Exercise 31

Aim: To detect the presence of albumin in the given sample of urine.

Principle: Nitric acid causes the precipitation of albumin. When heated or treated with sulphosalicylic acid, albumin undergoes coagulation.

Requirement: Glasswares: Test tubes, graduated pipette (5 mL capacity), spirit lamp; Chemicals: Concentrated nitric acid, acetic acid, Robert's solution, sulphosalicylic acid or a solution containing 13% salicylic acid and 20% sulphuric acid; Miscellaneous: test tube stand, test tube holder.

Procedure

(a) Nitric acid ring test
- Take 5 mL of concentrated nitric acid in a test tube.
- Incline the tube and add the urine sample with a dropper, so that the latter flows down slowly along the side of the test tube to form a separate layer.
- A white ring develops at the junction of the two liquids which indicates the presence of albumin in the urine sample.
  
  OR
- Take about 5 mL of Robert's solution in a test tube.
- Now incline the test tube and add 2 to 3 mL of the given sample of urine by means of a dropper along the inner side of the test tube so that it forms a layer over the Robert's solution.
- The presence of white ring at the junction of two layers indicates the presence of albumin in the sample.

(b) Heat coagulation test
- Take about 6 to 8 mL of urine in a test tube.
- Incline the test tube at an angle and heat the upper one-third of the test tube by a low flame.
- Turbidity develops in the heated portion of the urine.
- Add 1% acetic acid drop by drop and boil or simply add a drop of 33% acetic acid.
- If the turbidity persists it confirms the presence of albumin in the urine sample (disappearance of turbidity, confirms the presence of phosphates).
Exercise 31

(c) Sulphosalicylic acid test
- Take 3 mL of urine in a test tube.
- Add a few drops of sulphosalicylic acid and heat it gently.
- A whitish or cloudy turbid solution or precipitate (coagulation) in the solution indicates the presence of albumin in the urine sample.

Discussion

A trace of protein which is less than 250 mg (in 24 hours urine) is found in normal urine. Under pathological conditions like albuminuria, albumin is found in urine above normal level. This amount is so negligible that it escapes detection by any of the simple test. In kidney disturbance and in high blood pressure, albumin level in urine is significantly high.

Questions
1. What is the colour of the urine and name the pigments responsible for this characteristic colour?
2. In which organ of our body, highly toxic ammonia is converted into urea?
3. Name the disorder that shows presence of excess urea in the urine.
4. Name the disorder in which glucose level is high in urine.
5. What do you call those animals that eliminate nitrogen mainly in the form of urea?
6. Which other organ of our body also excretes urea in small amount?
7. Which reagents will demonstrate presence of protein in urine?
8. What is the significance of appearance of different colours while performing Benedict’s test?
9. What is the significance of performing Seliwanoff’s test?
Aim: To detect the presence of bile salts in the given sample of urine

Principle: Old and damaged RBCs are removed from the circulation mostly in the spleen and to some extent in the liver by macrophages. Hemoglobin of the RBCs is broken down in the cytoplasm of macrophages. When iron is removed from heme component of hemoglobin, the iron-free portion of heme is converted to biliverdin, a green pigment, and then into bilirubin, a yellow orange pigment. Bilirubin enters the blood and is transported to the liver from spleen. In the liver, bilirubin is secreted by liver cells into bile, which passes into the small intestine and then into the large intestine. Bilirubin is detected in urine in certain pathological conditions only.

Requirement: Test tubes, measuring cylinders (10 ml), funnel, dropping pipette or drop bottle, Lugol's iodine solution, barium chloride solution (10%), Fouchet reagent, sulphur powder, concentrated nitric acid, test tube holder, test tube stand, cotton, filter paper, distilled water.

Preparation of reagents

(i) Lugol's iodine solution: Dissolve 1g of iodine crystals and 2 g of potassium iodide in 100 mL of distilled water.

(ii) Fouchet reagent: Dissolve 25 mL of trichloroacetic acid (TCA) in 75 mL of distilled water. Now add 1g of ferric chloride to this solution and mix.

(iii) 10% Barium chloride solution: Dissolve 10 g of BaCl₂ in 90 ml of distilled water and make up the solution to 100 mL with water.

Procedure

(a) Lugol's Iodine Test
Pour 4 ml of urine sample into a test tube. Add 4 drops of lugol's iodine solution to this tube. Shake the tube well and observe. A faint yellow to brown colour indicates absence of bile pigments while light to dark green colour indicates the presence of bile pigments.

(b) Gmelins Test
Take 5 ml of concentrated nitric acid in a test tube. Add an equal volume of the given urine sample to it slowly along the sides of
the test tube. Formation of a green, blue, yellow or red ring at the junction of the two solutions indicates the presence of bile pigments.

**Procedure**

Bile pigments present in the urine react with concentrated nitric acid and induces formation of a coloured ring at the junction between the urine and acid layer.

