UNIT-I

Bio-potential Generation & Electrode types

Origin of bio potential & its propagation. Types of electrodes - Surface, needle & micro electrodes & their equivalent circuits. Recording problems - measurement with two electrodes.

Origin of Biopotential & its propagation

Biopotential are electric fields associated with the electrochemical activity of a certain class of cells & give information of the activity of the organs.

These potentials are

- → Electrocardiogram (ECG)
- → Electro encephalogram (EEG)
- → Electro myogram (EMG)
- → Electro oculogram (EOG).

Transducer → convert these ionic potentials into to electrical signal.

Bio potential signals:

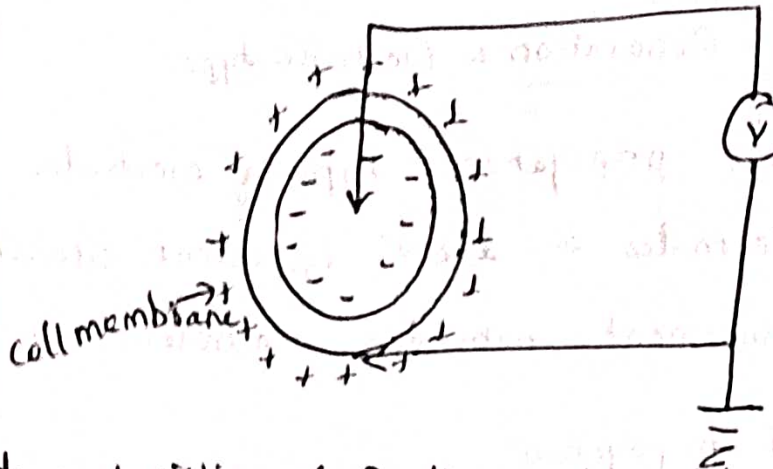
Bio electric signals are picked up by means of electrodes before the subsequent proces like amplification & filtering.

Resting potential

The membrane potential measured when an equilibrium with a potential difference across the cell membrane negative on the inside & positive on the outside is called Resting potential.

Resting potential → -60mV to -100mV

Cell is at resting state → polarized cell.



Characteristics of Resting potential

→ the resting potential is maintained as constant.

→ It depends on temperature.

→ permeability of different cells types will vary.

Resting potential → -60 to -100 mV

Resting potential V_R is derived by Goldman's equation.

$$V_R = -\frac{kT}{q} \ln \left[\frac{P_K [K^+]_i + P_{Na} [Na^+]_i + P_{Cl} [Cl^-]_o}{P_K [K^+]_o + P_{Na} [Na^+]_o + P_{Cl} [Cl^-]_i} \right]$$

o → outside the cell

i → inside the cell.

V_R → Resting potential.

T - Absolute temperature in kelvin.

k - Boltzman constant

q - charge

P_K - permeability of potassium ion

P_{Na} → " of sodium ion

P_{Cl} → " of chloride ion.

$[K^+]$ $[Na^+]$ $[Cl^-]$ → concentration of potassium, Sodium, chloride.

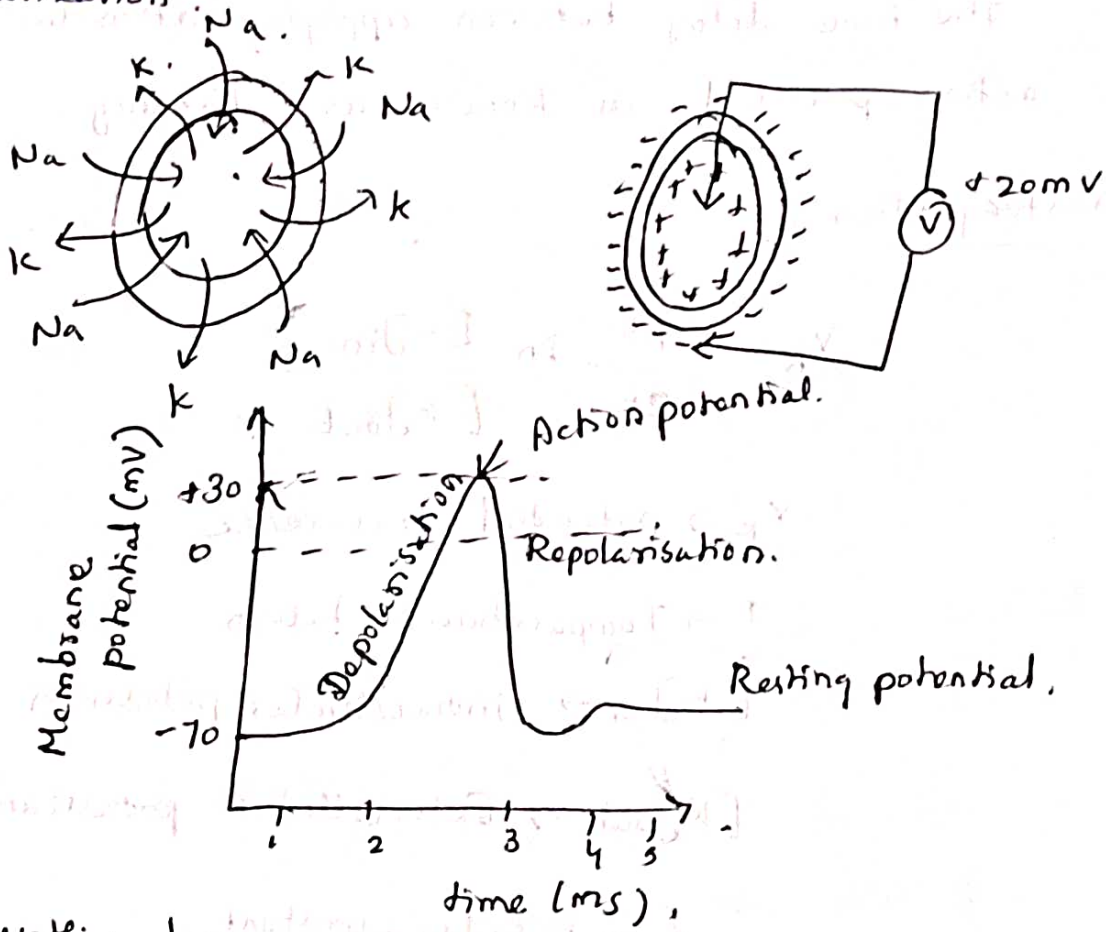
potential :

When the stimulus is applied to a cell at the resting stage, high concentration of positive ion inside & negative ion outside is called Action potential.

Action potential → +20mV.

→ A cell that has been excited & that display an action potential is said to be depolarized & process from Resting to Action potential is called depolarization.

→ The process from Action to Resting potential is called Repolarization.



All or Nothing Law

In the method of the excitation of cell, S greater than Stimulus of threshold, the action potential is always

Same for any given cell. this is known as the all or

Absolute Refractory period. time duration in which cell figuration cannot respond to any stimulus is called absolute Refractory period. It is 1ms in nerve cells.

Relative Refractory period:

During which another action potential can be triggered but a higher stimulus is required to reinitiate the action potential & the subsequent contraction of muscles.

It is several ms.

Latency

The time delay between applying stimulus & receiving the action potential is known as Latency.

