Research Article

Cystatin C in Evaluating Renal Function in Ureteral Calculi Hydronephrosis in Adults

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Keywords

Cystatin C · Serum creatinine · Hydronephrosis · Renal function · Ureteral calculi

Abstract

Background: Serum cystatin C (CysC) is still becoming used as a marker of renal function but is far from being commonly used worldwide. The purpose of this study was to characterize the ureteral calculi patients with hydronephrosis-caused CysC changes in renal function.

Methods: To better reflect the changes of renal function, we constructed models of ureteral obstruction in rats to mimic the hydronephrosis caused by human ureteral calculi. Moreover, our study included 200 patients diagnosed with ureteral calculi in our hospital between June 2017 and 2018. We compared the estimated glomerular filtration rate using different equations based on CysC and/or serum creatinine (SCr). **Results:** We found that the expression of CysC and SCr increased with the prolonged obstruction time by enzyme linked immunosorbent assay. Moreover, quantitative real-time polymerase chain reaction, Western blot and immunohistochemistry further demonstrated that the expression of CysC increases with the degree of hydronephrosis. Among 200 patients with ureteral calculi, 40 (20.0%) had no hydronephrosis, 110 (55.0%) had mild hydronephrosis, 32 (16.0%) had moderate hydronephrosis and 18 (9.0%) had severe hydronephrosis. As the degree of hydronephrosis increased, the expression of neutrophil percentage, CysC, blood urea nitrogen, SCr and serum uric acid also

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increased. Multivariate analyses demonstrated that only CysC was an independent risk factor for hydronephrosis ($p = 0.003$). In addition, CysC and the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) CysC equation showed the highest veracity in renal function estimation of patients with hydronephrosis caused by ureteral calculus. **Conclusion:** For patients with hydronephrosis caused by ureteral calculi, CysC better reflects the changes in renal function, and the CKD-EPI CysC equation has the highest accuracy.

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Introduction

Ureteral calculi is a common condition in urology [1, 2], that is often secondary to intra-renal calculi [3]. Calculi can cause urinary tract obstruction, resulting in upper urinary tract dilatation, upper urinary tract infection, and the deterioration of renal function. The principle of treatment is to remove the upper urinary tract obstruction, relieve the upper urinary tract expansion and accumulation of urine, control the progress of infection, and then perform the treatment to remove the calculi [4].

Hydronephrosis is urine retention caused by the obstruction of various causes and is a common complication of urogenital system diseases. Due to the lack of symptoms in the early stages of the chronic hydronephrosis, some renal function has been damaged when symptoms appear [5]. Prolonged hydronephrosis causes increased intra-renal pressure, resulting in progressive dilatation of the renal pelvis and renal calyces and even atrophy of the renal parenchyma [6]. Therefore, it is particularly important to evaluate the degree of renal function impairment in hydronephrosis.

The glomerular filtration rate (GFR) is widely accepted as an ideal indicator for evaluating renal function [7, 8]. Currently, the clearance of inulin is the gold standard used to estimate GFR. However, due to its complicated operation [9], blood urea nitrogen (BUN) β_2 -microglobulin (β_2 -MG) and serum creatinine (SCr) are often used to evaluate renal function clinically, but BUN, β_2 -MG and SCr are often affected by other physiological and pathological factors and cannot reflect renal function accurately [10, 11].

Cystatin C (CysC) is an endogenous cysteine protease inhibitor with a molecular weight of 13.3 kDa, and most nucleated cells are considered to be the source of secretion [12]. Normally, CysC is filtered through the glomerulus, reabsorbed and broken down into amino acids in the renal tubules. In recent years, serum CysC is a widely studied indicator of GFR. A large number of studies have shown that the metabolism of CysC is rarely affected by factors such as age, sex, weight, diet, infection and other factors, and has high specificity [13–15].

In this study, in order to better reflect the changes of renal function and the expression of CysC in kidney during hydronephrosis, we constructed models of ureteral obstruction in rats to mimic the hydronephrosis caused by human ureteral calculi. In addition, we analysed the patient's metabolic indicators and compared the estimated GFR (eGFR) using different equations based on CysC and/or SCr to provide clues for early detection of renal function changes in the presence of ureteral calculi.

