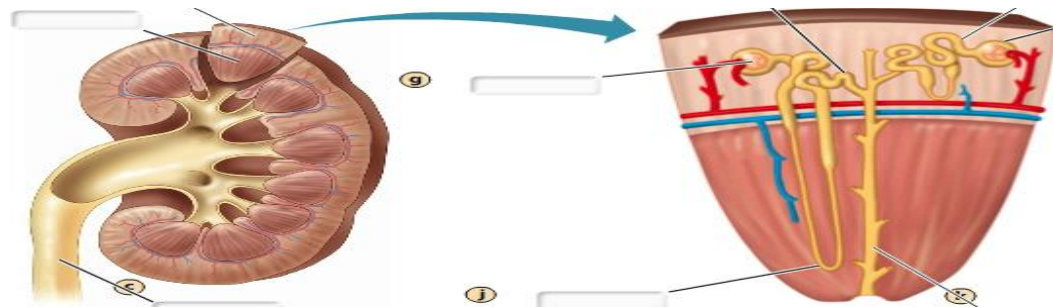
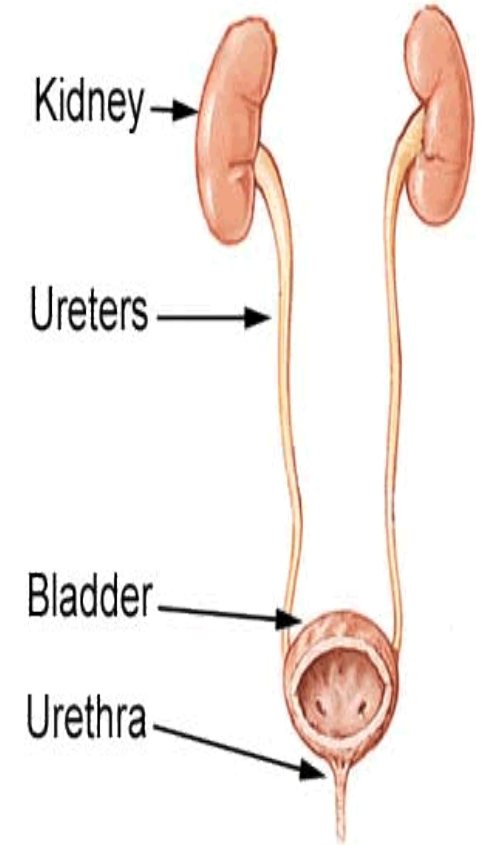


# Physical properties and detection of normal constituents of urine

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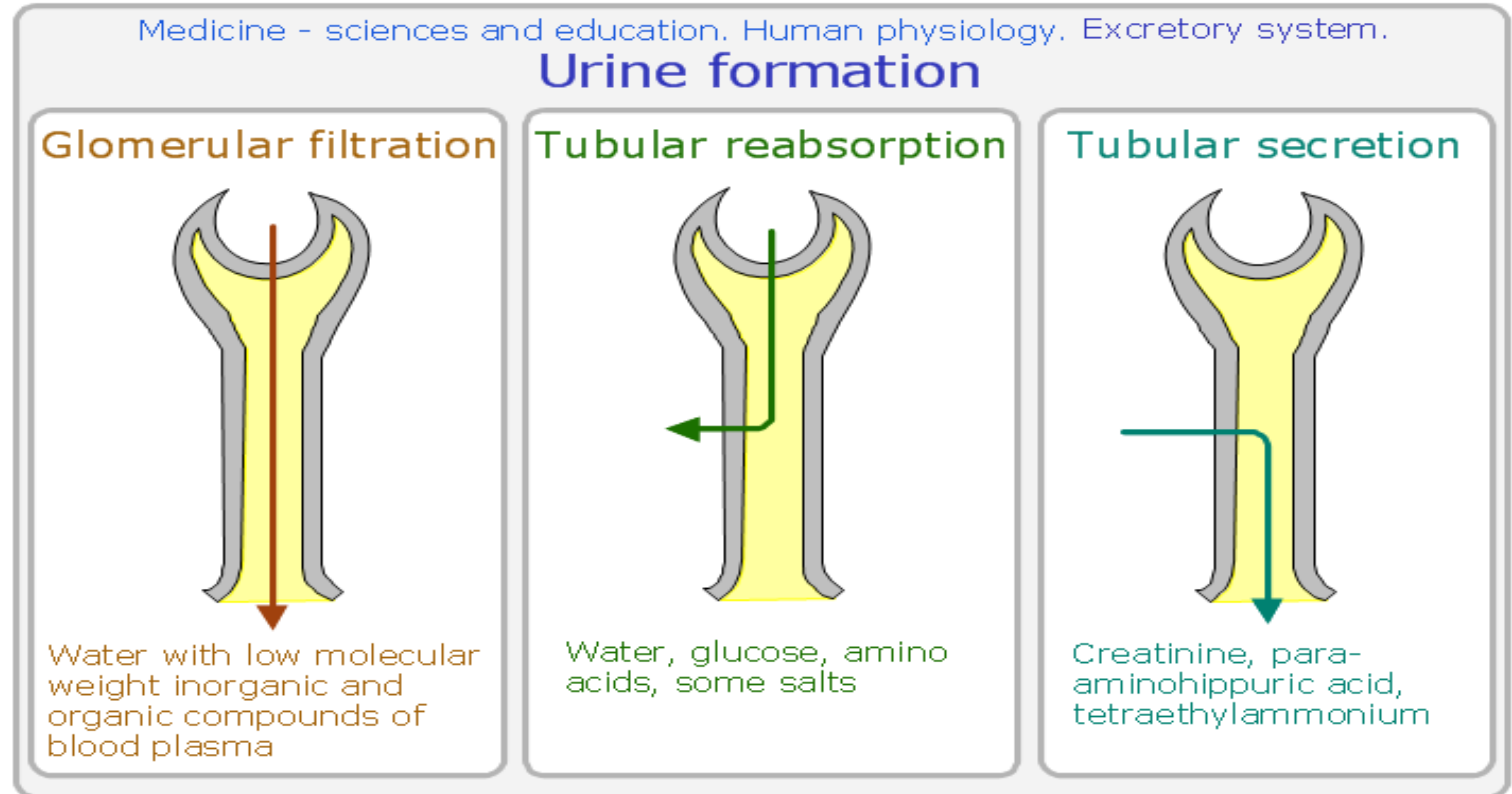
# -Urinary system :

