Risk of Overestimation of Kidney Function Using GFR-Estimating Equations in Patients with Low Inulin Clearance

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Background: Accurate estimation of the glomerular filtration rate (GFR) is very important in clinical practice. Although renal inulin clearance (Cin) is the gold standard for measuring GFR, the procedure for Cin measurement is complicated. Use of GFR-estimating equations has been increasing recently due to their simplicity. The objectives of the present study are to analyze the correlation between Cin and other GFR-estimating parameters and to investigate their clinical usefulness and limitation. **Methods:** Seventy-two Japanese patients were enrolled in this study. Cin was measured by the continuous infusion method. Serum creatinine (s-Cr), cystatin C, uric acid (UA), and hemoglobin (Hb) were measured. The Japanese formula of estimated GFR (eGFR) was as follows:

$$\text{eGFR (ml/min/1.73m}^2\text{)} = 194 \times \text{s-Cr}^{-1.094} \times \text{Age}^{-0.287} \times 0.739 \text{ (if female).}$$
 The endogenous creatinine clearance test was also performed. **Results:** Levels of Cin were highly correlated with those of endogenous creatinine clearance (Ccr) ($R^2 = 0.7585$) and eGFR ($R^2 = 0.5659$). However, patients with lower Cin showed unexpectedly elevated levels of endogenous Ccr and eGFR. Moreover, the levels of eGFR tended to be unexpectedly increased in patients with low body surface area. **Conclusion:** Although GFR-estimating equations are useful for estimating GFR accurately, they pose a risk of overestimation of kidney function in patients with decreased GFR or a poor physique. J. Clin. Lab. Anal. 26:248–253, 2012. © 2012 Wiley Periodicals, Inc.

Key words: glomerular filtration rate (GFR); inulin clearance; estimated GFR (eGFR); endogenous creatinine clearance

INTRODUCTION

The glomerular filtration rate (GFR) is the best indicator of overall kidney function (1). Accurate estimation of GFR is very important for diagnosis, especially in cases of deciding the dose of medication in clinical practice (2). Although renal inulin clearance (Cin) with continuous intravenous injection is the gold standard for measuring GFR (3), the Cin procedure is complicated. Measurement of endogenous creatinine clearance is performed in virtually every clinical center. In the Modification of Diet in Renal Disease (MDRD) Study, the measured creatinine clearance (Ccr) was not more accurate than the estimated GFR (eGFR) using GFR-estimating equations due to incomplete urine collection or creatinine secretion by the tubules (4). Therefore, use of GFR-estimating equations

has been recommended in clinical practice. The MDRD Study equation is the most commonly used worldwide. Recently, a new Japanese coefficient for the modified 4-variable (isotope dilution mass spectrometry [IDMS])-MDRD and a new Japanese equation for eGFR were proposed (5). Although the equation for eGFR is very convenient and useful in clinical practice because it consists

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of only serum creatinine (s-Cr), age, and sex, precautions are required so that body height or body weight is not reflected in the eGFR. Moreover, it was recently reported that the MDRD and Cockcroft–Gault formula have some limitations for proper GFR estimation (6). Serum cystatin C may have advantages over s-Cr for estimating GFR, and an equation for eGFR using serum cystatin C has been reported (7).

The objectives of the present study are (1) to analyze the correlation between inulin clearance and other GFR-estimating parameters such as s-Cr, Ccr, serum cystatin C, or GFR-estimating equations, and (2) to investigate their clinical usefulness and limitations.

METHODS

Patients

The study protocol was approved by the Ethics Review Committee of Juntendo University Faculty of Medicine, Tokyo, Japan. All individuals gave informed consent. In total, 72 Japanese patients were enrolled in this study. Patient characteristics are shown in Table 1.

Renal Inulin Clearance

Renal clearance of inulin was measured from samples obtained three times during 2 hr under fasting and hy-

drated conditions by the continuous infusion method (8). In brief, the patients drank 500 ml of water 30 min before getting intravenous injection of 1% inulin diluted with saline (INULEAD[®], Fujiyaku Co. Ltd., Saitama, Japan). For maintaining hydration, 60 ml of water was given at 30, 60, and 90 min after the start of inulin injection. The inulin solution was injected at 300 ml/hr for the first 30 min, followed by injection at 100 ml/hr for 90 min. Blood samples were obtained at 0, 45, and 105 min after the start of inulin administration. Urine collection was performed between 30 and 60 min, between 60 and 90 min, and between 90 and 120 min after complete urination at 30 min. Inulin samples were assayed by means of an enzymatic method using a kit (Diacolor Inulin[®], Toyobo Co., Osaka, Japan). Clearance was calculated by 30-min urine collection and serum concentration at the midpoint (60, 90, and 120 min) of each clearance period, as Cin60, Cin90, and Cin120. The mean of three clearances was used as inulin clearance (Cin-m) for the patient.

