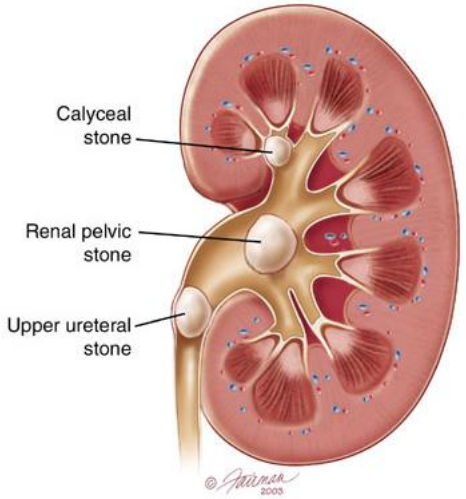
Identification and Qualitative analysis of

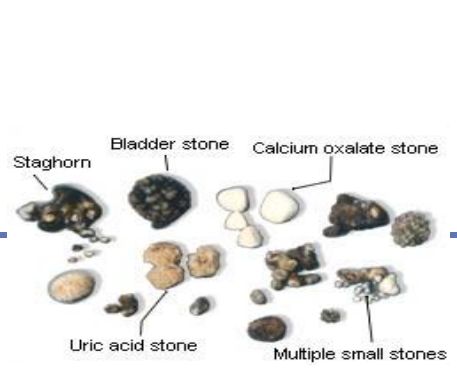
Renal Calculi

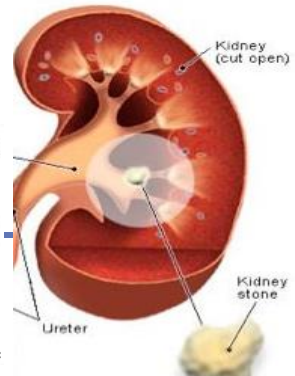
BCH 472



Renal Calculi:

- Kidney stones , renal calculi or renal lithiasis are small, hard deposits that form inside your kidneys.
- The stones are **made of mineral and acid salts**.
- Often, stones form when the urine becomes **concentrated**, allowing minerals to crystallize and stick together.
- A common cause of blood in the urine and pain in the abdomen, flank, or groin.
- Kidney stones have many causes and can affect any part of your urinary tract-kidneys, ureters, bladder, and urethra.





Pathogenesis of renal stones :

- There are **two basic aspects** in the pathogenesis of renal stones:
- **Increased urinary excretion of stone forming elements** like calcium, phosphorus, uric acid, oxalate, and cystine
- **Low fluid intake** (decreased urine volume). A low fluid intake results in the production of **concentrated urine**, causing supersaturation and crystallisation of stone-forming compounds.
- In addition, **low urine flow rates** favor crystal deposition on the urothelium.
- **Physio-chemical changes** which influence stone formation like: pH of urine, stone matrix, and protective substances in the urine.

Investigation of Renal Calculi

1- Urine analysis and Urine culture

It may show crystals, red blood cells, and/or pus cells in urine

2- Stone analysis

- It is important to know the **chemical composition** of urinary stone to understand the cause and plan appropriate treatment.
- Chemical analysis of stones is a simple test but is not an accurate method.

  (will be done in today's lab). Better method is crystallography.

3- Biochemical investigations

- Serum calcium, phosphorus, uric acid, and renal function tests.
- 24-hour urine for calcium, phosphorus, uric acid, oxalate, citrate, and cystine.
- Investigations for special clinical situations like hyperparathyroidism, gout, renal tubular acidosis should also be included.

Types of calculi

Stone composition	Cause	Note
Calcium stone	are the most common type of kidney stone and occur in two major forms: calcium oxalate and calcium phosphate. • **Calcium oxalate** stones are more common. • Calcium oxalate stone formation may be caused by high calcium "Hypercalciuria" and high oxalate Excretion. • **Calcium phosphate** stones are caused by the combination of high urine calcium and **alkaline** urine.	***Hypercalciuria caused by:** Hyperparathyrodisim. Vitamin D toxicity. *Excess vitamin C is converted into oxalate and excreted in the gut and urine. And some food eg.spinach.
Uric acid stones	• form when the urine is persistently **acidic**. • If uric acid becomes concentrated in the urine, it can settle and form a stone.	*Excessive urinary uric acid: a diet rich in **purines** found in protein such as meats may increase uric acid in urine, Gout.

