Practical Physiology Book
Understanding physiology has been the crux of learning medicine. The basic of medicine has greatly depended on physiology. No wonder learning of physiology has been strengthened and reinforced by the clinical and experimental methods. The leaps and bounds research, in medical physiology, has been made enormously due to the experimental physiology. It is mandatory for every medical student to understand physiology and all its branches in terms of practical. This currently updated edition will certainly satiate the academic needs of students as well as the teachers in physiology. The upcoming concepts in medical education, such as OSCE and OSPE, have been inducted to set the learning minds into further reading. These make physiology a much more interesting subject to read and comprehend.

The immense response that we received for the first edition has encouraged and inspired us to go ahead with the second improvised edition. The printed matter has undergone repeated scrutiny and perusal in order to present the best. Any suggestions and improvisation towards the betterment of this edition is solicited. It is a pleasure to realize that ultimately all the benefit goes to the learning community.

M Chandrasekar
Nitesh Mishra
Preface to the First Edition

I have learnt from my own experience in teaching over the years as well as gathered from mutual discussion with colleagues that there is quite a lack of uniformity in teaching of practical physiology to undergraduates in different institutions within the country as well as amongst different staff members within the same department in an institution, some teachers teaching ‘too much’ in the limited time meant for learning skills while others ‘too little.’

It was realized that in order to learn practical physiology, students had to bank upon the main textbook of physiology which is in no way complete for requirements of physiology practicals for undergraduates.

There has been a re-thinking during the past decade that the syllabus and the method of teaching of physiology are at variance with bedside medical practice. It is therefore not surprising that teachers in this discipline are blamed of bothering little to identify the needs of the undergraduate medical student or of not making any serious attempt to revitalize the concept of relevant teaching/training. Such re-thinking has prompted the author to reframe the list of practicals/demonstrations with the concurrence of teachers from several medical colleges in the country.

The purpose of laboratory techniques in hematology and clinical physiology is to develop a scientific foundation for the laboratory diagnosis and clinical diagnosis. In these manuals, an attempt has been made to acquaint the undergraduate medical student with the experimental and clinical approach to the science of physiology. On the basis of recommendation of MCI committee for revision of undergraduate curriculum in physiology, some conventional practical which had lost relevance have been deleted and new ones like ECG recording, PFT, RET and perimetry have been added.

An attempt is made to give the student an insight into the examination of the more important cardiovascular system, respiratory system and neurophysiology and to enable him, from the practicals outlined, to grasp the clinical applications of physiology.

The author gratefully acknowledges the help, cooperation and constructive criticism of all the teachers, colleagues and friends in physiology departments from many medical colleges. Special thanks are due to Dr Anandarajan, Dr Vijayalakshmi and my PGs for their useful suggestions and timely help.

We sincerely hope that this book will be of immense help to students in their practicals and viva voce in physiology.

Any suggestions for improvement would be greatly welcomed by us.

M Chandrasekar
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I–MBBS

PRACTICAL PHYSIOLOGY BOOK

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University Register number : ..........................................................................................................

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Examiners: ...........................................................................................................................................
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LABORATORY DISCIPLINE

Planning, experimentation, and observation are central concepts to all scientific advancement. Students are therefore required to:

• Read up relevant material on the practical for proper conduct of the experiment
• Collect the equipment needed for the experiment from the laboratory staff, which should be returned in good condition at the end of the practical
• Wear a clean white laboratory coat and have the following instruments: knee hammer, pen torch and measuring tape, stethoscope, etc.
• Keep the work area neat and clean during practical
• If in doubt, consult the instructor, without disturbing other students in the laboratory.

REPORT OF DATA AND PRACTICAL EVALUATION

Results or data collected by practical work should be neatly entered in observation notebook. The data should be presented to the concerned instructor, and their signature taken before leaving the laboratory.

Answer the questions given at the end of each experiment in the record. These are intended to stimulate further study, thereby, assisting the learning process.

Evaluation of the practical work for internal assessment will be based on periodic practical evaluation (20 marks) and the laboratory record (5 marks).

During the session, 4–6 practical examinations will be conducted for the purpose of evaluation. Practical examinations for internal assessment will be in the form of major experiment, minor experiment, comments, and calculations.

The marks for the practical record will be awarded on the basis of neatness, accuracy of work, conduct in the laboratory, the promptness in answering the questions, and weekly submission of the record, which will be assessed by the instructor.

OUR GOALS

• To provide the student with proper guidance to acquire a hands on knowledge of various laws and physiological principles—through the performance of experiments in physiology
• To acquire clinical, technical, and communication skills and to develop the art of careful observation and precise measurement
• To acquire training in scientific methods of thought and analysis of data
• To perform the various experiments, with a high degree of proficiency, in accordance with guidelines laid out by the Medical Council of India for the training of medical professionals.
Hematology Experiments
AIM
To study the parts of a compound microscope and to view the slides, under microscope.

APPARATUS REQUIRED
Compound microscope, cedar wood oil, stained slides, and light source.

PARTS OF THE COMPOUND MICROSCOPE
A typical compound microscope (synonyms: Light microscope, student microscope) consists of the following parts (Fig. 1.1):

1. **Base**: It is ‘U’ shaped. It serves to support the microscope and provides maximum stability.
2. **Pillars**: They are two in number and project upwards from the base and extend up to the handle of the microscope.
3. **Handle**: It is curved and is used to tilt the microscope. It supports the magnifying and adjusting systems.
4. **Body tube or barrel**: It is attached to the handle. It can be raised or lowered while viewing slides. The length of the tube is usually 160 mm.
5. **Adjustment screws**: They include the coarse adjustment screw and the fine adjustment screw. When viewing a slide, the field is initially focused using the coarse adjustment screw and then fine adjustment should be done.
6. **Stages**: They include the fixed stage and the mechanical stage. The fixed stage is a square platform with an aperture in its center that allows the light to pass from the light source to the lens. The slide to be viewed is placed on this stage. The mechanical stage is fitted
on the fixed stage. It is a calibrated metal frame with a spring mounted clip that holds the slide in position. It is fitted with two screws that can be adjusted to move the slide vertically or horizontally.

7. **Nosepiece:** It lies at the lower end of the body tube. It includes (i) a fixed nosepiece that is stationary and fitted to the lower end of the body tube and (ii) a revolving nosepiece that lies under the fixed nosepiece. The revolving nosepiece carries the objective lenses. Any lens can be rotated into position as desired. The correct position is indicated by a click.

8. **Objective lenses:** They are three in number and are fitted into the revolving nosepiece. It includes: (A) The low power objective lens, which magnifies the image 10 times, (B) The high power objective lens, which magnifies the image 40 or 45 times, and (C) The oil immersion objective lens that magnifies the image 100 times.

9. **Eyepiece:** It is fitted into the top of the body tube. Commonly used eyepieces have a magnifying power of 10 times.

10. **Mirror and illumination:** It lies at the lower end of the microscope just below the condenser. It can be adjusted to reflect light rays into the condenser. The mirror has two surfaces, namely a plane surface and a concave surface. The plane mirror is used when the light source is a distant one (for example, natural day light). The concave mirror is used when the light source is near the microscope (for example, an electrical lamp). Plane mirror is preferred when viewing an object via oil immersion lens. Certain microscopes have a built-in electrical light source.

11. **Condenser:** It is fitted under the fixed stage and can be raised or lowered using a screw. It has two lenses and an iris diaphragm that can be manipulated to control the amount of light entering the microscope. The condenser functions to convert parallel rays of light reflected from the mirror into a cone or condensed beam of light, which then passes towards the stage.
TOTAL MAGNIFICATION OF THE COMPOUND MICROSCOPE

With an eyepiece of 10× the magnification provided by a low power objective is 100, by a high power objective is 400 and by oil immersion objective is 1000. The maximum effective magnification obtained by compound microscope by altering either the eyepiece or the objective is 1500.

The first objective, to be used while viewing a slide, is the low power objective. The microscope is placed in upright position and an appropriate mirror is selected on the basis of the light source. The condenser is lowered and the diaphragm is partly shut to cut down the glare. The slide to be viewed is placed on the fixed stage. The objective is brought to about 1 cm above the slide. Now while looking into the eyepiece, the objective is raised with the coarse adjustment screw till the slide comes into focus. Using the fine adjustment screw finer details in the slide are focused.

Having focused the slide under the low power objective, a greater magnification can be obtained using the high power objective. While using this objective, the condenser is raised and the diaphragm is opened for optimal illumination. The high power objective is swung into position, and the area of the slide to be viewed is focused in a similar manner.

Further magnification can be obtained using the oil immersion objective. A drop of cedar wood oil is placed on the slide, and the oil immersion objective is swung into position. The objective is lowered until it touches the oil drop using the coarse adjustment screw. Now, the fine adjustment screw is used to obtain a clear image. After use, the lens is cleaned with xylol.

PRECAUTIONS WHILE USING THE COMPOUND MICROSCOPE

1. While transporting the microscope, one hand is placed under the base and the other hand on the handle. The microscope must not be used by placing it on the edge of the working table.
2. The microscope must not be tilted when counting cells in a counting chamber or when using the oil immersion objective.
3. The objective must not be moved down while looking into the eyepiece. Such a maneuver might risk breaking the slide.
4. All traces of cedar wood oil must be removed from the objective with a clean flannel dipped in xylol.
5. Foreign bodies, such as dust must be kept away from the eyepiece and objective.
6. While changing the objective, it should be noted that the objective clicks into its proper position.
7. Examination of the specimen under low and high power should always precede the examination under oil immersion objective.

OTHER TYPES OF MICROSCOPE

1. The simple microscope, which refers to a magnifying glass.
2. The binocular microscope, which has two eyepieces.
3. The dissection microscope, which is a type of binocular used for microdissection.
4. The ultramicroscope, which uses ultraviolet light instead of ordinary light and has a magnification of 4000 times.
5. The phase contrast microscope that uses principles of light refraction to produce an image with a good contrast.

6. The fluorescence microscope that uses a fluorescent dye to stain tissues, which are studied under the microscope.

7. The transmission electron microscope that uses a beam of electrons instead of light and electromagnetic field instead of glass lenses. This microscope obtains a magnification of up to 1 lakh times.

8. The scanning electron microscope is similar to the transmission electron microscope. It is used for three-dimensional study of surface topography of cells and objects.

<table>
<thead>
<tr>
<th>Procedure: Focus the given slide under oil-immersion in the microscope</th>
</tr>
</thead>
<tbody>
<tr>
<td>Put the slide on the stage and bring the object on the central aperture of the stage</td>
</tr>
<tr>
<td>Focus the light on the object by adjusting the plain mirror. Close the diaphragm partially and lower the condenser to the lowest position</td>
</tr>
<tr>
<td>Use the low power objective (10x) and bring it a few mm above the object</td>
</tr>
<tr>
<td>Look into the eyepiece and raise the objective slowly, using the coarse adjustment, till the object comes into focus</td>
</tr>
<tr>
<td>Use the fine adjustment to see a clear image</td>
</tr>
<tr>
<td>Scan the slide and select an area to observe under oil-immersion lens</td>
</tr>
<tr>
<td>Put a drop of cedar wood oil over the slide</td>
</tr>
<tr>
<td>Rotate the oil-immersion lens carefully over the slide. See that there is a layer of oil between the objective and the slide</td>
</tr>
<tr>
<td>Raise the condenser to the highest position and open the diaphragm fully</td>
</tr>
<tr>
<td>Focus a clear image using the fine adjustment.</td>
</tr>
</tbody>
</table>

**QUESTIONS**

1. Label the parts of the light/compound microscope in the given diagram.

2. What are the types of microscope?

3. What will be the total magnification—if a low power objective, high power objective or an oil immersion objective is used? (Eyepiece 10×)

4. What is the function of the condenser?

5. What is the function of the diaphragm?

6. When is concave and plane mirror used?

7. Why should the oil be removed from the oil immersion objective immediately after use?

8. What microscopic adjustments are made when the field of view is not clear?
AIM

To study the different methods of blood collection.

APPARATUS REQUIRED

Sterile lancet, disposable syringe and needle, anticoagulant container, cotton, spirit.

PROCEDURE

Collection of Capillary Blood by Finger Prick Method

1. Clean the tip of the ring or middle finger with a cotton swab dipped in spirit.
2. Allow the finger tip to dry as spirit can cause the hemolysis of blood and pain.
3. Using a sterile lancet prick the finger deep enough to ensure a free flow of blood (Fig. 2.1).
4. Wipe out the first few drops and collect the sample when the blood is flowing spontaneously. Do not squeeze the finger as tissue fluid can mix with blood and may cause error.

Fig. 2.1: Pricking the finger tip
**Collection of Venous Blood**

Venous blood is usually collected from antecubital vein. Clean the antecubital fossa with spirit and allow it to dry. After applying a tourniquet around the upper arm, introduce a needle under the skin and then puncture the vein from the side. During this procedure, open and close the fist repeatedly so that the vein gets engorged. When the blood flows into the syringe, release the tourniquet and draw the required amount of blood. Withdraw the needle and apply a cotton swab over the site of puncture and apply pressure till the bleeding stops (Fig. 2.2). To prepare serum, blood can be emptied into a container without anticoagulant. To prepare plasma blood has to be emptied into a container with anticoagulant.

**QUESTIONS**

1. What is the difference between serum and plasma?
2. Name the other places for capillary blood withdrawal.
3. Name five commonly used anticoagulants and explain the mode of action.
4. Name any two in vitro and in vivo anticoagulants.
5. Why is the first drop of blood discarded?
6. How does methylated spirit sterilize your fingertip?
AIM

To study the working and uses of the hemocytometer.

PRINCIPLE

A measured unit of blood is diluted quantitatively with diluents by using special measuring devices (pipettes). A mixture of blood and diluting fluid (dilution is known) is released smoothly on to the central platform of the chamber beneath the cover slip (depth of chamber is known). Cells are then counted under this known volume of mixture under the cover slip.

APPARATUS REQUIRED

Improved Neubauer’s counting chamber, RBC pipette, WBC pipette, diluents, cover slip.

DESCRIPTION

The hemocytometer is a box containing all the above apparatus. Improved Neubauer’s chamber is a glass, slide which has 2 platforms extending across its middle third. These platforms are separated from each other by shallow gutters or trenches. The central platforms are also known as the floor piece. It is 1/10 mm lower than the rest of the slide. The 2 platforms adjacent to the central platform support the cover slip. When a cover slip is placed in position, a space of 0.1 mm is present between the surface of the floor piece and the under surface of the cover slip (Fig. 3.1).
The central platform has 2 counting grids. Each counting grid has an area $9 \text{ mm}^2$. It is in turn divided into 9 squares with each square having an area of $1 \text{ mm}^2$. The 4 corner squares, which are used for WBC counting are each divided by single lines into 16 smaller squares. Each of them has a side measuring $1/4 \text{ mm}$ and an area of $1/16 \text{ mm}^2$. The central square, which is RBC counting area is divided into 25 medium squares, each of which is separated from the others by double or triple lines called tram lines. These tram lines extend in all directions. Each of the 25 squares is further divided into 16 small squares by single lines. These are the smallest squares on the grid with each small square having a side measuring $1/20 \text{ mm}$ and an area of $1/400 \text{ mm}^2$.

**FOCUSBING THE COUNTING GRID**

Using the standard protocol the counting grid is focused so that the lines are seen clearly. Initially, the lines are focused under low power; after which, the high power objective is used to view the squares.

**CHARGING THE HEMOCYTOMETER**

A cover slip is placed over the center of the hemocytometer on top of the pillars. After the blood is well mixed with the diluent, the first two drops are discarded and the pipette is placed on the floor piece so that its tip just touches the edge of the cover slip, at an angle of $30^\circ$ to the horizontal. The diluted blood is allowed to flow by capillary action evenly and slowly under the cover slip, making sure that there is no flow of the mixture into the trenches. If there is overflow of blood into the trenches, the hemocytometer must be charged again after cleaning it. Now, the cells are allowed to settle down for 2–3 minutes. After that the chamber is placed on the microscope. The lines are focused as desired, depending on the type of cell count; after which, the chamber is cleaned in running water and dried for reuse (Figs 3.2 and 3.3).

**Figs 3.2A and B:** Improved Neubauer’s chamber (A) Actual photograph; (B) Diagram

**Fig. 3.3:** Improved Neubauer’s chamber (side view)
DILUTING PIPETTES

Introduction

Blood must be adequately diluted before estimating its cell count. This is done using glass capillary pipettes. Knowing the dilution employed, the number of cells in undiluted blood can be calculated.

Description of the Pipettes

There are 2 types of diluting pipettes, namely the RBC pipette and the WBC pipette. Each pipette has a long narrow stem with a fine capillary bore and a ground conical tip.

The stem is graduated into 10 parts but has only 2 numbers etched on it namely 0.5 and 1.0. Smaller calibrations in tenths allow greater dilutions if needed. The stem widens into a small bulb, which contains a free rolling glass bead, red in RBC pipette, and white in WBC pipette. It helps in mixing the blood with the diluting fluid and for quick identification of the pipettes. The volume of the bulb is 10 times the volume of the stem in the WBC pipette, and 100 times the volume of the stem in the RBC pipette. The bulb narrows proximally into a glass capillary to which a long narrow soft rubber tube bearing a mouthpiece is attached. Beyond the bulb the figure 101 is etched on the RBC pipette and 11 on the WBC pipette (Figs 3.4 and 3.5).

Fig. 3.4: Diluting pipettes

Fig. 3.5: WBC and RBC pipettes
Filling the Pipette

Under aseptic precautions, the finger is pricked and the first 2–3 drops are wiped out following which a good sized drop of free flowing blood is made to form. Holding the mouthpiece between the lips and keeping the pipette horizontal its tip is placed within the edge of the blood drop. Blood is drawn into the pipette by gently sucking in the mouthpiece up to the 0.5 or 1.0 mark depending on the dilution required. The other end of the pipette is tightly closed by clenching the rubber tube. The outside of the pipette tip is wiped out (be careful not to touch the tip itself) with cotton and immersed in the diluting fluid while holding. The diluting fluid is sucked up to the 11 mark in case of the WBC pipette or 101 mark in case of the RBC pipette. Dilution of the blood must be done quickly otherwise blood is likely to clot in the stem. Following dilution, the pipette is kept horizontal so that the fluid does not run out. Now, by placing the pipette tip against the palm of the left hand and holding it just above the bulb, with the thumb and forefinger of the other hand, it is rotated to and fro for 3–4 minutes to allow the mixing of the blood with the diluting fluid.

Following this, the counting chamber is charged by discarding the stem fluid (about two drops) and placing the pipette at the edge of the cover slip that lies over counting chamber.

PRECAUTIONS

1. The pipette must be dry and free from clotted blood and the bead must roll freely.
2. The pipette must be almost horizontal while filling it with blood. The tip must not press against the subject’s skin or be lifted out of the blood drop (otherwise air will enter it).
3. The dilution of blood must begin immediately after the blood is drawn or else the blood may clot.
4. Leakage of fluid from the pipette must be avoided while mixing the bulb contents.
5. After charging the hemocytometer, the pipette must be washed in running water following which alcohol is used to remove water. Finally, the pipette is washed with acetone to remove the alcohol and left to dry.

### OSPE-I

**Procedure: Filling the RBC and WBC pipette**

<table>
<thead>
<tr>
<th>Step</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prick the finger using all aseptic precautions</td>
<td></td>
</tr>
<tr>
<td>Hold the mouthpiece of the pipette in your mouth and fill the pipette up to mark 0.5 with the blood drop formed at your fingertip</td>
<td></td>
</tr>
<tr>
<td>Wipeout the tip of the pipette</td>
<td></td>
</tr>
<tr>
<td>Fill the pipette with the given diluting fluid up to mark 101/11 mark</td>
<td></td>
</tr>
<tr>
<td>Roll the pipette horizontally in your palm to mix the contents thoroughly</td>
<td></td>
</tr>
</tbody>
</table>
**Hemocytometer**

**OSPE-II**

<table>
<thead>
<tr>
<th>Procedure: Charging the Neubauer's chamber for RBC and WBC count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Place the Neubauer's chamber horizontally on the table and cover it with cover slip</td>
</tr>
<tr>
<td>Discard two drops of fluid from the given pipette</td>
</tr>
<tr>
<td>Hold the pipette at 45° angle to the edge of the cover slip</td>
</tr>
<tr>
<td>Form a drop at the tip of the pipette and touch the drop to the edge of the cover slip</td>
</tr>
<tr>
<td>Avoid under/overcharging of the chamber</td>
</tr>
</tbody>
</table>

**QUESTIONS**

1. What are the uses of hemocytometer in hematology?
2. How do you identify the RBC and WBC pipette?
3. What are the steps for diluting the blood for various cell counts?
4. What are the precautions taken for pipetting?
5. Why the fluid is not sucked directly from the stock bottle?
**Experiment 4**

**AIM**
To enumerate the number of RBCs per cubic millimeter of blood.

**APPARATUS REQUIRED**
Hemocytometer, microscope, cover slips, Hayem’s fluid, cotton, spirit, and lancet.

**Composition of Hayem’s Fluid**
- Sodium chloride - 0.5 g to maintain isotonicity
- Sodium sulfate - 2.5 g to prevent Rouleaux formation
- Mercuric chloride - 0.25 g to fix the cells and act as a preservative.
- Distilled water - 100 mL

**PRINCIPLE**
The number of red cells in the blood is too many and the size of the cells is too small. It is, therefore, not possible to count the cells even under high power. This difficulty is overcome by diluting the blood to a known degree.

**PROCEDURE**
Collection and dilution of blood: The RBC pipette is cleaned with distilled water, alcohol and finally, with ether and dried. Sterilize the tip of the middle finger of left hand. A prick is made...
in the finger to a depth of 3–4 mm with a sterile lancet. The pricking should be done only in the middle or ring finger. Never use the thumb or index finger because the tendon is superficial. Blood is drawn up to 0.5 unit followed by the diluting fluid up to 101 units without any air bubble. Mixing is done by rotating the pipette between the palms. After mixing the blood with diluting fluid, keep it aside for 3 minutes. Discard the fluid from the stem of the pipette. Keeping it at 30°–45°, charge the fluid from the bulb between the cover slip and the central platform, so that it spreads uniformly. Care is taken not to induce any air bubbles. Wait for 2–3 minutes for the cells to settle down.

**Microscopic Adjustments**

1. The cells are focused under low power, and ultimately, the counting is done under high power.
2. Concave mirror to be used.
3. Condenser to be lowered.
4. Diaphragm is adjusted for adequate entry of light. Focus the RBC rulings under low power. Place the cover slip over the raised ridges.

**Counting the Cells**

Cells are to be counted from four corners and one central medium square of the RBC rulings. Each medium square is made up of 16 smaller squares. While counting, the cells lying inside as well as on the upper and left border of the smallest square are to be taken into account. In total, the cells lying in the 80 smallest squares have to be counted (Figs 4.1 and 4.2).

![Fig. 4.1: Rule of counting](image-url)
CALCULATION

- Area of 1 medium square: $\frac{1}{25} \text{ mm}^2 (\frac{1}{5} \times \frac{1}{5})$
- Depth of the chamber: $\frac{1}{10} \text{ mm}$
- Volume of the fluid over 1 medium square: $\frac{1}{25} \times \frac{1}{10}$
  $= \frac{1}{250} \text{ mm}^3$
- Volume of fluid present over 5 medium squares:
  $= \frac{1}{250} \times 5$
  $= \frac{1}{50} \text{ mm}^3$
- Number of RBC counted in $\frac{1}{50} \text{ mm}^3$ of diluted blood: $N$
- Number of RBC in $1 \text{ mm}^3$ of diluted blood: $N \times 50$
- Dilution factor: 1:200
- Number of RBC in $1 \text{ mm}^3$ of undiluted blood:
  $= N \times 50 \times 200$
  $= 10000 N$

PRECAUTIONS

1. Finger should not be squeezed once pricked, because the interstitial fluid will mix with blood and give wrong value.
2. As soon as the blood is drawn into the pipette, it should be immediately diluted with the RBC diluting fluid to prevent clotting.
3. There should not be any air bubbles in the pipette when blood or diluting fluid is sucked.
4. Do not tilt the microscope.
5. Cover slip should be placed uniformly over the ridges.
6. During charging care should be taken to prevent fluid from entering into the trenches.
DISCUSSION

Normal Value

Males: 4.5–6.0 million/cu mm  
Female: 4.0–5.5 million/cu mm

Physiological Variations

1. Diurnal variation: Variation of about 5% may occur per period of 24 hours. The count is least during sleep and maximum during evening.
3. Altitudes: RBC count increases with increase in altitude.
4. High external temperature: RBC count increases with increase of temperature.
5. RBC count also depends on the age and sex of a person.

Pathological Variations

1. Anemia: It means a significant reduction in number of red cells and/or the amount of hemoglobin in the blood as compared to the normal state.
2. Polycythemia: An increase in the number of red cells is called polycythemia. The blood count may be 6–7 millions/cu mm. It may be due to:
   a. Emphysema.
   b. Congenital heart disease.
   c. Poisoning by chemicals like arsenic and phosphorus.
   d. Repeated small hemorrhages.
   e. High altitudes.
3. Polycythemia vera: It is a primary condition due to alteration in the hemocytoblastic cell. RBC count may be 7–8 millions/cu mm.

RESULT

The RBC count of the given subject is .......... million cells/cu mm.