- The **urinary system** works with the **lungs, skin and intestines** to maintain the balance of chemicals and water in the body.
- The kidneys form urine, which passes through the ureters to the bladder for storage prior to excretion.
- Waste products are excreted **selectively**, electrolyte levels are controlled and pH (acid-base balance) is maintained by excretion of hydrogen ions .
- The **composition** of urine reflects **exchange** of substance between the nephron and blood in the renal capillaries.

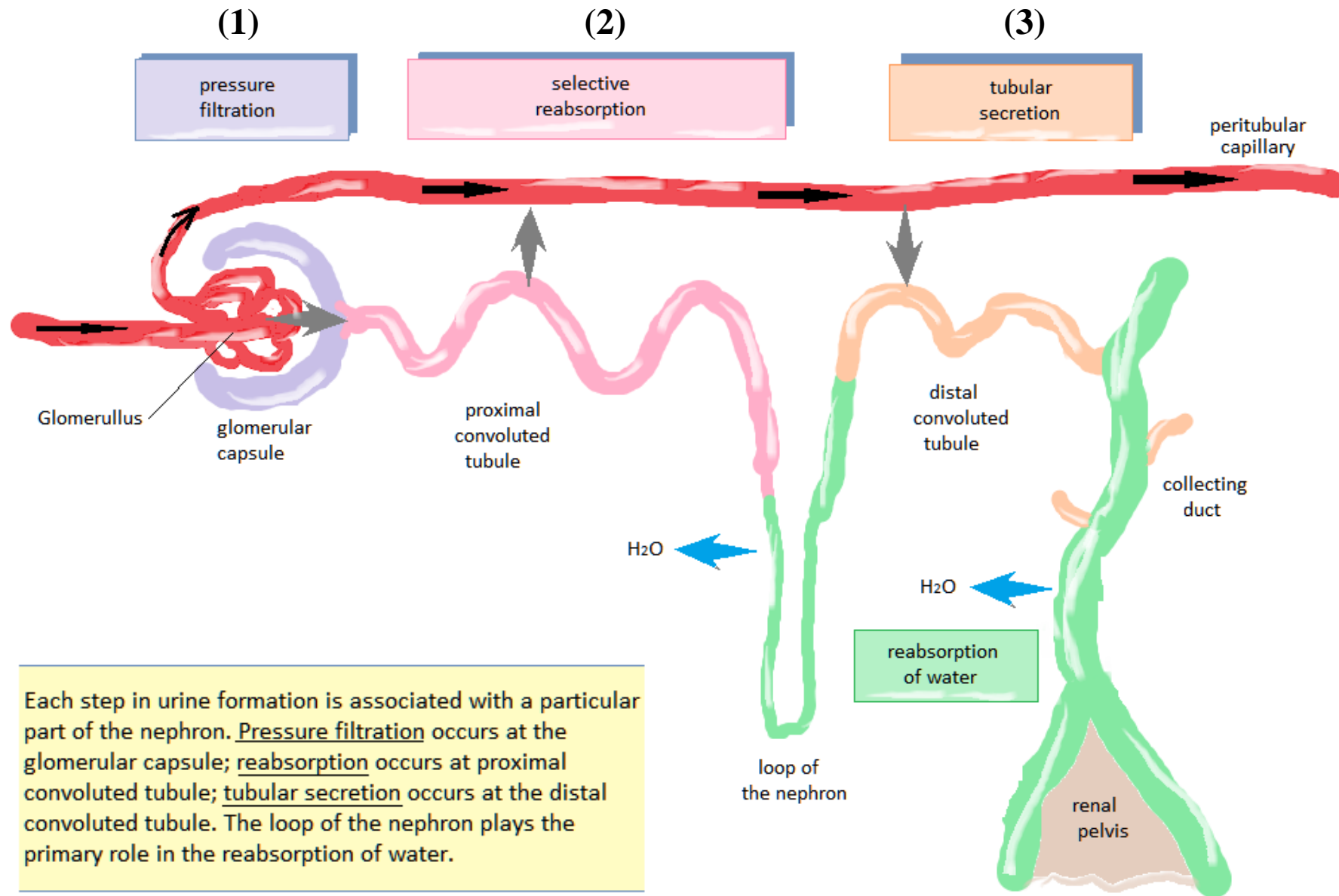


# -Urine Formation:

- There are three processes involved in the formation of urine:
  1. Filtration.
  2. Selective reabsorption.
  3. Secretion.

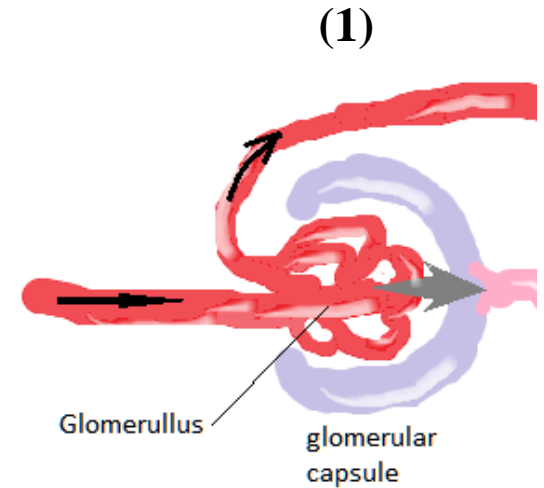