Types of calculi

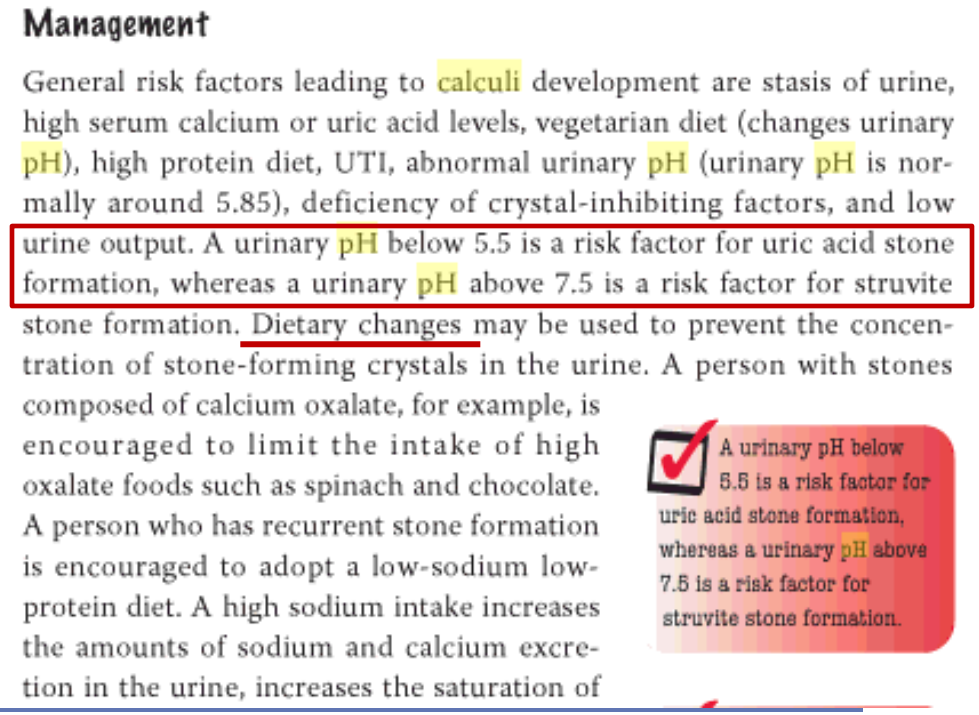
Stone composition	Cause	Note
Carbonate apatite (calcium carbonate and calcium phosphate)	These stones develop as a consequence of recurrent or chronic urinary tract **infections** caused by urease producing bacteria.	Some urinary bacteria can split the urea in urine to form ammonium and also to make urine **less acidic**.
Struvite (magnesium ammonium phosphate)		
Cystine stone	develop in patients with **cystinuria**.	caused by **mutations** in the genes, encode for two parts of a **transporter protein** that is made primarily in the kidneys. These defects prevent proper reabsorption of amino acids.

TABLE 33-2 Composition, Contributing Factors, and Treatment of Kidney Stones

TYPE OF STONE	CONTRIBUTING FACTORS	TREATMENT
Calcium (oxalate and phosphate)	Hypercalcemia and hypercalciuria Immobilization Hyperparathyroidism Vitamin D intoxication Diffuse bone disease Milk-alkali syndrome Renal tubular acidosis Hyperoxaluria Intestinal bypass surgery	Treatment of underlying conditions Increased fluid intake Thiazide diuretics Dietary restriction of foods high in oxalate
Magnesium ammonium phosphate (struvite)	Urea-splitting urinary tract infections	Treatment of urinary tract infection Acidification of the urine Increased fluid intake
Uric acid (urate)	Formed in acid urine with pH of approximately 5.5 Gout High-purine diet	Increased fluid intake Allopurinol for hyperuricosuria Alkalinization of urine
Cystine	Cystinuria (inherited disorder of amino acid-metabolism)	Increased fluid intake Alkalinization of urine

Treatment

Includes relief of pain, **hydration,** dietary changes and Alkalization or acidification of urine (depend on the type of stone). The majority of stones pass spontaneously within 48 hours. However, some stones may not. If a stone does not pass, **urologic intervention** may be needed.



Management

General risk factors leading to calculi development are stasis of urine, high serum calcium or uric acid levels, vegetarian diet (changes urinary pH), high protein diet, UTI, abnormal urinary pH (urinary pH is normally around 5.85), deficiency of crystal-inhibiting factors, and low urine output. A urinary pH below 5.5 is a risk factor for uric acid stone formation, whereas a urinary pH above 7.5 is a risk factor for struvite stone formation. Dietary changes may be used to prevent the concentration of stone-forming crystals in the urine. A person with stones composed of calcium oxalate, for example, is encouraged to limit the intake of high oxalate foods such as spinach and chocolate. A person who has recurrent stone formation is encouraged to adopt a low-sodium low-protein diet. A high sodium intake increases the amounts of sodium and calcium excretion in the urine, increases the saturation of