Materials and Methods

Constructing Models of Ureteral Obstruction in Rats

Male SD rats (aged 10 weeks; body weight 300–400 g, 3 rats per group) were purchased from Slaccas (Slaccas Laboratory Animal, Shanghai, China). The model of ureteral obstruction was constructed by ligating the unilateral ureter to simulate the change of hydronephrosis

after human ureteral obstruction. Rats were treated with cervical dislocation at 0 h, 1 day (D), 2 D, 3 D, 4 D, 5 D, 6 D, and 7 D after ligation. Bilateral kidney and cardiac blood were collected for the preparation of other experiments [16]. Animal research was conducted according to the ethical guidelines for animal experiment systems approved by the Animal Care and Use Committee of Tongji University.

RNA Extraction and Quantitative Real-Time Polymerase Chain Reaction

Total RNA was extracted from rat kidney tissues using Trizol reagent (Invitrogen, CA, USA) according to the manufacturer's protocol, and cDNA was generated using the cDNA synthesis kit (Takara Biotechnology, Dalian, China). Quantitative real-time polymerase chain reaction for CysC was performed using a SYBR Green PCR Kit (Takara Biotechnology, Dalian, China) with an ABI Prism 7500 Sequence Detection System (Applied Biosystems, Foster City, CA, USA). The primer sequences (Sango Biotech, China) were as follows: CysC forward, 5'-AACTACATGTAC-CAAGTCCCAG-3' and reverse, 5'-CTGAATTTTGTCTAGGGAGTGTG-3'. GAPDH acted as internal standards, and each sample was repeated 3 times. The relative quantification of expression of CysC was performed using the $2^{-\Delta\Delta C_t}$ method and compared with internal standards.

Enzyme Linked Immunosorbent Assay

Cardiac blood obtained from each group of rats was centrifuged at 3,000 rpm for 10 min. Then, the supernatant was collected and the levels of CysC and SCr in rat serum were detected according to the instructions of the enzyme linked immunosorbent assay kit (mlbio, Shanghai, China). The process was repeated 4 times for each sample.

Western Blot

Total protein was extracted from rat kidney tissues, separated using 10% sodium dodecyl sulfate-polyacrylamide gel and imprinted on the nitrocellulose membrane (Sigma-Aldrich; Merck KGaA). Membranes were sealed at room temperature for 1 h with 5% non-fat milk and then incubated with primary antibody against CysC (Santa Cruz Biotechnology CA, USA; 1:200). GAPDH (Abcam, Cambridge, MA, USA) was used as an internal control. After incubation with the secondary antibody (Jackson Immno Research; 1:5,000) for 1 h, the membranes were washed 3 times and the signals were visualized by the LI-COR Image Studio Lite imaging system.

Haematoxylin-Eosin Staining and Immunohistochemistry

The rat kidney tissues samples were fixed in 4% paraformaldehyde at room temperature and then dehydrated via an ethanol solution and embedded in paraffin. Haematoxylin-eosin staining and immunohistochemistry were performed according to the previous procedure [17]. The sections were incubated with previous antibody for expression of CysC. The sections were imaged using the microscope (Leica Microsystems, Mannheim, Germany).

Patients

A total of 200 eligible patients diagnosed with ureteral calculi were identified between June 2017 and 2018. The inclusion criteria were as follows: patients with unilateral ureteral calculi diagnosed by clinical history, symptoms, signs, and confirmed by CT scan and patients who understood the contents of this study and voluntarily participated and accepted the test methods used in this study. The exclusion criteria were as follows: patients with a history of recurrent kidney stones or with stones in the kidney at the screening visit; patients with immune dysfunction, severe impairment of liver function, chronic renal insufficiency; malignant tumours, diabetes, coronary heart disease or any other known condition that might influence the CysC level; and hydronephrosis due to other causes. All patients were divided into the following 4 groups according to the Noble hydronephrosis method [18]: the no

hydronephrosis group, the mild hydronephrosis group, the moderate hydronephrosis group and the severe hydronephrosis group. The study protocol was approved by the Biomedical Ethics Committee of the Tenth Hospital in Shanghai, and written informed consent was obtained from all patients or their relatives.

