Experiment No. 1.

①. Recording of Blood pressure using a Sphygmomanometer.

Blood pressure—Arterial blood pressure is the force of pressure which the blood is exerting against the walls of the blood vessels in which it is contained. The blood pressure in the arteries varies during the cardiac cycle. During ventricular systole, when the left ventricle is forcing blood in to the aorta, the pressure rises to a peak, referred to as *systolic pressure*. During diastole the pressure falls, the lowest value it reaches is referred to as diastolic pressure.

Measurement of blood pressure—Blood pressure is measured by blood pressure instrument called sphygmomanometer and a stethoscope is also needed.

Sphygmomanometer consists of a long glass manometer capillary tube. The upper end of this tube is open while its lower end is slightly bend. The manometer is mounted in the middle of a dark scale having markings on its both sides. The lower bend end of the manometer is connected to the ventral side of a large bulb filled with mercury. The upper open end of the manometer is exposed to atmospheric pressure but is guarded by a valve to check the spilling of mercury from the open end. The upper end of the bulb is connected with a nozel and nozel is

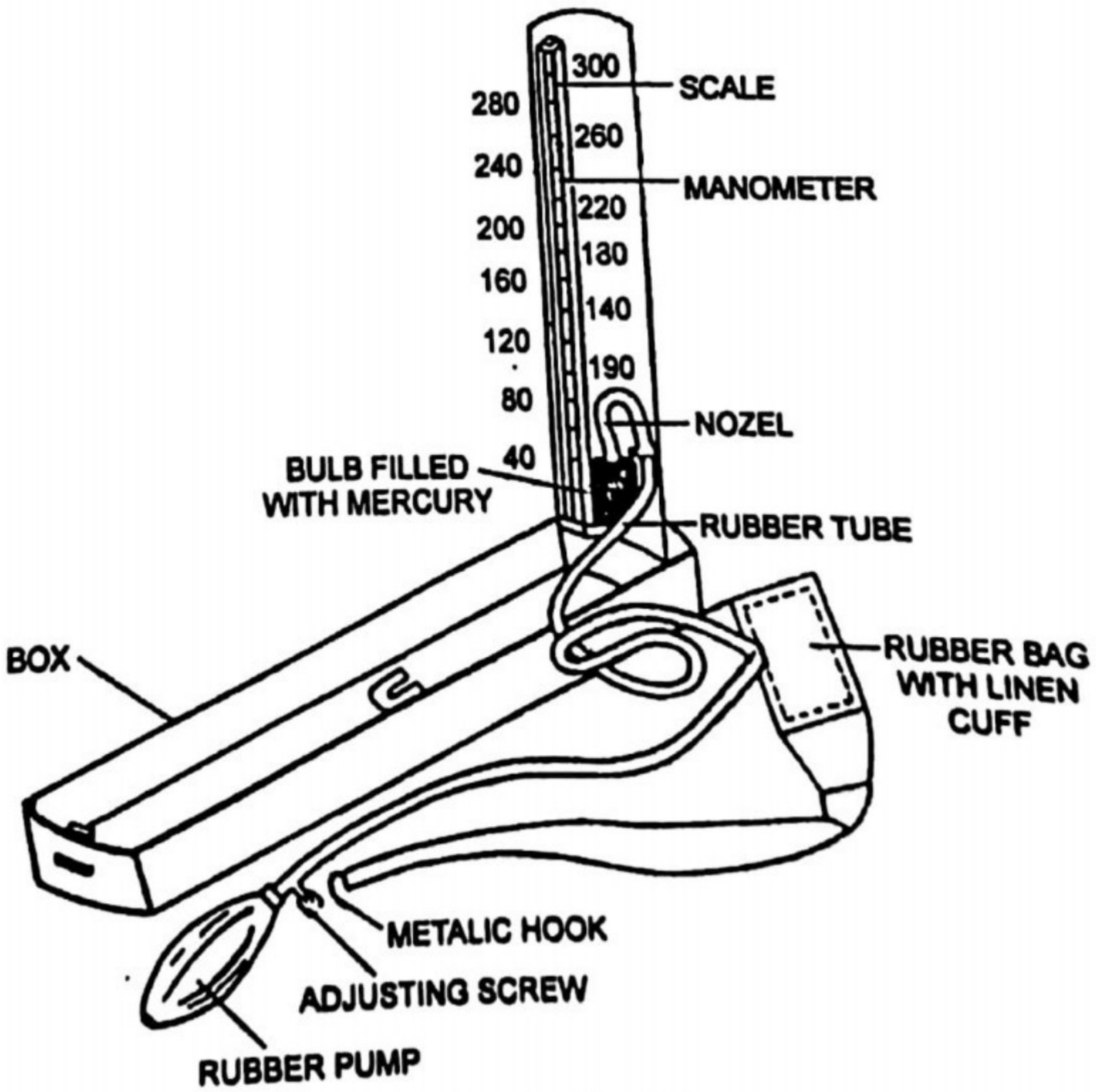


Fig. 21.14. Diagram showing the Sphygmomanometer.

connected with a narrow lumen rubber tube. The free end of the rubber tube is connected to a flexible rubber bag enclosed in a *cuff*. The cuff is further connected to **a rubber pumping bulb** with a rubber tube. the neck of the bulb is provided with an **adjusting screw**; the tightening or untightening of this screw ensures the flow of air in to the cuff (inflation) or out of the cuff (deflation).

