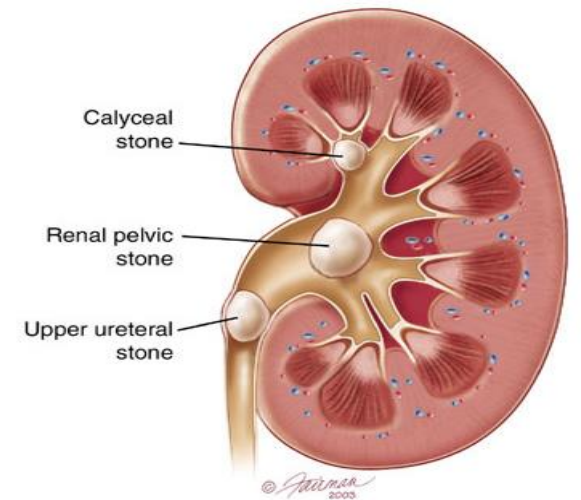


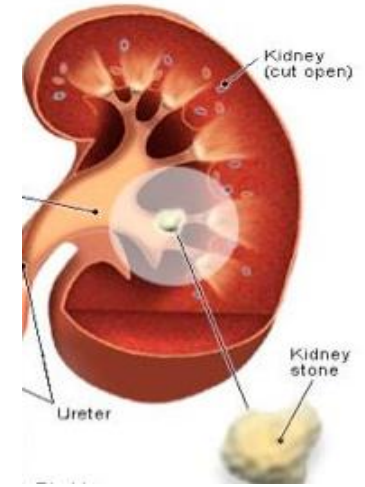
Identification and Qualitative analysis of Renal Calculi

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.
- It is 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).

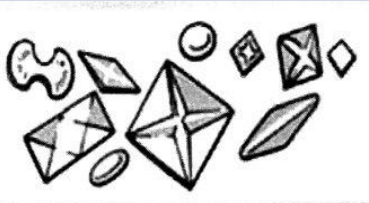



Pathogenesis of renal stones :

- 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** 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, and protective substances in the urine.

Types of calculi

Stone composition	Contributing factors	Note
<p>1) Calcium stone</p> <p>The most common type of kidney stone.</p> <p>Occurs in two major forms: calcium oxalate and calcium phosphate.</p>	<ul style="list-style-type: none"> • Calcium oxalate crystals are usually found in acidic urine • It may be caused <i>by high calcium</i> “Hypercalciuria” and <i>high oxalate</i> Excretion ‘Hyperoxaluria’. <div style="border: 1px solid black; padding: 10px; margin: 10px 0;"> <p>*Hypercalciuria is caused by:</p> <ul style="list-style-type: none"> - Hyperparathyroidism. - Vitamin D toxicity. <p>*Hyperoxaluria is caused by:</p> <ul style="list-style-type: none"> - High-oxalate foods e.g. spinach, strawberries and beets. - Large doses of vitamin C, since its excess amounts are excreted by the body in the oxalate form, and this may lead to more stones. </div> <ul style="list-style-type: none"> • Calcium phosphate stones are caused by the combination of high urine calcium and alkaline urine. (because phosphate level increase in alkaline urine). 	<ul style="list-style-type: none"> • <i>Calcium oxalate stones are more common</i> • Their crystals may occur as either bihydrated or monohydrated calcium oxalate. • Calcium oxalate bihydrate crystals appear as envelope form. • Calcium oxalate monohydrate crystals are colorless and can appear as ovoids, biconcave disks, rods or dumbbells 

Stone composition	Contributing factors	Note
<p>2) Uric acid stones (Urate)</p>	<ul style="list-style-type: none"> • Form in acidic urine with pH around 5. • Gout. • High purine diet. • Excessive urinary uric acid 	<ul style="list-style-type: none"> • Can treated by: <ul style="list-style-type: none"> - Increase fluid intake. - Alkalinisation of the urine. • Their crystals can look like barrels, rosettes, rhomboids, needles or hexagonal plates. 
<p>3) Struvite (magnesium ammonium phosphate)</p>	<ul style="list-style-type: none"> • These stones develop as a consequence of recurrent or chronic urinary tract infections caused by <u>urease producing bacteria</u>. • They can <u>split the urea in urine to form ammonium</u> and therefore make the urine less acidic (alkaline). 	<ul style="list-style-type: none"> • Can treated by: <ul style="list-style-type: none"> - Increase fluid intake. - Acidification of the urine
<p>4) Cysteine stone</p>	<p>-Develop in patients with cystinuria.</p> <p>-Caused by mutations in the genes that encode for two parts of a transporter protein that is made primarily in the kidneys <u>preventing proper reabsorption of amino acids</u>, and therefore make the urine more acidic.</p>	<ul style="list-style-type: none"> • Less common. • Can treated by: <ul style="list-style-type: none"> - Increase fluid intake. - Alkalinisation of the urine.

Investigation of Renal Calculi



1- Urine analysis and and Urine culture :

- High specific gravity, Low or high urine pH
- Microscopic or gross hematuria, white blood cells , nitrite or pus are often seen in the urine.
- Crystalluria can help in defining stone type under microscope (e.g. hexagonal cystine crystal, coffin lid phosphate crystal, rhomboidal uric acid crystal).
- Bacteriuria must be further evaluated with urine culture.

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. **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.

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

experiments

1) Test for Uric acid

2) Test for carbonate

3) Test for oxalate

4) Test for phosphates

5) Test for calcium

6) Test for magnesium

Objective:

- Identification and Qualitative analysis of Renal Calculi, to find out the presence and composition of stones.
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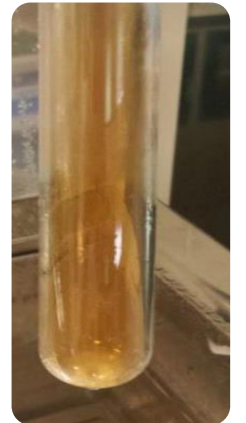
1) Test for Uric acid

Principle: Uric acid undergoes oxidation when treated with HNO_3 .

Method:

- 1-Put a small amount of the sample.
- 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:



Method:

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

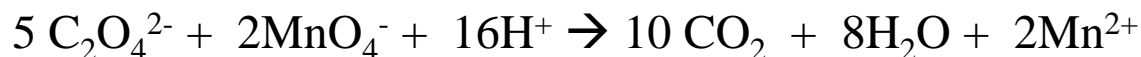
Gas bubbles will indicate the presence of carbonate.

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:



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

Method:

1-Heat a part of sample 3 with 2 ml dilutes sulphuric acid (2M H₂SO₄) for 1 min.

2-Add 2 drops (one by one) of, potassium permanganate (KMnO₄) solution and Mix

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

4) Test for phosphates

Principle:

Phosphate ions react with ammonium molybdate 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 HNO_3 .
- 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 phosphomolybdate 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**.

$\text{Mg}^{2+} + 2\text{OH}^- \rightarrow \text{Mg}(\text{OH})_2$, 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.