# The three processes of urine formation



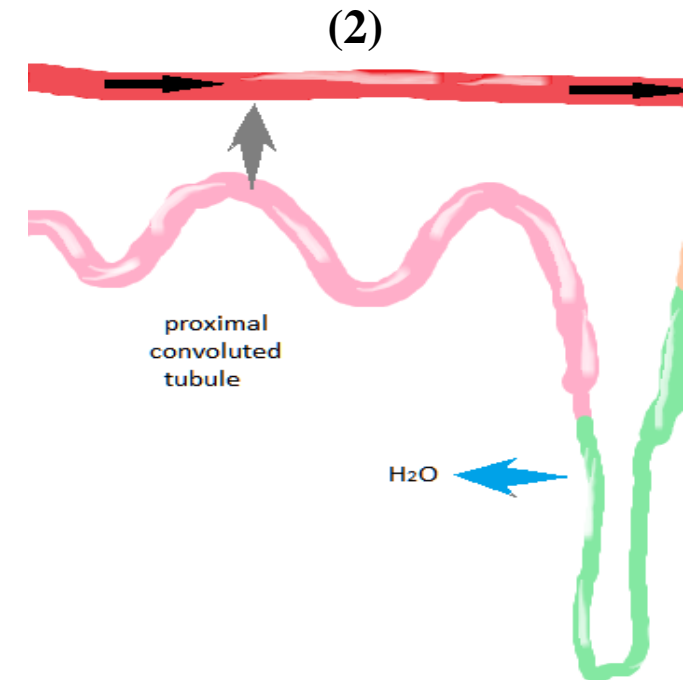
# one: Filtration :

- This takes place through the **semipermeable membrane** of glomerulus and **glomerular capsule (Bowman's capsule)**.
- **Water** and **small molecules** move from the glomerulus to the **inside** of the glomerular capsule.
- Molecules which have molecular weight **more than** 70,000 Dalton can **not** pass the glomerulus.
- Blood cells, plasma proteins and other large molecules are too large to filtrate (not filtrated).
- Inside the glomerular capsule now contains **glomerular filtrate** which is very similar in composition of plasma **except of plasma proteins and blood cells**.
- (non-selective filtration occurs).