There are 3 methods used for the measurement of blood pressure. These are as follows—

1. Oscillatory method—In this method a pressure cuff is wrapped over the branchial artery and the oscillations which are produced by pulsations are observed. Always the instrument is kept at the level of heart. When the cuff pressure is increased more than the systolic pressure, the oscillations disappear, but on releasing the pressure gradually, the oscillations become larger and prominent. The pressure at which the larger oscillations are seen, is considered as **systolic pressure**, But on further release of pressure, the oscillations becomes smaller and disappeared. The pressure at which the oscillation just becomes disappears is called the **diastolic pressure.**

2. Palpatory Method—In this method, the instrument is kept at the level of heart and the pressure cuff is wraped over the upper arm. The pressure is raised 200 mm of Hg and then gradually released. When the pulse just appear on the wrist, the pressure is noted which is called **systolic pressure.** This method is not accurate. The diastolic pressure can not be determined by this method.

3. Auscultatory method—In this method also, the instrument is kept at the level of heart and the cuff is wrapped over the upper arm. The pressure is raised to 200 mm of Hg and then releases gradually. Variations of sounds are heard with a stethoscope placing its chest piece on the branchial artery just below the cuff. The pressure is released gradually, following variations of sounds heart; **Ist phase**—Sudden-appearance of a clear tapping sounds which indicates **systolic pressure.** It persists up to the fall of pressure 15 mm of Hg. **2nd phase**—The tapped sound is replaced by murmur sound. It also persist another 15 mm of Hg. **3rd phase**—the murmur sound is replaced by a clear loud gong sound it persists for the next 20 mm of Hg. **4th phase**—the loud gong sound suddenly replaced by muffled sound and rapidly begins to fade. This point indicates the **diastolic pressure.**

Normal Range of Blood Pressure

	Diastolic	Systolic
In infants	50	70-90
In Childhood	60	80-100
During Adolescent stage	60	90-110
In young adult	60-70	110-125
As age advances it is increased	80-90	130-150

HAEMATOLOGY

EXPERIMENT NO. 2

ESTIMATION OF HAEMOGLOBIN

Principle—Hb is converted in to acid haematin by hydrochloric acid. The brown colour of the compound is matched against a brown glass standard in a comparator.

Requirements—Sahli-type haemoglobinometer consisting of the comparator with glass standards, a square Hb tube marked both in grams and percentage figures and Hb pipette marked at 20 cu mm., 0.1 N HCl and distilled water.

Procedure—1. Fill the Hb Calibrated tube up to the mark 20 (not less) with 0.1 N HCl by means of a droper.

2. Fill the Hb pipette exactly up to 20 cu mm mark by gentle controlled sucking; the pipette is held horizontally while taking the blood. If a slight excess is drawn in, it may be removed by touching the point of the pipette with the finger or gauge. If a great excess has been drawn in inaccuracy will result, in this case the pipette must be cleaned, dried and refilled. Wipe off with gauge the blood on the outside of the pipette.

