2023 لسنة A10 العدد
No10A - Septembr 2023
المجلة العراقية للبحوث الانسانية والاجتماعية والعلمية
Iraqi Journal of Humanitarian, Social and Scientific Research
Print ISSN 2710-0952 -Electronic ISSN 2790-1254

Estimated The Level Of Apelin As An Innovative Early Indicator Of Acute Kidney Injury And Its Linear Relationship With B_2- MG, FGF 23, And Egfr In Both Control Groups And Acute Kidney Injury Patients

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Abstract:

Background: There is a need for early and accurate diagnosis of acute kidney injury, and low level of apelin, high levels of B2-Micro globulin (B2-MG), and fibroblast growth factor 23 (FGF 23), in blood serum give early diagnosis of acute kidney injury in patients. **Objective:** estimated the level of apelin as innovative early indicator in acute kidney injury and measured it in both control and acute kidney injury patients groups. Also finding it's relationship with each of B2-MG, FGF23, and estimated glomerular filtration rate (eGFR) of both groups. **Meterials and Methods:** This study was conducted on 232 blood samples were collected from males and the females, 108 of which were control subject, and 124 of which were from Patients with acute kidney injury whose age ranged (≤35 years - ≥56 years). **Results:** decreased in apelin (pg/ml) and (eGFR), increased both B2-MG and FGF23 in the blood serum of acute kidney injury patients group compared to the control group. **Conclusions:** The results showed the apelin and (eGFR) decreased in the blood serum of acute kidney injury group corresponding to an increased in the level of both B2-MG and FGF23 compared to the control group, which indicates the possibility that apelin of may be an indicator of early prevention and protection for acute kidney injury, in addition to B2- MG and FGF 23.

Keywords: Acute Kidney Injury (AKI), Apelin, B2-MG, FGF 23, eGFR.

تقدير مستوى الأبلين كمؤشر مبكر مبتكر لإصابة الكلى الحادة وعلاقته الخطية مع B_2- MG ، FGF 23، و EGFR في كلا المجموعتين الضابطتين ومرضى إصابات الكلى الحادة

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الملخص:-

هناك حاجة للتشخيص المبكر والدقيق لإصابة الكلية الحادة ، وانخفاض مستوى الأبيلين وإرتفاع مستويات كلا من B2-microglobulin وعامل النمو الليفي FGF23،في مصل الدم يعطي تشخيص مبكر لإصابة الكلية الحادة لدى المرضى . الهدف: تقدير مستوى الأبيلين على أنه مؤشر مبكر مبتكر على الإصابة الكلوية الحادة وقياسهُ في كل من مجموعتي السيطرة ومرضى الإصابة الكلوية الحادة ،كما تم العثور على علاقته بكل من FGF23، B2MG، ومعدل الترشيح الكبيبي المقدر((eGFRلكلا المجموعتين ، المواد والطرق:- أجريت هذه الدراسة على ٢٣٢ عينة دم، تم جمع العينات من الذكور والإناث ، ١٠٨ منها كانت مجموعة سيطرة ، و ١٢٤ فيها كانت من مرضى إصابات الكلية الحادة ،

والذين تتراوح أعمارهم (بين أقل او يساوي ٣٥ إلى اكبر أو يساوي ٦٥سنة) النتائج:- انخفاض في مستوى الأبيلين ((pg/mlوال ((eGFR،وزاد كلا من FGF23و B2-MGفي مصل الدم لمجموعة مرضى إصابة الكلية الحادة مقارنة بجموعة السيطرة. الاستنتاجات:- أظهرت النتائج أنخفاض الابيلين و ((eGFRفي مصل الدم لمجموعة مرضى إصابة الكلية الحادة ، بما يتوافق مع زيادة في مستوى كل من ال FGF23وB2-MG مقارنة بمجموعة السيطرة ، فالابيلين هو مؤشر للوقاية المبكرة والحماية من إصابة الكلية الحادة بالاضافة الى FGF23وB2-MG

الكلمات الرئيسية: إصابة الكلى الحادة (AKI)، أبلين، B2-MG، FGF 23، eGFR.

Introduction:-

Acute Kidney injury (AKI) is a clinical syndrome that appears precisely in the rapid or sudden deterioration in kidney function and the resulting imbalance of electrolytes in the body and it's volume, In addition to the abnormal retention of nitrogenous wastes and residues in body [1].

In addition to keeping; Acute kidney injury, also called acute kidney failure, occurs when the kidneys stop working suddenly, which can happen withen a few hours or days, the Kidney loses its ability to rid the body of waste, which leads to its accumulation in the body, and this in turn leads to the kidney losing its ability to maintain the correct balance of question in the body [2].

