The 21st Budapest Nephrology School
Nephrology, Hypertension, Dialysis, Transplantation

Estimation and Measurement of GFR
Past, Present and Future

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Let me start with a question
As GFR falls, the creatinine and BUN rise.
Glomerular Filtration Rate (GFR)

- GFR estimates are used to:
  - Estimate measured GFR
  - Predict risk for adverse outcomes
  - Staging of CKD
  - ESRD risk
  - CV risk
  - Overall mortality risk
Glomerular Filtration Rate (GFR)

But, what is the best way to find exact GFR?
Le mieux est l'ennemi du bien.

The best is the enemy of the good.

François-Marie Arouet (November 21, 1694 – May 30, 1778)
Which one of the following does show your patient’s GFR most accurately?

A) Measured GFR
B) Estimated GFR
Does anyone of you measured GFR at any time?

A) Yes

B) No
Which one of the following markers does represent the gold standard for measuring GFR?

A) Inulin  
B) 51Cr-EDTA  
C) 99Tc-DTPA  
D) Iothalamate  
E) Ioxehol
Do you know what is inulin?

A) A synthetic sugar
B) A muscle breakdown product
C) A naturally occuring polysaccharide
D) A natural alcohol derivative
E) A metabolic by-product of intestinal bacteria
What will I tell?

- GFR and concept of renal clearance
- Measuring GFR: From past to present
- Estimating GFR: From past to present
- Measured vs Estimated GFR: Advantages vs disadvantages and clinical use
- Future prospects
Renal Clearance

• Renal clearance of a substance is defined as the volume of plasma cleared from this substance per time unit (mL/min).

\[
C_x = \frac{U_x \times \dot{V}}{P_x^a}
\]

• Clearance is thus a virtual volume but will permit to apprehend GFR and renal function.

The concept of clearance: History

• The Danish physiologist, Poul Brandt Rhehberg was the first to use and define the concept of clearance in 1926 even if this author did not use the word “clearance”.

• Rhehberg studied on himself the urea and creatinine clearances to prove that kidney has a filtrating and not only a secreting action.

LX. STUDIES ON KIDNEY FUNCTION.
I. THE RATE OF FILTRATION AND REABSORPTION IN THE HUMAN KIDNEY.

By POUL BRANDT REHBERG.

From the Laboratory of Zoophysiology, University of Copenhagen.

(Received April 26th, 1926.)
The concept of clearance: History

• The term clearance was used for the first time by Möller in 1929 and was then concerning the urea clearance which was proposed as the first evaluation of renal function.

STUDIES OF UREA EXCRETION. II.

RELATIONSHIP BETWEEN URINE VOLUME AND THE RATE OF UREA EXCRETION BY NORMAL ADULTS¹

By EGGERT MÖLLER, J. F. McINTOSH and D. D. VAN SLYKE
(From the Hospital of the Rockefeller Institute for Medical Research, New York)
(Received for publication August 21, 1928)
THE EXCRETION OF URINE IN THE DOG

III. The Use of Non-metabolized Sugars in the Measurement of the Glomerular Filtrate

NORMAN JOLLIFFE, JAMES A. SHANNON and HOMER W. SMITH

From the Department of Physiology, University and Bellevue Hospital Medical College,
New York City

Received for publication December 29, 1931

“No substance is known to be present normally in the plasma and urine of vertebrates which can be safely used to evaluate the quantity of glomerular filtrate under physiological conditions”
"Creatinine of exogenous origin has been recommended for this purpose by Rehberg, (1926) but when it is recognized that this substance is secreted by the renal tubules of the lower vertebrates (Marshall and Grafflin, 1932; Clarke and Smith, 1932) and that it is not present normally in significant quantities in the blood of mammals (Behre and Benedict, 1922; Gaebler and Keltch, 1928; Gaebler, 1930) its use is open to question."
The properties which we believe such a substance should possess are as follows:

1. It must be determinable in plasma and urine with quantitative accuracy.
2. It must be non-toxic and it must exert no local stimulating or depressing action upon the kidney.
3. It must be completely filtrable from plasma.
4. It must not be secreted by the renal tubules.
5. It must not be reabsorbed by the renal tubules.
They concluded that inulin was a near perfect filtration marker, which was eventually verified conclusively by micropuncture and tracer studies. It remains the standard against which filtration markers are gauged.
Measuring GFR

- Endogenous production
- Exogenous addition

Volume of distribution

Plasma concentration

- Tubule secretion
- Tubule reabsorption
- Glomerular filtration

Extrarenal elimination
Renal elimination

Plasma Clearance

Renal Clearance

Measured GFR Methods

1) Inulin
2) 51Cr-EDTA (Ethylenediaminetetraacetic acid)
3) 99Tc-DTPA (Diethylenetriaminopentaacetic acid)
4) Iothalamate
5) Ioxehol

Inulin

- Inulin is a polymer of fructose which is found in some plants.
- Its molecular weight is 5200 Da (Gaspari et al., 1997).
- Humans are not able to metabolize inulin.
- Inulin was freely and fully filtrated through the glomerulus.
- Absence of both tubular absorption and secretion.

Inulin

- **Limitations to its use** in daily practice.
  - Because its relatively **high molecular weight** (5200 Da), the molecule is relatively viscous and don’t quickly reach its volume of distribution. Therefore, only methods using **urinary clearance** with constant infusion rate seem accurate.
  - **Difficulty linked to its measurement** in urine and plasma.
  - There is **no standardization** in inulin measurement.

Inulin

The scarcity, cumbersome methodology and high cost of inulin all but eliminated its routine use.

Direct measurement of glomerular filtration rate (GFR) is impossible because the filtration process simultaneously takes place in millions of glomeruli and filtrate composition and volume change when passing through the kidney.
Which one is the best for measuring GFR?

**Index Tests**

- Endogenous creatinine clearance
- $^{51}\text{Cr-EDTA}$
  - Renal * Plasma
- DTPA
  - Renal * Plasma
- Ioxehol
  - Renal * Plasma
- Iothalamate
  - Renal * Plasma
- Inulin (plasma)

**Reference Test**

Renal inulin clearance measured under continuous inulin infusion and urine collection

- Endogenous creatinine clearance

- $^{51}$Cr-EDTA
  - Renal (strong evidence)
  - Plasma (strong evidence)

- Iothalamate
  - Renal (strong evidence)
  - Plasma (limited evidence)

- Inulin (plasma)
  - Sufficient accuracy (limited evidence)
Estimated GFR Methods
Estimated GFR Methods

• Clinically, GFR is often estimated from the serum concentration of endogenous filtration markers.
• Endogenous markers
  – Urea (used widely in the past)
  – Creatinine (most commonly used)
  – Cystatin C (promising new one)
Endogenous Marker

Serum Creatinine

- The most commonly used parameter to evaluate kidney function in routine clinical practice.

- Very convenient, cheap, and readily available technique.

Serum Creatinine

- **Generation**: Muscle mass, dietary meat, age, gender, racial differences
- **Tubular secretion** (5-10% of excreted creatinine)
- **Extrarenal elimination**
- **Measurement technique**
  - Alkaline picrate (Jaffe)
  - Enzymatic
  - HPLC
  - IDMS (*isotope dilution mass spectrometry*)

Creatinine Clearance

- 24 hour urine collection
  - Difficult
  - Less or much collection
- Tubular secretion
- Not accurate

Cockroft-Gault Equation

- The oldest (developed in 1973) but simplest equation for everyday clinical use
- Derived using data from 249 men with a creatinine clearance ranging from approximately 30–130 ml/min

\[
eCrCl = \frac{(140 - \text{Age}) \times \text{Weight (kg)}}{72 \times \text{Creatinine}_{\text{serum}} (\text{mg/dL})} \times 0.85 \text{ if female}
\]

Cockroft-Gault Equation

• Derived when standardized creatinine assays were not in use.
• In labs where standardized creatinine assays were used, this equation will cause an overestimation (10–40 %) of actual GFR.
• Not adjusted for body surface area.
• Less accurate in
  – obese patients (overestimate),
  – in patients with normal or mildly decreased GFR (underestimates), and
  – in the elderly (underestimates).