Nernst equation

$$V_K = -\frac{RT}{zF} \ln \frac{[K]_{in}}{[K]_{out}}$$

$V_K \Rightarrow$ potential difference

$T \rightarrow$ Temperature in kelvin

$[K]_{in} \rightarrow$ intracellular potassium

$[K]_{out} \rightarrow$ Extracellular potassium.

F – Faraday's constant.

$R \Rightarrow$ Gas constant.

$z \rightarrow$ valance of ion.

Electrodes:

→ Device that converts ionic potential into Electronic potentials are called electrode.

→ It is used to pickup the electrical signals of the body.

Types of electrode :-

a) Micro electrode → To measure the bioelectric potential near or single cell

b) Depth and Needle electrode → Measure biopotential of highly localized extracellular region eg brain.

c) Surface electrode.
↓
To measure the surface of skin
eg ECG, EEG, EMG.

a) Micro electrode :-

These are used to measure the biopotential near or within a single cell. These are also called as Intracellular electrodes.

Types of micro electrode :

a) Metal microelectrode

b) Non-metallic microelectrode.

Metal Microelectrode :-

* Metal microelectrode are formed by electrolytically etching the tip of a fine tungsten (or) stainless steel wire to a fine point.

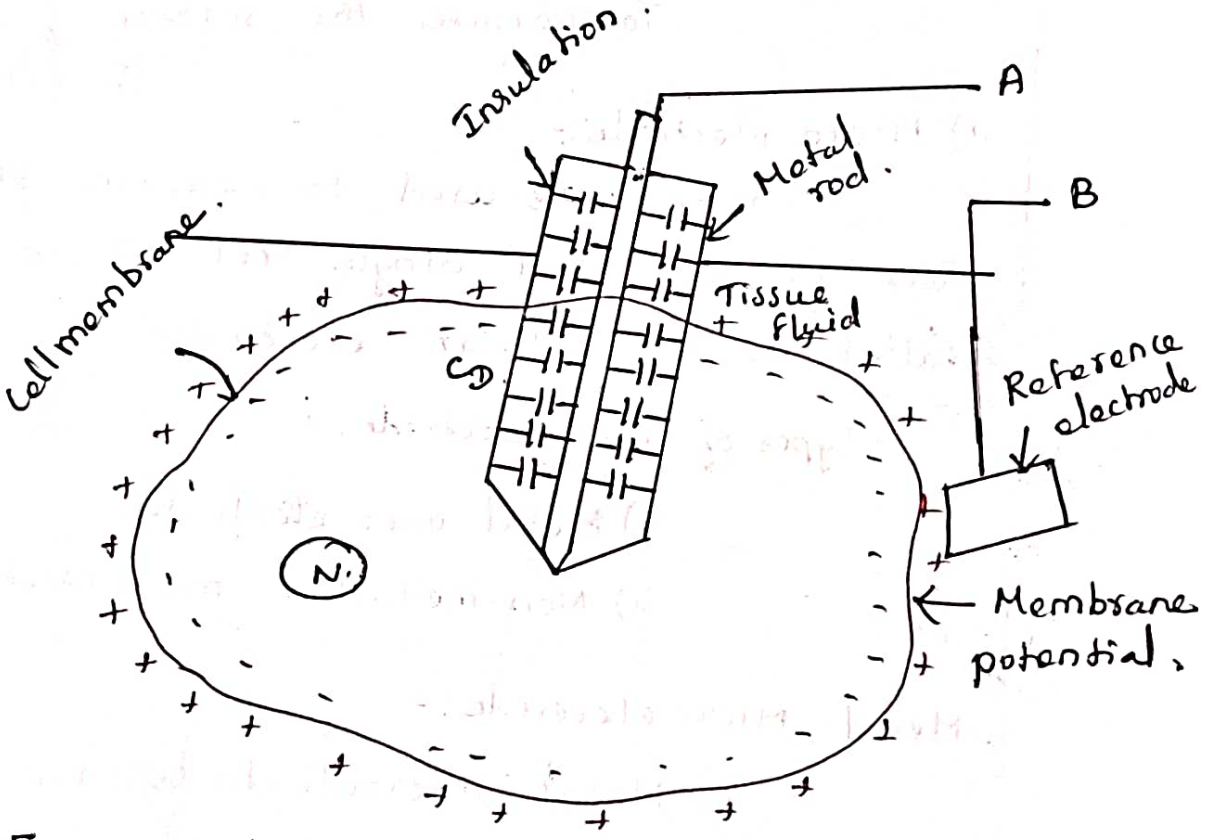
* This technique is known as electro - pointing.

* The metal microelectrode are almost to the micro tip with an insulating mate

To reduce the impedance

* Some electrolytic processing like chloridi the tip and then developing by the photographic developer can be performed.

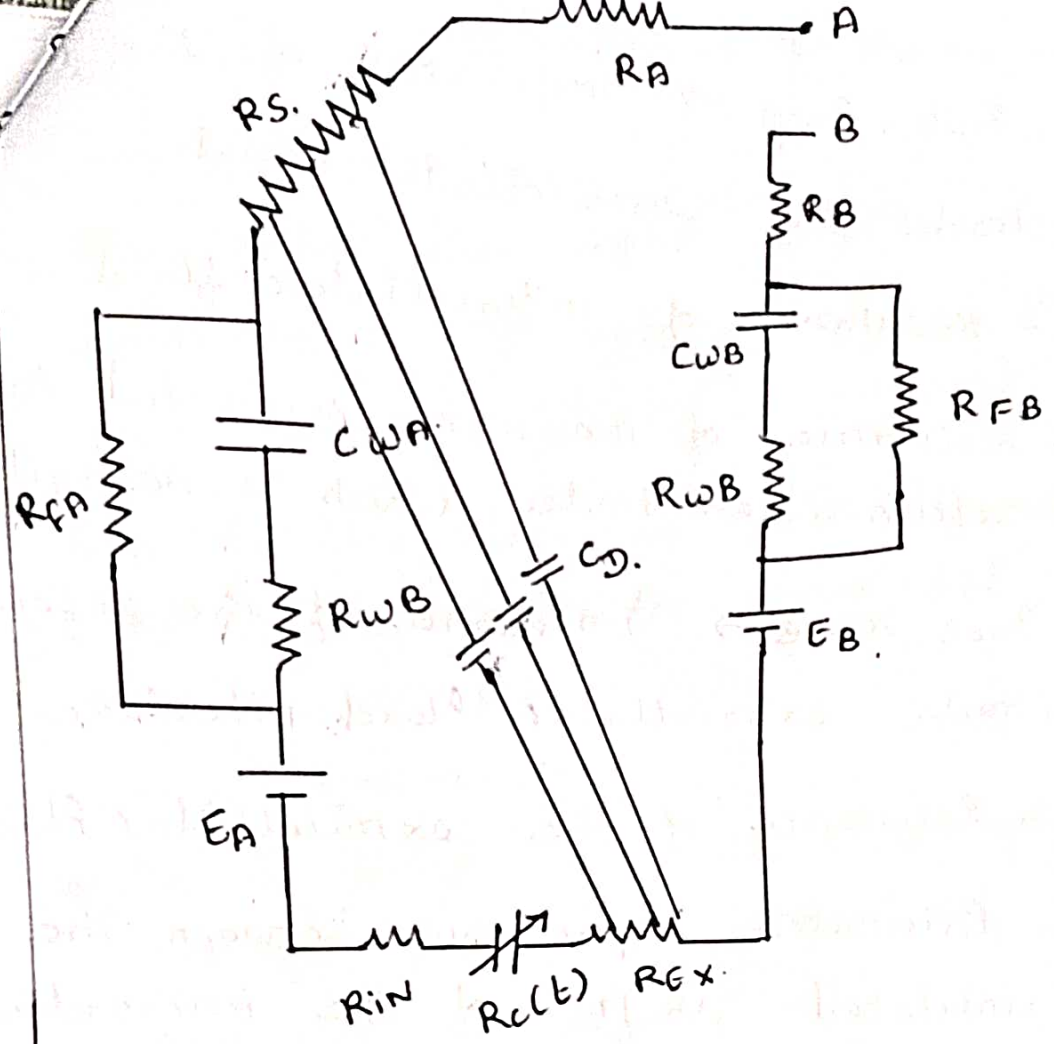
* The Measurement of bioelectric potential requires two electrodes, the voltage is measured as the difference between the micro electrode & reference electrode.

