Glomerular filtration rate measured by $^{99m}$Tc-DTPA renal dynamic imaging is significantly lower than that estimated by the CKD-EPI equation in horseshoe kidney patients

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SUMMARY AT A GLANCE
GFR determined by isotope renal dynamic imaging was compared to eGFR (CKD-EPI) in 19 subjects with horseshoe kidneys. Isotope renograms do not accurately estimate GFR in individuals with horseshoe kidneys.

Horseshoe kidney (HSK), a congenital anomaly of renal fusion, is one of the most common renal anomalies.1 This abnormality begins to develop at weeks 4 to 6 of gestation, as the metanephric blastema abnormally migrates across the midline and aggregates.2 Most HSK patients have abnormal kidney rotation and fusion of the kidneys at the lower poles to form an isthmus that usually lies anterior to the great vessels at the level of the third to fifth lumbar vertebra creating a U-shape.3–5 HSK patients always present with genitourinary and extra-genitourinary congenital abnormalities,2 such as vascular abnormalities. They are prone to a variety of complications, which are the main reasons for patients to visit a doctor, such as stone disease, ureteropelvic junction (UPJ) obstruction, trauma, infection, and a variety of benign and malignant tumors.3–7 Renal imaging, including abdominal ultrasonography (US), intravenous pyelography (IVP),

ABSTRACT:

Aim: Gate's glomerular filtration rate (gGFR) measured by $^{99m}$Tc-DTPA renal dynamic imaging and estimated GFR (eGFR) estimated by the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation are two indexes used to evaluate renal function. However, little is known about whether gGFR can be used to accurately assess renal function in horseshoe kidney (HSK) patients with renal fusion anomalies.

Methods: Nineteen HSK patients (HSK group) diagnosed by renal imaging and 38 CKD patients with “normal kidney shape” (non-HSK group) matched to the HSK patients in terms of gender, age and biochemical indicators at Chinese PLA General Hospital were enrolled in this study. Gender, age, serum total protein (TP), albumin (ALB), blood urea nitrogen (BUN), serum creatinine (Scr), gGFR and eGFR were recorded and analyzed using $\chi^2$ test, t-test, and Wilcoxon test which was presented as median(IQR).

Results: (1) There were no significant differences in gender, age, TP, ALB, BUN, Scr, or eGFR between these two groups. (2) In HSK patients, the renogram showed abnormal renal axis with the lower poles orientated medially. The timed uptake curve showed that the isotope excretion in the HSK group was slower than that in the non-HSK group. (3) For all HSK patients, gGFR was significantly lower than eGFR (range 12.52 mL/min per 1.73 m$^2$ to ~93.18 mL/min per 1.73 m$^2$). There was no significant difference in eGFR between the HSK [96.42 (36.02) mL/min per 1.73 m$^2$] and non-HSK groups [94.46 (33.00) mL/min per 1.73 m$^2$]. The gGFR of the HSK group [41.18 (16.60) mL/min per 1.73 m$^2$] was much lower than that of the non-HSK group [86.42 (26.40) mL/min per 1.73 m$^2$, $P < 0.001$] and the eGFR of the HSK group ($P < 0.001$). The gGFR and eGFR of the non-HSK group were not significantly different.

Conclusion: gGFR measured by $^{99m}$Tc-DTPA renal dynamic imaging is significantly lower than eGFR estimated by the CKD-EPI equation, which indicates that isotope renogram cannot accurately evaluate the GFR of HSK patients.
For HSK patients and patients with kidney diseases, it is important to accurately evaluate renal function to determine a suitable treatment plan. The best global index of renal function is glomerular filtration rate (GFR).\textsuperscript{9} Accurate assessment of GFR is essential for interpreting symptoms and signs and for drug dosing, detecting and managing kidney disease and assessing prognosis.\textsuperscript{10} Inulin clearance is the gold standard for drug dosing, detecting and managing kidney disease and of GFR is essential for interpreting symptoms and signs and important to accurately evaluate renal function to determine patients in clinical practice.