It is with amusement, that after almost 20 years as an academic “asthmatoologist,” that house officers recognize my name as that attached to the widely used “Cockcroft-Gault” formula for predicting creatinine clearance.
Developed in 1999 by using data from 1628 CKD patients, with nondiabetic kidney disease, with a GFR range between 5 and 90 ml/min/1.73 m².

The equation was re-derived in 2006 for use with the standardized serum creatinine assays.

MDRD Study Equation

\[
GFR \ (\text{ml/min/1.73 m}^2) = 186.3 \times \text{Scr}^{-1.154} \times \text{age}^{-0.203} \times (0.742 \text{ if female}) \times (1.210 \text{ if African American}),
\]

where Scr is expressed in mg/dl and age is expressed in years.

\[
GFR \ (\text{ml/min/1.73 m}^2) = 175 \times \text{Scr}^{-1.154} \times \text{age}^{-0.203} \times (0.742 \text{ if female}) \times (1.210 \text{ if African American}),
\]

where a standardized Scr (mg/dl) measurement is done.

MDRD Study Equation

- Most widely used formula
- Many laboratories automatically report
- More accurate in patients with lower GFR levels (<60 ml/min/1.73 m2)
- Less accurate in obese patients and in patients with normal or mildly decreased GFR.

CKD-EPI Equation

• Derived in 2009 from a large study population that included patients with or without kidney disease with a wide range of GFR.

• When compared with MDRD, CKD-EPI has found to be more accurate in people especially with higher GFR levels.

This is the formula...

\[
GFR \text{ (ml/min/1.73 m}^2) = 141 \times \min(\text{SCR}/\kappa, 1)^\alpha \times \max(\text{SCR}/\kappa, 1)^{-1.209} \times 0.993^{\text{Age}} \\
\times (1.018 \text{ if female}) \times (1.159 \text{ if African American}),
\]

where SCR is serum creatinine (in mg/dl), \(\kappa\) is 0.7 for females and 0.9 for males, \(\alpha\) is \(-0.329\) for females and \(-0.411\) for males, \(\min\) indicates the minimum of \(\text{SCR}/\kappa\) or 1, and \(\max\) indicates the maximum of \(\text{SCR}/\kappa\) or 1.

**Female**

\(< 0.7 \text{ mg/dl} \quad GFR = 144 \times (\text{Scr}/0.7)^{-0.329}\)

\(> 0.7 \text{ mg/dl} \quad GFR = 144 \times (\text{Scr}/0.7)^{-1.209}\)

\(\times (0.993)^{\text{Age}} \times 1.157 \text{ [if black]}\)

**Male**

\(< 0.9 \text{ mg/dl} \quad GFR = 141 \times (\text{Scr}/0.9)^{-0.411}\)

\(> 0.9 \text{ 7mg/dl} \quad GFR = 141 \times (\text{Scr}/0.9)^{-1.209}\)

Comparison of the Performance of the MDRD Study and CKD-EPI equations (Validation dataset)

Estimated GFR (mL/min/1.73 m^2)

<table>
<thead>
<tr>
<th>Method</th>
<th>All</th>
<th>60-89</th>
</tr>
</thead>
<tbody>
<tr>
<td>MDRD Study</td>
<td>5.5</td>
<td>11.9</td>
</tr>
<tr>
<td>CKD-EPI</td>
<td>2.5</td>
<td>4.2</td>
</tr>
<tr>
<td>%Δ</td>
<td>50%</td>
<td>59%</td>
</tr>
</tbody>
</table>


Slide courtesy of Lesley A Stevens, MD, MS
Comparison of distribution of estimated GFR for MDRD Study and CKD-EPI equations (NHANES 1999-2004)

Values are plotted at the midpoint.