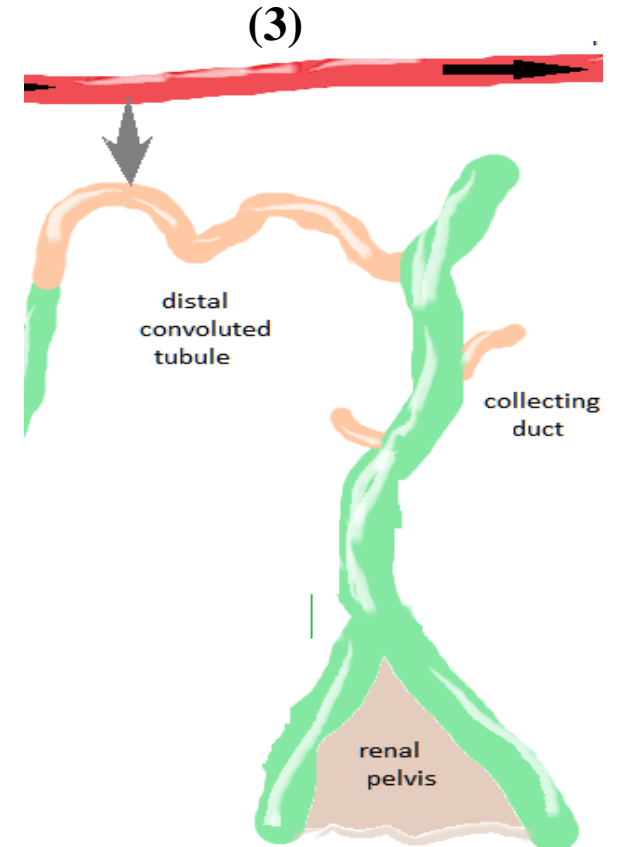
## two: Selective reabsorption:

- Is the process of restoration **water** and **some solutes** from the tubular fluid and returning them to the blood.
- **Reabsorption** is the movement of water and solutes from the tubule **back into the blood**.
- As molecules and ions are passively and actively reabsorbed from the nephron into the blood of the peritubular capillary network.
- Nutrients such as glucose and amino acids **return** to the peritubular capillaries almost exclusively at the proximal convoluted tubule.
- Every substance has a **maximum rate of transport**.



# three: Secretion:

- Is a second way by which substances are removed from blood and added to the tubular fluid.
- Is a process in which the renal tubule extracts chemicals from the capillary blood and secretes them into the tubular fluid.
- Hydrogen ions (H<sup>+</sup>), creatinine, and drugs such as penicillin are some of the substances moved by active transport from blood into the kidney tubule.
- Tubular secretion is now known to occur along the length of the kidney tubule.







- In the end, urine contains :

1-substances that have undergone glomerular filtration (step one) but have **not** been reabsorbed (step two).

2-substances that have undergone tubular secretion (step three).