Other Parameters

Biochemical analysis including s-Cr, serum cystatin C, uric acid (UA), and hemoglobin (Hb) was undertaken at the central laboratory of Juntendo University Hospital. s-Cr was measured with a kit (Aqua-auto Kainos CRE-II test kit, KAINOS Laboratories Inc., Tokyo, Japan) using an enzymatic method. The Japanese formula of eGFR was calculated as follows: $eGFR (ml/min/1.73m^2) = 194 \times s-Cr^{-1.094} \times Age^{-0.287} \times 0.739$ (if female) (5). The clearance of creatinine was calculated three times, in the same way as Cin. The mean of the three clearances was used as the renal creatinine clearance (Ccr-m). The body surface area (BSA) was calculated as follows: $BSA (m^2) = (body\ weight [kg])^{0.425} \times (body\ height [cm])^{0.725} \times 0.007184$ (9).

Urinary analysis including urine Cr (u-Cr) and proteinuria (u-Pro) was also undertaken at the central laboratory of Juntendo University Hospital. Urine samples of complete urination at 30 min were analyzed, and the levels of proteinuria (g/g-Cr) were calculated as u-Pro (mg/dl) / u-Cr (mg/dl).

Statistical Analysis

All statistical analyses were performed using the Windows version of StatView 5.0 software (Abacus Concepts Inc., Berkeley, CA).

TABLE 1. Patient characteristics

Characteristic	
Total number of patients	72
Male, n (%)	44 (61.1)
Age (years)	38.6 ± 15.2
Disease	49
IgA nephropathy	5
Diabetes	4
Hypertension	4
Membranous nephropathy	3
Mild proliferated glomerulonephritis	3
Chronic nephritic syndrome	2
Fabry's disease	1
Alport's syndrome	1
Tubulointerstitial nephritis	1
Nutcracker phenomenon	1
Amyloid kidney	1
Obesity related nephropathy	1
Mesangial proliferative glomerulonephritis	
Height (cm)	164.7 ± 7.9
Weight (kg)	60.1 ± 12.3
Body mass index (kg/m ²)	22.0 ± 3.37
Body surface area (m ²)	1.65 ± 0.19
Systolic blood pressure (mmHg)	112.0 ± 14.3
Diastolic blood pressure (mmHg)	66.4 ± 15.4
Proteinuria (g/g-Cr)	0.57 ± 0.76

Data are expressed as mean ± standard deviation (SD) or number (%).

Fig. 1. Inulin clearance. The mean ($\pm$ standard deviation; SD) Cin-m in all patients was 76.0 ± 34.2 ml/min/1.73m² and Cin values at each sampling point (Cin60, Cin90, and Cin120) were 70.9 ± 35.6 , 79.0 ± 38.0 , and 77.3 ± 41.4 ml/min/1.73m², respectively.

RESULTS

Inulin Clearance

The mean ($\pm$ standard deviation; SD) Cin-m in all patients was 76.0 ± 34.2 ml/min/1.73m² and Cin values at each sampling point (Cin60, Cin90, and Cin120) were 70.9 ± 35.6 , 79.0 ± 38.0 , and 77.3 ± 41.4 ml/min/1.73m², respectively (Fig. 1). These results suggest that our procedure for Cin was accurately performed.

Correlation Between Cin and Other Parameters

Levels of Cin-m were highly correlated with those of Ccr-m ($R^2 = 0.7585$) (Fig. 2A) and low Cin-m patients tended to show higher levels of Ccr-m / Cin-m (Fig. 2B).

The reciprocal of cystatin C or s-Cr ($1 / \text{cystatin C}$ or $1 / \text{s-Cr}$) was correlated with Cin ($R^2 = 0.4285$, 0.4952) (Fig. 3A and B). On the other hand, UA and Hb showed no correlation with Cin-m (Fig. 3C and D).

Although levels of Cin-m were highly correlated with eGFR ($R^2 = 0.5659$) (Fig. 4A), those of eGFR unexpectedly increased in patients with low Cin-m (Fig. 4B). Moreover, the levels of eGFR unexpectedly increased in patients with low BSA (Fig. 4C). However, body mass index (BMI) and proteinuria showed no correlation with eGFR/Cin-m (Fig. 4D). There was no correlation between BSA and proteinuria (Fig. 4E).