Slide courtesy of Lesley A Stevens, MD, MS

Levey et al *Ann Int Med* 2009; 150: 604 612
All GFR equations (incl. MDRD & CKD-EPI)

• Some imprecision and several limitations
  • Use of serum creatinine and its limitations
• All need to be used in **steady state**
  – Don’t use in AKI
• **Not recommended for use** in patients
  – under the age of 18,
  – with extremes in body size or muscle mass,
  – with severe alterations in dietary intake (vegetarians, using creatine supplements),
  – in very elderly (>85 years),
  – in pregnant patients.

Neither the CKD-EPI nor the MDRD Study equation is optimal for all populations and GFR ranges. Using a single equation for reporting requires a tradeoff to optimize performance at either higher or lower GFR ranges. A general practice and public health perspective favors the CKD-EPI equation.
CKD-EPI Equation

*KDIGO 2012 clinical practice guideline for the evaluation and management of chronic kidney disease recommends to use CKD-EPI equation for GFR estimation.*
Serum Cystatin C and GFR Equations

- A 122 amino acid with molecular weight (13-kDa) cysteine protease inhibitor
  - Produced by all nucleated cells
- Freely filtered by the renal glomerulus
- No tubular secretion
- Reabsorbed and completely catabolized by tubular cells, not excreted

Cystatin C.....drawbacks

- Cystatin C generation rate and serum levels have been influenced by age, sex, cell turnover rate, steroid use, body mass index, inflammation, and diabetes
- There is an extrarenal elimination of cystatin C at low levels of GFR
- Cystatin C measurements are not standardized yet and still evolving

Cystatin C.....drawbacks

• Other studies have suggested that inflammation, adiposity, thyroid diseases, certain malignancies, smoking, and use of glucocorticoids may increase cystatin C levels.

Cystatin C and Outcomes

• Higher levels of cystatin C are a better predictor of the risk of cardiovascular disease and total mortality.

Cystatin C use in GFR equations

- Cystatin C itself or equations based on cystatin C alone are not more accurate than creatinine-based estimating equations, rather, it is the combination of the two markers that results in the most accurate estimate in populations with and without CKD.

the risk of death was increased when values for both cystatin C–based eGFR and eGFR based on combined creatinine and cystatin C measurements were below a threshold of approximately 85 ml per minute per 1.73 m².
Formulas again....

CKD-EPI Cystatin C equation:

\[ GFR \text{ (ml/min/1.73 m}^2\text{)} = 133 \times \min(\text{SCysC/0.8, 1})^{-0.499} \times \max(\text{SCysC/0.8, 1})^{-1.328} \times 0.996 \text{Age [\times 0.932 if female]} , \]

where SCysC is serum cystatin C (in mg/l), min indicates the minimum of SCysC/0.8 or 1, and max indicates the maximum of SCysC/0.8 or 1.

CKD-EPI Creatinine-Cystatin C equation:

\[ GFR \text{ (ml/min/1.73 m}^2\text{)} = 135 \times \min(\text{SCR/\kappa, 1})^{\alpha} \times \max(\text{SCR/\kappa, 1})^{-0.601} \times \min(\text{SCysC/0.8, 1})^{-0.375} \times \max(\text{SCysC/0.8, 1})^{-0.711} \times 0.995 \text{Age [\times 0.969 if female]} [\times 1.08 \text{ if black}], \]

