iMedPub Journals
http://www.imedpub.com

Annals of Clinical and Laboratory Research
ISSN 2386-5180

2015
Vol. 3 No. 2:11

Clinical Significance of Cystatin C–Based Estimates of Renal Function in Type 2 Diabetic Patients: Review

Temesgen Fiseha
Department of Clinical Laboratory Science, College of Medicine and Health Science, Wollo University, Dessie, Ethiopia

Corresponding author: Temesgen Fiseha

temafiseha@gmail.com

Department of Clinical Laboratory Science, College of Medicine and Health Science, Wollo University, Dessie, Ethiopia

Tel: 033-3115204

Abstract

Chronic kidney disease (CKD) is a common and serious complication of diabetes associated with increased risk of mortality, progression to kidney failure, cardiovascular disease (CVD) and hospitalizations. Accurate estimation of glomerular filtration rate (GFR) is essential for the diagnosis, staging, and management of CKD. Serum creatinine (SCr) level, which is the most commonly used endogenous GFR marker in clinical practice, appears to be influenced by non-renal factors such as age, sex, race and muscle mass, and is not sufficiently sensitive for detecting early renal impairment in diabetics. Serum cystatin C, an alternative endogenous marker less influenced by non-renal factors, has been recently suggested as an early serum marker of detecting changes in GFR and assessing renal impairment at earlier stage. The levels of cystatin C in serum or urine may be elevated in diabetic patients even before the appearance of traditional CKD markers, and it can be used as useful marker for detecting nephropathy in patients with normoalbuminuria (early nephropathy). Moreover, cystatin C-based estimates of renal function may improve risk prediction in diabetics than the commonly used creatinine-based estimates. In this paper, we review from recent literatures the clinical efficiency and relevance of cystatin C as an endogenous renal marker for detecting early renal impairment and for predicting adverse outcomes in type 2 diabetic patients

Keywords: Glomerular filtration rate; Cystatin C; Creatinine; Diabetic nephropathy; Type 2 diabetes

Introduction

Chronic kidney disease (CKD) is a common and serious complication of diabetes associated with increased risk of mortality (both all-cause and cardiovascular), progression to kidney failure, cardiovascular disease (CVD) and hospitalizations [1,2]. Accurate estimation of glomerular filtration rate (GFR), which is usually accepted as the best overall index of kidney function, is essential for the diagnosis, staging and management of CKD [3,4]. GFR cannot be measured directly in humans. The "gold standard" for determining GFR is to measure the clearance of an exogenous substance, such as inulin, ^{51}Cr-EDTA, iohexol, ^{125}I-iothalamate and ^{99m}Tc-diethylelenetriaminepentaacetic acid (^{99c}Tc-DTPA) that are exclusively excreted via glomerular filtration [4,5]. However, these techniques are time-consuming, labor-intensive, expensive, and require administration of substances, so that cannot be generally applied for routine practice. Therefore, the measurement of an endogenous blood substance that is cleared by the kidney is used to estimate GFR.

In clinical practice, GFR is generally estimated based on measurement of endogenous blood substances, and serum creatinine (SCr) level is the most commonly used marker for estimating GFR and assessing renal impairment [6]. However, SCr does not depend solely on GFR, and its concentration is affected by non-renal factors including age, gender, race, muscle mass, medication use, and dietary meat intake [7,8]. SCr is not reabsorbed by the renal tubules, but it is secreted. The substantial tubular excretion of creatinine and the well-known sensitivity of the analytical methods, especially the Jaffe´ method, to interfering substances in the plasma (e.g., acetic acid, acetone, pyruvate, glucose, ascorbic acid, and bilirubin) are other factors that reduce the clinical utility of SCr as a marker of GFR estimate. Although, the measurement of creatinine clearance overcomes some of limitations of SCr, it requires a timed urine

collection, which has proven to be inconvenient and prone to collection errors [4,9]. Equations that take into account patients age, gender, race, and body weight in addition to SCr value have been developed to improve accuracy of creatinine in estimating GFR; however, the results not precise.

Recently, serum cystatin C has been proposed as an alternative marker for estimating GFR. Cystatin C is a single chain, non-glycosylated basic protein that is produced and secreted by all nucleated cells at a constant rate. The low molecular weight of 13 kD and the cationic nature assures the free passage of cystatin C through the glomerulus. It is not secreted, but is reabsorbed and subsequently catabolized by the proximal tubular cells without reentering the circulation [10,11]. These characteristics, together with its constant plasma concentration in the absence or variation of GFR make cystatin C an ideal endogenous marker of GFR. Although its clearance cannot be measured because of its catabolism, serum cystatin C is a good measure of GFR than the most commonly used endogenous marker, SCr [12]. Unlike creatinine, it is not secreted by the renal tubule, is not affected by age, gender and body mass, and does not suffer the same problems with analytical interference [11,13,14]. Because of these properties many investigators have proposed serum cystatin C as a more sensitive marker of GFR than SCr [11,15]. Furthermore, since cystatin C does not depend much on muscle mass, it shows superior performance over SCr as a measure of renal function in specific patient groups such as children, the elderly, and patients with reduced muscle mass [11,14,16]. Moreover, cystatin C may have a role in identifying persons with CKD who have the highest risk for complications than SCr [17].

Since a large proportion of individuals with type 2 diabetes passes through a period of pre-diabetes and may experience renal dysfunction, detection of CKD at early stage is important as early intervention can slow the loss of kidney function, so that improve survival and quality of life. Thus, cystatin C may have diagnostic importance in detecting earlier stages of renal dysfunction in these patients when SCr is unchanged. This review aims to evaluate from recent literature the clinical efficiency and relevance of the endogenous renal marker cystatin C in type 2 diabetic patients.

Cystatin C as a Marker of GFR

While SCr has been widely used to assess renal function in clinical practice, it is found to be defective in detecting early CKD in diabetes. For example in our recent study, up to 56.5% of patients with diabetes and normal SCr levels, have CKD [18]. The primary limitation of SCr as a marker of GFR is that it is affected by many non-renal factors that are not related to GFR. It has been reported that the proportion of variations in cystatin C attributable to extra renal factors in type 2 diabetic patients is considerably lower compared to creatinine [19]. Unlike creatinine, serum cystatin C concentrations in type 2 diabetics were reported to be independent of gender and BMI or by metabolic indices, such as high serum uric acid levels and obesity, but some authors suggest its relation with age [19–22]. Furthermore, the clinical and biochemical parameters associated with serum cystatin C levels in type 2 diabetes are closely linked to those associated with GFR [23].

