

# Comparison of Serum Cystatin C and Creatinine Levels to Evaluate Early Renal Function after Kidney Transplantation

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## Abstract

**Background:** Accurate and rapid assessment of allograft function is essential in renal transplant recipients in order to detect allograft rejection and to monitor drug nephrotoxicity. We aimed to evaluate the usefulness of cystatin C as a marker of kidney allograft function in the early post-transplant period and to compare this value with that of conventional serum creatinine concentration.

**Methods:** Twenty four patients scheduled for kidney transplantation at the Kidney Transplant Center of Ghaem Hospital, Mashhad, Iran from September 2006 to November 2007, were sequentially enrolled into the present study. Serum creatinine and cystatin C concentrations and urine output were measured daily after transplantation for 3 weeks or until discharge from the hospital.

**Results:** On the 3<sup>rd</sup> postoperative day, with a cut-off value of 75 mL/min for glomerular filtration rate, areas under the receiver operating characteristic (ROC) curves were 0.926 for creatinine (P=0.021) and 0.815 for cystatin C (P=0.088). At this point creatinine was more sensitive and specific than cystatin C in estimating glomerular filtration rate. On the 7<sup>th</sup> day after transplantation, areas under ROC curves were 0.893 for creatinine (P=0.066) and 1.000 for cystatin C (P=0.017). Therefore, cystatin C was more sensitive and specific than creatinine in estimating glomerular filtration rate. In two patients with acute rejection and arterial thrombosis, serum cystatin C concentrations increased earlier than serum creatinine.

**Conclusion:** There is a correlation between creatinine and cystatin C early after kidney transplantation. Serum creatinine levels seem to be more sensitive and specific for detecting transitory changes in renal function in the 1<sup>st</sup> week after transplantation. After the 1<sup>st</sup> week after transplantation, cystatin C was more sensitive and specific than serum creatinine concentration.  
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**Keywords** • Creatinine • cystatin C • kidney transplantation

## Introduction

**S**hort-term outcomes in kidney transplantation have improved significantly over the past decade.<sup>1,2</sup> Accurate and rapid assessment of allograft function is essential

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in renal transplant recipients in order to detect allograft rejection (acute and chronic) and to monitor drug nephrotoxicity. Measuring serum creatinine is the most widely used method to rapidly assess kidney function, but it has several disadvantages. First, it only detects renal function impairment of at least 50%.<sup>2</sup> Second, it is affected by several factors that are independent of changes in glomerular filtration rate (GFR) such as age, race, muscle mass, gender, medication use, and catabolic state. Third, different laboratories measure serum creatinine using different methods, producing results that are difficult to compare. Urinary creatinine clearance overcomes some of the limitations of serum creatinine concentration, but remains inaccurate because of collection errors and changes in creatinine excretion.<sup>2</sup> Measuring serum cystatin C concentration has been proposed as a suitable alternative to assess kidney function.

The use of serum cystatin C as an indicator of GFR was first suggested in 1985. Human cystatin C is a 132-amino-acid, 13-kd cysteine protease inhibitor, which is produced by all nucleated cells at a constant rate.<sup>2</sup> Production of cystatin C is not altered by inflammatory processes. In contrast to creatinine, serum cystatin C concentration appears to be independent of gender, age, nutritional status, medications, or body mass. Because of its low molecular weight and positive charge at physiologic pH, cystatin C freely passes the glomerular filter. It is not secreted into the renal tubules; however, proximal tubular cells reabsorb and catabolize the filtered cystatin C, resulting in very low urinary concentrations.<sup>1</sup> Given these characteristics, together with its constant production, cystatin C would be an ideal indicator of GFR. Renal transplant recipients, for whom precise determination of GFR is critical, may thus benefit from this information. Cystatin C can be measured by various radioimmunoassay and fluorescent or enzymatic immunoassays. Immunonephelometric methods appear to be superior to other assays for measuring cystatin C.<sup>3</sup>

It has been shown that serum cystatin C is highly sensitive in detecting impaired glomerular function in renal transplant patients. However, these results have been obtained mostly in the late postoperative period. In the present study we evaluated cystatin C as a marker of allograft function during the early post-transplantation period. In addition, we evaluated the relationship between serum levels of creatinine and cystatin C and urine output.