Determine a suitable method for evaluating the GFR of HSK patients in clinical practice.\textsuperscript{8,9} HSK diagnosed by renal imaging, including US, IVP, CT or MRI, play an important role in diagnosing these anomalies and their complications; the most valuable imaging modalities are CT and MRI.\textsuperscript{8,9}

SUBJECTS AND METHODS

Patients

The subjects enrolled in this retrospective clinical study were outpatients/inpatients at Chinese PLA General Hospital from May 2012 to April 2015. Inclusion criteria: (i) HSK patients: HSK diagnosed by renal imaging, including US, IVP, CT or MRI; HSK patients who undergo renal dynamic imaging and with complete gender, age and biochemical indexes (TP, ALB, BUN, Scr). (ii) Non-HSK patients: chronic kidney disease (CKD) patients with normal renal shape; non-HSK patients who undergo renal dynamic imaging and with complete gender, age and biochemical indexes which had no difference with HSK patients. The exclusion criteria were as follows: patients who did not undergo renal dynamic imaging, and/or patients with incomplete or without biochemical indexes, and/or patients whose biochemical indexes in non-HSK group had great difference from HSK patients ($P < 0.05$).

GFR Measurement

1. $^{99m}$Tc-DTPA renal dynamic imaging method (Gate’s method)

Gate developed a simple method to determine GFR in 1982 named the Gate’s method, and it was modified in 1983.\textsuperscript{12,16,17} It is also called the renal dynamic imaging method because GFR can be calculated simultaneously using a computer system when the patient is undergoing renal dynamic imaging. The most widely used radionuclide is $^{99m}$Tc-DTPA because it was filtered by the glomeruli without tubular absorption.\textsuperscript{18,19} Thus, GFR can be accurately measured using $^{99m}$Tc-DTPA.\textsuperscript{20} The patients were hydrated with 300–500 mL water 20 min prior to the examination.\textsuperscript{12,14} The patients laid down on the bed, and then a 5 mCi (185 MBq) count of the syringe containing $^{99m}$Tc-DTPA was performed before injection. After intravenous injection of a 5 mCi (185 MBq) $^{99m}$Tc-DTPA bolus, dynamic imaging was performed in the posterior position, usually at a rate of acquisition of 1–2 frames/s for 1 min, and then one frame/min for 14 min. Regions of the kidneys were placed in the centre view of the gamma camera. The syringe radioactive counting rate was measured using the central probe before and after injection. The total injected dose was determined by subtracting the post-count from the pre-count. The region of interest (ROI) was manually drawn on the frame of the kidney, and a semi-lunar background was placed around the lower, outer renal margin. After the patient’s weight and height were entered into an online computer, the GFR was automatically calculated by commercially available software according to the Gate’s algorithm. Single photon emission computerized tomography (SPECT) equipped with a low-energy general-purpose parallel-hole collimator was used to complete the examination. The timed uptake curves of the two kidneys provided notable information, such as renal blood flow, renal function and urinary tract patency. The renogram curve was divided into three sections: section a, steep slope increase, mainly reflecting renal blood perfusion conditions; section b, slower rate of slope increase, reflects renal function and renal blood flow; and section c, decreased slope after the peak, represents the decrease in the urine flow rate and urinary tract patency. It usually takes 20 min for the isotope to flow into the bladder under normal circumstances. Delayed excretion is often due to vascular disease, impaired renal function or urinary tract obstruction. GFR measured by the modified Gate’s method was calculated using the following equation:\textsuperscript{15}

\[
\text{Total renal uptake percent} = \frac{\left( R - RB \right) e^{-\mu R}}{\left( L - LB \right) e^{-\mu L}} + \frac{\left( L - LB \right) e^{-\mu L}}{\text{Pre} - \text{Post}}
\]

\[
\text{Global GFR} = \text{Total renal uptake percent} \times 100 \\
\times 9.81270 - 6.82319
\]

Pre: pre-count, Post: post-count, R: right kidney counts, RB: right kidney background counts, L: left kidney counts, LB: left kidney background counts, $\chi_R$: right kidney depth, $\chi_L$: left

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kidney depth, μ: attenuation coefficient of $^{99m}$Tc in soft tissue (0.153/cm), e: constant.

2. CKD-EPI equation method

Gender, age, race, and serum creatinine (Scr) are the main variables in the CKD-EPI formula, so any changes in these variables will affect the GFR obtained using this formula. However, many studies have shown that the CKD-EPI formula is more accurate and more widely applied than other methods of GFR determination in Chinese CKD patients and that it can be applied generally in clinical practice. The main blood biochemical indexes in this study were measured on the same day as gGFR. The normal range of Scr measured by the enzymatic method was 30–110 μmol/L (1 mg/dL = 88.4 μmol/L).

