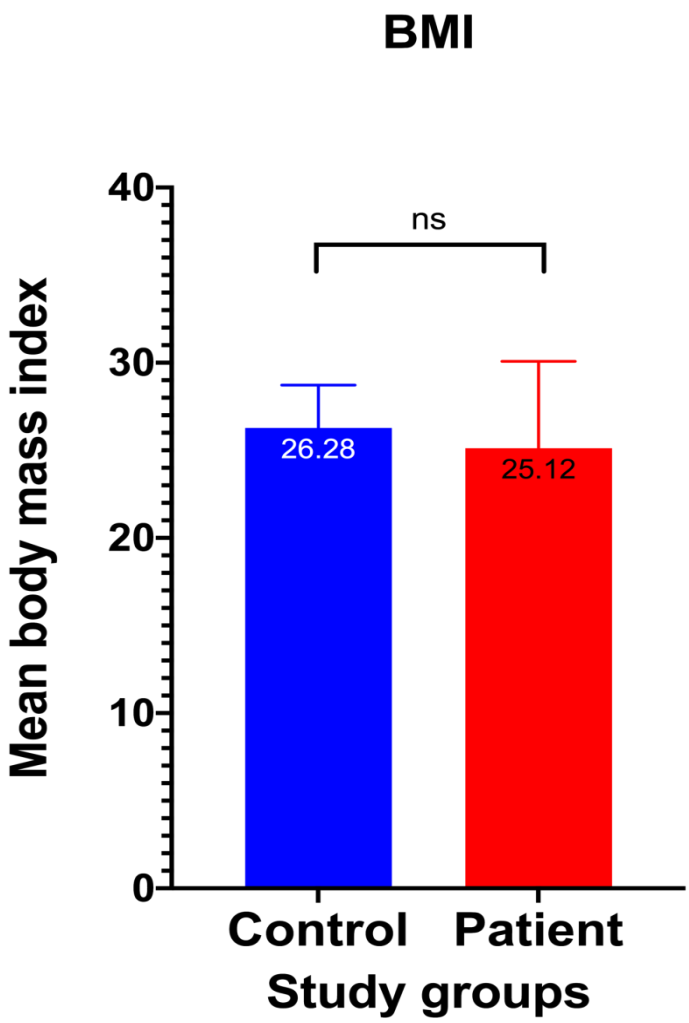


Figure (3-3) BMI of the study groups

According our study the BMI not statistically significant.

3.2 Descriptive Parameters:

The mean ± SD and the p-value of descriptive parameters in all studied groups with Duration (week) were 2.85±5.11 are presented in table (3.1).

The table (3-1) showed that the blood urea in serum levels was significantly higher (P<0.001) in patients (92.9±51.1) Compared with control group (26.62±4.6),

Table (3-1) showed that there was in serum levels significant (P<0.001) increase in creatinine levels of patients (3.55±2.23) compared; with control groups (0.8±0.14).

Table (3-1) showed that there was in levels a highly significant decrease (P<0.001) in GFR of patients (21.53±11.58) compared with control groups (97.86±17.98).as shown in the figure below (3.4).

Table (3-1) Comparison of serum urea, creatinine, GFR between patients & control group

	Study groups		P value
	Control mean ± SD	Patient mean ± SD	
Blood Urea (mg/dL)	26.62±4.6	92.9±51.1	<0.001**
Serum Creatinine (mg/dL)	0.8±0.14	3.55±2.23	<0.001**
Glomerular Filtration Rate (mL/min)	97.86±17.98	21.53±11.58	<0.001**
Fasting blood sugar (mg/dl)	110.38±17.54	115.48±19.17	0.201 NS
Potassium (mmol/L)	4.24±0.36	4.47±0.59	0.028*
Sodium (mmol/L)	140.63±4.91	134.16±4.48	<0.001**
Chloride (mmol/L)	105.47±4.4	106.13±18.41	0.819NS

NS: none statistical significance (p>0.05)
*: significant difference (p≤0.05)
**: High statistically significant difference (p≤0.001)

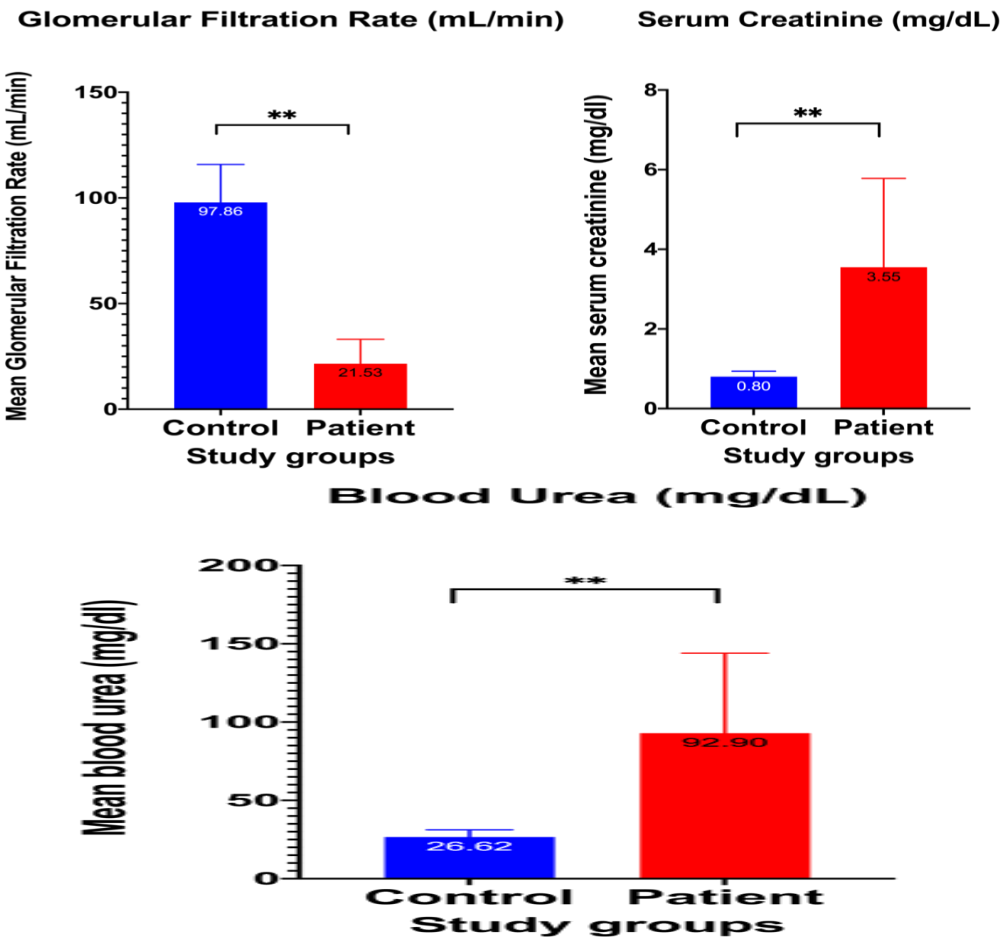


Figure (3-4): Comparison of serum urea, creatinine, GFR between patient & control group

The progression of kidney damage is marked by the rise in two important chemical substances in the blood, creatinine and urea whose evaluation in serum helps to assess GFR followed by renal function. However, creatinine nor urea is directly toxic and they are only a measure of kidney function[93]**.**

Results obtained in this study revealed that urea and creatinine levels in the serum of patients was significantly higher than healthy control groups. This finding is in agreement with that of Alvencar S et al, found A 78 year-old man with urinary retention upon evaluation by the medical staff a foley catheter was placed two liters of urine were drained within one

hour of catheter placement, Upon evaluation in hospital, the patient was confused and in respiratory distress. Initial laboratory studies were notable high level for blood urea and a serum creatinine [94].

The results demonstrated from this study illustrated that the level of GFR in patients lower than controls, P value (<0.001) agreement with Tolani et al. demonstrated decrease in GFR in patients with obstructive renal disease [95].

The table (3-1) showed that the fasting blood sugar was not significantly (p 0.201) patient (115.48±19.17) compared with control group (110.38±17.54). as summarized in (figure 3-5).

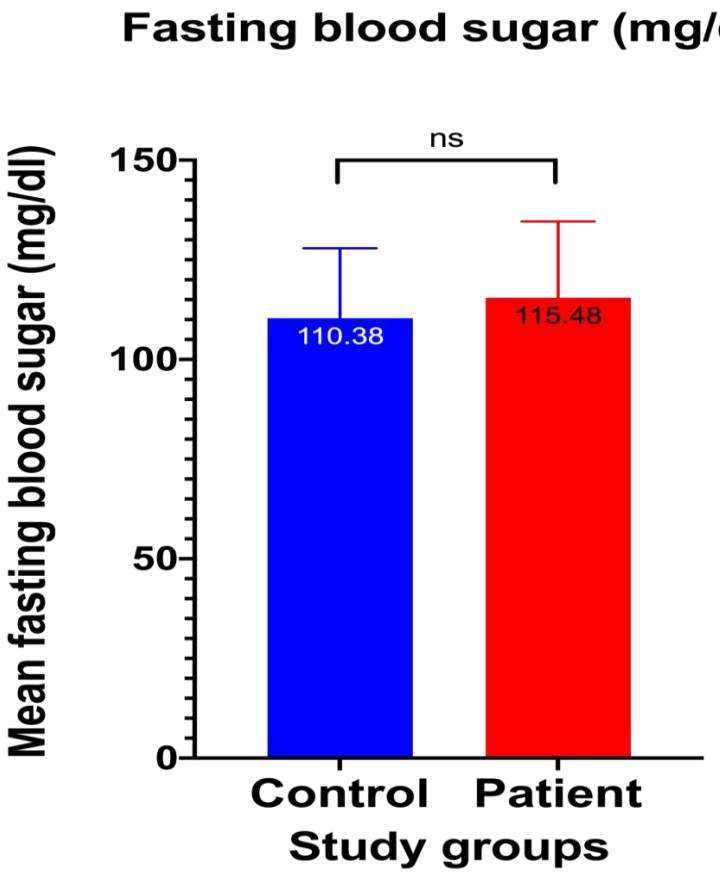


Figure (3-5): Comparison of FBS between patient & control groups.