Serum Collection for All Patients

Serum was collected in the morning, and all measurements were performed in the same laboratory. CysC was quantified using a fully automated particle-enhanced immunoturbidimetric assay with Sentinel Diagnostics reagents (Sentinel CH, Milan, Italy) on the Architect ci 16200 System (Abbott Laboratories, IL, USA) according to the manufacturer's instructions; (intra-assay CV <1.5%, inter-assay CV <1%). The baseline SCr was assessed in 200 participants and was analysed using an isotope dilution mass spectrometry traceable Jaffe kinetic assay for creatinine on a Hitachi 917 analyser (Roche Diagnostics GmbH, Mannheim Germany).

Statistical Analysis

To better evaluate changes in renal function, we used different GFR estimation equations such as the GFR, Cockcroft-Gault (CG) formula, modification of diet in renal disease (MDRD) equation, Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) CysC equation, CKD-EPI creatinine equation and CKD-EPI creatinine-CysC equation to calculate the eGFR [19–22].

Data was analysed using the SPSS software (version 20.0, SPSS, Inc., Chicago, IL, USA) and GraphPad Prism software (version 7.0, Inc., San Diego, CA, USA). Receiver operating characteristic curves were detected using MedCalc version 15.2.0 Software, which defined the sensitivity, specificity and differences in the area under the curve (AUC). A *p* value <0.05 was considered significant. All continuous data is summarized as the means ± SD, and chi-square tests were performed to evaluate differences in categorical variables.

Results

The Expression of CysC and SCr Increased with the Prolonged Obstruction Time in Rats with Ureteral Obstruction

We constructed rat models of unilateral ureteral obstruction, after which we obtained kidney tissue and cardiac blood at different obstruction time points. Using enzyme-linked immunosorbent assay, we found that the concentration levels of CysC and SCr in the blood of rats increased with the prolonged obstruction time (Fig. 1a, b). In addition, we assessed the relative expression of CysC by quantitative real-time polymerase chain reaction in the kidney tissues of rats at different obstruction times, and our results showed that the expression of CysC increased with the prolonged obstruction time (Fig. 1c). Similarly, Immunohistochemistry also confirmed that CysC expression increased with time extension (Fig. 1d).

With the Extension of Obstruction Time, Hydronephrosis was Aggravated, Kidney Volume Increased and the Kidney Parenchyma Thickness Decreased

As seen by comparing the appearance of the kidneys and the transverse anatomy of the kidney, hydronephrosis increased with the extension of obstruction time, the kidney expanded in volume, and the renal parenchyma thickness decreased (Fig. 2a). Haematoxylin-eosin staining of the renal cortex and renal medulla revealed that the renal glomeruli atrophied, and the renal tubules dilatated in the cortex with the aggravation of hydronephrosis (Fig. 2b). Additionally, intramedullary renal interstitial oedema was increased, and a large number of inflammatory cells were infiltrated in the interstitium (Fig. 2c).

Fig. 1. The concentration of CysC and SCr increased with the increase in the degree of hydronephrosis in rats with ureteral obstruction; **a** The concentration of CysC was determined by ELISA (** $p < 0.001$); **b** The concentration of SCr was measured by ELISA (** $p < 0.001$); **c** The relative expression of CysC was detected by qRT-PCR (** $p < 0.01$, *** $p < 0.001$); **d** IHC staining of CysC at different obstruction times; Scale bar = 200 μm for 10× and 100 μm for 20×. CysC, Cystatin C; Cr, creatinine.