### Patients and Methods

Twenty four patients (17 men and 7 women,

mean age  $36.2 \pm 12.87$  years, range 19-56 years) who received kidney transplants (19 from live donors and 5 from cadavers) treated at the Kidney Transplant Center of Ghaem hospital, Mashhad, Iran, were included in the study from September 2006 to November 2007. Informed consent was obtained from all patients. The study was approved by the Research Ethics Committee of Mashhad University of Medical Sciences. In one patient the transplanted kidney was removed because of arterial thrombosis at the end of the 1<sup>st</sup> week after transplantation. Serum creatinine, cystatin C, and urine output were measured daily during the 1<sup>st</sup> week after transplantation. Serum creatinine concentrations were measured by a modified kinetic Jaffe method. Enzyme linked immunosorbent assay (ELISA) using the microplate reader at a wavelength of 450 nm was used to measure serum cystatin C levels.

In order to obtain normal values and ranges of serum cystatin C, 40 apparently healthy adults mean age  $35.41 \pm 13.28$  years, range 20-53 years with normal serum creatinine levels (mean  $0.785 \pm 0.121$  mg/dL, range 0.5-1 mg/dL,) were selected as control group. Venous blood was collected and serum cystatin C was assayed by ELISA. The mean cystatin C plasma concentration in healthy adults (control group) was  $0.9 \pm 0.168$  mg/L (range 0.570 - 1.400 mg/L).

We used *Statistical Package for the Social Sciences* software (SPSS, version 15) for all analyses. Correlations between serum concentrations of cystatin C and creatinine and urine output were calculated with the Pearson coefficient. We estimated the GFR with the Cockcroft-Gault formula. To evaluate the sensitivity and specificity of serum creatinine and cystatin C levels in assessing renal impairment, a GFR <75 mL/min was used as the cut-off value to define renal failure. Receiver operating characteristic (ROC) curves were generated to estimate the accuracy of the different indicators of GFR using the cut-off value of 75 mL/min on the 3<sup>rd</sup> and 7<sup>th</sup> days after transplantation. One-way ANOVA was used to compare the percent reduction in serum creatinine cystatin C levels between live donors versus cadaver donors. A P value less than 0.05 was considered statistically significant.

### Results

In the healthy control group with normal GFR, mean serum cystatin C concentrations was  $0.9 \pm 0.168$  mg/L. There was no significant correlation between serum creatinine and cystatin C in the control group ( $r=0.049$ ,  $P=0.764$ ).

From the 3<sup>rd</sup> day after transplantation, a significant correlation was seen between serum concentrations of creatinine and cystatin C (table 1). There was a significant negative correlation between serum creatinine and urine output from the 4<sup>th</sup> to 7<sup>th</sup> day after transplantation (table 2). Similarly, a significant negative correlation was seen between serum cystatin C concentration and urine output from the 3<sup>rd</sup> to 7<sup>th</sup> day after transplantation (table 3).

**Table 1:** Correlation of serum creatinine and cystatin C levels in renal transplant recipients in the first week after transplantation

Day after transplantation	r* value	P value
Day 1	-0.5	0.021
Day 2	0.342	0.111
Day 3	0.650	0.001
Day 4	0.818	0.0001
Day 5	0.841	0.0001
Day 6	0.851	0.0001
Day 7	0.819	0.004

r\* = Pearson's correlation coefficient

**Table 2:** Correlation between serum creatinine level and urine output in renal transplant recipients in the first week after transplantation

Day after transplantation	r* value	P value
Day 1	0.317	0.07
Day 2	-0.178	0.202
Day 3	-0.306	0.073
Day 4	-0.377	0.035
Day 5	-0.403	0.026
Day 6	-0.480	0.009
Day 7	-0.510	0.006

r\* = Pearson's correlation coefficient

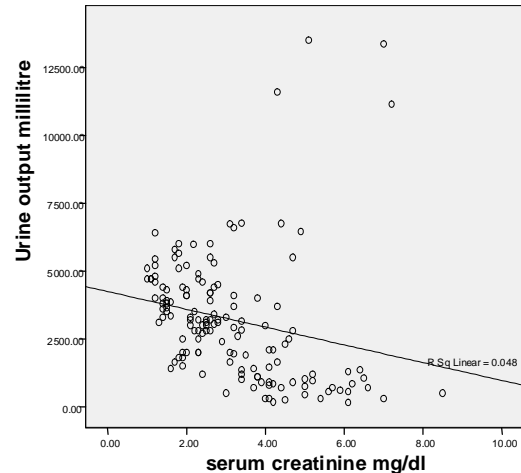
**Table 3:** Correlation between serum cystatin C and urine output in renal transplant recipients in the first week after transplantation