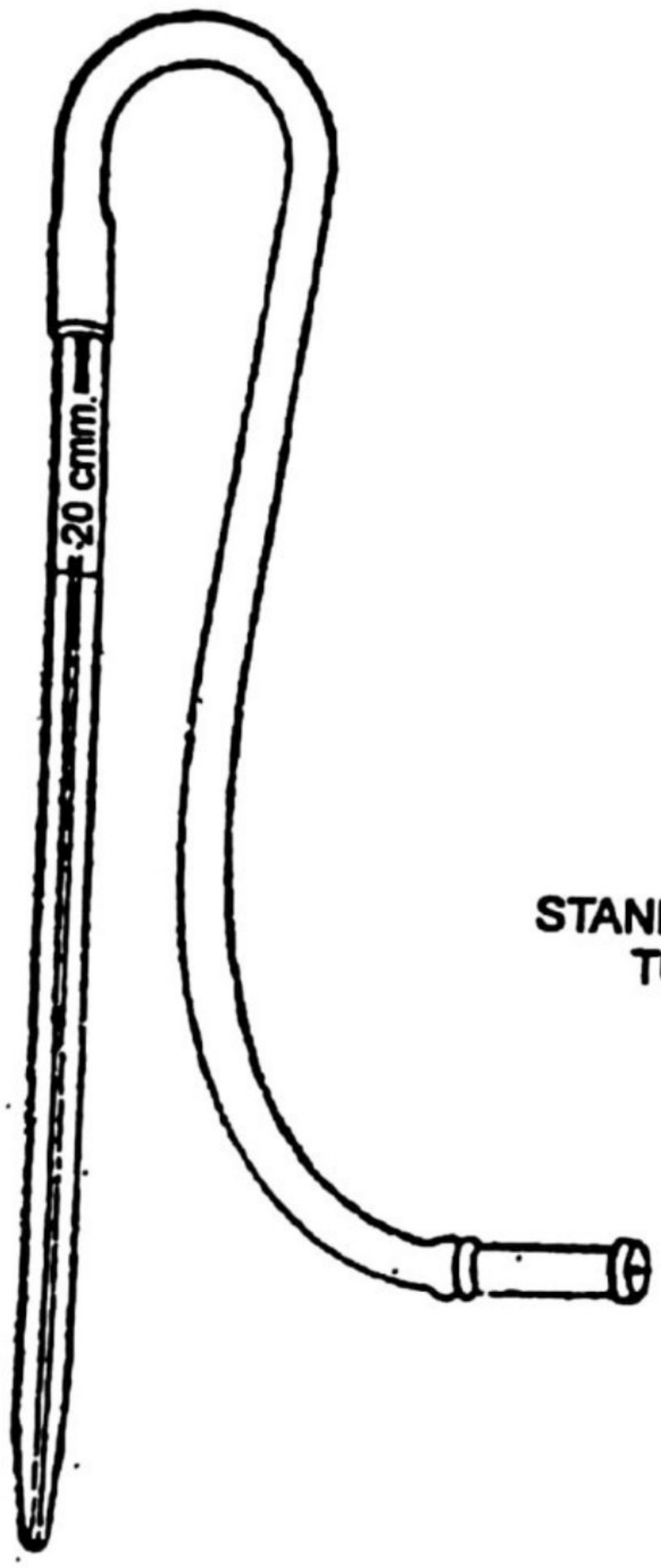
3. Empty the pipette in to acid in the tube by keeping the point of the pipette to the bottom of the tube and gently blowing off the blood without causing bubbles. Rinse the pipette at least three times by drawing in and discharging the blood acid mixture. Now withdraw the pipette half way up the tube and rinse the outside of pipette with a few drops of the acid.

4. Mix the acid haematin solution in the tube with the glass rod and allow the tube to stand for 10 minutes. In this interval at least 95% of the colour of acid haematin is developed.