OSPE

Procedure: Perform RBC count of your own blood

<table>
<thead>
<tr>
<th>Procedure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Focus the uncharged Neubauer’s chamber into low power of the microscope</td>
</tr>
<tr>
<td>Prick the finger and collect the blood in to the RBC pipette up to mark 0.5</td>
</tr>
<tr>
<td>Fill the pipette with the diluting fluid up to mark 101</td>
</tr>
<tr>
<td>Roll the pipette horizontally on your palm for about 2 minutes</td>
</tr>
<tr>
<td>Take out the Neubauer’s chamber from the microscope, without disturbing the focus, and charge it</td>
</tr>
<tr>
<td>Wait for about 5 minutes. Draw the RBC counting squares while waiting</td>
</tr>
<tr>
<td>Put the charged Neubauer’s chamber into the microscope, without disturbing the focus, and see a clear image using fine adjustment only</td>
</tr>
<tr>
<td>Rotate the high power into use and see a clear image using fine adjustment only</td>
</tr>
<tr>
<td>Count the RBCs into 5 RBC squares, observing the ‘rule of counting’</td>
</tr>
<tr>
<td>Calculate total RBCs counted into 5 RBC squares (X)</td>
</tr>
<tr>
<td>Calculate total RBC count of your own blood as X x 10,000 cells/cu mm</td>
</tr>
</tbody>
</table>
QUESTIONS

1. During dilution, blood is drawn up to the 0.5 mark and diluting fluid up to the 101 mark. Yet why do we say at the dilution is 1: 200? (and not 1: 202).
2. While mixing the fluids, why should you NEVER shake the pipette in the direction of its long axis?
3. Why is it important to discard the first few drops from the pipette before charging the counting chamber?
4. What is the normal range of erythrocyte count?
5. What are the physiological variations in RBC count?
6. Explain the terms “anemia” and “polycythemia” and classify in brief their various types. Describe the terms “physiological anemia of pregnancy” and “reticulocyte response.”
7. What is the most powerful stimulus for RBC production?
8. What is the normal size of RBC. Name one condition when RBC size is less than normal and one condition where the RBC size is more than normal?
9. What will happen if the RBC pipette is not dry before the experiment?
10. What is polycythemia vera rubra?
11. What is the composition and function of Hayem’s fluid?
12. What happens to leukocytes when blood is mixed with diluting fluid?
13. What is the morphology of RBC and why?
14. What is Rouleaux formation?
15. Mention the life span of RBC.
16. Enumerate the functions of RBC
AIM

To estimate the amount of hemoglobin in 100 mL of blood by Sahli’s method.

PRINCIPLE

The hemoglobin present in a measured sample of blood is converted by dilute HCl to acid hematin, which is brown in color. The intensity of this color depends on the amount of acid hematin, which in turn depends on the hemoglobin. The color of this solution is compared with that of a standard by direct vision.

APPARATUS REQUIRED

Sahli’s hemoglobinometer (Hemometer): This consists of (Figs 5.1A and B):

1. Comparator—a rectangular plastic box with a slot which accommodates the Hb tube and non-fading standard fixed on either side in front of an opaque white glass.
2. Hemoglobin tube—graduated in g % (2–24 g %) on one side and in percentage (20–140%) on the other.
3. Hemoglobin pipette with a 20 cu mm mark and rubber tubing with a mouthpiece.
4. A glass rod to stir.
5. N/10 hydrochloric acid.
6. Distilled water.
7. Lancet, cotton, and spirit.
PROCEDURE

N/10 HCl is taken up to the lowest mark in the graduated tube. The finger is pricked under aseptic precautions. The pipette is filled with blood up to 20 cu mm mark taking care not to allow air bubbles to enter. After wiping the blood sticking to the outside of the pipette, its tip is dipped in the acid and the blood is expelled. The pipette is rinsed 3 or 4 times with the acid solution, till all the blood is washed out. The blood and the acid are mixed well with the glass rod. The tube is allowed to stand for 8–10 min. for the formation of acid hematin. Distilled water is added drop by drop, mixed well and the color is compared with that of the standard. Care should be taken to remove the glass rod while matching and not to view through the graduations. This is continued till the color matches. The lowest point of the meniscus is read, which directly gives the hemoglobin concentration in 100 mL of blood.

PRECAUTIONS

1. All precautions mentioned for collection of blood and filling the pipette should be observed.
2. Enough time should be allowed for conversion of all the Hb into acid hematin.
3. The solution must be properly mixed.
4. Color matching should not be delayed and should be done in bright light.

Disadvantages of Sahli’s Method

1. As acid hematin is not in true solution some turbidity may result.
2. The method estimates only oxy Hb and reduced Hb.
3. The color of the glass standards may fade over time.
Estimation of Hemoglobin

Advantages of Sahli’s Method

1. Easy to perform.
2. Economical.
3. Less time consuming.

RESULT

The hemoglobin content of the given subject is.......... gm/dL.

QUESTIONS

1. What is the normal range of the hemoglobin in blood?
2. Record the Hb content of your blood. Is it normal?
3. Comment on the results of your group. Are there any abnormal values? Are there any attributable causes for the above mentioned?
4. Name the other methods to estimate hemoglobin?
5. What is the oxygen carrying capacity of the blood?
6. What is the color in the hemoglobinometer due to?
7. What are the functions of N/10 HCl in this experiment?
8. What are the advantages of doing red cell indices?
9. What is a picogram?
10. What is the structure of hemoglobin? What is it made up of?
11. Discuss the physiological factors which influence the Hb content of blood.
12. Define anemia.
13. List 4 pathological conditions where the Hb content is altered.
AIM
To find out the packed cell volume (PCV) and then, to calculate the following blood indices:
1. Color index
2. Mean corpuscular volume (MCV).
3. Mean corpuscular haemoglobin (MCH).
4. Mean corpuscular hemoglobin concentration (MCHC).

APPARATUS REQUIRED
Centrifuge machine, hematocrit tube (Wintrobe’s), Pasteur’s pipette, syringe with needle, 3.8% sodium citrate solution, etc.

PRINCIPLE
When blood mixed with an anticoagulant is centrifuged in a hematocrit tube, the corpuscles settle at the bottom and the ratio of packed red cells to that of whole volume in the hematocrit tube is called packed cell volume or hematocrit volume of blood.

PROCEDURE
1. Macro method—Wintrobe’s method.
Macro method—Wintrobe’s method (this is followed in lab): To determine the packed cell volume, in a clean dry syringe 2 mL of blood is drawn from the antecubital vein and mixed with 0.4 mL of 3.8% sodium citrate solution. The sodium citrate acts as the anticoagulant. The blood is then filled in the hematocrit tube up to the mark ‘0’ using a Pasteur’s pipette. The tube is centrifuged at the rate of 3,000 revolutions per minute for a period of 30 minutes. The packed cell volume is read in percentage directly as the column of blood in hematocrit tube is 100 cu mm (Fig. 6.1).

Normal values  Average  Range
Adult men  45%  40–50%
Adult women  42%  37–47%

RESULT

PCV of the given blood sample is ............%  

CALCULATION OF BLOOD INDICES

Following values are referred as RBC indices or hematological indices.
1. Mean corpuscular volume (MCV).
2. Mean corpuscular hemoglobin (MCH).
3. Mean corpuscular hemoglobin concentration (MCHC).

MCV, MCH, and MCHC are referred as absolute values. These values are widely used for the classification of anemia.

To calculate the blood indices, the RBC count and hemoglobin content are essential. For RBC count and hemoglobin content, the values are taken from the results of experiment number 4 and 5 respectively.

These are the value of my blood:

PCV =
RBC count =
Hemoglobin =
1. **Mean corpuscular volume (MCV):** It is the volume of an average RBC.

\[
MCV = \frac{PCV \text{ (in percentage)}}{\text{RBC Count (in millions/cm}^3) \times 10}
\]

**Normal MCV = 78–96 fl (femtoliters).**

In microcytic anemia (iron deficiency) MCV is low. In macrocytic (megaloblastic) anemia MCV is increased.

2. **Mean corpuscular hemoglobin (MCH):** Amount of hemoglobin that is present in an average RBC.

\[
MCH = \frac{Hb \text{ (in g%)}}{\text{RBC Count (in million/cm}^3) \times 10}
\]

**Normal MCH = 27–33 pg (picogram).** Cells with this value are considered as normochromic.

In iron deficiency anemia—MCH is low (hypochromic).

In megaloblastic anemia—MCH is high (not hyperchromic, why?)

3. **Mean corpuscular hemoglobin concentration (MCHC):** Amount of Hb present in 100 mL of RBCs.

\[
MCHC = \frac{Hb \text{ (in g%)}}{\text{PCB (in %)}} \times 100
\]

**Normal MCHC = 30–37 g % (or g/dL).**

MCHC denotes degree of saturation of Hb in cells. It is the more reliable index than other indices as it does not involve RBC count for its calculation.

4. **Color index:** Ratio of percentage of Hb to percentage of RBC is called as color index. 14.8 g% of hemoglobin is considered as 100% and 5 million/cubic mm of RBCs are considered as 100%. Color index of an individual with hemoglobin 14.8 gm% and RBC 5 million/cu mm will be 1.

\[
\text{Color index} = \frac{Hb \%}{RBC \%}
\]

where, 
\[
Hb\% = \frac{\text{Observed Hb}}{\text{Normal Hb (i.e. 14.8 gm %)}} \times 100
\]

\[
\text{RBC\%} = \frac{\text{Observed RBC count}}{\text{Normal RBC count (i.e. 5 million/cu mm)}} \times 100
\]

**Normal color index = 0.85–1.1**

Color index is useful to know the type of anemia. Low index indicates hypochromic anemia.

**QUESTIONS**

1. How do you do this experiment by micro hematocrit method?
2. Draw a diagram of the Wintrobe's hematocrit tube showing PCV.
3. Give the significance of PCV.
4. List the physiological conditions where the PCV is increased/ decreased.
5. What is the ideal anticoagulant to be used for PCV determination and why?
6. What are the uses of estimation of PCV?
7. Is there any change in PCV in arterial blood and venous blood?
8. What are the factors which influence PCV?
9. What is true hematocrit?
10. If you are given 1 mL of anticogulated blood to do only one investigation of maximum use, which should it be?
11. Classify anemia based on blood indices.
12. Why MCHC does have an upper physiological limit?
13. What is the practical use of red cell indices in clinical hematology?
AIM

To find out the erythrocyte sedimentation rate (ESR) of the given sample of blood.

PRINCIPLE

If blood containing an anticoagulant is allowed to stand in a tube placed vertically, the red cells settle down gradually to the bottom, since their specific gravity is greater than that of plasma. The rate at which the red cells settle down is noted.

METHODS

Two methods are used to determine ESR:
1. Westergren’s method
2. Wintrobe’s method.

WESTERGREN’S METHOD

Apparatus Required

A clean dry Westergren’s tube, stand for the Westergren's tube, sterile 3.8% sodium citrate solution.

Westergren's tube is graduated from 0–200, 30 cm in length with an internal bore of 2.5 mm. It is open at both ends (Figs 7.1A and B).
Determination of Erythrocyte Sedimentation Rate

**Procedure**

About 1.6 mL blood from the antecubital vein is diluted with 0.4 mL of 3.8% sodium citrate solution. It is drawn into the Westergren’s tube up to zero mark and fixed vertically in the Westergren’s stand. The cells settle down and the upper level of the cells is noted at the end of 1/2 hour and 1 hour.

**Normal Values**

Male = 3–5 mm after 1 hour  
Female and children = 5–12 mm after 1 hour

**WINTROBE’S METHOD**

**Apparatus Required**

1. A clean dry Wintrobe’s hematocrit tube. It is graduated in 1 mm up to 100 mm and is open at the top and closed at the bottom. Its length is 11 cm.
2. Wintrobe’s hematocrit tube stand.
3. A test tube containing 2 mg of solid potassium oxalate and 3 mg of solid ammonium oxalate
4. Pasteur’s pipette
5. Spirit, sterile 2 mL syringe with needle, cotton and tourniquet.
Procedure
Blood drawn from the anticubital vein is mixed with anticoagulant and filled in the Wintrobe's tube up to zero mark with the help of Pasteur's pipette. The tube is placed vertically in the stand. The upper level of cells is noted at the end of 1/2 and 1 hour.

Normal Values
Males—0–9 mm after 1 hour
Females and Children—0–20 mm after 1 hour.

Precautions
1. The tube should be kept perfectly vertical.
2. Avoid air bubbles in the column of blood.
3. Blood should be used within 2 hours.
4. Blood showing any signs of coagulation or hemolysis should be rejected.
5. The test should be carried out at room temperature.
6. Avoid exposure to direct sunlight.
7. If excess of anticoagulant is used, it will alter the ESR value.

DISCUSSION
Erythrocyte Sedimentation Rate
This test measures the rate at which red cells settle spontaneously in a standing column of blood. This sedimentation process is divided into 3 stages for convenience.
1. When, a column of blood stands undisturbed, there is a tendency of red cells to aggregate together in a peculiar ‘pile of coins’ arrangement called Rouleaux. This is the stage of aggregation.
2. The aggregates get heavy and begin to fall to the bottom of the suspension. This is the stage of sedimentation called the stage of fall.
3. The final phase is the stage of packing with the cells and aggregates low down due to increased cell concentration, crowding and packing.

FACTORS WHICH INFLUENCE ERYTHROCYTE SEDIMENTATION RATE
1. Plasma proteins: This affects the stage of Rouleaux formation. Fibrinogen and globulin increase Rouleaux formation opposite to albumin which has a retarding effect. When rouleaux formation increases, ESR increases.
   In conditions where there is an increase in fibrinogen or globulin, ESR is increased. Examples are chronic infections like TB and systemic inflammatory disorders like rheumatoid arthritis. All these conditions have tissue injury and immune reactions, both, leading to high levels of fibrinogen and globulins (All antibodies are globulins).
2. **Red cell count:** The total number of red cells also affects ESR; but not to such an extent as the plasma proteins. In anemia, there is more space between the red cells and it is easier for them to fall and for the displaced plasma to rise than when the cells are relatively more crowded in conditions like polycythemia. Therefore, an ESR in a severely anemic person should be given guarded consideration because, anemia will increase ESR more.

**SIGNIFICANCE OF ERYTHROCYTE SEDIMENTATION RATE IN DISEASE**

1. The increase in erythrocyte sedimentation rate (ESR) is not diagnostic of any disease but it has only prognostic value.
2. It only indicates increase in fibrinogen/globulin levels, as would happen in any tissue injury.
3. The ESR is used to check the progress of the patient. As the patient improves, the ESR tends to fall. If the patient’s condition is getting worse, the ESR tends to rise.

**PHYSIOLOGICAL VARIATIONS**

Decreased: Very rare in newborn babies.

**PATHOLOGICAL VARIATIONS**

Increased: i. Rheumatic fever, ii. Tuberculosis, iii. Malignancy, iv. Anemia

**FACTORS AFFECTING ERYTHROCYTE SEDIMENTATION RATE**

1. Specific gravity: Corpuscles of ‘high’ specific gravity settle down in plasma of low specific gravity.
2. Lowered viscosity of plasma increases ESR.
3. Size of RBC: Increased RBC size leads to increased ESR.
4. Clumping of cells increases ESR.
5. Spherocytes reduces ESR due to prevention of Rouleaux formation.
6. Increase of temperature leads to increase in ESR.
7. High RBC count decreases ESR.
8. Increase in plasma lecithin decreases ESR.
9. Increase in fibrinogen increases ESR. Increase in albumin decreases it.

**RESULT**

The erythrocyte sedimentation rate of the given subject is___________
QUESTIONS

1. Mention some common infections and inflammatory disorders, which show an increase in ESR. What is the cause of this increase?
2. In multiple myeloma, what is the reason for an increased ESR?
3. What is the importance of monitoring the ESR in a patient with tuberculosis?
4. What is the only normal condition in which ESR is decreased?
5. Compare the effects of anemia and polycythemia on the ESR.
6. What are the methods by which ESR can be determined?
7. What are the factors affecting ESR?
8. What is the clinical significance of this investigation?
9. What is the normal ESR value in Westergren’s method?
10. What is the normal ESR value in Wintrobe’s method?
11. Mention some pathological conditions, which decrease ESR.
12. What is the advantage of using the double oxalate mixture in Wintrobe’s method? Can we use any other anticoagulants?
13. Why the normal values of ESR are more with Wintrobe’s method as compared to Westergren’s?
14. Why ESR reading is taken normally after one hour?
AIM
To enumerate the white blood cells in 1 mm$^3$ of blood.

PRINCIPLE
The blood is diluted with a diluting fluid, which destroys the red blood corpuscles and stains the nuclei of the white blood cells. The leukocytes are then counted in a hemocytometer and their number in undiluted blood is calculated.

APPARATUS REQUIRED
Microscope, hemocytometer, WBC pipette, Turk’s fluid, cover slip, disposable lancet, cotton and spirit.

Composition of Turk’s Fluid
Glacial acetic acid—1.5 mL (or) 2 mL
Gentian violet—1 mL
Distilled water—100 mL
Glacial acetic acid destroys RBC; gentian violet colors the nuclei of WBC.

PROCEDURE
1. **Filling the pipette and diluting the blood sample:** Clean the finger tip with spirit and prick it with a sterile lancet. The first drop of blood is discarded as it contains tissue fluid. Allow a good sized drop to form and draw blood up to 0.5 in the WBC pipette. Suck the
Turk’s fluid up to the mark 11. Hold the pipette horizontally and roll it between the palms to ensure thorough mixing.

2. **Charging the chamber:** Place the cover slip over the chamber. Place the chamber on the microscope stage. Roll the pipette again before charging. Discard the first two or three drops of fluid as the stem of the pipette is filled only with diluting fluid and has no blood at all. Allow a moderate size drop of diluted blood to form at the tip of the pipette. Hold the pipette at an angle of 45° and make the drop to touch the slide and the cover slip. The fluid will run into the capillary space to fill it. The fluid should not run into the trenches. There should be no air bubbles in the chamber. After charging, allow the fluid then to settle for two to three minutes before actual counting is begun.

3. **Counting the cells:** Focus the WBC area under low power and ensure that there is no air bubble. The cells lying on the upper horizontal and left vertical lines should be counted along with those lying within the square under consideration. Count the cells in 4 large corner squares and enter in the observation table (Fig. 8.1).

**PRECAUTIONS**

1. There should be no air bubble in the column of the blood.
2. The cells should be evenly distributed.

![Observation table for WBC count](Fig. 8.1)
**CALCULATION**

<table>
<thead>
<tr>
<th>Description</th>
<th>Formula</th>
</tr>
</thead>
<tbody>
<tr>
<td>Side of 1 WBC square</td>
<td>1 mm</td>
</tr>
<tr>
<td>Area of 1 WBC square</td>
<td>1 mm²</td>
</tr>
<tr>
<td>Depth of chamber</td>
<td>1/10 mm</td>
</tr>
<tr>
<td>Volume of fluid present over 1 WBC square</td>
<td>1/10 mm³</td>
</tr>
<tr>
<td>Volume of fluid present over 4 WBC square</td>
<td>4/10 mm³</td>
</tr>
<tr>
<td>No. of cells in 4/10 cu mm of diluted blood</td>
<td>N</td>
</tr>
<tr>
<td>No. of cells in 1 cu mm of diluted blood</td>
<td>10/4 N</td>
</tr>
<tr>
<td>Dilution factor</td>
<td>1:20</td>
</tr>
<tr>
<td>No. of cells in 1 cu mm of undiluted blood</td>
<td>10/4 × 20 N</td>
</tr>
<tr>
<td></td>
<td>50 N</td>
</tr>
</tbody>
</table>

**DISCUSSION**

Total leukocytes count shows variation under certain physiological and pathological conditions (Normal WBC count = 4000–11000 cell/cu mm).

**Leukocytosis (Increase in WBC Count)**

*Physiological Conditions*

1. Age:
   a. Newborn will always have an increased count.
   b. Childhood, pregnancy and delivery shows increased count.
   c. There is no significant change in old age compared to adult values.
2. Females during pregnancy and parturition.
3. Stress like severe exercise, severe pain, and excitation.
4. Diurnal variation: WBC count may vary from hour-to-hour (highest count in evening and lowest count in morning).
5. Digestive leukocytosis (after digestion).
6. Injection of adrenaline.

*Pathological Conditions*

1. Acute pyogenic infections (e.g. pneumonia, appendicitis and tonsillitis).
2. Leukemia (abnormal increase with immature cells) count may go up to 1,00,000 to 3,00,000 per cu mm.
3. Acute hemorrhage.
4. Tissue damage resulting from burns, operations, myocardial infarction.
5. Malignant neoplasms.
6. Metabolic disorder (e.g. gout, diabetic acidosis).
Leukopenia (Decrease in WBC Count)

Physiological Condition

Rare but sometimes due to:
1. Exposure to cold
2. Aspirin.

Pathological Condition

1. Infection—Typhoid
2. Starvation—Malnutrition.
3. Viral infections—Measles, chickenpox, influenza and rubella
4. Drugs—Antimetabolites, antimicrobials (Sulfonamides, chloramphenicol)
5. Hematological disorders—Aplastic anemia, pernicious anemia, irradiation.

RESULT

The WBC count of the given subject is .......... cells/cu mm.

QUESTIONS

1. Is this a routine test? What is the usefulness of this test?
2. What other test has to be done with total count to make it more meaningful?
3. Why should you destroy the RBCs while doing WBC count?
4. What is the normal range of total WBC count? List three important physiological variations?
Differential Count of White Blood Cells

EXPERIMENT 9

DATE: ...........

AIM

To enumerate the different types of white blood cells in the given subject.

APPARATUS REQUIRED

Microscope, clean dry grease free glass slides with even edges, a drop bottle containing distilled water, a drop bottle containing Leishman’s stain, cedar wood oil, staining rack, lancet, cotton and spirit.

Composition of Leishman’s Stain

<table>
<thead>
<tr>
<th>Component</th>
<th>Function</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eosin</td>
<td>An acidic dye, which stains the basic protoplasmic material.</td>
</tr>
<tr>
<td>Methylene blue</td>
<td>A basic dye, which stains the acidic nuclear chromatin.</td>
</tr>
<tr>
<td>Acetone-free methyl alcohol</td>
<td>To fix the smear to the slide. Acetone being a strong lipid solvent it tends to damage the cell membrane. The stain is prepared by dissolving 750 mg of Leishman’s powder in 500 mL of acetone-free methyl alcohol.</td>
</tr>
</tbody>
</table>

PROCEDURE FOR THE PREPARATION OF BLOOD FILM

The finger is cleaned and pricked with a sterile lancet. The first drop of blood is discarded. A clean glass slide is touched to the newly formed drop of blood 1 cm away from the edge. The slide is placed on the table so that the blood drop is on the right side. It is supported with the left hand and the second slide (spreader) is held along its long edges with the right hand at an angle of 45°. The smooth narrow edge of the spreader slide is placed in front of the blood drop
and drawn back to touch the drop which then spreads along the edge of the spreader slide evenly. The spreader slide is pushed towards the left by a quick uniform motion with a light but even pressure. The blood follows along the spreader slide to form a blood film. It is dried quickly by waving in the air (Figs 9.1 and 9.2).

![Preparation of blood smear](image)

**Fig. 9.1:** Preparation of blood smear

![Diagram showing (A) Good and (B to D) Bad smears](image)

**Fig. 9.2:** Diagram showing (A) Good and (B to D) Bad smears
Differential Count of White Blood Cells

STAINING THE BLOOD FILM

The blood smear is placed on the staining rack. Leishman’s stain is added drop by drop till the entire film is covered by the stain. The number of drops added is counted (usually 8–10 drops). The undiluted stain is allowed to act for 2 minutes. It should not be allowed to dry up. The cells are fixed in these 2 minutes. Then, double the number of distilled water drops is added to dilute the stain. It is mixed by gently blowing through a pipette. The actual staining occurs now only. The stain should not be allowed to dry. After 7 minutes the stain is drained off. The slide is washed in a gentle stream of running tap water until the film turns pink. The slide is kept in a vertical position to drain and dry. Thick uneven smears should be discarded. It is preferable to make 3–4 smears. The best stained film should be selected for microscopic examination.

A good smear should:
1. Be buff colored.
2. Be uniform.
3. Be broader at the head and taper off into a tail.
4. Occupy the middle-third of the slide leaving a margin of about 5 mm along the edges.
5. Have no longitudinal or transverse striations or windows.
6. Have no stained granules or precipitates.
7. Have discrete red cells without overlapping each other.

MICROSCOPIC EXAMINATION

The microscope is adjusted for oil immersion lens. The condenser is raised, the diaphragm is completely opened and the plane mirror is used. Two drops of cedar wood oil are placed near the head end. The oil immersion objective lens is made to touch the oil by viewing from the side. The fine adjustment screw is adjusted till the cells are brought into focus.

Hundred squares are drawn for recording the cell count. The white blood cells are identified and entered using the letter N for neutrophil, E for eosinophil, B for basophil, L for lymphocyte and M for monocyte. The slide is slowly moved towards the tail end and the cells are counted. The slide is then shifted up and moved in the opposite direction. This pattern of movement of the slide (zigzag pattern) takes into consideration all the parts of the film and ensures that a cell is not counted more than once.

Hundred white cells are identified and entered. The number of each type of white blood cell is counted and expressed in percentage.

IDENTIFICATION OF THE CELLS

A leukocyte is identified from its size, the nucleus and the cytoplasm.

Size

The size of the white cell is assessed by comparing it to that of the red cells if the white cell is as big as the red cell, it probably is a small lymphocyte. If the cell is twice as big as the red cell, it may be a granulocyte or a large lymphocyte. If the cell is about 2.1/2 to 3 times, then it is possibly a monocyte.
**Nucleus**

The following characters of the nucleus must be observed:

1. Whether it appears as a single mass or lobed
2. If single, whether the shape is round, oval or kidney shaped
3. Whether partially or completely fills the cell

   A small cell with a single round or oval nucleus may be a small lymphocyte. A bigger cell with distinct nuclear lobes joined by chromatin strands should be a granulocyte. A big cell with a kidney shaped nucleus is a monocyte. The nucleus in a lymphocyte completely fills the cell.

**Cytoplasm**

1. The amount of cytoplasm in relation to the size of the nucleus is noted.
2. The presence of visible granules and their nature is observed.

   A small cell with a thin crescent of cytoplasm is a small lymphocyte. A bigger cell with a rim of cytoplasm all around is a large lymphocyte. A big cell with large amount of clear cytoplasm in relation to the nucleus is a monocyte (Fig. 9.3).

