Cystatin C as a renal function estimator in advanced chronic renal failure stages

M. V. Martín, S. Barroso, O. Herráez*, F. de Sande* and F. Caravaca
Nephrology and *Biochemistry Departments. Infanta Cristina Hospital. Badajoz. Spain.

SUMMARY

Serum cystatin C (CysC) has been shown to be more accurate than serum creatinine (Cr) in estimating renal function, especially in patients with mildmoderate chronic renal failure. However, it is unknown whether CysC provides or not any advantage over Cr in severe chronic renal failure. The aim of this study was to establish the accuracy of CysC in estimating the glomerular filtration rate (GFR) in advanced chronic renal failure patients. The study group consisted of 94 patients (57 females, mean age 61 ± 16 years) with advanced chronic renal failure. None of them had thyroid dysfunction or was on corticoid treatment. GFR was measured by Tc^{99m}DTPA, and simultaneously, serum CysC (particle enhanced immunonephelometry) and Cr (modified Jaffe's kinetic reaction) were also determined. Serum Cr and CysC levels were correlated with GFR, and the influences of age, sex and diabetes on these correlations were analyzed. The predictive value of CysC and Cr to estimate a GFR less than 15 ml/min/1.73 m^2 was analyzed by measuring the crea-under-the-curve (AUC) with Receiver-Operating Characteristics (ROC) plots. The mean GFR was 16.49 ± 4.65 ml/min/1.73 m^2. The mean concentrations of Cr and CysC were 4.19 ± 1.19 mg/dl and 3.44 ± 0.73 mg/dl, respectively. Both Cr and CysC correlated significantly with GFR (R = 0.49, p < 0.0001 and R = 0.52, p < 0.0001, respectively). Age and sex influenced on the correlation between Cr and GFR, but these demographic characteristics did not influence on the correlation between CysC and GFR. The AUC for the prediction of a GFR less than 15 ml/min/1.73 m^2 with serum Cr was 0.675 (p = 0.004), while with CysC was 0.633 (p = 0.030). In conclusion, both serum Cr and CysC are highly inaccurate markers of renal function in advanced chronic renal failure patients.

Key words: ***Chronic renal failure. Creatinine. Cystatin C. Glomerular filtration rate.***

CISTATINA C COMO ESTIMADOR DE LA FUNCIÓN RENAL EN ESTADIOS AVANZADOS DE ENFERMEDAD RENAL CRÓNICA

RESUMEN

Numerosos estudios han demostrado que los niveles plasmáticos de cistatina C (CisC) son más precisos que los niveles de creatinina (Cr) en la estimación del fil-

Correspondence: María Victoria Martín
Servicio de Nefrología
Hospital Infanta Cristina
06080 Badajoz
E-mail: mvmh-b(a)hotmail.com

trado glomerlar (FG). Este hecho parece particularmente cierto en casos de disfunción renal leve-moderada, aunque es menos conocida su precisión en casos de insuficiencia renal avanzada. El objetivo de este estudio fue determinar la precisión de la CisC plasmática para estimar el FG en pacientes con enfermedad renal crónica avanzada. Se estudiaron 94 pacientes (57 mujeres) con insuficiencia renal avanzada. La edad media fue 61 ± 16 años. Ningún paciente presentaba alteraciones funcionales tiroideas o estaba en tratamiento con corticoides. Se midió el FG mediante Tc^{99m}DTPA, determinando simultáneamente las concentraciones plasmáticas de Cr (reacción cinética modificada de Jaffé), y CisC (inmunonefelometría). Los niveles de Cr y CisC se correlacionaron con el FG, y se analizó la influencia del sexo, edad y diagnóstico de diabetes en los residuales de estas correlaciones. El valor predictivo tanto de la Cr como de la CisC para diagnosticar un FG < 15 ml/min/1,73 m^2 fue analizado mediante curvas ROC, determinando las áreas bajo las curvas y sus significaciones estadísticas. El FG medio fue 16,49 ± 4,65 ml/min/1,73 m^2. Las concentraciones medias de Cr y CisC fueron respectivamente: 4,19 ± 1,19 mg/dl y 3,44 ± 0,73 mg/dl. Tanto la Cr como la CisC se correlacionaron significativamente con el FG (R = 0,49; p < 0,0001 y R = 0,52; p < 0,0001 respectivamente). La edad y sexo influyeron en la correlación entre Cr y FG, pero no en la correlación entre CisC y FG. El área bajo la curva de predicción para un FG < 15 ml/min/1,73 m^2 con la Cr fue 0,675 (p = 0,004), mientras que con la CisC fue 0,633 (p = 0,030). En conclusión, las concentraciones séricas de CisC son igual de imprecisas que las de Cr para la estimación del FG en la enfermedad renal crónica avanzada.

