Estimation of Serum Creatinine, Urine Creatinine and Creatinine Clearance

-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, Ca2+ metabolism, and the regulation of blood pressure and blood flow.

-Renal function tests (RFT):

- 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 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** , where:

➔ U= concentration of any substance in urine.

P= concentration of the same substance in plasma .

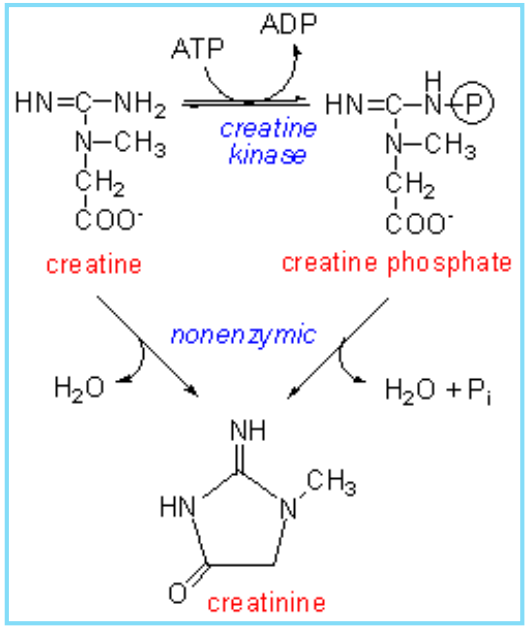
V= volume of urine(ml/min).

Substances clearance used for Measuring GFR

	Inulin Clearance	Creatinine Clearance	Urea Clearance
Source	Non-toxic fructose polymer.	End-product of skeletal muscle creatine metabolism.	End product 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 (exogenous).	Small amount is secreted .	-Partially reabsorbed. -synthesis varies with diet.

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

1. A high meat intake can cause a **temporary increase**.
2. Transient, small increases may occur **after vigorous exercise** .

- Urine Creatinine:

- **Decreased urine creatinine is found in:**

- Advanced renal disease.

- Renal stenosis.

- Hyperthyroidism.

Increased urine creatinine is found in:

- Diabetes mellitus.

- Hypothyroidism.

- Creatinine clearance:

- A measure of the amount of creatinine eliminated (filtered) 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.

- Clinical Implications:

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

a. Impaired kidney function.

b. Shock, dehydration.

c. Hemorrhage.

2. Increased creatinine clearance is found in:

a. Pregnancy.

-Reference Values:

- **Urine creatinine:** 1- 2 g/ 24h
- **(Serum) creatinine:** 0.6–1.2 mg/dL
- **Normal creatinine clearance=** 100-130 ml/min/1.73m2

- Note:

- What 1.73 m2 means?

- Kidney function is proportional to kidney size, which is proportional to body surface area. A of 1.73 m2 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 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 **520 nm.**

-Method:

1- Set up a series of 8 test tube as follows:

Chemical	Standard (3mg/dl) (serum)		Test (serum)		Standard (0.75mg/dl) (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.**

-Resuts:

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

- Calculation:

Patient information: 24h urine volume = 100ml, gender: women, body surface: 1.6m2, DF=100.

1-Serum creatinine =

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

2-Urine creatinine =

(Mean Absorbance of Urine test ÷ Mean Absorbance of Standard) X concentration of Urine standard X DF= …… mg / dl

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

3- Creatinine Clearance :

=U.V/ P

= [$(\boldsymbol{Urinary\ creatinine\ (mg/dl)})$ / $(\boldsymbol{plasmac\ creatinine\ (mg/dl)})$] $\boldsymbol{x\ Urine\ volume(ml/min)}$ = B

B ----------> 1.6 m2 (person surface area)

? ----------> 1.73 m2

-Corrected for surface area= **…… ml/min/1.73 m2**

-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 =100 ml and A "surface area"=1.6 m2 ?

➔ 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 m2, find the creatinine clearance for 1,73 m2 surface area :

=(14.6 ×1.73)÷1.6= 15.8 ml /min/1.73m2

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 /1.73m2

* 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 .**