   If a granulocyte has fine neutral granules of light violet color, the cell is a neutrophil. If the granules are coarse and orange or red colored, the cell is an eosinophil. In a basophil, the granules are large, coarse and deep blue in color. It is the smallest of granulocyte.
### Differential Count of White Blood Cells

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Size</th>
<th>Nucleus</th>
<th>Cytoplasm</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Granulocytes</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Eosinophil</td>
<td>10–14 µm</td>
<td>Bilobed connected by a chromatin strand.</td>
<td>Coarse orange to brick red granules.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(Spectacle shaped)</td>
<td></td>
</tr>
<tr>
<td>3. Basophil</td>
<td>10–14 µm</td>
<td>Irregular may be bilobed or S shaped.</td>
<td>Coarse deep blue granules completely filling the cell.</td>
</tr>
</tbody>
</table>

| **Agranulocytes**|          |                                              |                                                     |
|------------------|----------|----------------------------------------------|                                                     |
| 1. Lymphocyte    |          |                                              |                                                     |
| a. Small         | 8–10 µm  | Large-round nucleus completely, filling the cell, stains deep blue | Thin crescent of pale blue cytoplasm. No granules |
| b. Large         | 10–15 µm | Large round or indented nucleus, stains deep blue | Thin rim of pale blue cytoplasm all around. No granules seen |
| 2. Monocyte      | 12–24 µm | Large, placed centrally, kidney-shaped nucleus | Large amount of pale greyish blue cytoplasm. No granules seen |

### RESULTS

- Neutrophils – %
- Eosinophils – %
- Basophils – %
- Lymphocytes – %
- Monocytes – %

### DISCUSSION

The percentage of the different types of white blood cells is called the differential count. It is done to find out if there is an increase or decrease in a particular type of leukocyte. But it shows only a relative increase or decrease of the cell type with a corresponding change in the other cell types. Absolute values are more significant than relative values.

### NORMAL COUNT

- Neutrophils – 50–70%
- Eosinophils – 1–4%
- Basophils – 0–1%
- Lymphocytes – 20–40%
- Monocytes – 2–8%
VARIATIONS

Neutrophilia: Is an increase in the number of neutrophils
1. Acute pyogenic infection such as tonsillitis, appendicitis, pneumonia
2. Tissue necrosis as in myocardial infarction
3. Following hemorrhage
4. Trauma, postoperative burns
5. Hemolysis
6. Malignant neoplasm
7. Metabolic disorders like gout, diabetic acidosis, uremia.
8. Drugs such as glucocorticoids, adrenaline, digitalis, phenacetin

Neutropenia: Is decrease in the number of neutrophils.
1. Typhoid and paratyphoid fever, kala-azar
2. Viral infection
3. Depression of bone marrow due to irradiation
4. Drugs such as chloramphenicol
5. Autoimmune diseases.

Eosinophilia: Increase in the number of eosinophils.
1. Allergic conditions like asthma, hay fever, urticaria
2. Parasitic infestations—such as trichinosis, schistosomiasis, hookworm infestation
3. Tropical eosinophilia
4. Skin diseases like eczema
5. Loeffler’s syndrome
6. Collagen disorders
7. Chronic myeloid leukemia

Eosinopenia: Decrease in the number of eosinophils.
1. Administration of ACTH and glucocorticoids
2. Stress
3. Cushing’s syndrome

Basophilia: Increase in the number of basophils.
1. Viral infections such as small pox and chickenpox
2. Allergic diseases
3. Chronic myeloid leukemia and polycythemia vera.

Basopenia: Decrease in the number of basophils.
1. Acute pyogenic infection
**Monocytosis:** Increase in the number of monocytes.
1. Chronic infection like tuberculosis, syphilis, subacute bacterial endocarditis, brucellosis
2. Protozoal infections like malaria, kala-azar
3. Infectious mononucleosis
4. Monocytic leukemia and multiple myeloma
5. Collagen diseases

**Monocytopenia:** Decrease in the number of monocytes occurs rarely in:
1. Bone marrow failure
2. Aplastic anemia

**Lymphocytosis:** Increase in the number of lymphocytes.
1. Infants and young children (relative lymphocytosis)
2. Whooping cough, diphtheria
3. Chronic infections like TB, syphilis and malaria
4. Viral infection like chicken pox
5. Lymphocytic leukemia
6. Autoimmune diseases.

**Lymphocytopenia:** Decrease in the number of lymphocytes.
1. Steroid therapy
2. Acute infections and illnesses.
3. Hodgkin’s disease

### OSPE-I

**Procedure:** Prepare a neat smear with your own blood

- Select three clean slides for smear and a fourth slide with smooth edges as a spreader
- Prick your own finger with all aseptic precautions and put one drop of blood on each of the three slides 2–3 mm away from the edge
- Take a spreader and hold it at an angle of 45° in front of the blood drop
- Draw the spreader back towards blood drop till it touches the drop
- Push the spreader towards left with uniform motion and light pressure

### OSPE-II

**Procedure:** Stain the given smear for DLC

- Put all the smears on two parallel glass rods kept horizontally
- Cover the smears with the Leishman’s stain. Count the number of drops
- Wait for about 2 minutes
- Add double the number of drops of distilled water to the smear
- Allow uniform mixing of stain with distilled water by gentle blowing
- Wait for about 8–10 minutes
- Wash the slide by dipping in a beaker of plain water
- Allow it to dry by itself
QUESTIONS

1. What is the purpose of methyl alcohol in Leishman's stain?
2. What is the pH of the buffered water that is used?
3. Give the normal differential count. What are the physiological variations?
4. Explain the terms (a) neutrophilia and (b) neutropenia and list conditions where it occurs.
5. What are the terms which refer to (a) increase and (b) decrease in the lymphocyte count?
6. Name two conditions in which each occurs.
7. What is monocytosis? When does it occur?
8. What is the composition of Leishman's stain? What are the functions of each component?
9. Name some conditions where eosinophils and neutrophils increase in number.
10. Which WBC has got the phagocytic function?
11. What is the function of lymphocytes in the body?
12. What are the special features of monocytes? How will you differentiate monocytes from large lymphocytes?
13. How do you differentiate a bilobed eosinophil from a neutrophil?
14. What are microscopic adjustments you will do before starting the experiment?
15. Why methyl alcohol should be acetone free?
AIM

To estimate the absolute eosinophil count of the given sample of blood.

APPARATUS REQUIRED

Hemocytometer, microscope, cotton, spirit, sterile needle or lancet, Pilot’s fluid.

PILOT’S FLUID

- Phloxine 1% aqueous – 10 mL
- Propylene glycol – 50 mL
- Distilled water – 40 mL
- Sodium carbonate 10% – 1 mL

Phloxine stains the granules of the eosinophils. Propylene glycol and distilled water lyses the RBCs. Sodium carbonate accelerates staining.

DUNGER’S FLUID

- Eosin – 0.01 g
- Acetone – 10 mL
- Distilled water to make – up to 100 mL

May also be used to stain the eosinophil. It contains:

This Dunger’s fluid is used in this experiment (Fig. 10.1).
PROCEDURE

After sterilizing the finger, obtain a drop of blood by finger puncture. Draw blood up to mark 1 in the WBC pipette and immediately immerse into the diluting fluid and draw the fluid up to mark 11. Mix the fluid well by rolling the pipette in the palm. Keep it aside for 15 minutes for adequate staining of the cells.

Discard the fluid in the stem of the pipette and charge the counting chamber. Count the cells (eosinophils alone are stained) in all 9 large squares.

CALCULATION

<table>
<thead>
<tr>
<th>Calculation</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Side of one large square</td>
<td>1 mm</td>
</tr>
<tr>
<td>Area of one large square</td>
<td>1 × 1 = 1 mm²</td>
</tr>
<tr>
<td>Depth of chamber</td>
<td>1/10 mm</td>
</tr>
<tr>
<td>Volume over one large square</td>
<td>1/10 mm³</td>
</tr>
<tr>
<td>Volume over 9 large squares</td>
<td>9/10 mm³</td>
</tr>
<tr>
<td>Number of cells in 9/10 mm³ of diluted blood</td>
<td>N</td>
</tr>
<tr>
<td>Number of cells in 1 mm³ of diluted blood</td>
<td>10/9 N</td>
</tr>
<tr>
<td>Dilution factor</td>
<td>1:10</td>
</tr>
<tr>
<td>Number of cells in 1 mm³ of undiluted blood</td>
<td>10/9 × 10 N</td>
</tr>
<tr>
<td></td>
<td>100/9 N</td>
</tr>
</tbody>
</table>

PRECAUTIONS

Same as those observed for RBC and WBC count.

NORMAL VALUE

40–440 cells/mm³.
VARIATIONS

Eosinophilia: Increase in eosinophil count
1. Allergic Disorder :
   a. Bronchial asthma
   b. Hay fever
   c. Urticaria due to drug sensitivity especially sulfonamides and aspirin
2. Skin diseases, e.g. pemphigus
3. Parasitic infestations especially parasites that invade the tissues, e.g. trichinosis, echinococcus disease.
4. Tropical eosinophilia
5. Loeffler’s syndrome
6. Certain diseases of hemopoietic system, e.g. polycythemia vera, Hodgkin’s disease, pernicious anemia.
7. Following irradiation
8. Rheumatoid arthritis
9. Sarcoidosis
10. Idiopathic

Eosinopenia: Decrease in eosinophil count
1. Following corticosteroid therapy
2. Following ACTH injection
3. During neutrophilia

RESULT

The AEC count of the given subject is ........... cells/cu mm.

<table>
<thead>
<tr>
<th>OSPE</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Procedure:</strong> Estimate the absolute eosinophil count of your own blood</td>
</tr>
<tr>
<td>Focus the uncharged Neubauer’s chamber into low power of the microscope</td>
</tr>
<tr>
<td>With all aseptic precautions collect a sample of blood in WBC pipette up to mark 1</td>
</tr>
<tr>
<td>Immediately fill the pipette with diluting fluid up to mark 11</td>
</tr>
<tr>
<td>Mix the fluid by rolling the pipette in the palm</td>
</tr>
<tr>
<td>Cover the pipette with a wet filter paper and keep it aside for 15 minutes</td>
</tr>
<tr>
<td>Take out the Neubauer’s chamber from the microscope, without disturbing the focus</td>
</tr>
<tr>
<td>Discard first two drops of fluid in the pipette and charge the Neubauer’s chamber</td>
</tr>
<tr>
<td>Put the charged Neubauer’s chamber into the microscope, without disturbing the focus, and see a clear image using fine adjustment only</td>
</tr>
<tr>
<td>Count the eosinophils into all 9 large squares (X)</td>
</tr>
<tr>
<td>Calculate AEC of your own blood as X x 100/9 cells/cu mm</td>
</tr>
</tbody>
</table>
QUESTIONS

1. What is the normal total eosinophil count?
2. Mention the functions of eosinophils.
3. Name the conditions in which there is an increase in the absolute eosinophil count.
4. Name the hormone, which causes a lowering of eosinophil count.
5. What is the appearance of an eosinophil in a stained smear? What do the granules contain?
6. What is the significance of doing absolute eosinophil count?
7. What is major basic protein? What is its role?
8. Mention the composition of Dunger’s fluid.
9. With a diagram show the morphology of eosinophils.
10. What is the dilution used in eosinophil count?
11. What is the chemotactic factor for eosinophil?
12. How eosinophils combat infestation by worms?
13. Name the chemicals in the eosinophil granules.
Determination of Bleeding Time and Clotting Time

EXPERIMENT 11

DATE: ...........

BLEEDING TIME

AIM

To determine the bleeding time of the given subject by Duke's method.

APPARATUS REQUIRED

Filter paper, stop watch, lancet, cotton, spirit.

PROCEDURE

The tip of the left ring finger is pricked with aseptic precautions (3–4 mm). The blood should flow freely without squeezing. The time of puncture is noted. With a filter paper the blood is gently blotted every 30 seconds. The successive blots become smaller. This procedure is repeated until no blot appears on the filter paper. The time is noted again. The number of blots on the paper is counted. Number of blots × 30 seconds will be the bleeding time.

DISCUSSION

Bleeding time is the time interval between the skin puncture and the cessation of bleeding—the time in minutes, which it takes for a standardized skin wound to stop bleeding. 

Significance: Cessation of bleeding from a small wound as that inflicted during this procedure can be affected by vascular spasm and formation of platelet plugs. This test, therefore, measures the capillary and platelet functions in hemostasis.

Normal—1–5 minutes (by Duke's method)
The factors which affect the bleeding time are:
1. Size and nature of the injury
2. Condition of the vessel wall
3. Number of platelets.

Conditions where bleeding time is prolonged:
1. Decrease in the number of platelets (Thrombocytopenia).
2. Functional platelet defect.
   a. Drugs like aspirin, penicillin
   b. von Willebrand’s disease
   c. Uremia, cirrhosis, leukemia
3. Vessel wall defects.
   a. Prolonged treatment with corticosteroids
   b. Allergic purpuras
   c. Infections with hemolytic streptococci, bacterial endocarditis
   d. Deficiency of vitamin C
   e. Senile purpura.

OTHER METHODS
1. In Duke’s method the edge of the ear lobe can also be used.
2. Ivy's method: The cuff of the BP apparatus is applied to the upper arm and the pressure is raised to 40 mm Hg. The front of the forearm is used. The normal bleeding time by this method is up to 9 min.
3. The finger is pricked and dipped in a beaker containing normal saline at 37°C. The blood drops will be seen falling to the bottom of the beaker in a continuous slow stream. The time is noted when the bleeding stops.
4. Capillary fragility test of Hess or tourniquet test:
   The bleeding time may be performed by Duke’s method or Ivy’s method; Ivy’s method is a recommended method, but in the lab, we follow Duke’s method for the sake of convenience.

RESULT
The bleeding time of the given subject is...................

CLOTTING TIME

AIM
To determine the clotting time of the given subject by Wright’s method.

APPARATUS REQUIRED
Capillary glass tube 15 cm long with a bore of 0.8 mm, stop watch, lancet, cotton and spirit.
**PROCEDURE**

The tip of the left ring finger is pricked under aseptic precautions and the time of puncture is noted. The prick must be deep enough to allow free flow of blood without squeezing.

The blood is drawn into the capillary tube by dipping one end of the tube in the blood drop. The blood fills the tube by capillary action. After 2 minutes, a small bit of the tube is broken every 30 seconds until a fine thread of fibrin appears between the broken ends. The time is again noted and the interval between the prick and the appearance of fibrin thread gives the clotting time.

**DISCUSSION**

Clotting time is the time interval between the skin puncture and formation of fibrin thread. **Significance:** The clotting time is generally not affected by deficiency of platelets as only few platelets are required to provide enough platelet factor 3 for normal coagulation. This test assesses the intrinsic and common pathways of coagulation because the trigger for coagulation in this test is the surface activation of blood that comes into contact with the glass surface of the test tube (Therefore, for comparability, we should use standard size tubes and constant blood volumes so that the same amount of surface activation occur each time).

**Normal—2–8 minutes.**

**Conditions where clotting time is prolonged**

1. Hemophilia (Bleeding time is normal)
2. Christmas disease
3. Afibrinogenemia
4. Vitamin K deficiency
5. Liver disease
6. Anticoagulant therapy
7. Newborn baby.

**OTHER METHODS**

1. Lee and White method—1 mL of blood is placed in 8 mm diameter tube. The tube is tilted at definite time intervals until there is no displacement of blood, when the tube is inverted.
2. Drop method—A drop of blood is placed on a glass slide. Clotting is indicated by the absence of any change in the shape of the drop when the slide is held vertical.

Lee and White’s method is the most accurate and recommended method. But since it involves larger quantities of blood and venous sampling, this method will only be demonstrated to you. For individual exercise you will do the Wright’s capillary tube method.

**RESULT**

The clotting time of the given subject is....................
TESTS OF HEMOSTASIS

The first line tests:
1. Vascular integrity – Bleeding time
2. Platelet function – Bleeding time and platelet count
3. Coagulation mechanism – Clotting time
   - Partial thromboplastin time (PTT)
   - Prothrombin time (PT)
   - Fibrinogen assay.

In this course, you will learn to perform the following tests of hemostasis
1. Bleeding time
2. Clotting time
3. Platelet count

OSPE-I
Procedure: Determine the bleeding time of your own blood
With all aseptic precaution take a clean and bold prick and note the time
Blot the first drop of blood with the filter paper completely after 30 seconds
Repeat the same procedure until no blot appears on the filter paper
Count the number of blot on the filter paper
Calculate the bleeding time in seconds as number of blots x 30

OSPE-II
Procedure: Determine the clotting time of your own blood by Duke’s method
With all aseptic precaution take a clean and bold prick and note the time
Fill the 2/3rd length of an empty and clean capillary by holding the capillary tip to the edge of the blood drop
After 2 minutes break a small bit of the tube from filled end
Repeat the same procedure until a fine thread of fibrin appears between the broken ends
Calculate the clotting time as 2 minutes + (Number of bits-1 x 30) seconds

QUESTIONS
1. What happens to bleeding time in each of the following disorders of hemostasis and why?
   a. Thrombocytopenia
   b. Thrombasthenia
   c. Scurvy
   d. Afibrinogenemia.
2. List four conditions where clotting time is increased.
3. What happens to the following tests of hemostasis in each of the following conditions:

<table>
<thead>
<tr>
<th>Conditions</th>
<th>Bleeding time</th>
<th>Clotting time</th>
<th>Platelet count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thrombocytopenia</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hemophilia</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Obstructive jaundice</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heparin therapy</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>von Willebrand’s disease</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thrombasthenia</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pseudoxanthoma elasticum (Vascular disorder)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

4. What is the normal platelet count?
5. What are the functions of platelets?
6. Name two conditions where platelet counts may be abnormally:
   a. High
   b. Low
7. What are the clinical effects of a low platelet count?
8. Explain the term thrombasthenia. What is its clinical significance?
9. Name the different methods by which you can determine clotting time.
10. Name the diseases where bleeding time and clotting time are altered; explain briefly the reasons for the alterations.
11. Why the bleeding time is normally shorter than clotting time?
12. What is hemostasis? Name the different mechanisms involved in it.
13. What is the natural anticoagulant present in the human body?
14. Name some of the anticoagulants used clinically.
15. What is thrombosis?
16. What is an embolus?
17. Enumerate the various coagulation factors.
18. What is prothrombin time and what is its significance?
19. Name some other tests used in coagulation disorders.
SYNONYM

Blood typing.

INTRODUCTION

It is essential to know the blood group of a person if he is encountered by anyone of the following circumstances:
1. Blood transfusion
2. Paternity dispute
3. Medicolegal problem
4. Organ transplantation.

The current system of blood grouping was discovered by Landsteiner in 1900 and is known as the Landsteiner’s ABO system.

PRINCIPLE

The RBCs contain a series of antigens known as agglutinogens on their cell membrane while the plasma contains antibodies known as agglutinins. To find out a person’s blood group the RBCs are made to react with sera containing known agglutinins. The slide is then observed for the presence or absence of agglutination and hemolysis of RBC. This can be done with the naked eyes but is ideally done by viewing the slide under the microscope.
APPARATUS AND REAGENTS REQUIRED

1. Sterile lancet
2. Cotton swab
3. Alcohol
4. Glass dropper
5. Toothpicks
6. Microscope
7. Porcelain tile
8. 3.8% sodium citrate in normal saline
9. Anti-A serum
10. Anti-B serum
11. Anti-D serum.

Note: Anti-A serum is tinted blue and Anti-B serum is tinted yellow (Fig. 12.1).

PROCEDURE

With a glass marking pencil, the porcelain tile is divided into 3 portions. Under aseptic conditions the subject whose blood group is to be determined is asked to stick out his finger which is pricked and 3–4 drops of blood is then obtained. Following this the blood is diluted with 1 mL of 3.8% sodium citrate in normal saline taken in a test tube. A drop of anti-A, anti-B and anti-D sera is placed on each of the 3 portions on the tile. Now, 1 drop of the diluted blood is put on each of the 3 portions containing the sera. This is mixed with 3 separate toothpicks. After 10–15 minutes each portion is examined for clumping and agglutination first with the naked eyes and then under the microscope (Fig. 12.2).

OBSERVATION AND RESULTS

If any agglutination occurs, it is usually visible to the naked eye as dark red clumps of different sizes. The presence or absence of agglutination indicates the blood group of the subject as shown in the following table:

AGGLUTINATION OF SUBJECT’S BLOOD GROUP

<table>
<thead>
<tr>
<th>With anti-A serum</th>
<th>With anti-B serum</th>
<th>Blood group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Present</td>
<td>Absent</td>
<td>A</td>
</tr>
<tr>
<td>Absent</td>
<td>Present</td>
<td>B</td>
</tr>
<tr>
<td>Present</td>
<td>Absent</td>
<td>AB</td>
</tr>
<tr>
<td>Absent</td>
<td>Present</td>
<td>O</td>
</tr>
</tbody>
</table>
CONCLUSION

Having determined the subject’s blood group his Rh typing can be determined in a similar fashion using anti-Rh serum (Anti-D serum).

RESULT

The blood group of the given blood sample is _____________

OSPE

Procedure: Determine the blood group of your own blood

- Take a clean porcelain tile and mark the three wells as A, B and D.
- Add anti-A, anti-B and anti-D anti-sera to the three wells of the tile separately
- With all aseptic precautions add a drop of blood directly to each well, without touching the anti-sera
- Mix them properly and wait for 5–10 minutes
- See for the agglutination and report the findings
QUESTIONS

1. The approximate percentages of people in each of these blood groups in South India is as follows:
   - O group 45%
   - B group 30%
   - A group 20%
   - AB group 5%.
   Ascertain the blood group of a large part of your class as possible and calculate the percentages. How do these values vary compared to those of the general population?
2. What is your blood group? Illustrate your observations.
3. How are the A and B antigens inherited?
4. What is the practical importance of blood grouping?
5. What is a transfusion reaction? Differentiate between a major and a minor transfusion reaction.
6. Mention another important system of blood grouping. How does the ABO system differ from this system of grouping? Give the clinical importance of this system. What is Landsteiner’s law?
7. Name the different types of blood groups you know.
8. What is the main difference between the ABO group and the Rh factor?
9. What is cross-matching?
10. What are the uses of blood groups?
11. What is Bombay blood group?
12. Why is blood group ‘O’ known as universal donor?
13. What is the importance of MN group?
14. What is the composition of ACD mixture?
15. Which preservative (anticoagulant) is used for storing blood?
16. What is the difference between Rouleaux formation and agglutination?
AIM

To determine the osmotic fragility of red blood cells.

APPARATUS REQUIRED

Ten clean test tubes, test tube rack, disposable syringe with needle, spirit swab, dropper bottle, 3.8 % sodium citrate and saline solutions of following concentrations—0.70%, 0.65%, 0.60%, 0.55%, 0.50%, 0.45%, 0.40%, 0.35%, and 0.30%.

Preparation of Sodium Chloride Solution

A 1% stock solution is prepared by dissolving 1 g of NaCl in 100 mL of distilled water. To make 0.70% NaCl: 70 mL of stock solution is diluted with 30 mL of distilled water. To make 0.65% NaCl: 65 mL of stock solution is diluted with 35 mL of distilled water. Thus, sodium chloride solution of different concentrations is prepared accordingly.

PROCEDURE

Ten test tubes containing sodium chloride solutions of varying concentration from 0.70% to 0.30% are placed in a test tube rack. The blood drawn intravenously is mixed with the anticoagulant in the beaker. With the help of a dropper, a drop of blood is added into each of the test tube and mixed thoroughly by inverting the tubes several times. Alternatively, a finger is pricked and a drop of blood is squeezed into each of the test tube. The tubes are allowed to stand for one hour. At the end of one hour, the test tube where hemolysis has just begun (supernatant fluid just becomes red) and the test tube where hemolysis is incomplete (whole solution is uniformly red) are noted (Fig. 13.1).
Osmotic Fragility of Red Blood Cells

**DISCUSSION**

Fluid passes through the membrane into the red blood cells when it is placed in hypotonic solution. The movement of fluid depends on the surface area of the membrane. The cells get distended and lysed. Normal red blood cells commence lysing at 0.48% saline and end or complete at 0.34% saline solution.

**Osmotic Fragility is Increased in the Following Conditions**

1. Hereditary spherocytosis
2. Venous blood cells are more fragile than arterial blood cells
3. Stored blood
4. Increased intracellular osmotic pressure.

**Osmotic Resistance (Decreased Fragility) is Seen in the Following Conditions**

1. Thalassemia
2. Sickle cell anemia
3. Disorders in which leptocytes including ‘target’ cells are found.

**PRINCIPLE OF THIS EXPERIMENT**

This is a quantitative experiment assessing the degree of fragility of red blood cells. A series of test tubes of various concentrations of NaCl solutions are taken and a drop of blood is added to each of them. The tubes at which hemolysis begins and is complete, are noted and reported.

**DISCUSSION**

Osmosis is that process, which governs transport of solvent between two solutions separated by a membrane, which is relatively impermeable to the solute. When an RBC is placed in:

1. A solution of 0.9% saline, no exchange of water takes place. This solution is said to be
isotonic with the interior of the RBC.

2. Dilute NaCl solution (concentration of NaCl less than 0.9%), water enters the cell. This solution is said to be hypotonic with respect to the interior of the RBC.

3. Concentrated NaCl solution (concentration of NaCl more than 0.9%), water leaves the cell. This solution is said to be hypertonic with respect to the interior of the RBC.

As the RBC is a biconcave disc, when placed in hypotonic solution it is able to take up water till it attains the spherical shape. If the tonicity of the solution is very low then the cell is unable to contain the excess inflow of water and it bursts (hemolysis). On the other hand, when the cell is placed in a highly concentrated NaCl solution, water leaves the cell and the cell shrinks (crenation). On placing the cell in an isotonic solution no change occurs.

**SIGNIFICANCE**

1. In normal healthy individuals, osmotic fragility of RBCs begins at 0.48% NaCl and ends at 0.34% NaCl solution.

2. RBCs osmotic fragility increases when they become spherical, e.g. in spherocytosis (independent of the cause) it begins at 0.7% NaCl and ends at 0.45% NaCl solution.

3. RBCs osmotic fragility is decreased when they become slender, e.g. in iron deficiency anemia and in thalassemia, it begins at 0.36% NaCl and ends at 0.24% NaCl solution.

**RESULT**

Hemolysis begins at ..........% saline and ends at ..........% saline.

**QUESTIONS**

1. Would you say the fragility is increased or decreased in the following situations?
   a. Hemolysis begins at 0.55% and is complete at 0.45%.
   b. Hemolysis beings at 0.3% and is completed at 0.25%.

2. Name one condition where fragility is increased and say why?

3. Name one condition where fragility is decreased.

4. Name one other etiology where fragility is increased in the RBC.

5. What is the difference between isotonic and iso-osmotic solutions?

6. Define the terms hemolysis and crenation. When do they occur?

7. Name some hemolytic agents.

8. What is mechanical fragility and how is it tested?

9. Do all the normal RBCs present in the given sample of blood have the same osmotic fragility to hypotonic solutions?

10. What happens to the osmotic fragility of RBCs in an individual after splenectomy?

11. Define the terms osmosis and osmotic pressure. How much osmotic pressure is exerted by the blood and what is its importance in the body?