Palabras clave: ***Creatinina. Cistatina C. Filtrado glomerular. Insuficiencia renal crónica.***

INTRODUCTION

Measurement of plasma substances which concentration increases nearly in a proportional way as renal function level is the easiest and commonest way of indirect estimation of glomerular filtration. Measurement of serum creatinine level (Cr) is, to date, the most used method for estimating renal function. However, serum creatinine level depends on renal excretion and on factors related with muscle production, such as age, gender, anthropometrical and ethnic characteristics,[1,2] besides variable tubular secretion that occurs in advanced chronic renal failure stages.[3]

Cystatin C (CysC) is a substance constantly produced by most nucleated cells and exclusively excreted by the kidney.[4] These characteristics have made of it a perfect candidate to be a marker of renal function.[5] By contrast with Cr, CysC levels are not influenced by age, gender, or muscle mass.[6] A meta-analysis pooling together the experience of a number of investigations on the topic concludes that CysC is more accurate than Cr for renal function estimation.[7] CysC seems particularly useful to discriminate between mild to moderate renal function impairments,[8-10] although its usefulness to estimate glomerular filtration in more advanced renal failure stages is less well known.

The aim of the present study was to compare the accuracy of Cr versus CysC as a marker of glomerular filtration in advanced chronic renal failure stages.

MATERIAL AND METHODS

Patients

Ninety-four patients with advanced chronic renal failure referred to the pre-dialysis clinic were studied. Mean age was 61 ± 16 years (57 female and 37 male patients). There were no exclusion criteria. Causes of renal failure were: unknown (26 patients), glomerulopathy (17 patients), diabetic nephropathy (19 patients), chronic interstitial nephropathy (23 patients), polycystic renal disease (4 patients), ischemic nephropathy (3 patients), and other diagnoses (1 patient).

All patients were on a stable condition, with comorbidity indexes estimated by Davies's *et al.* method[11] as follows: grade 0 (51%), grade 1 (44%), and grade 2 (5%).

There were no ethnic differences in the study group, all were of Caucasian origin. None of the patients had thyroid functional impairment or was

on corticosteroid therapy. Anthropometrical characteristics were as follows: mean weight 72.4 ± 14.2 Kg; height 1.61 ± 0.10 m, body mass index 27.8 ± 5.4 kg/m^2, and body surface area 1.76 ± 0.19 m^2.

Glomerular filtration measurement and biochemical study

All patients had a determination of glomerular filtration (GF) by means of isotopic dilution of Tc^{99m} diethylenetriamine pentacetic acid (Tc^{99m}DTPA). It was intravenously administered at a dose of 50 µCi/Kg, and blood samples were obtained at 120, 180, and 240 minutes. The Bröchner-Mortensen method was used to calculate GF.[12]

Patients were fasting and in the first blood drawn urea, creatinine, albumin (Hitachi D2400 Roche Diagnostics, Germany) and cystatin C were also determined. Creatinine was measured by the modified Jaffé's kinetic reaction. Cystatin C was measured by immunonephelometry.

Design and statistical methods

Both Cr and CysC were correlated with glomerular filtration, determining Pearson's coefficient and graphically analyzing the influence of age and gender. The standardized residuals of these correlations were obtained, that is the error or random effect in predictive unfitness of the models. In order to determine which variables unfit the best correlation between Cr or CysC with glomerular filtration a multiple linear regression analysis was done in which the dependent variable was the standardized residual and the predictive studied variables were: age, gender, albumin, diabetes diagnosis, and body mass index.

To determine the usefulness of both Cr and CysC in predicting a glomerular filtration rate lower than 15 mL/min/1.73 m^2 the cut-off point to establish end-stage renal failure), ROC curves (Receiver-Operating Characteristics) were constructed determining the area under the prediction curve, its statistical significance, and Cr and CysC levels with the best predicted value for that level of renal function severity.

