TESTS FOR ABNORMAL CONSTITUENTS IN URINE

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- Under pathological conditions urine excreted by patient shows the presence of abnormal constituents along with normal constituents.
- Abnormal constituents of urine are sugar, proteins, blood, bile salts, bile pigments and ketone bodies.

A. Physical Characteristics

- **1. Volume :**
- a. **Polyuria**: Volume more than 3000 ml / 24 hours
- It is observed in Diabetes mellitus, Diabetes insipidus, Addison's disease, Chronic progressive renal failure, excess water intake, intake of diuretics like caffeine, alcohol etc.
- b. **Oliguria**: Volume less than 400 ml / 24 hours.
- It is observed in fluid deprivation, excess fluid loss as in hemorrhage and neurogenic shock, dehydration, acute glomerulonephritis, obstruction in the urinary tract, disease of heart and lungs & strenuous muscular exercise.

- c. **Anuria**: Less than 150ml / 24hrs Complete absence of urine output. It is observed in shock and renal failure.

- **2. Colour:-**
- The colour of urine is variable in following disease conditions as given following table

Sr. No	**Colour**	**possible causes/ disorder**
1	Colour less	Fatty disease, diabetes mellitus, Polyuria
2	Yellowish brown	Bile pigment, fever
3	Reddish brown	Hemoglobin in urine, hemorrhage, menstrual contamination
4	Milky	Presence of Fat
5	Dark Yellow	Fever
6	Dark green	typhoid and cholera
7	Black	Due to Melanin (Melanoma) or Homogentisic acid in Alkaptonuria

- **3. Odour:-** Normal urine has faint aromatic odour. On standing it has ammoniacal odour due to bacterial contamination. Odour of urine is variable in certain diseased condition.

Sr. No	**Odour**	**diseases**
1	Fruity odour	ketosis
2	Cabbage type odour	methionine Malabsorption
3	Maple sugar odour	maple sugar urine disease(MSUD)
4	Mousy	phenylketonuria
5	Rancid odour	tyrosine
6	Foul	Urinary Tract Infection, Vaginitis

Maple Syrup Urine Disease (MSUD)

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- **4. Reaction or pH:** Normal urine is slightly acidic in reaction.
- a) The urine is **acidic** in reaction in following disease conditions:
- 1. Diabetes mellitus
- 2. Ketoacidosis
- 3. Prolonged starvation
- 4. Febrile illness in children
- 5. Strenuous exercise

- b) The urine **alkaline** in reaction in following diseased condition:
- 1. Severe vomiting
- 2. Diarrhea
- 3. Hyper ventilation
- 4. Prolonged use of diuretic drugs.

- **5. Appearance** :- Normal urine is clear and transparent. It may be cloudy in appearance due to presence of large amount of phosphate, carbonate, ureate, WBC, RBC , oxalate, pus tissues and bacteria.

- **6. Sedimentation**: Normal urine does not contain any sediment, whereas sediment may be observe in certain disease conditions. They are of two type.
- i) Organic sediment- Ureate, Uric acid, oxalate
- ii) Inorganic sediment- Phosphate, Carbonate, Calcium
- Sediment may also be due to pus mucin, RBC, microorganism & excess of nucleoprotein.

- **7. Specific gravity**:- It depends upon the concentration of various solutes.
- Specific gravity usually lies between 1.015-1.025 (Specific gravity of water is taken as 1.000) Specific gravity is increased in Excessive sweating, acute nephritis albuminuria and all the cases of oliguria.
- Specific gravity is decreased in diabetes insipidus, chronic nephritis, and all cases of Polyuria.

B. Chemical Examination

1. Test for Reducing Sugar: (Glucose)

Test	Observation	Inference
1. Benedict's test : Take 5ml Benedict's reagent in test tube then add 8drops of urine sample and Boil it for 1-2 min and observe.	Green or yellowish green or orange or red precipitate obtained	Reducing sugar confirmed Therefore urine contains reducing sugar

Colour of the precipitate	Degree of Glycosuria	Approx. Glucose Conc. (gm %)
Green precipitate	+	0.5%
Yellow precipitate	+ +	1.0%
Orange precipitate	+ ++	1.5%
Brick Red precipitate	+ ++ +	2.0 or more

- **Clinical Interpretation:**
- Presence of glucose in urine is called as glycosuria. Colour of the precipitate indicates severity of glycosuria as follows:
- Presence of glucose in urine is found in,
- a. Diabetes mellitus.
- b. Hyperadrenalism.
- c. Renal glycosuria