12. Explain the mechanism of hemolysis of RBCs in the body.
AIM

To determine the specific gravity of the given sample of blood by Van Slyke’s falling drop method.

PRINCIPLE

The specific gravity of blood is determined by comparing the specific gravity of a drop of blood with that of solutions of known specific gravity.

APPARATUS AND REAGENT REQUIRED

1. Stock solutions of copper sulfate of 1.1 specific gravity. It is prepared by dissolving 170 g of blue vitriol (CuSO₄ + 5H₂O) in 1.0024 liters of distilled water.
2. Distilled water
3. Four test tubes
4. Clean dry sterilized syringe with needle
5. Cotton and rectified spirit.

PREPARATION OF SOLUTION

Twomiscible liquids of known but different specific gravity (copper sulfate and water) are mixed in varying proportions.

The stock solution of CuSO₄ with specific gravity 1.100 and distilled water with specific gravity 1.000 are mixed in varying proportions to give standard solutions of CuSO₄ with specific gravity varying from 1.049 to 1.064.
**Advantages of Copper Sulfate Solution**

1. It is cheap and easily available
2. It is not hygroscopic
3. It is not volatile or inflammable.
4. It can be repeatedly used.
5. The temperature coefficient of expansion of CuSO₄ solution and blood are equal and no correction for temperature is required.

**Other Solutions Used Are**

1. Glycerine and water
2. Chloroform and benzene.

**PROCEDURE**

1 mL of blood is collected from syringe; a drop of blood is delivered directly into each of labeled test tubes from a height of about 1 cm from the surface of CuSO₄. The drop of blood on entering CuSO₄ is surrounded by thin coat of copper proteinate. This layer does not produce any change in density. The drop first sinks for 2–3 cm below the surface of solution and loses momentum within a few seconds. If the specific gravity of the drop of blood is greater than CuSO₄ then it continues to sink for the next 15–20 sec. If its specific gravity is same as that of the solution it will remain stationary. If, however, the specific gravity is lower than that of the solution it will raise to the surface.

**PRECAUTIONS**

1. The stock solution and standard solution should be carefully prepared.
2. Blood should be dropped from 1 cm above the surface of standard solution. If blood is dropped from a greater height, it will sink to the bottom because of its greater momentum.
3. The reading should be taken 15–20 sec after adding the drop of blood to CuSO₄ solution.

**DISCUSSION**

The normal range of specific gravity of venous blood is 1.005–1.065; of serum 1.028–1.032; of corpuscles 1.092–1.095. The specific gravity is subjected to physiological and pathological variations.

**PHYSIOLOGICAL VARIATIONS**

1. It is highest in newborn babies due to polycythemia.
2. It is lower in women than men because of comparatively lower RBC count of women.
3. **Pregnancy:** It is lower during pregnancy because of hemodilution due to increase in plasma volume.
Specific Gravity of Blood

4. **Diurnal variation**: It is lower in afternoon and after meals and is higher during night.
5. **Exercise**: It is higher after strenuous exercise due to raised RBC count.
6. **Hemoconcentration**: Less water intake or excessive sweating raises the specific gravity of blood.

**PATHOLOGICAL VARIATIONS**

In conditions like vomiting, diarrhea and burns, the specific gravity of blood is raised due to hemoconcentration.

**RESULT**

The specific gravity of the sample of blood is ____________

**QUESTIONS**

1. What are the physiological conditions which affect the specific gravity of blood?
2. What are the uses of determining the specific gravity of blood?
3. What happens when blood reacts with CuSO₄?
4. In what pathological conditions the specific gravity increased and decreased?
5. What is the specific gravity of:
   a. Plasma
   b. Red cells
   c. Whole blood.
6. Define specific gravity.
7. What is the significance of determining the specific gravity of blood?
Clinical Physiology
All systemic examinations should be preceded by general examination, to get a general idea about the physical condition of the subject/patient. The student can refer the following books for detailed information regarding physical examination:

1. Hutchison’s clinical methods—Hunter and Bomford. Published by ELBS and Churchill Livingstone.
2. Physical diagnosis—Vakil and Golwalla. Published by MPP Bombay.
3. Clinical examination—John McLeod. Published by ELBS.

A properly elicited history followed by a general examination of the patient will clinch on the system which is mainly affected. The general examination findings, which have a bearing on a particular system should be given more importance and should not be missed during general examination. Before starting with general examination, the personal details of the subject should be recorded, i.e. Name, Age, Sex, Occupation and Address.

1. **Built and nourishment:** Normal person is well built and well nourished. Variations noted may be moderately built, poorly built; noting whether the person is over weight or under weight can assess nourishment.

2. **Height and weight:** As per Indian standard, the average height of an adult male is about 170 cm and adult female about 160 cm. Weight of adult male ranges between 55–75 kg and of female between 45–65 kg.

3. **Pallor:** It is the paleness that appears in skin and mucous membrane due to decrease in hemoglobin content. Examine the lower palpebral conjunctiva for verification of anemia. Anemia can occur due to decreased production of RBCs, increased destruction of RBCs, or blood loss (Fig. 15.1A).

4. **Icterus:** It is the yellowish discoloration of sclera, mucous membrane and skin due to increase in serum bilirubin above normal—usually above 2 mg%. Verification can be done on the upper sclera. For this, elevate the upper eyelid and ask the subject to look down. Bilirubin specifically stains elastic fibers and sclera has got plenty of elastic fibers.
Jaundice can develop due to prehepatic (hemolytic), hepatic (hepatocellular) and posthepatic (obstructive) causes (Fig. 15.1B).

5. **Cyanosis:** Cyanosis is bluish discoloration of skin and mucous membranes, due to increase in concentration of reduced hemoglobin above 5 gm% in capillary blood. Cyanosis can be central or peripheral. Central cyanosis is due to the defective oxygenation in the lungs or due to arteriovenous shunt in the heart. Peripheral cyanosis is due to the stagnation of blood in the periphery. Central cyanosis is verified on the tongue, and lips and peripheral on fingertips, earlobes, tip of nose, etc. Central cyanosis causes—respiratory diseases, congenital cyanotic heart disease. Peripheral cyanosis causes—congestive cardiac failure, inferior venacaval obstruction (cyanosis of lower extremities).

6. **Clubbing:** Clubbing is hyperplasia of soft tissue, in fingers and toes with obliteration of angle between the root of the nail and skin. The tips of fingers and toes appear like “club.” This is seen in chronic hypoxia, lung cancer, cirrhosis of liver, ulcerative colitis and subacute bacterial endocarditis (Figs 15.2A to C).

7. **Lymphadenopathy:** The lymph nodes in the drainage area of any organ are prone to be affected in diseases of that organ. There may be enlargement or tenderness or both. The usual groups of lymph nodes examined are as follows: Cervical group—vertical chain; axillary group—apical, anterior and posterior; para-aortic group; inguinal group, popliteal group and epitrochlear gland (Figs 15.3 and 15.4).
**Fig. 15.3**: Distribution of palpable lymph glands

**Figs 15.4A to E**: Palpation of lymph nodes: (A) Cervical; (B) Submandibular; (C) Submental; (D) Preauricular; (E) Axillary
8. **Edema**: It is the collection of excess fluid in the extracellular spaces, seen in diseases of cardiovascular system, renal system and gastrointestinal system. The mechanism of production varies. Edema is noted by pressing the area for a sustained period as edema pits on pressure. In an ambulant patient, edema develops over lower extremities. In a recumbent patient, edema develops over the sacral region and back (Fig. 15.5).

9. **Skin and hair**: The color and texture of skin and abnormalities, if any, should be noted. Similarly the color and distribution of hair also should be noted.

10. **Pulse**: By definition, it is the expansile impulse transmitted along the arteries, due to the intermittent ejection of blood from the aorta (Fig. 15.6). The features of the pulse to be noted:
   - **Rate**—how many per minute.
   - **Rhythm**—the regularity with which one pulse follows the other.
   - **Volume**—the amplitude of expansion of vessel wall.
   - **Character**—whether collapsing or not (rise, maintenance and fall); the contour of a pulse.
   - **Condition of vessels wall**—thickened or not.
   - **Other peripheral pulsations**, i.e. brachial, carotid, facial, femoral, popliteal, dorsalis pedis and posterior tibial.
   - **Radiofemoral delay**—whether there is any time lapse between the appearance of radial and femoral pulsations noted only in CVS diseases.

11. **Blood pressure**: The BP is noted in recumbent and erect postures on the upper limb.

12. **Jugular venous pressure (JVP)**: The right internal jugular vein is taken for the study. Keep the subject in a propped up position at 45° inclination to the bed and note the upper level of jugular pulse. The vertical height from sternal angle to the upper level of pulse is taken in centimeters and is expressed as the jugular venous pressure. Normally the jugular venous pulse is not visible above the clavicle (Figs 15.7 and 15.8).

13. **Respiratory rate**: Place the right palm over the upper part of the abdomen of the subject and count the number of respirations in one minute.

14. **Body temperature**: Body temperature is measured using the clinical thermometer. Usually the oral temperature is measured. The subject is asked to hold the thermometer under the tongue, gently supported by the lips for two minutes. The procedure should not be done immediately after taking hot or cold drinks. In unconscious patients, rectal temperature is usually taken and in young children, either axillary temperature or rectal temperature can be taken.
In addition to above mentioned points, the following should be specially noted if relevant:

**Along with cardiovascular and respiratory system:**
- Dyspnea
- Cough
- Chest pain
- Sputum

**Along with central nervous system:**
- Neurofibromas
- Tuft of hair on low back
- Short neck
- Pes cavus, pes planus
- Polydactyly, syndactyly
- Hemangiomas
GENERAL EXAMINATION (SAMPLE REPORT)

Name  
Age/sex  
Occupation  
Address  

GENERAL EXAMINATION

Well built and well nourished
Height 170 cm
Weight 60 kg
No pallor, No icterus, No cyanosis
No clubbing of fingers or toes
No significant lymph node enlargement
No edema
Skin, hair and nails appear healthy

**Pulse:** 72/min regular, normal in volume and character, no thickening of vessel walls, all peripheral pulsations are felt equally on both sides and there is no radio-femoral delay.

Blood pressure—120/80 mm Hg in the right upper limb in supine position.

**JVP:** Not raised

**Respiratory rate:** 18/min, regular

**Body temperature:** 37°C

**Impression:** On general examination, the subject appears to be normal.

### OSCE-I

**Procedure:** Examine the pallor of the subject

- Make the subject comfortable and instruct him/her properly
- Gently lower down the lower eyelid and look for the pallor in the palpebral conjunctiva
- Also check for the pallor on the nail beds, tip and dorsum of the tongue, palms and soles, and general skin surface
- Report the observation correctly

### OSCE-II

**Procedure:** Examine the cyanosis of the subject

- Make the subject comfortable and instruct him/her properly
- For peripheral cyanosis look for the bluish discoloration on the tip of the nose, ear lobule, tip of the fingers and toes, and palms and soles
- For central cyanosis look for the bluish discoloration on the tip of the tongue, inner aspect of the lips, soft palate, and lower palpebral conjunctiva
- Report the observation correctly
## General Examination

### OSCE-III

**Procedure: Examine the clubbing of the subject**

- Make the subject comfortable and instruct him/her properly
- Look for any obvious bulbous enlargement of the tip of the fingers
- Ask the subject to approximate the nail beds of the two index fingers in front of the eyes and look for the diamond shaped space formed between the fingers
- Report the observation correctly

### OSCE-IV

**Procedure: Examine the icterus of the subject**

- Make the subject comfortable and instruct him/her properly
- Look for the yellowish discoloration of skin and mucous membrane, sclera, under surface of the tongue, palm and sole and general skin surface. Also check for the pallor on the nail beds, tip and dorsum of the tongue, palms and soles, and general skin surface
- Report the observation correctly

### OSCE-V

**Procedure: Examine the edema of the subject**

- Make the subject comfortable and instruct him/her properly
- Expose the dorsum of the foot and press the area with the thumb for a minimum of 30 seconds
- Look for the depression at the point of pressure
- Report the observation correctly

### OSCE-VI

**Procedure: Examine the cervical lymph nodes of the subject**

- Make the subject comfortable and instruct him/her properly
- Inspect for any visible lymphadenopathy around the neck
- Stand behind the subject and by using both the hands palpate for submental, submandibular, tonsilar, pre-auricular, post-auricular and occipital group of lymph nodes, starting from submental region to occipital region
- Also palpate for superior cervical, deep cervical, posterior cervical and supraclavicular group of lymph nodes
- Report the size, site, consistency, tenderness and fixing of the nodes

### OSCE-VII

**Procedure: Examine the axillary lymph nodes of the subject**

- Make the subject comfortable and instruct him/her properly
- Inspect for any visible lymphadenopathy around the axilla
- Stand in front of the subject and support the arm on the side to be examined
- Palpate the left axilla with right hand and vice-versa
- Palpate the anterior, posterior, medial walls and roof of the axilla sequentially
- Report the size, site, consistency, tenderness and fixing of the nodes
OSCE-VIII

Procedure: Examine the JVP of the subject

<table>
<thead>
<tr>
<th>Step</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Make the subject comfortable and instruct him/her properly</td>
<td></td>
</tr>
<tr>
<td>Make the subject to lie on his back with the upper part of the body</td>
<td>supported at an angle of 45° to the horizontal with the chin turned slightly</td>
</tr>
<tr>
<td></td>
<td>to the left</td>
</tr>
<tr>
<td>Inspect the neck veins carefully and look for any visible pulsation</td>
<td></td>
</tr>
<tr>
<td>Mark the upper level of pulsation, if visible</td>
<td></td>
</tr>
<tr>
<td>Locate and mark the sternal angle</td>
<td></td>
</tr>
<tr>
<td>Measure the vertical height between the sternal angle and the visible</td>
<td>pulsation using two scales</td>
</tr>
<tr>
<td>Report the observation correctly</td>
<td></td>
</tr>
</tbody>
</table>
AIM

To perform the clinical examination of the respiratory system on a given subject. Examination of the respiratory system is done on the following headings:

1. Inspection
2. Palpation
3. Percussion
4. Auscultation.

Inspection

The subject is stripped up to the waist and is examined under good light.

The following points are noted:

1. Position of the trachea and the apex beat
2. Shape of the chest and deformities
3. Respiratory rate
4. Type of breathing
5. Movements of the chest
6. Any wasting of muscles or drooping of the shoulder.

The shape of the chest is normally bilaterally symmetrical and varies with the build of the individual. It is short, broad and deep in thick set individuals: Long, narrow, and flat in thin tall people. It is barrel shaped in emphysematous condition. The cross-section of the chest in a normal person is somewhat elliptical; the ratio between anteroposterior and lateral diameters being 5:7. The rate, rhythm, type of respiration and chest movement during respiration are noted. It is regular in rhythm and abdominothoracic in adults and in young children, and in women it is thoracoabdominal during pregnancy. The expansion of the chest is equal on both sides.
Palpation

**Mediastinal position:** The position of the trachea is determined by placing the index finger and the ring finger on the bony prominences of the suprasternal notch and the middle finger is gently pushed posteriorly and downwards between the sternocleidomastoid and the trachea on either side to see for any tracheal deviation (Fig. 16.1).

The position of the apex beat is also palpated. Displacement of trachea and/or apex beat may occur in mediastinal shift due to disease of the lungs or pleura.

**Chest movement:** To determine the chest expansion of the apical region of the lungs, the hands are placed over the shoulders close to the neck and the thumb at right angle to other fingers and approximated in the midline (This is carried out with the examiner standing behind the subject). To determine the expansion of the other regions, the chest is gripped on either side and thumbs approximated in the midline. When the subject takes full inspiration, the distance of departure of the thumbs from the midline indicate the extent of expansion of either half of the chest (Figs 16.2A to C).

**Vocal fremitus:** These are vibrations that are felt on the chest wall when the subject utters a sound. To test vocal fremitus, the subject is told to repeat “one, one.” The palm or ulnar border of the hand is applied flat on the chest. The intensity of vibration is compared on corresponding areas of the two sides of the chest. Vocal fremitus is increased in consolidation of lung and diminished in pleural effusion.

**Expansion of the chest:** The degree of expansion of chest may be measured by placing an inch tape just below the nipple around the chest with its mark at the middle of the sternum. Instruct the subject to breathe in and out as deep as possible. In females, the mammary tissue is avoided by making the measurement above or below the mammary gland. Initial respiratory efforts are often shallow. So readings are taken after 2 or 3 breaths. Measure the chest circumference at the end of deep inspiration and at the end of deep expiration. Five to eight cm difference is normal. It is decreased in bronchial asthma, pulmonary fibrosis, etc. (Fig. 16.3).
Percussion

There are two types of percussion (i) Direct and (ii) Indirect.

In direct percussion, the clavicle is percussed directly. In indirect percussion the middle finger of the left-hand (pleximeter finger) is placed firmly parallel to the intercostal space. The back of the middle phalanx is struck perpendicularly with the tip of the middle finger of the right hand (percussing finger) (Figs 16.4 and 16.5).

1. The stroke should be delivered at the wrist and finger joint, not at the elbow or shoulder.
2. The long axis of the pleximeter finger should be parallel to the edge of the organ being percussed.
3. Percussion should be done from a resonant area to a dull area.

During percussion of the sides of the chest, the subject’s hands are placed over the head and while percussing the back of the chest, the subject is asked to criss-cross his arms in front of him. Lung resonance is impaired in consolidations and fibrosis. In thickened pleura and pleural effusion, stony dullness occurs. Resonance is increased in pneumothorax.
Auscultation

Auscultation is performed all over the chest. Corresponding points on either side is to be auscultated simultaneously. The character of breath sounds, vocal resonance and abnormal sounds if any are noted (Figs 16.6A and B).

Figs 16.6A and B: Auscultation of the Anterior (A), and Posterior (B) chest wall

1. Character of breath sounds: There are two typical varieties of breath sounds.
   a. Vesicular
   b. Bronchial.

   **Vesicular breath sounds** are heard all over the chest under normal conditions. The inspiratory sounds are intense and audible during the whole of inspiration. Expiratory sound follows that of inspiration without a distinct pause, and is audible only during the early part of expiration. Vesicular breath sounds are low pitched and rustling in character (Fig. 16.7B).

   **Bronchial breath sounds** are produced by the passage of air through the trachea and large bronchi. The inspiratory sound is moderately intense, becomes inaudible before the end of inspiration. The duration extends through the greater part of expiration, being as long as the inspiratory sound (Fig. 16.7C).

   Table 16.1 shows the differences between bronchial and vesicular breath sounds.

   **Tubular breath sound:** High pitched bronchial breath sound, heard over the consolidated lung tissue.

   **Cavernous breath sound:** Low pitched bronchial breathing heard over large cavities. Diminution or absence of breath sounds are found when there is thickened pleura, pleural effusion and pneumothorax. It will also be found in conditions causing diminished air entry to the underlying lung.

   **Table 16.1:** Differences between bronchial and vesicular breath sounds

<table>
<thead>
<tr>
<th>Produced by</th>
<th>Bronchial breath sounds</th>
<th>Vesicular breath sounds</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pitch</td>
<td>Large airways</td>
<td>Small airways</td>
</tr>
<tr>
<td>Pause between I and E</td>
<td>High pitch and harsh</td>
<td>Low pitch (rustle)</td>
</tr>
<tr>
<td>Relative duration of I and E</td>
<td>Present</td>
<td>Absent</td>
</tr>
<tr>
<td>I = inspiration; E = expiration</td>
<td>Equal</td>
<td>E &lt; I</td>
</tr>
</tbody>
</table>


2. **Vocal resonance** is auscultatory sound when the subject is repeatedly saying “one, one, one”. Vocal resonance is increased in consolidation, diminished in pleural thickening, pleural effusion and pneumothorax.

3. **Abnormal sounds** are rhonchi, crepitations and pleural rub, which are heard during diseased condition of the lung. Rhonchi are heard when there is narrowing of the respiratory passage and crepitations or rales are heard when there is presence of fluid secretions. Rub is grating sounds heard during pleurisy.

### OSCE-I

**Procedure:** Examine the position of the trachea of the subject

- Make the subject comfortable and instruct him/her properly
- Make the subject to sit comfortably and ask him to extend the neck slightly
- Examine for the equal prominence of the sternocleidomastoid muscle on both the sides of the trachea
- Palpate the trachea with the middle finger and fell for the resistance between the trachea and sternocleidomastoid muscle on both the sides
- Report the observation correctly

### OSCE-II

**Procedure:** Examine the movements of the chest wall of the subject

- Make the subject sit comfortably and instruct him/her properly
- Stand in front of the subject and place the palms lightly over the chest on both sides with the fingers spread apart and the thumbs approximating each other in the midline
- Observe for the separation of the thumb with movements of the chest wall during the deep inspiration
- Report the observation correctly
### OSCE-III

**Procedure:** Examine the chest expansion of the subject

- Make the subject sit comfortably and instruct him/her properly
- Stand behind the subject and place the measuring tape at the level of the nipples and measure the girth of the chest wall
- Instruct the subject for deep inspiration
- Observe for the expansion of the chest wall (in cm)
- Report the observation correctly

### OSCE-IV

**Procedure:** Demonstrate the procedure of recording of vocal fremitus of the subject

- Make the subject sit comfortably and instruct him/her properly
- Keep the ulnar border of the hand sequentially over supraclavicular, infraclavicular, anterior intercostal spaces (2nd–6th), axillary, suprascapular, interscapular and infrascapular regions
- Simultaneously ask the subject to utter some monosyllable word (like one, one, one) and feel for the vibrations produced due to sound
- Compare the findings on the two sides of the chest
- Report the observation correctly

### OSCE-V

**Procedure:** Demonstrate the procedure of percussion of the respiratory system

- Make the subject sit comfortably and instruct him/her properly
- Expose the chest wall
- Place the palmer aspect of the middle finger of the left hand firmly over the chest wall, parallel to the ribs
- Strike the center of the middle phalanx with the tip of the right middle finger
- Movement should be at the wrist and not at the elbow
- Percuss all areas of the lung from resonant to dull area
- Report the observation correctly

### OSCE-VI

**Procedure:** Demonstrate the procedure of auscultation of the respiratory system

- Make the subject sit comfortably and instruct him/her properly
- Expose the chest wall
- Ask the subject to deeply inspire and expire through mouth
- Auscultate all the areas of the chest wall and compare the findings on both the sides
- Report the observation correctly
Examination of the Respiratory System

OSCE-VII

Procedure: Demonstrate the procedure of recording of vocal resonance of the subject

Make the subject sit comfortably and instruct him/her properly

Keep the stethoscope sequentially over supraclavicular, infraclavicular, anterior intercostal spaces (2nd–6th), axillary, suprascapular, interscapular and infrascapular regions

Simultaneously ask the subject to utter some monosyllable word (like one, one, one) and auscultate for the sound produced

Compare the findings on the two sides of the chest

Report the observation correctly

QUESTIONS

1. What is sternal angle?
2. What is the normal position of trachea? Why?
3. What is the difference between vocal resonance and vocal fremitus?
4. What are the abnormal shapes of the chest?
5. What are the rules of percussion?
6. Define the term eupnea, tahypnea, apnea, dyspnea and hyperpnea.
7. What is pleural effusion?
8. What are added sounds? When do they occur?
9. What are the causes for a shift in the mediastinum?
10. What is the cause for the slightly increased vocal resonance in the right upper zone of the normal lung?
11. Where are bronchial breath sounds heard in the normal human?
12. Where are vesicular breath sounds heard well in the normal human?
13. What is the significance of noting the positions of the trachea and apex?
14. Fill in the Table:

<table>
<thead>
<tr>
<th>Conditions</th>
<th>Percussion</th>
<th>Breath sounds</th>
<th>Vocal resonance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pleural effusion</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Consolidation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pneumothorax</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

15. Define BBS and VBS and what are the differences?
16. What are the investigations that can be done in diseases of the respiratory system?
RESPIRATORY SYSTEM EXAMINATION

Name: 
Age: 
Sex: 
Occupation: 

General Examination

Build and nutrition 
Pallor 
Icterus 
Cyanosis 
Clubbing 
Lymphadenopathy 
Pedal edema 
Pulse 
BP 
Temperature

Systemic Examination

Inspection
- Chest and spine shape
- Any obvious wasting
- Trachea
- Apex beat
- Type of breathing
- Respiratory rate
- Chest movements
- Chest symmetry
- Scars, sinuses

Palpation
- Trachea
- Apex beat
- Chest movements
- Chest expansion
- Vocal fremitus* on both sides

Percussion*
- Direct (both sides)
- Indirect

Auscultation
- Breath sounds*
- Vocal resonance*
- Adventitious sounds
+ Draw a table as given below for the points marked by star.

<table>
<thead>
<tr>
<th></th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Supraclavicular</td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>Infracavicular</td>
<td></td>
</tr>
<tr>
<td>3.</td>
<td>Mammary</td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>Axillary</td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>Infra-axillary</td>
<td></td>
</tr>
<tr>
<td>6.</td>
<td>Suprascapular</td>
<td></td>
</tr>
<tr>
<td>7.</td>
<td>Interscapular</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Upper</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Middle</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Lower</td>
<td></td>
</tr>
<tr>
<td>8.</td>
<td>Infra-axillary</td>
<td></td>
</tr>
</tbody>
</table>
AIM
To record the movements of respiration.

PRINCIPLE
It consists of a corrugated rubber tube with a stopper at each end and air outlet pipe connected to the tambour. If a rubber tube is tied round the chest, the movement of thorax during respiration brings about a change in the pressure of air contained in tube. This change in pressure of air inside the rubber tube can be recorded by connecting the tube to a suitable recording device.

REQUIREMENTS
1. Sherrington-Starling’s drum (SS drum)
2. Stethograph
3. Marey’s tambour.