The data are expressed as mean ± SD. A p value < 0.05 was considered statistically significant. The SPSS software, version 13.0, was used for the statistical analysis.

Table I. Biochemical data

Urea, mg/dL	150 ± 52
Creatinine, mg/dL	4.19 ± 1.19
Cystatin C, mg/dL	3.44 ± 0.73
Glomerular filtration, mL/min/1.73 m^2	16.49 ± 4.65
Serum albumin, g/dL	3.99 ± 0.39

RESULTS

The results of the studied laboratory parameters are shown in Table I. Both Cr and CysC significantly correlated with glomerular filtration (Figures 1 and 2). As it is shown in the graphs, age and gender had an influence on the correlation between Cr and glomerular filtration (figure 1) but did not had an influence on the correlation between CysC and glomerular filtration (figure 2). Diabetes diagnosis did not have an influence on the correlations between Cr or CysC and glomerular filtration.

By multiple linear regression, the best determinants of unfitness of the correlation between Cr and glomerular filtration (estimated by standardized residuals) were age (beta = -0,37; p < 0.0001) and gender (beta = 0.34; p < 0.0001). By contrast, none of the studied independent variables had an effect on the unfitness between CysC and glomerular filtration.

Figure 3 shows the ROC curves predicting a glomerular filtration rate < 15 mL/min/1.73 m^2 for Cr and CysC, respectively. The area under the curve for Cr was 0.675 (p = 0.004), whereas for CysC it was 0.633 (p = 0.030). A creatinine level of 4.12 mg/dL was the one having the best predictive value for diagnosing end-stage renal failure with a 63% sensibility and 66% specificity. The best predictive value for CysC was with a level of 3.41 mg/dL, with 58% sensibility and 61% specificity.

DISCUSSION

The study results confirm the inaccuracy of serum Cr to estimate glomerular filtration in patients with advanced chronic renal failure. As expected, age and gender were the main determinants of the unfitness in the correlation between Cr and glomerular filtration. However, although characteristics such as age, gender, and diabetes did not influence in the correlation between CysC and glomerular filtration, this new marker of renal function did not bring any advantages to the use of Cr in advanced chronic renal failure stages.

