

The Comparison of Glomerular Filtration Rate (GFR) Estimation by Cystatin C with Creatinine-Based Methods in Relation to Isotope-Based Method (^{99m}Tc DTPA Plasma Clearance) as Gold Standard in Patients with Chronic Kidney Disease

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Abstract

Original Research Article

Objective: In this study our main goal is to evaluate the comparison of glomerular filtration rate (GFR) estimation by cystatin C with creatinine-based methods in relation to isotope-based method (^{99m}Tc DTPA plasma clearance) as gold standard in patients with chronic kidney disease. **Method:** This cross-sectional study was carried out in the outpatient department of nephrology, Sir Salimullah Medical College and Mitford hospital; Bangabandhu Sheikh Mujib Medical University and National institute of nuclear medicine and allied sciences (NINMAS), BSMMU, Dhaka from June 2016 to May 2017. A total of 120 Chronic kidney disease patients attending outpatient departments in above institutions were included in the study. **Results:** During the study, in all CKD patients the mean value of MDRD (36 ± 14), CG (31 ± 14) and CKD-EPI (31 ± 15) which were significantly different ($p < 0.001$) from the mean value of m-GFR method (40 ± 14). Whereas significant difference was not observed ($p = 0.571$) between the value of e-GFR-Hoek's (39 ± 16) and m-GFR (40 ± 14). Significant correlation was found between m-GFR with MDRD ($r = 0.724$, $p < 0.001$), CG ($r = 0.697$, $p < 0.001$), CKD-EPI ($r = 0.721$, $p < 0.001$), e-GFR Hoek's ($r = 0.748$, $p < 0.001$). It was observed that e-GFR Hoek's ($r = 0.748$, $p < 0.001$) were more correlated with m-GFR than all other methods. Cystatin C based method [e-GFR Hoek's (AUC=0.965, Sensitivity 97 %, Specificity 76 %, $p < 0.0001$)] had a significantly higher diagnostic accuracy than creatinine based methods [MDRD (AUC=0.914, Sensitivity 100 %, Specificity 23%, $p < 0.0001$), CG (AUC=0.907, Sensitivity 99 %, Specificity 30 %, $p < 0.0001$) and CKD-EPI (AUC=0.908, Sensitivity 98 %, Specificity 30 %, $p < 0.0001$)]. **Conclusion:** From our study we can conclude that, serum cystatin C based estimated GFR showed better correlation with measured GFR in patients with CKD. Its diagnostic accuracy and agreement was found high with measured GFR than creatinine. Thus, cystatin C would be a good alternative marker for estimation of GFR in patients with chronic kidney disease.

Keywords: glomerular filtration rate (GFR), cystatin C, chronic kidney disease.

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INTRODUCTION

Chronic kidney disease (CKD) is a serious public health problem worldwide and is defined as abnormalities of kidney structure or function, present for > 3 months, with implications for health [1].

Glomerular filtration rate (GFR) is the best index available to assess kidney function in disease and in health in an individual. Determination of GFR with high accuracy requires the use of invasive techniques based on measuring the plasma clearance rate injected

substances that are exclusively excreted via glomerular filtration [2]. Several methods including inulin clearance, radioactive like [5, 1], Cr EDTA [1, 2, 5], I-iothalamate and non-radioactive agents like iohexol are used in GFR estimation. Inulin in GFR measurement is disadvantageous because it requires constant intravenous infusion to maintain plasma steady state level; analysis of inulin is technically time consuming, labor intensive, costly and unsuitable for outpatient use [3].

Cystatin C is produced at a constant rate and eliminated by glomerular filtration and it is not secreted, but is reabsorbed by tubular epithelial cells and subsequently catabolized so that it does not return to the blood flow. Its measurement has been proposed as an alternative and more sensitive marker of glomerular filtration rate than creatinine particularly in patients with mild to moderately decrease glomerular filtration rate. ⁴In elderly co-morbid patients and the critically ill patients, serum creatinine assay and exact calibration are variable. In this setting, cystatin C is a promising alternative [5].