where SCR is serum creatinine (in mg/dl), SCysC is serum cystatin C (in mg/l), \( \kappa \) is 0.7 for females and 0.9 for males, \( \alpha \) is –0.248 for females and –0.207 for males, \( \min(\text{SCR/\kappa, 1}) \) indicates the minimum of SCR/\kappa or 1, and \( \max(\text{SCR/\kappa, 1}) \) indicates the maximum of SCR/\kappa or 1; \( \min(\text{SCysC/0.8, 1}) \) indicates the minimum of SCysC/0.8 or 1 and \( \max(\text{SCysC/0.8, 1}) \) indicates the maximum of SCysC/0.8 or 1.
KDIGO 2012 clinical practice guideline for the evaluation and management of chronic kidney disease has recommended to measure cystatin C to confirm CKD in adults if eGFR based on serum creatinine was between 45 and 59 ml/min/1.73 m² without any markers of kidney damage.
To tie or conclude....
### Creatinine versus Cystatin C

<table>
<thead>
<tr>
<th>Variable</th>
<th>Creatinine</th>
<th>Cystatin C</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Molecular Properties</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weight</td>
<td>113 Da</td>
<td>13,300 Da</td>
</tr>
<tr>
<td>Structure</td>
<td>Amino acid derivative</td>
<td>Nonglycosylated basic protein</td>
</tr>
<tr>
<td><strong>PhysiologicDeterminants of Serum Level</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Generation</td>
<td>Varies according to muscle mass and dietary protein; lower in elderly persons, women, and whites</td>
<td>Made by all nucleated cells; thought to be mostly constant; increases in hyperthyroid states and with steroid use; lower in elderly persons and women</td>
</tr>
<tr>
<td>Handling by the kidney</td>
<td>Filtered, secreted, and excreted in urine</td>
<td>Filtered, reabsorbed, and catabolized</td>
</tr>
<tr>
<td>Extrarenal elimination</td>
<td>Yes; increases at reduced GFR</td>
<td>Preliminary evidence of increases at reduced GFR</td>
</tr>
<tr>
<td><strong>Use in GFR Estimating Equations</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Demographic and clinical variables as surrogates for physiologic determinants</td>
<td>Age, sex, and race; related to muscle mass</td>
<td>Age and sex</td>
</tr>
<tr>
<td>Factors associated with inaccurate estimates</td>
<td>Nonsteady state; GFR &gt; 60 mL/min/1.73 m²; conditions associated with alterations in muscle mass, drugs that inhibit tubular secretion, interferents with serum assays</td>
<td>Nonsteady state; GFR &gt; 60 mL/min/1.73 m²; conditions associated with alterations in thyroid or steroid hormones, possibly obesity</td>
</tr>
<tr>
<td><strong>Assay</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Method</td>
<td>Colorimetric and enzymatic</td>
<td>Immunoassays</td>
</tr>
<tr>
<td>Assay precision</td>
<td>Very good except at low range</td>
<td>Precision varies across assays</td>
</tr>
<tr>
<td>Clinical laboratory practice</td>
<td>Multiple assays; widely used; widely standardized</td>
<td>Not on most autoanalyzers; becoming standardized</td>
</tr>
<tr>
<td>Standardized reference materials</td>
<td>SRM 967</td>
<td>ERM-DA471/IFCC</td>
</tr>
<tr>
<td>Reference assay</td>
<td>IDMS</td>
<td>PENIA, PETIA, enzyme-amplified single radial immunodiffusion</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Endogenous Filtration Marker; Notation</th>
<th>Development Population</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Creatinine; eGFR\textsubscript{cr}</strong></td>
<td><strong>Group; Year</strong></td>
</tr>
<tr>
<td>MDRD Study; 1999</td>
<td>N = 1,628 with CKD; mean mGFR = 40</td>
</tr>
<tr>
<td>MDRD Study; 2006</td>
<td>Same as above</td>
</tr>
<tr>
<td>CKD-EPI; 2009</td>
<td>Diverse population, N = 8,254; mean mGFR = 68</td>
</tr>
<tr>
<td>CKD-EPI; 2008</td>
<td>N = 2,980 with CKD; mean mGFR = 48</td>
</tr>
<tr>
<td>CKD-EPI; 2011</td>
<td>Same as above</td>
</tr>
<tr>
<td>CKD-EPI; 2012</td>
<td>Diverse population, N = 5,352; mean mGFR = 68</td>
</tr>
<tr>
<td><strong>Cystatin C; eGFR\textsubscript{cys}</strong></td>
<td><strong>Group; Year</strong></td>
</tr>
<tr>
<td>CKD-EPI; 2008</td>
<td>N = 2,980 with CKD; mean mGFR = 48</td>
</tr>
<tr>
<td>CKD-EPI; 2011</td>
<td>Same as above</td>
</tr>
<tr>
<td>CKD-EPI; 2012</td>
<td>Diverse population, N = 5,352; mean mGFR = 68</td>
</tr>
</tbody>
</table>