Electrolyte distribution among the studied groups is presented in table (3-1). There was in serum levels a significant increase (P 0.028) of potassium (K+) concentration in patients (4.47±0.59) compared with control groups (4.24±0.36).

The serum levels of sodium (Na+) in patients were low significantly (134.16±4.48) (P<0.001) compared with control group (140.63±4.91). Summarized in table (3-1).

The serum levels of chloride (CL) in patients was not significantly (106.13±18.41) (P 0.819) compared with control group (105.47±4.4). Summarized in table (3-1). Summarized electrolyte in figures (3-6).

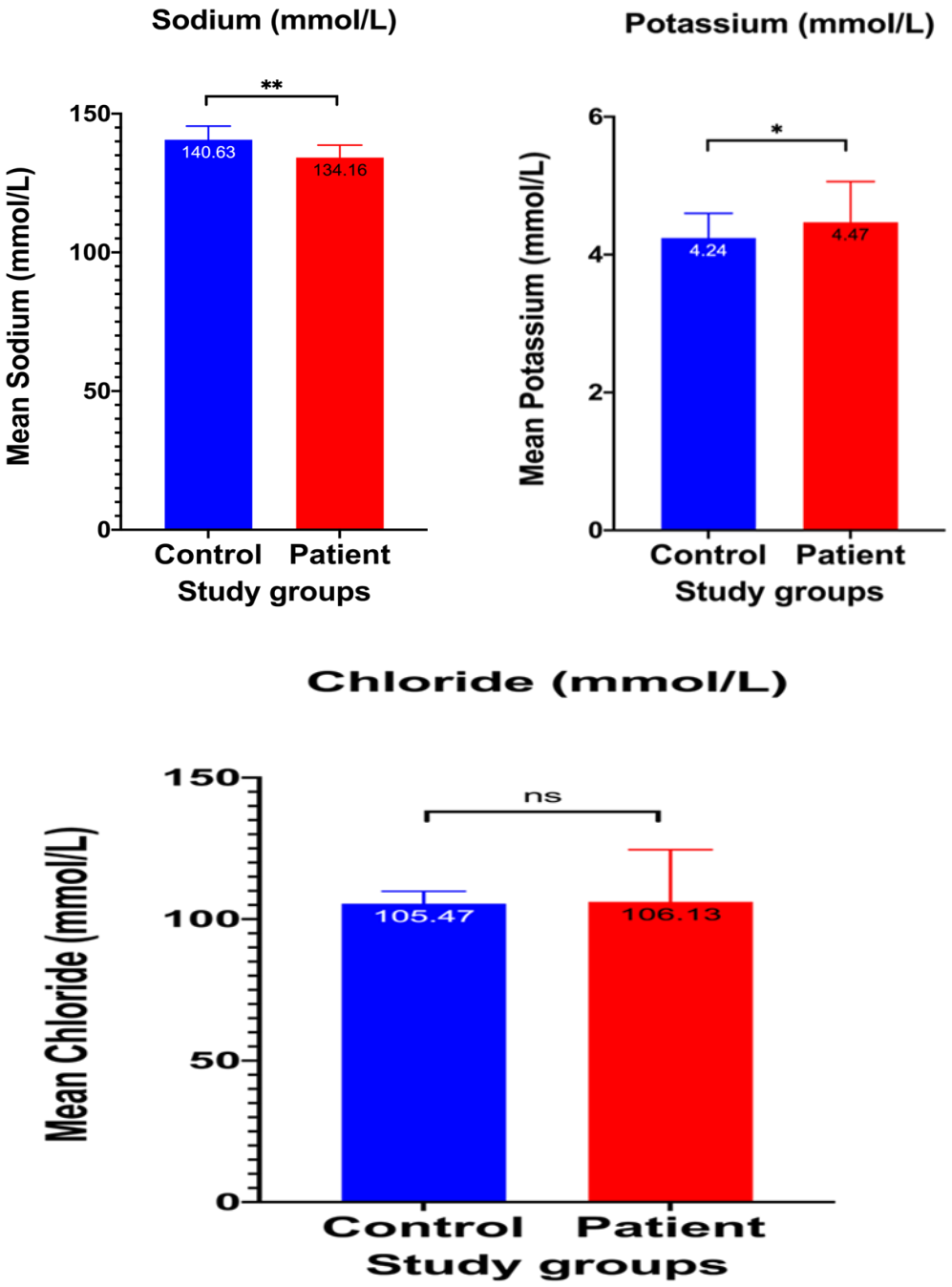


Figure (3-6): Comparison of electrolyte between patient & control groups

In current study found the level of K was show high significant difference between groups in patients and control, Sodium were found to be lower in patient group with highly significant when compared with control group. This study agrees with (Xiao-jieet al) & (Rosner et al).

(Rosner et al) suggesting that the abnormal of serum electrolyte Na+ and K+ may be related to the occurrence and severity of obstructive renal damage [96][97].

The levels of Fibroblast growth factor 23 (FGF23) in patients (501.3±230.89) was increasing significantly (P<0.001) compared with control groups (119.63±29.8) showed as (table 3-2) &figure (3-7)

The levels of Parathyroid hormone in patients (890.19±499.48) was increasing significantly (p<0.001) compared with control groups (261.96±86.84) summarized in (table 3-2) &figure (3-7).

Table (3-2) Comparison of FGF23, PTH, Hb, Po4, Ca levels between control group and patient group

	Study groups		P value
	Control mean ± SD	Patient mean ± SD	
Fibroblast growth factor 23 (pg/ml)	119.63±29.8	501.3±230.89	<0.001**
Parathyroid hormone (pg/ml)	261.96±86.84	890.19±499.48	<0.001**
Hemoglobin (mg/dL)	13.48±1.28	10.25±2.44	<0.001**
Phosphorus (mmol/L)	0.68±0.39	2.01±0.76	<0.001**
Calcium (mg/ dl)	9.03±0.54	8.21±1.04	<0.001**

**: High statistically significant difference (p≤0.001)

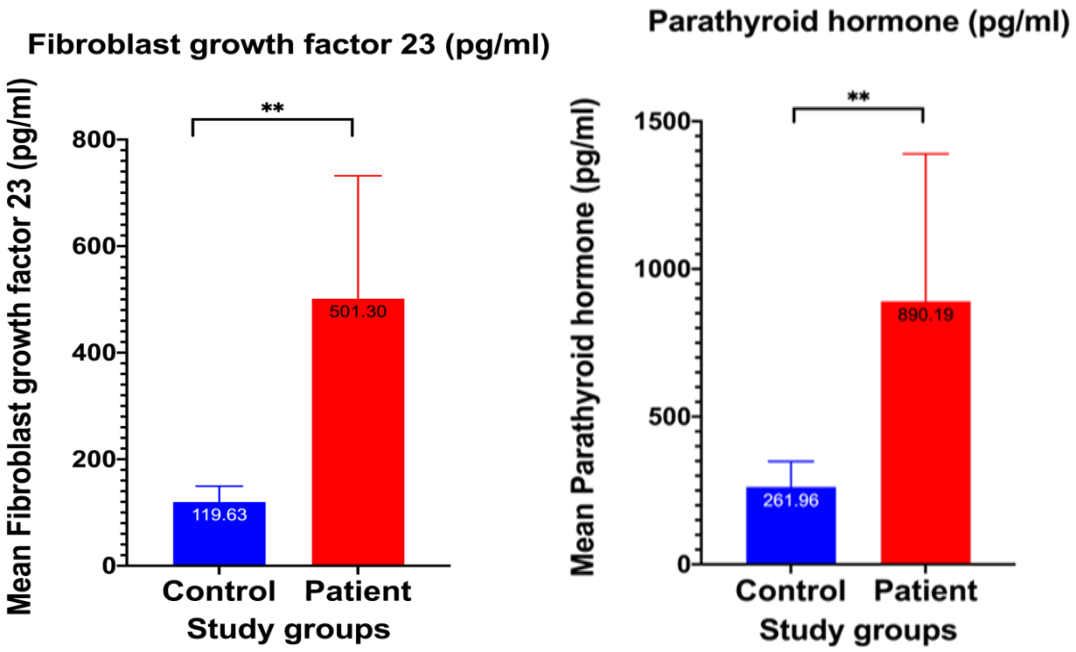


Figure (3-7): Comparison of FGF23,PTH between patient & control groups

Hb was significantly decreased (P<0.005) in patients (10.25±2.44) compared with control group (13.48±1.28), as summarized in (table 3-2), as summarized in (figure 3-8).

Table (3-2) &figure (3-8) showed that there was in serum levels significant difference of phosphate ($PO4^{-3}$) level in patients groups compared with control group. The levels of (PO4) in patients (2.01±0.76) was increasing significantly (P<0.001) compared with control groups (0.68±0.39).

