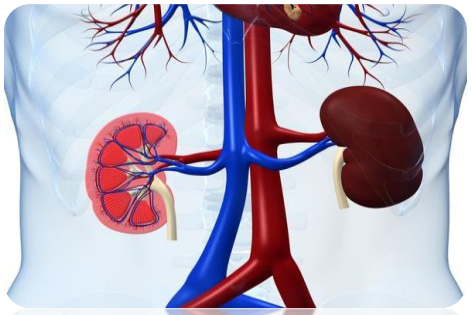
Estimation of Serum Creatinine,Urine Creatinine , and Creatinine Clearance

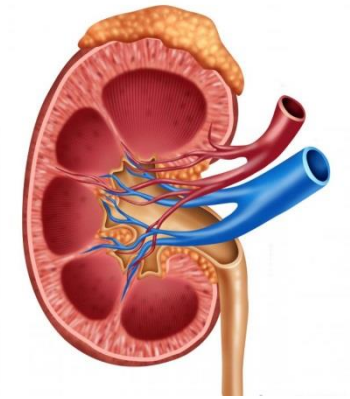
BCH 472



Kidney functions:

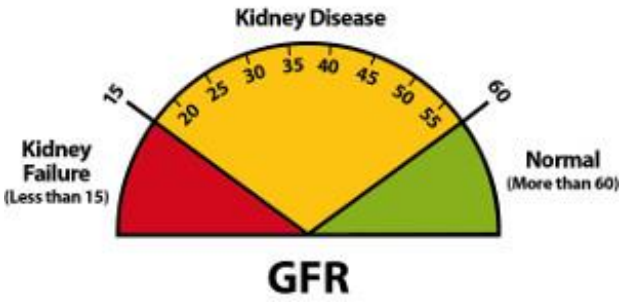
- **The kidneys serve three essential functions:**

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.

- The most widely used test is to measure the **glomerular filtration rate** (GFR), that is, the rate of filtrate formation by the kidneys.

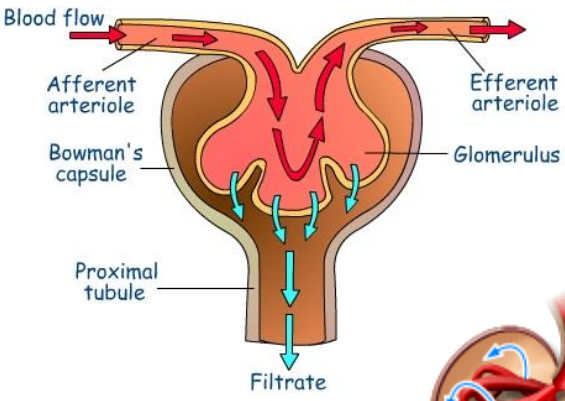


Glomerular Filtration Rate:

- 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.

- **GFR** is an important and the best overall measurement in the evaluation of kidney function.



Measuring the GFR:

- Accurate measurement of the GFR by **clearance 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.

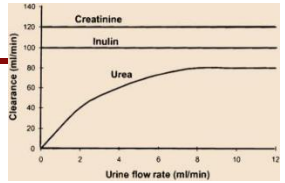
- This substance must **not** be reabsorbed nor secreted by renal tubules, broken down, or accumulated by the tubules and must remain at a constant concentration in the plasma throughout the period of urine collection.

- It's clearance is given by ,**clearance = U.V/ P**
- (U= urine creatinine, p= plasma creatinine, v= volume of urine)

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	An endogenous product of protein
Disadvantages	Not made by body; must be injected	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 <u>not reabsorbed</u> by the tubules as in the case of urea.

