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Original Research Article

Cystatin C As Marker for Detection of Renal Function in Comparison to Blood Urea and Serum Creatinine in Patient with Obstructive Uropathy.

Zuhair A. AL-Bayati Mufeed J. Ewadh* Emad H. AL-Jaff

College of Medicine, University of Babylon, Hilla, IRAQ

* E-mail: mewadh@yahoo.com

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Abstract

Diagnoses obstructive uropathy is usually based on changes in serum Creatinine, which is a poor marker of early renal dysfunction, instead used Cystatin C for this purpose. This study aimed to compare the efficacy of Cystatin C with Creatinine and urea in serum to diagnosis uropathy. The current study was preformed (50) patients (34 males and 16 females). Admitted to Al-Hilla teaching Hospital. Control group include (39) healthy person (21 males and 18 females) to measure kidney biochemical measurement including (Cystatin C, Creatinine and urea in serum). In present study results showed the rate of male more than female with non-significant relation at (P value >0.173) between patients and control groups, Obstructive Uropathy found 52% due to ureteric stone in male and female, 28% due to BPH and the other causes followed in different percentage. According to the kidney biochemical tests, results shows there are non-significant correlation between (cystatin C- Creatinine), and (Cystatin C-Urea). The sensitivity and specificity of Cys C marker were 90% and 97,43% respectively. According to S. Cr the sensitivity and specificity were 28% and 94.87% respectively. We concluded Cystatin C has been a more sensitive marker in detection of renal function in obstructive uropathy than Creatinine and urea in Serum.

Key Words: Obstructive Uropathy, Cystatin C, Creatinine and urea.

سيستاتين سي كعلامة للكشف عن وظيفة الكلى في مقارنة مع الكرياتينين واليوريا في مصل الدم للمرضى المصابين بالاعتلال الانسدادي

الخلاصة

تشخيص الاعتلال الانسدادي عادة ما يعتمد على التغيرات في مصل الكرياتينين، الذي هو علامة ضعيفة من القصور الكلوي في وقت مبكر. استخدم سيستاتين C بدلا من ذلك. هدفت هذه الدراسة إلى مقارنة فعالية المصل سيستاتين C مع مصل الكرياتينين واليوريا في تشخيص المرضى الذين يعانون من الاعتلال الانسدادي. اجريت الدراسة الحالية على خمسون مريض مصاب بالاعتلال الانسدادي (٣٤ الذكور و١٦ الإناث). اثناء دخولهم مستشفى الحلة التعليمي. وتشمل مجموعة السيطرة (٣٩) شخصا أصحاء (٢١ من الذكور و١٨ من الإناث). واستخدمت القياسات البيوكيميائية بما في ذلك (فحص السستاتين سي، والكرياتنين واليوريا في مصل الدم). أظهرت الدراسة الحالية أن معدل الاعتلال الانسدادي لدى الذكور أكثر من الإناث مع وجود علاقة غير معنوية عند (P. value> 0.173) بين المرضى والاصحاء، وجد أن نسبة الاعتلال هي ٥٢٪ بسبب انسداد الحالب في الذكور والإناث، و٢٨% بسبب ورم البروستات الحميدي وبقية الامراض كانت نسب قليلة مختلفة. أظهرت النتائج وجود علاقة غير معنوية بين السيستاتين سي والكرياتينين، وكذلك بين السيستاتين سي واليوريا. وكانت حساسية وخصوصية علامة السستاتين سي ٩٠٪ و٩٧،٤٣٪ على التوالي. وفقا لحساسية وخصوصية الكرياتنين كانت ٢٨٪ و ٩٤،٨٧٪ على التوالي. نستنتج ان فحص السستاتين سي في مصل الدم اكثر حساسة في الكشف عن وظيفة الكلى في الاعتلال الانسدادي من الكرياتينين واليوريا في مصل الدم.

الكلمات المفتاحية: الاعتلال الانسدادي، السيستاتين سي، الكرياتينين واليوريا.

Introduction

Obstructive uropathy is one of the most urgent clinical entities that both nephrologists and urologist have to diagnose [1]. Epidemiologically, obstructive uropathy accounts for 10% of the causes of acute renal failure and 4% of the cases of

chronic end stage renal failure [2]. It is classified on the basis of several criteria, including the degree, duration, site of obstruction and whether it is "bilateral or unilateral." The degree of obstruction prefers to whether the obstruction of the urine flow is partial or complete. Regarding the duration of the obstruction, obstructive uropathy is categorized in acute and chronic. Acute obstruction occurs for short period of time and therefore renal parenchyma lesions are mostly reversible, while chronic obstruction, after several weeks, causes permanent damage [3]. This obstruction may be due to intraluminal, intramural, and extramural causes. Renal calculi are the main etiological in young and middle aged patients, in female gynecological tract obstruction surgery and obstetrical trauma and in "old people malignancy contributes to upper obstructive uropathy"[4].

Serum Creatinine (Scr) has been widely used as a marker of renal function, but it is lacking enough sensitivity [5]. Consequently, early diagnosis of renal dysfunction is a major clinical challenge. Now various plasma low molecular weight proteins have been suggested to be of effective diagnostic value for decreased renal function instead of Scr [6, 7].

