Age-dependent reference intervals for estimated and measured glomerular filtration rate

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Abstract

Background: Defining mean and reference intervals for glomerular filtration rate (GFR) has been the subject of only a limited number of studies and review articles, with contradicting statements about the mean. Normal measured GFR (mGFR) values of \( \sim 120-130 \) mL/min/1.73 m\(^2\) have long been the referenced values for young adults but seem to be too high according to recent studies. Reference intervals are difficult to define because of the age decline of GFR, which is also observed in healthy subjects. Little data are available for subjects >70 years of age.

Methods: Based on the reference intervals for serum creatinine (SCr) and the recently published full-age spectrum (FAS) equation, we define simple age-dependent equations for the reference limits of GFR. The mGFR of 633 living potential kidney donors was used to validate the new formulae that define the reference interval.

Results: The reference limits for estimated GFR (eGFR), calculated by entering the reference limits for SCr into the FAS equation closely correspond with published reference limits for mGFR. Of the mGFRs of potential living kidney donors, 97.2% lie between the newly defined reference limits for GFR.

Conclusion: SCr reference limits may serve to define age-dependent reference limits for eGFR and mGFR.

Key words: age-dependent reference intervals; estimated and measured glomerular filtration rate
Introduction

We recently proposed a new full-age spectrum (FAS) estimating glomerular filtration rate (eGFR) equation [1, 2] for which the serum creatinine (SCr)-based version is presented as:

eGFR = 107.3/[SCr/Q] for ≤2 – ≤40 years of age,
eGFR = 107.3/[SCr/Q] × 0.988(Age-40) for age > 40 years.

where Q is the mean or median SCr concentration of an age-/sex-specific healthy population. For children, the value of Q linearly increases with age for both males and females. For adolescents, the Q-value increases with age at the same rate as for children in the case of females, but with a greater slope for males. For adult Caucasians, the value of Q is constant and equals 62 μmol/L (0.70 mg/dL) for females and 80 μmol/L (0.90 mg/dL) for males. We have also shown that the same form of the FAS equation can be applied for serum cystatin C (ScysC). The rescaling factor in ScysC/Q has been determined as Q’ = 0.82 mg/L for ages up to 70 years and Q’ = 0.95 mg/L beyond that age. The distributions of both rescaled biomarkers have specific properties that hold for all ages and sexes:

- The distribution is normal or Gaussian, that is, it has a symmetrical bell-shaped form with a mean of 1 and a standard deviation (SD) of ~0.1686.
- The 2.5th percentile equals 0.67 and the 97.5th percentile equals 1.33, and consequently the reference interval for the rescaled biomarkers is [0.67 – 1.33], independent of age and sex.

Since the mean value of the rescaled biomarkers for the healthy population equals 1, then, from the FAS equation, it is obvious that the corresponding mean eGFR equals 107.3 mL/min/1.73 m² for children, adolescents and young adults between 2 and 40 years of age. The referenced measured GFR (mGFR) values in white healthy young adults are ~120–130 mL/min/1.73 m², but these reference values date back to the 1950–70s [3, 4]. Also, the currently recommended Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) eGFR equation [5] predicts a value of 124 mL/min/1.73 m² for an 18-year-old male with SCr = 0.90 mg/dL. However, a recent meta-analysis analysing publications on GFR measurements in healthy living potential kidney donors challenged these prior thresholds and confirmed 107 mL/min/1.73 m² as an acceptable normal reference value [6]. For the older average healthy adult (with SCr/Q = 1), a simple age-dependent decline of eGFR = 107.3 × 0.988(Age-40) is obtained from the FAS equation. Analogously, the lower and upper limit for SCr/Q, of 0.67 and 1.33, respectively, can be entered in the above equation and yields age-dependent equations for FAS-eGFR that correspond to the lower and upper limit for SCr/Q.

