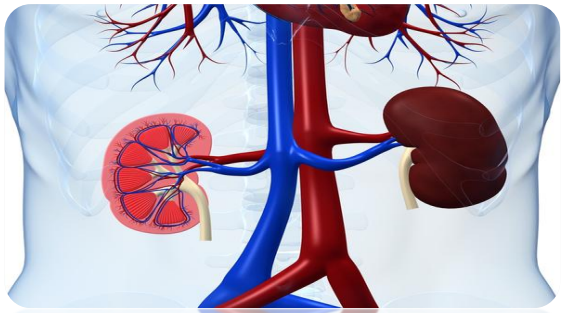
Estimation of Serum Creatinine,Urine Creatinine , and Creatinine Clearance

BCH 472



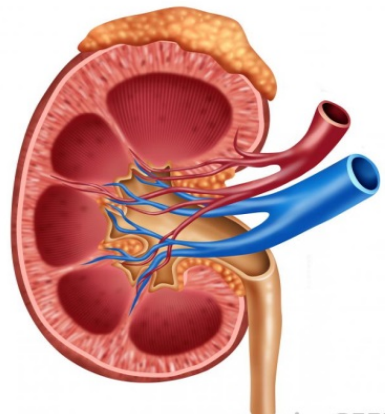
Kidney functions:

- **The kidneys serve three essential functions:**

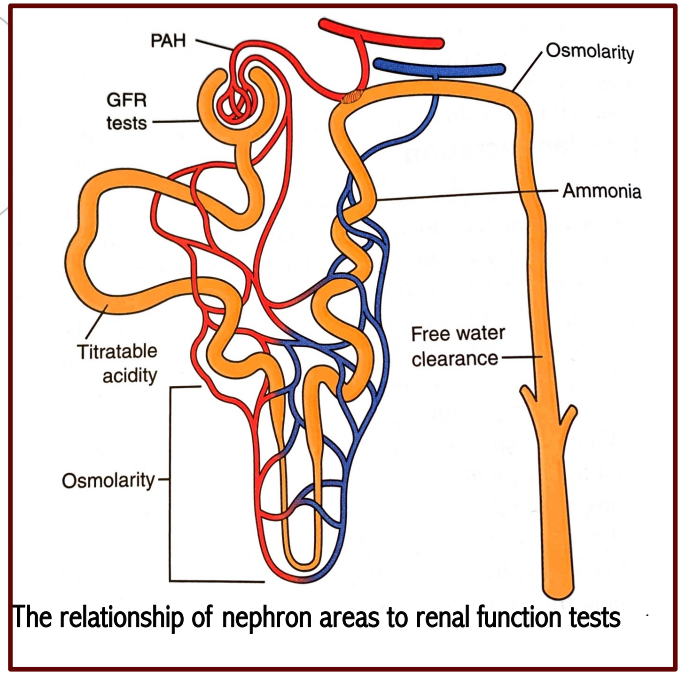
1. **They function as filters**, removing metabolic products and toxins from the blood and excreting them through the urine.

2. **They regulate the body's fluid status**, electrolyte balance, and acid-base balance.

3. **The kidneys produce or activate hormones** that are involved in erythrogenesis, Ca^{2+} metabolism, and the regulation of blood pressure and blood flow.



- **Renal function tests** are used to detect the presence of renal diseases and assess their progress.
- These tests include:
  - Glomerular Filtration Tests
  - Tubular Reabsorption Tests
  - Tubular Secretion Tests



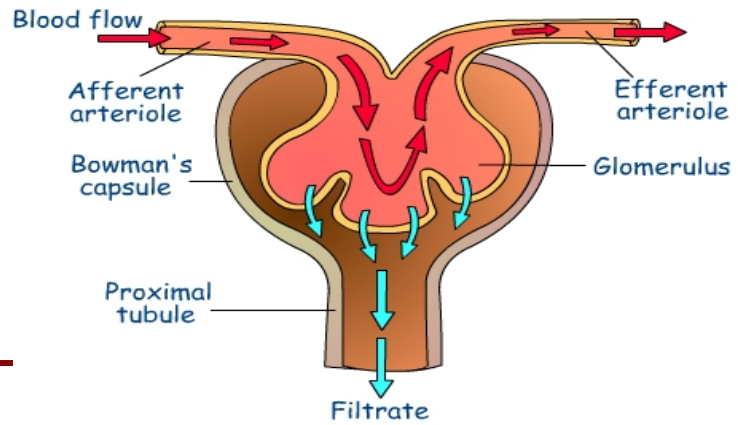
The relationship of nephron areas to renal function tests

- The most widely used test is to measure the **glomerular filtration rate** (GFR).
- **GFR** is an *important* and the *best* overall measurement in the evaluation of kidney function

Glomerular Filtration Rate:

- Under **normal** conditions, approximately 625 mL of plasma flow through the kidneys each minute and the volume of plasma **filtered** is 125 mL/ min which is called the glomerular filtration rate.