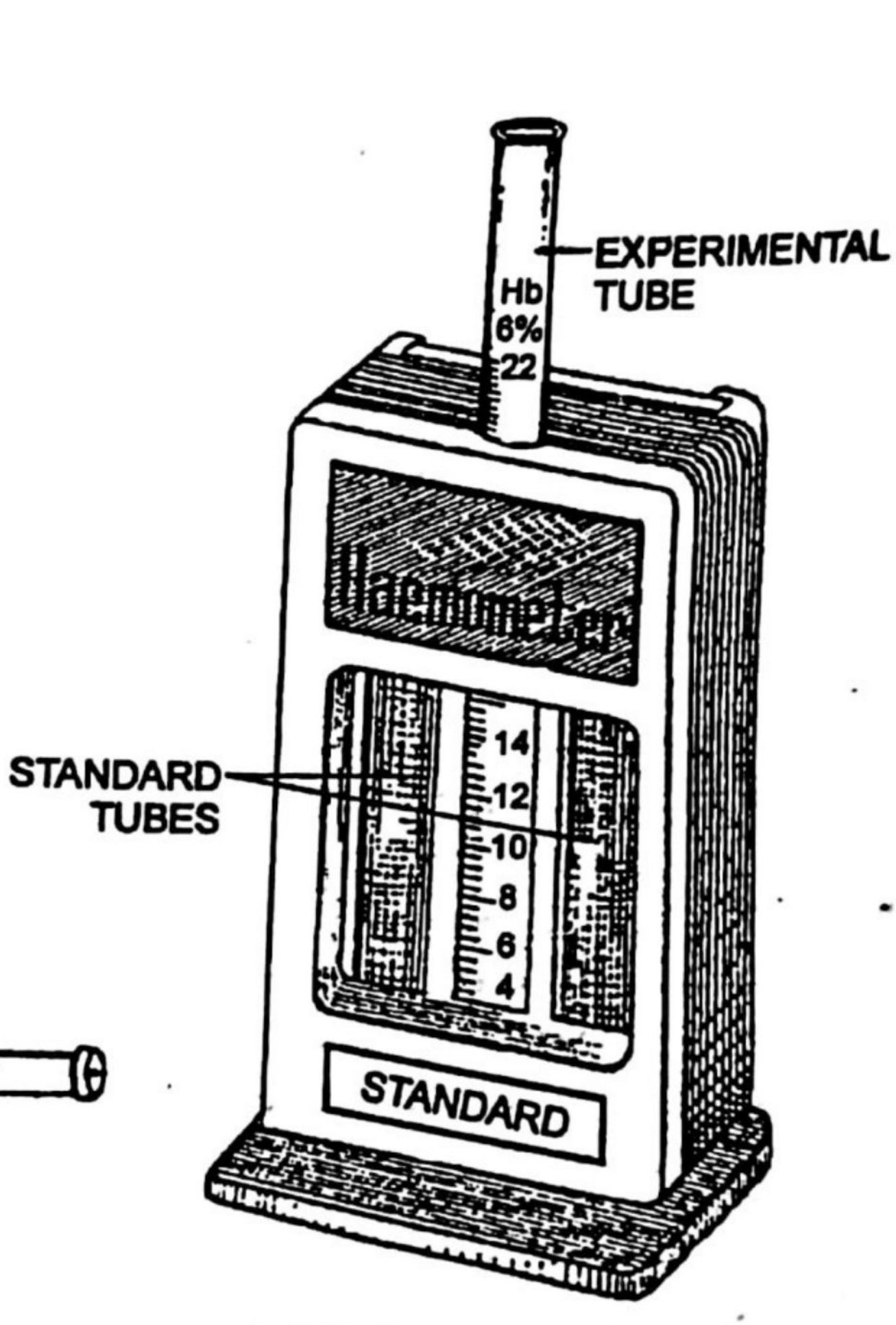
5. Now dilute the solution of acid haematin by adding distilled water, drop by drop, stirring the mixture all the time with glass rod. The comparator is held against good day light and the addition of water continued till the colour of solution matches perfactly with that of the standards. Take the reading in grams percent. The bottom of the meniscus is read.

Normal Range of Hb

Men	15.5 ± 2.5 g/dl
women	14.0 ± 2.5 g/dl
Infants (full term cord blood)	16.5 ± 3.0 g/dl
Children (3 months)	11.0 ± 1.5 g/dl
Children (3-6 years)	12.0 ± 1.0 g/dl
Children (10-12 years)	13.0 ± 1.5 g/dl



A. MICROPIPETTE



B. HALDANE'S HAEMOMETER

Fig. 21.1. Diagram showing the Hldane's haemometar with micropipette.

Experiment NO. 3

Object:- To prepair haemin crystal from Human blood.

Requirements—Blood sample, slide, cover-slip, microscope, Nipp's solution, spirit lamp.

Principle—Haemoglobin is a conjugated protein. It is made up of a protein molecule globin and a non-protein substance, haeme or haematin. When blood is heated in presence of acid, it splits into its components and forms haemin crystals which can be recognized under the microscope.

Procedure—1. Take a drop of blood on a clean slide.

2. Allow the blood to dry.

3. Put 2 drops of Nipp's solution and cover it with a cover slip.

4. Warm the slide gently over the spirit lamp.

5. Cool the slide and examine under the microscope.

Fig. 12.2 Haemin crystals.

Observations—Brown, shining rhombic crystals are seen.

Precaution—Too much heating should be avoided.

Significance—Preparation of haemin crystals is useful in medicolegal work, where it is difficult to differentiate between blood marks and other red coloured marks.