Among these markers, cystatin C was proposed as a new biomarker for the evaluation of renal function [8]. Serum cystatin C is a cysteine proteinase inhibitor with a low molecular weight "13 kDa", which is produced at a stable rate by all nucleated cells. It is freely filtered through the glomerular filtration membrane, and the filtration rate appears to be unaffected by external factors "e.g. muscle mass or meat intake"[7]. "Multiple studies have been performed to investigate" the accuracy of serum cystatin C for /assessing renal function [9, 10] and several pooled-analyses have evaluated the use of cystatin C to estimate GFR [11-13]. The aim of this study is to evaluate the efficacy of CystC in determine the kidney function in comparison with serum Creatinine and serum urea.

Materials and Methods:

In this study the Samples were collected from fifty patients 34 (68.0%) were male, and 16 (32.0%) were female aged ranging from 15-75 years, have been admitted to Al-Hilla Teaching Hospital, Urology Department. during the period August 2016 to January 2017. Thirty-nine apparently healthy individuals were taken as a control group. This group comprises of 21 (53.8%) males, and 18 (46.2%), females, age ranging from 15-70 years.

Samples Collection: 5 ml of blood were obtained from patients and controls, then collected in tube without anticoagulants and were left for 15 minutes at room temperature to clot. After that, the blood samples were centrifuged at 1000-2000 ×g approximately 15 minutes. Then the sera were aspirated and stored at (-20°C) until time of tests were done. All test had been performed on serum in biochemistry department in the College of Medicine/University of Babylon. Blood samples have been collected from patients and control subjects.

Blood samples were drawn with tourniquet. Clean and sterile vials without any anticoagulant have been used to collect (5) ml of blood sample in each tube. The blood has been allowed to clot and then centrifuged (1000×g for 10-15). Sera were separated, divided into four parts in sterile eppendrofs and frozen at -20°C until time of use.

We excluded patients with diabetes mellitus, hypertension, smoking and rheumatologic disease. Pregnant from the study group. All patients under went history and physical examination include: age, gender, family history of obstructive uropathy, past history of recurrent kidney diseases. The patients underwent ultrasonography (US), plan abdominal X-ray. Film of kidney, ureter and bladder (KUB), and CT scan. The serum Cyst. C assay, used Human CST3 (Cystatin C), ELISA Kit (BioSource/USA) in the present study, serum Creatinine was measurement via a modified "Jaffe method" with protein precipitation and the Kit company (BioLabo/France) [14]. Serum urea was measured using the kinetic urease method, the urea kit company is (Bioscience/ Germany) [15].

Data analysis:

Data entry and analysis was done using SPSS version 18 computer software (statistical

package for social sciences), categorical variables were presented as frequencies and percentages, continuous variables were presented as mean and standard deviation. Pearson chi square was conducted to determine the association between categorical variables and t-test was also used to determine the mean differences between groups. In addition correlation between continuous variables was carried out also. P value of ≤ 0.05 was considered as statistically significant.

Result and Discussion:

Cross Sectional study was done to randomly assessment of certain parameters among a group of patients (50 member) having obstructive uropathy as well as (39) apparently healthy as control group.

Obstructive uropathy is one of the commonest urological emergencies with incidence of 20%. This condition occurs due to any obstruction to urine flow, resulting in increased pressure within the collecting system, pain, infection, sepsis, and loss of renal function. This potentially life threatening condition requires immediate measures to divert the urine from obstructed kidney [16]. This obstruction may be due to intraluminal, intramural and extramural causes [17].

The present study targeting a convenient sample of patients and control at different age matched groups. The results were distributed according to different studied parameters such as the following:-

Relation of Gender with obstructive Uropathy:

This study showed 34 (68%) males and 16 (32%) females, while the control comprised (39) were 21 (53.8%) males and 18 (46.2%) females as shown in (Table 1). There is no significant relation at (P value > 0.173) between patients and control groups.

Table (1): Association between gender and study groups (N=89).

Sex	Study groups		Total	X^2	P value
	Patients Number(%)	Control Number(%)			
Male	34 (68.0 %)	21(53.8.0%)	55(61.8%)	1.859	0.173
Female	16 (32.0%)	18 (46.2%)	34(38.2%)		
Total	50 (100%)	39 (100%)	89(100%)		

*P value≤ 0.05 was significant

Obstructive uropathy in male higher than female due to more incidence of stone disease, BPH, carcinoma of bladder in males and due to anatomy of male ureter which is longer than ureter of female, caused outer bladder obstruction.

Karim *et al* [18], who found higher incidence of male than female patients.

Also the results were similar to the findings of other studies; Ayekpam *et al* [19], Apoku *et al* (2015) [20] and Guest *et al* [21].

Relation of Age with obstructive uropathy:

Regarding age group distribution, the results in Table 2, shows the age of patients group.

were 50 patients, 11 (22%) were between (15-29) years of age, while 7 (14%), 6 (12%) and 15 (30%) were in their (30- 44), (45- 59) and (60-74) age group respectively. 11 (22%) were aged above 75 years old. Statistically there was highly significant mean difference between patients and healthy control (P<0.001).

Variables	No. of patients	%
`Age in years		
(15-29)	11	22%
(30- 44)	7	14%
(45- 59)	6	12%
(60-74)	15	30%
➢ **75**	11	22%
Total	50	100%

Table 2: Age distribution of patients with obstructive uropathy

P <0.001 was significant

The highest rate of male obstructive uropathy in our study is similar to the finding observed in the study done by Katakwar (2017), Who found that from total of 100 patients the rate of male higher than female in which was 94% and 6% respectively, due to bladder outlet obstruction [22].