Calcium (Ca++) levels was in serum levels significantly low (P<0.001) in patients (8.21±1.04) compared with control group (9.03±0.54) as shown in table (3-2) &figure (3-8)

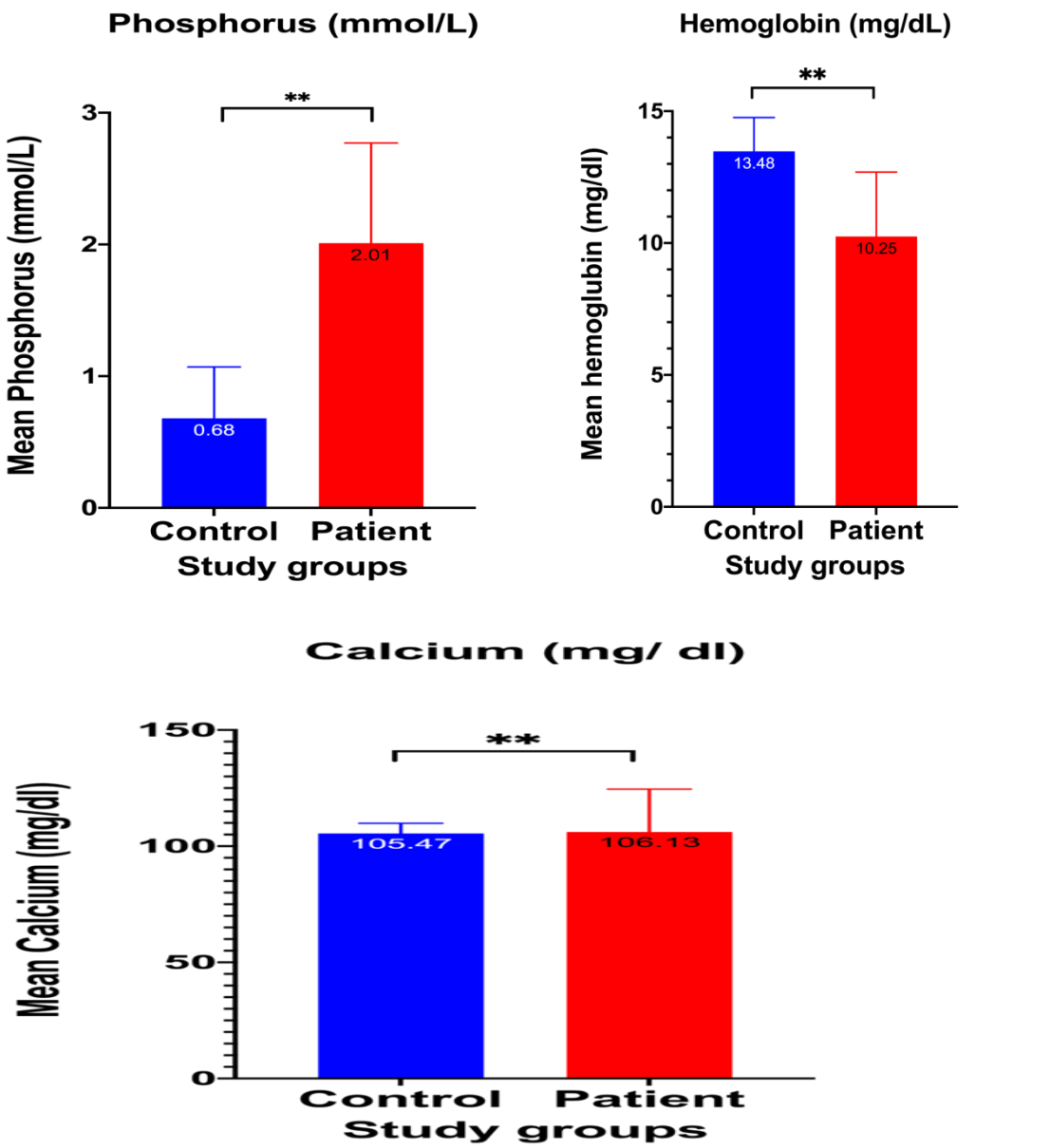


Figure (3-8): Comparison of Hb,Po4,Ca between patient & control groups

In this study, found the FGF23 levels was significantly elevated in patients compared to apparently healthy controls. This study agree with Javier A. Neyra *et al*. whose founds Acute kidney injury (AKI) is a state of high FGF23[98]. Elevated FGF23 levels have been observed in multiple studies of human AKI [99]. Plasma c-terminal FGF23 (cFGF23) levels were 5.6-fold higher in patients with AKI versus age-matched patients

without AKI [100]. When a patient has a reduced glomerular filtrate rate below 30% of normal, one nephron has to do the work of three. In this situation, there is a phosphate load since phosphate intake is maintained, but the number of glomeruli available to filter phosphate is decreased to 30%. The phosphate load stimulates FGF23, which decreases tubular absorption to 30%, so there is no increase in serum phosphate levels; however, a high concentration of phosphate in the tubule produces a decrease in renal Klotho[101]. The low Klotho level causes a further increase of FGF23 due to the resistance of the kidney to the action of FGF23, which leads to a significant decrease in calcitriol [102]. Thus, an increase in the levels of FGF23 and PTH, and a decrease in calcitriol and Klotho, are related to the onset of cardiovascular and other diseases [103] If the glomerular filtrate rate drops to 15% (approximately), a nephron has to do the work of 6, and the levels of FGF23 and PTH increase further. However, when 60% of the phosphate excretion fraction is exceeded (in humans), the kidney is not able to excrete enough phosphate so hyperphosphatasemia occurs [101]. FGF23 acts directly on the renal proximal tubular cells to suppress phosphate reuptake and expression of 1ahydroxylase, the rate-limiting enzyme for vitamin D production, by binding to FGFR1 and to FGFR3 and FGFR4, respectively [104]. Moreover, FGF23 acts directly on the distal tubular cells to stimulate calcium and sodium reabsorption [105].

the current study found the PTH was significantly higher in patients compared to apparently healthy controls. This study agree with Pazianas et al , who found the serum calcium and phosphate (Pi) remain normal until the late stages of CKD at the expense of elevate fibroblast growth factor-23 (FGF-23), a phosphatidic hormone, followed by reduced 1,25-

dihydroxy-vitamin D (1,25[OH]2D) and finally elevated parathyroid hormone (PTH)[106].

In present study found the phosphorus was significantly higher in patients compared to apparently healthy controls while calcium was significantly lower in patients compared to apparently healthy controls this study agree with (Yap et al) [107]

Loss of renal function results in reductions of Ca and Pi excretion. In individuals with normal renal function, the fractional urinary excretion of Ca is only 1% to 2% of the ultra-filterable Ca (ionized and complexed fractions) and Concurrently, FGF-23 could intervene to correct the Pi levels and prevent the development of hyperphosphatemia. This new pattern of response to Ca and Pi retention establishes FGF-23 as the dominant hormone in the regulation of Pi concentrations. Furthermore, FGF-23 will have inhibitory effects on PTH secretion whilst extracellular Ca2+ remains within normal range[108].At some stage, because of constantly raised Pi levels, FGF-23 will reach its limits on normalizing serum Pi and this could be the point when PTH will start rising, thus establishing a new equilibrium status in the homeostatic system.

The present study found the blood concentration of Hb significantly lower in patients compared with control group and this result agree with (Micarelli et al)[109]

Severe anemia is a common complication in CKD and is a risk factor for cardiovascular disease and heart failure in CKD patients. The cause of renal anemia is multifactorial and includes decreased erythropoietin production, iron deficiency, and inflammation. At previous studies, no association has been reported between high FGF23 levels and renal anemia according to animal or clinical studies. However, an observational study of

53 CKD patients showed that high FGF23 levels are negatively correlated with hemoglobin levels [110].

3.3 Relationships and correlation coefficients:

3.3.1correlation between GFR and another parameter

Table (3-3) correlation between GFR and another parameter

Glomerular Filtration Rate (ml/min)	Patient	
	r	P
Fibroblast growth factor 23 (pg/ml)	0.185	0.199
Parathyroid hormone (pg/ml)	0.083	0.565
Serum Creatinine (mg/dL)	-0.633**	0.000
Blood Urea (mg/dL)	-0.268	0.059
Hemoglobin (mg/dL)	0.219	0.127
Fasting blood sugar (mg/dl)	-0.275	0.053
phosphorus (mmol/L)	0.345*	0.014
Potassium (mmol/L)	-0.190	0.187
Sodium (mmol/L)	-0.035	0.809
Chloride (mmol/L)	0.098	0.498
Calcium (mg/ dl)	-0.152	0.291

Table(3-4) correlation between FGF23 and another parameter

Fibroblast growth factor 23 (pg/ml)	Patient	
	r	p
Parathyroid hormone (pg/ml)	0.029	0.843
Glomerular Filtration Rate (ml/min)	0.185	0.199
Serum Creatinine (mg/dL)	-0.084	0.562
Blood Urea (mg/dL)	0.163	0.258
Hemoglobin (mg/dL)	-0.088	0.542
Fasting blood sugar (mg/dl)	-0.159	0.271
phosphorus (mmol/L)	0.181	0.209
Potassium (mmoI/L)	0.043	0.769
Sodium (mmoI/L)	-0.045	0.757
Chloride (mmol/L)	-0.021	0.886
Calcium (mg/dl)	0.013	0.928

Table(3-5) correlation between Parathyroid hormone and another parameter

Parathyroid hormone (pg/ml)	Patient	
	r	P
Fibroblast growth factor 23 (pg/ml)	0.029	0.843
Glomerular Filtration Rate (ml/min)	0.083	0.565
Serum Creatinine (mg/dL)	-0.172	0.231
Blood Urea (mg/dL)	-.289*	0.041
Hemoglobin (mg/dL)	-0.068	0.637
Fasting blood sugar (mg/dl)	0.014	0.925
phosphorus (mmol/L)	0.117	0.418
Potassium (mmoI/L)	0.132	0.359
Sodium (mmoI/L)	0.096	0.506
Chloride (mmol/L)	-0.037	0.796
Calcium (mg/ dl)	-0.113	0.436

During our study, we found that there is a positive relationship between the levels of FGF23 and the GFR in the patients, where the value of (r = 0.185 p-value 0.199). Also, found positive relationship between the GFR& parathyroid hormone whom the study was conducted, where its value during the study 0.99, p-value <0.05. In figure (3.9).

Also, a clarification of the existence of positive relationship between GFR & phosphors whom the study was conducted, as we found its value during the study 0.345, p-value 0.014. In figure (3.10).