In this article we present original un published data from Lyon (France), Liège (Belgium) and Leuven (Belgium) for mGFR in healthy potential living kidney donors to confirm our results from the meta-analysis [6]. The goal of this work is to validate the correspondence between the FAS-eGFR reference limits derived from the upper and lower SCr reference limits, with reference limits for mGFR as obtained from the literature. We also want to verify if the mean GFR for healthy young adults is close to 107.3 mL/min/1.73 m² and not 120–130 mL/min/1.73 m² as is mostly reported.

Materials and methods

We collected mGFRs of 633 potential living kidney donors from Lyon (inulin, n = 514; iohexol, n = 36), Liège, Belgium (51Cr-EDTA, n = 41) and Leuven, Belgium (51Cr-EDTA, n = 42). All subjects have been consecutively included in this study, although the entry periods in Leuven, Lyon and Liège were not the same. Any disproportion in gender or age is therefore a consequence of pure chance. The inulin measurement in Lyon was based on the continuous infusion method and the collection of timed urine (U) and plasma (P) samples, from which the mGFR is calculated as GFR = [U × V/P], with [U] and [P] the concentration of inulin (in mg/mL) and V the urine production per time interval (in mL/min) [7, 8]. Iohexol measurements in Lyon and Liège were based on the plasma clearance protocol using three and four time points, respectively, to obtain the area under the concentration–time curve (AUC) from the mono-exponential slow compartment decay. The GFR calculated from this AUC was then corrected with the Bröchner–Mortensen formula [9]. 51Cr-EDTA measurements in Leuven were also based on the plasma clearance protocol, but using eight time points to obtain the concentration–time curve and the AUC was obtained from bi-exponential decay (fast and slow compartment). All presented GFR methods have sufficient accuracy compared with the inulin gold standard reference method [10, 11]. Estimating GFR with the FAS equation is based on standardized assays for Scr and ScysC [1, 2]. A general informed consent was obtained from all patients, including the information that their data could be used anonymously in retrospective data analyses.

We also performed an extensive literature study to obtain reference data for mGFR to support our findings. This literature search resulted in 12 publications that have previously been presented in a meta-analysis study [6]. Mean mGFR and standard deviations (SDs) from this meta-analysis study were used to calculate lower (LRL) and upper (URL) reference limits as mean mGFR ± 2 × SD. Statistical analysis [descriptive statistics, t-tests, analysis of variance (ANOVA)] was performed with SAS 9.4 (SAS Institute, Cary, NC, USA).

Results

Table 1 and Figure 1 demonstrate the age decline for both male and female potential kidney donors in our dataset (n = 633). No difference was observed between males and females in the defined age groups younger than 50 years of age. However, we noticed a difference between males and females older than 50 years, with a faster decline in females than in males, an observation that we also found in our meta-analysis study [6].

When combining the mGFR data of males and females in age decades, then, for the decades 20–30 years (mean mGFR = 107.3, [95% confidence interval (CI) 103.5–111.0] (n = 44)), 30–40 years [mean mGFR = 107.4, (95% CI 104.7–110.1) (n = 127)] and 40–50 years [mean mGFR = 103.7 (95% CI 101.4–106.0) (n = 202)], no difference between the mean mGFRs of these age groups was observed (ANOVA, P = 0.0814) and no difference in the age decades 20–30 years and 30–40 years was observed with a mean value of 107.3 mL/min/1.73 m², as predicted from the FAS equation. On the other hand, all age groups showed a significant difference from 120 to 130 mL/min/1.73 m² (P < 0.0001).

Using the LRL (0.67) and URL (1.33) for SCr/Q, we can calculate corresponding eGFR limits as:

URL:

FAS/SCr/Q = 0.67 = 107.3/0.67 = 160 mL/min/1.73 m² for ≤2 – ≤40 years of age.