PROCEDURE
The subject is allowed to sit on a stool. The stethograph, which is connected to a Marey's tambour is tied around the chest of the subject. The SS drum is allowed to move at 1.2 mm/sec speed and the writing point of piston recorder is brought into contact with the smoked paper on the drum. First, the normal respiratory movements are recorded. The subject is asked to take forceful, deep and rapid respirations for 3 seconds and the record
Recording of Respiratory Movements (Stethography)

is continued till normal movement returns. The subject is asked to swallow a glass of water and its effect on respiratory movements is recorded. The subject is asked to breathe in and out into a polythene bag and a record of the effect of CO₂ excess is obtained. The subject is asked to take maximum forceful inspiration and to hold his breath for as long as he can. The movement of the chest during breath holding and the return to normal respiration is recorded. The effect of exercise on respiration are also recorded (effects of other maneuvers may also be recorded). A time tracing is taken by an electrical time marker (frequency l/sec) below the record of respiratory movements and the normal respiration rate is calculated. The tracings after being labeled are fixed in the varnishing fluid (Figs 17.1 and 17.2).

**Fig. 17.1:** Stethograph

**Fig. 17.2:** Stethograph recordings
QUESTIONS

1. What changes occur following hyperventilation, swallowing and apnea?
2. Outline the mechanism by which arterial blood PCO$_2$ affects respiration.
3. What are the effects of exercise on respiration?
4. How else may respiration be recorded?
5. What is the mechanism of the apnea or hypoventilation, which follows voluntary hyperventilation?
6. What is periodic breathing? Mention the conditions where this develops. What is its mechanism?
7. What is the cause of the hyperventilation following breath holding?
9. What are the uses of stethography?
10. What is the principle involved in stethography?
11. What is the normal breath-holding time in adults?
12. What is breaking point?

STETHOGRAPHY

<table>
<thead>
<tr>
<th>Name:</th>
<th>Age:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex:</td>
<td>Occupation:</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>SI No.</th>
<th>Maneuvre</th>
<th>Effect seen (Draw the stethography tracings)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Normal breathing</td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>Hyperventilation</td>
<td></td>
</tr>
<tr>
<td>3.</td>
<td>Breath-holding</td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>Deglutition</td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>Coughing</td>
<td></td>
</tr>
<tr>
<td>6.</td>
<td>Sneezing</td>
<td></td>
</tr>
<tr>
<td>7.</td>
<td>Laughing</td>
<td></td>
</tr>
<tr>
<td>8.</td>
<td>Talking</td>
<td></td>
</tr>
<tr>
<td>9.</td>
<td>Jogging</td>
<td></td>
</tr>
</tbody>
</table>

Inference
AIM

To record and measure vital capacity, tidal volume and expiratory reserve volume in a given subject.

Several models of spirometers are available out of which student’s spirometry is very commonly used in the laboratory. It consists of a metal cylinder of adequate capacity, which is filled with water. A metal tube pierces the bottom of the cylinder and rises above the level of the water. This metallic tube at one end is connected by a rubber tubing with mouth piece, in to which the subject breathes. The spirometer bell is another metallic cylinder of slightly smaller diameter. It is completely submerged in the water and its weight is balanced by a counter weight. The top of the spirometer bell is connected by a cord running over a pulley to a weight. The subject’s nose is clipped and through the mouthpiece breathes out into the spirometer and the air raises the bell. The volume of air is read from the scale marked on the pulley. The inner cylinder is raised initially and the subject may be asked to breathe in from it to measure inspiratory volumes.

PROCEDURE

1. Sterilize the mouthpiece with spirit (Fig. 18.1).
2. Adjust the pointer on the pulley to the zero mark with the drum pushed down and confirm that the pointer moves in the correct direction on pulling the drum up.
3. Breathe at tidal volume. At the end of normal inspiration, breathe out as usual but into the mouthpiece. The volume recorded is the tidal volume.
4. Readjust the drum at the lower position with pointer at zero. Breathe again at tidal volume. At the end of normal expiration, breathe out forcibly into the mouthpiece till you can breathe out no more. This volume is the expiratory reserve volume.
5. Readjust the spirometer as in step 4. After a few tidal breaths, breathe in maximally and breathe out maximally into the mouthpiece. This reading is the vital capacity (Fig. 18.2).

Fig. 18.1: The student’s spirometer

Fig. 18.2: A normal spirogram
QUESTIONS

1. Define tidal volume, vital capacity, IRV, ERV, maximum breathing capacity (MBC).
2. What are the factors, which will affect the vital capacity?
3. What is functional residual capacity and residual volume (FRC and RV)?
4. How would you measure FRC and RV?
5. What is respiratory reserve?
6. What is dyspneic index?
7. Draw a schematic diagram of a spirometer.
8. What is the effect of change in posture on vital capacity? Explain.
9. Give the normal values of vital capacity in an adult male and female.

SPIROMETRY

Name:    Age:
Sex:    Occupation:

<table>
<thead>
<tr>
<th>Lung volume/capacity</th>
<th>Reading 1</th>
<th>Reading 2</th>
<th>Reading 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vital capacity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Inspiratory reserve volume</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Expiratory reserve volume</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tidal volume</td>
<td></td>
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</tbody>
</table>

Inference
AIM
To determine the efficiency of respiratory system.

REQUIREMENTS
1. Sphygmomanometer
2. Stopwatch
3. Mini-peak flow meter (Figs 19.1A and B).

PROCEDURE
a. Breath-holding test: Ask the subject to breath out as deeply as possible and to take in air as much as possible. Now, ask him to hold his breath to the breaking point and determine the maximum time with a stop watch, noting the subjective symptoms during the experiment.

b. 40 mm Hg endurance test: After a deep expiration, ask the subject to inspire to the maximum. Now apply a clip to the nose and ask him to expire into the mercury manometer upto the level 40 mm Hg and to maintain it as long as possible.

c. Peak expiratory flow rate (PEFR): After a deep inspiration, ask the subject to expire forcefully into the mouthpiece of the mini-peak flow meter after adjusting the knob to the zero level.

d. Candle test (or) expiratory blast test.
Respiratory Efficiency Tests

Figs 19.1A and B: (A) Wright’s peak flow meter; (B) Mini-peak flow meter

RESULT

1. Maximum breath holding time = 
2. Maximum endurance time = 
3. PEFR = 

QUESTIONS

1. What is the standard breath holding time?
2. What is the normal maximal endurance time?
3. What is breaking point?
4. Define PEFR. What is the normal value?
5. Define VC, FEV₁. How is it useful?
6. Define MVV or MBC.
7. What is dyspneic index.
8. In what situation is respiratory efficiency test assessed?
9. What is expiratory blast test? (Candle test)
10. Does the PEFR and VC correlate well?

RESPIRATORY EFFICIENCY TESTS

<table>
<thead>
<tr>
<th>Test</th>
<th>Reading 1</th>
<th>Reading 2</th>
<th>Reading 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximum breath-holding test</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maximum endurance test</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PEFR</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Inference
Examination of the Cardiovascular System

AIM

Examination of the cardiovascular system (CVS) includes examination of the heart as well as the blood vessels. The cardiovascular system is assessed in the following order:

1. Arterial pulses
2. Blood pressure
3. Venous pulses
4. Examination of the precordium (part of the chest which overlies the heart).

ANATOMICAL LANDMARKS

1. Sternal angle or the angle of Louis: This marks the junction of the manubrium with the body of the sternum. The second costal cartilage articulates with the sternum at this level. The space immediately below this is the second intercostal space.
2. Midclavicular line: Vertical line descending from the center of clavicle.
3. Anterior, mid and posterior axillary lines: These are vertical lines descending respectively from the anterior border, the center and posterior border of the axilla.

ARTERIAL PULSES

The presence or absence of the main peripheral pulses—the radial, brachial, carotid, femoral, popliteal, posterior tibial and dorsalis pedis pulses are noted. The volume of each is compared with the other side.
Examination of the Cardiovascular System

METHOD OF EXAMINING THE PULSE

The pulse is usually felt at the wrist and over the radial artery because of its superficial position. It is best felt with the subject’s forearm slightly pronated and the wrist flexed. The examiner’s three fingers, viz. the index, middle and ring finger should be over the radial artery. Both radial arteries are palpated simultaneously to detect the irregularities of the pulse on the two sides. The following observations are to be made with regard to cardiac function.

1. Rate of pulse: It is stated as number of beats per minute. The normal pulse rate is about 70–80 per minute (to be precise 60–100 beats is considered normal). Increase in pulse rate (tachycardia) occurs during fever and in thyrotoxicosis, decrease in pulse rate (bradycardia) occurs in myxedema and following regular athletic training. Abnormalities of the function of SA or AV nodes may reduce the heart rate. Pulse deficit: The difference between the heart rate and the pulse rate is called as pulse deficit.

2. Rhythm: The rhythm could be regular or irregular. It could be regularly irregular or irregularly irregular. Regularly irregular pulse would be seen during atrial extrasystole which occurs at a regular interval. Irregularly irregular pulse would be seen in atrial fibrillation.

3. Character: The character or form of the pulse wave is studied by palpating the carotid pulse.
   a. Collapsing (water-hammer) pulse: Characterized by a rapid upstroke and descent of the pulse wave. It would be seen in large arteriovenous communications (Figs 20.1A to C).
   b. Pulsus paradoxus: There is a marked diminution in an arterial pressure as seen in pericardial effusion.
   c. Pulsus alternans: Occur when the left ventricular muscle is severely failing. The ventricle beats strongly and then weakly, in successive beats of normal rhythm.

Figs 20.1A to C: Types of pulses: (A) Normal; (B) Anacrotic; (C) Water hammer pulse

4. Volume: This gives a rough guide to pulse pressure, which depends upon the stroke volume and the compliance of the arteries. With normal vessels, the pulse volume gives an indication of the stroke volume.

5. Nature of the vessel wall: Vessel wall is thickened in old age due to atherosclerosis.

6. Radiofemoral delay: Delay of the femoral pulse compared with the right radial pulse is found in coarctation of the aorta.

BLOOD PRESSURE

It is to be measured by the following methods:
1. Palpatory
2. Auscultatory
VENOUS PULSES

Examination of the neck veins, especially the internal jugular vein is done with the patient in a good light and reclined at an angle of about 45°. The neck is supported so that the neck muscles, especially the sternocleidomastoid are relaxed. The normal slight pulsations of the veins and the three waves (a, c and v waves) are distinguished.

There is a mean level and the perpendicular height of this level above the right atrium indicates the mean hydrostatic pressure within the right atrium. The normal upper limit is 4 cm vertically above the sternal angle. This is about 9 cm above the right atrium. In a healthy person reclining at an angle of 45°, the mean level will be invisible because it is just below the clavicle.

Elevation of the JVP indicates elevation of the right atrial pressure.

Causes of Elevation of JVP

1. Congestive heart failure
2. Pulmonary embolism
3. Constrictive pericarditis
4. Tricuspid valve disease.

Arterial pulsation may also be visible in the neck and has to be distinguished from various pulsations:

1. Venous pulse has a definite upper level
2. This level falls during inspiration
3. Firm but gentle pressure on the upper abdomen will raise the level due to transient increase in the venous return (hepatojugular reflex)
4. Venous pulsation is not palpable
5. It is more sinuous and less sharp than arterial pulsation.

The venous pulsations are more easily recognized in external jugular veins than in internal jugular veins but the former is less reliable because:

1. Of the presence of venous valves within the external jugular system
2. It passes through more fascial planes so that it is more likely to be affected by external compression.

EXAMINATION OF THE PRECORDIUM

The precordium is to be examined in the following order:

1. Inspection
2. Palpation
3. Percussion
4. Auscultation.

INSPECTION

The subject is stripped up to the waist and the chest wall is examined for any deformities. Some of the common deformities include pigeon chest, barrel chest (cylindrical chest), funnel shaped chest (exaggeration of the normal hollow over the lower end of sternum).
Examination of the Cardiovascular System

The position of trachea is noted. The position and the extent of the apical impulse are made out. The apical impulse is the lowermost and the outermost point of definite cardiac pulsation. The normal position of the apical beat is 1 cm internal to the midclavicular line in the left fifth intercostals space.

Any other pulsations or engorged veins over the chest wall or the presence of extrathoracic pulse are also noted for.

PALPATION

The position of trachea is confirmed. This method confirms the inspection findings. The apical impulse is to be felt initially with the palmar aspect of the hand and finally localized by the digital method. Abnormal impulse, if any, like ‘Thrill’ is to be made out by palpation. ‘Thrill’ is palpable murmur.

The apex beat may be displaced due to disease of the surrounding viscera which ‘push’ or ‘pull’ from the usual site.
- Causes of ‘pushing’ are: (1) Pleural effusion (2) Pneumothorax.
- Causes of ‘pulling’ are: (1) Pulmonary fibrosis (2) Collapse of lung.

Enlargement of the heart can also cause displacement of the apex beat outwards.

PERCUSSION

The left border of the heart is percussed first and then the right border.

**Left border of the heart:** The apex beat is to be marked by percussing from a resonant area in the axilla and moving inside at the fifth intercostal space. Likewise the left border is to be marked by percussing intercostal space above, moving from axilla inwards.

Right border of the heart: Percussion is to be done from above downwards in the intercostal spaces on the right side till the liver dullness is reached. Then percussion in the intercostal space just above the liver dullness is made from outside inwards to mark the lower boundary of the organ. Similar percussion is to be made on the intercostal space at a higher level, from outside inwards to mark the right border.

Joining the lower limit of the left and right borders, and the upper limit of the same, gives the inferior and superior borders of the heart. Superior border may be percussed for starting at the topmost intercostal space on the left side.

AUSCULTATIONS

Auscultation is done by means of a stethoscope. The important auscultatory areas are:

1. **Mitral area:** Corresponds to the apex beat.
2. **Tricuspid area:** Lies just to the left of the lower end of sternum.
3. **Aortic area:** Lies to the right of the sternum in the second intercostal space.
4. **Pulmonary area:** Lies to the left of the sternum in the second intercostal space.

These areas are customarily called by the name of the valve from which murmurs and sounds arise. This does not correspond to the anatomical location of the valves (Figs 20.2A and B).
Figs 20.2A and B: (A) Anatomical position of cardiac valves; (B) Physiological basis of heart sounds
HEART SOUNDS

Clinically, two heart sounds are easily distinguishable. The first heart sound is due to the closure of the AV valves (mitral and tricuspid), usually pronounced as ‘Lubb’. It synchronizes with the carotid pulse beat. The second heart sound is due to the closure of the semilunar valves (aortic and pulmonary), usually pronounced as ‘Dub.’

In disease, the following deviations from the normal occur:
1. The sounds may have different intensity, either increased or decreased
2. The sounds may be abnormally split
3. Low frequency sounds in diastole—third and fourth heart sounds may be heard
4. Additional sounds, often related to abnormal valves may be heard.

| OSCE-I |
|---|---|
| Procedure: Examine the pulse of the subject |
| Make the subject comfortable and instruct him/her properly |
| Keep the hand in semipronated and semiflexed position |
| Place the three fingers (index, middle and ring fingers) on the radial artery over the flexor aspect of the radial styloid process |
| Record the pulse rate for complete one minute |
| Also look for the rhythm, volume, character of the pulse, radio-femoral delay, radio-radial delay, and condition of the vessel wall |
| Palpate other peripheral pulsations |
| Report the observation correctly |

| OSCE-II |
|---|---|
| Procedure: Examine the apex impulse of the subject |
| Make the subject comfortable and instruct him/her properly |
| Expose the chest completely |
| Inspect the most prominent pulsation over the precordium tangentially |
| Report the observation correctly |

| OSCE-III |
|---|---|
| Procedure: Examine the apex beat of the subject |
| Make the subject comfortable and instruct him/her properly |
| Expose the chest completely |
| Warm the hands and keep the palmer aspect of the hand over the area of the apex impulse inspected previously and feel for any prominent pulsation |
| Confirm your finding by placing the ulnar border of the right hand |
| Locate this prominent pulsation as apex beat with the tip of the thumb |
| Report the position of the apex beat in relation to the intercostals spaces and midclavicular line |
OSCE-IV

Procedure: Examine the heart sounds of the subject

Make the subject comfortable and instruct him/her properly
Expose the chest completely
Locate all four auscultatory areas (mitral, tricuspid, aortic and pulmonary)
Place the diaphragm of the stethoscope over the areas sequentially and auscultate for the heart sounds
Confirm the first heart sound by palpating the carotid artery simultaneously
Report the observation correctly

QUESTIONS

1. What is precordium?
2. How will you identify the intercostal space?
3. What is the normal shape of the chest?
4. What is orthopnea and paroxysmal nocturnal dyspnea?
5. Name the four auscultatory areas and their exact positions?
6. What is apical impulse? What is the normal site?
7. Why JVP is seen on the right side of the neck? What is their normal level?
8. When does JVP get increased? Name the jugular venous waves after drawing it.
9. What are the types of abnormal pulses you know?
10. Why is radial artery always chosen? What information can you get on palpating the artery?
11. When does the vessel wall get thickened?
12. What are the types of irregular pulse you know?
13. What is a murmur?
14. Clinically, how will you identify the 1st heart sound?
15. When is 1st heart sound loud?
16. When is 2nd heart sound loud?
17. What is the cause of IIIrd HS and IVth HS? What is its significance?
18. When do you get a:
   - Systolic murmur
   - Diastolic murmur
   - Continuous murmur.
19. What is phonocardiogram and echocardiogram?
20. What is the physiological basis for the “split” of the 2nd heart sound?
21. What are the heart sounds other than those mentioned above caused by?
22. Which portion of the heart corresponds to the:
   a. Apex
   b. Parasternal area.
23. What is a “Bruit”?
24. Enter the timing of the murmurs in the table provided:

<table>
<thead>
<tr>
<th>Valves</th>
<th>Lesion</th>
<th>Timing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aortic</td>
<td>Stenosis</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Regurgitation</td>
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<tr>
<td>Pulmonary</td>
<td>Stenosis</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Regurgitation</td>
<td></td>
</tr>
<tr>
<td>Mitral</td>
<td>Stenosis</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Regurgitation</td>
<td></td>
</tr>
<tr>
<td>Tricuspid</td>
<td>Stenosis</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Regurgitation</td>
<td></td>
</tr>
</tbody>
</table>

25. What other positions would you like to put the patient to feel the apical impulse?

**CARDIOVASCULAR SYSTEM EXAMINATION**

Name:     Age:
Sex:     Occupation:

**General Examination**
- Build and nutritional status
- Pallor
- Icterus
- Cyanosis
- **Arterial pulse**: Rate, rhythm, volume, character, condition of vessel wall, radio-femoral delay, pulse-apex deficit, all peripheral pulses.
- BP
- Temperature
- JVP.

**Systemic Examination**

**Inspection**
- Trachea
- Apex-beat
- Venous engorgement
- Chest and spine shape
- Scars

**Palpation**
- Trachea
- Apex-beat
- Thrills

**Percussion**
- Right border
- Left border
- Superior border

**Auscultation**
- Mitral area
- Tricuspid area
- Pulmonary area
- Aortic area
- Other sounds

Report
DEFINITION

Blood pressure (BP) is the lateral pressure exerted by the column of blood on the wall of the artery.

AIM

To determine the blood pressure of the given subject at rest and after moderate exercise.

APPARATUS

Sphygmomanometer and stethoscope.

PRINCIPLE

The pressure of blood in the artery (brachial artery) is balanced against the pressure of air in a rubber cuff surrounding the artery. The pressure of air in the cuff is then measured by means of a mercury manometer.

METHODS

1. Palpatory method
2. Auscultatory method
3. Oscillatory method.
Determination of the Blood Pressure

PROCEDURE

1. Palpatory method: The subject is asked to sit on a stool. The cuff is tied around the upper arm with the lower border of the cuff not less than 2.5 cm above the cubital fossa. The outlet valve of the bulb is closed. The radial pulse is palpated while the cuff is being inflated to a pressure slightly above the level at which the radial pulsation is no longer felt. The pressure at which the pulsation was obliterated is read in the mercury manometer. The outlet valve is opened. The manometric reading is noted at the point where the pulsation reappears. The average of the two readings gives the systolic pressure. The diastolic pressure cannot be determined by this method.

2. Auscultatory method: By palpatory method, only the systolic blood pressure could be measured. By auscultatory method, both the systolic and diastolic blood pressure can be measured. The chest piece of the stethoscope is placed over the brachial artery. The pressure in the cuff is raised above the systolic pressure (by about 30 mm Hg) previously determined by the palpatory method. The pressure is then lowered gradually (2–3 mm per second). The sounds that are heard are the Korotkoff’s sounds. The first sound (phase one) that occurs is a sharp tapping sound, indicates the peak systolic pressure, the second and third phases, initially murmurous in quality and then louder and more banging, are due to turbulent flow of blood through a partially occluded vessel. In the fourth phase, the sound becomes muffled and dull and the fifth phase accurately gives true diastolic pressure, which is disappearance of the sound (Figs 21.1A to D).

Figs 21.1A to D: (A) Mercury sphygmomanometer; (B) Aneroid sphygmomanometer; (C) Riva-Rocci cuff; (D) Korotkoff’s sounds
3. **Oscillatory method:** This is another method of determining blood pressure. By this method, both the systolic and diastolic blood pressure are determined. The pressure at which oscillations appear in the mercury manometer gives the systolic pressure and the pressure at which it disappears give the diastolic blood pressure. However, this method is not accurate.

**Important precautions in the use of sphygmomanometer:**
1. The manometer should be placed at the level of the heart.
2. The lower border of the cuff should be 2.5 cm above the cubital fossa. For children, a narrow cuff should be used.
3. Blood pressure should be preferably taken in the left arm.

**NORMAL VALUES**

The average systolic pressure in healthy adults is 100–140 mm Hg. The average diastolic pressure is 60–90 mm Hg. In children it is closer to the lower end of the scale and in the elderly, it reaches or even exceeds the higher figure. The difference between the systolic and diastolic pressure is the pulse pressure 30–60 mm Hg.

**Physiological Variations**

Blood pressure is slightly lower in women than men. Persons with slender build have got a lower blood pressure than those of heavy build. During sleep, systolic pressure is less. Emotional excitement and muscular exercise cause an increase in the blood pressure. It is also increased after meals. The blood pressure especially the diastolic is highest in the standing position, lower in the sitting and lowest while the subject is lying down.

**Pathological Increase in Blood Pressure**

1. Essential hypertension
2. Adrenal tumor
3. Hyperthyroidism
4. Pheochromocytoma.

**Pathological Decrease in Blood Pressure**

1. Shock
2. Hypothyroidism
3. Adrenal insufficiency.

**DISCUSSION**

Blood pressure is the lateral pressure exerted by the column of blood on the wall of the vessels while flowing through it.
**Determination of the Blood Pressure**

**Systolic pressure** is the maximum pressure in the arteries during systole. It indicates:

a. The extent of work done by the heart
b. The force with which the heart is working
c. The degree of pressure which the arterial walls have to withstand. Normal systolic pressure approximately is 100 + age.

**Diastolic pressure** is the minimum pressure at the end of ventricular diastole. It is the measure of constant stretch to which walls of the arteries are subjected. It is more important than systolic pressure because:

a. It is less fluctuating
b. It is the constant load against which the heart has to work
c. It is the pressure of peripheral resistance and depends mainly on the tone of the arteries.

**Pulse pressure** is the difference between the systolic and diastolic pressures. It is the rise in pressure caused by the ejection of blood into the aorta by ventricular contraction. It is a measure of stroke volume and compliance of arteries. Mean arterial pressure is the average pressure present throughout the cardiac cycle. It is responsible for pushing the blood through the systemic circulatory system. It is equal to diastolic pressure + 1/3 pulse pressure.

**EFFECT OF MODERATE EXERCISE ON BLOOD PRESSURE**

During exercise, there is a moderate increase in systolic blood pressure. This is due to an increase in cardiac output caused by an increased heart rate and myocardial contractility (stroke volume increases) due to increased sympathetic activity, and increased venous return. The increase in blood pressure is not proportionate to the increase in cardiac output because there is a reduction in total peripheral resistance. The effects of vasoconstriction in inactive regions are overcome by vasodilatation in active muscles. Hence, the diastolic pressure at the pre-exercise level is slightly reduced. Pulse pressure is increased.

<table>
<thead>
<tr>
<th>OSCE-I</th>
<th>Procedure: Tie the sphygmomanometer cuff for the measurement of blood pressure</th>
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<tbody>
<tr>
<td></td>
<td>Take the appropriate cuff size, according to the built of the subject</td>
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<tr>
<td></td>
<td>Instruct the subject and position him/her in the sitting position with arm at the level of the heart and well supported</td>
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<tr>
<td></td>
<td>Expose the arm properly and palpate the brachial artery in the cubital fossa</td>
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<td></td>
<td>Wrap the cuff around the medial aspect of the arm ensuring that the middle of the cuff lies over the brachial artery</td>
</tr>
<tr>
<td></td>
<td>Ensure that the cuff does not overlie the cubital fossa and is not too tight or too loose</td>
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</table>

<table>
<thead>
<tr>
<th>OSCE-II</th>
<th>Procedure: Estimate the systolic blood pressure by palpatory method</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Instruct the subject and tie the cuff properly</td>
</tr>
<tr>
<td></td>
<td>Place the sphygmomanometer at the level of the heart</td>
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<tr>
<td></td>
<td>Feel the radial pulse</td>
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<tr>
<td></td>
<td>Inflate the cuff till the pulse disappears. Note the reading and further inflate the cuff by 10–20 mm Hg</td>
</tr>
<tr>
<td></td>
<td>Deflate the cuff at appropriate speed till the pulse appears again. Note the reading as systolic blood pressure</td>
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<tr>
<td></td>
<td>Deflate the cuff completely</td>
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</tbody>
</table>
QUESTIONS

1. Define
   a. Systolic blood pressure
   b. Diastolic blood pressure
   c. Pulse pressure
   d. Mean arterial pressure.
2. Mention the different methods of determining blood pressure.
3. What are the physiological factors affecting blood pressure?
4. What is the value of normal blood pressure? Give its acceptable range.
5. Comment on the mean values you obtained from your group.
   a. Is it within the normal range?
   b. Are there any members of your group who have abnormal values? Are there any attributable causes?
6. What are the possible sources of error in this experiment?
7. Why is the systolic pressure determined by the palpatory method lower than that determined by the auscultatory method?
8. Why is it important to first check the BP by the palpatory method always?
9. How are the sounds of Korotkoff produced?
10. What are the precautions to be taken while measuring blood pressure in:
    a. Children
    b. Obese individuals.
11. Explain how the BP readings change in the following conditions. Explain the basis for these changes.
    a. When the arm is raised above the head
    b. If the cuff is applied to the thigh.
12. While calculating the mean arterial pressure, what formula do we use and why?
13. Give two conditions when blood pressure is abnormally
    a. Increased
    b. Decreased.
14. Briefly describe two other methods of measuring blood pressure.
15. What is the physiological basis for postural hypotension?
16. What are the BP changes you observe during exercise in the subjects?
17. What are the changes in heart rate you observe after exercise? What is the physiological basis for these changes? How are they mediated?
18. Who first recorded the blood pressure?
19. Who invented the mercury manometer?
20. How is hypertension classified? What are their percentages?
Determination of the Blood Pressure

BLOOD PRESSURE

Name:               Age:               
Sex:               Occupation:          

<table>
<thead>
<tr>
<th>Procedure</th>
<th>Pulse (beats/ min)</th>
<th>Systolic BP (mm Hg)</th>
<th>Diastolic BP (mm Hg)</th>
<th>Pulse pressure (mm Hg)</th>
<th>MAP (mm Hg)</th>
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<tbody>
<tr>
<td>Before exercise</td>
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<tr>
<td>After exercise</td>
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Effect of exercise on BP

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<tr>
<th>Procedure</th>
<th>Pulse (beats/ min)</th>
<th>Systolic BP (mm Hg)</th>
<th>Diastolic BP (mm Hg)</th>
<th>Pulse pressure (mm Hg)</th>
<th>MAP (mm Hg)</th>
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<td>Lying</td>
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<tr>
<td>Sitting</td>
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Effect of posture on BP (from lying to sitting)

<table>
<thead>
<tr>
<th>Procedure</th>
<th>Pulse (beats/ min)</th>
<th>Systolic BP (mm Hg)</th>
<th>Diastolic BP (mm Hg)</th>
<th>Pulse pressure (mm Hg)</th>
<th>MAP (mm Hg)</th>
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<tbody>
<tr>
<td>Lying</td>
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<tr>
<td>Standing</td>
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Effect of posture on BP (from lying to standing)

<table>
<thead>
<tr>
<th>Procedure</th>
<th>Pulse (beats/ min)</th>
<th>Systolic BP (mm Hg)</th>
<th>Diastolic BP (mm Hg)</th>
<th>Pulse pressure (mm Hg)</th>
<th>MAP (mm Hg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lying</td>
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<tr>
<td>Standing</td>
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Inference
AIM

To record and analyze electrocardiogram (ECG) of a normal subject.