The inability of creatinine to detect early decline in GFR is due to the fact that SCr levels only begin to rise above the normal range when approximately 50% of renal function is already lost, suggesting that GFR can change before SCr becomes abnormal [24]. Evidences suggest that serum cystatin c may rise faster than creatinine after a fall in GFR and is a reliable endogenous marker for assessing renal function in type 2 diabetic patients with renal impairment [25]. In one study, including 52 type 2 diabetic patients, an early and more significantly increased levels of serum cystatin C than SCr was observed as GFR decreases, which indicated that serum cystatin C might be a useful marker for detecting early renal impairment in diabetic patients.

The overall relationship between the reciprocal cystatin C and GFR was significantly stronger (r = 0.84) than those between SCr and GFR (r = 0.65) and between Cockcroft and Gault (C-G) estimated GFR (eGFR) and GFR (r = 0.70). From this, they concluded that cystatin C may be considered as an alternative and more accurate serum marker than SCr for early identification of patients with reduced GFR [26]. Another study also suggested that serum cystatin C is a better marker for GFR than is SCr in type 2 diabetic patients with reduced GFR, especially in the "creatinine-blind area", reflecting the greater sensitivity of cystatin C as a predictor of GFR in type 2 diabetic patients [27].

It has been also suggested that serum cystatin C is a more sensitive marker for detecting early changes in glomerular filtration in type 2 diabetic patients than creatinine-based measurements [28]. In the above study, cystatin C increased more significantly than SCr as GFR decreased from 120 to 20 mL/min/1.73 m^2, giving a stronger signal in comparison to that of creatinine over the range of the measured GFR [26]. In a study by *Pucci et al.,* the mean cystatin C concentrations showed a step-by-step statistically significant increases as measured GFR decreases [29]. In another similar study from type 2 diabetic patients, mean cystatin C concentrations also showed step wise statistically significant increases as GFR reduces, indicating the sensitivity of serum cystatin C as a marker for detecting early changes in GFR [19]. Thus, any change in GFR is reflected by a step-by-step change in serum cystatin C when creatinine-based measurements are unchanged, and, for this reason, increased levels of serum cystatin C are detectable much earlier in the course of CKD, allowing very early detection of reduction in renal function.

Zhang et al. suggested that cystatin C may be a better indicator of GFR than SCr in diabetic patients, in both the early hyper transfusion stage and in the late renal dysfunction stage [21]. One study from type 2 diabetic patients found that, at 90 and 75 ml/min/1.73 m^2 cut-points, the diagnostic efficiencies of cystatin C (90% and 93%) were better than those of SCr and eGFR calculated from SCr (80%-84% and 86%-89%, respectively; P = 0.01), indicating ability of serum cystatin C for detecting very early renal dysfunction [19]. Another study also found similar findings suggesting that the diagnostic efficiencies of cystatin C (89% and 92%) were better than those of the other variables SCr and calculated eGFR (79%–82% and 85%–86%, respectively; P = 0.01) [29]. From these findings, the above studies suggested convincing evidence that serum cystatin C measurement is an excellent diagnostic test both for identifying diabetic patients

with normal (>90) or near normal (>75 ml/min/1.73 m^2) GFR and for detecting patients with early (<75) or very early (<90 ml/min/1.73 m^2) impairment of GFR than creatinine or eGFR derived from creatinine.

It has been also suggested that cystatin C may be considered as an alternative and more accurate serum marker than creatinine-based estimates in discriminating type 2 diabetic patients with normal GFR from those with reduced GFR [26]. In this study the maximum diagnostic accuracy of serum cystatin C (90%) was significantly better than those of SCr (77%) and eGFR by C-G (85%) in discriminating between the type 2 diabetic patients with a normal GFR (>80 mL/min/1.73 m^2) and those with a reduced GFR (≤ 80 mL/min/1.73 m^2). In this study the authors suggest that, both when the object is to exclude with certainty type 2 diabetic patients with normal or near-normal GFR (specificity) and when it is important to identify those with GFR impairment (sensitivity), serum cystatin C is a better diagnostic tool than SCr and eGFR calculated by C-G [26].

Since a large proportion of individuals with type 2 diabetes passes through a period of pre-diabetes and may experience renal impairment, early diagnosis and management of patients are essential for delaying progression to renal failure and improving outcomes. In *MacIsaac et al.* study, serum cystatin C had the best test characteristics as a screening tool for detecting mild and moderate CKD (iGFR < 90 and < 60 ml/min/1.73 m^2) when compared with creatinine-based methods, highlighting the potential usefulness of screening for moderate or mild CKD in subjects with diabetes by simply measuring serum cystatin C levels [23]. *Walczak et al.* also suggested that the advantage of cystatin C over SCr may be found in early stages of CKD in diabetics, when GFR is still normal or elevated, and cystatin C may be used for early detection of renal function impairment [30].

Furthermore, cystatin C has a potential to be used as an endogenous marker to detect the presence of a progressive loss of renal function over time. *Perkins et al.* suggested that serial measures of serum cystatin C accurately detect trends in renal function in patients with normal or elevated GFR and provide means for studying early renal function decline in diabetes. In type 2 diabetic patients with baseline of 153 mL/min/1.73 m^2, trends in the reciprocal of serum cystatin C concentration have been shown to more closely reflect changes in GFR measured by iothalamate clearance than have creatinine-based estimates of renal function. In this study, the trends in GFR derived from 100/cystatin C were closely correlated with an iothalamate-based reference method over a follow-up period of four years (r = 0.77), whereas the trends for SCr and creatinine-based eGFR (C-G and MDRD) compared poorly with trends in iothalamate clearance (r < 0.35) [31]. Over all, serum cystatin C has superior diagnostic accuracy than those of creatinine-based estimates for the assessment of changes in GFR in type 2 diabetes. Thus, measurement of renal function on the basis of cystatin C levels will optimize early detection, follow-up and monitoring of renal dysfunction in type 2 diabetic patients.