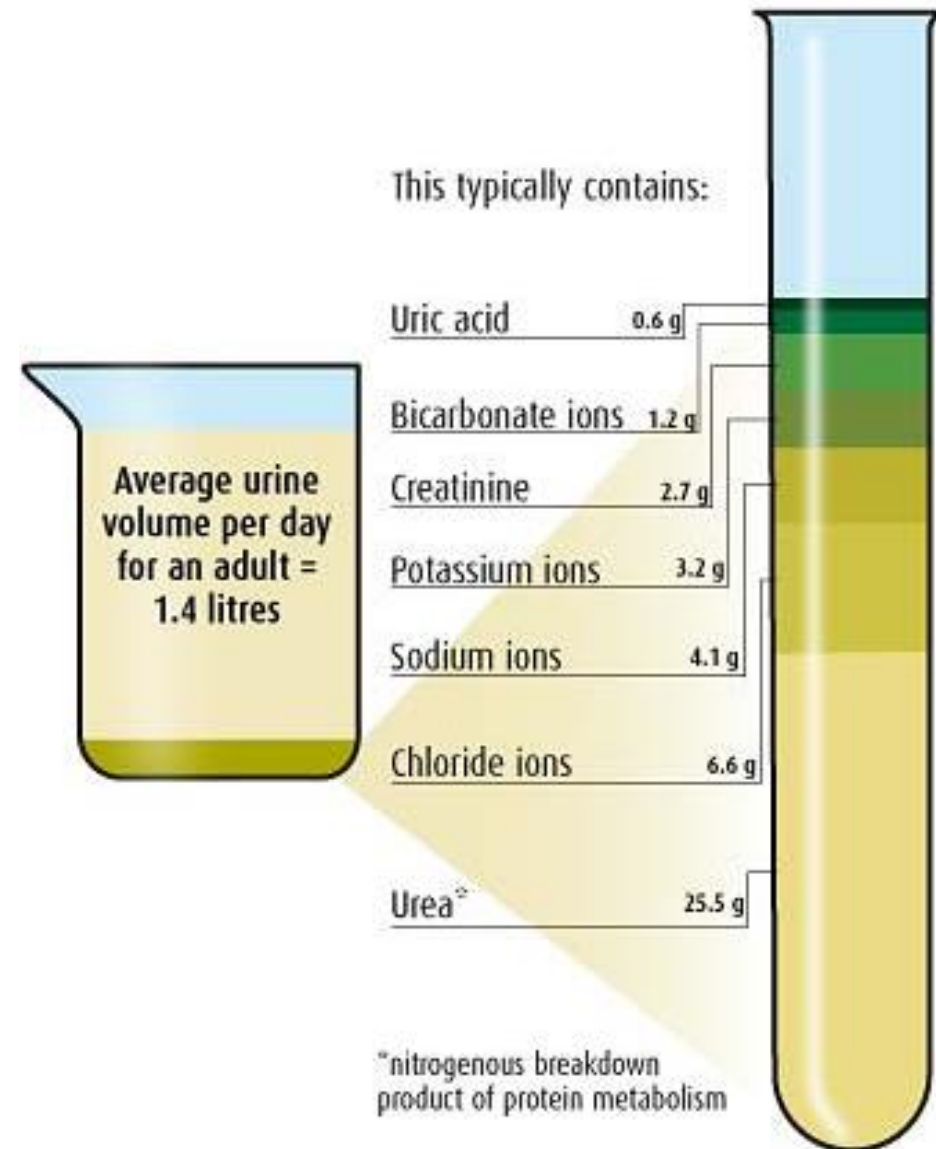
# Glomerular filtrate VS Urine

Constituent	Daily Excretion	
	Glomerular Filtrate	Urine
Water	130,000 ml 	1500 ml
Sodium	20,000 mmol 	150 ml
Albumin	4 g (60 $\mu$ mol) 	0.04 g (6 $\mu$ mol)
Urea	900 mmol 	400 mmol

# -Composition of Normal Urine:

- Water 96%
- Urea 2%
- Uric acid
- Creatinine
- Ammonia
- Sodium
- Potassium
- Chloride
- Phosphate
- Sulphate
- Oxalate

2%



# -Urinalysis:

- Urinalysis (UA) is one of the most frequently ordered tests.
- Two unique characteristics of urine specimens :
  - 1- Urine is readily available and easily collected specimen.
  - 2- Urine contains information about many of the body's major metabolic functions, and this information can be obtained by simple laboratory tests.

## -Laboratory testing for routine urinalysis (types of testing):

- First, the **physical characteristics** of the urine are noted and recorded .
- Second , a series of **chemical tests** is run . A chemically impregnated dipstick can be used for many of these tests.
- Third, the urine sediment is examined under **microscopic** to identify components.

Patient Name: \_\_\_\_\_

Age: \_\_\_\_\_  M  F

Physician's Name: \_\_\_\_\_

Collection Date: \_\_\_\_\_ Test Date: \_\_\_\_\_ Tester's Initials: \_\_\_\_\_

**Physical Characteristics:**

Color:  colorless  yellow  amber  other  orange  green  red

Appearance:  clear  hazy  cloudy  turbid

**Chemical Measurements:** (circle one)

urobilinogen (mg/dL)	normal	2	4	8			
glucose (mg/dL)	neg	50	100	250	500	1000	
ketone (mg/dL)	neg	trace/5	+ /15	++ /40	+++ /80	++++ /160	
bilirubin	neg		+	++	+++		
protein (mg/dL)	neg	trace		+ /30	++ /100	+++ /300	++++ /2000
nitrite	neg	pos (any pink color is considered positive)					
leukocytes	neg	trace	+	++	+++		
blood	neg	trace	moderate	trace	+ /small	++ /mod	+++ /large
		Non-Hemolyzed		Hemolyzed			
pH	5	6	6.5	7	8	9	
specific gravity	1.000	1.005	1.010	1.015	1.020	1.025	1.030

