Identification and qualitative Analysis of Renal Calculi
Renal Calculi:

- Kidney stones, renal calculi or renal lithiasis (stone formation) are small, hard deposits that form inside your kidneys.

- The stones are made of mineral and acid salts.

- 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:

1. Increased urinary excretion of stone forming elements: like calcium, phosphorus, uric acid, oxalate, and cysteine.

2. Low fluid intake: a low fluid intake results in the production of concentrated urine, causing supersaturation and crystallisation of stone-forming compounds.

- Physio-chemical changes which influence stone formation like: pH of urine, stone matrix, and protective substances in the urine.
- Risk factors:

1. **Low fluid intake:**
   - The single most important determinant of stone formation is low fluid intake.
   - A low fluid intake results in the production of concentrated urine.

2. **High Salt diet.**

3. **Repeating, or recurrent, urinary tract infections.**

4. **Blockage of your urinary tract.**
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:
- 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 and gout should also be included.
• **The main objectives in investigation are to find out:**

1. The composition of stones.
2. Cause of stone formation.
5. Evidence of possible urinary infection.
-Types of Calculi:

• **There are four basic types of kidney stones:**
  1. Calcium stones (calcium oxalate and calcium phosphate).
  2. Uric acid stones.

• Most kidney stones (70% to 80%) are calcium stones – calcium oxalate, calcium phosphate, or a combination of the two materials.
## Types of Calculi:

<table>
<thead>
<tr>
<th>Stone type and composition</th>
<th>Contributing factors</th>
<th>Notes</th>
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<tbody>
<tr>
<td><strong>Calcium stones</strong>&lt;br&gt;- calcium oxalate&lt;br&gt;- calcium phosphate</td>
<td>• Hyperparathyroidism.&lt;br&gt;• Hypercalcemia and Hypercalciuria.&lt;br&gt;• Hyperoxaluria. (some food eg. spinach, strawberries and large doses of Vitamin C may increase the amount of oxalate in your urine).&lt;br&gt;• Vitamin D toxicity.</td>
<td>• Calcium oxalate stones are more common.&lt;br&gt;• Calcium phosphate stones are caused by the combination of high urine calcium and alkaline urine (because phosphate level increase in alkaline urine ).&lt;br&gt;• <strong>Carbonate apatite (calcium carbonate and calcium phosphate)</strong> is one kind of calcium phosphate stone.</td>
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<td><strong>Uric acid stones</strong>&lt;br&gt;(Urate)</td>
<td>• Form in acid urine with pH around 5.&lt;br&gt;• Gout.&lt;br&gt;• High purine diet.&lt;br&gt;• Excessive urinary uric acid.</td>
<td>• Can treated by:&lt;br&gt;- Increase fluid intake.&lt;br&gt;- Alkalinization of the urine.</td>
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<td><strong>Struvite stones</strong>&lt;br&gt;(magnesium ammonium phosphate stones)</td>
<td>• Urea-splitting urinary tract infection UTIs&lt;br&gt;(Some urinary bacteria can split the urea in urine to form ammonium and also to make urine less acidic).</td>
<td>• They can also be called <strong>infection stones</strong> if they occur with kidney or urinary tract infections (UTIs).&lt;br&gt;• Can treated by:&lt;br&gt;- Increase fluid intake.&lt;br&gt;- Acidification of the urine</td>
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<td><strong>Cystine stones</strong></td>
<td>• Develop in patients with cystinuria.</td>
<td>• Less common.&lt;br&gt;• Can treated by:&lt;br&gt;- Increase fluid intake.&lt;br&gt;- Alkalinization of the urine.</td>
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Practical Part
- **Objective:**
  
  - Identification and qualitative analysis of kidney stones.

- **General Principle:**
  
  - Each test based on the chemical properties of the stone-forming substance.
- 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.
(1) Test for Uric acid:

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

• **Method:**
  1. Put a small amount of the sample.
  2. Add 5-7 drops of concentrated nitric acid (Carefully).
  3. Heating in a water bath.

  ➔ (the positive result is yellow to orange color on the inner surface of the test tube).
(2) Test for carbonate:

• **Principle:**
  
  \[ 2 \text{HCl} + \text{CaCO}_3 \rightarrow \text{CaCl}_2 + \text{H}_2\text{O} + \text{CO}_2 \]

• **Method:**
  
  1. Add 0.5 ml con. hydrochloric acid to a small portion of the sample (Carefully).

  ➔ *(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.

• **Method:**
  1- Heat a part of the sample 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 decolonization and evolution of bubbles will confirm the presence of oxalate).
(4) Test for phosphates:

• **Principle:**
Phosphate ions react with ammonium molybdate to produce a characteristic yellow precipitate, ammonium phosphomolybdate.

• **Method:**
1- Dissolve a little of the sample 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 phosphomolybdate is obtained)
(5) Test for calcium:

- **Principle:**
  calcium is precipitated as calcium oxalate using ammonium oxalate.

- **Method:**
  1- Dissolve small amount of the sample 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:**
The combination between titan yellow and magnesium hydroxide to produce an orange colour.

• **Method:**
1- On a few amount of magnesium, add 1ml of titan followed by 1 ml potassium hydroxide (to be strongly alkaline).

⇒ (An orange to red color indicates the presence of magnesium).
-Results:

<table>
<thead>
<tr>
<th>Component</th>
<th>Observation</th>
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</thead>
<tbody>
<tr>
<td>Uric acid</td>
<td></td>
</tr>
<tr>
<td>Carbonate</td>
<td></td>
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<tr>
<td>Oxalate</td>
<td></td>
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<tr>
<td>Phosphate</td>
<td></td>
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<tr>
<td>Calcium</td>
<td></td>
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<tr>
<td>Magnesium</td>
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</table>

-Discussion:

• Comment in each results you obtained and mention whether the sample contains these component or not?
• What type of stone can be formed by each substance.
• What the disease that cause each type of stone.