CKD-EPI equation:21

\[
\text{Female: } \text{Scr} \leq 0.7 \text{mg/dl} \times 144 \times (\text{Scr}/0.7)^{-0.329} \\
\times 0.993^{\text{age}} \times 1.159; \text{if black} \\
\text{Scr} > 0.7 \text{mg/dl} \times 144 \times (\text{Scr}/0.7)^{-1.209} \\
\times 0.993^{\text{age}} \times 1.159; \text{if black} \\
\text{Male: } \text{Scr} \leq 0.9 \text{mg/dl} \times 141 \times (\text{Scr}/0.9)^{-0.411} \\
\times 0.993^{\text{age}} \times 1.159; \text{if black} \\
\text{Scr} > 0.9 \text{mg/dl} \times 141 \times (\text{Scr}/0.9)^{-1.209} \\
\times 0.993^{\text{age}} \times 1.159; \text{if black}
\]

**Ethics statement**

This study was performed in compliance with the Helsinki Declaration with the approval of the Ethics Committee of Chinese PLA General Hospital (No. 2012-001).

**RESULTS**

General information about HSK patients and non-HSK patients

A total of 77 patients diagnosed with HSK by renal imaging were screened from May 2012 to April 2015 at Chinese PLA General Hospital. Fifty-eight HSK patients without renal dynamic imaging and with incomplete or without blood biochemical indexes were excluded. Finally, 19 HSK patients were enrolled, including 10 males and 9 females (mean age 31.95 ± 11.37 years, range 15 to 54 years). Thirty-eight non-HSK patients were enrolled as controls, including 20 males and 18 females (mean age 35.21 ± 8.57 years, range 15 to 50 years). The reasons of the HSK patients to visit a doctor were physical examination (six cases, 31.6%), waist and abdomen pain (three cases, 15.8%), abnormal urinalysis (four cases, 21.1%), elevated serum creatinine (two cases, 10.5%), hydronephrosis

| Patients | Gender | Age (years) | TP (g/L) | ALB (g/L) | BUN (mmol/L) | Scr (μmol/L) | eGFR (mL/min per 1.73 m²) | eGFR (mL/min per 1.73 m²) | d (mL/min per 1.73 m²) |
|----------|--------|-------------|---------|-----------|-------------|-------------|--------------------------|--------------------------|---------------------|
| 1        | Male   | 42          | 75.60   | 49.30     | 6.33        | 85.30       | 37.60                     | 96.42                    | -58.82              |
| 2        | Male   | 25          | 74.50   | 52.20     | 4.87        | 95.90       | 37.20                     | 94.89                    | -57.69              |
| 3        | Male   | 22          | 78.90   | 47.60     | 6.63        | 77.10       | 39.00                     | 122.51                   | -83.51              |
| 4        | Male   | 24          | 82.20   | 53.50     | 5.00        | 84.90       | 28.70                     | 110.18                   | -81.48              |
| 5        | Female | 25          | 77.40   | 45.10     | 5.49        | 81.30       | 28.38                     | 86.82                    | -58.44              |
| 6        | Female | 29          | 74.30   | 47.90     | 3.92        | 62.00       | 63.50                     | 117.46                   | -53.96              |
| 7        | Male   | 44          | 76.00   | 45.20     | 6.46        | 105.00      | 37.00                     | 73.85                    | -36.85              |
| 8        | Male   | 17          | 65.00   | 47.30     | 5.32        | 69.40       | 40.70                     | 133.88                   | -93.18              |
| 9        | Female | 24          | 73.50   | 47.00     | 5.56        | 72.00       | 46.16                     | 101.98                   | -55.82              |
| 10       | Male   | 37          | 65.80   | 43.20     | 6.60        | 96.00       | 46.81                     | 86.25                    | -39.44              |
| 11       | Female | 15          | 69.80   | 44.20     | 2.70        | 66.90       | 49.80                     | 117.33                   | -67.53              |
| 12       | Female | 31          | 60.50   | 35.80     | 11.04       | 191.00      | 17.14                     | 29.66                    | -12.52              |
| 13       | Male   | 26          | 59.20   | 40.80     | 5.18        | 107.90      | 41.18                     | 81.31                    | -40.13              |
| 14       | Male   | 41          | 64.80   | 39.90     | 6.06        | 143.50      | 19.41                     | 51.94                    | -32.53              |
| 15       | Female | 51          | 72.50   | 44.00     | 4.47        | 65.50       | 53.60                     | 94.10                    | -40.50              |
| 16       | Female | 46          | 77.80   | 51.00     | 5.18        | 62.60       | 46.40                     | 103.47                   | -56.07              |
| 17       | Male   | 27          | 71.30   | 45.10     | 7.10        | 81.20       | 53.69                     | 113.58                   | -59.89              |
| 18       | Male   | 54          | 69.00   | 36.00     | 7.29        | 84.20       | 54.00                     | 68.12                    | -14.12              |
| 19       | Female | 27          | 64.60   | 43.10     | 4.28        | 62.10       | 96.10                     | 119.12                   | -23.02              |