The Concentration of CysC and SCr Is Associated with the Degree of Hydronephrosis

Patients were divided into 4 groups according to the degree of hydronephrosis, of which 40 (20.0%) patients had no hydronephrosis, 110 (55.0%) had mild hydronephrosis, 32 (16.0%) had moderate hydronephrosis, and 18 (9.0%) had severe hydronephrosis. The variables associated with the degree of hydronephrosis, including the demographic and clinical characteristics, are described in Tables 1 and 2. Univariate analyses demonstrated that the degree of hydronephrosis was associated with the disease course, body weight, proteinuria, neutrophil percentage, CysC, BUN, SCr and serum uric acid (SUA; All $p < 0.05$). The 200 ureteral calculi patients included in the study cohort had a median age of 52 years, median neutrophil percentage of 63.47%, median CysC of 0.95 mg/L, median BUN of 6.59 mmol/L, median SCr of 102.88 μmol/L and median SUA of 342.02 μmol/L. In addition, as the degree of hydronephrosis increased, the concentration of neutrophils, CysC, BUN, SCr and SUA also increased (Table 2). From Figure 3a and b, we could conclude that CysC has correlated between no hydronephrosis group and mild hydronephrosis group, while SCr has no correlation.

Stratified univariate and multivariate log regression were used to analyse the factors associated with hydronephrosis. As shown in Table 3, CysC and SCr were factors that affected hydronephrosis in the univariate log regression. However, the multivariate log regression model showed that only CysC (OR 6.28; 95% CI 1.85–21.31; $p = 0.003$) was an independent risk factor for hydronephrosis.

Fig. 2. With the extension of obstruction time, hydronephrosis was aggravated, kidney volume increased, and the kidney parenchyma thickness decreased; **a** The renal volume and degree of hydronephrosis increased with time; **b** Renal cortical HE staining showed that the renal tubules in the cortex were atrophied with the aggravation of hydronephrosis; **c** Renal medullary HE staining showed that intramedullary renal interstitial oedema was increased with the aggravation of hydronephrosis.

CysC and the CKD-EPI CysC Equation Better Reflect the Changes in Renal Function

The means of determining the patients' eGFR using 6 methods are shown in Table 4. We could conclude that the eGFR determined using various equations was associated with the degree of hydronephrosis. In addition, the diagnostic sensitivity of our evaluation by the receiver operating characteristic curve (Table 5) showed that the AUC for CysC detection of

Table 1. Univariate analyses of patients with different hydronephrosis according to various clinicopathological variables

Characteristics	Total	Hydronephrosis group, n (%)	Mild hydronephrosis group, n (%)	Moderate hydronephrosis group, n (%)	Severe hydronephrosis group, n (%)	p value
Total	200	40 (20.0)	110 (55.0)	32 (16.0)	18 (9.0)	
Age, years						0.099
<40	33 (16.5)	7 (21.2)	17 (51.5)	7 (21.2)	2 (6.1)	
40–60	111 (55.5)	20 (18.0)	69 (62.7)	16 (14.4)	6 (5.4)	
>60	56 (28.0)	13 (23.2)	24 (42.9)	9 (16.1)	10 (17.9)	
Gender						0.151
Female	69 (34.5)	9 (13.0)	43 (62.3)	13 (18.8)	4 (5.8)	
Male	131 (65.5)	31 (23.7)	67 (51.1)	19 (14.5)	14 (10.7)	
Course, day						0.007
<7	124 (62.0)	23 (18.5)	79 (63.7)	13 (10.5)	9 (7.3)	
≥7	76 (38.0)	17 (22.4)	31 (40.8)	19 (25.0)	9 (11.8)	
Body weight, kg						0.047
<60	39 (19.5)	4 (10.3)	20 (51.3)	13 (33.3)	2 (5.1)	
60–80	133 (66.5)	29 (21.8)	74 (55.6)	16 (12.0)	14 (10.5)	
>80	28 (14.0)	7 (25.0)	16 (57.1)	3 (10.7)	2 (7.1)	
Height, cm						0.539
<160	38 (19.0)	6 (15.8)	22 (57.9)	8 (21.1)	2 (5.3)	
160–170	93 (46.5)	17 (18.3)	56 (60.2)	12 (12.9)	8 (8.6)	
>170	69 (34.5)	17 (24.6)	32 (46.4)	12 (17.4)	8 (11.6)	
BMI, kg/m ²						0.218
<18.5	9 (4.5)	2 (22.2)	3 (33.3)	4 (44.4)	0 (0.0)	
18.5–23.9	72 (36.0)	12 (16.7)	40 (55.6)	11 (15.3)	9 (12.5)	
≥24	119 (59.5)	26 (21.8)	67 (56.3)	17 (14.3)	9 (7.6)	
Primary site						0.593
Right	93 (46.5)	17 (18.3)	51 (54.8)	14 (15.1)	11 (11.8)	
Left	107 (53.5)	23 (21.5)	59 (55.1)	18 (16.8)	7 (6.5)	
Urine protein						0.040
Negative	157 (78.5)	29 (18.5)	93 (59.2)	20 (12.7)	15 (9.6)	
Positive	43 (21.5)	11 (25.6)	17 (39.5)	12 (27.9)	3 (7.0)	
Urine sugar						0.855
Negative	182 (91.0)	37 (20.3)	101 (55.5)	28 (15.4)	16 (8.8)	
Positive	18 (9.0)	3 (16.7)	9 (50.0)	4 (22.2)	2 (11.1)	