Day after transplantation	r* value	P value
Day 1	-0.162	0.248
Day 2	-0.081	0.357
Day 3	-0.406	0.034
Day 4	-0.563	0.005
Day 5	-0.628	0.006
Day 6	-0.515	0.052
Day 7	-0.462	0.023

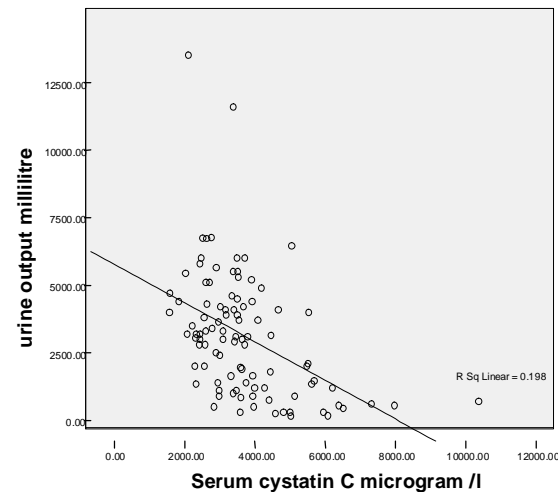
r\* = Pearson's correlation coefficient

The results were verified by comparing the scatter plot of urine output as the dependant factor with serum creatinine and cystatin C levels as independent factors. The slope of the line describing the relationship between changes in serum cystatin C levels and changes in urine output was steeper than the slope for serum creatinine and urinary output (figures 1 and 2). The percent reductions in serum creatinine and cystatin C on each day compared with the previous day were calculated, and are reported in tables 4 and 5.

Table 6 summarizes the results of the effect of transplantation from live donors versus cadaver donors on percent reduction in serum



**Figure 1:** Scatter plot of the relationship between serum creatinine level and urine output.



**Figure 2:** Scatter plot of the relationship between serum cystatin C level and urine output.

creatinine and cystatin C levels on each day compared with the previous day, according to one-way ANOVA. Our comparison between live and cadaver kidney recipients showed that there was a significant difference in the percent reduction in serum creatinine on the 2<sup>nd</sup> day versus 1<sup>st</sup> day after transplantation. The reduction in serum creatinine on the 2<sup>nd</sup> day was significantly more obvious in live-donor kidney recipients versus cadaver donors. There were no significant differences between the two groups on subsequent days. Comparisons of the percent reduction in serum cystatin C concentrations showed no significant differences between live and cadaver kidney recipients on subsequent days. There were no gender differences in percent reduction in serum creatinine or serum cystatin C levels.

**Table 4:** Comparisons of the percent reduction in serum creatinine levels per day versus the previous day during the first week after transplantation

Day after transplantation	Mean percent reduction in creatinine level $\pm$ SD	P value
2 <sup>nd</sup> vs 1 <sup>st</sup>	-103.24 $\pm$ 81.85	0.0001*
3 <sup>rd</sup> vs 2 <sup>nd</sup>	-62 $\pm$ 52.63	0.00001*
4 <sup>th</sup> vs 3 <sup>rd</sup>	-12.63 $\pm$ 20.45	0.27
5 <sup>th</sup> vs 4 <sup>th</sup>	-6.21 $\pm$ 20.55	0.81
6 <sup>th</sup> vs 5 <sup>th</sup>	-2.22 $\pm$ 18.51	0.97
7 <sup>th</sup> vs 6 <sup>th</sup>	11.22 $\pm$ -9.25	0.99

\*Significant reduction in serum creatinine level continued to day 3 after transplantation.