Cystatin C as a Marker for early Diabetic Nephropathy

Traditionally, the earliest manifestation of diabetic nephropathy (DN) in patients with type 2 diabetes is the determination of a small amount of protein albumin in the urine, called microalbuminuria and it is associated with significant renal damage. However, a significant proportion of individuals with type 2 diabetes could have renal impairment as defined by decreased GFR to levels < 60 mL/min/1.73 m^2 without microalbuminuria, the gold standard for diagnosis, and these patients can commonly progress to a significant degree of renal impairment while remaining normoalbuminuric [32,33]. It has been suggested that cystatin C may be elevated in diabetic patients even before the appearance of traditional CKD markers such as albuminuria and creatinine, and can be used as useful marker for detecting nephropathy in patients with normoalbuminuria (early nephropathy).

One recent study *Rao et al*. indicated that levels of cystatin C are related to subclinical renal damage and can be an earlier measurable marker of renal involvement in type 2 diabetes even before the onset of albuminuria [34]. Another study by *Surendar et al.* found that cystatin C levels were highest in type 2 diabetic patients without microalbuminuria, indicating that cystatin C can detect early renal damage even before the development of albuminuria. In this study, cystatin C and cystatin GFR levels were highest and lowest, respectively in type 2 diabetes mellitus subjects with microalbuminuria, which suggested that quantification of cystatin C in serum can be used for predicting onset of nephropathy in type 2 diabetic patients with normoalbuminuria (early nephropathy) [35]. Another study by *Singla et al.* also reported that cystatin C estimation is quite useful and practical method for evaluation of renal impairment in type 2 diabetics even before the onset of microalbuminuria; i.e., early nephropathy [20].

In another study, *Wang et al.* assessed the usefulness of serum homocysteine (Hcy, a major metabolite of methionine and cysteine) and cystatin C assays combined with urine microalbumin excretion rate (UMAER) for detecting early stage renal damage in type 2 diabetic patients. Serum cystatin C level and UMAER were significantly increased in the normoalbuminuric group (UMAER < 30 mg/24 h), the early DN group (EDN; UMAER of 30–300 mg/24 h), and the clinical DN group (CDN; UMAER >300 mg/24 h) as compared to the control group. The serum levels of cystatin C increased more significantly in CDN patients as compared to EDN and normoalbuminuric patients and showed a positive correlation with UMAER, suggesting that cystatin C may contribute synergistically to the occurrence and progression of DN in diabetic patients. Therefore, they suggested that serum cystatin C assays in combination with the UMAER test are clinically useful for detecting early-stage DN and monitoring disease progression, and that this combined assay method will allow a more sensitive and accurate evaluation of renal damage in early-stage DN [36]. In another recent study, levels of cystatin C in serum were significantly higher in microalbuminuric type 2 diabetic patients (1.74 ± 0.66) than normoalbuminuric group

(1.19 ± 0.62, $P < 0.05$) and was found to be higher in patients with GFR ≤ 60 ml/min/1.732 m^2, suggesting that cystatin C is a predictor of early renal damage in patients even before the appearance of microalbuminuria. Authors of this study suggested that the determination of serum cystatin C is a valuable tool to describe GFR loss independently and together with ACR among the patients with diabetes and can optimize the early detection of renal damage [37].

Serum cystatin C is also a sensitive marker for detecting early renal impairment and is a stronger predictor of early onset of nephropathy and its progression than SCr measurements. *Uslu et al.* conducted a study to determine whether the serum cystatin C and activities of some tubular enzymes could be used as screening markers for renal dysfunction in diabetic patients. The levels of serum cystatin C were found elevated in normo- and microalbuminurics as compared to controls. The ROC plot of this study indicated that serum cystatin C had higher sensitivity than SCr and met the criteria for detecting glomerular and tubular dysfunction as screening tests for early diagnosis of DN, which suggested serum cystatin C might be a promising early marker for detecting DN [38]. In one study, evaluating cystatin C as a marker of nephropathy in normo-, micro-, and macroalbuminuric type 2 diabetic patients, cystatin C identified 40% of the patients with DN as compared to 12% by SCr, which indicated that serum cystatin C was a better predictor of nephropathy than SCr in patients with type 2 diabetes [39]. Another study from type 2 diabetic patients found that the diagnostic accuracy of cystatin C (0.76) was greater than that of SCr (0.66) and, as an early prognostic marker of type 2 DN, serum cystatin C was better than SCr in terms of sensitivity and specificity. This indicated that the levels of serum cystatin C may predict early prognostic stages of patients with type 2 DN [40].

In another study, *Suzuki et al.* examined the effectiveness of serum cystatin C as a marker of early DN and CKD in Japanese type 2 diabetic patients. Abnormal serum cystatin C level was found in 28.7%, 54.8% and 80.0%% of normo-, micro-, and macroalbuminuric DN patients, respectively compared to abnormal SCr: 5.7%, 19.0% and 40.0%, respectively. This indicated that SCr increased significantly in macroalbuminuric stage with the proportion of abnormal values being 40.0%, while serum cystatin C had already increased significantly in microalbuminuric stage with the proportion of abnormal values reaching 54.8%, which suggested that serum cystatin C is a good marker for assessing renal injuries earlier than the appearance of SCr. The ROC curve analysis of this study also demonstrated that serum cystatin C had a superior diagnostic ability for detecting all stages of CKD in normo-, micro- and macroalbuminuric DN patients than SCr. The sensitivity and specificity, for example, of serum cystatin C (%) to detect stage 2 CKD (normo: 71.1% and 77.3%; micro- and macroalbuminuric, 78.7% and 83.0%, respectively) were markedly higher than the corresponding values for SCr (normo-, 50.5% and 48.9%; micro- and macroalbuminuric, 56.0% and 54.5% , respectively [41].

Similarly, *Borges eta al.* evaluated cystatin C as a marker of diabetic kidney disease in normoalbuminuric diabetic patients without CKD. Elevated levels of cystatin C was present in 45.9% of normoalbuminuric type 2 diabetic patients with eGFR > 60 mL/min/1.73 m^2, indicating that cystatin C levels in serum could be used as an early biomarker of DN when albuminuria and creatinine-based GFR estimates are insensitive. From this, they concluded that elevated cystatin C levels in diabetics may identify a certain degree of renal dysfunction even when albuminuria and creatinine-eGFR do not reflect CKD [42]. Another recently study from type 2 diabetic patients found that serum cystatin C was the most sensitive and specific marker of macroalbuminuria and damage progress with sensitivity of 70.8% and specificity of 83.3%. Authors from this study demonstrated that, for damage progress, serum cystatin C is the most sensitive and specific marker for follow-up and monitoring DN [43].