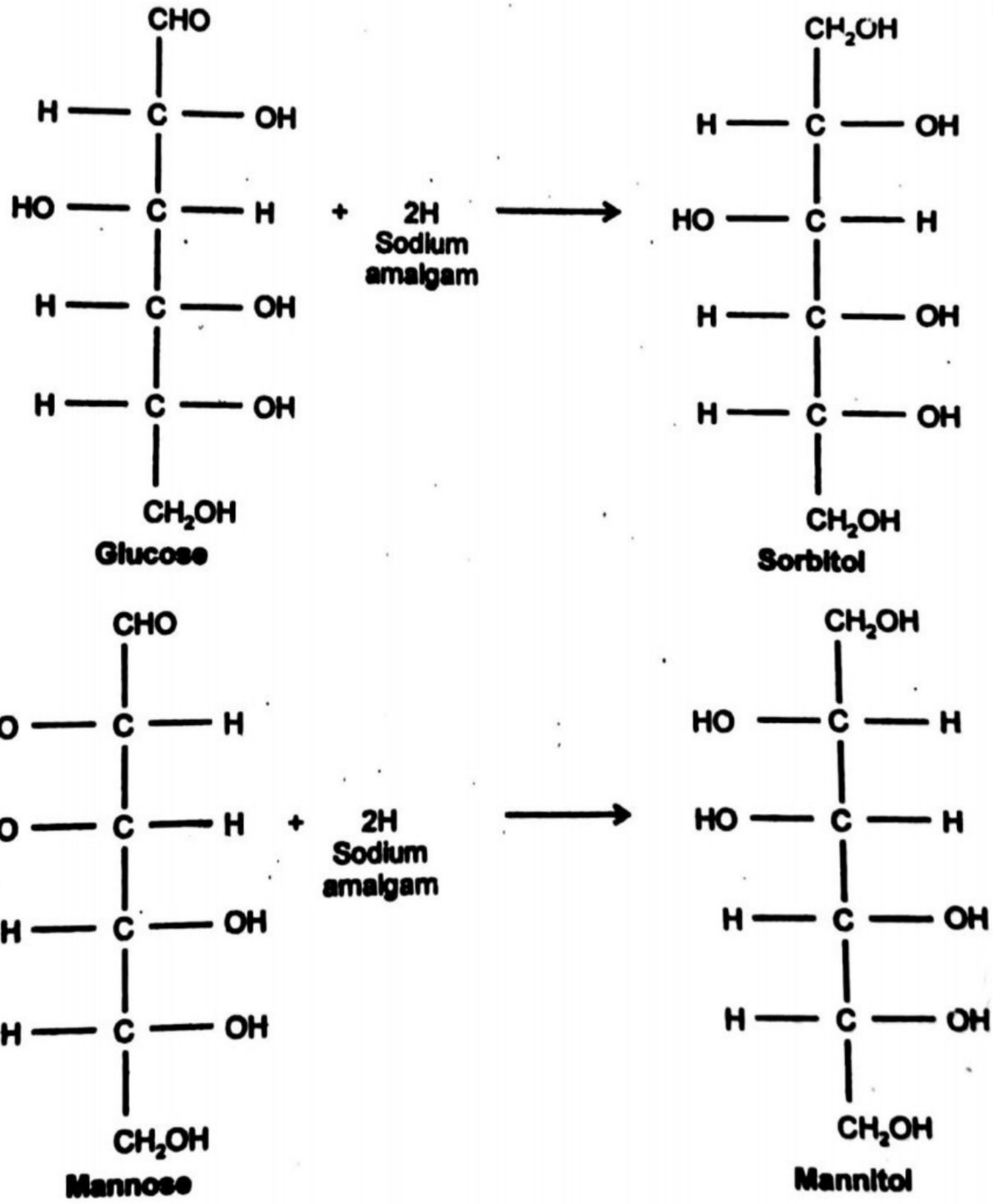
Experiment No.4

Benedict's test for reducing sugar and Iodine test for starch.

Fehling test—Take 1 ml. of Fehling's solution in a test tube, a few drop of the sugar solution is added and the mixture is heated. The production of reddish precipitate shows that reduction has taken place.

⇒ **Benedict test**—The above test is repeated, but the Benedict solution is used instead of Fehling solution. A reddish brown precipitate is formed.

Reduction—The monosaccharides are reduced to their corresponding alcohols by treating them with reducing agents like sodium amalgam. Thus, glucose yields sorbitol, mannose yields mannitol, glalactose yields



