Cystatin C as Marker for Detection of Renal Function Change in Obstructive Uropathy Due to Ureteric Stone

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Abstract

Objective: To Detection of Ureteric Stone Using Cystatin C marker in Compare with serum Creatinine.

Methods: Between September 2016 to February 2017 at Al-Hilla Teaching Hospital, Urology Department. The study group consisted of 25 healthy subjects as control with age between (20-66)year and 25 patients of Ureteric Stone with age from (20-62)year, the biochemical measurement including human CSTs (Cystatin C), ELISA kit used in the present study and serum Creatinine test used as biochemical diagnostic assessment. Results: Statistical analysis show significant difference (P<0.005) in the mean age (40.64±11.47), 21-30 years was the most influential of these disease compared with the other age groups. The percentages of males and females in patients group 16(64%) and 9(36%) respectively, while in the control group female were 14(56%), and male 11(44%) respectively. In total twenty five patients 60% from the total number were unilateral and 40% bilateral ureteric stone, study reveals that there was a significant increase (P. value<0.001) in the Cystatin C marker between study groups the mean value was (2.56 ±0.81) compared to control group (0.82 ±0.17) with (P. value<0.001). Creatinine level revealed no significant association (p>0.05) between the level in the control and patients. In order to confirm the results of cystatin C levels using ELISA , the characteristic of the technique :Sensitivity (92%) , Specificity (96%).The characteristic of colorimetric methods test as compare to clinical diagnosis in detection of Creatinine :Sensitivity (40%) , Specificity (88%). Conclusion: Cystatin C found to be a more accurate than Creatinine in detection the obstruction due to ureteric stone.

Keywords: Creatinine, Stone, Cystatin C, Biochemical study, Urinary Tract.

Introduction

Urinary stones are the third most common affliction of the urinary tract. "Stone disease has been a major problem afflicting human population ever since the antiquity. The disease is both very common among men and women with estimated prevalence among the population of 2–3%. The life time recurrence rate is approximately 50%, Urinary tract stones begin to form in a kidney and may enlarge in a ureter or the bladder. Depending on where a stone is located, it may be called a kidney stone, ureteral stone, or bladder stone [1].

Increased incidence of urinary stones in the industrialized world is associated with improved standards of living "mainly including the high dietary intake of proteins and minerals" as well as with race, ethnicity and region of residence [2]. Moreover there are a number of diseases associated with stone formation "i.e., hyperparathyroidism, renal tubular acidosis cystinuria, hyperoxaluria, intestinal mal absorptive conditions" as well as medications "i.e., calcium supplements," vitamin D, triamterene, indinavir". Predisposing factors for stone formation also include anatomical abnormalities "i.e., ureteral strictures, vesico-ureteral reflux, ureteropelvic stenosis, extrinsic ureteral compression and ureterocele among others"[3].

A varying degree of obstruction of the lower ureter due to urinary lithiasis has been explained by the size of the calculus, ureteral oedema and degree of impaction. Most of the cases with urinary lithiasis present with colicky pain although they may sometimes remain asymptomatic and identified only on routine assessment [4]. Cystatin C is inhibitor protease, a non-glycosylated, and
low molecular weight protein. Cystatin C has been proposed to be a marker as it is produced by all nucleated cells at a constant rate and is freely filtrated by the glomeruli and completely catabolized in the proximal tubules. The concentration of serum Cystatin C is mainly determined via glomerular filtration, which makes Cystatin C an endogenous marker of glomerular filtration rate [5].

In a Meta-analysis study by Dharnidharka et al [6] found Cystatin C was superior to serum Creatinine as a marker of glomerular filtration rate. Serum creatinine (Scr) has been widely used as a marker of renal function, but it is lacking enough sensitivity [7].

In this study, we studied serum Creatinine and serum cystatin C levels in Ureteric Stone patients to establish its relevance in the early period of Ureteric Stone The purpose of this study was to assess serum cystatin C as a marker of early Ureteric Stone. Detection in critically patients with normal serum Creatinine.