On the other hand, the level of urinary cystatin C has been recognized as a marker of early renal damage among patients with type 2 diabetes mellitus [44]. More recently, *Ibrahim et al.* assessed the possible value of urinary cystatin C in early detection of DN in type2 diabetes mellitus. Urinary cystatin C has a diagnostic accuracy of 71.4% to predict the presence of microalbuminuria in early DN. Levels increased significantly in patients with microalbuminuria without any other urinary abnormality and with normal SCr as compared to those without microalbuminuria or any other urinary abnormality, and showed a positive correlation with urinary ACR. They concluded that urinary cystatin C level may be valuable marker for detection of microalbuminuria independent on any other tubular markers and independent of the degree of tubular dysfunction, and that it can be used as a good predictor for the presence of microalbuminuria in early DN [45]. Another study from type 2 diabetic patients found that increased urinary cystatin C was associated with decline in GFR, particularly at the early stages of DN in patients with an eGFR of ≥60 mL/min/1.73 m^2 and was associated with progressed to CKD stage 3 or greater, which indicated that higher urinary cystatin C excretion was a better predictor of early nephropathy [46].

In another recent study, *Jeon et al.* evaluated clinical usefulness of cystatin C levels of serum and urine in predicting renal impairment in normoalbuminuric patients with type 2 diabetes. Cystatin C levels of serum and urine were identified as independent factors associated with eGFR < 60 mL/min/1.73 m^2 in patients with normoalbuminuria, which indicated serum/urinary levels of cystatin C could be a useful marker for early DN in type 2 diabetics with normal albuminuria excretion. The cystatin C levels of serum and urine increased with increasing degree of albuminuria, reaching higher levels in macroalbuminuric patients, indicating that levels of cystatin C in serum/urine might be a useful biomarker for predicting progression of type 2 DN. From this, *Jeon et al.* suggest that cystatin C measurement in urine and serum is a useful, practical, non-invasive tool for the evaluation of renal involvement in the course of diabetes, especially in normoalbuminuric patients [22]. Thus, being elevated in serum or urine, even before the appearance of albuminuria and creatinine-based estimates, cystatin C may serve as a more sensitive early renal marker for predicting onset of nephropathy in patients with

normoalbuminuria (early nephropathy) and its progression in type 2 diabetic patients.

Cystatin C-versus Creatinine-based estimating equations

Recently, several cystatin C-based equations have been developed for estimating GFR, and has been recommend to be used as a confirmatory test for the diagnosis of CKD in patients with mild to moderate decreased GFR as estimated from creatinine (eGFR 45-59 ml/min/1.73 m^2) and no other markers of kidney damage (e.g. ACR <30 mg/g) [47,48]. A recent study also suggest that cystatin C-based estimated GFR (eGFRcys) can be a useful confirmatory marker in those with creatinine-eGFR (eGFRcr) < 60 ml/min/1.73 m^2 and whose ACR is <30 mg/g [49]. Since cystatin C is less affected by non-GFR factors, GFR estimates based on serum cystatin C may have advantages over the creatinine-based estimates in diabetics. In light of this, a few studies have compared the performance of cystatin C- and creatinine-based equations in diabetic patients. *Rigalleau et al.* suggest that the addition of cystatin C measurements to creatinine measurements in assessment of renal function significantly improves the diagnosis and stratification of CKD, and the estimation of GFR in diabetes [50].

The Modification of Diet in Renal Disease (MDRD) and the 2009 CKD Epidemiology Collaboration (CKD-EPI) equations are the most widely used creatinine-based equations to estimate GFR in clinical practice [51,52]. Although these equations are simpler, less costly and easily available compared with the clearance method and are superior to the C-G equation, they pronouncedly underestimated GFR in type 2 diabetic patients [53]. In this regard, it has been suggested that creatinine-based eGFRs are unable to predict the early renal dysfunction in type 2 diabetics compared to cystatin C based eGFR [54]. Creatinine-based the MDRD formula has been recommended by current guidelines for the annual evaluation of renal function in all patients with type 2 diabetes [55]. However, it significantly underestimate GFR, especially at the high-normal range, and therefore lacks sufficient sensitivity to detect an early GFR declines in these patients [53]. Few investigators have compared cystatin C-based equations to the MDRD equations in type 2 diabetes.

In a recent study, *Chudleigh et al.* compared the performance of GFR estimations obtained by cystatin C-based formulae with those obtained by the MDRD formula in predicting isotopically measured GFR (^{51} Cr-EDTA) in 106 patients with type 2 diabetes and normoalbuminuria. The MDRD formula significantly underestimated isotopic GFR, but there was no statistically significant difference between mean eGFRcys and isotopic GFR, and was less biased than the MDRD formula. In this study, the proportion of eGFR results within 10% of isotopic GFR were greater using cystatin C-based formulae than the MDRD [56]. Similar findings were also found in *Hamed et al.* study, demonstrated that the MDRD formula underestimates normal-to-high GFRs (both $P < 0.01$). According to this study, one practical advantage is that basing the diagnosis of CKD on cystatin C-based equation avoids the erroneous estimation of GFR in type 2 diabetic patients without CKD that tends to result with the MDRD [25].

In another recent study, *Oh et al.* compared the performances of GFR estimations measured by cystatin C-based formula with those measured by the creatinine-based formulae in predicting isotopically measured GFR in type 2 diabetic patients according to glycaemic status. Cystatin C-eGFR was less biased and more accurate than creatinine-based eGFR in all patient groups (HbA1c level of ≤ 75 and >95 mmol ⁄ mol). The MDRD-eGFR underestimated isotopic GFR in the group with HbA1c > 95 mmol⁄ mol; however, there was no statistically significant difference between eGFRcys and isotopic GFR. Although the performance of eGFRcys in this study was not superior to MDRD-eGFR in the patient groups with HbA1c < 95 mmol⁄ mol, there is an improved performance of eGFRcys in the groups with HbA1c > 95 mmol⁄ mol [57]. Cystatin C-based eGFR was also found to be correlating well with glycemic status (expressed by the fasting glucose and the HbA1c) in type 2 diabetes in *Mathew et al.* study, indicating that it is the marker of choice for the detection of renal involvement in patients with type 2 diabetes [54]. In routine clinical practice, the prediction of GFR becomes crucial as many diabetics are often poorly controlled and hence GFR and its estimations correlate with HbA1c [58]. In these conditions, the above studies have shown that the cystatin C-based equation has a better diagnostic accuracy, and this accuracy was not altered by high HbA1c level, whereas the MDRD becomes less accurate when HbA1c is >95 mmol⁄ mol.