Experiment No-5

V.S. OF PITUITARY GLAND

Comments

1. The pituitary gland is situated on the ventral side of the brain, attached to the hypothalamus by infundibular stalk.

2. Pituitary gland has 2 lobes : anterior and posterior.

3. The anterior lobe constitutes adenohypophysis while posterior lobe constitutes neurohypophysis.

4. The adenohypophysis has 3 parts—pars distalis, pars tuberalis and pars intermedia.

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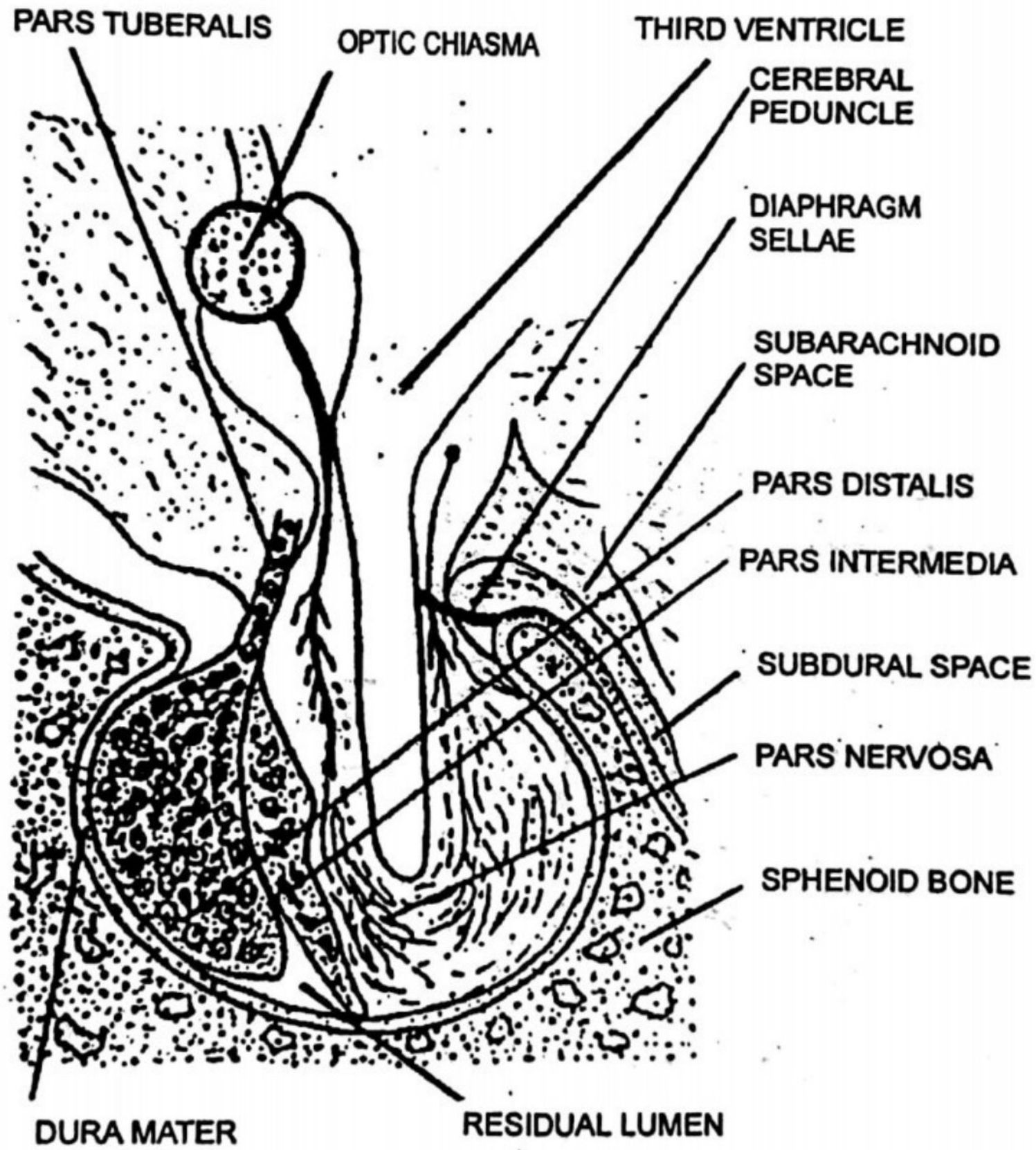

Fig. 15.50 Vertical section of Pituitary gland of mammal

EXPERIMENT NO. 6

TO DEMONSTRATE ACTION OF SALIVARY AMYLASE

Principle—The enzyme salivary amylase is present in the saliva. It is also known as ptyalin. It changes starch in to glucose and maltose. It acts steadily at body temperature but acts optimally between 40-50^{0}C. The optimum pH for its activity is between 6 to 7 and that of salivary amylase of man is 6.6

Requirement—Test tube, test tube holder, sprit lamp, 1% starch solution, Man's saliva, Iodine solution, Benedict's solution, pipette, incubator set at 37^{0}C.

Procedure—Take 4 to 5 drops of Man's saliva in clean test tube and add 1.0 ml of 1% starch solution shake the test tube between the palm for 30 minutes or placed it in a set incubator at 37^{0}C for 30 minutes. Then add 1.0 ml of iodine solution. No change in colour of iodine solution will indicate the presence of amylase enzyme. Boil the test tube till the colour of iodine is disappear. Then add 1.0 ml of Benedict's solution and heat it till boils. A brick red precipitate will form, which confirm the presence of amylase in saliva. The brick-red precipitate of cuprous oxide shows the presence of glucose in the test tube, because glucose is a reducing sugar, it reduces copper sulphate in to cuprous oxide.

Result—The presence of amylase is confirmed in Man's saliva.

Experiment No- 7

T.S. OF TESTIS OF MAMMAL

Comments

1. Testes are small, oval glands, covered by a dense fibrous connective tissue, called tunica albuginea.

2. The tunica albuginea is surrounded by peritoneum.

3. Each testis is divided by fibrous septa into lobules.

4. Each lobule contains seminiferous tubules, lying in connective-tissue matrix. Connective tissue contains blood vessels, lymph vessels, nerves and interstitial cells.