REQUIREMENTS

Electrocardiograph, ECG leads (electrodes), ECG paper, straps and ECG jelly.

DESCRIPTION

Electrocardiograph

This is the equipment that can pick up, amplify and record from the body surface the electric potentials generated by the heart. Einthoven (1903) devised and used string galvanometer for this purpose. This instrument must be sensitive to potential changes of the order of micro-volts and have a frequency response of 50–100 Hz. At present, transistors, amplifiers and cathode ray tubes are used for this purpose. Biopotentials are amplified to drive a galvanometer. Recording is obtained on a heat-sensitive paper using a hot stylus, an ink-writing pen or a photographic paper. Speed of the paper can be adjusted at 25 or 50 mm/sec. Using a polygraph or an oscilloscope, recording can be obtained at higher speeds. The sensitivity can be adjusted to $1 \text{ mv} = 1 \text{ cm}$ (or $2 \text{ cm}$ or $0.5 \text{ cm}$).

Electrocardiogram

It is the graphic record of algebraic sum of action potentials developed during a cardiac cycle and recorded from the body surface: ECG paper has horizontal and vertical rulings at 1 mm distance. Every fifth line (5 mm) is thicker. At a paper speed of 25 mm/sec, 1 mm (vertical lines) = 40 m sec. At a calibration of $1 \text{ cm} = 1 \text{ mv}$, 1 mm (horizontal lines) = 0.1 mV (Figs 22.1 and 22.2).
ECG LEADS

A lead or an electrode is a metal plate applied snugly over an appropriate body part. For better contact, saline or ECG jelly is applied after the skin surface is cleaned thoroughly. The leads are classified as (Fig. 22.3):

1. Direct leads
2. Indirect leads
   a. Limb leads: Unipolar and bipolar
   b. Chest leads: Unipolar and bipolar
   c. Esophageal leads
DIRECT LEADS

Direct leads are applied directly to the heart during surgery or an experiment. An electrode can also be fixed on the tip of a catheter and introduced into the heart via a blood vessel. This is done for recording His bundle electrocardiogram (HBE).

INDIRECT LEADS

Bipolar Limb Leads

These are leads I, II and III. Here, both the electrodes pick up the potentials from the body surface. The electrodes are attached to the right arm, left arm and left foot as indicated in the Einthoven triangle.

Unipolar Limb Leads

These are aVL, aVR and aVF. Here ‘a’ denotes augmented (i.e. aVR is 50% more amplitude than VR). ‘V’ denotes voltage and ‘R’, ‘L’ and ‘F’ indicate that the exploring electrode is on the right arm, left arm and left foot respectively. The another one electrode is connected to the right foot through a high resistance.

Unipolar Chest Leads

These are V1 to V6. Here, the passive or indifferent electrode is connected to Wilson terminal which is maintained at near zero potential and the exploring electrode is on the chest surface.

BIPOLAR CHEST LEADS

Here, one electrode is positioned on the chest wall as in V1 to V6 and the other electrode is on the right arm. Records obtained by bipolar chest leads are very similar to those obtained by unipolar chest leads. For routine electrocardiography, 12 leads (I, II, III, aVR, aVL, aVF and V1 to V6) are recorded and analyzed.

Esophageal Leads

Here, a miniature electrode is fixed on the tip of an esophageal catheter and positioned in esophagus so as to be close to cardiac chambers (e.g. posterior wall of right atrium). The lead is denoted by the letter E and a number indicates the distance of the electrode from the incisor teeth. For example, E20 indicates that the esophageal electrode is 20 cm from the incisor teeth.

PROCEDURE

Before fixing the lead, clean the skin thoroughly and apply ECG jelly. The subject should lie down supine and be completely relaxed to avoid muscle potentials. The right leg is earthed. Record in order of leads I, II, III, aVR, aVL, aVF and V1 to V6 (Fig. 22.4).
Calculation of heart rate (HR):

\[
HR = \frac{60 \text{ seconds}}{\text{RR interval (in seconds)}}
\]

At a paper speed of 25 mm/second,

\[
HR = \frac{25 \times 60}{\text{RR interval in no. of small squares in ECG}} = \frac{1500}{\text{RR (mm)}} = \frac{300}{\text{RR interval in no. of large squares in ECG}}
\]

**CALCULATION OF VENTRICULAR VECTOR (MEAN ELECTRICAL AXIS)**

Calculate the net value of the QRS (R-S in mm) from leads I, II and III. Draw an equilateral triangle. From the midpoint of its each arm, mark in millimeters on the + and – sides. Draw vertical lines from the midpoints so that they meet at the midpoint of the triangle. Mark on each limb (lead) the net QRS values and draw vertical lines from these points. Let the lines meet at a point. Connect this point with the midpoint of the triangle. This gives the magnitude (depending on the length) and the direction (depending on the angle) of the net ventricular complex. Normal axis is about +60° (range: –30° to +110°).

**QUESTIONS**

1. List the differences between action potential and ECG.
2. With the help of diagrams, show how various leads are connected for recording ECG.
3. Draw a normal lead II ECG and label it.
4. Measure and tabulate durations and amplitudes of various waves, intervals and segments from lead II of ECG.
5. Calculate QRS axis using Einthoven’s triangle.
6. Name the physiological and pathological conditions that shift the ventricular axis to the
left/right.
7. Calculate heart rate from the recorded ECG.
8. Draw a diagram showing ECG, left ventricular pressure changes and heart sounds on the same time scale.
9. What is ’J’ point?
10. What conditions cause ST elevation and ST depression?
11. What are the causes of tall T-waves and inverted T-waves?
AIM

To check the following sensations in the given subject:
1. Tactile sensibility—touch, localization, discrimination
2. Pain and temperature
3. Position and vibration sense
4. Stereognosis.

PROCEDURE

The following tests are done with the subject’s eyes closed.

TACTILE SENSIBILITY

This includes light touch, pressure, tactile localization and discrimination.

Light touch: Corresponding points of the different parts of the body on the two sides are touched with a wisp of cotton wool and the subject is asked to say whether he feels the sensation of touch.

Pressure touch: It is tested with the point of a finger or any blunt object.

Tactile localization: The subject is asked to localize the stimulus by indicating the exact position of the spot touched with eyes closed. Sensibility to touch may be altered in various ways.
1. If entirely abolished, it is called anesthesia
2. Partial loss is called hypoesthesia
3. When touch produces painful, irritating, tingling sensation, it is referred to as hyperesthesia

**Tactile discrimination:** The two points of a compass are applied to the finger tips, arms and to the back of the subject. The minimum distance in millimetres at which the two points of the compass are felt separately is noted. There is a marked difference in the distances at which the two points of the compass are felt separately at the finger tip, palmar surface of finger, the forearm or middle of the back. Normally 2 mm of separation of the points can be recognized as two separate stimuli on the finger tips, 1 cm on the palmar surface and more in other area (Figs 23.1 to 23.3).
PAIN

The sensation of superficial pain is examined by giving pricks with a steel pin to the different parts of the body. The subject is asked to describe the nature of sensation.

Deep pain is tested by pressing a muscle between thumb and index finger or the tendo-achilles and the sensation felt is enquired about.

Absence of pain sensibility is termed as analgesia, partial loss is termed as hypoalgesia and an exaggerated sensibility is called as hyperalgesia.
TEMPERATURE

In two test tubes, warm and cold water is taken. The test tubes are then applied to different parts of the body separately. The subject is asked to tell whether he feels warmth or cold. Test tube containing cold water is tested first. May use thermoesthesiometer (Figs 23.4A and B).

VIBRATION

A vibrating tuning fork of 128 Hz is applied to the various bony prominences of the body and an enquiry is made regarding the sensation, felt by the subject. Normally he would be able to feel the vibration. Usually the tuning fork is placed on dorsum of great toe, lateral malleolus, dorsum of the finger and top of the acromion (Figs 23.4A and B).

SENSE OF POSITION AND PASSIVE MOVEMENT

With the eyes of the subject closed, one of his fingers are flexed and extended passively. The subject is asked to say whether he felt any movement; if so, what movement it was. In this way, different joints of the body are moved and the sense of passive movement is tested. The finger or a toe of the subject is moved up and down and is to be held there. The position of the part moved is enquired from the subject. Normally the subject would be able to appreciate the passive movements and the position of his limb. The subject may also be asked to imitate with the opposite limb or digit. The position of the limb is also tested (Fig. 23.5).
**STEREOGNOSIS**

With the subject's eyes closed, he is given some article which is commonly used, e.g. coins, pens, etc. in his hand and he is asked to name them. Normally, he would be able to appreciate them with his eyes closed by their size, weight, shape, texture, temperature, etc. Loss of this power is known as astereognosis. The subject should use one hand only to feel the given object.

**ABNORMAL SENSATION**

Paresthesias are the various sensations experienced in the absence of any outward stimulus; there is a feeling of pins and needles, of numbness or insects crawling over the body.

### OSCE-I

**Procedure:** Demonstrate the procedure of testing the fine touch in the given subject

- Make the subject comfortable and instruct him/her properly
- Ask the subject to close both the eyes and to say 'yes' whenever he/she feels touch sensation
- Take Von Frey's hair asthesimeter/cotton wisp and touch over the different dermatomes sequentially
- Compare on the other side
- Report correctly

### OSCE-II

**Procedure:** Demonstrate the procedure of testing the deep pain in the given subject

- Make the subject comfortable and instruct him/her properly
- Squeeze a large muscle belly between the thumb and the index finger
- Enquire about the sensation felt
- Compare on the other side
- Report correctly

### OSCE-III

**Procedure:** Demonstrate the procedure of testing the vibration sense in the given subject

- Make the subject comfortable and instruct him/her properly
- Vibrate a tuning fork (128 Hz) and place the base of the tuning fork on bony prominences
- Enquire about the sensation felt
- Compare on the other side
- Report correctly
OSCE-IV

Procedure: Demonstrate the procedure of testing the position sense in the given subject

Make the subject comfortable and instruct him/her about the flexed and extended positions of the thumb/great toe

Ask the subject to close the eyes

Stabilise the thumb/great toe by holding it at the sides of the distal interphalangeal joint

Do the passive flexion and extension of thumb/great toe and ask the subject about the movement felt

Ask the subject to imitate the same movement actively on the opposite limb

Report correctly

QUESTIONS

1. What are epicritic and protopathic sensations?
2. What is proprioception? Name the receptors.
3. What is the receptor for pain and cold, hot, touch sensation?
4. What are pacinian corpuscles?
5. What is stereognosis?
6. Define analgesia, anesthesia, hyperalgesia and hyperesthesia.
7. Name the tract carrying pain and touch sensation.
8. What is the Brodmann’s area number for primary sensory area and secondary sensory area?
9. What is tactile localization and discrimination?
10. Name the nucleus of the thalamus for conscious sensations.
11. Differentiate between fine and crude touch?
12. Can tuning fork with higher frequency (above 256 Hz) be used to test vibration sense?
13. Name few clinical conditions where vibration sense is lost.
The image contains a page from a document titled "Examination of Sensory System". The page outlines a sensory system examination with sections for general examination, higher functions, and sensory system examination. There is a table with various sensory parameters such as touch, pain, temperature, joint position sense, vibration sense, stereognosis, and Romberg's sign, along with columns for upper limb (left and right) and lower limb (left and right). The table indicates whether each parameter is intact or not. The page also includes an inference section at the end.
The motor system should be examined under the following aspects:
1. Bulk or nutrition of the muscles
2. Tone of the muscles
3. Strength of the muscles
4. Reflexes
5. Coordination of movements
6. Gait
7. Involuntary movements.

**BULK OF MUSCLES**

Wasted and atrophic muscles are not only smaller but also softer and more flabbier than normal when they are contracted. Bulk of muscles are also assessed by measuring the circumference of the limbs with a tape at certain points with reference to definite bony landmarks and comparing on both sides.

The different muscles of the body are inspected and palpated and any wasting of muscles is to be noted. Similarly any contracture of the muscles is also to be observed. Muscle wasting and contracture are characteristic of poliomyelitis and muscular dystrophy (Figs 24.1A to C).
TONE OF THE MUSCLES

Muscle tone is a state of tension or contraction found in healthy muscles. It is tested by passively moving the limbs at different joints and noting the resistance offered by the muscles during this process (Figs 24.2A and B).

A. **Hypertonia**—increase in tone. They are of two types.
   1. **Clasp knife/spastic rigidity**—due to lesion in the corticospinal system
   2. **Cog-wheel/extrapyramidal rigidity**—due to disease of the basal ganglia.

B. **Hypotonia**—decrease in tone, e.g., poliomyelitis.

STRENGTH OF THE MUSCLES

Tested by asking the subject to carry out the action of muscle against resistance offered by examiner (Figs 24.3A to C).
TESTING MUSCLES OF THE UPPER LIMB

1. **Abductor pollicis brevis**: The subject is asked to abduct the thumb against resistance in a plane at right angle to palmar aspect of the index finger.

2. **Opponens pollicis**: The subject is asked to touch the tip of his little finger with the point of his thumb against resistance.

3. **First dorsal interosseous**: The subject is asked to abduct his index finger against resistance.

4. **Interosseous and lumbricals**: The subject is asked to flex the metacarpophalangeal joints and to extend the distal interphalangeal joints. The subject is also asked to abduct the finger. In ulnar nerve paralysis, “Claw hand” deformity is produced. The first phalanges are overextended and the distal two are flexed. The fingers are slightly separated.

5. **Flexors of the fingers**: The subject is asked to squeeze the examiners index and middle finger.

6. **Flexors of the wrist**: The subject is asked to bring the tips of his fingers towards the front of the forearm.

7. **Extensor of the wrist**: The subject is asked to make a fist and the examiner tries to forcibly extend the wrist against the subject’s effort to maintain this posture. Weakness or paralysis of extensor (Wrist drop) occurs in radial nerve palsy.

8. **Brachioradialis**: The arm is placed midway between the prone and supine position and the subject is asked to bend the forearm against resistance. The muscle (if healthy) will stand out prominently at its upper part.

9. **Biceps**: The subject is asked to bend up the forearm against resistance with the forearm in full supination. The muscle will stand out clearly.

10. **Triceps**: The subject is asked to straighten out his flexed forearm against resistance.

11. **Supraspinatus**: The subject is asked to lift his arm straight at right angles to his side. The first 30° of this movement is carried out by the supraspinatus. The remaining 60° is by the deltoid.

12. **Deltoid**: The anterior and posterior fibers help to draw the abducted arm forwards and backwards respectively. The middle fibers abduct the shoulder (Fig. 24.4).

13. **Infraspinatus**: The subject is asked to tuck his elbow into his side with the forearm flexed at right angle. He is then asked to rotate outwards against resistance. The elbow being held against the side throughout. The muscle can be seen and felt to contract.
14. **Pectorals**: The subject is asked to stretch his arms out in front of him and then clap his hands together while the examiner endeavors to hold them apart.

15. **Serratus anterior**: The subject is asked to push forward his hands against resistance such as wall. If there is paralysis of serratus anterior, there would be ‘winging’ of the scapula (Fig. 24.4).

16. **Latissimus dorsi**: The subject is asked to clasp his hands behind his back while the examiner standing behind the patient offer passive resistance to the downward and backward movement (Fig. 24.4).

**Testing Muscles of the Trunk**

**Abdominal muscles**: The subject lying in a spine position is asked to lift up his head from the pillow against resistance (Fig. 24.5). In paralysis of the lower segment, the umbilicus moves upward, but when the upper segment is involved, the umblicius is pulled downwards. This is called as Beevor’s sign.

In weakness of the abdominal muscles, the patient would be unable to sit up in bed from the supine position without the aid of his arms (Babinski’s rising up sign).

**Muscles of the back**: The subject is asked to lie on his face and is asked to raise his head from the bed by extending the neck and back. The muscles stand out prominently.

**Trapezius**: The patient is asked to shrug his shoulders against resistance.
Testing Muscles of the Lower Limb

Dorsiflexion and plantar: Flexion of the feet and toes are tested by asking the subject to elevate or depress the distal foot against resistance.

Extensors of the knee: The subject is asked to bend the knee. And then pressing the shin with the examiner’s hand the patient is asked to straighten it out again.

Flexors of the knee: The patient is asked to raise the leg up from the bed, the thigh being supported with the examiners left hand and the ankle with the right. The patient is then asked to bend his knee.

Extensors of the hip: With the knee being extended, the patient’s foot is lifted off the bed. The subject is asked to push it down against resistance.

Flexors of the thigh: With the leg extended, the patient is asked to raise his leg of the bed against resistance.

Abductors of thigh: The subject’s legs are placed together. He is then asked to separate them against resistance.

Adductors of the thigh: Abduct the limbs and ask the subject to bring them back to midline against resistance.

Rotators of the thigh: With the lower limb extended on the bed, the patient is asked to roll it outwards or inwards against resistance.

Grading of Weakness

Grade 0   : Complete paralysis
Grade 1   : Flicker of contraction only
Grade 2   : Power detectable only when gravity is excluded by appropriate adjustment
Grade 3   : The limb can be bent against the force of gravity, but not against the examiner’s resistance
Grade 4   : There is some degree of weakness, usually described as poor, fair, or moderate strength
Grade 5   : Normal power.

Patterns of Weakness

Hemiplegia : Paralysis of one side of the body, especially of the arm and leg and usually of the jaw
Paraplegia  : Paralysis of both legs
Quadriplegia: All four limbs are paralyzed.

REFLEXES

1. Superficial reflexes
2. Deep reflexes
3. Visceral reflexes.
Grading of Deep Reflexes

0. Absent
1. Present (as a normal ankle jerk)
2. Brisk (as a normal knee jerk)
3. Very brisk

COORDINATION OF MOVEMENT

By coordination, it is meant the smooth recruitment, interaction and cooperation of separate muscles or groups of muscles in order to accomplish a definite motor act. If coordination is imperfect, ataxia is said to be present.

The coordination of groups of muscle is a function of various factors, among which are different impulses coming from the muscle spindles and joint receptors, cerebellar function and the state of tone of the muscles.

Tests for Coordination

Upper Limbs

1. The subject is asked to touch his nose first with open eyes and then with the closed eyes repeatedly (Finger-nose test).
2. The subject is asked to touch his nose first and then the examiner’s forefinger with his index finger (Finger-finger-nose test).
3. The subject is asked to flex his elbows to a right angle and then alternately supinate and pronate his forearms as rapidly as possible. In dysdiadochokinesia (a sign of cerebellar ataxia), the movements are slow, awkward and incomplete and often impossible after a few attempts. In addition, the rhythm of the movement is characteristically irregular (Fig. 24.6).
4. Coordination can also be assessed by everyday movements like dressing or undressing, handling a book or picking up pins or threading a needle.

Fig. 24.6: Test for dysdiadochokinesia
Lower Limbs

1. The subject is asked to walk along a straight line. If incoordination is present, he will soon deviate to one side or the other.

2. **Knee-heel test:** The subject is asked to lie in bed and to lift one leg high in the air and to place the heel of his leg on the opposite knee and then to slide the heel down his shin towards the ankle. The test should be performed with the eyes open. In cerebellar ataxia a characteristic, irregular side to side series of error in the speed and direction of movement occurs.

3. **Draw a circle test:** The subject is asked to draw a large circle in the air with his great toe. The circles should be drawn smoothly and accurately and the subject is asked to put a dot in the center.

4. **Romberg’s sign:** This test is for loss of position sense. It is not a test of cerebellar function. The subject is asked to stand with his feet close together and if he can do this, he is then asked to close his eyes. If Romberg’s sign is present, as soon as his eyes are closed he begins to sway about or may even fall to the same side.

With defective position sense in the legs, as in tabes dorsalis (where posterior column is affected) or sensory neuropathy, the patient is unable to maintain his posture without the aid of vision. Patient with labyrinthine or cerebellar lesions do not show instability in this test.

**GAIT**

To observe gait, the legs are adequately exposed and freed of constricting clothing and the feet bare. The subject is asked to walk away from the observer, to turn round at a given point and then to come towards him again. Certain points are to be noted:

1. Whether the subject could walk at all? If he could,
2. Whether he could walk in a straight line or deviated to one side or the other.
3. If the subject deviates to one side, the side of deviation is noted.

During examination of the gait, care is taken to see that the abnormal gait is not due to some surgical cause or to any local disease of the joint, e.g. osteoarthritis of the hip.

**Well-recognized Abnormal Types of Gait**

1. **Spastic gait or the hemiplegic gait:** This type of gait is seen in hemiplegic patient.
2. **Stamping gait occurs during sensory ataxia:** This gait is seen in tabes dorsalis.
3. **Reeling, or ‘Drunken man’s gait** occurs in cerebellar ataxia.
4. **Festinant gait** is seen in Parkinson’s disease.
5. **Waddling gait** is seen in muscular dystrophy and myopathies.
6. **High stepping gait** is seen in weakness of extensor muscles of the feet, e.g. in common peroneal nerve palsy.

**INVOLUNTARY MOVEMENTS**

The involuntary movements occur either at rest or during voluntary movement. Most are due to diseases of the basal ganglia and extrapyramidal system. The different involuntary movements are:
Examination of Motor System

1. Epilepsy
2. Myoclonus
3. Tremors:
   a. Fine tremors—seen in anxiety, hyperthyroidism
   b. Coarse tremors
      i. Intentional tremor seen in cerebellar diseases
      ii. Resting tremor seen in Parkinsonism
4. Hysterical—intentional
5. Athetosis
6. Chorea
7. Hemiballismus
8. Tics.

**SPEECH**

Speech is higher function. The subject is asked to repeat a sentence put forth by the examiner and the pattern of speech is observed. Certain common disorders of speech are:

- **Aphasia**: Absence of speech
- **Dysphasia**: Disturbance of speech
- **Dysarthria**: Difficulty in articulation of speech.

Aphasia and dysphasia are usually associated with higher center lesion. Dysarthria is usually associated with lesion in the phonation apparatus.

### OSCE-I

**Procedure:** Demonstrate the procedure of testing the nutrition/bulk of the muscle in the given subject

1. Make the subject comfortable and instruct him/her properly
2. Expose the part to be examined
3. Inspect the obvious atrophy or hypertrophy of the muscle and compare on the other side
4. Measure the girth of the upper limb, 4” above and 3” below the olecranon process of the elbow. Compare the findings on the other side
5. Measure the girth of the lower limb, 9” above and 6” below the tibial tuberosity. Compare the findings on the other side
6. Report correctly

### OSCE-II

**Procedure:** Demonstrate the procedure of testing the tone of the muscle in the given subject.

1. Make the subject comfortable and instruct him/her properly
2. Expose the part to be examined
3. Feel the tautness of the muscle by holding the muscle to be examined between the thumb and the remaining 4 fingers
4. Feel for the tone of the muscle by performing passive movements at different joints on all the limbs
5. Report correctly
OSCE-III
Procedure: Demonstrate the procedure of testing the power of the muscle in the given subject

- Make the subject comfortable and instruct him/her properly
- Expose the part to be examined
- Instruct the subject to perform flexion and extension of the limbs actively, at different joints
- Repeat the same procedure by applying moderate resistance against the movement
- Compare the findings on the other side

OSCE-IV
Procedure: Demonstrate the procedure of testing the coordination of movement in the upper limb in the given subject

- Make the subject comfortable and instruct him/her properly
- Ask the subject to extend one arm on the side and then to touch his/her nose with the index finger of the same arm, first with the eyes open and then with the eyes closed
- Put your index finger in front of the subject and ask the subject to first touch this finger with his/her index finger and then to touch his/her nose
- Ask the subject to thread a needle
- Ask the subject to perform rapid pronation and supination
- Report correctly

QUESTIONS

1. Define muscle tone in physiological terms.
2. Distinguish between spasticity and rigidity (types of hypertonia). Mention in which condition each occurs.
4. Describe two tests of coordination (other than those mentioned in your record).
5. What is hypertrophy, hyperplasia and atrophy of muscles?
6. What is rigidity? Give example.
7. How do you grade the power of muscles?
8. What is the normal grade?
9. Which is the major descending motor tract?
10. Name some extrapyramidal tracts.
11. Which is the primary motor area of the cortex?
12. What is paralysis and paresis?
13. When does winging of scapula occurs?
14. What is ‘Saturday night’ palsy?
15. What is chorea? Give examples.
17. What is hemiballismus? Give examples.
18. What is Parkinsonism?
Examination of Motor System

20. What happens when radial and ulnar nerves are paralyzed?
21. Name some abnormal gaits and mention the conditions where it occurs.
22. What is a stretch reflex?
23. What is Jendrassik’s maneuver?
24. What are the different aspects of motor functions that are assessed while examining the motor system?
25. How do you measure the bulk of the muscles in the upper and lower limbs? Name four bony landmarks.
26. How do you estimate the strength of the intrinsic muscles of hand?
27. How do you assess the strength of the trunk muscles?
28. How do you assess the strength of extensors and flexors of the knee and thigh?