p value <0.05 are shown in bold. BMI, body mass index.

renal function in patients was larger than that of BUN, SCr and SUA, and CysC had higher sensitivity than BUN, SCr and SUA (Fig. 4a). Based on the CysC and SCr equations, the CKD-EPI CysC equation detected a larger AUC and highest sensitivity for renal function (Fig. 4b).

Discussion

Clinically, the urine above the calculi cannot flow smoothly beyond the obstruction caused by ureteral calculi, and the urine collects in the renal pelvis and calyces. Excessive pressure leads to renal pelvis and calices expansion, renal unit cell degeneration, atrophy and necrosis. Subsequently, this leads to thinning of the renal parenchyma, which increases the hemodynamic resistance of the renal parenchyma and cortical internal arteries, aggravating the degree of hydronephrosis and reducing renal perfusion, thus damaging renal function [23].

Table 2. Demographics and renal function of the study population

Characteristics	Total (n = 200)	No hydronephrosis group (n = 40)	Mild hydronephrosis group (n = 110)	Moderate hydronephrosis group (n = 32)	Severe hydronephrosis group (n = 18)	p value
Mean age, years	52.79±12.74	53.53±12.18	52.35±11.54	51.28±16.02	56.44±14.77	0.721
BMI, kg/m ²	24.72±3.29	24.91±3.61	24.93±3.22	23.59±3.32	25.00±2.74	0.387
BSA, m ²	1.75±0.18	1.79±0.16	1.75±0.17	1.70±0.21	1.81±0.16	0.581
White cell (10 ⁹ /L)	7.15±2.70	6.70±1.98	7.19±2.48	7.82±3.82	6.74±2.89	0.443
Neutrophil, %	63.47±11.12	60.58±10.70	62.79±10.42	67.13±12.30	67.52±12.12	0.004
CysC, mg/L	0.95±0.31	0.82±0.14	0.90±0.19	1.00±0.26	1.53±0.53	<0.001
BUN, mmol/L	6.59±3.71	5.66±2.28	6.05±2.18	7.99±5.40	9.44±7.01	<0.001
SCr, μmol/L	102.88±57.40	86.32±25.12	92.87±46.01	117.09±52.68	175.56±105.90	<0.001
SUA, μmol/L	341.02±94.83	321.73±90.08	341.96±88.81	334.04±75.76	390.56±148.48	0.037

p value <0.05 are shown in bold. BMI, body mass index; BSA, body surface area; GFR, glomerular filtration rate; CysC, cystatin C; BUN, blood urea nitrogen; SCr, serum creatinine; SUA, serum uric acid.