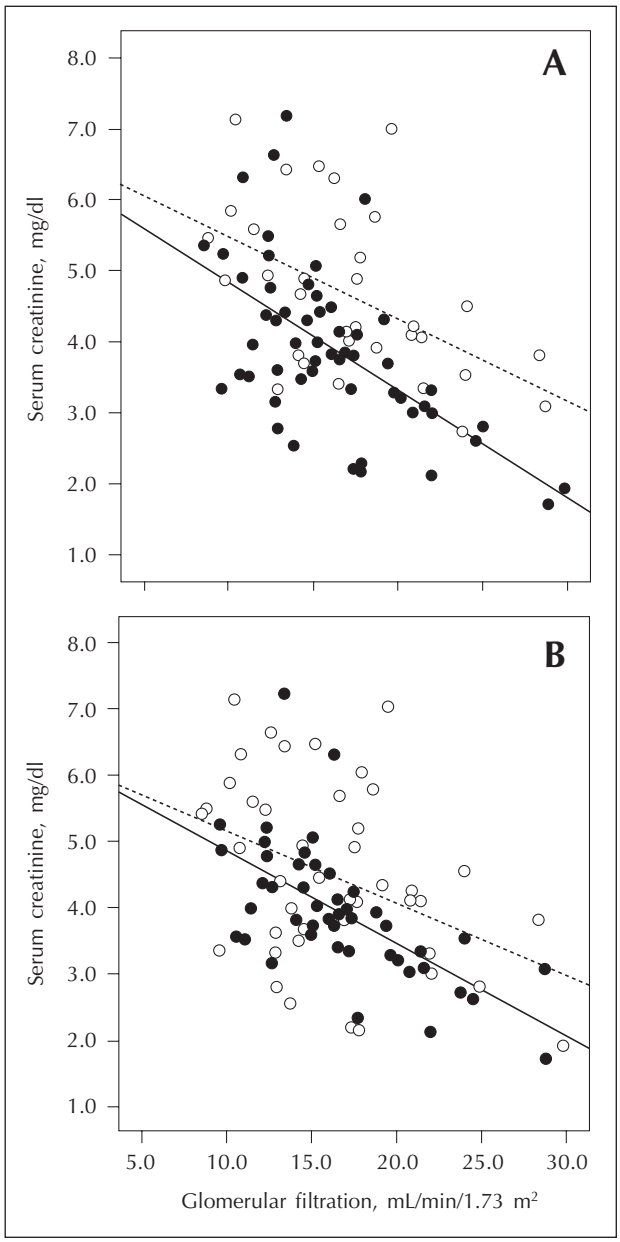

Fig. 1.—Correlation between serum creatinine and glomerular filtration. The characteristics of this correlation by gender are shown in Graph A (females: black continuous line; males: white dotted line). In Graph B patients are divided by age > 65 years (black continuous line) or < 65 years (white dotted line). Pearson's correlation coefficient: 0.489.

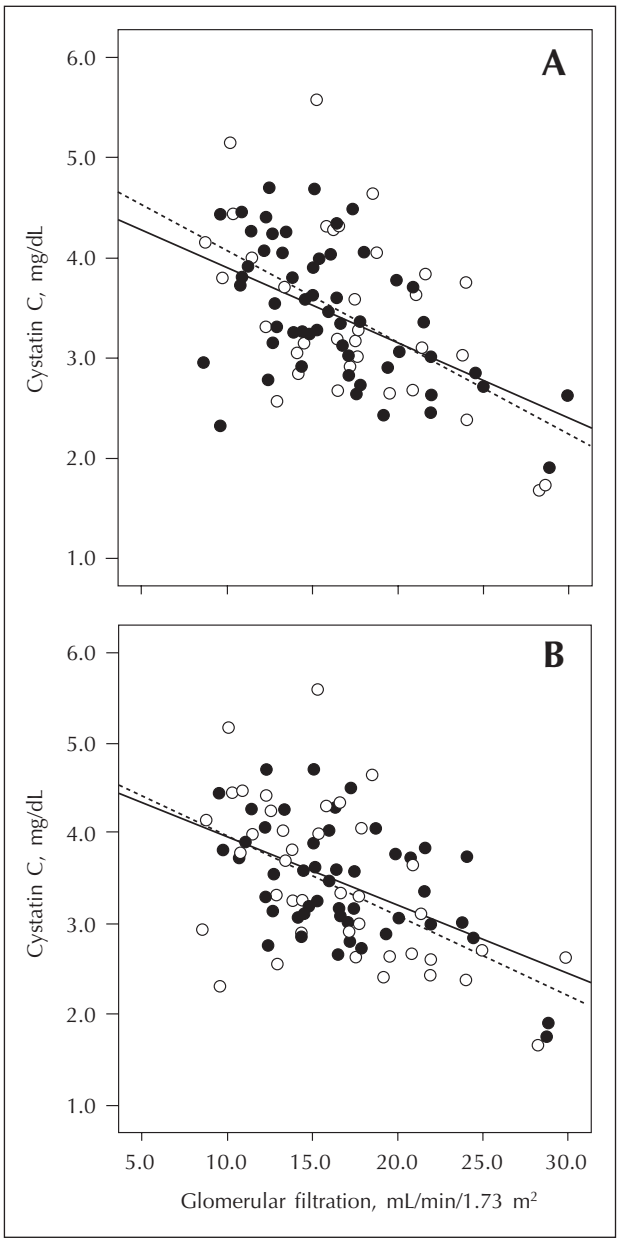

Fig. 2.—Correlation between plasma cystatin C and glomerular filtration. The characteristics of this correlation by gender are shown in Graph A (females: black continuous line; males: white dotted line). In Graph B patients are divided by age > 65 years (black continuous line) or < 65 years (white dotted line). Pearson's correlation coefficient: 0.517.