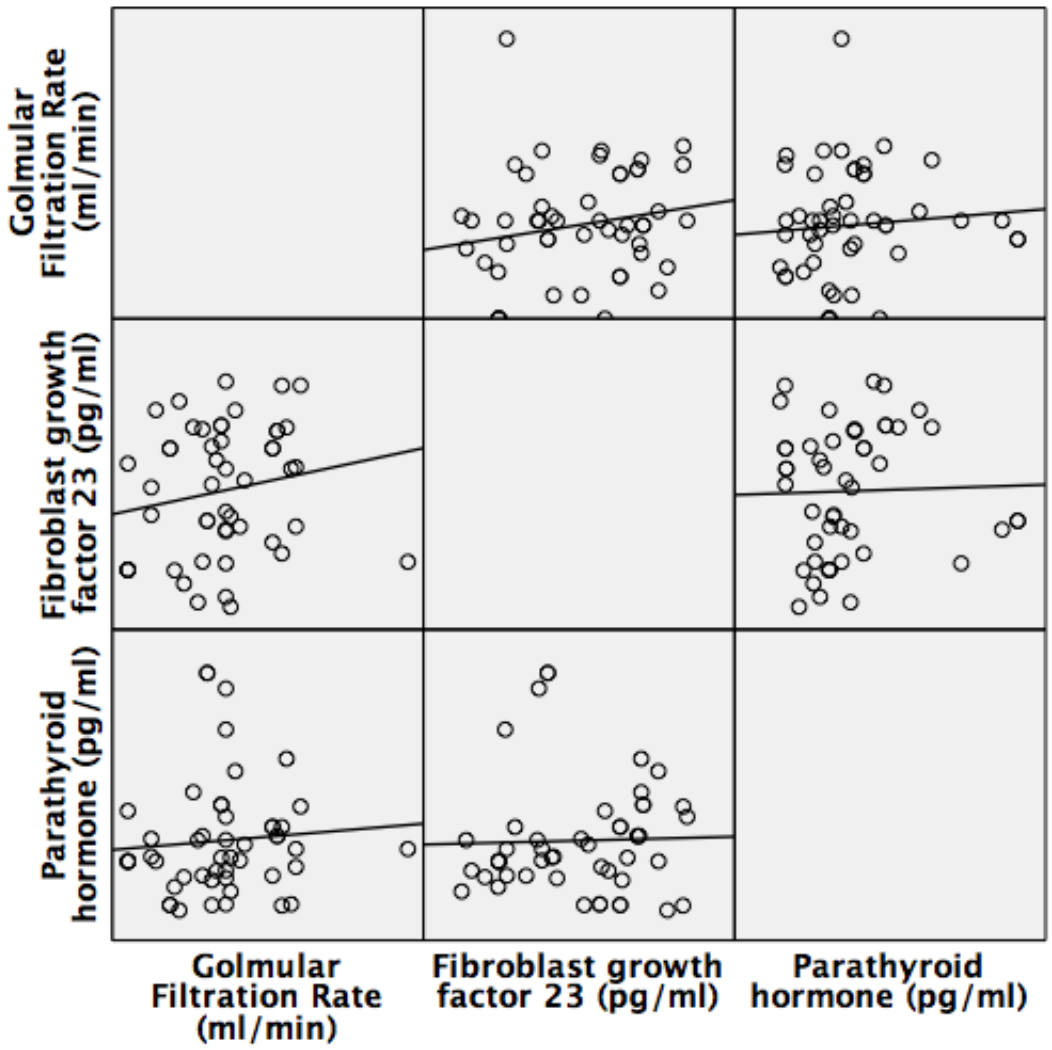


Figure (3.9). Correlation between GFR, FGF23&PTH

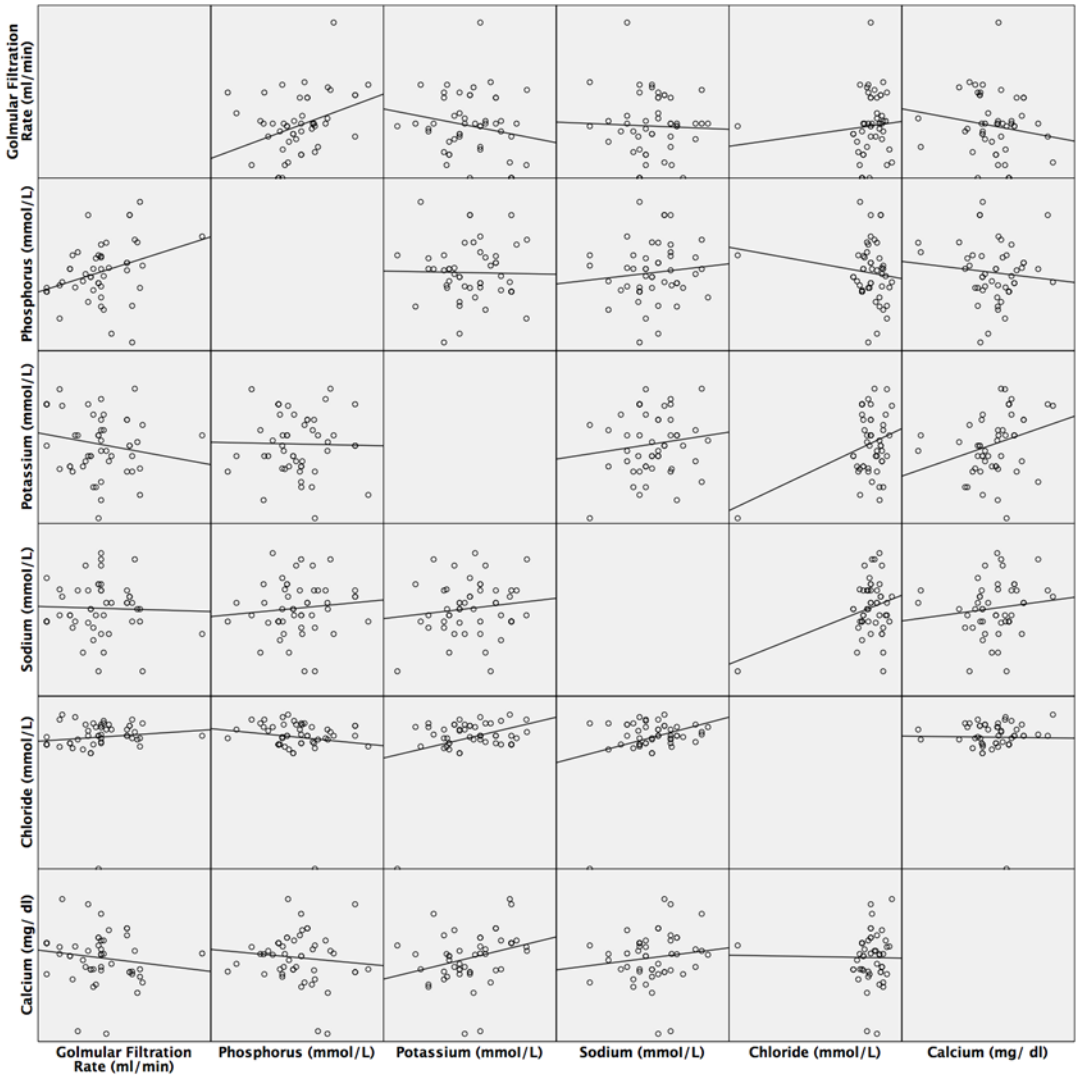


Figure (3.10).correlation between parameters

During the results of our study, there is a decrease in GFR and an increase in FGF23, PTH and PO4, and we believe that the reason for the positive relationship between them is that due to the sudden blockage in the kidneys, the rapid rise of FGF23, PTH and PO4 leads to sudden onset of acute kidney disease stages leads to a rapid decrease in GFR. Interestingly, limited available studies proving , we would also like to point out that in our new study.

And most of the previous studies in CKD have an inverse relationship FGF23, PTH, PO4 and GFR which means that an increase in one leads to a decrease in the other because These data suggest that the increase in PTH with decreasing GFR described in textbooks of kidney physiology reflects primarily the elevation of oxidized PTH(oxPTH) but not of Non-oxidized PTH (n-oxPTH). this data indicate that the progressive increase in FGF23 with decreasing GFR seems not to be associated with the moderate rise in n-oxPTH, but that other factors might play a more important role in the regulation of FGF23 levels in CKD[111].

Also in present study a clarification of the existence of positive relationship between FGF23 & parathyroid hormone whom the study was conducted, as we found its value during the study r 0.029, p-value 0.843. In figure (3.9).

In recent studies, they show that serum FGF23 is directly related with PTH which is in keeping with experimental studies in animal models indicating a stimulating effect by FGF23 on the parathyroid glands . However, in contrast with observations in hemodialysis patients, findings in this trial also show that, independently of FGF23, the parathyroid glands of patients with moderate to severe CKD maintain an intact ability to respond to vitamin D receptor (VDR) activation. However, as discussed above, whether FGF23 contributes to secondary hyperparathyroidism in CKD is difficult to assess, because 1,25 VD suppression by FGF23 may have a major role in raising PTH in these patients[112].

Along with previous studies in CKD, we observed a close association between baseline serum FGF23 and PTH in CKD which was largely independent of the GFR and of biomarkers of the bone mineral disorder in this population including serum calcium, phosphate and 1,25 vitamin D.

Such an association is compatible with the hypothesis that FGF23 has a direct stimulatory on PTH secretion [113] [114].

In current study we also found strong inverse relationship between GFR and Blood urea,serum creatinin between the paitents for whom the study was conducted, where it found that the increase of one of them was dependent on the decrease of the other, as we found its value during the study(r -0.633, p-value 0.000) (r -0.268, p-value 0.059) in serum creatinin & blood urea respectively In figure (3.11).