Table 3. Univariate and multivariate analyses of factors associated with hydronephrosis

Characteristics	Univariate analyses		Multivariate analyses	
	OR (95% CI)	p value	OR (95% CI)	p value
Neutrophil, %				
<75	1	0.065	-	0.107
≥75	3.21 (0.93–11.05)			
CysC, mg/L				
<1.02	1	0.003	1	0.003
≥1.03	6.28 (1.85–21.31)		6.28 (1.85–21.31)	
BUN, mmol/L				
<8.07	1	0.149	-	0.681
≥8.07	2.50 (0.72–8.71)			
SCr, μmol/L				
<84 (female)/<104 (male)	1	0.037	-	0.518
≥84 (female)/≥104 (male)	2.36 (1.05–5.28)			
SUA, μmol/L				
<357 (female)/<428 (male)	1	0.184	-	0.413
≥357 (female)/≥428 (male)	1.89 (0.74–4.83)			

p value <0.05 are shown in bold. CysC, Cystatin C; BUN, blood urea nitrogen; SCr, serum creatinine; SUA, serum uric acid.

Fig. 3. The relationship between the degree of hydronephrosis and the concentration of CysC (a) and SCr (b; $n = 200$, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$). CysC, Cystatin C; Cr, creatinine.

GFR is a commonly used indicator of renal function changes. Currently, the commonly used indicators include BUN, β_2 -MG, SCr, creatinine clearance, urinary microalbumin, urinary N-acetyl-D-aminoglucosidase and CysC [10, 24, 25]. However, due to the influence of many physiological and pathological factors such as age, sex, body size, and complicated steps of specimen collection, BUN, β_2 -MG, SCr, creatinine clearance, urine microalbumin and N-acetyl-D-aminoglucosidase are not accurate and reliable in evaluating renal function.

CysC is a low molecular weight protein containing 120 amino acids. It was found that CysC sustain constant velocity in all nuclear cells in vivo as an endogenous GFR marker [12]. CysC can be completely filtered by the glomeruli and do not get degraded and reabsorbed in the renal tubules [26]. Unlike SCr, CysC values are not affected by muscle mass, age, sex and have high specificity making CysC an ideal indicator to evaluate GFR [14].

To improve the accuracy and effectiveness of GFR detection, many specifically designed prediction equations have been proposed as reliable alternatives for evaluating GFR. Commonly used equations are the CG formula, the nephropathy diet improvement equation (MDRD), and the equation for CKD-EPI based on CysC and/or SCr [20–22]. Previously, 84% of laboratories in United States recommended using the MDRD study equation to estimate GFR [27, 28]. Studies have found that CKD-EPI equation is a better predictor of risk than MDRD equation in CKD cohort [29], and the CKD-EPI equation is widely used in the North America, Europe, and Australia [30]. However, some studies have shown that MDRD equation has the smallest deviation and the highest accuracy compared with CG formula and CKD-EPI in diabetic cohort [31]. In addition, researchers have also developed new equations that are more accurate than CKD-EPI to eGFR in the elderly in China [32]. This is the first time to explore the expression of CysC and SCr in patients with hydronephrosis caused by ureteral calculi.

It has been found that patients with mild, moderate and severe renal hydronephrosis caused by ureteral calculi were all impaired in renal glomerular and renal tubular filtration function [33]. However, for patients with mild and moderate hydronephrosis, SCr may still

Fig. 4. CysC and the CKD-EPI CysC equation can better reflect the changes in renal function; **a** ROC curve analysis for CysC, BUN, SCr and SUA; **b** The ROC curve analysis for the equations based on CysC and SCr. CysC, Cystatin C; BUN, blood urea nitrogen; SCr, serum creatinine; SUA, serum uric acid; GFR, glomerular filtration rate; MDRD, modification of diet in renal disease; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration.

be in the normal clinical range, but renal function has been impaired. In our study, we judged the degree of hydronephrosis based on the patient's renal ultrasound results [18]. But this is an experienced and professional ultrasound doctor who needs to be involved. Ultrasound doctor with different experiences have different grasps of the degree of hydronephrosis, and the degree of hydronephrosis will be mixed with artificial uncertainty. However, as an indicator in the blood, CysC can be quantified, more convenient and cheaper, and not subject to artificial factors, and CysC as a sensitive indicator can reflect the degree of hydronephrosis.

Our research aims to find a sensitive indicator to reflect the degree of hydronephrosis in patients with ureteral calculi, and then to make an intervention in the patients. By constructing rat models of unilateral ureteral obstruction and statistically analysed various metabolic parameters and clinical variables of patients diagnosed with unilateral ureteral calculi in our hospital, we found that CysC and CKD-EPI CysC equation were more sensitive and accurate for assessing renal function in patients with hydronephrosis caused by ureteral calculi, patients would be more inclined to intervene surgically sooner rather than later compared to a patient in whom no compromise in renal function is detected.