**(c) Fouchets Test**

Take 5 mL of the given urine sample. Add 2-5 mL of BaCl₂ to this test tube and mix the two solution. A precipitate will appear. Now filter the mixture. The precipitate containing the bile pigments remains on the filter paper. Add 2 drops of Fouchet reagent to the precipitate on the filter paper. If the precipitate turns green, it shows the presence of bile pigments.

**Discussion**

The colourless bilirubin is oxidised by the ferric ion of ferric chloride (present in the Fouchet Reagent) to green biliverdin.

\[
\text{Bilirubin} + \text{Fe}^{+++} \rightarrow \text{Biliverdin} + \text{Fe}^{++} \\
\text{(colourless)} \quad \text{Biliverdin} \quad \text{(green)}
\]

**Questions**

1. Give the names of the pigments found in bile.
2. Which organ of the body produces bile pigments?
3. Which pigment provides colouration to the bile?
4. What are the functions of bile pigments?
5. How are the bile pigments produced?
6. Mention the name of the diseases during which excretion of bile pigments occurs in urine.
7. What are the different tests to detect the presence of bile pigments in urine?
Aim: To study the human skeleton

Principle: Human skeleton in adults is composed of 206 bones. It is divisible into two categories: Axial and appendicular skeleton. The axial skeleton consists of the bones of the skull, vertebral column, sternum and ribs. The appendicular skeleton consists of the bones of the limbs along with their girdles.

Requirement: Specimen of human skeleton

Procedure

(i) Observe the different types of bones and joints present in a human skeleton.

(ii) Draw labeled diagram of your observations.

Observation

(a) Human Skull

(i) It is composed of two sets of bones - cranial and facial (Fig. 33.1).

(ii) Cranial bones are occipital, parietal, frontal, temporal, sphenoid and ethmoid bones.

(iii) Corresponding to their location in the body, the cranial bones have strong bone case for eyes called orbit.

(iv) Facial bones form the front part (i.e., face) of the skull.

(v) A single U-shaped bone called hyoid is present at the base of the buccal cavity.

(vi) A nasal passage formed by nasal bones is present just below the orbit.

(vii) Maxilla and pre-maxilla bones form the upper jaw, and the mandible bone forms the lower jaw. These two bones also form the face, and into them are lodged teeth in special sockets. Teeth are not bones.

(viii) Distinct sutures in zig-zag fashion are present at the junctions of the frontal with the two parietals, as well as between the two parietals.
(ix) The occipital bone has a very big foramen at its posterior base, the foramen magnum, through which the brain is continued posteriorly as a spinal cord.

(x) The skull is dicondylic, i.e., it has two occipital condyles for articulation with the first cervical vertebra.

**Note:** The cranium forms the hard protective outer covering for the brain. All the bones of the cranium are articulated by fibrous or fixed or immovable joints. Mandible is the strongest bone of the body.

(b) **Vertebral Column**

(i) It consists of 26 serially arranged units (Fig. 33.2) called vertebrae (singular: vertebra).

(ii) Each vertebra has a central hollow portion called neural canal through which the spinal cord passes. The first vertebra is the atlas and it articulates with the occipital condyles of skull.

(iii) Vertebral column has several types of vertebrae: cervical (7), thoracic (12), lumbar (5), sacral (1 which is fused), and caudal or coccygeal (1 which is fused).

(iv) A typical vertebra (Fig. 33.3) has a — (i) centrum, the modified notochord (ii) two laterally projecting transverse process (iii) a neural canal through which passes the spinal cord (iv) a mid dorsal neural spine formed by the union of neural arch. Depending upon their location in the body, secondary modifications are seen in the length of transverse process and the length of neural spine. The two neighbouring vertebrae articulate with each other through their anterior and posterior zygapophyses. Intervertebral discs are present between the centra of two neighbouring vertebrae.

**Note:** The vertebral column forms the central axis of the body and keeps it erect. It encloses and protects the spinal cord and provides surface for the attachment of skull, ribs, pectoral and pelvic girdles, as well as several muscles in the neck, thorax, abdomen and waist.

(c) **Rib Cage and Sternum**

(i) Sternum forms the floor of branchial basket. It bears 7 (seven) notches for articulation with ribs. It has hexagonal disc at the top called manubrium. Lower end has a reduced bone called xiphoid process (Fig. 33.4).
(ii) Ribs can be put under two classes: the thoracic ribs, and the sternal ribs. The thoracic ribs articulate with the thoracic vertebrae, and the sternal ribs do so with the sternum. Some (7) of the thoracic ribs are attached to the sternal ribs with the help of ligaments, enabling the increase and decrease in volume of the thoracic chamber during respiration.

(iii) There are 12 (twelve) pairs of thoracic ribs. Each rib is a thin flat bone and is carried ventrally from the vertebral column. It has a head articulating with the centrum, and tubercle articulating with transverse process of vertebrae (Fig. 33.4).

(iv) 7 (seven) pairs of thoracic ribs are attached to the sternal ribs.

(v) Last 5 (five) pairs of thoracic ribs do not articulate with sternal ribs, and are called false ribs. Among these, the last 2 (two) pairs of false ribs are free and are called floating ribs.