**Microscopic Examination:**

WBC \_\_\_\_\_ /HPF Crystals \_\_\_\_\_ Parasites \_\_\_\_\_

RBC \_\_\_\_\_ /HPF Bacteria \_\_\_\_\_ Spermatozoa \_\_\_\_\_

Casts \_\_\_\_\_ /LPF Yeast \_\_\_\_\_ Artifacts \_\_\_\_\_

Epithelial Cells \_\_\_\_\_ /HPF Other \_\_\_\_\_

Physical characteristics

Chemical tests

Microscopic examination

# Urine dipstick / Urine test strips:

- The test strips consist of a ribbon made of absorbent microfiber cellulose pads attached to it.
- Each pad contains the dried **reagents** needed for a specific test that react with the compounds present in urine producing a characteristic colour.
- The depth of color produced relates to the concentration of the substance in the urine.
- It provides quick **Semi-quantitative determinations** of pH, protein, glucose, ketones, bilirubin, hemoglobin (blood), nitrite, leukocyte , urobilinogen, and specific gravity.
- Color changes then matched to the control chart at the correct time after each stick is dipped into the urine specimen.



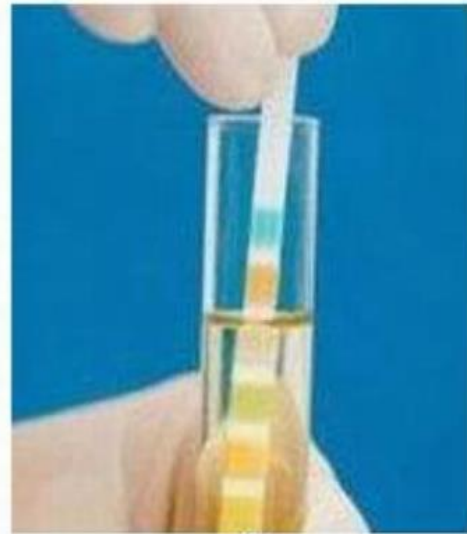
# Urine dipstick / Urine test strips

## How to test your urine(visual read)?



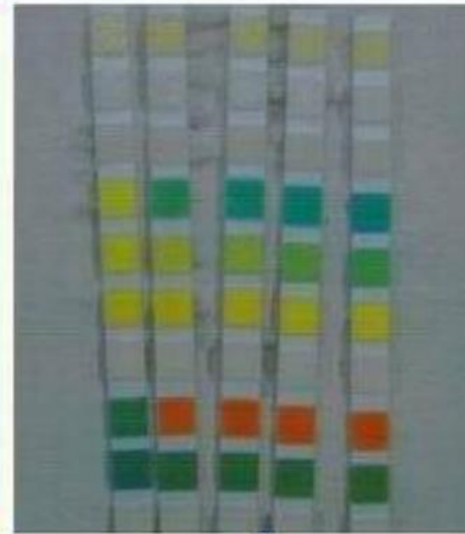
**A**

**Prepare some fresh urine sample.**



**B**

**Dip the dry strip into the urine.**



**C**

**Absorb the excess urine with absorbent paper.**



**D**

**Contrast color chart, close to which color?**

# Simple Examination of the Urine

## Physical Examination

Volume, Specific gravity, Color, Appearance, odor, pH

## Chemical Examination

### Organic

Uric acid, Creatinine

### Inorganic

Chloride, Phosphate, Bicarbonate, Sulphate

# Physical Examinations:

## 1- Volume:

- The daily output of urine on an average diet and normal fluid intake is between **800-2500 ml** with an average of **1500 ml/day**.
- Effected by : 1) Physiological factors. 2) pathological factors.

### Polyurea

- **More than 2500 ml/day.**
- Diabetes mellitus.
- Chronic renal insufficiency.

### Oligurea

- **Below 500 ml /day.**
- In case of deficient intake of water or excessive loss of fluids by other routs like haemorrhage or as diarrhea and vomiting.