MOTOR SYSTEM EXAMINATION

Name: ____________________________ Sex: ____________________________
Age: __________ Occupation: ____________________________

General Examination
Higher Functions
Motor System Examination

<table>
<thead>
<tr>
<th>I. Bulk of muscles (in cm)</th>
<th>Right</th>
<th>Left</th>
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<tbody>
<tr>
<td>Upper limb</td>
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<tr>
<td>Upper arm</td>
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<td>Forearm</td>
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<td>Lower limb</td>
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<td>Thigh</td>
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<tr>
<td>Leg</td>
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<table>
<thead>
<tr>
<th>II. Tone of the muscle</th>
<th>Right</th>
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</thead>
<tbody>
<tr>
<td>Upper limb</td>
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<tr>
<td>At small joints</td>
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<tr>
<td>At large joints</td>
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<tr>
<td>Lower limb</td>
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<td>At small joints</td>
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<tr>
<td>At large joints</td>
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</table>
### III. Strength power of the muscle

<table>
<thead>
<tr>
<th>Grade</th>
<th>Right</th>
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<tbody>
<tr>
<td>1. Abductor pollicis brevis</td>
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<td>2. Opponens pollicis</td>
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<td></td>
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<td>3. First dorsal interossei</td>
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<td></td>
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<td>4. Interossei and lumbricals</td>
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<tr>
<td>5. Flexors of the fingers</td>
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<td>6. Flexors of the wrist</td>
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<tr>
<td>7. Extensors of wrist</td>
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<tr>
<td>8. Brachioradialis</td>
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<tr>
<td>9. Biceps</td>
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<td></td>
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<tr>
<td>10. Triceps</td>
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<td></td>
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<tr>
<td>11. Supraspinatus</td>
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<td>12. Deltoid</td>
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<tr>
<td>13. Infraspinatus</td>
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<tr>
<td>14. Pectorals</td>
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<tr>
<td>15. Serratus anterior</td>
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<td></td>
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<tr>
<td>16. Latissimus dorsi</td>
<td></td>
<td></td>
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<tr>
<td>17. Abdominal muscles</td>
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<td></td>
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<tr>
<td>18. Trapezius</td>
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<tr>
<td>19. Dorsiflexion of foot</td>
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<tr>
<td>20. Plantar flexion of foot</td>
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<td></td>
</tr>
<tr>
<td>21. Extensors of knee</td>
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<td></td>
</tr>
<tr>
<td>22. Flexors of knee</td>
<td></td>
<td></td>
</tr>
<tr>
<td>23. Extensors of hip</td>
<td></td>
<td></td>
</tr>
<tr>
<td>24. Flexors of thigh</td>
<td></td>
<td></td>
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<tr>
<td>25. Abductors of thigh</td>
<td></td>
<td></td>
</tr>
<tr>
<td>26. Adductors of thigh</td>
<td></td>
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<tr>
<td>27. Rotators of thigh</td>
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</tbody>
</table>

IV. Reflexes

V. Coordination of muscles

VI. Involuntary movements

VII. Gait

Inference
AIM
To elicit the superficial, deep and visceral reflexes on the subject.

REQUISITES
1. Knee hammer (Figs 25.1A and B)
2. Torch light
3. Cotton swab

The various reflexes are elicited under the following headings:
1. Superficial reflexes
2. Deep reflexes
3. Visceral reflexes.
SUPERFICIAL REFLEXES

On stimulation of a particular part of skin or mucous membrane, contraction of certain muscles results. A lesion at any part of the reflex path causes the reflexes to disappear, e.g. anesthesia of skin, disease of sensory fibers or posterior nerve root, changes in gray matter of the cord, lesions of motor fibers or muscles. In UMN lesion, the reflexes are affected on the paralyzed side.

1. **Conjunctival reflex (Aff-V, Eff-VII cranial):** The conjunctiva of the eye is touched with a wisp of cotton. Observation: Closure of eyelids.

2. **Corneal reflex (Aff-V, Eff-VII cranial):** The sclerocorneal junction is touched with a wisp of cotton. Observation: Closure of eyelids.

3. **Pupillary reflexes (Aff-II, Eff-III cranial):**
   a. **Light reflex:** Direct and indirect
   b. **Accommodation reflex**
      i. **Direct light reflex:** The subject is asked to sit in a dimly lit room. A beam of bright light using a torch is thrown on one eye.
         Observation: Constriction of the pupil on the illuminated eye. Similarly the other eye is tested.
      ii. **Indirect light reflex (consensual light reflex):** A beam of light is thrown on one eye the other eye is observed.
         Observation: Constriction of the pupil on the opposite side.

4. **Ciliospinal reflex (cervical symp):** The skin on the side of the neck is stroked or pinched. Observation: Dilatation of the pupil.

5. **Abdominal reflex (T6–T12):** The skin of the anterior abdominal wall is gently stroked on the four quadrants with the sharp end of the knee hammer obliquely.
   Observation: Contraction of the muscles on the side stroked.

6. **Cremastric reflex (L1, L2):** The medial side of the thigh is stroked in the male subject.
   Observation: Elevation of the testicle or the scrotal sac of the same side.

7. **Anal reflex (S3, S4):** The skin around the anus is stroked.
   Observation: Contraction of the anal sphincter.

8. **Plantar reflex (L5, S1):** The outer edge of the sole of the foot is stroked gently from the heel towards the ball of the little toe and continued medially upto the ball of the great toe in one stroke with the blunt end of a knee hammer (Figs 25.2A and B).
   Observation: Plantar flexion of the great toe and the other four toes are flexed and drawn together.

**Babinski sign:** Seen in upper motor neuron lesion. In this condition, there is dorsiflexion of the great toe and fanning of other toes. It is also seen in infants below 1 year.
DEEP REFLEXES

When the tendon of a lightly stretched muscle is struck, a single sharp blow with soft rubber hammer, the muscle contracts briefly, this is tendon or stretch reflex.

1. **Jaw jerk (Cranial V):** The subject is asked to keep his mouth half opened. A finger is placed on chin of the subject and the finger struck with a knee hammer.
   Observation: Closure of the mouth.

2. **Biceps jerk (C5, C6):** The elbow of the subject is slightly flexed. The examiner’s thumb is placed over the biceps tendon and the thumb struck with a knee hammer.
   Observation: Contraction of the biceps (Fig. 25.3A).

3. **Triceps jerk (C6, C7):** The elbow of the subject is flexed and the forearm is allowed to rest across the subject’s chest. The triceps tendon just above the olecranon is tapped gently with a knee hammer (Fig. 25.3B).
   Observation: Contraction of triceps.

4. **Supinator jerk (brachioradialis jerk) (C5, C6):** The elbow of the subject is gently flexed and the forearm half pronated. The tendon overlying the radial styloid process is gently tapped with a knee hammer (Fig. 25.3C).
   Observation: Contraction of the brachioradialis muscle.

5. **Knee jerk (L2, L3):** The subject is allowed to be seated on a stool with one leg crossed over the other and hanging loosely. Examiner’s hand is placed over the quadriceps muscle and the patellar tendon is gently tapped with a knee hammer.
   Observation: A forward kick of the leg would be observed due to contractions of the quadriceps muscle.

   When the subject is lying supine, the knee is supported with the left hand and the patellar tendon is tapped.
Figs 25.3A to C: Deep reflexes: (A) Biceps jerk; (B) Triceps jerk; (C) Supinator jerk

Patellar clonus: With the leg fully extended at the knee joint, the patella is ripped between the thumb and the other fingers and pushed upwards. Observation: In a normal person there may be a few contractions which immediately relaxes. In upper motor neuron lesion sustained contractions may be seen.

Ankle clonus: The subject’s knee is bent slightly and supported with left hand. The forefoot is grasped with the hand and suddenly dorsiflexed (Fig. 25.4).
Observation: Intermittent jerking movement can be observed which would be persistent in upper motor neuron lesion.

6. Ankle jerk (S1, S2): The subject is asked to sit on a stool with the leg crossed and with the foot slightly doriflexed by the examiner so as to apply slight tension on tendo achillis. The tendo achillis is tapped with a knee hammer (Figs 25.5A and B).
Observation: Contraction of calf muscles with plantar flexion.

Figs 25.5A and B: Ankle jerk: (A) In kneeling position; (B) In supine position
Grading of Deep Reflexes

0 - Absent
1 - Present (as a normal ankle jerk)
2 - Brisk (as a normal knee jerk)
3 - Very brisk
4 - Clonus.

VISERAL REFLEXES

1. **Defecation reflex:** The subject is questioned about his bowel habits to rule out incontinence.
2. **Micturition reflex:** The subject is questioned about his micturition habits to rule out bladder incontinence.
3. **Swallowing reflex (Deglutition reflex):** The subject is questioned about.
   - i. Any difficulty in swallowing
   - ii. Nasal regurgitation of food during swallowing.

**Reinforcement Technique (Jendrassik’s Maneuver)**

In healthy subjects, the knee jerk is hardly seen or entirely absent. But in case of other tendon reflexes, sometimes it may be necessary to apply reinforcement—Jendrassik maneuver (Fig. 25.6). It is done by asking the subject to make a strong voluntary muscular effort such as...
hooking the fingers of the two hands together and pulling them apart. Reinforcement acts by increasing the excitability of anterior horn cells and by increasing the sensibility of the muscle spindle to stretch.

**Abnormal Tendon Reflex**

**Hyporeflexia:** Diminished or absent tendon reflexes. Seen in lower motor neuron lesion, e.g. tabes dorsalis, poliomyelitis.

**Hyperreflexia:** Exaggerated tendon reflexes. Occurs in upper motor neuron lesion, anxious individuals, thyrotoxicosis, tetanus. In cerebellar disease, the reflexes have a pendular quality.

**OSCE-I**

Procedure: Demonstrate the abdominal reflex in the given subject

- Make the subject lie down comfortably and instruct him/her properly
- Expose the abdomen of the subject
- Stroke the abdomen lightly with a blunt object along 3 lines, from lateral towards medial side
- Look for the contraction of the underlying muscles
- Compare on the other side
- Report correctly

**OSCE-II**

Procedure: Demonstrate the plantar reflex in the given subject

- Make the subject lie down comfortably and instruct him/her properly
- Expose the sole of the subject completely
- Stroke the sole of the subject from the heel towards the ball of the little toe and continue medially up to the base of the great toe
- Look for the movements of the toes
- Compare on the other side
- Report correctly

**OSCE-III**

Procedure: Demonstrate the biceps reflex in the given subject

- Make the subject sit comfortably and instruct him/her properly
- Expose the upper arm of the subject
- Support the arm of the subject on your forearm
- Place your thumb over the bicep’s tendon and strike the thumb with the pointed end of the knee hammer
- Look for the contraction of the Biceps muscle/or any movement at the elbow
- Compare on the other side
- Report correctly
Reflexes

OSCE-IV
Procedure: Demonstrate the knee jerk/reflex in the given subject
- Make the subject sit comfortably with the legs hanging and instruct him/her properly
- Expose the lower limb of the subject
- Feel for the patellar tendon and strike it with the pointed end of the hammer
- Look for the contraction of the quadriceps muscle/or any movement at the knee joint
- Compare on the other side
- Report correctly

OSPE-V
Procedure: Demonstrate the ankle jerk/reflex in the given subject
- Make the subject lie down comfortably and instruct him/her properly
- Expose the leg of the subject
- Cross one leg over the other and position the knee semiflexed and the hip externally rotated
- Slightly dorsiflex the foot with the other hand and strike the tendon with the broad end of the knee hammer
- Look for the contraction of the calf muscle/or any movement at the ankle joint
- Compare on the other side
- Report correctly

QUESTIONS

1. Define a reflex.
2. Draw and label a reflex arc.
3. How will you grade a reflex?
4. List the differences between the upper motor neuron lesions and lower motor neuron lesions.
5. What is clonus?
6. What is Babinski sign?
7. Give the different classification of reflexes.
8. What is Jendrassik maneuver? Give its importance.
9. What is the root value of knee jerk, biceps jerk and ankle jerk?
10. What is crossed extensor reflex?
11. What is the root value of abdominal and plantar reflex?
12. How is the reflexes examination going to be useful to the physician?
EXAMINATION OF REFLEXES

Name:      Age:       Sex:      Occupation:

General Examination:
Higher functions:

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<thead>
<tr>
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<td>Corneal reflex</td>
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<td>Pupillary reflex</td>
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<tr>
<td>Accomodation reflex</td>
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<td>Upper abdominal reflex</td>
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<tr>
<td>Lower abdominal reflex</td>
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<td>Plantar reflex</td>
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<tr>
<td>2. Biceps jerk</td>
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<tr>
<td>3. Triceps jerk</td>
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<tr>
<td>4. Brachioradialis/supinator jerk</td>
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<tr>
<td>5. Knee jerk</td>
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<tr>
<td>6. Ankle jerk</td>
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VISCERAL REFLEXES

1. Defecation reflex
2. Micturition reflex
3. Swallowing reflex

Inference
AIM

To examine the cranial nerves I, II, III, IV, V, and VI.

Olfactory Nerve

Anatomy: Olfactory nerves gives the sense of smell. Receptors are found in the mucous membrane of the roof of the nasal cavity. From here, the fibers pass via the cribriform plate of the ethmoid bone to the glomeruli of the olfactory bulb, and later, it reaches the cerebral cortex to end in the uncus of the parahippocampal gyrus.

Tests for smell: The sense of smell is tested in each nostril separately with eyes closed. Substances like oil of peppermint, oil of clove, tincture of asafoetida are used (Fig. 26.1). Irritating substances like ammonia are avoided since it would partially stimulate the trigeminal nerve.

Abnormalities

Anosmia  –  Loss of sensation of smell as seen in cases of meningitis, fracture of anterior cranial fossa, etc.
Parosmia  –  Perverted smell as seen in head injuries.
Hyposmia  –  Decrease in a perception of smell.
OPTIC NERVE

Anatomy: The receptors for visions are the rods and cones of retina. From here the fibers pass backward as the optic nerve. The two optic nerve then form the optic chiasma. Fibers of the optic nerve continue as optic radiations and end in the striate area of the occipital cortex.

TESTS

1. Acuity of vision (Figs 26.2A to C)
2. Field of vision
3. Color vision
4. Pupillary response.

Figs 26.2A to C: (A) Snellen’s chart; (B) Jaeger’s chart; (C) Principle behind Snellen’s chart
Acuity of Vision

1. **Test for distant vision:** Using Snellen's test types. Each eye is tested separately. A chart containing letters of different sizes is kept at a distance of six meters from the subject. Under each line on the chart is a number which indicates the distances at which the line should be read. Visual acuity is determined by the formula \( V = \frac{d}{D} \) where 'd' is the distance at which the letters are read and 'D' is the distance at which they should be read (Fig. 26.2A).

   **When ‘V’ is less than 1/60, other tests are done like:**
   
   i. Finger counting test
   
   ii. Hand movement test
   
   iii. Perception or no perception of light.

2. **Test for near vision:** This is assessed by using Jaeger's chart which has letters of different sizes. The smallest print is 0.5 mm. The near vision is recorded as the smallest type which the subject can read comfortably (Fig. 26.2B).

Field of Vision

This represents that portion of space in which other objects are visible while fixing the gaze on a particular object.

1. **Confrontation method:** The test is done first for each eye individually. The subject and the examiner is seated at an arm's distance (If the subject’s left eye is to be tested, the subject should close his right eye and the examiner his left eye, so that the subject’s left eye and the examiner’s right eye confront each other). The examiner holds the index finger of his outstretched left hand to a plane midway between him and the patient. The finger is brought nearer to the midline until the examiner first perceives the moving finger. The subject is asked if he is also able to see the finger. If he fails to do so, the finger is brought nearer until he sees it. The field is tested in each eye separately in every direction—upwards, downwards, to right and to left—using the extent of the examiner’s own field for comparison (Fig. 26.3).

2. **Instrumental method:** Using perimeter.

Abnormalities in the Field of Vision (Fig. 26.4)

1. **Central scotoma:** Loss of vision confined to the center of the field.

2. **Hemianopia:** Loss of vision in one half of visual field.
   
   a. **Heteronymous hemianopia:** Same half of both the fields are affected.
      
      i. **Binasal hemianopia:** Loss of vision in the nasal field in both eyes.
      
      ii. **Bitemporal hemianopia:** Loss of vision in the temporal field in both eyes.
   
   b. **Homonymous hemianopia:**
      
      i. **Right homonymous hemianopia:** Right half of field of each eye is affected.
      
      ii. **Left homonymous hemianopia:** Left half of field of each eye is affected.

3. **Quadrantanopia:** Loss of vision in a quadrant of the visual field.
Color Vision

1. Holmgren’s wool test
2. Ishihara’s pseudoisochromatic charts (Fig. 26.5).

Fig. 26.4: Visual pathway and its lesions

Fig. 26.5: Ishihara’s pseudoisochromatic chart
Pupillary Response
It is dealt with under next section.

OCULOMOTOR, TROCHLEAR, AND ABDUCENT (III, IV AND VI) NERVES

Oculomotor Nerve Supplies
1. Extraocular muscles other than supplied by IV and VI cranial nerves.
2. Sphincter pupillae
3. Muscles of accommodation
4. Levator palpebrae superioris.
Edinger-Westphal nucleus supplies parasympathetic fiber to ciliary muscles and iris.
Trochlear nerve supplies superior oblique (SO)
Abducent nerve supplies lateral rectus (LR).

EXAMINATION OF EXTRAOCULAR MOVEMENTS

The superior and inferior recti act as elevators and depressors alone when the eye is in abduction. The obliques act similarly when the eye is in adduction (Superior oblique acts as depressors of the eye and inferior oblique acts as elevators) (Figs 26.6 and 26.7).

Fig. 26.6: Actions of extraocular muscles

Figs 26.7A and B: Examination of extraocular movements
Supranuclear Lesion

1. Loss of conjugate movements of eyes
2. Skew deviation of eyes
3. Nystagmus

Infranuclear lesion: There is paralysis of individual muscles of III N: There is ptosis or drooping of eyelid—the eyeball is displaced downward and outward. The pupil is dilated and there is loss of accommodation.

Paralysis of IV N: Impaired downward movement. Eyeball rotated outwards due to unopposed action of inferior rectus when attempting to look downwards and diplopia below the horizontal plane.

Paralysis of VI N: Inability to move the eye outwards and diplopia on looking in that direction. Sometimes convergent squint is due to unopposed action of medial rectus.

EXAMINATION OF THE PUPIL

1. Size—compare both sides.
2. Shape—may be irregular due to adhesion of iris to the lens following iritis.
3. Mobility.

Reaction to Light

1. Direct light reflex: There would be constriction of the pupil when a bright light is shown in that eye.
2. Indirect or consensual light reflex: When light is shown in one eye there would be constriction of the pupil of the opposite eye even when a hand or a book is placed between the eyes (Figs 26.8A and B).

Accommodation Reflex

When a person seeing a distant object is asked to focus on a near object, accommodation
Examination of Cranial Nerves I to VI

occurs. Together with accommodation, there is miosis and convergence of the eyes.

**Argyll Robertson pupil:** Seen in neurosyphilis. In this condition, the lesion is in the pretectal region of mesencephalon. Accommodation reflex is present whereas the light reflex is absent (Figs 26.9A and B).

![Figs 26.9A and B: Accommodation reflex](A) Resting condition; (B) Accommodation

**TRIGEMINAL NERVE**

**Sensory pathway:** From trigeminal (Gasserian) ganglion the fibers enter the lateral surface of pons about its middle. Fibers for touch and postural sensibility terminate large nucleus on pons (lateral to motor nucleus).

Fibers for pain and thermal sensation enter the descending or bulbospinal tract, which extends up to C2 level and ascends in medial lemniscus.

The fibers arise from three divisions (Fig. 26.10):

1. Ophthalmic
2. Maxillary
3. Mandibular.

The mandibular division supplies parasympathetic fibers to the salivary glands.

![Fig. 26.10: Sensory divisions of trigeminal nerve]
**Motor pathway:** It originates from the small nucleus medial to sensory nucleus. It emerges from the side of pons and passes inferior to trigeminal ganglion and joins the mandibular division to supply muscles of mastication, namely masseter, temporalis, lateral pterygoid and medial pterygoid.

**Tests for Sensory Function**

1. To test for cutaneous sensation.
2. To test for corneal reflex and conjunctival reflex (Fig. 26.11).
3. To test for palatal reflex.

**Tests for Motor Functions**

1. The subject is asked to clench the teeth. The prominence of the masseter and temporalis is to be felt (Fig. 26.12).
2. The subject is asked to open the mouth—in paralysis of the motor division of trigeminal nerve the jaw is deviated to the paralyzed side due to action of pterygoid muscle.

**Lesion of Trigeminal Nerve**

1. Loss of sensation of skin and mucous membrane of jaw and nasopharynx.
2. Salivary, buccal, and lacrimal secretion decreases.
3. Trophic ulcers in mouth, nose, and cornea.
4. Weakness of muscles of mastication.

---

### OSCE-I

**Procedure:** Demonstrate the procedure of testing the first cranial nerve in the given subject

1. Make the subject sit comfortably and instruct him/her properly.
2. Ask the subject to close both the eyes and one nostril.
3. Bring the test tube containing the odorous substance close to the open nostril and ask him/her to sniff deeply.
4. Ask the subject to identify the smell.
5. Repeat the procedure with the other nostril.

### OSCE-II

**Procedure:** Demonstrate the procedure of testing the visual acuity for distant vision in the given subject

1. Make the subject stand comfortably at a distance of 6 meters from a well lit Snellen's chart and instruct him/her properly.
2. Ask the subject to close one eye with the cup of the palm.
3. Ask the subject to read the chart from top to bottom and note the lowest line which he/she can read correctly.
4. Repeat the procedure with the other eye.
5. Report correctly.
### OSCE-III
**Procedure:** Demonstrate the procedure of testing the visual acuity for near vision in the given subject

1. Make the subject sit comfortably and instruct him/her properly.
2. Ask the subject to close one eye with the cup of the palm.
3. Ask the subject to read the Jaeger’s chart from top to bottom, by holding it in other hand at the normal reading distance (25 cm).
4. Note the smallest font size which he/she can read correctly.
5. Repeat the procedure with the other eye.

### OSCE-IV
**Procedure:** Demonstrate the procedure of testing the color vision in the given subject

1. Make the subject sit comfortably and instruct him/her properly.
2. Ask the subject to close one eye with the cup of the palm.
3. Ask the subject to read the numbers and trace the paths depicted on the color plates of the Ishihara chart.
4. Note whether he/she can identify the numbers and paths correctly.
5. Repeat the procedure with the other eye.

### OSCE-V
**Procedure:** Demonstrate the procedure of testing the field of vision by confrontation method in the given subject

1. Sit face to face in front of the subject at a distance of ½ to 1 meters and instruct him/her properly.
2. Ensure that the eyes of both, the subject and the examiner are at the same level.
3. Ask the subject to close one eye with the cup of the palm of the same side. The examiner will close the opposing eye similarly.
4. Ask the subject to look at the tip of the examiner’s nose.
5. Stretch your arm on the temporal side of the subject’s test eye, in a plane mid way between the subject and you.
6. Point out the index finger and bring it slowly towards the midline and ask the subject to report when he/she first sees the finger.
7. Also check the field of vision for the superior, inferior and nasal quadrants.
8. Repeat the procedure with the other eye.

### OSCE-VI
**Procedure:** Demonstrate the procedure of testing the direct light reflex in the given subject

1. Make the subject sit comfortably and instruct him/her properly.
2. Ask the subject to close one eye with the cup of the palm and to look at some distant point.
3. Shine a bright light into the open eye by bringing it from the side.
4. Look for the constriction of the pupil.
5. Repeat the procedure with the other eye.
### OSCE-VII
**Procedure: Demonstrate the procedure of testing the indirect light reflex in the given subject**
- Make the subject sit comfortably and instruct him/her properly.
- Ask the subject to put a cardboard over the bridge of his/her nose and to look at some distant point.
- Shine a bright light into one of the eyes by bringing it from the side.
- Look for the constriction of the pupil in the opposite eye.
- Repeat the procedure with the other eye.
- Report correctly.

### OSCE-VIII
**Procedure: Demonstrate the procedure of testing the accommodation reflex in the given subject**
- Make the subject sit comfortably and instruct him/her properly.
- Ask the subject to look at some distant point.
- Bring your index finger in front of the subject, at the level of his/her eyes, at a distance of about 15-20 cm.
- Ask the subject to shift the gaze from the distant point to the tip of your finger.
- Look for the change in the papillary size and convergence of the visual axis.
- Report correctly.

### OSCE-IX
**Procedure: Demonstrate the procedure of testing the III/IV/VI cranial nerve in the given subject**
- Sit face to face in front of the subject and instruct him/her properly.
- Place your index finger in front of the subject and move it to the six cardinal positions of the gaze.
- Ask the subject to follow the finger with the eyes without moving his/her head.
- Look for the simultaneous and symmetrical conjugate movements of both the eyes.
- Ask the subject for any diplopia.
- Report correctly.