In this study our main goal is to evaluate the comparison of glomerular filtration rate (GFR) estimation by cystatin C with creatinine-based methods in relation to isotope based method (^{99m}Tc DTPA plasma clearance) as gold standard in patients with chronic kidney disease.

OBJECTIVE

General Objective

- To assess the comparison of glomerular filtration rate (GFR) estimation by cystatin C with creatinine-based methods in relation to isotope-based method (^{99m}Tc DTPA plasma clearance) as gold standard in patients with chronic kidney disease

Specific Objective

- To detect demographic status of the patients.
- To identify clinical characteristic of the patients.

METHODOLOGY

Study type: This was a cross sectional study.

Place and period of the study: This study was carried out in the outpatient department of nephrology, Sir Salimullah Medical College and Mitford hospital; Bangabandhu Sheikh Mujib Medical University and National institute of nuclear medicine and allied sciences (NINMAS), BSMMU, Dhaka from June 2016 to May 2017.

Study population: A total of 120 Chronic kidney disease patients attending outpatient departments in above institutions.

Inclusion Criteria

- Age > 18 years
- Diagnosed cases of chronic kidney disease.

Exclusion Criteria

- Acute deterioration of kidney function.
- Patients suffering from Hypothyroidism or hyperthyroidism
- Drugs taken like steroid that altered serum cystatin C and serum creatinine level

Study Procedure

All study subjects were informed about the potential risk and benefit of the procedure and informed consent was taken from each patient before the procedure. Good hydration (300-500 ml water) and voiding prior to beginning of study was maintained. 10 ml of blood was taken for serum creatinine and serum cystatin C prior isotope (^{99m}Tc-DTPA) injection. Two syringe counts (pre and post syringe) were taken. Blood was drawn after 1 hour and 3 hour for ^{99m}Tc-DTPA plasma clearance.

Statistical Analysis

Computer based statistical analysis was carried out with appropriate techniques and systems. All data were recorded systematically in preformed data collection form (questionnaire) and quantitative data were expressed as mean and standard deviation and qualitative data were expressed as frequency distribution and percentage. Statistical analysis was performed by using window-based computer software devised with Statistical Packages for Social Sciences (SPSS-20) (SPSS Inc., Chicago, IL, USA). 95% confidence limit was taken. According to the result, the inferential analysis like ANOVA, paired t-test, chi-square test, Pearson's correlation test, linear regression, ROC curve analysis, kappa co-efficient test and data were presented as tables and graphs in result section.

RESULTS

In Table-1 shows demographic status of the patients where mean value among CKD stage 3 and 4 patients was 52 ±12 and 51 ±10, where as in CKD 5 was 46 ±14. The following table is given below in detail:

Table-1: Demographic status of the patients

Variable	CKD 3 (n=63)	CKD 4 (n=44)	CKD 5 (n=5)	P value
Age	52 ±12	51 ±10	46 ±14	0.498
Sex ratio (M:F)	58 : 42	48 : 52	40 : 60	0.439

In Table-2 shows the average values of GFR in different method were calculated. In all CKD patients the mean value of MDRD (36±14), CG (31±14) and CKD-EPI (31±15) which were significantly different (p<0.001) from the mean value of m-GFR method

(40±14). Whereas significant difference was not observed (p=0.571) between the value of e-GFR-Hoek's (39±16) and m-GFR (40±14). The following table is given below in detail:

Table-2: Comparison of e-GFR by different methods (creatinine and cystatin C based) in relation to m-GFR (isotope based) in different stages of CKD

CKD	MDRD	Creatinine based (enzymatic)		Cystatin C based	Isotope based
		CG	CKD-EPI	e-GFR Hoek's	m-GFR
Stage 3	(42±12)	(41±13)	(42±13)	(46±12)	(47±12)
P value	< 0.001	< 0.001	< 0.001	0.062	-
Stage 4	(20±5)	(21±6)	(19±5)	(23±5)	(25±4)
P value	< 0.001	< 0.001	< 0.001	0.134	-
Stage 5	(10±3)	(10±1)	(10±4)	(12±1)	(13±1)
P value	0.022	0.012	0.023	0.061	-