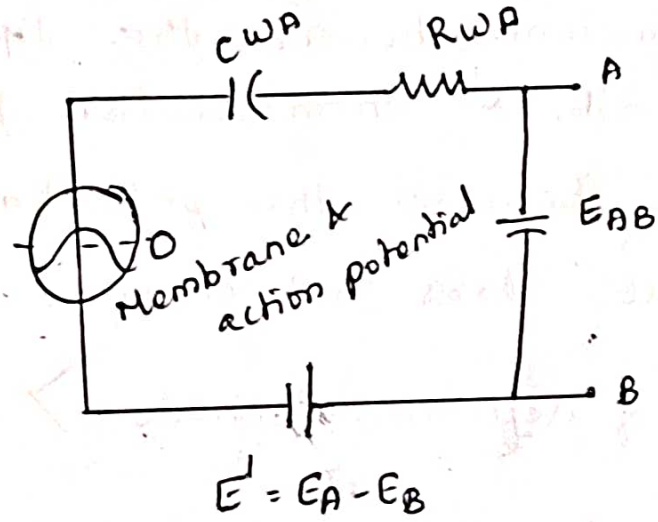


E_A - metal electrode - electrolyte potential at the micro electrode tip.

E_B - Reference electrode - electrolyte potential,

E_C - membrane potential.





$E' = E_A - E_B$

$R_A \rightarrow$ Resistance of the connecting wire which is negligible.

$R_S \rightarrow$ Resistance of the shaft of micro electrode which is negligible.

$\therefore E = E_A + E_B + E_C$.

R_{FA}, R_{WA}, C_{WA} → impedance of the electrode tip intracellular fluid.

R_{IN} - Resistance of intracellular fluid.

R_B - Resistance of the wire connected to the reference electrode which is negligible.

R_{FB}, R_{WB}, C_{WB} → impedance of the reference electrode - extracellular fluid interface.

R_{EX} - Resistance of the extracellular fluid.

C_D - distributed capacitance between the insulated shaft of the microelectrode & extracellular fluid.

* The capacitance between the tip of the microelectrode & intracellular fluid is negligible because the potential difference across it does not change.

Area of reference electrode > Metal electrode

* Impedance of micro electrode is inversely proportional to the Area of the tip & frequency

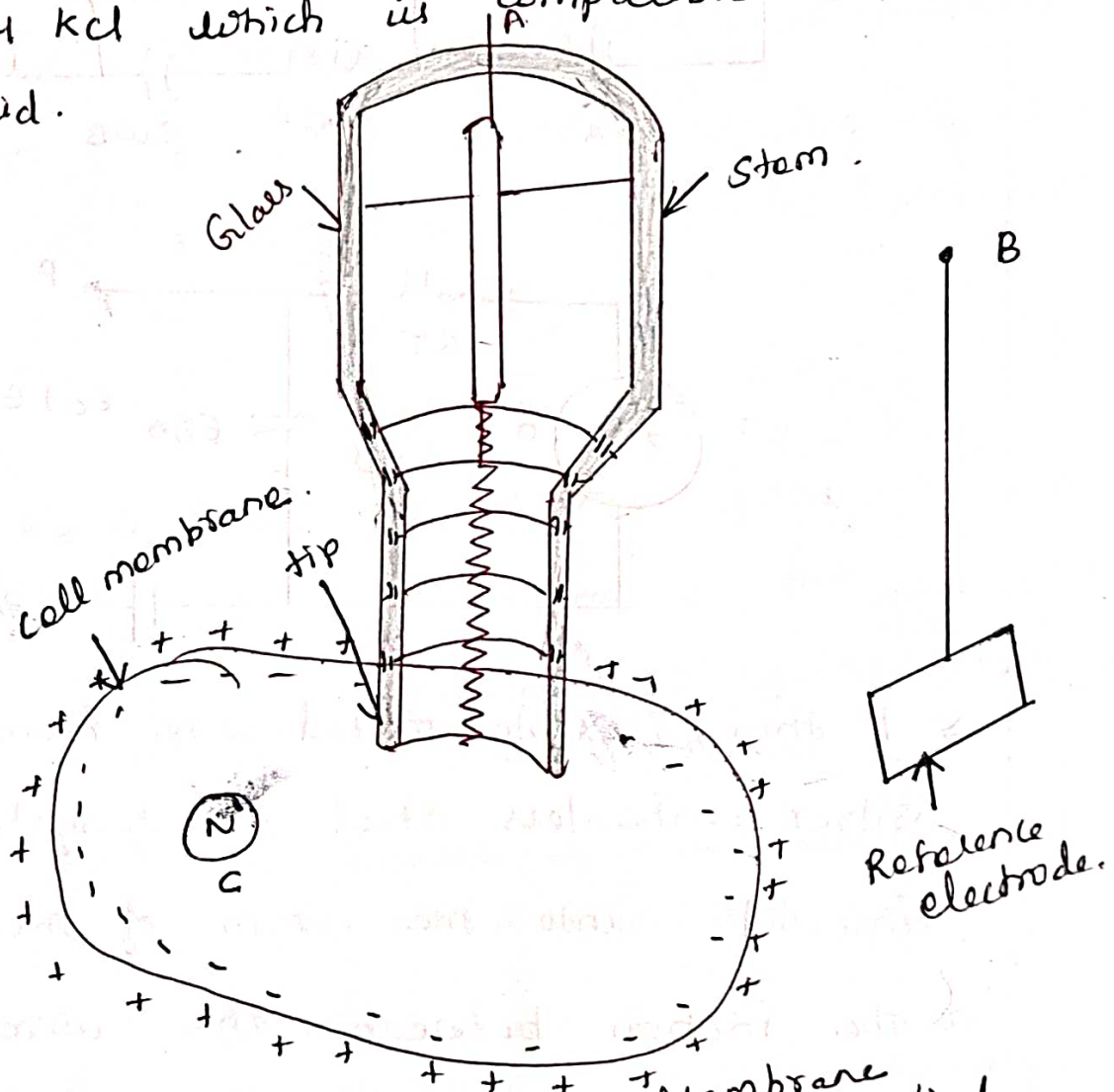
* The electrode output is coupled with an amplifier, Low frequency component of Biopotential will be attenuated.

if the input impedance of the amplifier is not high.