### Comparison of Formulas

<table>
<thead>
<tr>
<th>Study Reference</th>
<th>Characteristics</th>
<th>GFR Measurement Method</th>
<th>Endogenous Filtration Marker Assays</th>
<th>Equation</th>
<th>Bias $^a$ (mL/min/1.73 m$^2$)</th>
<th>$P_{30}$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stevens et al$^{27}$ (JASN, 2007)</td>
<td>Diverse population; N = 5,504; mean mGFR = 68</td>
<td>Urinary clearance of iothalamate</td>
<td>Standardized</td>
<td>eClCr by Cockcroft-Gault 1976$^{28,b}$; eGFR$^{c}$_cr by 2006 MDRD Study</td>
<td>11.4</td>
<td>69</td>
</tr>
<tr>
<td>Levey et al$^{12}$ (Annals, 2009)</td>
<td>Diverse population; N = 3,771; mean mGFR = 68</td>
<td>Urinary clearance of iothalamate or EDTA, plasma clearance of iohexol</td>
<td>Standardized</td>
<td>eGFR$^{c}$_cr by 2006 MDRD Study$^{b}$; eGFR$^{c}$_cr by 2009 CKD-EPI</td>
<td>5.8$^c$</td>
<td>83$^c$</td>
</tr>
<tr>
<td>Inker et al$^{15}$ (NEJM, 2012)</td>
<td>Diverse population; N = 1,119; mean mGFR = 70</td>
<td>Urinary clearance of EDTA, plasma clearance of iohexol</td>
<td>Standardized</td>
<td>eGFR$^{c}$_cr by 2009 CKD-EPI$^b$; eGFR$^{c}$_cr-cys by 2012 CKD-EPI</td>
<td>3.7</td>
<td>87.2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Average of eGFR$_{cr}$ and eGFR$^{c}$_cr-cys by 2012 CKD-EPI</td>
<td>3.5</td>
<td>91.8$^g$</td>
</tr>
</tbody>
</table>

Newer Equations, Markers and Methods

- **Berlin Initiative Study (BIS)** equations for elderly based on standardized creatinine (BIS1) and cystatin C (BIS2) methodology. *Ann Intern Med* 2012;157:471-481.

- **Beta trace protein** (BTP, also known as prostaglandin D2 synthase)

- **Symmetric dimethylarginine** (SDMA)

- Dynamic contrast-enhanced **magnetic resonance imaging**
What about the opponents?

There are three kinds of people in the world, the wills, the won'ts and the can'ts. The first accomplish everything; the second oppose everything; the third fail in everything...

- Unknown
Opponents of GFR formula

- The use of eGFR should be limited to settings where knowing actual GFR is relevant and eGFR is more informative about GFR than serum creatinine or cystatin C alone. Such settings include staging CKD severity by GFR and dosing medications cleared by glomerular filtration.

- Alternatively, the diagnosis of CKD, the longitudinal progression of CKD, and prognostic models for CKD are settings where serum creatinine and cystatin C can be better applied and interpreted without eGFR.
What shall we do, then?
Guidelines

What shall we do, then?

It is not the numbers who diagnoses or treats a patient....but a skilled physician.
Thank you very much for your attention!