Acute Kidney failure is not present permanetly in the event of immediate treatment and in the event that the affected person does not have serious health problems, as the kidneys can return to work normally [2, 3].

For this reason, in order to facilitate the early diagnosis of acute kidney failure we must develop a more accurate and deeper explanation of acute kidney failure, as we must detect more sensitive and capable marker than creatinine for early detection of acute kidney failure [4], one of the recent signs in the discovery of acute kidne kidney injury is apelin, B2-Miroglobulin, FGF23, and eGFR.

Apelin:

It is an endogenous ligand-peptide of the orphan G protein-coupled J (APJ) receptor, which was discovered in 1998 and isolated by the scientist Tatemoto [5]. Apelin belongs to the family of adipokines [6], which represent biologically active mediators secreated by adipose tissue, the wide tissue distribution of apelin and its receptors indicates that it can be involved in many physiological processes including blood pressure regulation, body fluid homeostasis, endocrine stress response, angiogenesis, cardiac contractility kidney disease, and others [7], whereas, the production of apelin peptides by cleavage of the C-end of the precursor of the 77-amino acid preproapelin, the peptide segments are of vary lengths and spread in vivo, as the main isoforms are (Apelin-36, apelin-17, apelin-13) Represent the main asian forms [8].

B2- migroglobulin (B2-MG):

B2 - migroglobulin is a non glycosylated polypeptide weighting (11.8KD), present in all cells with a nucleust [9], this protein found on the surface of nucleated cells, it works as part of the human immune system, as it is synthesized and secreted by many cells, especially lymphocytes [10]. This protein is routinely released into the blood at a constant rate by cells and is present in most fluids in the body, it is at its highest level in the bloods, and is found at a lower level in the lower level in the cerebrospinal fluid in general, while it is found in negligible levels in the urine, in the kidney B2-migroglobulin (B2-MG) passes through the blood filtering units known as the glomeruli and is then completaly reabsorbed and catabolized by the near by Kidney tubules, which are known as the structures that retrieve waters proteins, vitamins, minerals and other vital substances, often, there are very small amounts of (B2-MG) in the urine, but if the kidney, tubules are damaged injured, the concentration of (B2-MG) in the blood rise as a result of the decreased ability to re-absorb this (B2-MG) protein, as the glomeruli in the kidney [11]. B2- migroglobulin protein levels may be associated with the risk of cardiovascular disease and may be the case of death in patients with kidney disease or dialysis. Also, an increase in the level of (B2-MG) protein in people exposed to high levels of cadmium of mercury may indicate early impairment of kidney function [11].

B2-MG can be long protein chains that can deposit in joints and tissues, Causing stiffness and pain. This Condition is called B2-M dialysis - associated amyloidosis and occurs in people with kidney disease [11]. B2-MG levels predict the development of acute kidney injury (AKI), as the hypothesis that (B2-MG) is an independent predict of the development of (AKI) and its out come in patients with intracerebral hemorrhage (ICU) in the neurotransmission (NICU) were explored to [12]. B_2- MG is not affected by Sex of age, as a low molecular weight protein known as a thiotoxin [13]. Where it is released into the circulation, these propeties may make B2-microglobulin in ideal endogenous biomarker for estimating glomerular filtration rate [14].

Fibroblast growth factor 23 (FGF23):-

It is a glycoproteins (32 KD) containing 251 amino acids [15], and is a member of the fibroblast growth family (FGF), which is involved in the regulation of plasma phosphate, and vitamin D metabolism. In humans, it is encoded by the FGF-23 gene. It reduces phosphate reabsorption in the kidney mutations in FGF-23 Can increase its activity and thus lead to causing rickets which results in a decrease in the level of phosphate (hypophosphatemic), it also leads to neoplastic to calcification [16]. The main regulators of FGF-23 levels are calcitriol, parathyroid hormone, and dietary phosphate [17]. Where it was found that bone-derived fibroblast growth factor 23 (FGF 23) is on important

regulator of endocrine activity of mineral homeostasis [18], where this hormone works in concert with parathyroid hormone (PTH) and 1,25 $(OH)_2$, vitamin D (vit D) on phosphate and calcium homeostasis, where elevated levels of phosphate and vitamin D lead to the production of FGF-23 in bone, which acts to circulate to the kidney through its binding with FGF receptors and enhance phosphorylation and reduce turnover Vit D levels [19].