FAS/SCr/Q = 0.67 = 160 × 0.988(Age-40) mL/min/1.73 m² for age > 40 years.
If the correspondence between reference intervals for SCr/Q and GFR is indeed true, then we would expect 95% of healthy subjects to have an mGFR between these limits. We therefore plotted the FAS-eGFR reference limits together with the mGFRs of the 633 potential kidney donors in Figure 2, resulting in 97.2% of mGFRs of the kidney donors lying between the limits defined above.

From the meta-analysis table, published previously [6], we calculated 2.5th and 97.5th percentiles for mGFR as mean ± 2 × SD for 12 studies (males and females separately) and 6 different age groups. The means of the LRLs and URLs with their SDs obtained from these 12 studies are presented in Table 2.

We plotted the results of Table 2 in Figure 3. The lower solid line is the FAS prediction corresponding to SCr/Q = 1.33; the upper solid line is the FAS prediction corresponding to SCr/Q = 0.67. An alternative upper limit for FAS is shown as the dashed line. This alternative upper limit is based on symmetrical properties (normality for mGFR) and defined by using the ΔFAS (i.e. mean FAS prediction – LRL) to define the URL as mean FAS + ΔFAS.
In this study we challenged the mean GFR of 107.3 mL/min/1.73 m² defined by the new FAS equation for healthy young people, corresponding to a mean SCr/Q of 1. The age/gender subgroup analysis of the mGFRs of a group of 633 potential live kidney donors did not provide evidence to reject the hypothesis that the mean GFR equals 107.3 mL/min/1.73 m² for individuals up to 50 years of age, but there was statistical evidence to reject the hypothesis that the mean GFR equals 120 mL/min/1.73 m² in all age groups. This finding is also supported by our recent meta-analysis [6].

The mean value of 107.3 mL/min/1.73 m² was first introduced by Pottel et al. [12, 13] and was based on non-indexed mean mGFR values in children published by Piepsz et al. [14]. Using Belgian national growth curves for children [15], Pottel et al. indexed these mGFR values for body surface area (BSA) and fitted the obtained results against age, which resulted in the constant mean value for children 2–14 years of age of 107.3 mL/min/1.73 m². This finding resulted in a very simple height-independent eGFR formula for children [13], which has been extended by Hoste et al. [16] to adolescents and young adults. In two recent publications [1, 2], this simple equation was further extended to adults and older adults and to other biomarkers (cystatin C). The coefficient of 107.3 mL/min/1.73 m² is obviously the most important value in the equation and it was assigned the interesting property of being the average GFR value for healthy children, adolescents and young adults.

The new FAS equation claimed that this value could be maintained up to the age of 40 years, which was defined as age threshold where age-dependent renal decline begins. The findings we have observed here correspond very well with the findings in our recent meta-analysis study [6]: the mean mGFR is 107.3 mL/min/1.73 m² for young adults (up to 40 years of age) and then declines, with an average decline rate of 0.92 mL/min/1.73 m²/year between 40 and 100 years of age.

In this study on healthy kidney donors, we did not find a difference between males and females <50 years of age, but beyond 50 years there is a faster renal decline in females compared with males, a finding that we also observed in the meta-analysis study [6]. Poggio et al. [17] found a statistically significant difference between men and women <50 years of age, but the authors underlined that this difference of 3% was not clinically relevant. Rule et al. [18] also presented mean values (2.5th–97.5th percentiles) of mGFR (iothalamate) for 365 healthy potential living kidney donors. In his analysis, Rule et al. did not observe differences between men and women. They implicitly assumed a linear renal decline starting at 18 years of age and regressed the mGFR data over the full age range of 18–71 years, observing a slope or decline of 4.9 mL/min/1.73 m²/decade. The FAS equation would predict a renal decline of 6.3 mL/min/1.73 m² over the same age range.

Grewal and Blake [19] performed a study on 428 subjects (218 females, 210 males; age range 19–72 years) undergoing assessment as live kidney donors. GFR was evaluated from 51Cr-EDTA uptake (Fig. 2).