CysC is a non-glycosilated protein with a molecular weight of 13.5 kDa, constantly produced by nucleated cells, freely filtered by the glomeruli, being reabsorbed by the túbulo-interstitial system.[4-6] The presence of CysC in the urine with an excessive Cr excretion is a sign of túbulo-interstitial damage, and thus, some investigators consider that its urinary measurement may be also used as a diagnostic tool of renal damage.[13]

By contrast with Cr, CysC is not influenced by muscle mass.[4-6] This fact has been put forward as an

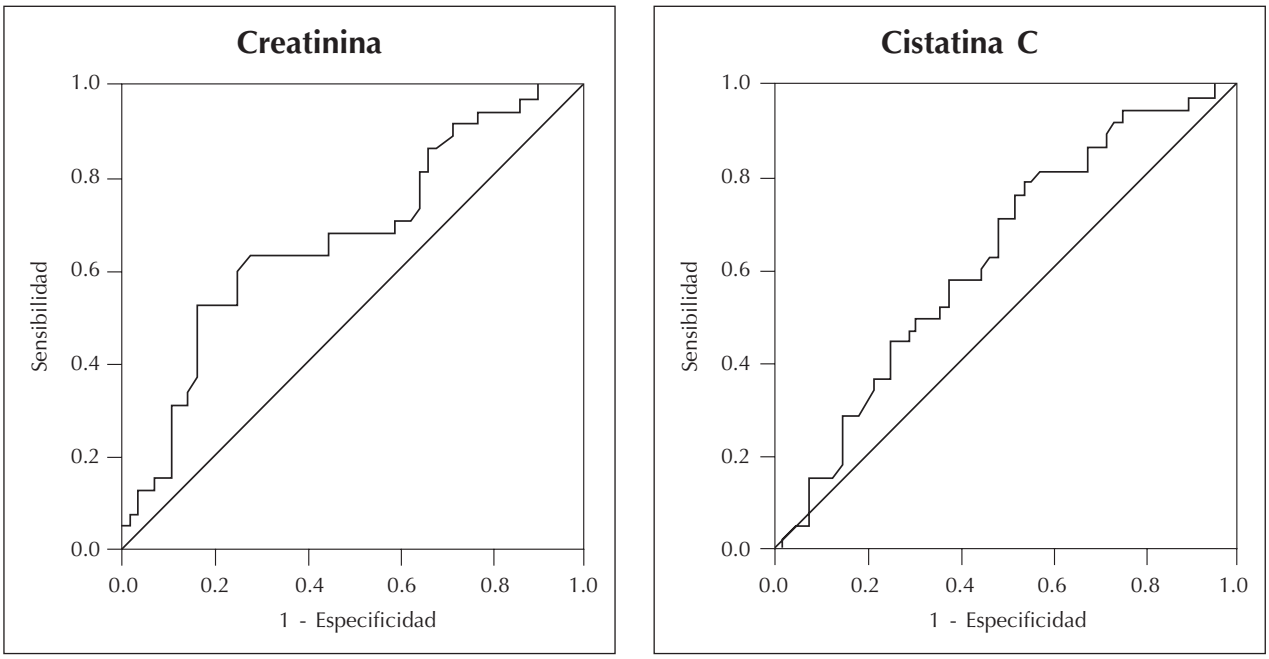

Fig. 3.—ROC curves of the analysis of the predictive value for serum creatinine and cystatin C for diagnosing a glomerular filtration rate < 15-mL/min/1.73 m^2.

advantage in renal function estimation in those patients in whom a defect in endogenous Cr production may underestimate glomerular filtration.[7-10] In this way, the use of CysC has been well accepted by pediatric nephrologists,[14] and some of the best results have been obtained in groups of patients in whom muscle atrophy or hyponutrition are more prevalent, such as diabetics[7,8,10] and cirrhotic patients.[15]

The experience has shown, however, that CysC, although not subjected to muscle mass influences, is subjected to other factors that modify its plasma concentration, independently of glomerular filtration. Hyper- and hypothyroidism, smoking, age, gender, body weight, inflammatory state, and corticosteroid therapy all have shown to alter the relationship between CysC and glomerular filtration.[16-18]

May one to the main advantages of CysC over Cr is its greater accuracy to detect mild to moderate descents in renal function, especially in patients with reduced muscle mass.[8-10] An earlier increase with CysC over Cr has also been observed in critically ill patients with acute renal failure.[19] However, its accuracy to estimate glomerular filtration in advanced chronic renal failure stages is less well known since most of the published studies excluded patients with a glomerular filtration rate below 20 mL/min. The results of the present study show that, although not influenced by age, gender, anthropometrical characteristics, or diabetes, CysC presents high scattering in the correlation with glomerular filtration, similarly to what occurs with Cr. This fact, together with the uncertainty of those factors influencing its variability, make difficult to justify its use as a glomerular filtration estimator in advanced chronic renal failure stages.