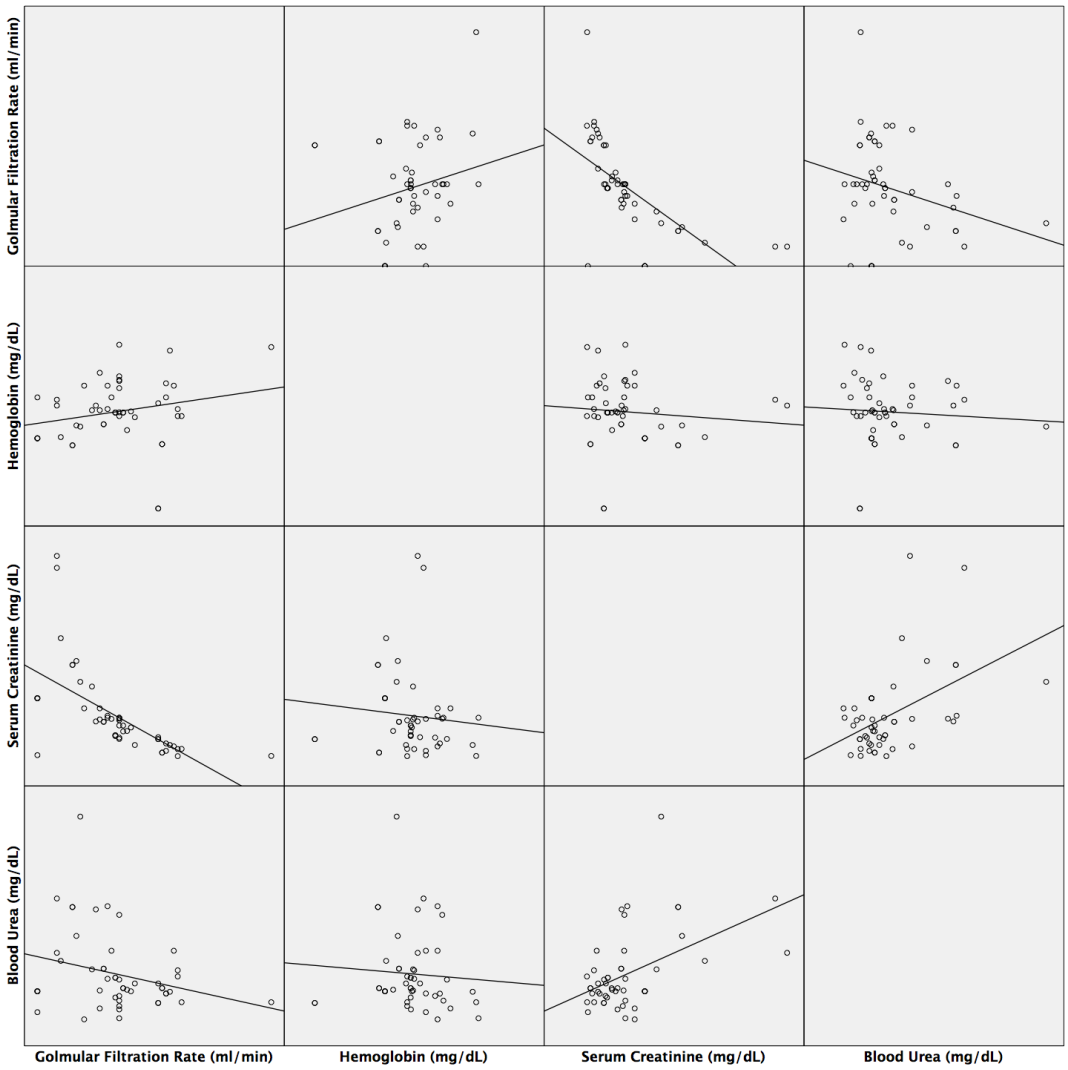


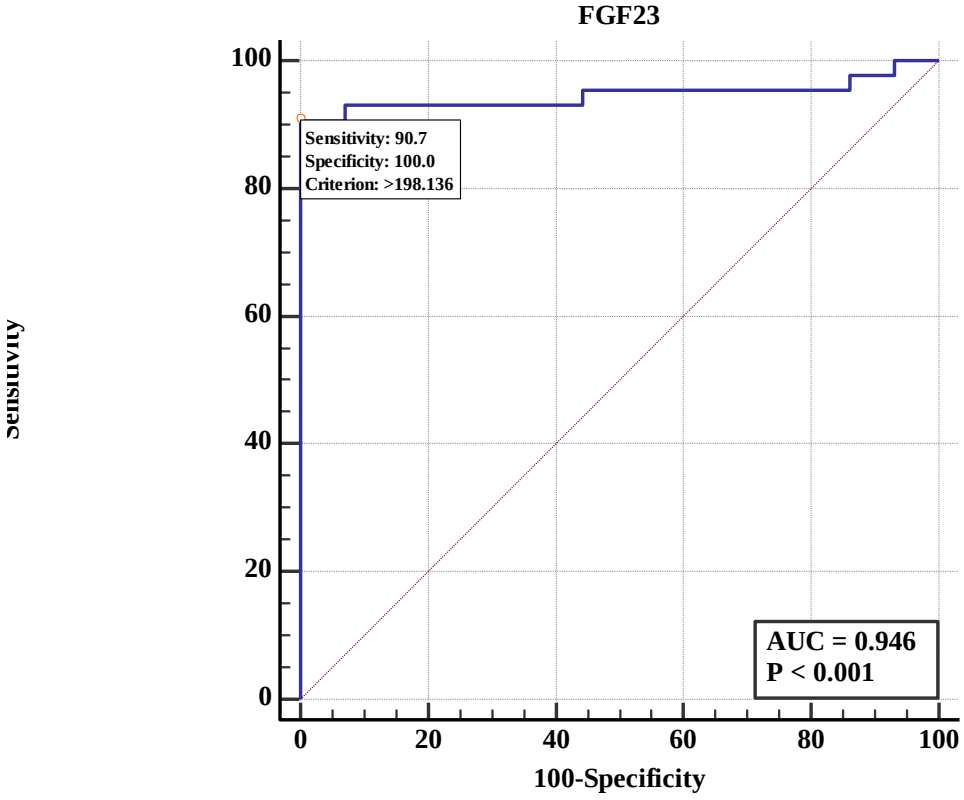
Figure (3.11).correlation between parameters

Creatinine is the anhydrous form of creatine that is present in muscles. Creatinine is filtered by the glomerulus and thus, serum creatinine estimation is considered as an indirect measure of glomerular filtration. Diminishing of GFR results in the rise of serum creatinine and urea levels [115] .

And we found inverse relationship between GFR and calicum (r -0.152, p-value 0.291) In figure (3.10).

3.4 Receiver operating characteristic(ROC Test)

It is a special program that determines the sensitivity and specificity of the marker with patients . Because of the highly significant difference in the level of FGF23 in patients groups when compared control groups, the authors of the current study suggested the presence of a cutoff value that can predict a diagnosis of obstractive renal disease with certain level of accuracy. For that reason, receiver operator characteristic (ROC) curve analysis was carried out and the results are shown in figure (3-12) and table (3-6) (3-7).



(Figure 3-12) Criterion values and coordinates of the ROC curve analysis for FGF23 as differentiating patients from control subjects

(Table 3-6) Comparison and Criterion values and coordinates of the ROC curve for FGF23 as differentiating patients from control subjects

Area Under the Curve				
Test Result Variable(s): Fibroblast growth factor 23 (pg/ml)				
Area	Std. Error	Asymptotic Sig.	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
0.954	0.026	0.000	0.903	1.000

Table (3-7) best discriminative (with maximum sensitivity + specificity values) cut-off values and their criteria of the serum FGF23

		Study groups	
		Control	Patient
Fibroblast growth factor (pg/ml)	< 200 (pg/ml)	50	4
	%	100.0%	8.0%
	> 200 (pg/ml)	0	46
	%	0.0%	92.0%
Total		50	50
Effect size			
Sensitivity		92 (81.16-96.85)	
Specificity		100 (92.87-100)	
Positive Predictive Value		100 (92.29-100)	
Negative Predictive Value		92.59 (82.455-97.08)	

FGF23 serum with (AUC=0.946) in patients and CI between (0.903-1.000) in patients, the cut-off point for serum FGF23 = (>198.136).The sensitivity was (92%). Specificity was (100%) Table (3-13) (3-14).

We show there highly specific which indicates the FGF23 diagnostic for obstructive renal disease &highly sensitive for renal impairment that The AUC is close to one that It can separate the patients from the healthy this study agree with (Rygasiewicz et al).

Rygasiewicz et al also shown prognostic value of FGF23 in a cohort of ICU patients with regard to incident AKI as well as in-hospital mortality, even after

adjustments for multiply confounders[116]. This finding is in concert with previously shown predictive value of FGF23 with regard to cardiovascular events and mortality, in cohorts of patients with chronic kidney disease and general population [117]. other data show also utility of FGF23 in outcome projection in acute settings such as in patients after cardiac arrest, cardiogenic shock or hospitalized in ICU [118].Noteworthy, FGF23 exerts its predictive value beyond cardiovascular system showing utility in prognostication of such life threating complication as AKI [119].

Conclusions

This study concluded that:

1- Patients with obstructive renal failure had greater levels of fibroblast growth factore-23, parathyroid hormone, and phosphors
2- The fibroblast growth factor and parathyroid hormone have a beneficial association.
3- FGF23 could be used as a prognostic marker in individuals with obstructive renal failure to predict the development of chronic kidney disease.
4- The high prevalence of obstructive renal disease in old age, with males outnumbering women.

Recommendations:

1- Preferably taken larger number of subjects.
2- The follow up study give a better result about the variability in the level of FGF23.
3- Always wear a mask and gloves to avoid infection with the Corona virus from the patient's relatives or the patient himself.
4- Make sure that the patient is fasting before make any analyzed such as fasting blood sugar because most patients are not fasting before performing the analysis so it may cause a possible sample loss due to the impact on of some results.
5- Do not sample patients until be sure that they are in a good psychological condition, because they are in severe pain and we may hurt them during withdrawal.
6- Most of the patients have lost large amounts of blood or have dehydration, so their veins are weak, so it is preferable to seek help from the health staff to ensure that they are not pricked more than once and hurt them.
7- Talk with family members about the history of the disease and the patient's habits, because we noticed that most patients suffer from kidney stones for a long time and there is no consultation from the doctor, which led to a sudden and severe blockage.
8- The hospital staff is a good source of information for the researcher. used them to find out the presence of potential patients and the dates of their arrival to the hospital.
9- The research recommend to do comparative study with non-abstractive renal failure for same parameter.