As early as 2010, circulating levels of FGF-23 were increased in acute kidney injury (AKI) [20]. In recent years FGF-23 has been found to be positively associated with the risk of rheumatoid arthritis and morality in critically ill patients [21]. where it was found that high levels of the hormone can predict or detect in acut kidney injury [21].

It was found that patients who recover from acute kidney failure have 25% increased risk of developing chronic kidney disease (CKD) and a 50% increase in mortality after the disease was followed up for about 10 years [18]. It also found that circulating FGF-23 concentrations increase very rapidly in acute kidney impairment, in addition to that it precedes other mineral markers and traditional measures of kidney function [22]. Blood phosphat levels will rise which subsequently leads to the secretion of FGF-23. In addition, high phosphate, levels. are present in acute kidney injury (AKI) [22, 23].

Estimated glomerular filtration rate (eGFR):-

Is usualy associated with decreased kidney blood flow. Inflammation is an important additional component of acute kidney injury (AKI) resulting in extended infection, since injury to kidney cells can be fatel, sublethal injury represents an important component of acute kidney impairment as it is likely to significantly affect glomerular filtration rate and kidney blood flow [24], where the decrease in the glomerular filtration rate (GFR) is considered the defining characteristic of (AKI), as it works to increase the levels of both blood urea nitrogen (BUN) and creatinine clinically, it was found that the ability to measure the glomerular filtration rate very quellickly and with high accuracy at an early stage of acute kidney failure will quickly determine the severity of the injury, which paves way the for drug or early kidney treatment or both [25, 26].

Material and methods

Subjects

During this study, 232 blood samples were colected from males and females 108 of which. were as control group and 124 of which were from patients with acute kidney injury, whose ages ranged from (≤35 years - ≥56 years), During a period from [May 2022 to February 2023], from (Ibn sina, Al-Salam and Al-Jumhuries) teaching Hospitals and the central blood Bank in mosul city, Iraq.

Collection of blood serum samples from patients:

Ten ml of blood were drawn from the subjects Serum samples were separated, and then kept in clean and tightly covered tubes at a temperature of -20C° until use.

Exclusion criteria

The collected samples are free from decomposition and turbidity to avoid any interference with the results, as well as free from viral hepatitis.

Methods

Serum human apelin is determined by using ELISA kit assayed according to the manufactured procedure (Bioassay laboratory technology, Cat. No. EH2174, China) [27].

Serum human B2- Microglobulin (β2_MG) is determined by using ELISA kit assayed according to the manufactured procedure (Bioassay laboratory technology , Cat. No. EHO424, China) [27].

Serum human Febroblast growth factor 23 (FGF 23) is determined by using ELISA kit assayed according to the manufactured procedure (Bioassay laboratory technology, Cat. No. EH 3058, China) [27].

The Estimated glomerular filtration rate was at calculated using the Cockcroft - Gault formula [28].

The study was conducted at the department of life sciences at the university of Mosul, Iraq, in partnership with Al-Ghufran laboratory in Mosul.

Statistical analysis

In the study, the SPSS statistical system was used to analyze the results of the study statistically, as it was study through the (independent T test) analysis program to analyze the results of studies that include two variables, while the completely randumized Design (CRD) was used to analyze the variance of traits that included more than two variables and that through one way analysis of variance, as well as extracting the correlation coefficient for some of the school characteristics, and the correlation factor, as well as extracting the (means), as well as extracting the standard errors, while using Duncan's multiple range test, in order to find the difference between means significant reduction score ($P \leq 0.05$) [29].

Ethical approval

The study was conducted by the ethical principles that have origin in the declaration of Helsinki. It was carried out with patients verbal and analytical approval before sample was taken. The study protocol information and consent form were reviewed and approved by a local ethics committee according to document number 14459 (including the number and the date on (26/4/2022) to get this approvel.

Results: The results shown in table 1 indicate that there is a significant decrease in the level of apelin at the level of probability (P≤0.05) in patients with acute kidney injury (148.600 ± 11.67 pg/ml) compared to control group (17777.99 ±88.14 pg/ml). The result in table 1 also show that there is a significant increase in the level of B2-microglobulin (β2-MG) at the level of probability (P≤0.05) in the blood serum of acute kidney injury (18.74 ± 1.75 ng/ml) compared to control group (11.18 ± 1.63 ng/ml) Also, the results in table (1) indicate that there is a significant increase at the level of probability (P≤0.05) in the level of FGF 23 in patients with acute kidney injury (360.22 ± 16.13 pg/ml) compared to control group (24.90 ± 1.39 pg/ml), also a significant decrease was found at the probability level (P≤0.05) in the level of estimated glomerular filtration rate (eGFR) (48.22 ± 3.97 ml/minute) in patients with acute kidney injury compared to control group (155.33 ± 2.85 mL/minute).