![Fig. 2. Potential (healthy) kidney donors: n = 633 [550, Lyon, France (solid gray circles); 41, Liège, Belgium (open squares); 42, Leuven, Belgium (solid black circles)]. Of mGFR values, 97.2% lie between the upper and lower limit defined by the FAS equation with SCr/Q = 1.33 and SCr/Q = 0.67, respectively (upper and lower solid lines). Of mGFR values, 91.8% lie between the symmetrical upper and lower eGFR limit (lower solid line and dashed line). The middle solid line corresponds with SCr/Q = 1, the average FAS prediction for GFR.](https://academic.oup.com/ckj/article-abstract/10/4/545/3782683/548-H-Pottel-et-al)
plasma clearance and they presented a mean GFR of 103.4 mL/min/1.73 m² for 187 subjects 19–40 years of age, for whom they also found that the dependence of GFR on age was not statistically significant, a finding that confirms our results. They also found a difference between men and women of 1.3 mL/min/1.73 m², which was not statistically significant, another finding that was confirmed here. In 241 subjects, 40–72 years of age, they found that GFR decreased by 0.91 mL/min/1.73 m²/year. They presented a model for the decline of GFR with age in which GFR remains constant at 103.4 mL/min/1.73 m² until the age of 40 years and then declines at a rate of 0.91 mL/min/1.73 m²/year. These reference data have been used as the basis for defining minimal age-dependent GFRs in living donors by the British Transplantation Society [20]. These results are very similar to our findings. Our mean value of 107.3 mL/min/1.73 m² is 4% higher than the mean value of 103.4 mL/min/1.73 m² reported by Grewal and Blake, a difference that can be attributed to the difference in measurement method [6]. At the age of 40 years, the FAS equation predicts a mean value of 107.3 mL/min/1.73 m², and at the age of 100 years the mean value is predicted to be 107.3 × 0.986(100–40) = 52.0 mL/min/1.73 m², resulting in an average decline rate calculated over 60 years of (107.3–52.0)/60 = 0.92 mL/min/1.73 m²/year, nearly exactly the value reported by Grewal and Blake. The British Transplantation Society also proposes a safety limit of 80 mL/min/1.73 m² for adults up to the age of 46 years (corresponding with FAS-eGFR = 107.3/1.33 = 81 mL/min/1.73 m²) and declining to 50 mL/min/1.73 m² at the age of 80 years, which is exactly the value predicted by the FAS equation, corresponding to Scr/Q = 1.33, eGFR = 107.3/1.33 × 0.986(60–40) = 50 mL/min/1.73 m². Grewal and Blake presented mean GFR and the upper and lower boundaries were defined as a mean ± 2SD of 103.4 ± 28.9 for individuals <40 years of age (thus ranging from 75 to 132.3 mL/min/1.73 m²) and for those >40 years of age, there is an additional decline of 0.91 mL/min/1.73 m²/year.

Hamilton et al. [21] presented mean total plasma clearance values of 51Cr-EDTA in 201 potential kidney donors 16–60 years of age of Saudi Arabian origin, categorized in 10-year groups, showing no decline with age in the 20–50 years age range. They also did not observe a difference between men and women.

Soares et al. [22] measured GFR by the 51Cr-EDTA single-injection method in 285 healthy Brazilian individuals 19–70 years of age. After stratifying data by age decades, they found that GFR starts to decline significantly after 45 years of age. Regression analysis of GFR versus age for subjects <45 years of age did not give a slope significantly different from zero, whereas the slope of the older groups was consistent with a significant decrease in GFR. They defined reference GFR values (mean ± 2SD) ranging from 76 to 148 mL/min/1.73 m² for individuals <45 years of age and 68 to 128 mL/min/1.73 m² for subjects >45 years of age. After adjustment for BSA, they also did not find a difference in mean GFR between men and women, <45 years of age (111 ± 18 versus 113 ± 18 mL/min/1.73 m²). For subjects >45 years of age, they observed an age decline in both men and women, but with a trend towards lower GFRs in older women.