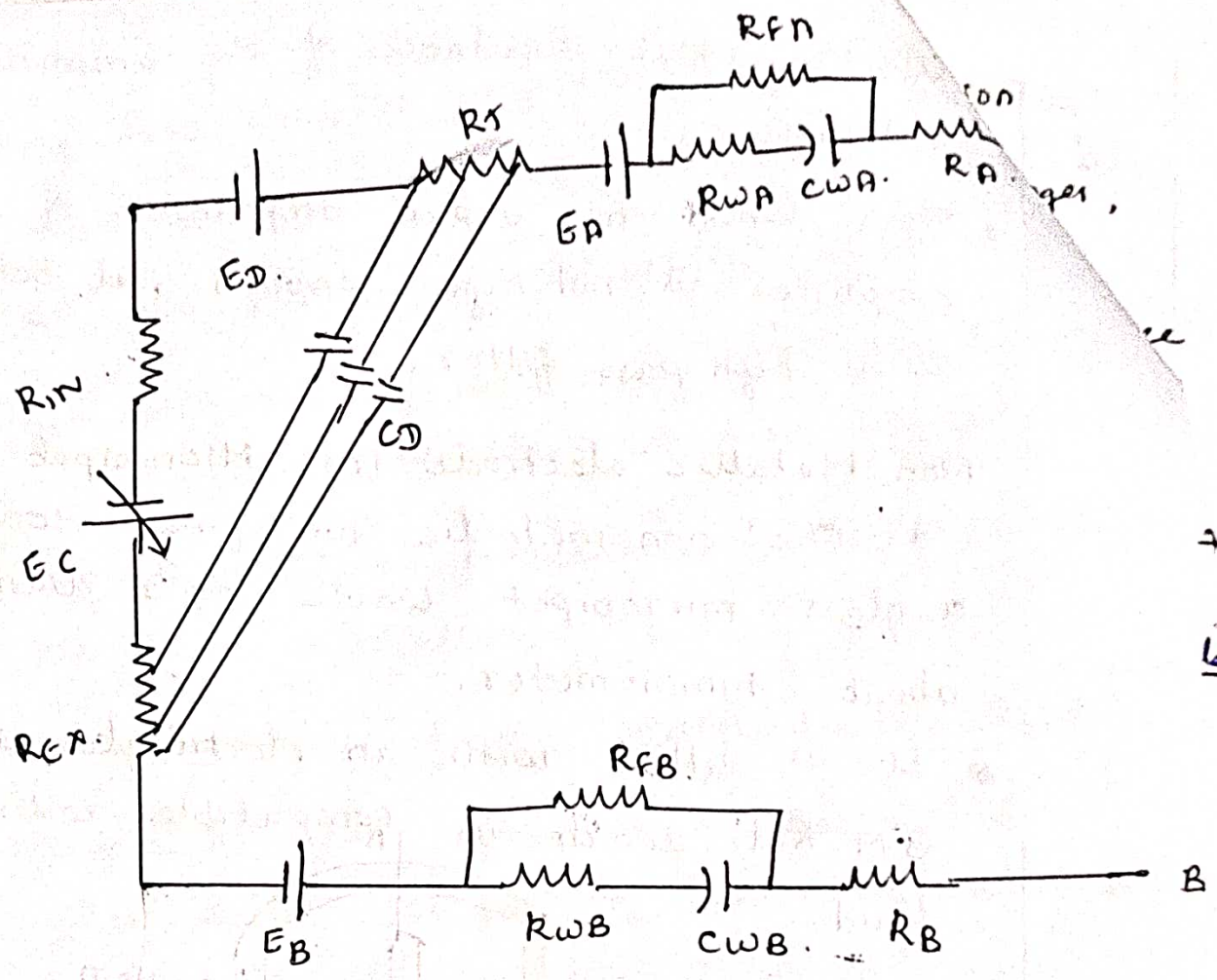
* Thus when the input impedance of the amplifier is not high enough, it behaves as a high pass filter.

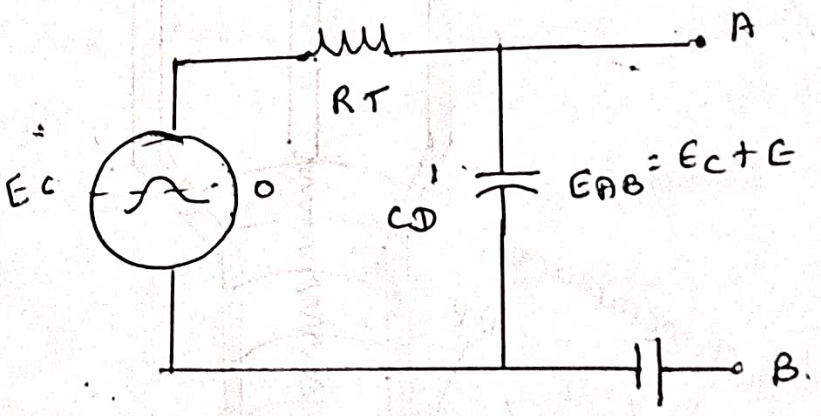
Non Metallic electrode (or) Micropipet.

* The non metallic micropipet consist of a glass micropipet whose tip's diameter in about 1 micrometer.
* It is filled with an electrolyte usually 3 M KCl which is compatible with cellular fluid.



N → Nucleus.
C → cytoplasm.





→ A thin, flexible metal wire from chlorided silver, stainless steel or tungsten is inserted into the stem of the micropipet.

→ the friction between the wire & stem of the micropipet & the fluid surface

tension hold the micropipet on the wire.

→ the other end of the metal wire is mounted to a rigid support and the other free end is mounted to a rigid support and the other free end of its resting on the cell.

E_A - potential between the metal wire & electrolyte filled in micropipet.

E_B - potential between the reference electrode & the extracellular fluid.

E_C - variable cell membrane potential.

E_D - potential existing at the tip due to different electrolyte present in the pipet.

$$E = E_A + E_B + E_D + E_C.$$

R_B → Resistance of the connecting wire

R_{FA}, R_{WA}, C_{WA} → impedance of the electrolyte filling the tip of the micropipet. Which is very large.

R_{IN}, R_{EX} → Resistance of the electrolyte inside the cell & electrolyte outside the cell.

R_{FB}, R_{WB}, C_{WB} → reference electrode - electrolyte interface impedance.

R_B - resistance of the wire connected with reference electrode.

C_D - distributed capacitance existing between fluid in the pipet & the extracellular fluid.

→ When the micropipet is coupled with the amplifier terminal A & B, then the membrane potential E_C is coupled with it via a high series Resistance 'R_T'. & a moderate shunt capacitance C_D along with electrode potential.

→ The impedance of the electrode places a limit on the response time of the circuit such that it behaves as a Low pass filter when the input impedance of the amplifier is not high enough.

Depth electrode:-

It is used to study the electrical activity of neuron in superficial Layer of the Brain & also the oxygen tension.

- Bundle of Teflon insulated platinum (90%) & iridium alloy (10%) wires.
- End of supporting wire is rounded.

Area: $0.5 mm^2$.