ALB, albumin; BUN, blood urea nitrogen; d, gGFR–eGFR; eGFR, estimated glomerular filtration rate; gGFR, Gate's glomerular filtration rate; Scr, serum creatinine; TP, total protein.
(one case, 5.3%), renal cyst (two cases, 10.5%) and kidney neoplasms (one case, 5.3%). The reasons the non-HSK patients visited a doctor were chronic renal insufficiency (thirteen cases, 34.2%), nephrosis or hydronephrosis (seven cases, 18.4%), kidney stones (two cases, 5.3%), kidney neoplasms (six cases, 15.8%) and other reasons (ten cases, 26.3%).
Main biochemical parameters and GFR of HSK patients

Table 1 shows the gender, age, main biochemical parameters and GFR of the 19 HSK patients. As listed in Table 1, the gGFR values of all HSK patients were significantly lower than their eGFR values, with a minimum difference of ~12.52 mL/min per 1.73 m² (No.12) and a maximum difference of ~93.18 mL/min per 1.73 m² (No.8).

Comparison of isotope renogram and the timed uptake curves between HSK and non-HSK patients

The isotope renogram images were different between the HSK and non-HSK patients because of the differences in their kidney anatomies. The renograms of the HSK patients showed abnormal renal axis with the lower poles orientated medially (Fig. 1a), while the renograms of the non-HSK patients showed that the lower poles were divided (Fig. 1c). The timed uptake curves were obviously different between the HSK and non-HSK groups. In the HSK patients, isotope excretion was slower, and the curve stayed at a high level (Fig. 1c). In the non-HSK patients, isotope excretion without urinary tract obstruction was smooth, and the curve was tilted downward (Fig. 1g).

Comparison of gender, age, blood biochemical indexes and GFR between HSK and non-HSK patients

Gender, age, TP, ALB, BUN, and Scr of the patients were not significantly different between the HSK and non-HSK groups. eGFR was not significantly different between the HSK and non-HSK groups [96.42(36.02) mL/min per 1.73 m² vs 94.46 (33.00) mL/min per 1.73 m², \( P=0.35 \)]. gGFR was much lower in the HSK group than in the non-HSK group [41.18(16.60) mL/min per 1.73 m² vs 86.42 (26.40) mL/min per 1.73 m², \( P<0.001 \)], and in the HSK group, gGFR was significantly lower than eGFR [41.18(16.60) mL/min per 1.73 m² vs 96.42(36.02) mL/min per 1.73 m², \( P<0.001 \) ], the correlation coefficient was 0.537 (\( P=0.018 \)) (Fig. 2a). In the non-HSK group, gGFR and eGFR were not significantly different [86.42(26.40) mL/min per 1.73 m² vs 94.46(33.00) mL/min per 1.73 m², \( P=0.12 \)] (Table 2), the correlation coefficient was 0.827 (\( P<0.001 \)) (Fig. 2b).

DISCUSSION

Our study verified firstly that for all HSK patients, gGFR was significantly lower than eGFR (range ~12.52 mL/min per 1.73 m² to ~93.18 mL/min per 1.73 m²). The gGFR and eGFR of the non-HSK group were not significantly different. There was no significant difference in eGFR between the HSK and non-HSK groups, which indicates that isotope renogram cannot accurately evaluate the GFR of HSK patients.