REFERENCES

1. James GD, Sealey JE, Alderman M, Ljungman S, Mueller FB, Pecker MS y cols.: A longitudinal study of urinary creatinine and creatinine clearance in normal subjects. Race, sex, and age differences. *Am J Hypertens* 1: 124-31, 1988.
2. Perrone RD, Madias NE, Levey AS: Serum creatinine as an index of renal function: new insights into old concepts. *Clin Chem* 38: 1933-53, 1992.
3. Levey AS, Berg RL, Gassman JJ, Hall PM, Walker WG: Creatinine filtration, secretion and excretion during progressive renal disease. *Kidney Int* 36 (Supl. 27): 573-80, 1989.
4. Abrahamson M, Olafsson I, Palsdottir A, Ulvsback M, Lundwall A, Jensson O y cols.: Structure and expression of the human cystatin C gene. *Biochem J* 268: 287-94, 1990.
5. Simonsen O, Grubb A, Thysell H: The blood serum concentration of cystatin C (gamma-trace) as a measure of the glomerular filtration rate. *Scand J Clin Lab Invest* 45: 97-101, 1985.
6. Laterza OM, Price CP, Scott MG: Cystatin C: an improved estimator of glomerular filtration rate? *Clin Chem* 48: 699-707, 2002.
7. Dharnidharka VR, Kwon C, Stevens G: Serum cystatin C is superior to serum creatinine as a marker of renal function: a meta-analysis. *Am J Kidney Dis* 40: 221-6, 2002.

8. Christensson AG, Grubb AO, Nilsson JA, Norrgren K, Sterner G, Sundkvist G: Serum cystatin C advantageous compared with serum creatinine in the detection of mild but not severe diabetic nephropathy. *J Intern Med* 256: 510-8, 2004.
9. Artunc FH, Fischer IU, Risler T, Erley CM: Improved estimation of GFR by serum cystatin C in patients undergoing cardiac catheterization. *Int J Cardiol* 102: 173-8, 2005.
10. Perkins BA, Nelson RG, Ostrander BEP, Blouch KL, Krolewski AS, Myers BD y cols.: Detection of renal function decline in patients with diabetes and normal or elevated GFR by serial measurements of serum cystatin concentration: results of a 4-year follow-up study. *J Am Soc Nephrol* 16: 1404-12, 2005.
11. Davies SJ, Phillips L, Naish PF, Russell GI: Quantifying comorbidity in peritoneal dialysis patients and its relationship to other predictors of survival. *Nephrol Dial Transplant* 17: 1085-92, 2002.
12. Bröchner-Mortensen J: Current status on assessment and measurement of glomerular filtration rate. *Clin Physiol* 5: 1-17, 1985.
13. Uchida K, Gotoh A: Measurement of cystatin-C and creatinine in urine. *Clin Chim Acta* 323: 121-8, 2002.
14. Filler G, Foster J, Acker A, Lepage N, Akbari A, Ehrich JH: The Cockcroft-Gault formula should not be used in children. *Kidney Int* 67: 2321-4, 2005.
15. Orlando R, Mussap M, Plebani M, Piccoli P, De Martin S, Floreani M y cols.: Diagnostic value of plasma cystatin C as a glomerular filtration marker in decompensated liver cirrhosis. *Clin Chem* 48: 850-8, 2002.
16. Fricker M, Wiesli P, Brandle M, Schwegler B, Schmid C: Impact of thyroid dysfunction on serum cystatin C. *Kidney Int* 63: 1944-1947, 2003.
17. Knight EL, Verhave JC, Spiegelman D, Hillege HL, De Zeeuw D, Curhan GC y cols.: Factors influencing serum cystatin C levels other than renal function and the impact on renal function measurement. *Kidney Int* 65: 1416-1421, 2004.
18. Wasen E, Isoaho R, Mattila K, Vahlberg T, Kivela SL, Irjala K: Serum cystatin C in the aged: relationship with health status. *Am J Kidney Dis* 42: 36-43, 2003.
19. Herget-Rosenthal S, Marggraf G, Husing J, Goring F, Pietruck F, Janssen O y cols.: Early detection of acute renal failure by serum cystatin C. *Kidney Int* 66: 1115-1122, 2004.