Depth & Needle electrode:

Needle electrode are used in clinical electromyography, neurography & other electrophysiological investigation. of the muscle tissue underneath the skin & in the deeper tissues.

Material :- Stainless steel.

Types of needle electrode.

→ monopolar needle electrode
→ Bipolar needle electrode.
→ concentric (coaxial) core needle electrode
→ Multielement needle electrode.

Monopolar Needle Electrode:-

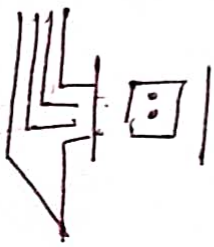
It consist of Teflon coated stainless steel wire which is bare only at the tip.

Bipolar (double coaxial) needle electrode:-

* It contain two insulated wire within a metal cannula. The two wires are bared at the tip & provide contact to the patient.

* The cannula act as a ground.

Bipolar electrodes are electrically symmetrical & have no polarity sense.



concentric (coaxial) core needle electrode:-

* It consist of both the active & ref. electrode within the same structure.

* It consist of an insulated wire contained within a hypodermic needle.

* The inner wire is exposed at the tip & this form one electrode.

* very stable characteristics.

Material :- platinum wire.

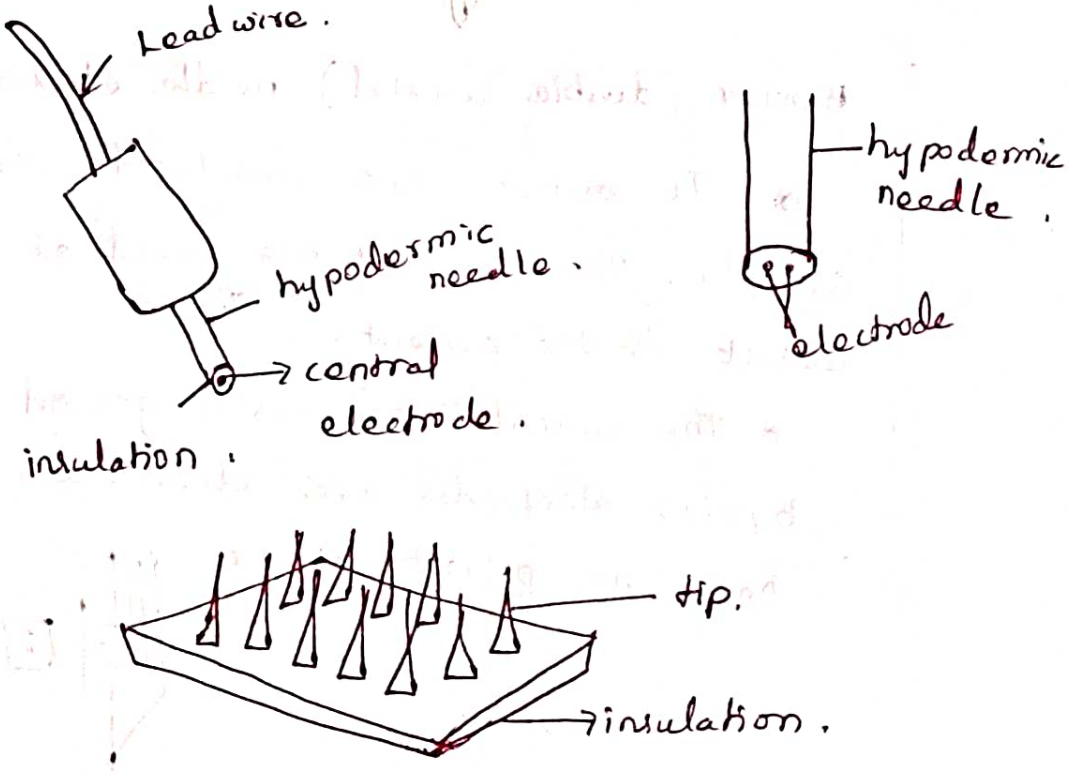
Area = 0.0005 mm^2.

diameter = Less than 0.6 mm.

multielement needle :-

It is used to pick up the signal from the individual fibres of the muscle tissue.

Diameter - 25 micron.



B Surface Electrode:-

It is used to sense ECG potential and smaller area surface electrode are used to sense EEG & EMG potentials.

Types of electrodes.

→ Metal plate electrode.
→ Suction cup
→ Multipoint
→ Adhesive tape.
→ Floating electrode.

Metal plate electrode:-

Shape: Rectangular (3.5cm x 5cm)
Circular (4.75 dia)

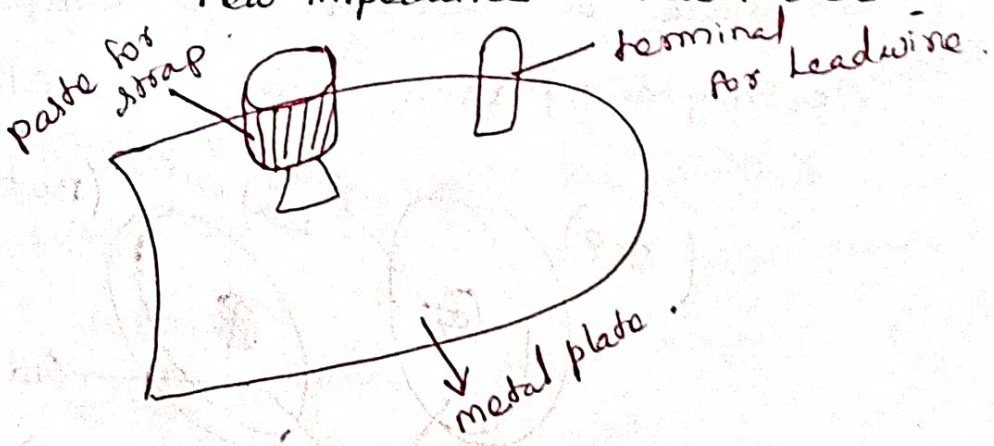
Material:- German silver, nickel silver or nickel plated steel.

→ ECG measurement.

These electrodes are applied on the skin with electrode paste.

dc resistance value - 2-10kΩ

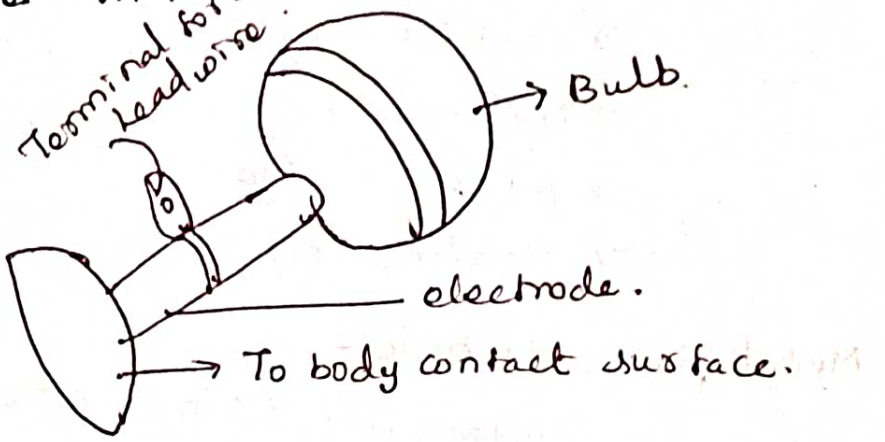
Few impedance - Few 100 Ω



Suction cup electrode:-

→ It is attached to the flat surface of the body & to region where the underlying tissue is soft.