The mean age of the study patients was 53.48 ± 21.66 years (mean ± SD), while that of control was 39.21±13.99 years, as shown in table 3.

The obstructive uropathy in our study found have been started in the age above 53 years, this result is in agreement with a cross-sectional study done in shiraz Iran by (Sagheb *et al* (2014*)* [23], who found that the mean standard deviation age in their study was 45.14±18.16 years. Also by (Shukla *et al* 2017) [24], Who found the mean age of patients was 56.54±10.04 years with majority of the population were male (81.42%).

Table (3): Mean difference of age of the respondents according to study groups.

Variable	Study group	No.	Mean±SD	t-test	P-value
Age(year)	Patients	50	53.84±21.66	3.855	<0.001*
	Control	39	39.21±13.99		

Causes of obstructive uropathy:

Depending on the causes of obstructive uropathy was found highest due to; ureteric stone, benign prostatic hyperplasia (BPH), urethral stricture, post-trans urethral resection (TUR) Urethral stricture, and vesico-ureteric reflux (VUR) respectively as summarizes in Figure 1. The figure shows that three quarters of the causes of obstructive uropathy in respondent patients are ureteric stone (52%) and benign proststic hypertrophy (28%), while the other quarter are due to VUR (2%), urethral stricture (6%), PUJ stone (4%), bladder tumour (6%) and (2%) for post-TUR urethral stricture.

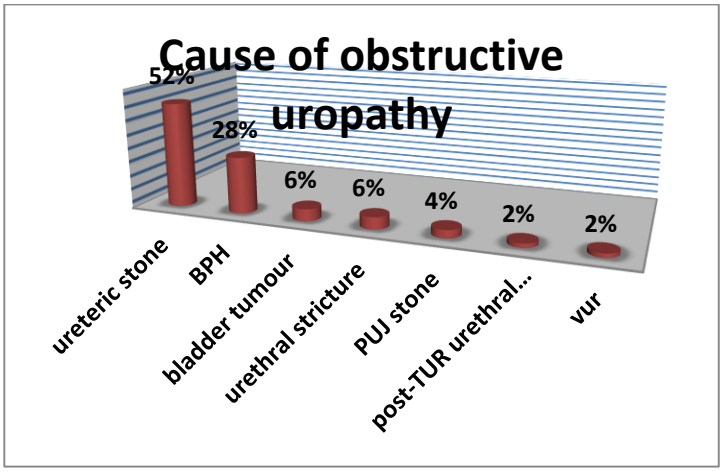

Figure (1): Causes of Obstructive uropathy

(*) (vur: vesico-ureteric reflux, BPH: benign prostatic hypertrophy. PUJ: pelvi-ureteric junction. TUR: trans-urethral resection.

Obstructive uropathy due to ureteric stone in this study found to be 52% higher percentage than other causes. In this study the most common cause of ureteric obstruction was the ureteral stone. This finding was in agreement with Shakeir *et al* [25] who reported that ureteral obstruction is usually a consequence

of nephrolithiasis which is the most common cause of urinary obstruction.

The incidence depends on geographical, climatic, ethnic, Dietary, fluid intake and genetic factor [26]. The recurrence risk is basically, determined by the disorder or disease causing the stone formation [27].

In addition to dietary habitat which may High intake of proteins among male patients [28]. The endogenous estrogen and estrogen treatment in postmenopausal women may decrease the risk of stone recurrence by lowering urinary calcium and calcium oxalate saturation. Estrogen may also help to prevent the formation of calcium stones by raising protective citrate levels. Experiments in animals demonstrated that testosterone promoted crystal growth by suppressing osteopontin expression in the kidney and increasing urinary oxalate excretion while estrogen possibly inhibited stone formation by increasing osteopontin expression in the kidney and decreasing urinary oxalate excretion [29]. The lower serum testosterone level may contribute to some of the protection women and children have against oxalate stones. This factor could lead to the higher incidence of urinary stones cases in males than females observed in same study [30].

Benign prostatic hyperplasia (BPH) in this study found to be 28% above 45 years old age group. It is a very common disorder age-dependent with initial development usually after 40 years of age [31].

This study finding is in agreement with a cross sectional study done in Cameroon, which found that main etiologies of obstructive uropathy was 35% urolithiasis and 27% benign prostatic hyperplasia [32].

A study done in Iraq by Al-Saadi. Illustrate the elevation of biochemical and immuno-logical parameters in BPH elderly patients who had significantly higher risk due to an obstructive uropathy, which the major public health problem among men especially over 55 years [33].

Chronic kidney disease has been consistently proved to be a significant risk factor for bladder cancer in the population, because of kidney function alteration and that inflammation would stimulate the cellular proliferation [34].

The percentage of obstructive uropathy due to urethral stricture and bladder tumor was 6%, while due to pelvi-ureteric junction 4%, vesico-ureteric reflux (VUR) and post-trans-urethral resection was 2% similar finding was observed in the studies of the other workers Katakwar *et al* [35], Alosta [36], Halle *et al* [37].

Estimation of kidney function test.

In the presented study, we primarily aimed to determine the utility of serum Cys C in compare with serum Creatinine and blood urea to detect renal function in obstructive uropathy. According to Table 4 results shows there are significant differences at (P-value < 0.05, P value < 0.01) of Cystatin C and blood urea respectively represented as mean ±SD. While there are no significant difference (P-value = 0.093) of serum Creatinine by study group.