**Note:** Rib cage and sternum covers and protects the vital organs such as heart and lungs. The ribs are also helpful in breathing.

(d) **Pectoral Girdle**

(i) It consists of a clavicle and a scapula (Fig. 33.5).

(ii) Scapula is a large triangular flat bone with a slightly elevated ridge called spine. The spine projects as a flat, expanded process called the acromion.

(iii) The clavicle is a long slender bone with two curvatures. The clavicle articulates with the acromion.

(iv) Below the acromion is a depression called the glenoid cavity, for articulation of the head of the humerus to form the shoulder joint.

**Note:** Pectoral girdle is formed of two halves. Each half consists of a scapula and a clavicle. The clavicle is commonly called collar bone. The third element of vertebrate pectoral girdle, the coracoid is highly reduced in man, and is present only by a small projection over the glenoid cavity.

(e) **Pelvic Girdle**

(i) It consists of two halves.

(ii) Each half is formed by the fusion of three bones - ilium, ischium and pubis (Fig. 33.6).

(iii) At the point of fusion of the above bones is a cavity called acetabulum to which the thigh bone articulates.
(v) The two halves of the pelvic girdle meet ventrally to form the pubic symphysis.

**Note:** Pelvic girdle is commonly called hip bone. The ilium articulates anteriorly with the flat transverse process of sacral vertebrae.

(f) **Bones of the Hand or Fore Limb**

(i) It is made up of bones consisting of humerus, radius and ulna, carpals, metacarpals and phalanges (Fig. 33.7).

(ii) Humerus is a straight bone with a long shaft, and forms the upper arm. The head of the humerus fits into the glenoid cavity of the pectoral girdle. It has a crest at its proximal end in the form of deltoid ridge for the attachment of arm muscles. The distal end has a foramen and a trochlear process, which forms elbow joint with radius and ulna.

(iii) Radius-ulna consists of 2 (two) separate bones of the forearm namely radius and ulna. Ulna is more developed and has olecranon process at its proximal end, which forms elbow joint with humerus.

(iv) Carpals consist of 8 (eight) small bones arranged in two rows. It forms the wrist (Fig. 33.8).

(v) Metacarpals are made up of 5 (five) long bones forming the palm of hand.

(vi) Phalanges consist of 2 (two) in the thumb and, 3 (three) bones in the remaining four fingers, thus totalling 14 (fourteen) bones.

(g) **Bones of the Leg or Hind Limb**

(i) It is made up of femur, tibia and fibula, patella (knee cap) tarsals, metatarsals, phalanges (Fig. 33.9).

(ii) The femur is the longest bone. The head of femur fits into the acetabulum of the pelvic girdle. The proximal end has trochanters for attachment of thigh muscles. The distal end has two condyles, which articulate with triangular shaped patella and proximal part of tibia to form knee on the ventral side.
(iii) Tibia-fibula consists of two separate bones namely tibia and fibula and is present in the shank region of leg. Tibia is more developed than fibula. Its proximal end articulates with femur and patella and forms knee.

(iv) There are 7 (seven) tarsal bones, which are arranged in two rows to form the ankle. The largest bone of these is calcareous which form heel (Fig. 33.10).

(v) Metatarsals consist of 5 (five) bones and form foot.

(vi) Phalanges consist of 2 (two) bones in big toe and three bones in each of the remaining toes thus totaling 14 (fourteen) bones.
Exercise 34

**Aim:** To study different types of joints in human skeleton.

**Principle:** Bones may be movable, slightly movable or immobile depending on the nature of joints. Joints are defined as regions/surfaces of contact between bone and cartilage.

**Requirement:** Specimen of human skeleton, charts and models of skeleton.

**Procedure**

(i) Observe the different types of bones and joints present in a human skeleton.

(ii) Draw labeled diagram of your observations.

**Observation**

(a) **Gliding Joints**

(i) These are flat joints, which allow back and forth or side-to-side movement of all or a few joining elements. However, twisting is not possible.

(ii) These joints are found between bones of tarsals and carpals (Fig. 34.1).

(b) **Pivot Joints**

(i) These joints allow rotational movement.

(ii) These joints are found between the atlas and axis vertebrae of backbone. It is the odontoid process of the axis vertebra over which the atlas along with the skull rotate (Fig. 34.2).

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**Fig. 34.1** Gliding Joints (a) hind limb bones showing the joint (b) Diagramatic representation

**Fig. 34.2** Pivot Joints (a) Skull and vertebral column showing the joint (b) Diagramatic representation
(c) **Hinge Joints**

(i) These joints allow movement in one plane only.

(ii) These joints are present in elbow and knee (Fig. 34.3).

(d) **Saddle Joints**

(i) These joints allow movement in two planes.

(ii) These joints are found in bones of metacarpals and carpals of thumb (Fig. 34.4).

(e) **Ball and Socket Joints**

(i) These joints allow movement in more than two planes (Fig. 34.5).

(ii) These joints are present between humerus with pectoral girdle, femur with pelvic girdle, and maleus with incus (in ear ossicles).