* P value of all methods comparing with * P value m-GFR method Paired t test were done to measure the significance level

In Table-3 shows Pearson's correlation of m-GFR and e-GFR (Creatinine and cystatine C based method). Pearson's correlation was carried out between m-GFR and e-GFR (Creatinine and cystatine C based method) among all CKD patient. Significant correlation was found between m-GFR with MDRD ($r=0.724$,

$p<0.001$), CG ($r=0.697$, $p<0.001$), CKD-EPI ($r=0.721$, $p<0.001$), e-GFR Hoek's ($r=0.748$, $p<0.001$). It was observed that e-GFR Hoek's ($r=0.748$, $p<0.001$) were more correlated with m-GFR than all other methods. The following table is given below in detail:

Table-3: Pearson's correlation of m-GFR and e-GFR (Creatinine and cystatine C based method)

Method		Correlation coefficient r	P value
Creatinine based (enzymatic)	MDRD	0.724	< 0.001
	CG	0.697	< 0.001
	CKD-EPI	0.721	< 0.001
Cystatin C based	e-GFR Hoek's	0.748	< 0.001

Pearson's correlation was carried out to measure the significance level.

In Table-4 shows linear regression between m-GFR and e-GFR (creatinine and cystatine C based method). Linear regression between m-GFR and e-GFR (creatinine and cystatin C based method) were carried

out. In comparison of the correlation coefficient it was found that correlations between m-GFR and MDRD method ($p<0.001$), m-GFR and CG method ($p<0.001$), m-GFR and CKD-EPI method ($p<0.001$) were inferior to m-GFR and e-GFR Hoek's method ($p<0.001$). The following table is given below in detail:

Table-4: Linear regression between m-GFR and e-GFR (creatinine and cystatine C based method)

Method		m-GFR B (unstandardized coefficients)	Beta (Standardized coefficients)	F	P value (95% CI)
Cystatin C	e-GFR Hoek's based	0.790	0.686	97	<0.001 (.631-.948)
Creatinine Based (enzymatic)	MDRD	0.727	0.725	121.84	<0.001 (.597-.858)
	CG	0.698	0.695	103.01	<0.001 (.561-.834)
	CKD-EPI	0.759	0.722	119.635	<0.001 (.621-.896)

In Table-5 shows diagnostic accuracy (area under the ROC curves, Sensitivity, Specificity) and comparison of ROC curves at a cut-off value for GFR 30ml/min/1.73m². ROC curve analysis showed that cystatin C based method [e-GFR Hoek's (AUC=0.965, Sensitivity 97 %, Specificity 76 %, $p<0.0001$)] had a significantly higher diagnostic accuracy than creatinine

based methods [MDRD (AUC=0.914, Sensitivity 100 %, Specificity 23%, $p<0.0001$), CG (AUC=0.907, Sensitivity 99 %, Specificity 30%, $p<0.0001$) and CKD-EPI (AUC=0.908, Sensitivity 98 %, Specificity 30 %, $p<0.0001$)]. The following table is given below in detail:

Table-5: Diagnostic accuracy (area under the ROC curves, Sensitivity, Specificity) and comparison of ROC curves at a cut-off value for GFR30ml/min/1.73m²

Method		AUC	Sensitivity	Specificity	P value
Creatinine based (enzymatic)	MDRD	0.914	100%	23%	<0.001
	CG	0.907	99%	30%	<0.001
	CKD-EPI	0.908	98%	30%	<0.001
Cystatin C based	e-GFR Hoek's	0.965	97%	76%	<0.001

GFR determined with m-GFR method was used as gold standard.

In Figure-1 shows ROC curve on m-GFR with creatinine based and cystatin C based methods. ROC

curve analysis of diagnostic accuracy (sensitivity and specificity) of calculated clearance from the creatinine based (CG, MDRD, and CKD-EPI) and cystatin C based (e-GFR-Hoek's) methods.