→ The electrode has small area because only the rim of it is in contact with the skin.

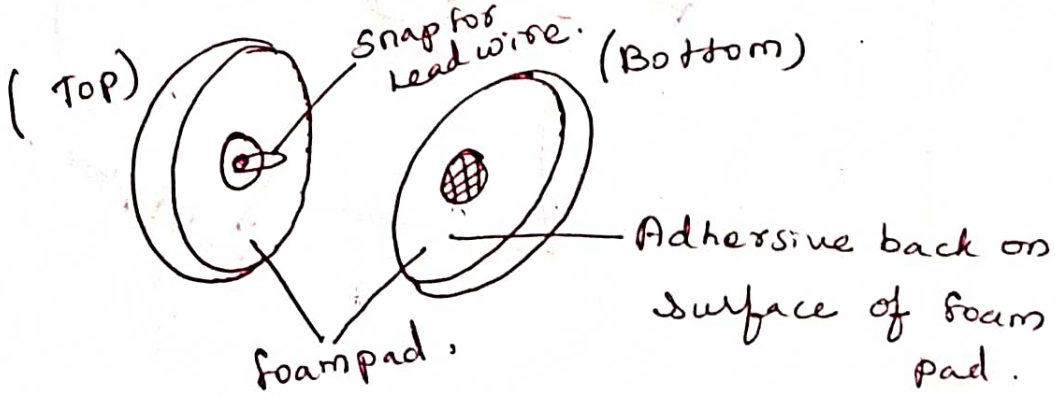


Adhesive tape electrode:-

To avoid the electrode paste squeezing from the skin, adhesive type electrode is used.

→ It consist of light weight, metallic screen backed by a pad for electrode paste.

→ The adhesive backing hold the electrode in place & retards the evaporation of the electrolyte present in the electrode paste.



Multipoint electrode :-

- ECG measurement
- It contain nearly 1000 fine active contact point.
- very small & very low Resistance contact.
- If the subject has hair on the region of interest, then one can use the multipoint electrode without removing the hair.
- use under any environmental condition.

Floating electrode :-

The interface can be stabilized by the use of floating electrode in which metal electrode doesn't make direct contact with the skin.

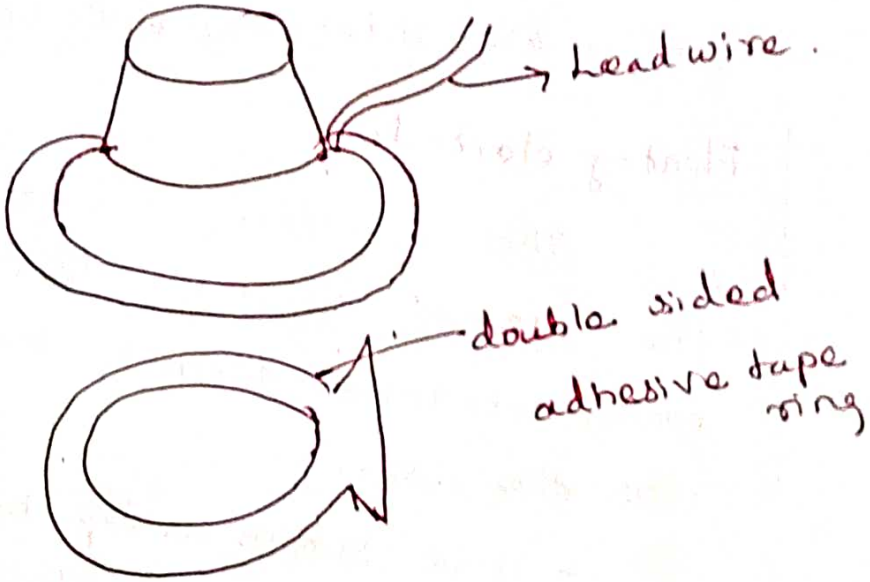
- It is known as top-hat electrode.

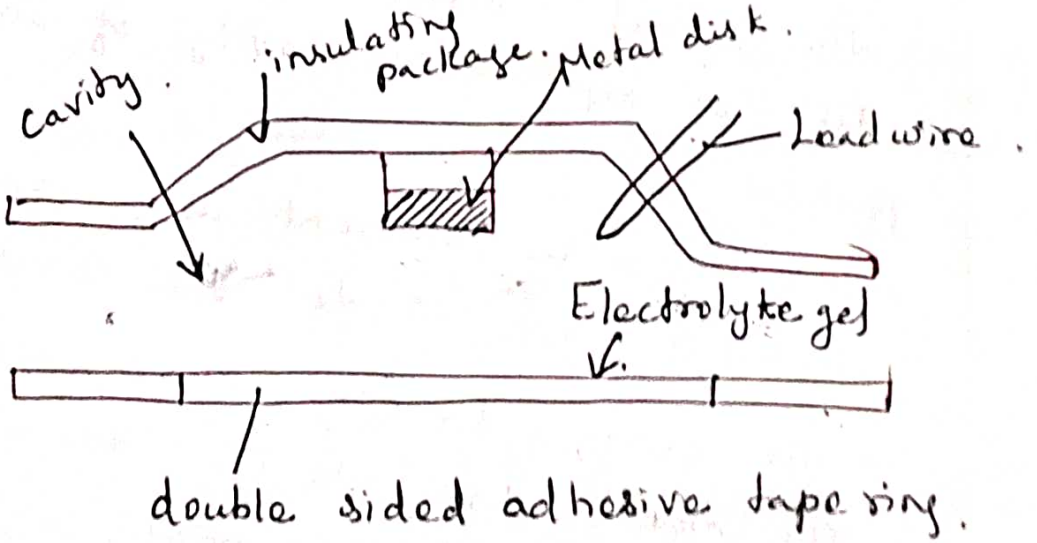
The actual electrode element or metal disk is kept in a cavity of insulated package.

→ The electrode element is surrounded by electrolyte gel inside the cavity.

→ The cavity doesn't move with respect to the metal disk, so it doesn't produce any Mechanical movement.

- Electrolyte gel is attached to the [illegible]
  by means of a double sided adhesive tape ring.
- the electrode disk is made of silver metal & coated with silver chloride.
- impedance of the electrode is 50 kΩ.
- Light weight & donot make use of electrode Jelly.





Recording Problem :-

Electrodes play an important part in the satisfactory recording of bioelectric signals & their choice requires careful consideration. they should be comfortable for the patient to wear over long periods & should not produce artifacts.