**Table 5:** Comparisons of the percent reduction of serum cystatin C levels per day versus the previous day during the first week after transplantation

Day after transplantation	Mean/median percent reduction of Cystatin C level $\pm$ SD	P value
2 <sup>nd</sup> vs 1 <sup>st</sup>	-64 $\pm$ NA*	0.00001
3 <sup>rd</sup> vs 2 <sup>nd</sup>	-33.42 $\pm$ 52.54	0.020
4 <sup>th</sup> vs 3 <sup>rd</sup>	1.74 $\pm$ 24.65	0.99
5 <sup>th</sup> vs 4 <sup>th</sup>	-62 $\pm$ 24.79	0.95
6 <sup>th</sup> vs 5 <sup>th</sup>	3.7 $\pm$ 32.92	0.88
7 <sup>th</sup> vs 6 <sup>th</sup>	1.6 $\pm$ 10.12	96.

\*Not applicable because of non-normal distribution of this parameter on day 2 compared with day 1.

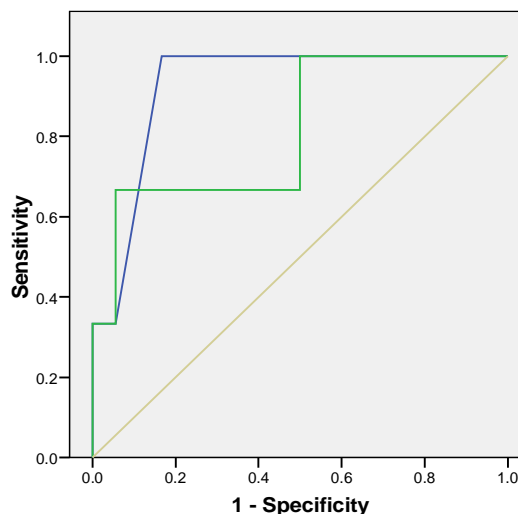
When we compared percent reduction in serum creatinine and cystatin C levels on each day after transplantation versus the previous day, the maximum correlation between the two values was observed between the 2<sup>nd</sup> and 3<sup>rd</sup> post-transplantation day ( $r=0.510$   $P=0.044$ ).

On the 3<sup>rd</sup> postoperative day, with a cut-off value of 75 mL/min for GFR, areas under the ROC curves (AUCs) were 0.926 for creatinine ( $P=0.021$ ) and 0.815 for cystatin C ( $P=0.088$ , figure 3). At this time point, creatinine was more sensitive and specific than cystatin C in estimating GFR with acceptable sensitivity and specificity.

**Table 6:** The effect of transplantation from live donors and cadaver donors on percent reduction in serum creatinine and cystatin C levels per day versus the previous day

Day after transplantation	Mean	SD	P value
Cr <sup>1</sup> 2 <sup>nd</sup> vs 1 <sup>st</sup>	L* -107.91	79.65	0.017
	C** -12.98	31.44	
Cys <sup>2</sup> 2 <sup>nd</sup> vs 1 <sup>st</sup>	L -109.01	227.27	0.860
	C -89.97	102.54	
Cr 3 <sup>rd</sup> vs 2 <sup>nd</sup>	L -53.61	38.107	0.088
	C -97.72	82.36	
Cys 3 <sup>rd</sup> vs 2 <sup>nd</sup>	L -30.69	56.36	0.684
	C -18.34	37.27	
Cr 4 <sup>th</sup> vs 3 <sup>rd</sup>	L 11.04	14.49	0.090
	C -28.02	32.45	
Cys 4 <sup>th</sup> vs 3 <sup>rd</sup>	L 3.41	28.86	0.898
	C 1.43	17.29	
Cr 5 <sup>th</sup> vs 4 <sup>th</sup>	L -7.52	20.02	0.898
	C -8.75	12.70	
Cys 5 <sup>th</sup> vs 4 <sup>th</sup>	L 1.04	31.91	0.729
	C 9.40	17.33	
Cr 6 <sup>th</sup> vs 5 <sup>th</sup>	L -79.19	19.36	0.473
	C -7.64	15.36	
Cys 6 <sup>th</sup> vs 5 <sup>th</sup>	L 3.20	37.76	0.939
	C 5.44	10.39	

\*L: Live donors, \*\* C: Cadaver donors



**Figure 3:** Non-parametric ROC plots for the diagnostic accuracy of serum concentrations of (a) cystatin C (green line) and creatinine (blue line) on the 3<sup>rd</sup> postoperative day, with a cut-off value of 75 mL/min for GFR. The areas under the curve were 0.926 for creatinine ( $P=0.021$ ), and 0.815 for cystatin C ( $P=0.088$ ).