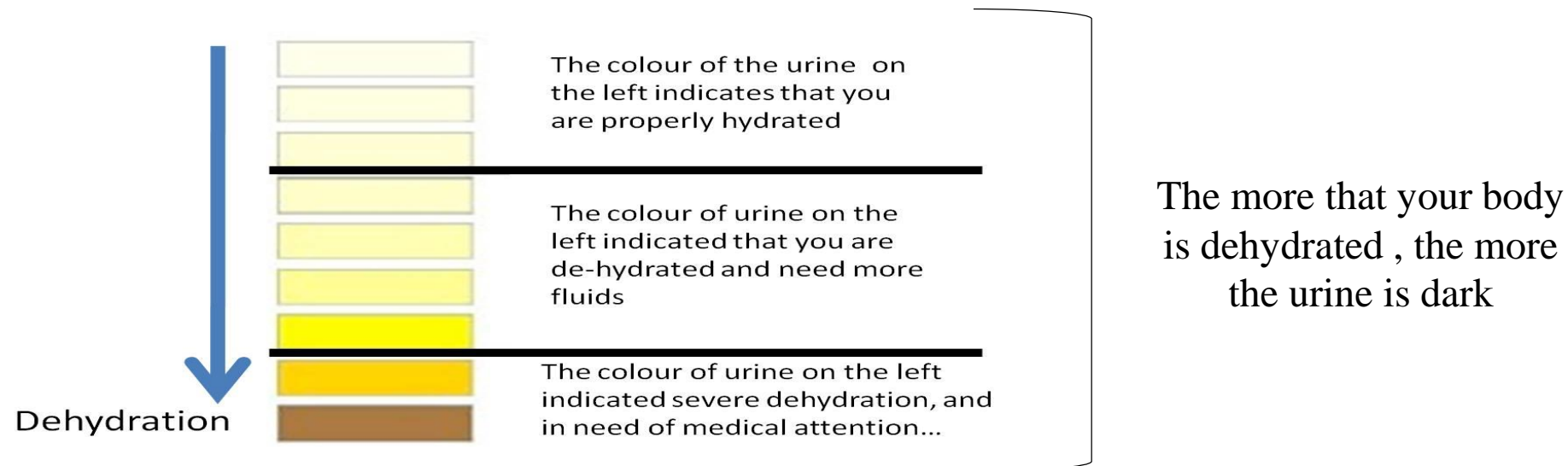
### Anurea

- **Below 100 ml /day.**
- Stones or tumors in the urinary tract can also cause it by creating an obstruction to urinary flow.



## 2- Colour :

- Normally, Urine is **amber** in color due to the presence of urobilin (urochrome).
- **Pale** urine has a **low** specific gravity, a **dark** line has a **high** specific gravity ( a **direct relationship** between the colour and the specific gravity).
- **Coloured urines** occur in certain diseases or metabolic disorders, and after the administration of many drugs.



# 3- Appearance :

- Normal urine is clear.
- **Urine clarity is typically classified as:** clear, mildly cloudy, cloudy, or turbid.  
➔**Note:** cloudy or turbid urine can indicate dehydration, urinary tract infection or presence of RBCs, WBCs, epithelial cells or bacteria.



## 4- Odour:

- Normally Urine smells **aromatic** due to the presence of volatile organic acids.
- The urine of patients with **diabetes mellitus** may have a fruity (acetone) odor **because of ketosis.**
- Urine which is infected with **Gram-negative organisms** often has a distinctive unpleasant smell.
- Certain drugs impart a typical odour.

# 5- pH:

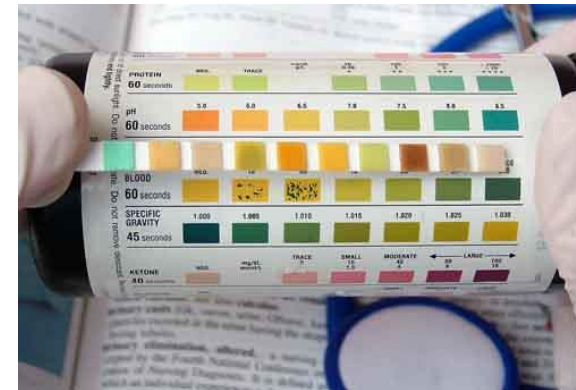
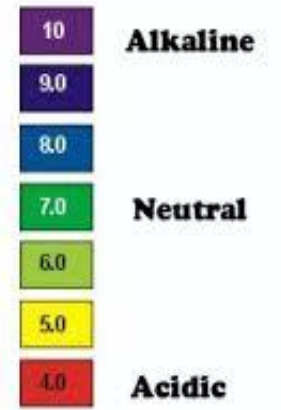
- On a normal mixed diet the urine is **usually acid**, generally varying in pH between **5.5 and 8.0**, with a **mean of 6 in 24 hours**.

## - Acidic Urine :

- Diabetic ketosis, urinary tract infection, diarrheal and starvation.

## - Alkaline Urine:

- A vegetarian diet which causes a tendency to alkalosis.
- It may also be grossly increased by **bacterial infection** of the urinary tract.



# 6- Specific gravity (SG):

- The normal specific gravity (correctly called relative density) of a pooled 24 hour urine sample is between **1.010 and 1.025**.
- There are **direct relation ship** between **concentration** of substance in urine (Concentration of urine) and **SG**.
- ➔ The concentration of urine is highest in the a morning specimen (overnight urine) and is lowest in a specimen passed an hour after much fluid has been taken.