There are several limitations in this study. First, all patients need to be followed up prospectively, and the sample numbers need to further expand. Moreover, approximately 28% of patients are over the age of 60, which might have repercussion on physiological renal function decline. Additionally, the underlying etiologic disorders for the development of urinary calculi are multiple, which requires to accurately select patients and identify the aetiology of stone disease.

In conclusion, we found that the expression of CysC and SCr increased with the aggravation of hydronephrosis. Moreover, CysC and the CKD-EPI CysC equation can better reflect the changes in renal function in patients with hydronephrosis caused by ureteral calculi.

Table 4. eGFR by various equations of patients with different hydronephrosis

eGFR, mL/min/1.73 m ²	Total (n = 200)	No hydronephrosis group (n = 40)	Mild hydronephrosis group (n = 110)	Moderate hydronephrosis group (n = 32)	Severe hydronephrosis group (n = 18)	p value
GFR	66.64±25.48	73.14±24.00	70.61±23.29	57.89±25.96	43.49±25.67	<0.001
Cockcroft-Gault	80.41±32.11	90.18±31.37	84.82±29.44	67.85±31.76	54.03±32.14	<0.001
MDRD	72.66±26.03	81.87±22.48	77.15±24.27	61.28±23.51	44.99±24.64	<0.001
CDK-EPI creatinine equation	77.60±25.65	85.21±20.96	81.26±22.86	67.74±29.06	55.83±29.89	<0.001
CDK-EPI CysC equation	93.80±27.96	104.82±21.62	96.52±22.05	89.74±38.45	59.87±26.13	<0.001
CDK-EPI creatinine-CysC equation	76.40±24.67	84.96±20.55	80.05±20.66	69.54±28.37	47.25±26.48	<0.001

p value <0.05 are shown in bold. eGFR, estimated glomerular filtration rate; MDRD, modification of diet in renal disease; CKD-EPI, Chronic Kidney Disease-Epidemiology Collaboration; CysC, Cystatin C.

Table 5. Analysis of predictive accuracy through the evaluation of the AUC

Characteristics	AUC	Sensitivity, %	Specificity, %	p value	95% CI
CysC	0.657	76.9	50.0	0.002	0.587–0.723
SCr	0.585	30.0	95.0	0.096	0.514–0.654
BUN	0.605	53.7	70.0	0.040	0.534–0.673
SUA	0.601	63.7	60.0	0.048	0.529–0.669
GFR	0.588	53.1	70.0	0.084	0.517–0.657
Cockcroft-Gault	0.609	41.2	80.0	0.034	0.537–0.677
MDRD	0.619	39.4	87.5	0.020	0.548–0.686
CDK-EPI CysC equation	0.651	77.5	52.5	0.003	0.580–0.717
CDK-EPI creatinine equation	0.585	36.9	87.5	0.097	0.513–0.654
CDK-EPI creatinine-CysC equation	0.610	53.7	67.5	0.032	0.539–0.678

p value <0.05 are shown in bold.

AUC, area under the curve; GFR, glomerular filtration rate; MDRD, modification of diet in renal disease; CKD-EPI, CKD-epidemiology collaboration; CysC, Cystatin C; BUN, blood urea nitrogen; SCr, serum creatinine; SUA, serum uric acid.

Statement of Ethics

The study protocol was approved by the Biomedical Ethics Committee of the Tenth Hospital in Shanghai. The study conforms to the ethical standards of the Declaration of Helsinki.

Disclosure Statement

All authors declare no conflicts of interest.

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Author Contribution

W.M., S.L., and J.G.: designed the research. K.W., M.W., H.S., Q.L., and M.B.: acquired the data. W.M. and K.W.: analysed the results. W.M., S.L., and K.W.: wrote the article. B.P. and J.G.: revised and provided critical comments. All authors read and approved the final manuscript.

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