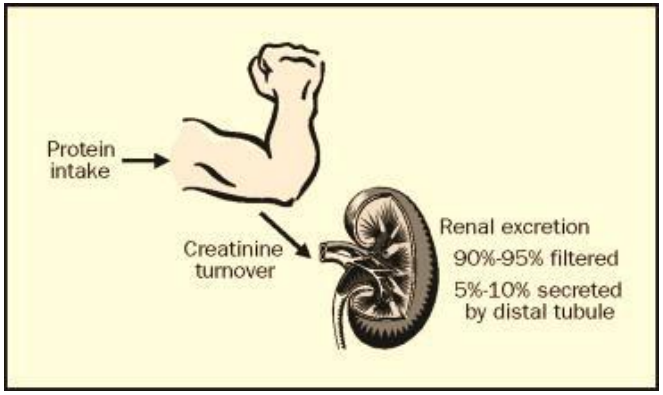


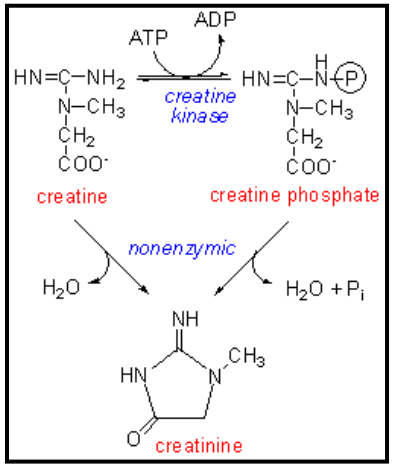
Creatinine:

- 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.

- **"Creatinine"** is the byproduct of muscle energy metabolism and is produced at a constant rate according to the muscle mass of the individual. It is a substance that, in health, is easily excreted by the kidney.

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





Serum Creatinine:

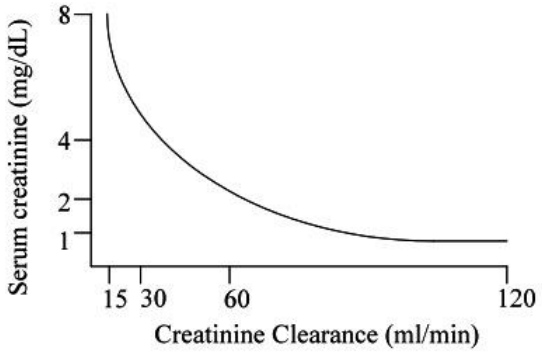
High plasma creatinine

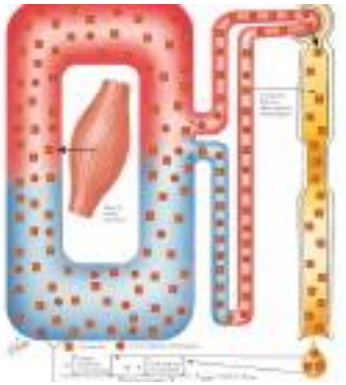
Plasma creatinine tends to be higher in subjects with a large muscle mass.

Other non-renal causes of increased plasma 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 can be excluded, an **increased** plasma creatinine indicates a fall in GFR





Urine Creatinine

Decreased urine creatinine is found in:

- There are many cases leads to a decrease in urine creatinine eg. Advanced renal disease, renal stenosis.

Increased urine creatinine is found in:

There are many cases eg. Diabetes mellitus

Creatinine clearance :

A measure of the amount of creatinine eliminated from the blood by the kidneys.

- Creatinine is cleared from the body fluids almost **entirely by glomerular filtration** (small amount is secreted by kidney tubules).
- Therefore, the clearance of creatinine can be used to assess **GFR**.

- Because measurement of creatinine clearance does **not** require intravenous infusion into the patient, this method is much more **widely** used than inulin clearance for estimating GFR clinically.