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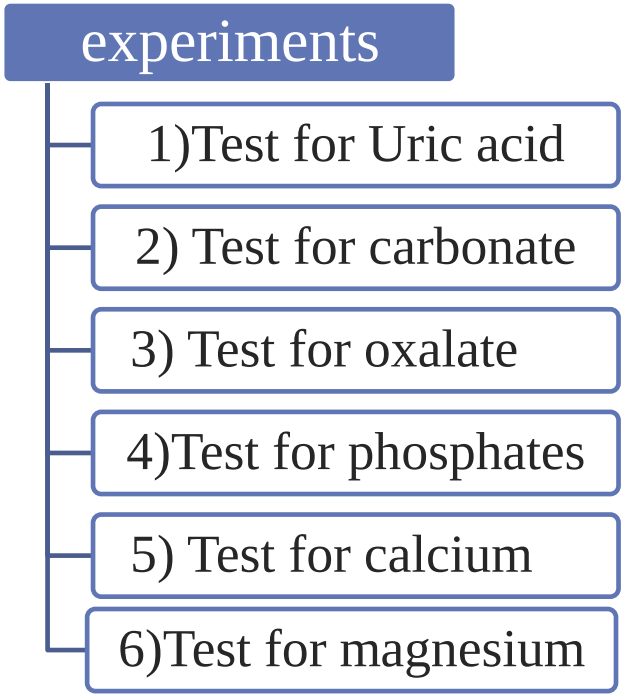
Practical Part



Objective:

- Identification and Qualitative analysis of Renal Calculi, to find out the presence and composition of stones.

1)Test for Uric acid

Principle: Uric acid undergoes oxidation when treated with **HNO3.**

Method:

1-Put a small amount of the sample1.

2-Add 5-7 drops of concentrated nitric acid.

3-Heat in a water bath.

yellow to orange color on the inner surface of the test tube.

2) Test for carbonate

Principle:

2 HCl + CaCO3 --> CaCl2 + H2O + CO2

Method:

1-Add 0.5 ml of conc. hydrochloric acid to small portion of sample2.

Gas bubbles will indicate the presence of carbonate.

3) Test for oxalate

Principle:

In sulfuric acid solution, oxalate combines with hydrogen to form oxalic acid.

Potassium permanganate reacts with oxalate ions to produce carbon dioxide and water in an acidic solution, and the permanganate ion is reduced to manganese (II) as follows:

$5C_2O_4^{2-} + 2MnO_4^- + 16H^+ \rightarrow 10CO_2 + 8H_2O + 2Mn^{2+}$

The permanganate ion is intensely **purple**, whereas the manganese (II) ion is nearly **colorless**.

Method:

1-Heat a part of sample3 with 2 ml dilutes sulphuric acid (2M H_2SO_4) for 1 min.

2-Add 2 drops (one by one) of, potassium permanganate ($KMnO_4$) solution and Mix

The **decolonization** and **evolution of bubbles** will confirm the presence of oxalate.

4)Test for phosphates

Principle:

Phosphate ions react with ammonium molybdateto produce a characteristic yellow precipitate, ammonium phosphomolybdate.

Method:

1-Dissolve a little of the sample 4 in about 1.5 ml of concentrated nitric acid HNO3.

2-Add an equal volume (1.5 ml) of ammonium molybdate solution.

3-Heat to boiling.

(If phosphates are present, a **yellow precipitate** of ammonium phosphomolybdateis obtained).

5) Test for calcium

Principle:

calcium is precipitated as calcium oxalate using ammonium oxalate

Method:

1-Dissolve small amount of the sample 5 by heating with 2 ml dilute hydrochloric acid (2M HCL)

2-Add 1 ml ammonium oxalate.

A **white precipitate** of calcium oxalate shows the presence of calcium).

6) Test for magnesium

Principle:

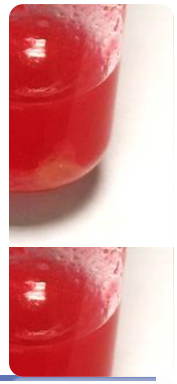
When magnesium hydroxide precipitated in the presence of titan yellow by sodium hydroxide the yellow color of reagent changes to **red or orange-red**.

$Mg^{2+} + 2OH^- \longrightarrow Mg(OH)_2$, titan yellow form a red absorption complex when magnesium hydroxide is precipitated in its presence.

Method:

1-add 1 ml of titan to small amount of sample 6.

2- add 1ml of sodium hydroxide until strongly alkaline. A **red or orange-red color** indicates the presence of magnesium.



Results:

Components	Result
Uric acid	
carbonate	
oxalate	
phosphates	
calcium	
magnesium	

Discussion:

Comment in each results you obtained and mention whether the sample contains these component or not? And the disease that cause each type of stone.

Questions :

How change in urine pH can influence the type of stone formed?

Why Hyperparathyrodisim increases chance of calcium stone formation?