Table 4: Mean difference of cystatinC, creatinine, urea, serum K, serum Na and TSH according to study groups (N=89)

Variable	Study group	N	Mean±SD	t-test	P-value
Cystatin C (mg/l)	patients	50	2.63±0.72	15.937	<0.001*
	Control	39	0.87±0.26		
Creatinine (mg/dl)	patients	50	1.23±1.07	1.713	0.093
	Control	39	0.96±0.16		
Urea (mg/dl)	patients	50	38.34±24.25	2.591	0.012*
	Control	39	28.92±7.51		

Beegum *et al* found that cystatin c has important association with sensitivity, early detection and accurate serum marker than serum Creatinine [38].

villa *et al* reported that cystatin c is better than serum Cr for assessing critically ill patients in which, only 20% of patients were found to have elevated S. Cr. level, whereas 76% of them had elevated S CystC level [39].

Creatinine production changes significantly according to the muscle mass of the body, age and gender; while Cyst C not affected by age, gender and body mass [40].

There was a strong association of S. Cys C with age in compare with serum Creatinine, since CysC doesn't cross the placental barrier as creatinin, which comes from both mothers and Newborn NB [41]. It is found that CysC in higher at (NB) and after 1 years of age the values remain constant until approximately age (70), when there is a gradual age-related decline in GFR and a corresponding increased CysC. In contrast, Creatinine values increase gradually throughout childhood as body mass increases, and there is a wide inter-individual range for Creatinine [42]. In contrast to serum Cr, serum Cys C does not correlate with body weight or fat free mass or level of physical activity [43].

Cystatin C is freely filtered from glomeruli; nearly all is reabsorbed and metabolized by the proximal tubular cells. Therefore, Cys C seems to be a better surrogate marker of GFR than serum Cr when its cellular production was accepted to be constant [44].

Cystatin c provides its greatest utility in the detection of both acute and chronic kidney disease [45].

Many studies over the past several years which supports the use of Cystatin C as an alternative and more sensitive endogenous marker for the estimation of GFR than Serum Creatinine and Serum Creatinine based GFR estimations [46].

Table (5): Measuring of cystatin C test using ELISA method.

Cystatin C	Obstructive uropathy		Total
	patient	control	
Elevated	45	1	46
Normal	5	38	43
Total	50	39	89

Cystatin C as marker for kidney function.

Sera of (45) patients with obstructive uropathy were positive for cystatin C analysis; while (5) of them false negative. Control group represented (39) healthy persons, the result of analysis were (38) true negative and 1 of them were false positive, table 5 Serum Cys C showed a faster elevation in patients at early stages of obstructive uropathy compered to serum blood urea and serum Creatinine and may considered as a screening test for detection [40].

Table (6): The characteristic of ELISA test as compare to clinical diagnosis.

Sensitivity	Specificity	Accuracy	PPV	NPV
90%	97.43%	93.25%	97.82%	88.37%

The sensitivity 90% and specificity 97.43% for detection obstructive uropathy, as seen in the table (3-6). According in the table (3-6); the accuracy was 93.25%, positive predictive value 97.82% and negative predictive value was 88.37%. Cystatin C Identifying an endogenous marker of renal function with appropriate accuracy is an urgent demand. The results of a meta-analysis on 13 studies demonstrated that serum cystatin C appears to be a good biomarker for prediction of Acute Kidney Injury (AKI) development both overall and across a range of subgroups [47]. In the current study, we examined the hypothesis that serum CystC is more accurate than serum creatinine for detection of early AKI, defined as GFR < 80 mL/min/1.73 m^2, in critically ill patients.

Estimation serum creatinine test:

In this study sera of patients with obstructive uropathy were the result of creatinine analysis gave (14) seropositive; while (36) of them false negative. Control group represented (39) healthy persons, the result of analysis was (37) true negative and (2) of them were false positive, Table 7.

Table (7): Measuring serum Creatinine test using colorimetric method.

Creatinine	Obstructive uropathy		Total
	patient	control	
Elevated	14	2	16
Normal	36	37	73
Total	50	39	89

Even though serum Creatinine determination remains the most commonly used renal marker for estimation of GFR, these include the fact that measurement of GFR by Creatinine is influenced by multiple non-renal factors including gender, muscle mass and tubular secretion which can result in an overstatement of GFR up to 20%. Unlike Creatinine, cystatin c serum levels are virtually unaffected by age (1years), muscle mass, gender and race. Multiple studies have found cystatin c to be more sensitivity to actual change in GFR in the early stages of kidney disease than Creatinine based GFR estimation [48].

The sensitivity 28% and specificity 94.87% for detection obstructive uropathy, as seen in the table 8. According in the table 8; the accuracy was 57.30%, positive predictive value 87.50% and negative predictive value was 50.68%. Serum Creatinine remains in the normal range until 50% of renal function is loss [49].

Evaluate the diagnostic value of S. Cr. and S. Cys C to detect the more reliable marker for detection of renal function.

Our analysis showed that S. Cys C was a favorable marker than S.C this finding is in agreement with Garlipp *et al* (2008), a study carried on (82) patients from (5) to (80) years (median, 44 years) with diagnostic renal diseases they confirmed that Cystatin C appears to be an efficient and a sensitivity marker for kidney function (r = 0.82, sensitivity=100%, Specificity= 75%, efficiency = 95%) [50].