Like cystatin C levels in serum/urine, eGFRcys has been suggested to show better clinical utility for detecting nephropathy in patients with normoalbuminuria (early nephropathy) as well as for predicting progression of nephropathy in patients with normo-, micro- or macroalbuminuria. In one study *Yarkova et al.* found that CKD defined by eGFRcys < 60 ml/min/1.73 m^2 was higher in 34.3% patients at the normoalbuminuric stage and 37% at the microalbuminuric stage compared to 17.1% and 15.2% by the MDRD. GFR values <90 ml/min/1.73 m^2 were also found in 82.9% and 93.5% of patients with normoalbuminuria and microalbuminuria, respectively using the cystatin C equation compared to 57.1% and 47.8% using the MDRD. From this, *Yarkova et al.* concluded that the determination of early stages of DN by cystatin C–based estimating equations, even before the appearance of albuminuria, is more accurate than that calculated by the MDRD [59]. Another study by *Lee et al*. also found that levels of serum cystatin C and eGFRcys were significantly related to the onset or presence of DN in type 2 diabetic patients with microalbuminuria, which indicated that cystatin C might be a useful marker for detecting early renal damage. The serum levels of cystatin C were statistically related to all DN stages with decrease of 11.7% and 21.0% in the normoalbuminuric group than in the micro- and macroalbuminuric groups ($P = 0.004$ and $P < 0.001$). From this, they suggested that serum cystatin C might rise earlier than SCr in the presence or progression of type 2 DN, and that eGFRcys might be more valuable than eGFRcr in the prediction of the microalbuminuric stage [60].

The creatinine-based CKD-EPI equation, initially developed to correct the systematic underestimation of the MDRD equation, presented a poor performance to estimate GFR in type 2 diabetics, especially for high-normal GFRs [53]. It was also reported to achieve better results in the non-diabetic cohort comparing with

those in the type 2 diabetic cohort [61]. *Singla et al.* evaluated the role of cystatin C as an early biomarker of renal impairment in type 2 DM before the onset of microalbuminuria. They found that eGFRcys is a good biomarker to detect early stage of DN resulting from diabetes than eGFRcr using CKD-EPI, confirming the superiority of the cystatin C-based equations over the CKD-EPI equation in patients with type 2 diabetes [20]. However, the superiority of the cystatin C-based equations over the MDRD and CKD-EPI creatinine equations has not been confirmed by *Iliadis et al*. study. In this study, the proportion of eGFR results within 10% and 30% of mGFR (defined by ^{51}Cr-EDTA-measured GFR) was the same when using the MDRD, CKD-EPI and cystatin C-based formulae for the diagnosis of mild and moderate CKD (mGFR <90 and <60 ml/min/1.73 m^2, respectively). Furthermore, the CKD-EPI creatinine-based equation was found to be the least biased, the most precise and the most accurate equation in type 2 diabetic patients with mGFR ≥90 ml/min/1.73 m^2 [62].

Cystatin C as a Marker for Predicting Adverse Outcomes

Although all the reviewed studies suggest that cystatin C is a promising renal marker, it is important to document the advantages of cystatin C to improve patient outcome, which ultimately depends on the results of patient outcome studies. There is growing evidence suggesting that the addition of cystatin C to traditional CKD markers such as albuminuria and eGFRcr significantly improves risk prediction [49]. Being less influenced by non-renal factors, cystatin C might be a promising early marker for predicting adverse clinical outcomes in diabetic patients. Recently, a study by *Tsai et al.* showed that a higher prevalence of reduced kidney function was found among persons with diabetes using eGFRcys compared to eGFRcr (22.0% and 16.5%, respectively). More persons with diabetes were reclassified from preserved kidney function by eGFRcr to reduced kidney function by eGFRcys, and it was strongly associated with diabetic complications, including DR than that based on creatinine. In this study, the relationship between eGFRcys and all-cause and cardiovascular mortality was stronger than the corresponding relationship with eGFRcr [63].

In another similarly, eGFRcys identified a quite different population with CKD compared with the eGFRcr (79 identical/181 not identical for MDRD, and 86 identical/178 not identical for CKD-EPI). In this study, only the eGFRcys based definition of CKD was an independent risk predictor for cardiovascular events in diabetic patients and indicated a potentially better clinical utility for cardiovascular risk prediction than the commonly used estimates, eGFRcr [64]. In another study, *Krolewski et al.* showed that CKD staging by both eGFRcr and eGFRcys resulted in significant discrepancies in approximately 25–35% of patients. For young or middle-aged diabetic patients in CKD stages 1–3 (based on routine measurements of SCr); a secondary assessment of renal function based on serum cystatin C significantly improves ESRD risk stratification. From this they suggested that the measurement of serum cystatin C in diabetic patients has value in assessing risk of ESRD [65].

Recently, CKD outcome studies from type 2 diabetic patients have established that cystatin C can improve risk stratification. In a recent study, *Senghor et al*. demonstrated that serum Cystatin C, a preclinical marker of renal dysfunction, can be used as a predictive marker of diabetic dyslipidemia and cardiovascular risk in poorly controlled Type 2 Diabetic patients [66]. In another study, *Lee et al.* evaluated the association between cystatin C and various biomarkers in estimating risk for CVD in type 2 diabetic patients, and found that cystatin C significantly correlated with various emerging biomarkers for CVD [67]. In the same context, a recent study, including 42 patients with type 2 diabetes, showed that only cystatin C level was associated with increased risk of CVD, which indicated that cystatin C may be a valuable and useful marker for predicting CVD in type 2 diabetic patients [68]. Another study also demonstrated that measurement of the levels of serum cystatin C is a useful, practical, noninvasive technique for the evaluation of renal involvement and might be related with a risk for cardiovascular events in patients without nephropathy in the course of diabetes, especially in patients with normoalbuminuria [34].