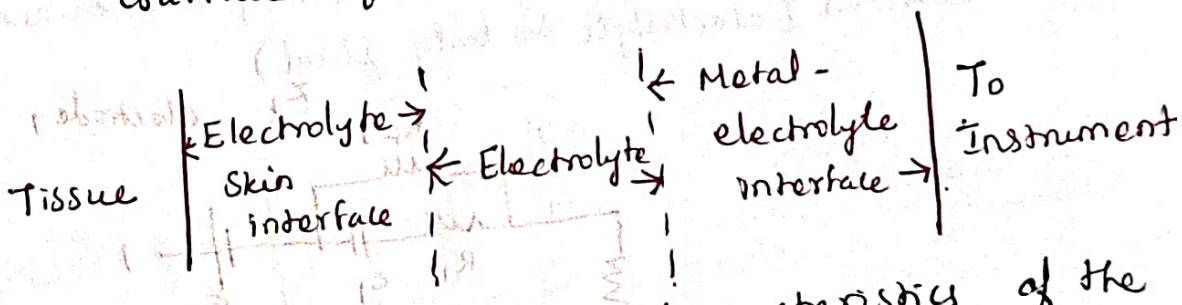
1. Electrode - Tissue Interface.
   - metal - electrolyte interface
   - electrolyte - skin interface.
2) polarization
3) Skin contact impedance
4) motion artefacts

1) Electrode - Tissue Interface.

→ To avoid the movement artefacts.

→ Low contact impedance

→ electrode paste is usually employed as an interface between the electrode & the surface of the source of event.



these are different characteristics of the surface electrode.

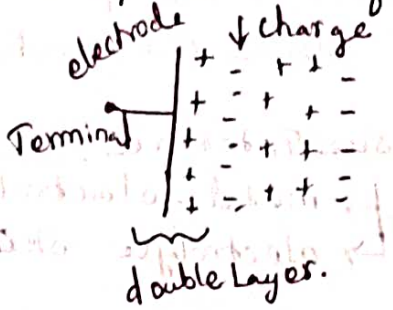
→ metal - electrolyte interface

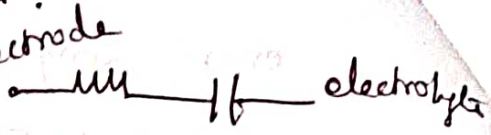
→ electrolyte - skin interface

→ quality of the electrode.

Metal - electrolyte interface:

At metal - electrolyte transition, th
a electrical double layer is formed at the adja
to the electrode. At metal - electrolyte interface the
voltage developed is half cell potential.





The difference in half cell potential that exist between two electrode is called offset potential.

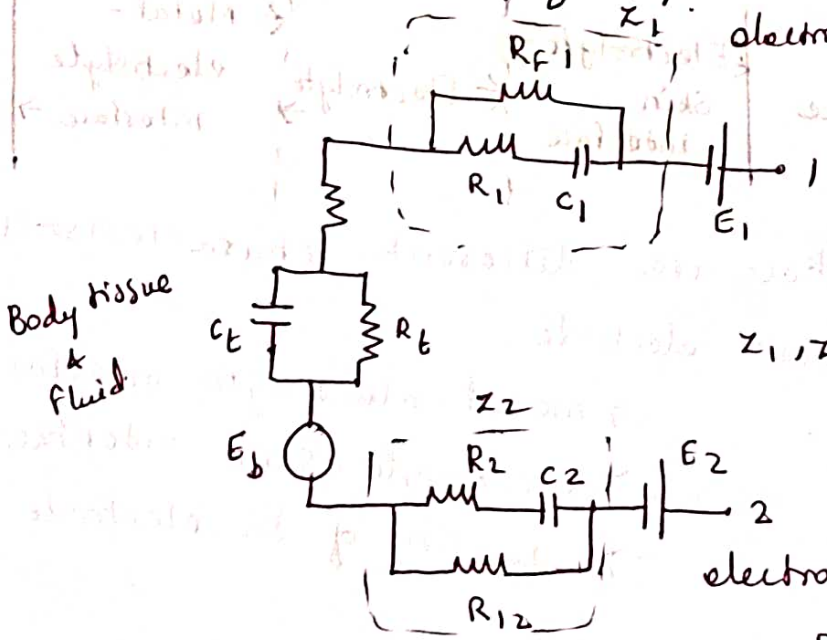
Eg Aluminium → -1.66 V

Iron Fe^{++} → -0.44 V

electrode metal	electrolyte	potential difference between electrode.
Stainless steel	saline	10 mV
silver	saline	94 mV.

Electrolyte - skin interface:

Skin is act as diaphragm between two solution (electrolyte & body fluid).



E_1, E_2 - half cell potential.

Z_1, Z_2 → skin contact impedance.

R_t → tissue Resistance

C_t → tissue Capacitance

E_b - Bioelectric event

To minimize the potential drop across the electrode impedance,

→ Skin contact impedance as low as possible.

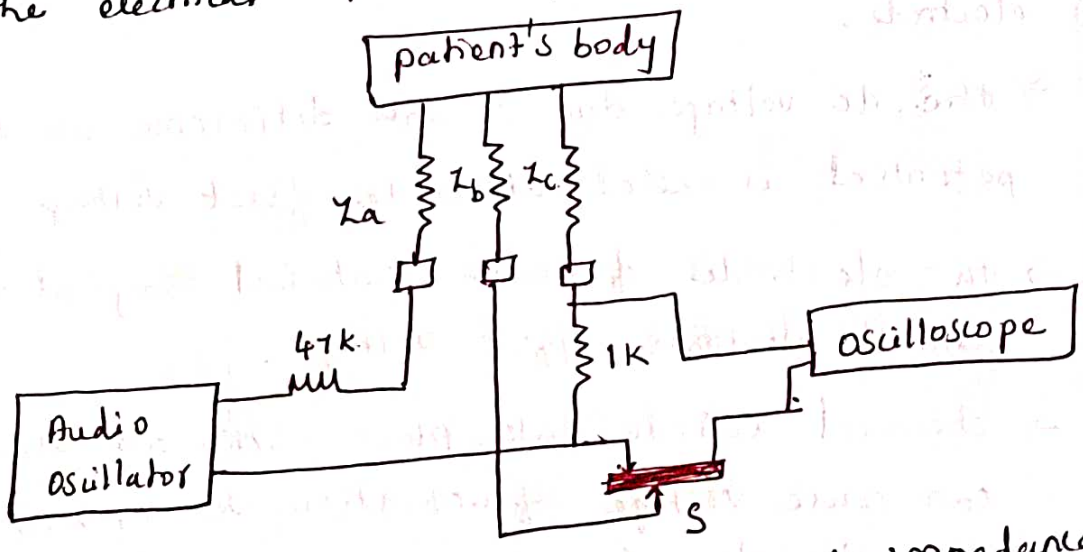
→ input impedance of the measuring device is high.

polarization:-

If a low voltage is applied to two electrode placed in a solution, the electrical double layer are distributed. Steady state current may or may not take place. double layer is disturbed, this cause the electrode measurement of slowly varying potential in the tissue. they are said to be polarized (or) non reversible.

Skin contact impedance:-

Skin electrode impedance is known as skin contact impedance. It must be greater than the electrical impedance of the body tissue.



(A,B,C) three electrode, Za, Zb, Zc { contact impedance of 3 electrode): F= 0.1 - 100Hz. The voltage drop across 1k resistor is calculated & skin contact impedance is measured.

Motion artefacts:-

Motion artefacts is a problem in biopoten measurement. This problem is occur in ECG. artefacts cause the signal → display unreadable
→ recording instrument ove its range,
→ incorrect output
→ False alarm.

Skin electrolytic paste interface major motion artefacts.
→ Skin irritation.
→ Magnitude of skin abration is reduced.