On the 7<sup>th</sup> day after transplantation, AUCs were 0.893 for creatinine and 1.000 for cystatin C ( $P=0.066$  and  $0.017$ , respectively). Cystatin C was more sensitive and specific than creatinine for estimation of GFR with acceptable sensitivity and specificity.

On the 3<sup>rd</sup> postoperative day, Pearson correlation coefficients ( $r$ ) between GFR and serum creatinine and cystatin C showed a more significant negative correlation between GFR and serum creatinine ( $r=-0.859$ ) than cystatin C ( $r=-0.689$ ) ( $P=0.021$  and  $0.088$  respectively). This suggests that serum creatinine concentration was a better predictor of GFR at this time.

On the 7<sup>th</sup> day after transplantation, the results were the opposite, with serum cystatin C being the better predictor (data not shown).

In one patient with acute rejection in the 2<sup>nd</sup> week after transplantation, the increase in cystatin C level began approximately 4 days before serum creatinine began to increase. In another patient with renal artery stenosis and good primary allograft function in the 2<sup>nd</sup> week after transplantation, the increase in cystatin C concentration appeared 2 days earlier than the increase in creatinine.

## Discussion

The sensitive and reliable recognition of changes in renal function is of primary importance in the immediate post-transplant period. Recent studies have suggested that cystatin C might be a potentially better marker of GFR than plasma creatinine.<sup>1,2</sup> The lack of correlation between serum creatinine and cystatin C levels in healthy persons with normal GFR in our study was also reported earlier by others.<sup>3</sup> This lack of correlation might be caused by the fact that serum creatinine level is influenced more than cystatin C by gender and body mass index.

Our results revealed that in the early post-transplantation period, the concentration of plasma cystatin C paralleled creatinine levels. During the first 3 days after transplantation, we observed a strong correlation between serum creatinine levels and GFR. From the 3<sup>rd</sup> day after transplantation, a significant correlation was seen between serum levels of creatinine and cystatin C. It therefore seems that serum cystatin C can be considered a better marker than creatinine for assessing allograft function. Another reason why cystatin C may be a better indicator of GFR than creatinine is that the former is not affected by age, sex, muscle mass, or height.<sup>3</sup>

Bricon et al. reported that cystatin C and creatinine were significantly correlated during the postoperative period in adult patients with renal transplantation.<sup>4</sup> In this study, during the first 4 days after transplantation, plasma cystatin C concentration decreased more rapidly than creatinine because of transtubular leakage of creatinine. Later the decrease in plasma concentration became more prominent for creatinine than cystatin C. By contrast, Bökenkamp and colleagues have claimed several reasons for the observed increase in serum cystatin C levels during the immediate post-transplant period, such as the steroid effect, leakage of cystatin C from the tubuli to the circulation, and impaired filtration of cystatin C

caused by increased protein binding.<sup>5</sup> In our study we observed a significant difference in the percent reduction in creatinine between cadaver versus live donor kidney recipients during the 1<sup>st</sup> post-transplantation day. This difference can be explained by the delayed resumption of graft function caused by possible ischemia or reperfusion injury of renal tubules, which interferes with creatinine secretion.

In the present study, during the first few days after transplantation the reduction in serum creatinine levels was greater than the reduction in serum cystatin C levels. This may be due to the increase in cystatin C serum levels subsequent to high oral and intravenous doses of corticosteroids administered during the first 3 days after transplantation, as reported by others.<sup>6,7</sup> Risch and coworkers demonstrated that patients with renal transplantation who received glucocorticoid medication had higher cystatin C concentrations than two comparable groups with glucocorticoid-free immunosuppression.<sup>6</sup> From the 3<sup>rd</sup> day after transplantation, the correlation between serum creatinine and cystatin C levels became more obvious. Thus the declining effect of corticosteroids on serum cystatin C level and the decrease in creatinine secretion by the 7<sup>th</sup> day after transplantation are also factors that can also contribute to the better sensitivity and specificity of cystatin C compared with creatinine for estimating GFR.

Oddeze and others claimed that cystatin C was not more sensitive than creatinine in detecting early renal impairment in patients with diabetes mellitus with rather stable kidney function and no rapid or significant fluctuations in GFR.<sup>8</sup> Our results showed that on the first few days after kidney transplantation, creatinine was probably still the better marker for detecting transitory changes of GFR.