5. Each seminiferous tubule is oval in shape, surrounded by basement membrane and lined by germinal epithelium.

6. Some larger cells, called sertoli cells are present in seminiferous tubules, which provide nourishment to developing spermatozoa.

7. Sperms are seen in various stages of development as follows—

(i) Spermatogonia : lie towards the peripheral region of the tubule.

(ii) Primary spermatocytes : lie just inside the spermatogonia.

(iii) Secondary spermatocytes : lie just inside the primary spermatocytes.

(iv) Spermatids : clusters of cells between primary spermatocytes and spermatozoa.

Experiment-7

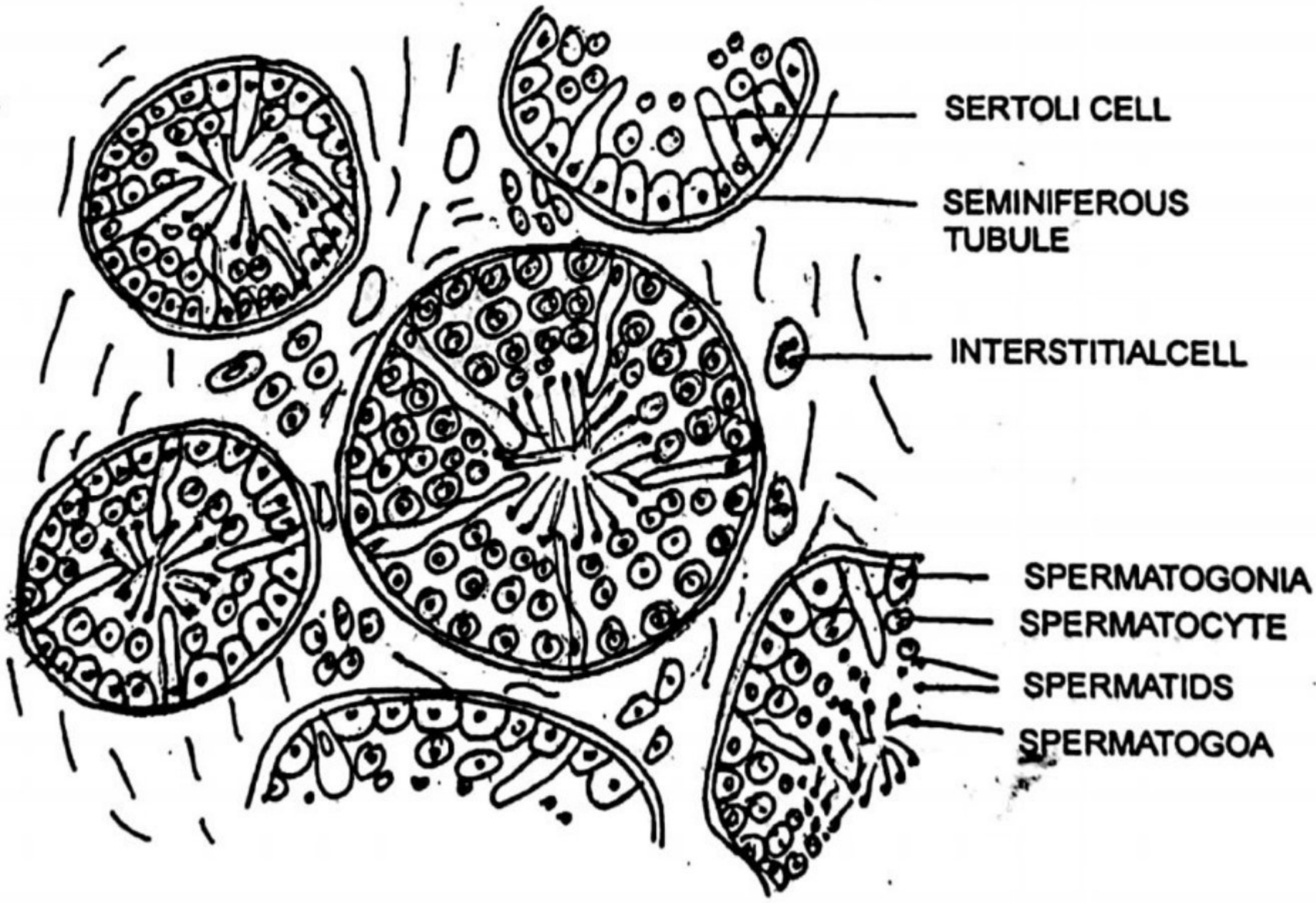

Fig. 15.52 T.S. of testis of mammal.