More recently, *Pavkov et al.* compared values of baseline serum cystatin C, SCr, and measured GFR (mGFR) for predicting ESRD in patients with type 2 diabetes and elevated albuminuria. They found that serum cystatin C was a better predictor of ESRD than mGFR or SCr, and the predictive ability of serum cystatin C remained superior to the other filtration markers in subjects with normal or high-normal GFR, suggesting that cystatin C may allow earlier stratification of patients at high risk for progression to kidney failure. *Pavkov et al.* also suggested that the predictive value of serum cystatin C for ESRD in patients with type 2 diabetes may be enhanced beyond the gold-standard measurement of GFR because of additional renal and non-renal information cystatin C may impart [69]. Another recent study by *Liu et al*. showed that the prevalence of coronary artery disease (CAD), cerebral infarction (CI) and lower limb ischemia (LLI) caused by peripheral arterial disease (PAD) increased with cystatin C, especially the prevalence of LLI. From this, *Liu et al*. concluded that apart from renal function the detection of cystatin C concentration is of great value for screening out the patients with the angiostenosis risk of lower limb to prevent foot ulceration and amputation [70].

Conclusion

Diagnostic markers which reflect renal impairment at early stage is important as early intervention can slow the loss of kidney function and reduce adverse clinical outcomes. Serum cystatin C rise faster than SCr after a fall in GFR and has the potential to accurately detect earlier changes in GFR compared to SCr, serving as an excellent endogenous marker of early renal dysfunction in type 2 diabetes. Its levels in serum or urine might be also elevated in diabetic patients even before the appearance of microalbuminuria, and can be used as useful marker for detecting nephropathy in patients with normoalbuminuria (early nephropathy), which will allow early intervention and management of type 2 diabetic patients with DN. Cystatin C

may serve as a marker for monitoring kidney function in type 2 diabetic patients with normo-, micro-and macroalbuminuric DN. Cystatin C and cystatin C-based estimates of GFR predicted risk of progression to end stage renal disease (ESRD), cardiovascular events, CVD, and diabetic lower arterial lesion (one of the major causes for foot ulceration and amputation in diabetes) in type 2 diabetes more strongly than the commonly used creatinine-based estimates. We suggest that future studies utilize cystatin C to study the onset and progression of diabetic nephropathy and to predict outcomes, in addition to further assessing its ability to predict early renal dysfunction in type 2 diabetic patients and for predicting nephropathy in patients with normoalbuminuria (early nephropathy).

References

1 Ninomiya T, Perkovic V, de Galan BE, Zoungas S, Pillai A, et al. (2009) Albuminuria and kidney function independently predict cardiovascular and renal outcomes in diabetes. J Am Soc Nephrol 20: 1813-1821.

2 Go AS, Chertow GM, Fan D, McCulloch CE, Hsu CY (2004) Chronic kidney disease and the risks of death, cardiovascular events, and hospitalization. N Engl J Med 351: 1296-1305.

3 National Kidney Foundation (2002) K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. Am J Kidney Dis 39: S1-266.

4 Stevens LA, Coresh J, Greene T, Levey AS (2006) Assessing kidney function--measured and estimated glomerular filtration rate. N Engl J Med 354: 2473-2483.

5 Stevens LA, Levey AS (2009) Measured GFR as a confirmatory test for estimated GFR. J Am Soc Nephrol 20: 2305-2313.

6 Miller WG (2009) Estimating glomerular filtration rate. Clin Chem Lab Med 47: 1017-1019.

7 Perrone RD, Madias NE, Levey AS (1992) Serum creatinine as an index of renal function: new insights into old concepts. Clin Chem 38: 1933-1953.

8 Ferguson MA, Waikar SS (2012) Established and emerging markers of kidney function. Clin Chem 58: 680-689.

9 Salgado JV, Neves FA, Bastos MG, França AK, Brito DJ, et al. (2010) Monitoring renal function: measured and estimated glomerular filtration rates - a review. Braz J Med Biol Res 43: 528-536.

10 Séronie-Vivien S, Delanaye P, Piéroni L, Mariat C, Froissart M, et al. (2008) Cystatin C: current position and future prospects. Clin Chem Lab Med 46: 1664-1686.

11 Filler G, Bökenkamp A, Hofmann W, Le Bricon T, Martínez-Brú C, et al. (2005) Cystatin C as a marker of GFR--history, indications, and future research. Clin Biochem 38: 1-8.

12 Laterza OF, Price CP, Scott MG (2002) Cystatin C: an improved estimator of glomerular filtration rate? Clin Chem 48: 699-707.

13 Pham-Huy A, Leonard M, Lepage N, Halton J, Filler G (2003) Measuring glomerular filtration rate with cystatin C and beta-trace protein in children with spina bifida. J Urol 169: 2312-2315.

14 Fliser D, Ritz E (2001) Serum cystatin C concentration as a marker of renal dysfunction in the elderly. Am J Kidney Dis 37: 79-83.

15 Christensson AG, Grubb AO, Nilsson JA, Norrgren K, Sterner G, et al. (2004) Serum cystatin C advantageous compared with serum creatinine in the detection of mild but not severe diabetic nephropathy. J Intern Med 256: 510-518.

16 Zaffanello M, Franchini M, Fanos V (2007) Is serum Cystatin-C a suitable marker of renal function in children? Ann Clin Lab Sci 37: 233-240.

17 Peralta CA, Katz R, Sarnak MJ, Ix J, Fried LF, et al. (2011) Cystatin C identifies chronic kidney disease patients at higher risk for complications. J Am Soc Nephrol 22: 147-155.

18 Fiseha T, Kassim M, Yemane T (2014) Chronic kidney disease and underdiagnosis of renal insufficiency among diabetic patients attending a hospital in Southern Ethiopia. BMC Nephrol 15: 198.

19 Godwill O, Ntuen N (2013) Use of Serum Cystatin C in Assessment of Early Deterioration of Renal Function in Type 2 Diabetic Industrial Workers in Port Harcourt, Nigeria. IOSR-JDMS 8: 27–35.