- **Glomerular filtration rate(GFR),** is the volume of plasma filtered by the kidneys in per unit of time.



Measuring the GFR:

- Accurate measurement of the GFR are done by **clearance tests.**
- **These tests** requires determination of the concentration, in plasma and urine, of a substance is known to be:
  - **completely filtered** from the plasma at the glomerulus.
  - must **not be reabsorbed nor secreted** by renal tubules, broken down, or accumulated by the tubules.
  - and must remain at a **constant** concentration in the plasma throughout the period of urine collection.

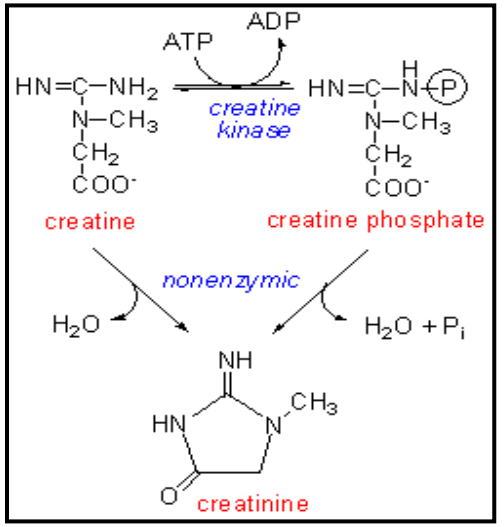
Substances clearance used for Measuring GFR

	Inulin Clearance	Creatinine Clearance	Urea Clearance
Source	Non-toxic fructose polymer	End-product of skeletal muscle creatine metabolism	endproduct of protein Metabolism
Advantages	Not reabsorbed or secreted	An endogenous product of muscle metabolism; near constant production, not reabsorbed	An endogenous product of protein
Disadvantages	Not made by body; must be injected (exogenous)	Small amount is secreted	Partially reabsorbed synthesis varies with diet

* **Creatinine clearance** is preferred because it is a normal constituent of blood and **no infusion** is needed unlike inulin. Moreover it is not reabsorbed by the tubules as in the case of urea.

Creatinine:

- Creatinine is derived from “creatine” which is synthesized in the liver, kidney and pancreas it moves through the circulation and is taken up entirely by muscles.



- In the muscles “**creatine**” is converted to creatine phosphate which becomes the source of a high energy phosphate bond for the immediate reformation of ATP.

- Endogenous creatinine production is **constant** as long as the muscle mass remains constant.

- If the filtration in the kidney is deficient, **creatinine blood levels rise**.

Creatinine clearance :

- Creatinine is cleared from the body fluids **almost entirely** by glomerular filtration

- **Tubules to variable degree secrete creatinine**, which by itself, would lead to an ~20% overestimate of GFR in humans.

- However, chromogens present in human plasma react in the chemical analysis helping to counteract the falsely elevated rates caused by tubular secretion.

- **Clearance is given by :**

Clearance = U.V / P

Where:

U= concentration of any substance in urine.

P= concentration of the same substance in plasma.

V= volume of urine (ml/min).

Serum Creatinine:

- **High serum creatinine**

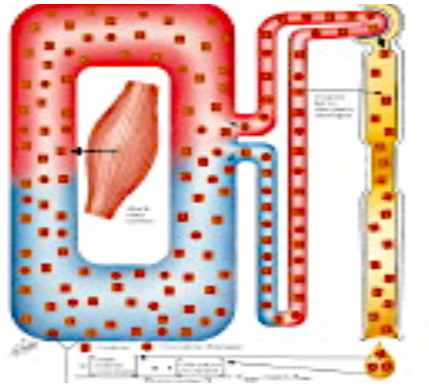
  Plasma creatinine tends to be higher in subjects with a large muscle mass.

- **Other non-renal causes of increased serum creatinine include the following :**

  • A high meat intake can cause a temporary increase.

  • Transient, small increases may occur after vigorous exercise .