Blake et al. [23] presented a non-linear relationship between mean mGFR and age of GFR of 103.9 – 0.0061 × Age² based on a retrospective study of 904 subjects (468 women, 436 men; age range 18–84 years) undergoing assessment as prospective living kidney donors. GFR was evaluated from 51Cr-EDTA plasma clearance using the slope-intercept method [9]. Their analysis was based on evaluating models of the form mGFR = A – B × Age², which implicitly assumes that decline starts at the lowest age in the range under study. Blake et al.’s model and the FAS model are clearly different from a physiological point of view, as the FAS model assumes a decline starting at the age of 40 years. Blake et al. defined the upper and lower limit as ±2SD (±25.8 mL/min/1.73 m²) from the mean (mGFR = 103.9 – 0.0061 × Age²). Although the lower limit defined by Blake et al. is only
slightly lower than the lower limit defined by FAS, the upper limit is much higher for FAS than the upper limit defined by Blake et al.

Herein we also used the data from our meta-analysis study [6] to calculate URls and URLs and found a very good correspondence between the mGFR = 2 × SD and the FAS lower limit obtained from SCr/Q = 1.33. The upper limit for GFR (mGFR + 2 × SD) is much lower than the FAS prediction corresponding to SCr/Q = 0.67. A possible reason could be that the FAS equation overestimates GFR for low SCr/Q values due to the power coefficient of 1 in the inverse relationship of GFR with SCr/Q. Children with a muscular disorder (e.g. like Duchenne Muscular Dystrophy patients) have extremely low SCr/Q values while still maintaining normal kidney function [24]. Consequently, these subjects have normal mGFR but overestimated (too high) eGFR values (for SCr-based eGFR equations). Thus, too low SCr/Q values may produce incorrect GFR estimates, but the question remains where this threshold lies. However, this is only one possible explanation, as it should also be noted that SCr/Q has a well-established bell-shaped distribution, in other words, it has a Gaussian distribution. Because of the inverse relationship between eGFR and SCr/Q, eGFR can never be distributed in a Gaussian manner when SCr/Q is normally distributed. On the other hand, all studies on kidney donors aimed at defining reference intervals for mGFR assume that the mGFR distribution is Gaussian and thus that mGFR reference intervals can be defined as mean ± 2SD. The value of this assumption is difficult to evaluate since GFR declines with age. Clearly, the lower limit (LL) of GFR is the most important reference limit and can be easily expressed as a simple mathematical equation:

\[
\text{GFR(LL)} = 80.7 \times \text{min} /1.73 \text{m}^2 \quad \text{for} \quad < 40 \text{years of age and }
\]

\[
\text{GFR(LL)} = 80.7 \times 0.988^{\text{Age} - 40} \text{min} /1.73 \text{m}^2 \quad \text{for} \quad \leq 40 \text{years of age}
\]

**Conflict of interest statement**

None declared.

**References**

1. Pottel H, Hoste L, Dubourg L et al. An estimating glomerular filtration rate equation for the full age spectrum. Nephrol Dial Transplant 2016; 31: 798–806

2. Pottel H, Delaney P, Schaeffer N et al. Estimating glomerular filtration rate for the full age spectrum from serum creatinine and cystatin C. Nephrol Dial Transplant 2017. doi:10.1093/ndt/gfw425

3. Smith H. Comparative physiology of the kidney. In: Smith H (ed). The Kidney: Structure and Function in Health and Disease. New York: Oxford University Press, 1951, pp. 520–574

4. Wesson L. Physiology of the Human Kidney. New York: Grune & Stratton, 1969

5. Levey AS, Stevens LA, Schmid CH et al. A new equation to estimate glomerular filtration rate. Ann Intern Med 2009; 150: 604–612

6. Pottel H, Hoste L, Yayo E et al. Glomerular filtration rate in healthy living potential kidney donors: a meta-analysis supporting the construction of the full age spectrum equation. Nephron 2017; 135: 105–119