Reference

1. Undre S, Marks SD. Obstructive Uropathy. In: Pediatric Kidney Disease. Springer; 2016. p. 1121–33.

2. Cheungpasitporn W, Rossetti S, Friend K, Erickson SB, Lieske JC. Treatment effect, adherence, and safety of high fluid intake for the prevention of incident and recurrent kidney stones: a systematic review and meta-analysis. J Nephrol. 2016;29(2):211–9.

3. Stevens S. Obstructive Kidney Disease. Nurs Clin North Am. 2018;53(4):569–78.

4. Dousdampanis P, Trigka K, Stefanidis I. Obstructive Nephropathy. 2020;

5. Klahr S. The geriatric patient with obstructive uropathy. Nephrol Geriatr Integr. 2000;167–77.

6. Van Biesen W, Yegenaga I, Vanholder R, Verbeke F, Hoste E, Colardyn F, et al. Relationship between fluid status and its management on acute renal failure (ARF) in intensive care unit (ICU) patients with sepsis: a prospective analysis. J Nephrol. 2005;18(1):54–60.

7. Tseng TY, Stoller ML. Obstructive uropathy. Clin Geriatr Med. 2009;25(3):437–43.

8. Yap E, Salifu M, Ahmad T, Sanusi A, Joseph A, Mallappallil M. Atypical causes of urinary tract obstruction. Case reports Nephrol. 2019;2019.

9. Rodgers AL. Physicochemical mechanisms of stone formation. Urolithiasis. 2017;45(1):27–32.

10. Santucci RA, Joyce GF, Wise M. Male urethral stricture disease. J Urol. 2007;177(5):1667–74.

11. Heyns CF. Urinary tract infection associated with conditions causing urinary tract obstruction and stasis, excluding urolithiasis and neuropathic bladder. World J Urol. 2012;30(1):77–83.

k12. Rishor-Olney CR, Hinson MR. Obstructive Uropathy. StatPearls [Internet]. 2020;

13. Gyang AN, Gomez NA, Lamvu GM. Endometriosis of the bladder as a cause of obstructive uropathy. JSLS J Soc Laparoendosc Surg. 2014;18(2):357.

14. Bakari AA, Gadam IA, Aliyu S, Suleiman I, Ahidjo AA, Pindiga UH. Use of Mitrofanoff and Yang–Monti Techniques as Ureteric Substitution for Severe Schistosomal Bilateral Ureteric Stricture: A Case Report and Review of the Literature. Niger J Surg. 2012;18(1).

15. Wrona AJ, Zgajewski J, Kopeć N, Chodor D, Kopcza P, Klekot S. Subcutaneous pyelovesical bypass–Detour bypass–as a solution for ureteric obstruction. Cent Eur J Urol. 2017;70(4):429.

16. Klahr S. Obstructive nephropathy. Intern Med. 2000;39(5):355–61.

17. Kouba E, Wallen EM, Pruthi RS. Management of ureteral obstruction due to advanced malignancy: optimizing therapeutic and palliative outcomes. J Urol. 2008;180(2):444–50.

18. Friedlander JI, Duty BD, Okeke Z, Smith AD. Obstructive uropathy from locally advanced and metastatic prostate cancer: an old problem with new therapies. J Endourol. 2012;26(2):102–9.

19. Batlle DC, Arruda JAL, Kurtzman NA. Hyperkalemic distal renal tubular acidosis associated with obstructive uropathy. N Engl J Med. 1981;304(7):373–80.

20. Colgan R, Williams M, Johnson JR. Diagnosis and treatment of acute pyelonephritis in women. Am Fam Physician. 2011;84(5):519–26.

21. Hickling D, Aponte M, Nitti V. Evaluation and management of outlet obstruction in women without anatomical abnormalities on physical exam or cystoscopy. Curr Urol Rep. 2012;13(5):356–62.

22. Girardi M, Martin N. Obstructive uropathy. Hosp Med Clin. 2015;4(3):328–41.

23. Choyke PL. Radiologic evaluation of hematuria: guidelines from the American College of Radiology's appropriateness criteria. Am Fam Physician. 2008;78(3):347.

24. Nakanishi S, Miyazato M, Miyagi R, Saito S. Acute renal failure in a neonate due to bilateral primary obstructive megaureters. J Pediatr Surg case reports. 2018;31:69–70.

25. Marshall WJ, Lapsley M, Day A, Shipman K. Clinical chemistry. Elsevier Health Sciences; 2020.

26. Etafy MH, Saleh FH, Ortiz-Vanderdys C, Hamada A, Refaat AM, Aal MA, et al. Rapid versus gradual bladder decompression in acute urinary retention. Urol Ann. 2017;9(4):339.

27. Nemoy NJ, Fichman MP, Sellers A. Unilateral Ureteral Obstruction: A Cause of Reversible High Renin Content Hyertension. JAMA. 1973;225(5):512–3.

28. Sun Z. Aging, arterial stiffness, and hypertension. Hypertension. 2015;65(2):252–6.

29. Santoro J, Kaye D. Recurrent urinary tract infections: Pathogenesis and management. Med Clin North Am. 1978;62(5):1005–20.

30. Byers LA, Rochon PJ. Obstructive uropathy. In: IR Playbook. Springer; 2018. p. 477–87.

31. Lerma E V, Sparks MA, Topf J. Nephrology Secrets E-Book. Elsevier Health Sciences; 2018.

32. Truong L, Mai K. Obstructive nephropathy: what the surgical pathologist should know. AJSP Rev Reports. 2015;20(6):250–5.

33. Bascands J-L, Schanstra JP. Obstructive nephropathy: insights from genetically engineered animals. Kidney Int. 2005;68(3):925–37.

34. Chevalier RL. Pathogenesis of renal injury in obstructive uropathy. Curr Opin Pediatr. 2006;18(2):153–60.

35. Khalaf IM, Shokeir AA, El-Gshi FI, Amr HS, Amin MM. Recoverability of renal function after treatment of adult patients with unilateral obstructive uropathy and normal contralateral kidney: a prospective study. Urology. 2004;64(4):664–8.

36. Watson WJ, Brost BC. Maternal hydronephrosis in pregnancy: poor association with symptoms of flank pain. Am J Perinatol. 2006;23(08):463–6.

37. Cheung KL, Lafayette RA. Renal physiology of pregnancy. Adv Chronic Kidney Dis. 2013;20(3):209–14.

38. Scales Jr CD, Smith AC, Hanley JM, Saigal CS, Project UD in A. Prevalence of kidney stones in the United States. Eur Urol.

2012;62(1):160–5.

39. SATHICK IJ. 31 Obstructive Nephropathy in Cancer. Onco-Nephrology E-b. 2019;281.

40. Prabhakar H, Rajan S, Kapoor I, Mahajan C. Problem Based Learning Discussions in Neuroanesthesia and Neurocritical Care. Springer; 2020.

41. Klahr S. New insights into the consequences and mechanisms of renal impairment in obstructive nephropathy. Am J kidney Dis. 1991;18(6):689–99.

42. Palmieri PA. Obstructive nephropathy: pathophysiology, diagnosis, and collaborative management. Nephrol Nurs J. 2002;29(1):15–26.

43. Chávez-Iñiguez JS, Navarro-Gallardo GJ, Medina-González R, Alcantar-Vallin L, García-García G. Acute Kidney Injury Caused by Obstructive Nephropathy. Int J Nephrol. 2020;2020.

44. Purkerson ML, Klahr S. Prior inhibition of vasoconstrictors normalizes GFR in postobstructed kidneys. Kidney Int. 1989;35(6):1306–14.

45. Gilbert S, Weiner DE. National Kidney Foundation Primer on Kidney Diseases E-Book. Elsevier Health Sciences; 2013.

46. Li C, Wang W, Knepper MA, Nielsen S, Frøkiær J. Downregulation of renal aquaporins in response to unilateral ureteral obstruction. Am J Physiol Physiol. 2003;284(5):F1066–79.

47. Murer L, Addabbo F, Carmosino M, Procino G, Tamma G, Montini G, et al. Selective decrease in urinary aquaporin 2 and increase in prostaglandin E2 excretion is associated with postobstructive

polyuria in human congenital hydronephrosis. J Am Soc Nephrol. 2004;15(10):2705–12.

48. Chapple CR, Steers WD, Evans CP. Urologic Principles and Practice. Springer; 2020.

49. Ill OU in C, 64 CP, Abudayyeh COK and A. Obstructive Uropathy in Critically Ill Cancer Patients. Oncol Crit Care. 2020;969–75.

50. Ophascharoensuk V, Giachelli CM, Gordon K, Hughes J, Pichler R, Brown P, et al. Obstructive uropathy in the mouse: role of osteopontin in interstitial fibrosis and apoptosis. Kidney Int. 1999;56(2):571–80.

51. Chevalier RL, Forbes MS, Thornhill BA. Ureteral obstruction as a model of renal interstitial fibrosis and obstructive nephropathy. Kidney Int. 2009;75(11):1145–52.

52. Hiatt MJ, Ivanova L, Trnka P, Solomon M, Matsell DG. Urinary tract obstruction in the mouse: the kinetics of distal nephron injury. Lab Investig. 2013;93(9):1012–23.

53. Goetz R, Mohammadi M. Exploring mechanisms of FGF signalling through the lens of structural biology. Nat Rev Mol cell Biol. 2013;14(3):166.

54. Ornitz DM, Itoh N. The fibroblast growth factor signaling pathway. Wiley Interdiscip Rev Dev Biol. 2015;4(3):215–66.

55. Dolegowska K, Marchelek-Mysliwiec M, Nowosiad-Magda M, Slawinski M, Dolegowska B. FGF19 subfamily members: FGF19 and FGF21. J Physiol Biochem. 2019;1–12.