DISCUSSION

Renal inulin clearance (Cin) with continuous intravenous injection is the gold standard for measuring GFR (3). Although it was recently reported that the simple method for measurement of renal Cin is sufficiently accurate for use in clinical practice (10), the procedure for Cin measurement is complicated. Therefore, in the clinical situation, GFR-estimating equations or clearance of endogenous creatinine are frequently used. However, it has been reported that Ccr is not so accurate for GFR estimation (4).

Very few reports have compared Cin with other GFR-estimating makers. In this study, we analyzed the correlation between Cin and other GFR-estimating parameters, and investigated their clinical usefulness and limitations. In the present study, there was a risk of overestimation of kidney function in patients who had decreased GFR, although Ccr is a helpful tool to estimate GFR. Although Cin and Ccr showed a high correlation, Ccr was about 10–20% higher than Cin. Moreover, low Cin patients showed a tendency for unexpectedly high Ccr. Creatinine is not only filtered from glomeruli but also secreted from

Fig. 2. Correlation between Cin and Ccr. (A) Values of Cin-m were highly correlated with those of Ccr-m ($R^2 = 0.7585$). Dotted line indicates line of identity. (B) Low Cin-m patients tended to show higher values of Ccr-m/Cin-m.

Fig. 3. Correlation between Cin and cystatin C, s-Cr, UA, or Hb. (A) The reciprocal of cystatin C (1 / cystatin C) was correlated with Cin-m ($R^2 = 0.4952$). (B) The reciprocal of s-Cr (1 / Cr) was correlated with Cin-m ($R^2 = 0.4285$). (C) UA showed no correlation with Cin-m. (D) Hb showed no correlation with Cin-m.

tubules, and secretion from tubules increases with the deterioration of GFR (11, 12). These mechanisms may contribute to these discrepancies.

The relationship between Cin and eGFR showed similar tendencies. In this study, Cin and eGFR showed a high correlation, and low Cin patients tended to have unexpectedly high eGFR. These results suggest that there is a risk of overestimation of kidney function in patients who had decreased GFR, similar to Ccr, although the Japanese formula of eGFR is a useful tool to estimate GFR. Since eGFR is calculated using only s-Cr, age, and sex (5), it is very convenient and useful in clinical practice. However, such convenience and usefulness may result in overestimation of GFR. It is a logical result for eGFR to be overestimated in low Cin patients because of the comparatively high contribution of s-Cr to eGFR. Moreover, it is necessary to consider the physique of the patient because it is not reflected in the eGFR formula. Patients with lower BSA tended to show unexpectedly high eGFR. However, differences among individuals were also observed. These results suggest that there is a risk of overestimation of kidney function in patients who have a poor physique, not depending on obesity or emaciation.

Although there are some reports that increasing of proteinuria causes unexpectedly high Ccr (11), proteinuria

showed no correlation with eGFR/Cin-m in this study. Moreover, there was no correlation between BSA and proteinuria, although patients with lower BSA tended to show unexpectedly high eGFR.

In daily clinical practice, accurate estimation of GFR is very important, especially when deciding doses of medications for patients. Moreover, the stage of chronic kidney disease (CKD) is decided using eGFR. Therefore, it is important to educate CKD patients concerning their diseases and dietary restrictions (13), and a simple method of GFR-estimation is needed in clinical practice. Measurement of Cin is the best way to estimate GFR, but a disadvantage is that the procedure is complicated. Although measurement of the clearance of endogenous creatinine was performed in the past, the number of patients for whom eGFR is used due to its simplicity is increasing recently. From this study, although it was proven that endogenous creatinine clearance or the GFR-estimating equation is a useful tool to estimate GFR in clinical practice, the risk of overestimation of kidney function in patients who had decreased GFR was suggested. Moreover, it was also suggested that there was a tendency for overestimation of kidney function using the GFR-estimating equation in patients who have a poor physique.

Fig. 4. Correlation between Cin and eGFR. (A) Cin-m was highly correlated with eGFR ($R^2 = 0.5659$). Dotted line indicates line of identity. (B) Low Cin-m patients tended to show higher values of eGFR/Cin-m. (C) eGFR tended to be unexpectedly high in patients with low body surface area (BSA). (D) Body mass index (BMI) showed no correlation with eGFR/Cin-m. (E) Proteinuria also showed no correlation with eGFR / Cin-m. (F) There was no correlation between BSA and proteinuria.

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