Materials & Methods

Study Group Case and Sampling

The study was carried out from September 2016 to February 2017 at Al-Hilla Teaching Hospital, Urology Department. The study group consisted of 25 healthy subjects as control with age between (20-66) year and 25 patients of Ureteric Stone with age from (20-62) year. After obtaining an informed consent, a screening questionnaire was filled to collect information such as age, gender and laboratory investigations.

A detailed physical examination was carried out. The serum cystatin C and serum creatinine values were measured simultaneously and analyzed in the Ureteric Stone using commercial kit. 5 ml of blood collected then centrifuge in order to use serum in detection Cystatin C and serum creatinine Evaluation.

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 (Catalogue No.: MBS761747) (BioSource – USA) used in the present study Principle of the Assay was based on sandwich enzyme-linked immuno-sorbent assay technology [8], serum Creatinine was measured via a modified "Jaffe method" with protein precipitation using the Kit from (Bio Labo – France) [9].

Statistical 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.

Ethical Issues

This study was performed with Ethical permission from College of medicine in Babylon University and Ministry of health in Iraq. All patients and family members were counseled. They signed a written informed consent before testing.

Results and Discussion

The correlation between the Ureteric Stone and various factors were studied. The distributions of subjects according to these factors were summarized in Table (1).

<table>
<thead>
<tr>
<th>Sex</th>
<th>Patients</th>
<th>Control</th>
<th>Total</th>
<th>X²</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Number</td>
<td>Number</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Percent %</td>
<td>Percent %</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>16(64%)</td>
<td>14(56%)</td>
<td>30(60%)</td>
<td>3.804</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Female</td>
<td>9(36%)</td>
<td>11(44%)</td>
<td>20(40%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>25(100%)</td>
<td>25(100%)</td>
<td>50(100%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Among 25 patients group the percentages obstruction due to ureteric stone in males higher than females; 16 (64%) and 9 (36%) respectively. In the control group the distribution of male and female were 14 (56%), 11 (44%) respectively. depending on gender, Table (1) have shown the statistical analysis of results and a significant difference at the level of probability (P<0.05) between gender and infection with this.

According to our study the ureteric stone were more common in male than female with ratio of 2:1. This finding is comparable to other studies done by Strope, S. A. et al (2010) [10], who found male more than female ratio of 3.4:1.3., also Khan, G., et al (2014) [11].and found estimated male to female ratio of 2.5:1. According to Table (2), The current study investigates the Ureteric Stone among 25 patients with mean age (47.40±22.14) and 25 apparently healthy subjects from different areas of Al-hilla city with mean age (40.64±11.47), Table (2) have shown Mean difference of Age according to the patients and control, there is significant difference at the level of probability (P<0.005).

### Table 2: Mean difference of Age according to the study groups (patients and control)

<table>
<thead>
<tr>
<th>Age</th>
<th>Study group</th>
<th>No;</th>
<th>Mean ±Std. D</th>
<th>t-test</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>patients</td>
<td>25</td>
<td>47.40±22.14</td>
<td>2.89</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>control</td>
<td>25</td>
<td>40.64±11.47</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

In this study the obstructive uropathy due to have found to be highest in the age 20-40 (52%), and above 60 years (36%). This is comparable to study conducted by Khan et al [11], where the commonest age group was 21 to 40 years, and study conducted by Rajput et al [12] found a mean age of 29 years for renal stones while Grases, F., et al [13] had maximum incidence in age group 30 to 50 years. Peschel et al.(2008), observed in north Iranian population a decreasing gradient of Ureteric Stone patients between younger and older groups with the 80-100 age group showing a significantly lower frequency of Ureteric Stone patients, with respect to the younger group. This contradictory among studies may be due to the difference in sampling, population, life style as well as the difference in the age range to be identified of individuals as an inclusion criteria [15,16].

In total twenty five patients with ureteric stone, figure (1) show 60% from the total number unilateral and 40% was Bilateral ureteric stone Unilateral It may be more common than bilateral previously thought looking at the number of cases diagnosed in our study mechanisms have been proposed to explain Ureteric Stone.