2. TEST FOR PROTEIN:-

Test	Observation	Inference
2. **Heat coagulation test:** Fill 3/4th of test tube with urine sample solution. Heat it at the top and observe. Then Add 1-2 drop of acetic acid to it and again observe.	White Coagulum or turbidity is obtained which after the addition of acetic acid gets intensified.	Albumin is confirmed. Therefore urine contains albumin

- **Clinical Interpretation**
- Presence of detectable amount of protein in urine is known as proteinuria. It is of two types, **Physiological proteinuria** : Less than 0.5 gm%. It occurs due to sever exercise, pregnancy, high protein diet etc.
- **Pathological proteinuria:** It occurs due to kidney damage (Glomerulonephritis and Nephrotic syndrome). In some patients suffering from multiple disease such as leukemia lymphosarcoma. Urine & blood contain specific type of protein called Bence Jones protein. They are globulin & posses peculate solubility property i.e. they coagulated at 40-60 ^{o}C & go in to the solution at 80 ^{o}C and reappear after cooling

3. TEST FOR Ketone bodies :-

- **Principle:** Acetone in urine after saturation with ammonium sulphate crystals dissociate into acetate ions which combines with sodium nitropursside in alkaline medium to from permagnate colour sodium nitropursside acetate ion complex.

3. TEST FOR Ketone bodies :-

Test	Observation	Inference
3. Rothera's test : Saturate 5 ml of urine with Rothera's mixture and then add 1-2 ml of conc. Ammonia solution. Gently mix by rotation and allow to stand and observe.	Permagnate colour or deep purple ring is observed at the junction of two layers	Acetone and acetoacetic acid present. Therefore urine contains ketone bodies.

- **Clinical Interpretation**
- Acetoacetic acid, β -hydroxy butyric acid & acetone are collectively known as ketone bodies. Normal urine of 24hrs may contain ketone bodies upto 1 mg. Excess of ketone bodies in blood is called as ketonemia and higher excretion of ketone bodies in urine is known as ketonuria and overall condition is known as ketosis. Ketone bodies are observed in fasting, untreated diabetes mellitus, high fat, low carbohydrate diet, prolonged starvation, pregnancy & lactation.

4. Test for Blood:-

Test	Observation	Inference
4. Benzidine Test : Take 2ml of urine in a test tube and boil for 5min. Cool it. Mix equal volume of benzidine solution (2-3ml) and H_2O_2 in a test tube and add the boiled cooled urine into the test tube	Blue colour solution later turns green	Blood is present in urine. Therefore urine contains blood

- **Note:** -Normal urine contains any RBC but few leucocytes & epithelial cells may be present.

- **Clinical Interpretation**
- Normal urine does not contain any RBC but few leukocytes and epithelial cells may be present **Haematuria** - It is a condition in which RBCs is present in urine & occurs in acute glomeruli nephritis, stone in kidney and ureter, renal tuberculosis and carcinoma of kidney.
- **Hemoglobinuria:**-It is a condition in which free haemoglobin is present in urine and occurs in malaria, Typhoid, hemolytic jaundice, yellow fever, intravascular hemolysis, due to mismatch blood transfusion and autoimmune hemolytic anaemia.

5. Test for Bile salts and bile pigments

- **a). Hay's Sulphur powder test for bile salts**
- **Principle:** Sulphur powder sinks down in test tube containing urine sample because bile present in urine contain bile salts which acts as emulsifying agent reduces the surface tension of urine sample.

Test	Observation	Inference
5. Hay's Sulphur powder test : Take 3ml urine in a test tube and sprinkle a pinch of sulphur powder and observe. **Control:** Take 3ml water in a test tube and sprinkle a pinch of sulphur powder and observe	Sulphur powder sinks down to bottom of the test tube. Sulphur powder floats.	Bile salts confirmed. Therefore urine contains bile salts.

b). Fouchet's test for bile pigments

- **Principle:** Bile pigments present in urine solution reacts with $BaCl_2$ to form white precipitate. When this is dried and reacted with fouchet's reagent green or bluish green colour precipitate is obtained due to formation of complex derivative.

Test	Observation	Inference
6. **Fouchet's test :** Take 3ml urine solution in test tube. add 2 ml 10% BaCl2. until thick white precipitate is obtained. Filter it and to the precipitate on the filter paper add few drops of Fouchet's reagent and observe.	Colour changes from yellow to pista green	Bile pigments confirmed. Therefore urine contains bile pigments.

- **Clinical Interpretation:**
- Bile salts are Na+ or K+ salts of glycocholates & taurocholates.
- These are found in urine in obstructive jaundice & amount of bile salt is proportional to the degree of obstruction. These are found to some extent in infective hepatitis due to partial obstruction.
- In hemolytic jaundice bile pigments are found in urine without bile salts.