### OSCE-X
**Procedure: Demonstrate the procedure of testing the motor division of the V cranial nerve in the given subject**
- Make the subject sit comfortably and instruct him/her properly.
- Ask the subject to clench the teeth and you feel for the tautness of the masseter and temporalis muscle on both the sides.
- Ask the subject to open and close the mouth and to perform side-by-side movement of the jaw.
- Ask the subject to repeat the same movements against the slight resistance.
- Report correctly.
Examination of Cranial Nerves I to VI

OSCE-XI

Procedure: Demonstrate the procedure of testing the corneal/conjunctival reflex in the given subject

<table>
<thead>
<tr>
<th>Step</th>
</tr>
</thead>
<tbody>
<tr>
<td>Make the subject sit comfortably and instruct him/her properly</td>
</tr>
<tr>
<td>Ask the subject to look up at the ceiling and gently depress the lower eyelid</td>
</tr>
<tr>
<td>Very lightly touch the lateral edge of the cornea/lower palpebral conjunctiva with a wisp of damp cotton wool</td>
</tr>
<tr>
<td>Look for both direct and consensual blinking</td>
</tr>
<tr>
<td>Repeat the procedure on the opposite side</td>
</tr>
<tr>
<td>Report correctly</td>
</tr>
</tbody>
</table>

QUESTIONS

1. Define anosmia, hyperosmia, and parosmia. Mention the conditions in which these occur.
2. List the effects of various lesions in the visual pathway.
3. What is color blindness? What are the types of color blindness? How is it transmitted in man?
4. How are supranuclear (UMN) and infranuclear (LMN) lesions of IIIrd, IVth, and VI cranial nerves differentiated?
5. What are the effects of lesions of the Vth cranial nerve?
6. Describe Argyll Robertson pupil.
7. What is Horner’s syndrome?
8. What are the receptors involved in color vision?
9. Why NH₃ is not used for smell test?
10. What is the instrument used for smell test?
11. Define visual acuity and visual field. How is it tested?
12. What are the tests for color vision?
13. What is blind spot?
14. What is the cause for indirect light reflex?
15. What is squint/strabismus?
16. What is cataract?
17. What are the manifestations of vitamin ‘A’ deficiency?
18. What are the muscles of mastication?
19. Which cranial nerve is commonly affected by increase in intracranial pressure?
20. What is glaucoma, types and how is it tested?
21. What is the afferent and efferent nerve for corneal reflex?
22. What are the divisions of Vth cranial nerve?
23. How will you test the motor function of the Vth cranial nerve?
# EXAMINATION OF CRANIAL NERVES I TO VI

<table>
<thead>
<tr>
<th>Name:</th>
<th>Age:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex:</td>
<td>Occupation:</td>
</tr>
</tbody>
</table>

## General Examination

- Higher functions

### CRANIAL NERVE I

No history of cold or nasal block

<table>
<thead>
<tr>
<th></th>
<th>Right nostril</th>
<th>Left nostril</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peppermint oil</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clove oil</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tincture of asafoetida</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nicotine in solution</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### CRANIAL NERVE II

#### Visual acuity

<table>
<thead>
<tr>
<th></th>
<th>Distant vision (Snellen’s chart)</th>
<th>Near vision (Jaeger’s chart)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Right eye</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left eye</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

#### Field of vision

<table>
<thead>
<tr>
<th></th>
<th>Superior</th>
<th>Inferior</th>
<th>Medial</th>
<th>Lateral</th>
</tr>
</thead>
<tbody>
<tr>
<td>Right eye</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left eye</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Color Vision*  

- Right eye
  - Left eye

Pupillary responses (See below in cranial nerve III, IV and VI)

### CRANIAL NERVES III, IV AND VI

#### Pupils

<table>
<thead>
<tr>
<th></th>
<th>Right eye</th>
<th>Left eye</th>
</tr>
</thead>
<tbody>
<tr>
<td>Size and shape</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Direct light reflex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Indirect light reflex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Accommodation reflex</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Examination of Cranial Nerves I to VI

#### Ocular movements

<table>
<thead>
<tr>
<th>Action</th>
<th>Right eye</th>
<th>Left eye</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adduction</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Abduction</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Elevation while adducted</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Depression while adducted</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Elevation while abducted</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Depression while abducted</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

#### CRANIAL NERVE V

No history of loss of sensation in mouth or ulcers in mouth nose and cornea

<table>
<thead>
<tr>
<th>General sensation</th>
<th>Ophthalmic division</th>
<th>Maxillary division</th>
<th>Mandibular division</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Right</td>
<td>Left</td>
<td>Right</td>
</tr>
<tr>
<td>Touch</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pain</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vibration</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Temperature</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tactile localization</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tactile discrimination</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Reflexes</th>
<th>Right eye</th>
<th>Left eye</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corneal reflex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Conjunctival reflex</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

#### Motor function

<table>
<thead>
<tr>
<th></th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Open the mouth</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Closure of the mouth</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clench the teeth</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Jaw jerk</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Inference
AIM

To examine the cranial nerves VII, VIII, IX, X, XI, and XII.

FACIAL NERVE

Anatomy

From facial nucleus (situated lateral to abducent) fibers wind round the abducent nucleus and emerge medial to vestibulocochlear nerve. In close contact with the VIII nerve, it enters the auditory meatus. During its course in the temporal bone, it gives off a branch to stapedius muscle. It is joined by chorda tympani, which contains taste fibers of the anterior 2/3rd of tongue at the geniculate ganglion. It emerges at a point opposite the junction of the anterior border of the mastoid process and spreads to supply the facial muscles. Facial nerve is a motor nerve. It supplies all the muscles of the face and scalp except levator palpebrae superioris. It also supplies platysma.

Pathway for Taste

From anterior 2/3rd of the tongue, the fibers pass from lingual nerve to chorda tympani and through the geniculate ganglion of the facial nerve into medulla oblongata (sometimes passes in maxillary division of V nerve).

From the nucleus of tractus solitarius, the fibers pass to the thalamus and then to the temporal part of postcentral gyrus and to amygdala.
**Examination of Cranial Nerves VII to XII**

**Tests for Taste**

The subject is asked to protrude his tongue. The test substance used are sugar and salt. The substance is placed on the tongue and the subject is asked to indicate the perception of the taste without withdrawing the tongue. After each test, the mouth must be rinsed. Quinine and citric acid are used to test bitter and sour tastes. Quinine test must be applied last.

The subject is asked for abnormal tastes or hallucinations of taste.

**Abnormalities**

Ageusia—loss of taste sensation.

**Tests for Motor Effects of Facial Nerve (Figs 27.1A to C)**

1. The subject is asked to raise the eyebrows. The furrows on the forehead are noted.
2. The subject is asked to shut his eyes as tightly as he can. An attempt to open the closed eyes against resistance is made.
3. The subject is asked to whistle. This is impossible in facial palsy.
4. The subject is asked to smile or show his upper teeth. In paralysis, the angle of the mouth is drawn to the healthy side.
5. He is asked to blow his cheeks and an attempt is made to deflate it by tapping on it with a finger. Air escapes more easily on the paralyzed side.

![Figs 27.1A to C](image)

Figs 27.1A to C: Examination of facial nerve: (A) Closure of the eyes; (B) Raising the eyebrows; (C) Showing the upper teeth

**Features of Facial Nerve Palsy (Motor Effects)**

**Affected side shows:**

1. Loss of expression.
2. Nasolabial fold would be less pronounced.
3. Furrows of the forehead would be smoothened.
4. Eye would be more widely open than on the normal side.
5. Mouth is drawn towards the normal side when the patient smiles.
6. Unable to whistle.
7. Food collects between the teeth and cheek.
8. Saliva escapes from the affected angle of the mouth.
Supranuclear Facial Paralysis

Lower part of the face is chiefly affected since the upper part is bilaterally innervated. Taste is not affected.

Infranuclear Facial Paralysis

Both upper and lower part equally involved. Since it is a lower motor neuron lesion, the facial muscles show atrophy at a later date (Bell’s palsy). Chorda tympani branch affected if the lesion is in the facial canal—loss of taste sensation in the anterior 2/3 of the tongue. If the stapedius is paralyzed the sound on the affected side may be unusually loud (hypercusis) (Figs 27.2A and B).

VESTIBULOCOCHLEAR NERVE

It contains two sets of fibers:
1. Auditory
2. Vestibular.

Auditory Pathway

From cochlear ganglion, the fibers enter the brainstem and distribute in the dorsal and ventral cochlear nuclei. The secondary auditory tracts after partial decussation terminate in the inferior colliculi and medial geniculate bodies. The fibers then pass via the internal capsule to the center for hearing in the auditory cortex in the temporal lobe.

Vestibular Pathway

From the semicircular canals and labyrinth (sense of equilibrium and balance) and through the vestibular ganglion the fibers terminate in the vestibular nuclei of pons and medulla. This has got close connections with the cerebellum and temporal lobe of cerebral cortex.

Tests for Hearing

1. The subject’s capacity for hearing conversational and whispered voice is to be found.
2. **Tuning fork tests:** A tuning fork with a frequency of 256 or 128 Hz is used.
   a. **Rinne’s test:** The prongs of the tuning fork are struck and the base is kept on the mastoid process. Once the sound is no more heard the tuning fork is held near the pinna of the ear to perceive the sound once again. In a normal person air conduction is better than bone conduction (Rinne’s positive). In sensorineural deafness (Lesion of cochlear branch of VIII nerve) Rinne’s test is positive as long as the deafness is partial. In conductive deafness Rinne’s test is negative (bone conduction is better than air conduction) (Figs 27.3A and B).

   ![Rinne's test](image)

   **Figs 27.3A and B:** Rinne’s test: (A) Testing the bone conduction followed by (B) Air conduction

   b. **Weber’s test:** The vibrating tuning fork is kept in the vertex of the forehead. In a normal person the sound would be heard well on both sides. In conductive deafness (middle ear deafness) sound is perceived better in the affected ear. In sensorineural deafness the sound is louder in the better hearing ear (Fig. 27.4).

   ![Weber's test](image)

   **Fig. 27.4:** Weber’s test

   c. **Schwabach’s test:** In this test, the bone conduction is compared to that of the examiner. The prongs of the tuning fork are struck and the base is kept on the mastoid process. Once the subject is unable to hear, the base is brought to the examiner’s mastoid process to assess if the sound is still heard or not.

   d. **Audiometry tests:** It is used to assess the degree of hearing loss and the likely site of pathology in the auditory pathway.
      a. Pure tone audiometry.
      b. Speech audiometry.
Abnormal Auditory Sensations

1. Tininitus.
2. Hyperacusis—even slight sounds are heard with painful intensity (due to paralysis of stapedius muscle).

Vestibular Nerve Tests

Caloric test: Water is added 7°C above and below the normal body temperature (37°C) with the head positioned at 30° and nystagmus is looked for.

GLOSSOPHARYNGEAL NERVE

Anatomy

Receives sensory supply from the posterior 1/3rd of the tongue and the mucous membranes of the pharynx. It also receives taste sensation from posterior 1/3rd of the tongue.

It also innervates the middle pharyngeal sphincter and stylopharyngeus muscle.

Tests

a. The taste sensation in posterior 1/3rd of the tongue is examined.
b. The back of the pharynx is tickled (pharyngeal reflex). Reflex contraction of pharyngeal muscles results.

VAGUS NERVE

Anatomy

It gives motor innervation for soft palate (except tensor palati), pharynx and larynx. Sensory and motor supply for respiratory passage and heart. Parasympathetic supply to the abdominal viscera.

Tests

1. The subject is asked whether there is any regurgitation of fluids through the nose while swallowing. This occurs in total paralysis of soft palate.
2. The subject is asked to say high pitched word like ‘egg’ and ‘rub’ which requires complete closure of nasopharynx. In paralysis of vagus ‘egg’ is pronounced as ‘eng’ and ‘rub’ as ‘mm.’
3. The subject is asked to say ‘Ah.’ In bilateral paralysis, the soft palate remains motionless. In unilateral paralysis the affected side remains motionless.
4. Visceral reflex—oculocardiac and carotid sinus reflex (not done in class).
ACCESSORY NERVE

Anatomy
It is purely motor nerve. It has got a cranial and a spinal origin. It innervates some muscles of pharynx and larynx as well as sternocleidomastoid and trapezius.

1. **Test for trapezius muscle:** The subject is asked to shrug his shoulder against resistance (Fig. 27.5).
2. **Test for sternocleidomastoid muscle:** The subject is asked to rotate his chin against resistance.

HYPOGLOSSAL NERVE

Anatomy
It is a motor nerve. It supplies the tongue and the depressors of the hyoid.

Test
1. The subject is asked to protrude the tongue. In unilateral lesion, the tongue would deviate to the paralyzed side.
2. The subject is asked to move the tongue from side-to-side and to lick each lip with it. Observe whether this can be done freely. Strength may also be assessed by pressing against the tongue with a finger as the subject protrudes it into each cheek.
3. Fasciculation and wasting of the muscle is also looked for. Fasciculation is assessed with the tongue relaxed in the mouth not when protruded. Wasting—indicates lower motor neuron paralysis.

OSCE-I

**Procedure:** Demonstrate the procedure of testing the taste sensation in anterior 2/3rd of the tongue in the given subject

- Make the subject sit comfortably and instruct him/her properly
- Ask the subject to close the eyes and protrude the tongue
- Take the testing material in a dropper and place it over the dorsum of the tongue on either side
- Ask the subject to identify the taste without taking the tongue inside the mouth
- Repeat the procedure with all 4 tasting material after rinsing the mouth each time with the plain water
- Repeat the procedure on the opposite side
- Report correctly

Fig. 27.5: Test for trapezius muscle
### OSCE-II

**Procedure:** Demonstrate the procedure for Rinne’s test in the given subject

- Make the subject sit comfortably and instruct him/her properly.
- Vibrate a tuning fork and keep the base of the tuning fork over the mastoid process behind the ear to be tested.
- Ask the subject to raise a finger when he/she stops hearing the sound.
- Immediately hold the prongs of the tuning fork parallel to the external ear and ask the subject whether he/she can still hear the sound.
- Repeat the procedure on the other side.
- Report correctly.

### OSCE-III

**Procedure:** Demonstrate the procedure for Webber’s test in the given subject

- Make the subject sit comfortably and instruct him/her properly.
- Vibrate a tuning fork and keep the base of the tuning fork in the midline high on the forehead/vertex.
- Ask the subject which side the sound is loudest.
- Report correctly.

### OSCE-IV

**Procedure:** Demonstrate the procedure for examining the X cranial nerve in the given subject

- Make the subject sit comfortably and instruct him/her properly.
- Ask the subject about any present history of nasal regurgitation of the food, nasal intonation of the voice or hoarseness of the voice.
- Ask the subject to widely open the mouth and say “aaah” and look for the movement the palate and uvula, using a torch.
- Perform the ‘Gag Reflex’ by tickling the posterior pharyngeal wall with cotton tipped thin stick and look for the reflex contraction of the posterior pharyngeal wall or any vomiting sensation reported by the subject.
- Report correctly.

### OSCE-V

**Procedure:** Demonstrate the procedure for examining the XI cranial nerve muscle in the given subject

- Make the subject sit comfortably and instruct him/her properly.
- For testing the sternocleidomastoid ask the subject to turn the head both the sides actively.
- Repeat the procedure against moderate resistance.
- For testing the trapezius, ask the subject to shrug the shoulders.
- Repeat the procedure against moderate resistance by standing behind the subject.
- Report correctly.
Examination of Cranial Nerves VII to XII

OSCE-VI

Procedure: Demonstrate the procedure for examining the XI cranial nerve muscle in the given subject

- Make the subject sit comfortably and instruct him/her properly
- For testing the sternocleidomastoid, ask the subject to turn the head both the sides actively
- Repeat the procedure against moderate resistance
- For testing the trapezius, ask the subject to shrug the shoulders
- Repeat the procedure against moderate resistance by standing behind the subject
- Report correctly

OSCE-VII

Procedure: Demonstrate the procedure for examining the XII cranial nerve in the given subject

- Make the subject sit comfortably and instruct him/her properly
- Ask the subject to open the mouth and look for any fasciculation or atrophy/hypertrophy, while the tongue resting on the floor
- Ask the subject to protrude the tongue and look for any deviation from the midline
- Ask the subject to push the cheeks from inside, on the either side, by pushing it with tongue
- Repeat the above procedure by applying mild resistance with the thumb from outside the cheek
- Report correctly

QUESTIONS

1. Differentiate between, lower motor and upper motor neuron lesions of the facial nerve.
2. What are the muscles of facial expressions?
3. What is the receptor for taste sensation?
4. Trace the pathway for taste sensation from the tongue.
5. What is aguesia and paraguesia? Give examples.
6. What are the bedside tests for hearing?
7. What is hyperacusis? When does a person get hyperacusis?
8. What is the principle behind tuning fork test?
9. What is the receptor for hearing and center for hearing?
10. Mention two types of deafness and list the tests to differentiate these types.
11. What is the afferent nerve and efferent nerve for palatal reflex?
    
    - What is gag reflex?
12. What is nystagmus? What are its components? Name some conditions, which cause nystagmus.
13. Name the visceral reflex for the X nerve.
14. What are the muscles supplied by the XI nerve?
15. Name some tests for the vestibular division of VIII nerve.
16. Name the intrinsic muscles of the tongue.
17. Which side is the tongue deviated if the XII nerve is paralyzed on the left side’?
18. What is Bell’s palsy?
19. How will you treat Bell’s palsy?
20. What are the tests for motor division of facial nerve?
21. Why does the upper part of the face not affected in supranuclear lesion of the VIIth cranial nerve?
22. What is vertigo?
23. What is fasciculation? How will you test it?
24. What are the effects of paralysis of IX cranial nerve?
25. What are the two types of nerves which supply its areas?
26. What are primary taste sensation and where are they found?
27. What is ptosis? In which type of facial nerve palsy it is found?
28. What is common cause of loss of hearing?
29. What is dysarthria?

EXAMINATION OF CRANIAL NERVES VII TO XII

Name:       Age:       Sex:       Occupation:
General Examination
Higher function

CRANIAL NERVE VII

<table>
<thead>
<tr>
<th>Sensory examination (Taste sensation in anterior 2/3rd of tongue)</th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sweet (sugar)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salty (salt)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sour (vinegar)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bitter (quinine)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Motor examination</th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raise the eyebrows</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Shut the eyes tightly</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whistle</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Smile (nasolabial fold)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Show upper teeth (angle of mouth)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Cranial Nerve VIII

<table>
<thead>
<tr>
<th>Test</th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bedside test</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whispering</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Watch test</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rubbing of fingers</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tuning fork tests</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rinne’s test</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weber’s test</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Schwabach’s tests</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Cranial Nerve IX

<table>
<thead>
<tr>
<th>Test</th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Taste sensation in posterior 1/3rd of tongue</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sweet</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salt</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bitter</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sour</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pharyngeal reflex</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Cranial Nerve X

<table>
<thead>
<tr>
<th>Test</th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Regurgitation of fluids</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Say high pitched sound</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Say “ah”</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pharyngeal reflex</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
## CRANIAL NERVE XI

<table>
<thead>
<tr>
<th></th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shrug shoulder</td>
<td>against resistance</td>
<td></td>
</tr>
<tr>
<td>Rotate chin</td>
<td>against resistance</td>
<td></td>
</tr>
</tbody>
</table>

## CRANIAL NERVE XII

<table>
<thead>
<tr>
<th></th>
<th>Right</th>
<th>Left</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protrude the</td>
<td>tongue</td>
<td></td>
</tr>
<tr>
<td>Move tongue</td>
<td>from side-to-side</td>
<td></td>
</tr>
<tr>
<td>Fasciculation</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wasting</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Inference
AIM

To chart the uniocular visual field of a given subject using Lister’s perimeter.

Definition of Visual Field

The field of vision is defined as the sum of the objects that form images on the retina when the eye is fixed at a particular point. Various methods are available for measuring the field of vision.

Perimetry

1. The instrument used for accurate charting of the field of vision is called a perimeter (Fig. 28.1). It consists of metal arc graduated in degrees. The metal arc is either a quadrant or semicircle of a circle. The concavity of the arc faces the subject. A holder sliding in the arc carries a test object, which can be glided along the length of the arc. The arc is pivoted at the center and can be rotated through various angle. For plotting the field of vision, test object of different colors and sizes can be used. The subject is asked to rest his chin on one of the two chin rests. When the field of the right eye is to be plotted the subject rests his chin on the left chin rest. This brings his eye in line with the fixation point.

2. Before mapping the field of vision the perimetry chart is studied. The lines through the center of the circle denotes the various meridian in degrees. Each concentric circle around the center denote points of equal visual acuity called isopters and are marked in degrees from the central fixation point. The perimter is used to chart the visual field between the periphery
and the 30° isopter circle. The metal arc is adjusted in one meridian and the object is then brought gradually from the periphery (90°) towards the center (fixation point). Keeping the eye on the central fixation point, the subject is asked to point at which he just sees the object. This reading in degrees on the arc is marked on the chart for that particular meridian. In this way the procedure is repeated at every 15° intervals till the field of vision is plotted in four quadrants. Care is to be taken to mark the blind spot along the horizontal meridian in the temporal quadrant (Figs 28.2A and B).

**Figs 28.2A and B:** (A) The perimeter; (B) The perimetric chart

**QUESTIONS**

1. Give the medial, lateral, superior and inferior extent of the field of vision. Explain the differences in the various extents.
2. Draw a diagram to show the possible lesions in the optic tract and their effects on the field of vision.
3. Mention the precautions for performing perimetry.
4. Mention the factors affecting the field of vision.
5. What is blind spot?
6. What is tubular vision?
7. What happens when there is damage to optic chiasma centrally and peripherally?
8. What is optic radiation?
9. What are the uses for perimetry?
10. What is binocular vision?
11. What is bitemporal hemianopia and its cause?
12. What is binasal hemianopia and its cause?
13. What is homonymous hemianopia?
14. What is stereoscopic vision?
15. What is scotoma?
16. Name any other method of determining the field of vision.
Cerebellum plays a major role in coordination of rapid muscular activities and helps to sequence the motor activities and also monitors and makes corrective adjustments in the body motor activities as they are performed.

In cerebellar dysfunction, the movements ordinarily overshoot their intended mark. This effect is called dysmetria, and it results in incoordinated movements that are called ataxia.

Tests for ataxia and dysmetria
1. **Past pointing:** When the subject attempts to touch an object with a finger, it results in overshooting to one side or the other.
2. **Finger-nose test:** The subject is asked to touch his nose with one index finger and then with the other with eyes open.
3. **Finger-finger-nose test:** The subject is asked to touch his nose first and then the examiner’s forefinger with his index finger.
4. **Counting:** The subject is asked to touch each finger in turn with the thumbs rapidly.
5. **Dysdiadochokinesia:** It is the inability to perform rapidly alternating opposite movements. The subject is asked to flex the elbow to a right angle and then alternately supinate and pronate the forearm as rapidly as possible.

Other tests for coordination:
1. Everyday movements like dressing (or) undressing, handling a book, picking up pins, threading a needle is observed.
2. **Knee-heel test:** In cerebellar ataxia, a characteristic irregular side to side series of errors in the speed and direction of ataxia occurs. The test should be performed with eyes open.
3. **Draw a circle test:** The subject is asked to draw a large circle in the air with the toe and put a dot in the center. In cerebellar ataxia, it is squared off irregularly.
4. The subject is asked to walk along a straight line.
Dysarthria

It is the defective articulation of speech. The subject speaks slowly and deliberately syllable by syllable as in scanning a line. "EGG" is pronounced as "ENG". This staccato speech rhythm is characteristic of severe cerebellar dysarthria. Minor cerebellar dysarthria causes slowing and slurring of speech.

Intentional Tremor (Action Tremor)

When a person with cerebellar dysfunction performs a voluntary act, the movements oscillate, especially when they approach the intended mark. The subject is asked to lift an object, such as a glass of water, and the presence of tremors is looked for.

Nystagmus

Cerebellar nystagmus is a tremor of the eyeballs that occurs usually when one attempts to fix the eyes on a scene to one side of the head.

Hypotonia

In cerebellar disease, there is decreased tone of the peripheral body musculature on the side of the lesion.

Rebound Phenomenon

The subject is asked to flex the forearm against resistance. If the resistance is suddenly withdrawn, the patient cannot break the movement of the limb and the forearm sways backward in the wide arc. This is because in cerebellar diseases, the patient is unable to stop movement promptly.

Decompensation of Movement

In performing a complex action involving more than one joint, e.g. touching the back of the head with the finger, it is not done smoothly as one movement is split into its component parts.

Pendular Knee Jerk

After eliciting patellar tendon reflex, the limb will swing back and forth, like a pendulum a few times, because of hypotonia.

Gait

Reeling (or) drunken gait occurs in cerebellar ataxia. Patients with this gait disorder walks on a broad base with the feet placed widely apart and irregularly.
**QUESTIONS**

1. Name the organs responsible for equilibrium.
2. Name the vestibular apparatus.
3. Name the three types of cerebellum phylogenetically.
4. Name the nucleus of the cerebellum.
5. What is dysdiadochokinesia? Examples.
6. What is intentional tremor and resting tremor?
7. What is tetany?
8. What is Romberg’s sign?
9. Name the two ascending spinocerebellar tracts.
10. What is dysmetria?
11. What is nystagmus?
12. What is tinnitus?
13. What is the receptor in the semicircular canals and otolith organs?
14. How is the cerebellum connected to the brain stem?
15. Which nucleus of the cerebellum is concerned with coordination? Name the tract connecting this to the cortex.
16. Mention some abnormal gaits.

**CEREBELLAR FUNCTION TEST**

<table>
<thead>
<tr>
<th></th>
<th>Eyes open</th>
<th></th>
<th>Eyes closed</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Tone</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Finger-nose test</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Finger-finger-nose test</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Counting of fingers</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Dysdiadochokinesia</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Rebound phenomenon</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
<tr>
<td>Threading a needle</td>
<td></td>
<td>**********</td>
<td></td>
<td>**********</td>
</tr>
</tbody>
</table>

Name:      Age:  
Sex:      Occupation:
### LOWER LIMB

<table>
<thead>
<tr>
<th>Eyes open</th>
<th>Eyes closed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Right</td>
<td>Left</td>
</tr>
<tr>
<td>Tone</td>
<td>X</td>
</tr>
<tr>
<td>Gait</td>
<td>X</td>
</tr>
<tr>
<td>Walk along a straight line</td>
<td>X</td>
</tr>
<tr>
<td>Tandem walk</td>
<td>X</td>
</tr>
<tr>
<td>Knee-jerk</td>
<td>X</td>
</tr>
<tr>
<td>Knee heel test</td>
<td>X</td>
</tr>
<tr>
<td>Draw a circle</td>
<td>X</td>
</tr>
<tr>
<td>Romberg's sign</td>
<td>X</td>
</tr>
</tbody>
</table>

**Others**
- Speech
- Nystagmus

**Inference**
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