An accurate, convenient, and reproducible GFR estimation method is important for clinical practice.\(^{22}\) It helps clinicians to make the best treatment decisions. The \(^{99m}\)Tc-DTPA renal dynamic imaging method is simple, fast and less expensive for the determination of GFR than other methods and has been used as a reference method for GFR estimation.\(^{21}\) Multiple nuclear medicine techniques for measuring renal glomerular filtration using \(^{99m}\)Tc-DTPA currently exist,\(^ {23}\) because it is the least expensive and the most practical radiopharmaceutical agent.\(^ {24}\) Most importantly, \(^{99m}\)Tc-DTPA is filtered by the glomeruli without reabsorption by the renal tubules. Thus, it is more accurate to measure GFR with \(^{99m}\)Tc-DTPA than with other radiopharmaceutical agents. The valuable and accurate information provided by the renogram can help clinicians to diagnose urinary system obstruction and monitor transplanted kidneys and the effects of treatment and operation.\(^{25}\) However, many factors affect the accuracy of gGFR measurement: for example, delay or reduction of radionuclide \(^{99m}\)Tc-DTPA clearance due to the binding of \(^{99m}\)Tc-DTPA to plasma proteins;\(^ {15}\) artificial bias in the drawing of the regions of interest; different doses of radionuclides injected; attenuation coefficient of radionuclide in the kidney; depth of the kidney, etc. Whether \(^{99m}\)Tc-DTPA renal dynamic imaging can accurately assess renal function in HSK patients has not been reported because of the abnormal anatomic locations and morphological structures of the kidneys in HSK patients.

The CKD-EPI equation, which was developed based on a large dataset pooled from research and clinical populations, including renal disease patients and healthy individuals, has been proven to be suitable for the determination of GFR in many countries, such as Japan, America, Australia, Spain, and China.\(^ {12,26-28}\) Multiple studies have shown that the CKD-EPI formula can accurately evaluate GFR and effectively reduce the false positive rate of CKD diagnosis, especially at GFR > 60 mL/min per 1.73 m².\(^ {29}\) Moreover, the National Kidney Foundation (NKF) recommends the CKD-EPI formula rather than the MDRD formula.\(^ {30}\) Therefore, the CKD-EPI equation was chosen as a reference to evaluate the accuracy of GFR estimation using \(^{99m}\)Tc-DTPA renal dynamic imaging in HSK patients in this study.

Based on the above results, \(^{99m}\)Tc-DTPA renal dynamic imaging significantly underestimated renal function in HSK patients, which could result in a premature diagnosis of renal failure. The possible causes of this underestimation are as follows. Firstly, the kidneys are connected by an isthmus anterior to the lumbar vertebra, which is either a band of fibrous tissue or a thick rim of functional renal tissue. If the isthmus is composed of functional renal tissue, the gGFR would be lower than usual because the functional isthmus shielded by the lumbar vertebra could be overlooked by technicians. Secondly, the isthmus usually lies anterior to the lumbar vertebra and great vessels, so the distances from the kidneys to the back waist are much longer than normal, which results in signal attenuation. The weaker isotopic signal obtained by the receiver will lead to a lower gGFR. Furthermore, whether the abnormal internal structure of the HSK itself leads to abnormal isotope absorption or excretion and subsequent underestimation of
gGFR is still unclear. Therefore, premature diagnosis of renal failure could occur among HSK patients when the isotope renogram is used to measure GFR. This may also increase the incidence of renal failure due to over-treatment and can increase the economical and mental burden of HSK patients. The renogram curves of the HSK patients revealed delayed isotope excretion, which may be due to other urinary system malformations or renal dysfunction. Therefore, for HSK patients, both clinical manifestations and blood biochemical indicators should be taken into consideration to comprehensively assess renal function.

This study had some limitations. Firstly, we did not choose a gold standard method for the measurement of GFR as a reference, such as the inulin clearance rate and double plasma methods. Although these two methods are highly accurate, they are less accepted because they require more time and labor than other methods and have poor patient compliance. Secondly, we did not determine the exact causes of the underestimation of renal function by $^{99m}$Tc-DTPA renal dynamic imaging in HSK patients. Therefore, we could not propose a modification to the Gate’s formula, but this is still being studied by our group.

In conclusion, HSK has become one of the most common urinary malformations, and accurate evaluation of renal function in these patients is essential. $^{99m}$Tc-DTPA renal dynamic imaging is not suitable for HSK patients because it significantly underestimates GFR in those patients, resulting in an increased rate of diagnosis of renal failure. Determining a suitable method to evaluate GFR in HSK patients will be helpful to accurately determine renal function and split renal function to aid in diagnosis and treatment. The eGFR estimated by the CKD-EPI equation is suitable for HSK and non-HSK patients.

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