7. Hadj-Aissa A, Bankir L, Frayssé M et al. Influence of the level of hydration on the renal response to a protein meal. Kidney Int 1992; 42: 1207–1216

8. Dubourg L, Hadj-Aissa A, Ferrier B. Adaptation of an enzymatic polyfructosan assay to clinical practice. Anal Biochem 2010: 405: 266–268

9. Bröchner-Mortensen J, Haahr J, Christoffersen J. A simple method for accurate assessment of the glomerular filtration rate in children. Scand J Clin Lab Invest 1974; 33: 139–143

10. Soveri I, Berg UB, Björk J et al. Measuring GFR: a systematic review. Am J Kidney Dis 2014; 64: 411–424
11. Delanaye P, Ebert N, Melsom T et al. Iohexol plasma clearance for measuring glomerular filtration rate in clinical practice and research: a review. Part 1: how to measure glomerular filtration rate with iohexol? Clin Kidney J 2016; 9: 682–699
12. Pottel H, Mottaghy FM, Zaman Z et al. On the relationship between glomerular filtration rate and serum creatinine in children. Pediatr Nephrol 2010; 25: 927–934
13. Pottel H, Hoste L, Martens F. A simple height-independent equation for estimating glomerular filtration rate in children. Pediatr Nephrol 2012; 27: 973–979
14. Piepsz A, Tondeur M, Ham H. Escaping the correction for body surface area when calculating glomerular filtration rate in children. Eur J Nucl Med Mol Imaging 2008; 35: 1669–1672
15. Roelants M, Hauspie R, Hoppenbrouwers K. References for growth and pubertal development from birth to 21 years in Flanders, Belgium. Ann Hum Biol 2009; 36: 680–694
16. Hoste L, Dubourg L, Selistre L et al. A new equation to estimate the glomerular filtration rate in children, adolescents and young adults. Nephrol Dial Transplant 2013; 29: 944–947
17. Poggio ED, Rule AD, Tanchanco R et al. Demographic and clinical characteristics associated with glomerular filtration rates in living kidney donors. Kidney Int 2009; 75: 1079–1087
18. Rule AD, Gussak HM, Pond GR et al. Measured and estimated GFR in healthy potential kidney donors. Am J Kidney Dis 2004; 43: 112–119
19. Grewal GS, Blake GM. Reference data for $^{51}$Cr-EDTA measurements of the glomerular filtration rate derived from live kidney donors. Nucl Med Commun 2005; 26: 61–65
20. Fleming JS, Zivanovic MA, Blake GM et al. Guidelines for the measurement of glomerular filtration rate using plasma sampling. Nucl Med Commun 2004; 25: 759–769
21. Hamilton D, Riley P, Miola U et al. Total plasma clearance of $^{51}$Cr-EDTA: variation with age and sex in normal adults. Nucl Med Commun 2000; 21: 187–192
22. Soares AA, Prates AB, Weinert LS et al. Reference values for glomerular filtration rate in healthy Brazilian adults. BMC Nephrol 2013; 14: 54
23. Blake GM, Sibley-Allen C, Hilton R et al. Glomerular filtration rate in prospective living kidney donors. Int Urol Nephrol 2013; 45: 1445–1452
24. Braat E, Hoste I, De Waele L et al. Renal function in children and adolescents with Duchenne muscular dystrophy. Neuromuscul Disord 2015; 25: 381–387
25. Inker LA, Shaﬁ T, Okparavero A et al. Effects of race and sex on measured GFR: the multi-ethnic study of atherosclerosis. Am J Kidney Dis 2016; 68: 743–751
26. Berg UB. Differences in decline in GFR with age between males and females. Reference data on clearances of inulin and PAH in potential kidney donors. Nephrol Dial Transplant 2006; 21: 2577–2582
27. Sasson AN, Cherney DZ. Renal hyperﬁltration related to diabetes mellitus and obesity in human disease. World J Diabetes 2012; 3: 1–6