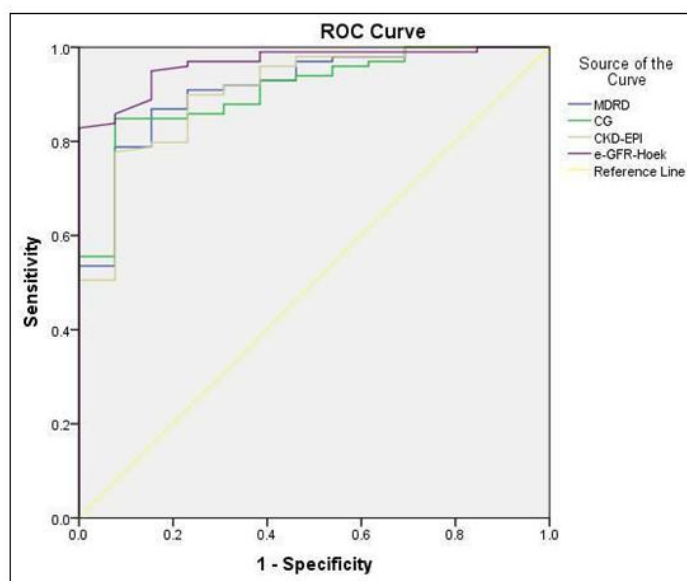


Fig-1: ROC curve on m-GFR with creatinine based and cystatin C based methods

In Table-6 shows accuracy of test methods within 30% of estimated ^{99m}Tc-DTPA (Isotope based) clearance. In patients with CKD 3 statistically higher accuracy found for e-GFR Hoek's (76.4%) compared to accuracy for MDRD method (64.1%), CG (59.3%) and CKD-EPI method (53.2%). In patients with CKD 4 statistically higher accuracy was found for e-GFR

Hoek's (74.2%) compared to accuracy for MDRD method (63.2%), CG (57.1%) and CKD-EPI method (50.2%). In patients with CKD 5 statistically higher accuracy was found for e-GFR Hoek's (64.2%) compared to accuracy for MDRD method (50.2%), CG (53.3%) and CKD-EPI method (51.1%). The following table is given below in detail:

Table-6: Accuracy of test methods within 30% of estimated ^{99m}Tc-DTPA (Isotope based) clearance

Methods		CKD	CKD	CKD
		n=63	n=44	n=5
		50% Accuracy for stage 3	30% Accuracy for stage 4	15% Accuracy for stage 5
Creatinine Based (enzymatic)	MDRD	64.1%	63.2%	50.2%
	CG	59.3%	57.1%	53.3%
	CKD-EPI	53.2%	50.2%	51.1%
Cystatin C based	e-GFR Hoek's	76.4%	74.2%	64.2%

DISCUSSION

Significant correlation was found between m-GFR with MDRD(r=0.724, p<0.001), CG(r=0.697, p<0.001), CKD-EPI(r=0.721, p<0.001) and e-GFR Hoek's (r=0.748, p<0.001). It was observed that e-GFR

Hoek's (r=0.748, p<0.001) were more correlated with m-GFR than all other methods.

ROC curve analysis showed that e-GFR Hoek's had a significantly higher diagnostic accuracy

(AUC=0.966, Sensitivity 97%, Specificity 76%, $p < 0.0001$) than creatinine-based methods (MDRD, CG & CKD-EPI). This findings supports with previous few studies [6].

It has been unambiguously proved that creatinine varies with age, gender and body mass. But in the case of cystatin C, there are conflicting views, some evidence supporting, other evidence opposing [5].

The accuracy estimated ^{99m}Tc -DTPA clearance values differ according to stages of CKD. In patients with CKD 3 statistically higher accuracy found for e-GFR Hoek's (76.4%) compared to accuracy for MDRD method (64.1%), CG (59.3%) and CKD-EPI method (53.2%).