![Figure 1: Distribution of ureteral stone according to type](image)

This finding in agreement with a study examined a total of 65 ureters, which found that 35% unilateral and 15%bilateral, the most common causes of ureteric obstruction was ureteric stone, followed ureteric stricture [17].This was in agreement with Chevalier et al., and Shokeir et al., [18,19]. Who reported that ureteral obstruction is usually a consequence of nephrolithiasis which is the most common cause of urinary obstruction? Mean difference of Cystatin C according to study groups; this study reveals that there was a significant increase (P<0.001) in the According to Cystatin C marker there is significant difference between study groups the mean value was (2.56 ±0.81) compared to control group (0.82 ±0.17) with probability value <0.001. At the other hand Creatinine level in the study groups show mean difference of serum Creatinine according to study group reveals that there was non-significant difference (P>0.12) between
patients the mean value was (1.52 ± 1.13) compared to control group (0.97 ± 0.28). According to patients group there is no significant correlation between the Serum Cystatin C and with Serum Creatinine (P>0.955).

Table 3: Mean difference of Cystatin C and serum Creatinine according to study groups (N=50)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Study group</th>
<th>N</th>
<th>Mean ±Std.D</th>
<th>t-test</th>
<th>p. Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cystatin C</td>
<td>patients</td>
<td>25</td>
<td>2.56 ± 0.81</td>
<td>10.48</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>control</td>
<td>25</td>
<td>0.82 ±0.17</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Creatinine</td>
<td>Patients</td>
<td>25</td>
<td>1.52±1.13</td>
<td>2.20</td>
<td>0.12</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>25</td>
<td>0.97±0.28</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Serum cystatin C is a non-glycosylated, 13.3-kDa protein belonging to cystatin protease inhibitors [20]. It is fully catabolized in the proximal renal tubule and is not returned to blood. Concentration of serum cystatin C is not affected by gender, age, race, protein intake, and muscle mass, unlike serum Creatinine. Which changes significantly according to the muscle mass of the body and dietary factors? Creatinine is increased when there is 50% loss of kidney function [21, 22]. All of these factors explain why serum creatinine concentration may not be a good parameter for accurate. 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 [23]. In order to confirm the results of cystatin C levels using ELISA, and using colorimetric method test to detect serum Creatinine, the characteristic of these techniques, subjects analyze showed in Table (4), for cystatin C the results were found to be; Sensitivity (92%) , Specificity (96%), Accuracy (94%) , PPV (95%) and NPV (92%). While the result for serum Creatinine found to be, Sensitivity (40%) , Specificity (88%), Accuracy (64%) , PPV (77%) and NPV (59.4%).

Table 4: the compare between cystatin c and Creatinine depending on sensitivity and specificity

<table>
<thead>
<tr>
<th>Variable</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Accuracy</th>
<th>PPV</th>
<th>NPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cystatin C</td>
<td>92%</td>
<td>96%</td>
<td>94%</td>
<td>95%</td>
<td>92%</td>
</tr>
<tr>
<td>Creatinine</td>
<td>40%</td>
<td>88%</td>
<td>64%</td>
<td>77%</td>
<td>59.4%</td>
</tr>
</tbody>
</table>

In the Ureteric Stone patients group, more than half the patients (56.2%) had normal Creatinine values, but they had elevated cystatin C and were in Creatinine. This confirms the finding that serum cystatin C is elevated much before serum Creatinine levels start rising. Serum cystatin C has a higher sensitivity in identifying early kidney dysfunction, which is missed by relying on serum Creatinine alone [24].

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 [25].

In the current study, we found that serum Cyst C is more accurate than serum creatinine for detection obstructive uropathy due to ureteric stone. Beegum, M. S., et al (2017) found that cystatin c has important association with sensitivity, early detection and accurate serum marker than serum Creatinine [26]. Our analysis showed that S. Cys C was a favorable marker than S.C this finding is in agreement with Garlipp, C. R., 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 a efficient and a sensitivity marker for kidney function (r = 0.82, sensitivity=100%, Specificity = 75%, efficiency = 95%) [27].

Yang, S.K et al. (2016) [28], their study was conducted according to the guide-line of Meta-analysis of observational studies on 17(for S. CysC) 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.
References


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