20 Singla K, Sodhi KS, Pandey R, Singh J, Sharma P (2014) The utility of serum cystatin C in the diagnosis of early diabetic nephropathy. J Pharm Biomed Sci 4: 84–87.

21 Zhang PP, Zhan JF, Xie HL, Li LS, Liu ZH (2010) Evaluation of glomerular filtration rate using cystatin C in diabetic patients analysed by multiple factors including tubular function. J Int Med Res 38: 473-483.

22 Jeon YK, Kim MR, Huh JE, Mok JY, Song SH, et al. (2011) Cystatin C as an early biomarker of nephropathy in patients with type 2 diabetes. J Korean Med Sci 26: 258-263.

23 Macisaac RJ, Tsalamandris C, Thomas MC, Premaratne E, Panagiotopoulos S, et al. (2007) The accuracy of cystatin C and commonly used creatinine-based methods for detecting moderate and mild chronic kidney disease in diabetes. Diabet Med 24: 443-448.

24 Shemesh O, Golbetz H, Kriss JP, Myers BD (1985) Limitations of creatinine as a filtration marker in glomerulopathic patients. Kidney Int 28: 830-838.

25 Hamed EO, Zaky NA, Kamal YM (2011) Simple cystatin-C formula for estimation of glomerular filtration rate in chronic kidney disease in diabetic patients. AAMJ 9: 81–89.

26 Mussap M, Dalla Vestra M, Fioretto P, Saller A, Varagnolo M, et al. (2002) Cystatin C is a more sensitive marker than creatinine for the estimation of GFR in type 2 diabetic patients. Kidney Int 61: 1453-1461.

27 El-Kafrawya NA, Shohaiba AA, El-Deenb SMK, Barbarya HE, Seleema AS (2014) Evaluation of serum cystatin C as an indicator of early renal function decline in type 2 diabetes. Menoufia Med J 27: 60–65.

28 Rinnno H, Iijima T, Takeda Y, Shimizu A, Kobata M, et al. (2002) Serum cystatin c levels in hypertensive type 2 diabetic nephropathy patients after treatment with angiotensin converting enzyme (ACE) inhibitor temocapril. Acta Nephrol 16: 62–65.

29 Pucci L, Triscornia S, Lucchesi D, Fotino C, Pellegrini G, et al. (2007) Cystatin C and estimates of renal function: searching for a better measure of kidney function in diabetic patients. Clin Chem 53: 480-488.

30 Walczak K, Rosniak-Bak K, Paradowski M, Moczulski D (2009) Cystatin C as a marker of renal function in diabetic kidney disease. Diabet Dosw Klin 9: 69–72.

31 Perkins BA, Nelson RG, Ostrander BEP, Blouch KL, Krolewski AS, et al. (2005) Detection of Renal Function Decline in Patients with Diabetes and Normal or Elevated GFR by Serial Measurements of Serum Cystatin C Concentration: Results of a 4-Year Follow-Up Study. J Am Soc Nephrol 16: 1404–1412.

32 Macisaac RJ, Tsalamandris C, Panagiotopoulos S, Smith TJ, Mcneil KJ (2004) Nonalbuminuric Renal Insufficiency in Type 2 Diabetes. Diabetes Care 27: 195–200.

33 Walczak K, Sodolska M, Materek I, Krysicka A, Moczulski D (2008) Impaired renal function in type 2 diabetes patients in the absence of increased urine albumin excretion rate. Diabet Dosw Klin 8: 165–168.

34 Rao GSN, Abayambigai J, Sruti E, Sowmiya K (2014) Early prediction of nephropathy and cardiovascular diseases in indian patients with type 2 diabetes mellitus. Int J Med Sci Public Heal 3: 1523–527.

35 Surendar J, Anuradha S, Ashley B, Balasubramanyam M, Aravindhan V, et al. (2009) Cystatin C and cystatin glomerular filtration rate as markers of early renal disease in Asian Indian subjects with glucose intolerance (CURES-32). Metab Syndr Relat Disord 7: 419–425.

36 Wang T, Wang Q, Wang Z, Xiao Z, Liu L (2013) Diagnostic value of the combined measurement of serum hcy, serum cys C, and urinary microalbumin in type 2 diabetes mellitus with early complicating diabetic nephropathy. ISRN Endocrinol 2013: 407452.

37 Dayanidhi S, Ramadevi K, Periyandavar I (2015) Serum Cystatin C and Microalbumin in the detection of early Nephropathy in type 2 Diabetic patients. Int J Pharm Bio Sci 6: 1208–1214.

38 Uslu S, Efe B, AlataÅŸ O, KebapÃ§i N, Colak O, et al. (2005) Serum cystatin C and urinary enzymes as screening markers of renal dysfunction in diabetic patients. J Nephrol 18: 559-567.

39 Mojiminiyi OA, Abdella N, George S (2000) Evaluation of serum cystatin C and chromogranin A as markers of nephropathy in patients with type 2 diabetes mellitus. Scand J Clin Lab Invest 60: 483-489.

40 Shimizu A, Horikoshi S, Rinnno H, Kobata M, Saito K, et al. (2003) Serum cystatin C may predict the early prognostic stages of patients with type 2 diabetic nephropathy. J Clin Lab Anal 20: 164–167.

41 Suzuki Y, Matsushita K, Seimiya M, Yoshida T, Sawabe Y, et al. (2012) Serum cystatin C as a marker for early detection of chronic kidney disease and grade 2 nephropathy in Japanese patients with type 2 diabetes. Clin Chem Lab Med 50: 1833-1839.

42 Borges RL, Hirota AH, Quinto BM, Ribeiro AB, Zanella MT, et al. (2010) Is cystatin C a useful marker in the detection of diabetic kidney disease? Nephron Clin Pract 114: c127-134.

43 Assal HS, Tawfeek S, Rasheed EA, El-Lebedy D, Thabet EH (2013) Serum cystatin C and tubular urinary enzymes as biomarkers of renal dysfunction in type 2 diabetes mellitus. Clin Med Insights Endocrinol Diabetes 6: 7-13.

44 Rao X, Wan M, Qiu C, Jiang C1 (2014) Role of cystatin C in renal damage and the optimum cut-off point of renal damage among patients with type 2 diabetes mellitus. Exp Ther Med 8: 887-892.