Yang *et al* (2016) [51], their study was conducted according to the guide-line of Meta-analysis of observational studies on (17) (for S. Cys C) and (12) (for S. creatinine) published studies respectively, the pooled sensitivity and specificity of serum Cystatin C for renal dysfunction were 95% respectively. Their results indicated that serum Cystatin C is an effective index in diagnosing renal dysfunction comparing serum creatinine, and more sensitivity for evaluation of renal dysfunction patients.

Correlation Between Cystatin C and serum Creatinine in patients group.

Statistically there is no significant between cystatin c and serum creatinine (P value =0.371), r =-0.12.

Figure 2 shows that there is no significant correlation between creatinine and cystatin C. The present study was comparable with Sagheb (2014) [23], who found that significant correlation between false negative rates was 95.33% and 80% for S. creatinine and S. Cys. C respectively.

Colombian Narvaez-Sachez *et al.* [52]. found that Cystatin C is a very interesting marker, and could be a replacement to Serum Creatinine for diagnosing and follow up kidney function in children.

Several studies have reported the superior diagnostic accuracy of serum cystatin C.

based formulae over other markers in detecting mild and sever renal impairment in patients with liver cirrhosis, contrast induced nephropathy [53].

Table (8): The Characteristic of Colorimetric Methods Test as Compare to Clinical Diagnosis.

Sensitivity	Specificity	Accuracy	PPV	NPV
28%	94.87%	57.30%	87.50%	50.68%

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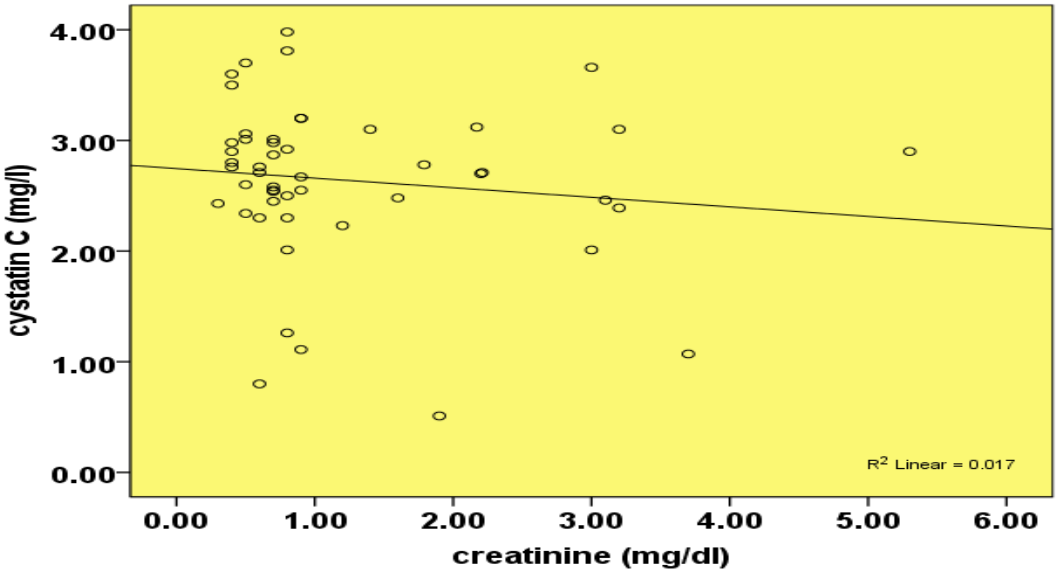

Figure (2): Correlation Between Cystatin C and serum Creatinine in patients group.

Correlation Between Serum Cystatin C and Blood Urea.

Figure 3 shows that there is P value = 0.073, r = 0.191.

Until now, traditional measures for evaluation of renal function such as measuring S. Cr and S.U have been widely used, although they diagnostic efficacy of using serum cystatin c level and compared these results to those obtained.

In our found the S. Cys C has been more accuracy in assessment of renal function in general population, than S. Cr and S. U respectively.

Although serum creatinine has become the most used serum marker; may be unreliable because it is frequently affected by protein intake, age, gender, ethnicity and muscle mass [54]. and because Creatinine synthesized in the liver, any cause of hepatic parenchymal dysfunction will directly reduce creatinine production [55].

Thus any injury that impaired with GFR lead to slowly increase in the level of S. Cr acute deterioration of renal function, thus, the serum level can be expected to rise slowly until reflected in an elevated level and that is require 24-48 hours [56].

In contrast to Cys C, is secreted by all nucleated cells at a constant rate, low molecular weight and positive charged at physiological PH are the factor facilitating its glomerular filtration. Another advantage of S. Cyst C. that is not affected by age, sex, diet and muscle mass, therefore any elevation in the serum level may be detect more rapidly [57].