* If non-renal cause does not exist, an **increased** plasma creatinine indicates a fall in GFR (renal disease)



Urine Creatinine

- **Decreased urine creatinine is found in:**
  - Advanced renal disease,
  - renal stenosis, narrowing of arteries that carry blood to one or both of the kidneys

- **Increased urine creatinine is found in:**
  - Diabetes mellitus
  - Starvation and fever

Clinical Implications:

1. Decreased creatinine clearance is found in any condition that decreases renal blood flow:

- Impaired kidney function.
- Shock, dehydration.
- Hemorrhage.
- Hypothyroidism.

2. Increased creatinine clearance is found in:

- Pregnancy.
- Hyperthyroidism.

Reference Values:

- Urine creatinine :1- 2 g/ 24h
- Serum creatinine: 0.6 – 1.2 mg/dL
- Normal creatinine clearance= 100-130 ml/min/1.73m^2

Note: What 1.73 m^2 means?

Kidney function is proportional to kidney size, which is proportional to body surface area. A 1.73 m^2 is the normal mean value of body surface area for young adults.

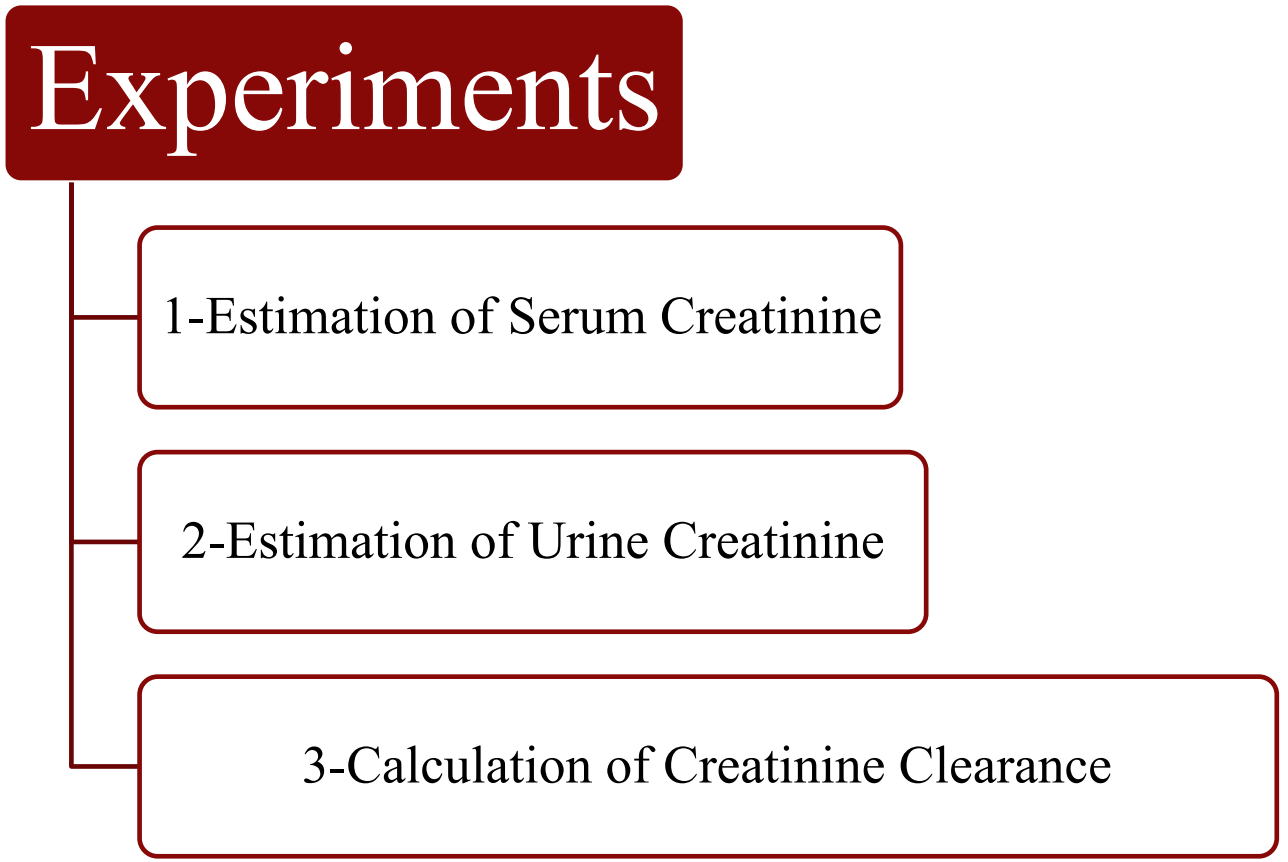
Adjustment for body surface area is necessary when comparing a patient's estimated GFR to normal values or to the levels defining the stages of Chronic kidney disease (CKD).