56. Leifheit-Nestler M, Haffner D. Paracrine effects of FGF23 on the

heart. Front Endocrinol (Lausanne). 2018;9.

57. Takashi Y, Fukumoto S. FGF23 beyond phosphotropic hormone. Trends Endocrinol Metab. 2018;

58. Kanbay M, Vervloet M, Cozzolino M, Siriopol D, Covic A, Goldsmith D, et al. Novel faces of fibroblast growth factor 23 (FGF23): iron deficiency, inflammation, insulin resistance, left ventricular hypertrophy, proteinuria and acute kidney injury. Calcif Tissue Int. 2017;100(3):217–28.

59. Prié D, Friedlander G. Reciprocal control of 1, 25-dihydroxyvitamin D and FGF23 formation involving the FGF23/Klotho system. Clin J Am Soc Nephrol. 2010;5(9):1717–22.

60. Kocełak P, Olszanecka-Glinianowicz M, Chudek J. Fibroblast growth factor 23—structure, function and role in kidney diseases. Adv Clin Exp Med. 2012;21(3):391–401.

61. Urakawa I, Yamazaki Y, Shimada T, Iijima K, Hasegawa H, Okawa K, et al. Klotho converts canonical FGF receptor into a specific receptor for FGF23. Nature. 2006;444(7120):770.

62. Hu MC, Shi M, Zhang J, Pastor J, Nakatani T, Lanske B, et al. Klotho: a novel phosphaturic substance acting as an autocrine enzyme in the renal proximal tubule. FASEB J. 2010;24(9):3438–50.

63. Neyra JA, Hu MC. Potential application of klotho in human chronic kidney disease. Bone. 2017;100:41–9.

64. Kurosu H, Ogawa Y, Miyoshi M, Yamamoto M, Nandi A, Rosenblatt KP, et al. Regulation of fibroblast growth factor-23

signaling by klotho. J Biol Chem. 2006;281(10):6120–3.

65. Imura A, Iwano A, Tohyama O, Tsuji Y, Nozaki K, Hashimoto N, et al. Secreted Klotho protein in sera and CSF: implication for post-translational cleavage in release of Klotho protein from cell membrane. FEBS Lett. 2004;565(1–3):143–7.

66. Courbebaisse M, Lanske B. Biology of fibroblast growth factor 23: from physiology to pathology. Cold Spring Harb Perspect Med. 2018;8(5):a031260.

67. Strewler GJ. FGF23, hypophosphatemia, and rickets: Has phosphatonin been found? Proc Natl Acad Sci. 2001;98(11):5945–6.

68. Ito N, Fukumoto S, Takeuchi Y, Yasuda T, Hasegawa Y, Takemoto F, et al. Comparison of two assays for fibroblast growth factor (FGF)-23. J Bone Miner Metab. 2005;23(6):435–40.

69. Schumacher D, Alampour-Rajabi S, Ponomariov V, Curaj A, Wu Z, Staudt M, et al. Cardiac FGF23: new insights into the role and function of FGF23 after acute myocardial infarction. Cardiovasc Pathol. 2019;40:47–54.

70. Bär L, Stournaras C, Lang F, Föller M. Regulation of fibroblast growth factor 23 (FGF 23) in health and disease. FEBS Lett. 2019;

71. Rodelo-Haad C, Santamaria R, Muñoz-Castañeda JR, Pendón-Ruiz de Mier M, Martin-Malo A, Rodriguez M. FGF23, Biomarker or Target? Toxins (Basel). 2019;11(3):175.

72. 10.1016/j.semnephrol.2018.10.005. Fibroblast Growth Factor 23 and Klotho in AKI. Semin Nephrol [Internet]. 2019;39(1):57–75.

Available from: https://doi.org/10.1016/j.semnephrol.2018.10.005

73. Andrukhova O, Slavic S, Smorodchenko A, Zeitz U, Shalhoub V, Lanske B, et al. FGF23 regulates renal sodium handling and blood pressure. EMBO Mol Med. 2014;6(6):744–59.

74. Yuan B, Takaiwa M, Clemens TL, Feng JQ, Kumar R, Rowe PS, et al. Aberrant Phex function in osteoblasts and osteocytes alone underlies murine X-linked hypophosphatemia. J Clin Invest. 2008;118(2):722–34.

75. Benet-Pagès A, Orlik P, Strom TM, Lorenz-Depiereux B. An FGF23 missense mutation causes familial tumoral calcinosis with hyperphosphatemia. Hum Mol Genet. 2004;14(3):385–90.

76. Sitara D, Razzaque MS, Hesse M, Yoganathan S, Taguchi T, Erben RG, et al. Homozygous ablation of fibroblast growth factor-23 results in hyperphosphatemia and impaired skeletogenesis, and reverses hypophosphatemia in Phex-deficient mice. Matrix Biol. 2004;23(7):421–32.

77. Wein MN, Kronenberg HM. Regulation of bone remodeling by parathyroid hormone. Cold Spring Harb Perspect Med. 2018;8(8):a031237.

78. Rosen RD, Bordoni B. Embryology, Parathyroid. 2020;

79. Byrd C, Kashyap S, Kwartowitz G. Parathyroid Cancer. In: StatPearls [Internet]. StatPearls Publishing; 2020.

80. Khan M, Sharma S. Physiology, parathyroid hormone (PTH). 2018;

81. Chu V, Goggs R, Bichoupan A, Radhakrishnan S, Menard J. Hypophosphatemia in Dogs With Presumptive Sepsis: A

Retrospective Study (2008–2018). Front Vet Sci. 2021;8:207.

82. Rudin A V, McKenzie TJ, Wermer R, Thompson GB, Lyden ML. Primary hyperparathyroidism: redefining cure. Am Surg. 2019;85(2):214–8.

83. Varacallo M, Mair SD. StatPearls [Internet] StatPearls Publishing. Treasure Isl Jun. 2019;4.

84. Hans SK, Levine SN. Hypoparathyroidism. StatPearls [Internet]. 2020;

85. Goltzman D. Physiology of parathyroid hormone. Endocrinol Metab Clin. 2018;47(4):743–58.

86. Searcy RL, Reardon JE, Foreman JA. Enzymatic serum urea determination. Am J Med Technol. 1967;33:15–20.

87. Bernard JH. Clinical diagnosis and managment by laboratory methods. Saunders; 1996.

88. Levey AS, Bosch JP, Lewis JB, Greene T, Rogers N, Roth D. A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation. Ann Intern Med. 1999;130(6):461–70.

89. Moss DW, Henderson AR, Kachmar JF. Enzymes In: Fundamentals of clinical chemistry 3rd ed n. Tietz, NW Ed. Philadelphia, WB Saunders Company; 1987.

90. Burtis CA, Ashwood ER. Tietz textbook of clinical chemistry. 1999;

91. Sati C, Chattor AJ, Watts N. Fundamentals of Clinical Chemistry. Ed. Tietz, NW 3rd Ed., pg. 586. Saunders press Phila; 1987.

92. Hillmann G, Beyer G. Rapid determination of serum potassium by turbidity measurement with kalignost after protein precipitation. Z Klin Chem Klin Biochem. 1967;5(2):93–4.

93. Elnokeety MM, Shaker AM, Fayed AM. Creatinine, cystatin, and combined-based equations in assessment of renal functions in type 2 diabetic Egyptian patients. Egypt J Intern Med. 2017;29(3):105.

94. Alvencar S, Gayen S, Kohli A, Stuewe E, Rabin A, Anderlind C. A Cruise to Remember: Unilateral Urinothorax Secondary to Obstructive Uropathy. In: D40 AIR AND BODILY FLUIDS IN THE THORACIC SPACE: CASE REPORTS IN PLEURAL DISEASE. American Thoracic Society; 2020. p. A6703–A6703.

95. Tolani MA, Ahmed M, Nasir O, Sudi A. Obstructive uropathy and intrinsic renal disease in patients with benign prostatic obstruction: analysis of burden and associations in a university teaching hospital in Nigeria. J Med Trop. 2020;22(2):127.

96. Rosner MH, Capasso G, Perazella MA. Acute kidney injury and electrolyte disorders in the critically ill patient with cancer. Curr Opin Crit Care. 2017;23(6):475–83.

97. Xiao-jie Y, You-fang L, Dong Z, Qian W, Peng Z. Study on role of serum electrolyte combined with GFR evaluation equation in predicting the prognosis of severe obstructive renal injury. J Hainan Med Univ. 2019;25(17):60–5.

98. Neyra JA, Hu MC, Moe OW. Fibroblast Growth Factor 23 and αKlotho in Acute Kidney Injury: Current Status in Diagnostic and Therapeutic Applications. Nephron. 2020;144(12):665–72.

99. Christov M, Neyra JA, Gupta S, Leaf DE. Fibroblast growth factor

23 and klotho in AKI. In: Seminars in nephrology. Elsevier; 2019. p. 57–75.

100. Leaf DE, Wolf M, Waikar SS, Chase H, Christov M, Cremers S, et al. FGF-23 levels in patients with AKI and risk of adverse outcomes. Clin J Am Soc Nephrol. 2012;7(8):1217–23.

101. Muñoz-Castañeda JR, Herencia C, Pendón-Ruiz de Mier MV, Rodriguez-Ortiz ME, Diaz-Tocados JM, Vergara N, et al. Differential regulation of renal Klotho and FGFR1 in normal and uremic rats. FASEB J. 2017;31(9):3858–67.

102. Rodríguez-Ortiz ME, Díaz-Tocados JM, Muñoz-Castañeda JR, Herencia C, Pineda C, Martínez-Moreno JM, et al. Inflammation both increases and causes resistance to FGF23 in normal and uremic rats. Clin Sci. 2020;134(1):15–32.