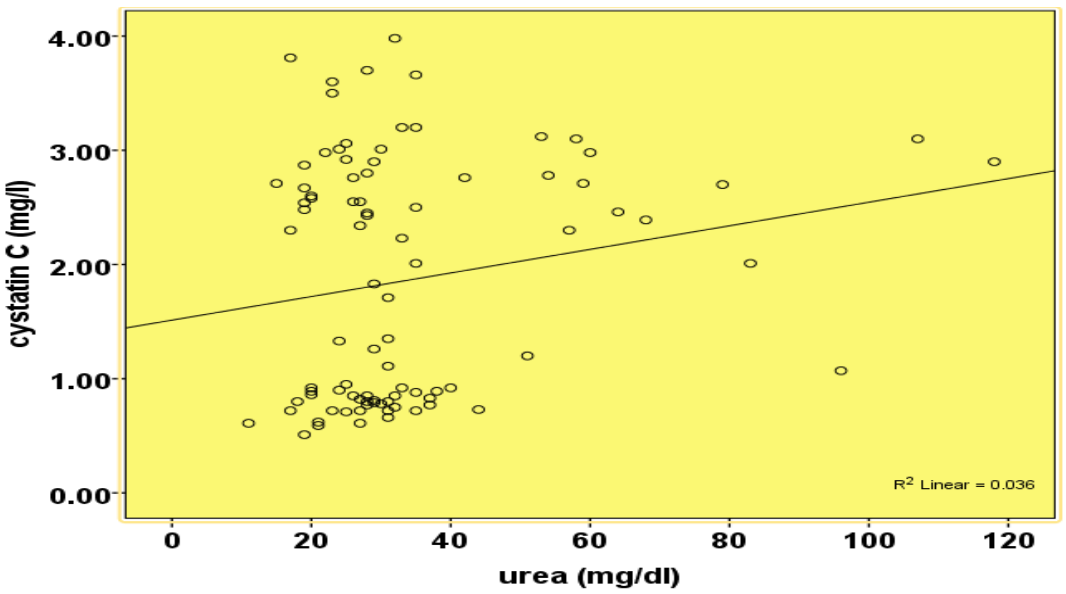

Figure (3) : Correlation of Urea and cystatin C of the patients' group.

Conclusions

1- Serum cystatin c is a favorable marker for identifying renal dysfunction in obstructive uropathy. Compared with serum Creatinine and serum urea, the diagnostic sensitivity and specificity of serum cystatin C is high.

2- Serum Cys C. is the best measures that reflect the actual renal performance in obstructive uropathy, also the most accurate one in detecting early stages of renal impairmentin these patients.

3- Cystatin c in present study has been identified as a more sensitive marker than Creatinine and serum urea in detection of renal function.

References:

1. Tseng, T. Y., & Stoller, M. L. (2009). Obstructive uropathy. *Clinics in geriatric medicine*, *25*(3), 437-443.
2. Siddiqui, M. M., & McDougal, W. S. (2011). Urologic assessment of decreasing renal function. *Medical Clinics of North America*, *95*(1), 161-168.
3. Samodai, L., & Andualem, D. (2014). Pattern and Outcome of Surgical Management of Postrenal Acute Renal Failure Over Three Years Period at Tikur Anbessa Specialized Hospital. East and Central African Journal of Surgery, 19(3), 65-69.
4. Ahmad, I., Pansota, M. S., Tariq, M., & Shahzad, M. (2014). Complication Of Percutaneous Nephrostomy (Pcn) In Upper Obstructive Uropathy: Our Ecperience.
5. Filler, G., Yasin, A., & Medeiros, M. (2014). Methods of assessing renal function. Pediatric nephrology, 29(2), 183-192.
6. Lim, W. H., Lewis, J. R., Wong, G., Teo, R., Lim, E. M., Byrnes, E., & Prince, R. L. (2015). Plasma neutrophil gelatinase-associated lipocalin and kidney function decline and kidney disease-related clinical events in older women. American journal of nephrology, 41(2), 156-164.
7. Onopiuk, A., Tokarzewicz, A., & Gorodkiewicz, E. (2015). Chapter Two-Cystatin C: A Kidney Function Biomarker. Advances in clinical chemistry, 68, 57-69.
8. Shlipak, M. G., Matsushita, K., Ärnlöv, J., Inker, L. A., Katz, R., Polkinghorne, K. R., ... & Levey, A. S. (2013). Cystatin C versus creatinine in determining risk based on kidney function. New England Journal of Medicine, 369(10), 932-943.
9. Allen, A. M., Kim, W. R., Larson, J. J., Colby, C., Therneau, T. M., & Rule, A. D. (2015). Serum cystatin C as an indicator of renal function and mortality in liver transplant recipients. Transplantation, 99(7), 1431.