Experiment No-8

Experiment. No (8),

T.S. OF OVARY OF MAMMAL

Comments

1. Ovary is the most important endocrine gland in females. There are two ovaries in female.

2. Each ovary is surrounded by germinal epithelium. Just below the germinal epithelium is tunica albuginea.

3. The connective tissue and spindle shaped cells form the stroma of ovary, in which a large number of egg cells in various stages of development are seen.

4. The cells of germinal epithelium grow to form primary follicles.

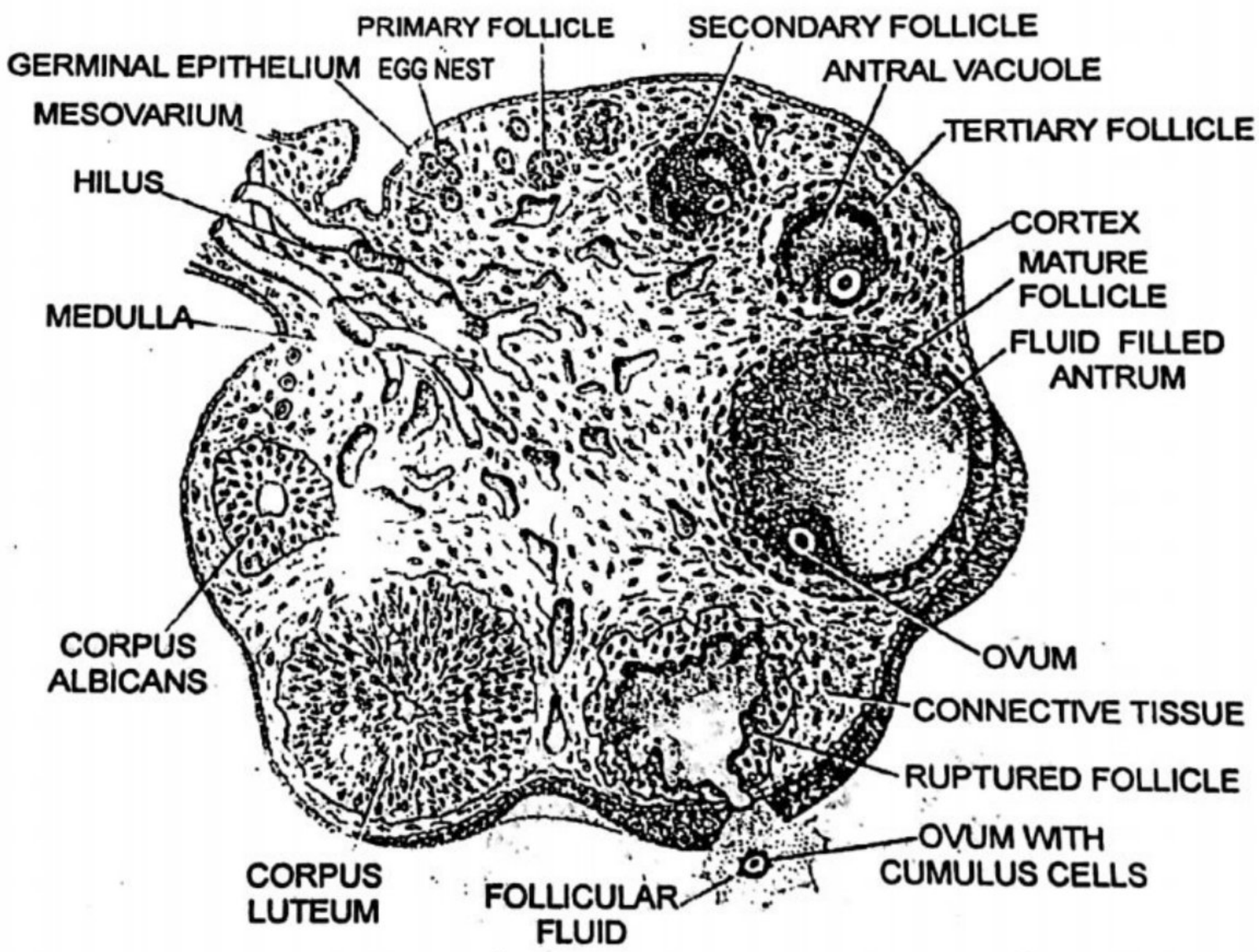

Fig. 15.53 T.S. of Ovary of mammal

5. Under the influence of FSH, primary follicles begin to mature and one of them forms Graffian or mature follicle, containing an ovum.

6. Graffian follicle has 3 layers : theca externa, theca interna and membrana granulosa.

7. The cavity of Graffian follicle is known as antrum, which is filled with liquids containing estrogen hormones.