103. Rodríguez M. FGF23: Is It Another Biomarker for Phosphate–Calcium Metabolism? Adv Ther. 2020;37(2):73–9.

104. Andrukhova O, Zeitz U, Goetz R, Mohammadi M, Lanske B, Erben RG. FGF23 acts directly on renal proximal tubules to induce phosphaturia through activation of the ERK1/2–SGK1 signaling pathway. Bone. 2012;51(3):621–8.

105. Andrukhova O, Smorodchenko A, Egerbacher M, Streicher C, Zeitz U, Goetz R, et al. FGF 23 promotes renal calcium reabsorption through the TRPV 5 channel. EMBO J. 2014;33(3):229–46.

106. Pazianas M, Miller PD. Current understanding of mineral and bone disorders of chronic kidney disease and the scientific grounds on the use of exogenous parathyroid hormone in its management. J bone Metab. 2020;27(1):1–13.

107. Yap H-K, PResontoc L. Obstructive uropathy. IAP Spec Ser Pediatr Nephrol. 2019;268.

108. Mace ML, Gravesen E, Nordholm A, Olgaard K, Lewin E. Fibroblast growth factor (FGF) 23 regulates the plasma levels of parathyroid hormone in vivo through the FGF receptor in normocalcemia, but not in hypocalcemia. Calcif Tissue Int. 2018;102(1):85–92.

109. Micarelli D, Cristi E, Taddei AR, Della Rovere FR, Mercanti C, Feriozzi S. A case of acute renal failure with multiple origins of the renal injury. CEN Case Reports. 2020;9(4):437–41.

110. Agoro R, Montagna A, Goetz R, Aligbe O, Singh G, Coe LM, et al. Inhibition of fibroblast growth factor 23 (FGF23) signaling rescues renal anemia. FASEB J. 2018;32(7):3752–64.

111. Zeng S, Querfeld U, Feger M, Haffner D, Hasan AA, Chu C, et al. Relationship between GFR, intact PTH, oxidized PTH, non-oxidized PTH as well as FGF23 in patients with CKD. FASEB J. 2020;34(11):15269–81.

112. D'Arrigo G, Pizzini P, Cutrupi S, Tripepi R, Tripepi G, Mallamaci F, et al. FGF23 and the PTH response to paricalcitol in chronic kidney disease. Eur J Clin Invest. 2020;50(2):e13196.

113. Kawakami K, Takeshita A, Furushima K, Miyajima M, Hatamura I, Kuro-o M, et al. Persistent fibroblast growth factor 23 signalling in the parathyroid glands for secondary hyperparathyroidism in mice with chronic kidney disease. Sci Rep. 2017;7(1):1–14.

114. Burnett-Bowie SM, Henao MP, Dere ME, Lee H, Leder BZ. Effects of hPTH (1-34) infusion on circulating serum phosphate, 1, 25-

dihydroxyvitamin D, and FGF23 levels in healthy men. J Bone Miner Res. 2009;24(10):1681–5.

115. Ray AS, Kare PK, Makwane HS, Saxena T, Garg C. Estimation of serum creatinine, serum urea, glomerular filtration rate and proteinuria among apparently healthy adults to assess the renal impairment and its association with body mass index: An observational hospital-based study. Studies. 2020;6:7.

116. Rygasiewicz K, Hryszko T, Siemiatkowski A, Brzosko S, Rydzewska-Rosolowska A, Naumnik B. C-terminal and intact FGF23 in critical illness and their associations with acute kidney injury and in-hospital mortality. Cytokine. 2018;103:15–9.

117. Scialla JJ, Xie H, Rahman M, Anderson AH, Isakova T, Ojo A, et al. Fibroblast growth factor-23 and cardiovascular events in CKD. J Am Soc Nephrol. 2014;25(2):349–60.

118. Leaf DE, Jacob KA, Srivastava A, Chen ME, Christov M, Jüppner H, et al. Fibroblast growth factor 23 levels associate with AKI and death in critical illness. J Am Soc Nephrol. 2017;28(6):1877–85.

119. Leaf DE, Christov M, Jüppner H, Siew E, Ikizler TA, Bian A, et al. Fibroblast growth factor 23 levels are elevated and associated with severe acute kidney injury and death following cardiac surgery. Kidney Int. 2016;89(4):939–48.

APPENDIX A

Questionnaire

Case No. Date:

Hospital name:

Patient name:-

Age: -

Sex:-

Weight: - Kg

BMI:

Height: - cm

Telephone number:-

Profession:-

Residence:-

Duration of disease:-

Final diagnosis of the disease:-

Other diseases:-

Type of Treatment: -

Smoking:-

الخلاصة

اعتلال المسالك البولية الانسدادي هو انسداد لتدفق البول قد يكون كليًا أو جزئيًا ، وقد يتسبب في تلف تدريجي ومتزايد للكلى. يمكن أن يحدث الانسداد في أحد الحالبين أو كليهما وقد يحدث على مستوى عنق المثانة أو بعيدًا عن ذلك (مثل الإحليل). الانسداد هو أحد الأسباب الرئيسية للفشل الكلوي وقد ركزت العديد من الدراسات على فهم مسببات وعواقب اعتلال المسالك البولية الانسدادي.

ان الهدف من الدراسة هو التحقيق في العلاقة بين مستويات المصل لعامل نمو الخلايا الليفية -٢٣ و ,وهرمون الغدة الجار درقية والفوسفور في المرضى الذين يعانون من الفشل الكلوي الانسدادي واخرون اصحاء.

اشتملت الدراسة على جمع عينات دم من ١٠٠ متطوع ، و ٥٠ شخصًا يتمتعون بصحة جيدة اعتبروا كمجموعه مقارنة (٣٨ رجلاً و ١٢ امرأة) ، (٣٤ رجلاً و ١٦ امرأة) يعانون من الفشل الكلوي الانسدادي . كان العمر (٢٥-٦٥) عامًا مؤشر كتلة الجسم (٢٤-٢٧) كجم / م ٢. تم ايجاد المرضى في مستشفى الحلة التعليمي قسم جرحة الكلية من الفترة من ١ ديسمبر ٢٠٢٠ إلى ١ يونيو ٢٠٢١ وتم قياس مستويات عامل خلايا النمو الليفية ٢٣ , هرمون جار درقي و فسفور بتقنية ايلايزا بينما تم قياس مستويات المعلمات الأخرى بطريقة القياس اللوني إلى دليل الشركة المصنعة.

كما أظهرت نتائج الاختبارات التي أجريت أن مستويات الهيموجلوبين والصوديوم و معدل الترشيح الكبيبي والكالسيوم لدى الأشخاص المصابين بمرض الانسداد الكلوي كانت أقل بشكل ملحوظ من مجموعة المقارنة وبشكل ملحوظ حيث كانت قيمة $P < 0.05$. كانت مستويات عامل نمو الخلايا الليفية ٢٣ ، هرمون الغدة الجار درقية ، الفوسفور ، البوتاسيوم ، في الأشخاص المصابين بمرض انسداد الكلى أعلى بكثير من مجموعة المقارنة وبشكل ملحوظ ، حيث أن قيمة $P < 0.05$ ، كما تم دراسة العلاقة بين مستويات عامل نمو الخلايا الليفية ٢٣ و GFR حيث تم العثور على قيمة $r = 0.185$ و $P\ 0.199$ ووجدت علاقة ارتباط موجبة بينهما وعلاقة موجبة بين عامل نمو الخلايا الليفية ٢٣ وهرمون الغدة الجار درقية $r = 0.029$، $P\ 0.843$.

يمكن الاستنتاج من الدراسة الحالية التي أجريناها أن مستوى حدوث عامل نمو الخلايا الليفية٢٣ ، وهرمون الغدة الجار درقية والفوسفور أعلى في المرضى الذين يعانون من الفشل الكلوي الانسدادي قياسا بالمجموعة المقارنة ، ويمكن استخدام عامل نمو الخلايا الليفية٢٣ كعلامة

تشخيصية في مرضى الفشل الكلوي الانسدادي للتنبؤ بإمكانية الإصابة بأمراض الكلى المزمنة ، حدوث مرض الانسداد الكلوي بمعدل كبير في الشيخوخة وعند الرجال أكثر من النساء ، أظهرت الدراسة الحالية أن مؤشر كتلة الجسم ليس ذات معنى لدى المريض مقارنة بمجموعة المقارنة. ، كان هناك انخفاض في مستويات الصوديوم والكالسيوم و معدل الترشيح الكبيبي في المرضى مقارنة مجموعة المقارنة ووجود علاقة إيجابية بين عامل نمو الخلايا الليفية ٢٣ و هرمون الغدة الدرقية ، وجود علاقة إيجابية بين معدل الترشيح الكبيبي وعامل نمو الخلايا الليفية ، وهرمون الغدة الجار درقية. والفوسفور والهيموجلوبين الذي تم إجراؤه والعلاقة العكسية بين معدل الترشيح الكبيبي والكريتيانين واليوريا والكالسيوم والبوتاسيوم، هنالك علاقة العكسية بين مستويات الهيموجلوبين والكريتيانين واليوريا وفي الدراسة الحالية يوجد علاقة بين الفوسفور والبوتاسيوم والصوديوم والكالسيوم المستويات والمعلمات، وتوجد بينهما علاقة عكسية وإيجابية

قوية.

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جامعة بابل/ كلية الطب
فرع الكيمياء الحياتية السريرية

دراسة كيميائية حيوية لعامل النمو الليفي (٢٣) وهرمون الغدة الجار درقية والفسفور لمرضى الفشل الكلوي الانسدادي للبالغين في محافظة بابل العراق

رسالة

مقدمة الى مجلس كلية الطب في جامعة بابل

كجزء من متطلبات نيل شهادة الماجستير

في العلوم/ الكيمياء الحياتية السريرية

من قبل

محمد كامل كاظم عباس

بكالوريوس تقنيات التحليلات المرضية
كلية الاسراء الجامعة
٢٠١٧

إشـــراف

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