10. Woo, K. S., Choi, J. L., Kim, B. R., Kim, J. E., & Han, J. Y. (2014). Clinical usefulness of serum cystatin C as a marker of renal function. Diabetes & metabolism journal, 38(4), 278-284.
11. Pan, Y., Hu, B., Li, M., Fan, L., Ni, Y., Zhou, J., & Shi, X. (2014). A Meta-analysis on diagnostic value of serum cystatin C and creatinine for the evaluation of glomerular filtration function in renal transplant patients. African health sciences, 14(4), 1025-1035.
12. Zhang, M., Cao, X., Cai, G., Wu, D., Wei, R., Yuan, X., ... & Chen, X. (2013). Clinical evaluation of serum cystatin C and creatinine in patients with chronic kidney disease: a meta-analysis. Journal of International Medical Research, 41(4), 944-955.
13. Roos, J. F., Doust, J., Tett, S. E., & Kirkpatrick, C. M. (2007). Diagnostic accuracy of cystatin C compared to serum creatinine for the estimation of renal dysfunction in adults and children—a meta-analysis. Clinical biochemistry, 40(5), 383-391.
14. Tietz, N. W. (1995). Clinical guide to laboratory tests. WB Saunders Co.
15. Wu, A. H. (2006). Tietz Clinical Guide to Laboratory Tests-E-Book. Elsevier Health Sciences4 ed., p. 316-321.
16. Sood G, Sood A, Jindal A, Verma DK, Dhiman DS. Ultrasound guided percutaneous nephrostomy for obstructive uropathy in benign and malignant diseases. Intl Braz J Urol. 2006;32(3):281-6.
17. Ahmad, I., Pansota, M. S., Tariq, M., & Shahzad, M.,(2014). Complication of Percutaneous Nephrostomy (pcn) in Upper Obstructive Uropathy: our ecperience.
18. Karim, R., Sengupta, S., Samanta, S., Aich, R. K., Das, U., & Deb, P. (2010). Percutaneous nephrostomy by direct puncture technique: An observational study. Indian journal of nephrology, 20(2), 84.
19. Ayekpam, M., Keretsu, T., & Singh, A. K., (2015). Evaluation of Obstructive Uropathy with Computed Tomography Urography and Magnetic Resonance Urography-A Clinicoradiological study. IOSR Journal of Dental and Medical Sciences (IOSR-JDMS), 1(14), 1-5.
20. Apoku, I. N., Ayoola, O. O., Salako, A. A., & Idowu, B. M. (2015). Ultrasound evaluation of obstructive uropathy and its hemodynamic responses in southwest Nigeria. International braz j urol, 41(3), 556-561.
21. Guest AR ., Cohan RH ., Korobkin M . , et al. ;(2001)."Assessment of the clinical utility of the rim and comet-tail signs in differentiating renal stones from phleboliths". AJR Am J Roentgenol;177 (6).P: 1285-91.
22. Katakwar, P., & Thakur, R. (2017). Clinical study and management of bladder outlet obstruction. International Surgery Journal, 4(4), 1272-1275.
23. Sagheb, M. M., Namazi, S., Geramizadeh, B., Karimzadeh, A., Oghazian, M. B., & Karimzadeh, I. (2014). Serum cystatin C as a marker of renal function in critically ill patients with normal serum creatinine. Nephro-urology monthly, 6(2).
24. Shukla, A. N., Juneja, M., Patel, H., Shah, K. H., Konat, A., Thakkar, B. M., ... & Prajapati, J. (2017). Diagnostic accuracy of serum cystatin C for early recognition of contrast induced nephropathy in Western Indians undergoing cardiac catheterization. Indian Heart Journal, 69(3), 311-315.
25. Shokeir, A. A., El-Diasty, T., Eassa, W., Mosbah, A., El-Ghar, M. A., Mansour, O., ... & El-Kappany, H. (2004). Diagnosis of ureteral obstruction in patients with compromised renal function: the role of noninvasive imaging modalities. The Journal of urology, 171(6), 2303-2306.
26. Kalaitzidis, R. G., Damigos, D., & Siamopoulos, K. C. (2014). Environmental and stressful factors affecting the occurrence of kidney stones and the kidney colic. International urology and nephrology, 46(9), 1779-1784.
27. Chae, J. Y., Kim, J. W., Kim, J. W., Yoon, C. Y., Park, H. S., Moon, D. G., & Oh, M. M. (2013). Increased fluid intake and adequate dietary modification may be enough for the successful treatment of uric acid stone. Urolithiasis, 41(2), 179-182.
28. Martin, W. F., Armstrong, L. E., & Rodriguez, N. R. (2005). Dietary protein intake and renal function. Nutrition & metabolism, 2(1), 25.
29. Abbagani S., Gundimeda S. ,Varre S. ,and et al ;(2010).''Kidney Stone Disease:Etology AND Evaluation'' .International Journal of Applied Biology and Pharmaceutical Technolgy.(1).P:175-182.
30. Cavendish M.; (2008). "Kidney disorders". Diseases and Disorders 2 (1st ed.). Tarrytown, New York: Marshall Cavendish Corporation. P:490-3.