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

Clinical Implications:

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

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

2. Increased creatinine clearance is found in:

- Pregnancy

Reference Values:

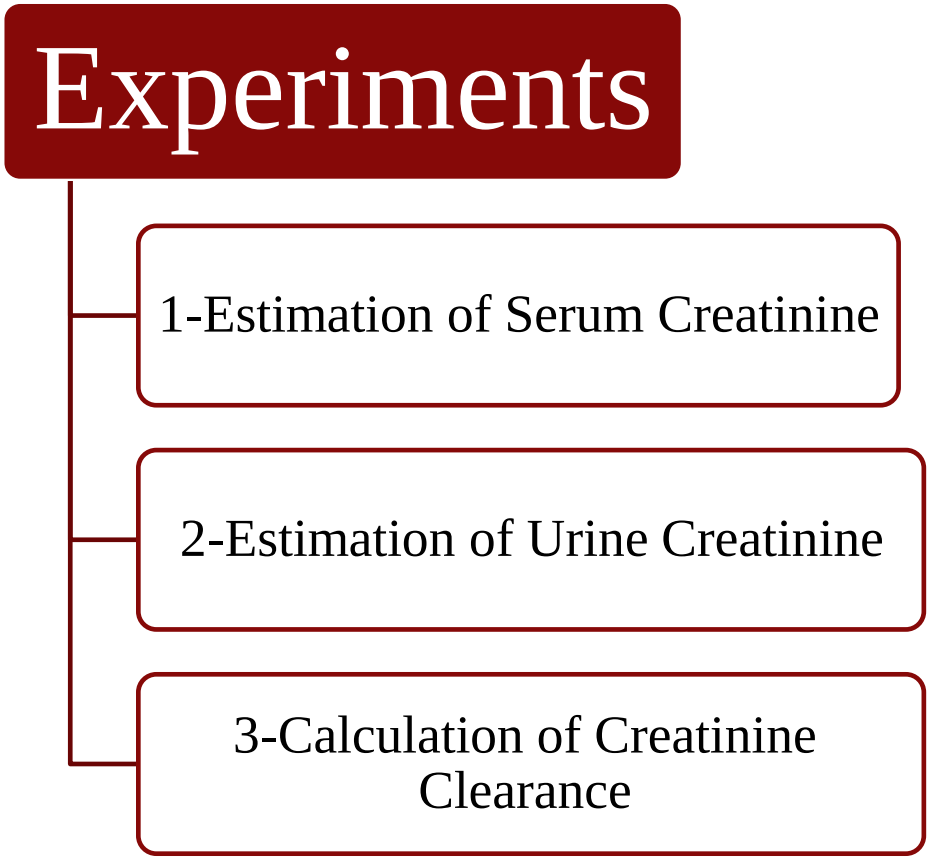
- Urine creatinine :1- 2 g/ 24h
- (serum) creatinine: 0.6–1.2 mg/dL
- Normal creatinine clearance= 100-130 ml/min/1.73m^2

Note: What 1.73 m^2 means?

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

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 $\longrightarrow$ 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:

1-Serum creatinine =

(Mean Absorbance of sample serum ÷ Mean Absorbance of Standard serum) X concentration of standard serum(3 mg/dl) = …… mg / dl

2-Urine creatinine =

(Mean Absorbance of sample urine ÷ Mean Absorbance of Standard) X concentration of standard urine(0.75 mg/dl) X DF (100) = …… mg / dl

To compare with normal range, convert from mg/dl to g/24 h

3- Creatinine Clearance : = U.V/ P

$$= \frac{\text{Urinary creatinine (mg/dl)}}{\text{plasmac reatinine (mg/dl)}} \text{ x Urine volume(ml/min)= A}$$

Note: 24h urine volume = 100ml, body surface= 1.6m2

- A----------→ 1.6 m2
- ?----------→ 1.73 m2
- **-Corrected for surface area= ml/min/1.73 m^2**

Creatinine Clearance:

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

A- Creatinine Clearance: = U.V/ P

For example: V = 100 ml/24 h **>> to convert hours to minutes /1440 "24x60"**

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

14.6 ml/ min **in 1.6 m^2**, find Creatinine clearance in **1.73m^2 :**

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

B- 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

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-Home work:

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