45 Ibrahim MA, Ahmed YS, El-Shinnawy HA, Maseeh IYAA, Makkeyah YM, et al. (2015) Value of Urinary Cystatin C in early detection of Diabetic nephropathy in type 2 diabetes mellitus. Int J Adv Res Biol Sci 2: 211–223.

46 Kim SS, Song SH, Kim IJ, Jeon YK, Kim BH, et al. (2013) Urinary cystatin C and tubular proteinuria predict progression of diabetic nephropathy. Diabetes Care 36: 656-661.

47 (2014) National Institute for Health and Care Excellence. Chronic kidney disease: early identification and management of chronic kidney disease in adults in primary and secondary care, NICE.

48 Inker LA, Schmid CH, Tighiouart H, Eckfeldt JH, Feldman HI, et al. (2012) Estimating glomerular filtration rate from serum creatinine and cystatin C. N Engl J Med 367: 20–9.

49 Waheed S, Matsushita K, Astor BC, Hoogeveen RC, Ballantyne C, et al. (2013) Combined Association of Creatinine, Albuminuria, and Cystatin C with All-Cause Mortality and Cardiovascular and Kidney Outcomes. Clin J Am Soc Nephrol. 8: 434–442.

50 Rigalleau V, Beauvieux M-C, Moigne F, Lasseur C, Chauveau P, et al. (2008) Cystatin C improves the diagnosis and stratification of chronic kidney disease, and the estimation of glomerular filtration rate in diabetes. Diabetes Metab 34: 482–489.

51 Levey AS, Bosch JP, Lewis JB, Greene T, Rogers N, et al. (1999) A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation: Modification of Diet in Renal Disease Study Group. Ann Intern Med 130 :461–470.

52 Levey A, Stevens L, Schmid C, Zhang L, Castro AF, et al. (2009) A new equation to estimate glomerular ltration rate. Ann Intern Med150: 604–614.

53 Silveiro SP, Araújo GN, Ferreira MN, Souza FDS, Yamaguchi HM, et al. (2011) Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) Equation Pronouncedly Underestimates Glomerular Filtration Rate in Type 2 Diabetes. Diabetes Care 34: 2353–2355.

54 Mathew R, Pai VR, Vijayakumar T (2013) Comparative evaluation of conventional and new biomarkers of renal involvement in patients with type 2 diabetes mellitus and hypertension-a pilot study. Int J Pharm Bio Sci 4: 742–750.

55 (2011) American Diabetes Association. Standards of medical care in diabetes - 2011. Diabetes Care 34: S11–S61.

56 Chudleigh RA, Ollerton RL, Dunseath G, Peter R, Harvey JN, et al. (2009) Use of cystatin C-based estimations of glomerular filtration rate in patients with type 2 diabetes. Diabetologia. 52: 1274–1278.

57 Oh S, Lee J, Ha W, Jeong S, Yim H, et al. (2012) Comparison of cystatin C- and creatinine-based estimation of glomerular filtration rate according to glycaemic status in Type 2 diabetes. Diabet Med 29: e121–125.

58 Rigalleau V, Lasseur C, Raffaitin C, Perlemoine C, Barthe N, et al. (2006) Glucose Control Influences Glomerular Filtration Rate and Its Prediction in Diabetic Subjects. Diabetes Care 29: 1491–1495.

59 Yarkova N, Borovkov N, Zanozina O, Nosov V (2013) Cystatin C in the Diagnosis of Chronic Kidney Disease in Patients with Type 2 Diabetes Mellitus. Clinical Med 5: 89–92.

60 Lee B-W, Ihm S-H, Choi M-G, Yoo H-J (2007) The comparison of cystatin C and creatinine as an accurate serum marker in the prediction of type 2 diabetic nephropathy. Diabetes Res Clin Pract 78: 428–434.

61 Liu X, Gan X, Chen J, Lv L, Li M (2014) A New Modified CKD-EPI Equation for Chinese Patients with Type 2 Diabetes. PLoS ONE 9: e109743.

62 Iliadis F, Didangelos T, Ntemka A, Makedou A, Moralidis E, et al. (2011) Glomerular filtration rate estimation in patients with type 2 diabetes: creatinine- or cystatin C-based equations? Diabetologia 54: 2987–2994.

63 DeBoer IH, Ronit K, Jie J, Linda F, Bryan K, et al. (2009) Cystatin C, Albuminuria, and Mortality among older adults with diabetes. Diabetes Care 32:1833–1838.

64 Schöttker B, Herder C, Müller H, Brenner H, Rothenbacher D (2012) Clinical Utilityof Creatinine and Cystatin C–Based Definition of Renal Function for Risk Prediction of Primary Cardiovascular Events in Patients With Diabetes. Diabetes Care 35: 879–886.

65 Krolewski AS, Warram JH, Forsblom C, Smiles AM, Thorn L, et al. (2012) Serum Concentration of Cystatin C and Risk of End-Stage Renal Disease in Diabetes. Diabetes Care 35: 2311–2316.

66 Senghor A, William E, Jeevanathan, Naveen C, Krishanan S (2013) Correlation of Cystatin C and Cardiovascular Risk Markers in Uncontrolled Type 2 Dm. IJPCR 5: 79–82.

67 Lee S-H, Lee K-W, Kim ES, Park YR, Kim H-S, et al. (2008) Cystatin C is a Valuable Marker for Predicting Future Cardiovascular Diseases in Type 2 Diabetic Patients. Korean Diabetes J 32: 488–497.

68 Triki S, Fekih O, Hellara I, Neffati F, Douki W, et al. (2013) Association between serum cystatin C levels and cardiovascular disease in type 2 diabetic patients. Ann Biol Clin 71: 438–442.

69 Pavkov ME, Knowler WC, Hanson RL, Williams DE, Lemley KV, et al. (2013) Comparison of Serum Cystatin C, Serum Creatinine, Measured GFR, and Estimated GFR to Assess the Risk of Kidney Failure in American Indians With Diabetic Nephropathy. Am J Kidney Dis 62: 33–41.

70 Liu F, Shen J, Zhao J, Zeng H, Li L, et al. (2013) Cystatin C: A Strong Marker for Lower Limb Ischemia in Chinese Type 2 Diabetic Patients? PLoS ONE 8: e66907.