Table (1): Level of biochemical parameters measured in the blood serum of patients with acute kidney injury compared to control group.

Biochemical parameters	Control groups N=108 Mean ± SE	Patients group N=124 Mean ± SE
Apelin (pg/ml)	17777.99*± 88.14	148.600± 11.6 7
β_2-MG (ng/ml)	11.18*± 1.63	18.74± 1.75
FGF 23 (pg/ml)	24.90*± 1.39	360.22± 16.13
eGFR (ml/minute)	155.33*± 2.85	48.22±3.97

*The difference is significant horizontally at the level of significant (P≤0.05)

Correlation between apelin and brochemical parameters measured in the serum of patients with acute kidney injury:

The relation of apelin with the biochemical variables measured in the blood serum of patients with acute kidney injury and group and control group shown in table (2).

Table 2: the linear correlation between apelin and biochemical parameters measured in the two groups of control and acute kindrey injury patients.

Clinical parameter	Apelin (pg/ml)			
	Control group Mean ±S.E		Patients group Mean ±S.E	
	r-value	p-value	r-value	p-value
β_2-MG (ng/ml)	-0.007	0.007	-0.126	0.163
FGF 23 (pg/ml)	-0.195**	0.043	-0.103	0.254
eGFR(ml/minute)	0.329*	0.001	0.300*	0.001

**Significant at (p≤0.05)

* significant at (p≤0.01)

Discussion:

The reason for the decrease in the level of apelin is attributed to increases in plasma osmolality were accumpained by a simultaneous increase in plasma arginine vaso paress in levels (AVP) and a decrease in plasma apelin levels, Thus, there is an abnormal apelin / AVP balance in acute kidney injury patients, which leads to higher levels of water and reabsorption of water by the kidney tubules. Decreased watery diuresis and thus leads to an elevation of (AVP) and a decrease in apelin levels [30], the reason for the increase in the level of β_2-MG is consistent with what was found [9], that levels of β_2-MG predict acute kidney injury since it is a protein low molecular weight (11.8 KD). it is released at a steady rate into the circulation as well as then freely filtered by the glomeruli. It is completely absorbed and catabolized in the renal tubules. These properties make it an ideal endogenous biomarker for the estimation of glomerular filtration rate [14], and thus an early indicator for patients with acute renal insufficiency. The reason behind the increased in the level of (FGF 23) in acute kidney injury patients compared to the control group may be multiple factors according to [31], including increased production in the bone and decreased elimination, but not vitamin D or parathiroid hormone (PTH) - activated both ways, were early work showed that (FGF 23) acts as a hormone on the kidney.

To increase phosphate secretion and reduce (25-hydroxy) vitamin D activating enzyme; 1-alpha hydroxylase [32]. The reasion is a ttributed to the decrease in the estimated glomerular filtration rate (eGFR) in the case of acute kidney injury (AKI), which is mainly caused by several causes, and the main causes are inchemia, hypoxia, or nephotoxicity, which results in a lower (eGFR) usually associated with decreased renal blood flow, inflammation is an important additional component of (AKI) as it leads to an extended stage of injury, which may be consistent with insensitivity to vasodilator therapy, as the main underlying cause of renal injury could be impaired energy of the highly, metabolically active nephron segments (ie proximal tubules and thick ascending tips), in the external renal medulla, which can lead to a shift from transient hypoxia to intrinsic renal failure, and this is consistent with what was stated by [33].

In the table (2) It was found that there is anon - significant inverse relationship between the level of β_2-MG and the level of apelin in both groups of patients and control groups, and the results also showed a non-significant inverse relationship between the level of FGF 23 and the level of apelin in the group acute kidney injury patients, while the results showed a significant inverse relationship at the level of probability ($P \leq 0.05$) between the level of FGF23 and the level of apelin in the control group. The reason is that the association between apelin and (eGFR) is positive and significant at the probability level ($p \leq 0.05$), the concentration of apelin decreases in acute kidney injury and is associated with a decrease in the estimated glomerular filtration rate (eGFR) [34].

Conclustion

The results showed the apelin and (eGFR) decreased in the blood serum of acute kidney injury group, corresponding to an increased in the level of both β_2-MG and FGF23 Compared to the control group, which indicates the possibility that apelin may be an indicator of early provention and protection for acute kidney injury, in addition to β_2-MG and FGF23.

Financial Support and sponsorship

NIL.

Conflicts of interest

There are no Conflicts of interest

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