31. sharma, A. K. (2015). Benign Prostatic Hyperplasia(BPH) and It's Management in Ayurveda World J. Pharmacy and Pharmaceutical scien. ; 4(11) ; 768-774.
32. Halle, M. P., Toukep, L. N., Nzuobontane, S. E., Ebana, H. F., Ekane, G. H., & Priso, E. B. (2016). The profile of patients with obstructive uropathy in Cameroon: case of the Douala General Hospital. Pan African Medical Journal, 23(1):1-6.
33. Al-Saadi, E. (2013). Study of Some Biochemical and Immunological Parameter in Iraqi Benign Prostatic Hyperplasia and Lower Urinary Tract Symptoms Patients. Kerbala Journal of Phrmaceutical, 5, 24-33.
34. Tseng, C. H. (2011). Diabetes and risk of bladder cancer: a study using the National Health Insurance database in Taiwan. Diabetologia, 54(8), 2009-2015.
35. Katakwar, P., & Thakur, R. (2017). Clinical study and management of bladder outlet obstruction. International Surgery Journal, 4(4), 1272-1275.
36. Alosta, A., Xi, C., & Gordon, K. (2014). Conservative Management of Obstructive Uropathy Secondary to Stones.
37. Halle, M. P., Toukep, L. N., Nzuobontane, S. E., Ebana, H. F., Ekane, G. H., & Priso, E. B. (2016). The profile of patients with obstructive uropathy in Cameroon: case of the Douala General Hospital. Pan African Medical Journal, 23(1).
38. Beegum, M. S., Mohan, V., Kailasanathan, C. P., & Cyrilraj, E. E. (2017). Serum Cystatin C Compared to Serum Creatinine as an Early Marker of Renal Failure. Int. J. Curr. Microbiol. App. Sci, 6(2), 1687-1693.
39. Villa, P., Jiménez, M., Soriano, M. C., Manzanares, J., & Casasnovas, P. (2005). Serum cystatin C concentration as a marker of acute renal dysfunction in critically ill patients. Critical Care, 9(2), R139.
40. İnal, S., Altuntaş, A., Kidir, V., Özorak, A., İlgin, Y., & Sezer, M. T. (2014). Utility of serum creatinine/cystatin C ratio in diagnosis of postrenal acute kidney injury. Journal of research in medical sciences: the official journal of Isfahan University of Medical Sciences, 19(11), 1086.
41. Cruzado, L. B., González, E. P., Martos, Z. M., Guitarte, C. B., Asencio, M. G., Lagares, S. L., & Padilla, J. P. (2015). Serum cystatin C levels in preterm newborns in our setting: Correlation with serum creatinine and preterm pathologies. Nefrología (English Edition), 35(3), 296-303.
42. Gantzer, M. L. (2003). Cystatin C: Analysis and utility in monitoring GFR. Laboratory Medicine, 34(2), 118-123.
43. Baxmann, A. C., Ahmed, M. S., Marques, N. C., Menon, V. B., Pereira, A. B., Kirsztajn, G. M., & Heilberg, I. P. (2008). Influence of muscle mass and physical activity on serum and urinary creatinine and serum cystatin C. Clinical Journal of the American Society of Nephrology, 3(2), 348-354.
44. Finney, H., Newman, D. J., & Price, C. P. (2000). Adult reference ranges for serum cystatin C, creatinine and predicted creatinine clearance. Annals of clinical biochemistry, 37(1).
45. Herget-Rosenthal, S., Bökenkamp, A., & Hofmann, W. (2007). How to estimate GFR-serum creatinine, serum cystatin C or equations?. Clinical biochemistry, 40(3), 153-161.
46. Hojs, R., Bevc, S., Ekart, R., Gorenjak, M., & Puklavec, L. (2006). Serum cystatin C as an endogenous marker of renal function in patients with mild to moderate impairment of kidney function. Nephrology Dialysis Transplantation, 21(7), 1855-1862.
47. Zhang Z, Lu B, Sheng X, Jin N. Cystatin C in prediction of acute kidney injury: a systemic review and meta-analysis. Am J Kidney Dis. 2011;58(3):356-65.
48. Pucci, L., Triscornia, S., Lucchesi, D., Fotino, C., Pellegrini, G., Pardini, E., ... & Penno, G. (2007). Cystatin C and estimates of renal function: searching for a better measure of kidney function in diabetic patients. Clinical chemistry, 53(3), 480-488.
49. Inker, L. A., Schmid, C. H., Tighiouart, H., Eckfeldt, J. H., Feldman, H. I., Greene, T., ... & Coresh, J. (2012). Estimating glomerular filtration rate from serum creatinine and cystatin C. New England Journal of Medicine, 367(1), 20-29.
50. Garlipp, C. R., Bottini, P. V., Queiroz, R. R., & Afaz, S. H. (2008). Estimating glomerular filtration rate by measurement of cystatin C serum concentrations: A comparison with chrome-EDTA clearance. Clinica Chimica Acta, 393(2), 125-127.
51. Yang, S. K., Liu, J., Zhang, X. M., Hu, C., Zhang, W., Sun, L., & Zhang, H. (2016). Diagnostic Accuracy of Serum Cystatin C for the Evaluation of Renal Dysfunction in Diabetic

Patients: A Meta-Analysis. Therapeutic Apheresis and Dialysis, 20(6), 579-587.
52. Newman, D. J. (2002). Cystatin c. Annals of clinical biochemistry, 39(2), 89-104.
53. Swan, S. K. (1997). The search continues—an ideal marker of GFR.;43:913–4.
54. Kim, D. J., Kang, H. S., Choi, H. S., Cho, H. J., Kim, E. S., Keum, B., ... & Yim, H. J. (2011). Serum cystatin C level is a useful marker for the evaluation of renal function in patients with cirrhotic ascites and normal serum creatinine levels. The Korean journal of hepatology, 17(2), 130.
55. McCullough, P. A. (2008). Contrast-induced acute kidney injury. Journal of the American College of Cardiology, 51(15), 1419-1428.
56. Harjai, K. J., Raizada, A., Shenoy, C., Sattur, S., Orshaw, P., Yaeger, K., ... & Stapleton, D. (2008). A comparison of contemporary definitions of contrast nephropathy in patients undergoing percutaneous coronary intervention and a proposal for a novel nephropathy grading system. The American journal of cardiology, 101(6), 812-819.
57. Murty, M. S. N., Sharma, U. K., Pandey, V. B., & Kankare, S. B. (2013). Serum cystatin C as a marker of renal function in detection of early acute kidney injury. Indian journal of nephrology, 23(3), 180.