# Practical Part

---

# -Objectives:

1. Simple examination of Urine.
2. To detect some of the normal organic constituents of urine (Qualitative) .
3. To detect some of the normal inorganic ions present in urine (Qualitative) .

- **Note:**

All the examination in 24 hour collection of urine.



# Physical Examinations:

## **method:**

### **1- Volume:**

- Measure the volume of the 24 hour collection of normal urine.

### **2- Odour:**

- State whether it is normal urine like ammonical, or not.

### **3- Colour:**

- Visually examine its colour.

### **4- Appearance:**

- State whether it is clear, cloudy or whether deposits or precipitates are present.

### **5- pH:**

- Record the pH of the sample by test strips.

### **6- Specific gravity:**

- Record the specific gravity of the sample by test strips

# Chemical Examinations:

**Principle:** Each test based on the chemical properties of the substance + test strip.

## 1- Organic:

### A. Uric acid:

- Uric acid is the end product of purine metabolism.

#### -Method:

1. To 2 ml of urine add 1 ml of Benedict reagen.
2. Then heated in a boiling water bath for three minutes .
3. Changes to the **white precipitate** indicates the presence of uric acid.



### B. Creatinine:

#### -Method:

1. To about 5 ml of urine add a few drops of a saturated solution of picric acid.
2. On rendering the solution alkaline with a few drops of 10% sodium hydroxide solution, a deep **red color** or orange due to creatinine picrate appears.
3. On acidification, with 2N HCl, the color changes to **yellow**.





# Chemical Examinations cont':

## 1- Inorganic:

### A. Chloride:

#### -Method:

Add 5 ml of Urine +5 drops of 2N nitric acid+ 2N silver nitrate solution

→ A white precipitate of silver chloride is formed (Silver chloride is precipitated in the presence of nitric acid and silver nitrate).

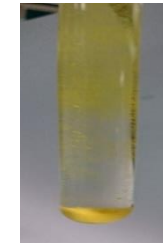


### B. Phosphate:

#### -Method:

Add 5 ml of urine +5ml nitric acid+4 ml of sodium molybdate , then heat in water bath.

→ A yellow crystalline precipitate of ammonium phospho-molybdate appears.



### C. Bicarbonate:

#### -Method:

Add 4 drops of concentrate hydrochloric +5 ml of urine.

→ A slight effervescence occurs due to CO<sub>2</sub> evolution. Test the gas evolved with lime water.

### D. Sulphate:

#### -Method:

To Acidify add 10 ml of urine with 1ml dilute hydrochloric acid + 4 drops of 5% barium chloride solution

→ A white precipitate sulphate is precipitated as of barium sulphate is formed.



### E. Ammonia:

#### -Method:

Add 1 ml of 10% sodium hydroxide solution +5 ml or urine, then heat in water bath.

→ The evolved ammonia may be detected its occur in confirmed by turning the moist red litmus paper to blue.



# Summery:

<b>Physical examination</b>	
The normal constituent of 24 hour urine	
<b>Volume</b>	800-2500 ml with an average of 1500 ml
<b>Color</b>	Amber in color
<b>Appearance</b>	Clear
<b>Odour</b>	Urine like
<b>pH</b>	5.5 - 8.0, with a mean of 6
<b>Specific gravity</b>	1.010 - 1.025
<b>Chemical examination</b>	
<b>Chemical</b>	<b>Positive result</b>
<b>Uric acid</b>	White precipitate
<b>Creatinine</b>	Deep orange color
<b>Chloride</b>	White precipitate
<b>Phosphate</b>	Yellow precipitate
<b>Bicarbonate</b>	CO <sub>2</sub> bubble appeared
<b>Sulphate</b>	White precipitate
<b>Ammonia</b>	Litmus paper turns to blue

Test For	reagent	Color
Creatinine	saturated solution of picric acid in alkaline condition	Red-orange color
Uric acid	Benedict reagent after heating	White precipitate
Chloride	nitric acid and silver nitrate	White precipitate
Phosphate	concentrated nitric acid and saturated ammonium molybdate	Yellow precipitate
<b>Bicarbonate</b>	concentrate hydrochloric acid	gaseous carbon dioxide.
<b>Sulphate</b>	dilute hydrochloric acid + 1 ml drops of 5% barium chloride solution	white precipitate
<b>Ammonia</b>	sodium hydroxide	<b>ammonia gas</b> with <b>sodium hydroxide</b> . This is an alkaline <b>gas</b> . It turns red <b>litmus</b> paper blue

## Questions:

- Albumin is not normally detected in any appreciable quantity in urine. Why not?
  - What 2 symptoms are seen in a patient developing diabetes mellitus ?
  - What is the simplest and quickest method of detecting the presence of blood in urine?
-