Zahran, Coll, and their coworkers have shown that cystatin C-based equations were more accurate in predicting GFR in renal transplant recipients than traditional creatinine-based equations.<sup>9,10</sup> Small reductions in GFR appear to be detected more easily by measuring cystatin C than creatinine.<sup>11-15</sup> These findings suggest that measuring serum cystatin C may be a useful way to estimate GFR, especially to detect smaller reductions, and therefore may be useful in the detection of early renal insufficiency in a variety of renal diseases for which early treatment is critical. In our patients, changes in cystatin C preceded change in creatinine in patients with acute rejection or renal artery thrombosis.

In conclusion, we found a good correlation between serum creatinine and cystatin C levels

in the early post-transplantation period. Serum creatinine concentration is probably more sensitive and specific for detecting transitory changes in renal function during the first 3 days after transplantation. However, cystatin C is potentially a better marker for detecting impaired renal function in cases of early allograft dysfunction due to acute rejection or vascular thrombosis. The sensitivity and specificity of cystatin C in detecting changes in GFR increased at the end of the 1<sup>st</sup> week after transplantation.

**Conflict of Interest:** None declared

## References

- 1 Keevil BG, Kilpatrick ES, Nichols SP, Maylor PW. Biological variation of cystatin C: implications for the assessment of glomerular filtration rate. *Clin Chem* 1998; 44: 1535-9.
- 2 Sawyer WT, Canaday BR, Poe TE, et al. A multicenter evaluation of variables affecting the predictability of creatinine clearance. *Am J Clin Pathol* 1982; 78: 832-8.
- 3 Xu X, Zou J, Ding X, Xin D, Ren Y. Clinical value of serum cystatin C by ELISA for estimation of glomerular filtration rate. *J Clin Lab Anal* 2004; 18: 61-4.
- 4 Le Bricon T, Thervet E, Benlakehal M, et al. Changes in plasma cystatin C after renal transplantation and acute rejection in adults. *Clin Chem* 1999; 45: 2243-9.
- 5 Bökenkamp A, van Wijk JA, Lentze MJ, Stoffel-Wagner B. Effect of corticosteroid therapy on serum cystatin C and beta2-microglobulin concentrations. *Clin Chem* 2002; 48: 1123-6.
- 6 Risch L, Herklotz R, Blumberg A, Huber AR. Effects of glucocorticoid immunosuppression on serum cystatin C concentrations in renal transplant patients. *Clin Chem* 2001; 47: 2055-9.
- 7 Risch L, Huber AR. Glucocorticoids and increased serum cystatin C concentrations. *Clin Chim Acta* 2002; 320: 133-4.
- 8 Oddeze C, Morange S, Portugal H, Berland Y, Dussol B. Cystatin C is not more sensitive than creatinine for detecting early renal impairment in patients with diabetes. *Am J Kidney Dis* 2001; 38: 310-6.
- 9 Zahran A, Qureshi M, Shoker A. Comparison between creatinine and cystatin C-based GFR equations in renal transplantation. *Nephrol Dial Transplant* 2007; 22: 2659-8.
- 10 Coll E, Botey A, Alvarez L, Pouch E, et al. Serum cystatin C as a new marker for non-invasive estimation of glomerular filtration rate and as a marker for early renal impairment. *Am J Kidney Dis* 2000; 36: 29-34.
- 11 Newman DJ, Thakkar H, Edwards RG, et al. Serum cystatinC, a replacement for creatinine as biochemical marker maker of GFR. *Kidney Int Suppl* 1994; 47: S20-1.
- 12 Newman DJ, Thakkar H, Edwards RG, et al. Serum cystatin C measured by automated immunoassay: a more sensitive marker of changes in GFR than serum creatinine. *Kidney Int* 1995; 47: 312-8.
- 13 Tian S, kusano E, Ohara T. Cystatin C measurement and its practical use in patients with various renal diseases. *Clin Nephrol* 1997;48:104-8.
- 14 Randers E, Erlandsen EJ, Pedersen OL, et al. Serum cystatin C as an endogenous parameter of the renal function in patients with normal to moderately impaired kidney function. *Clin Nephrol* 2000; 54: 203-9.
- 15 Zahran A, El-Husseini A, Shoker A. Can cystatinC replace creatinine to estimate glomerular filtration rate? A literature review. *Am J Nephrol* 2007; 27: 197-205.