Chart 2 - **Chronic kidney disease staging**

Stage	Description	GF (ml/ min/1.73m²)
I	Kidney lesion with normal or increased GF	≥ 90
II	Kidney lesion with mild GF decrease	60-89
III	Kidney lesion with moderate GF decrease	30-59
IV	Kidney lesion with marked GF decrease	15-29
V	Functional kidney failure or undergoing SRT	< 15

SRT- substitutive renal therapy. Source: National Kidney Foundation, 2002.

Practical Part



Objective:

1- To estimate creatinine in serum and urine.

2- To calculate creatinine clearance value.

Principle:

(Jaffe's method):

Colorimetric estimation of creatinine using the alkaline picrate method.

Creatinine + picric acid ⟶ **Creatinine picrate** (orange)

Absorbance at 520nm

Method:

1-Set up a series of test tube as follows:

Chemical	Standard (serum)		Test (serum)		Standard (Urine)		Test (urine)		Blank
	(A)	(B)	(C)	(D)	(E)	(F)	(G)	(H)	
Water	1.5 ml	1.5 ml	1.5 ml	1.5 ml	1.5 ml	1.5 ml	1.5 ml	1.5 ml	2 ml
Standard (serum)	0.5 ml	0.5 ml	-	-	-	-	-	-	-
Serum Sample	-	-	0.5 ml	0.5 ml	-	-	-	-	-
Standard (Urine)	-	-	-	-	0.5 ml	0.5 ml	-	-	-
Urine Sample	-	-	-	-	-	-	0.5 ml	0.5 ml	-
Picric acid	6 ml	6 ml	6 ml	6 ml	6 ml	6 ml	6 ml	6 ml	6 ml

2-Immerse the Tubes carefully in the boiling water bath for 40 seconds.

4- Pipette 0.6 ml of NaOH to all tube

5- Let the tubes stand for 20 min.

6- Read the absorbance at **520 nm**.

Results:

Tube	Standard (serum)		Test (serum)		Test (urine)		Standard(Urine)	
	(A)	(B)	(C)	(D)	(E)	(F)	(G)	(H)
Absorbance at 520 nm								
Average (Mean of Absorbance)								

Calculation:

Patient information: 24h urine volume = 100ml, gender: women, body surface: 1.6m^2 , DF=100.

1-Serum creatinine =

$$\frac{\textit{Mean Absorbance of sample serum}}{\textit{Mean Absorbance of standard serum}}$$ X concentration of standard serum (3 mg/dl) = …… mg / dl

2-Urine creatinine =

$$\frac{\textit{Mean Absorbance of sample urine}}{\textit{Mean Absorbance of standard urine}}$$ X concentration of standard urine (0.75 mg/dl) X DF (100) = …… mg / dl

To compare with normal range of **urine creatinine** , convert from mg/dl to g/24 h

3- Creatinine Clearance : = U.V/ P

$$= \frac{\textbf{Urinary creatinine (mg/dl)}}{\textbf{serum creatinine (mg/dl)}}$$ **x Urine volume (ml/min)= A**

Adjustment for body surface area

A----------> 1.6 m^2 (person surface area)

? ----------> 1.73 m^2

-Corrected for surface area= **…….. ml/min/1.73 m^2**

Example:

Find the Creatinine Clearance = if you know that the Urine creatinine **U** = 488 mg/dl , Serum creatinine **P**= 2.32 mg/dl, Volume of urine in 24 h **V** =100 ml and A (surface area)=1.6 m^2

➔ Creatinine Clearance: = U.V/ P

= (488 mg/dl ÷2.32 mg/dl) × (100÷1440*)= 14.6 ml/min

14.6 ml/ min in 1.6 m^2, find the creatinine clearance for 1,73 m^2 surface area :

=(14.6 ×1.73)÷1.6= **15.8 ml /min/1.73m^2**

--------OR-------

➔ Creatinine Clearance: = (U XV X1.73)/ (P X 1440 X A)

= (488 mg/dl× 100× 1.73)/ (2.32× 1440× 1.6)

= **15.8 ml/min /1.73m^2**

* To convert 24 hour to min (24x60 = 1440)

Discussion:

- Comment on the concentration of **creatinine in serum**.
- Comment on the concentration of **creatinine in urine**.
- Comment on the value of **Creatinine Clearance** .

Question:

A man aged 35 years has a serum creatinine of 3 mg/dl. A 24 h urine of 2160 ml is collected and found to a creatinine concentration of 400 mg/dl

Calculate the Creatinine Clearance.