Cystatin C based equation has less bias (1.9 vs. 12.4 ml/min/1.73 m²), and higher precision (25.6 vs. 13.1 mL/min/1.73 m²) and accuracy (92.1% vs. 75.7%) than creatinine based equation (Sun Lee H et al, 2014). Hari P et al, 2014 showed that serum cystatin C based prediction equations appear more precise than those of serum creatinine. Accuracy within 30% ranged from 68.6 to 80.4% for creatinine based formula and 54.0 to 82.9% for cystatin C based formula respectively [7].

A systematic review, identified 10 studies, evaluating the accuracy of 14 different cystatin C based eGFR equations in chronic kidney disease patients. They concluded that the Hoek's equation was the most accurate S cystatin C based equation; most of the S cystatin C based equations showed little improvements in performance compared with the creatinine based MDRD equation [8].

S cystatin C based equations may recommend and advantage over the MDRD equation in chronic kidney diseases [7]. The e-GFR Hoek's based equation has been shown similar to previous reviewed results. As per reviewed this type of comparison study on Bangladeshi population in which GFR were assessed comparing creatinine and cystatin C based equations, employing measured GFR(^{99m}Tc -DTPA plasma clearance) as the Gold standard was not remarkable [9].

The results of this study showed that serum cystatin C is the most useful endogenous marker of GFR. This study compared the diagnostic value of cystatin C, creatinine, and CG and MDRD formulae for GFR in assessment of renal function. The correlation of cystatin C with GFR was comparable to that of creatinine, CG and MDRD formulae. These results suggest that cystatin C is a good marker of renal function in patients with renal impairment

CONCLUSION

From our study we can conclude that, serum cystatin C based estimated GFR showed better correlation with measured GFR in patients with CKD. Its diagnostic accuracy and agreement was found high with measured GFR than creatinine. Thus, cystatin C would be a good alternative marker for estimation of GFR in patients with chronic kidney disease.

REFERENCES

1. WorkGroup CK. Clinical Practice Guideline for the Evaluation and Management of Chronic Kidney Disease. *Kidney Int*, vol. supplement. 2013;30-150.
2. Perrone RD, Steinman TI, Beck GJ, Skibinski CI, Royal HD, Lawlor M, Hunsicker LG. Utility of Radioisotopic filtration markers in chronic renal insufficiency: simultaneous comparison of 125I-Iothalamate, 169Yb-DTPA, ^{99m}Tc -DTPA, and inulin. *American Journal of Kidney Diseases*. 1990 Sep 1;16(3):224-35.
3. Shannon JA, Smith HW. The excretion of inulin, xylose and urea by normal and phlorizinized man. *The Journal of clinical investigation*. 1935 Jul 1;14(4):393-401.
4. Köttgen A, Selvin E, Stevens LA, Levey AS, Van Lente F, Coresh J. Serum cystatin C in the united states: The third national health and nutrition examination survey (NHANES III). *American Journal of Kidney Diseases*. 2008 Mar 1;51(3):385-94.
5. Hojs R, Bevc S, Ekart R, Gorenjak M, Puklavec L. Serum cystatin C-based equation compared to serum creatinine-based equations for estimation of glomerular filtration rate in patients with chronic kidney disease. *Clinical nephrology*. 2008 Jul;70(1):10-7.
6. Tian S, Kusano E, Ohara T, Tabei K, Itoh Y, Kawai T, Asano Y. Cystatin C measurement and its practical use in patients with various renal diseases. *Clinical nephrology*. 1997 Aug;48(2):104-8.
7. Teo BW, Xu H, Wang D, Li J, Sinha AK, Shuter B, Sethi S, Lee EJ. Estimating glomerular filtration rates by use of both cystatin C and standardized serum creatinine avoids ethnicity coefficients in Asian patients with chronic kidney disease. *Clinical chemistry*. 2012 Feb 1;58(2):450-7.
8. Prasad N, Barai S, Gambhir S, Parasar D, Ora M. Comparison of glomerular filtration rate estimated by plasma clearance method with modification of diet in renal disease prediction equation and Gates method. *Indian J Nephrol*, 2012; 22(2):103–107.
9. Anwarul HF. Cystatin C as an endogenous marker of renal function in patients with chronic kidney disease. MD (Thesis) Nephrology. 2010.