These all problems are reduced by proper design of electrodes.

Measurement of two electrodes:-

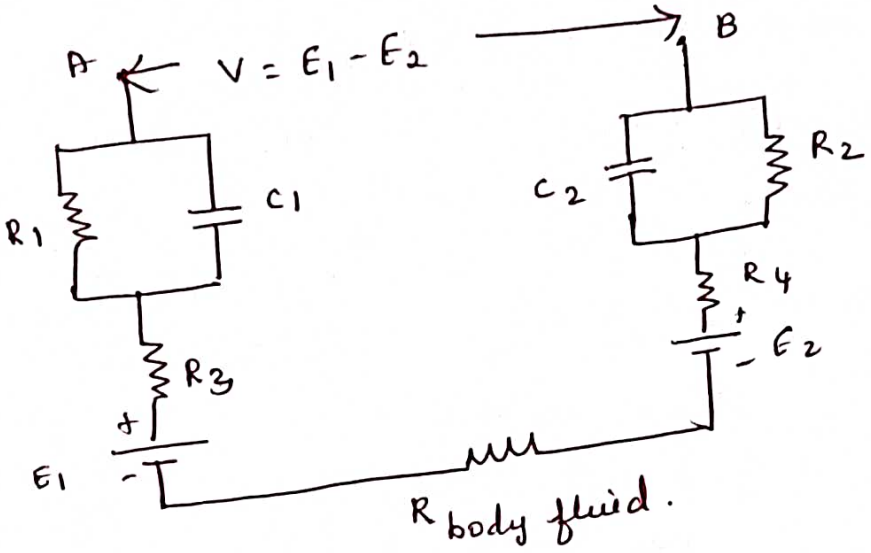
The voltage measured is the difference between the instantaneous potential of the two electrode.

→ the dc voltage due to the difference in electrode potential is called electrode offset voltage.
→ two electrodes of same material may also produce small electrode offset voltage.
→ chemical activity take place within an electrode can cause voltage fluctuation to appear without any physiological input.
→ such variation may cause noise in bio electric signal.

noise can be reduced by proper choice of material
(or) by coating the electrode to improve stability.

→ Ag - AgCl is very stable

It is prepared by coating a pure silver with silver chloride. This coating is done by placing silver into bromide free sodium chloride solution.



R3, R4 - series Resistance

impedance of electrode A

$$Z_A = R_3 + \frac{R_1}{1 + j2\pi f C_1 R_1}$$

impedance of electrode B

$$Z_B = R_4 + \frac{R_2}{1 + j2\pi f C_2 R_2}.$$

The value of voltage & impedance depend on the electrode metal, its area, the electrolyte, charge density & frequency of current.

unit - II

Bio signal characteristics & Electrode configuration

Bio signal characteristics, frequency & amplitude ranges, ECG - Einthoven's triangle, Standard 12 Lead system. EEG, 10-20 electrode system, unipolar, bipolar & average mode. EMG - unipolar & bipolar mode.

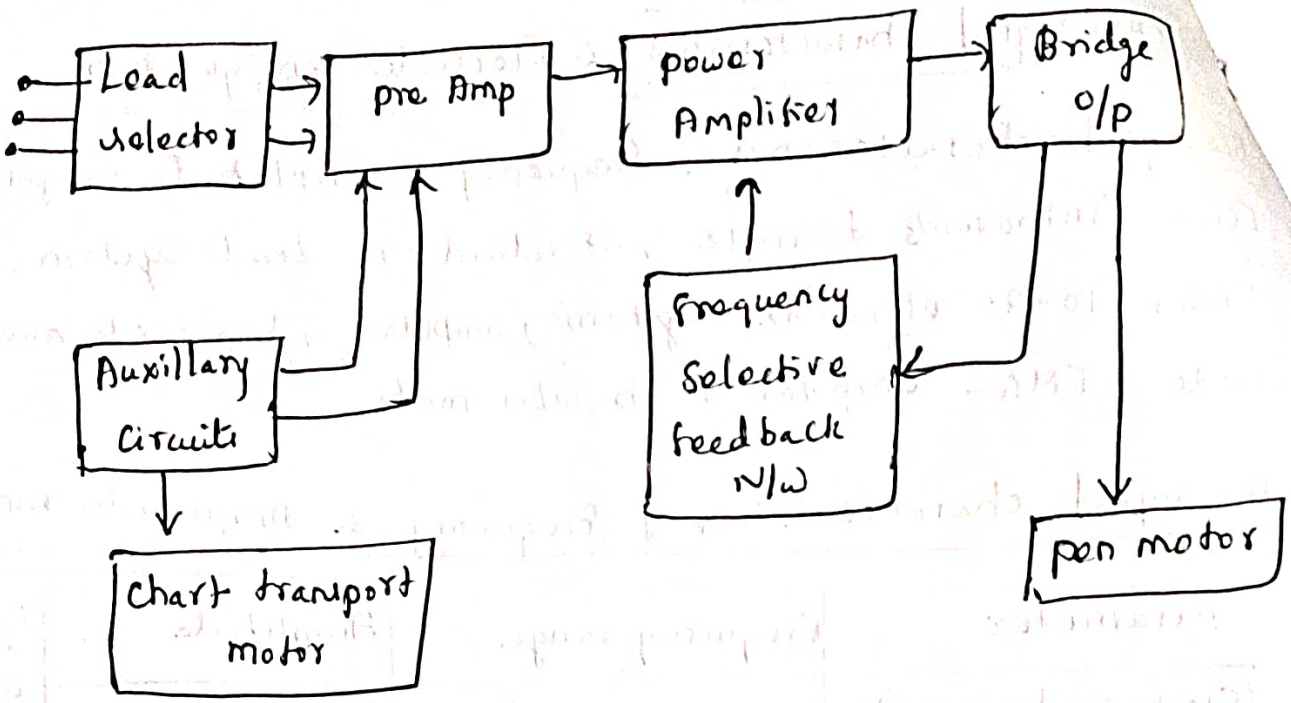
Bio signal characteristics, frequency & Amplitude ranges

parameter	frequency range	Amplitude	Type of electrode
(Electrocardiography) ECG	0.05 - 120 Hz	0.1 - 5 μV	surface
Electroencephalography (EEG)	0.1 - 100 Hz	2 - 200 μV	Scalp
Electromyography (EMG)	5 - 2000 Hz	0.1 - 5 μV	Needle
Electro retinography (ERG)	dc - 20 Hz	0.5 - 1 μV	contact
Electro oculography (EOG)	dc - 100 Hz	10 - 3500 μV	contact

ECG Electrocardiography

Bio electric potential generated by the heart muscle are called Electro cardiogram. sometimes (EKG).

The recording produced by this non invasive procedure is known as electrocardiogram.



Lead Selector :-

The potential picked up by the patient electrode are taken to the Lead selector switch.

preamplifier:

The signal from Lead selector switch given to preamplifier. A preamplifier is an electronic amplifier which prepares an electronic signal for further amplification.

power Amplifier:

It is a push pull differential amplifier.

o/p is single ended.

o/p of the power Amplifier is fed to the pen motor. Which deflects the writing arm of the paper

Frequency selective network It is an RC network which provide necessary damping for the pen.

Auxillary circuits: It provides 1mV calibration signal & automatic blocking of the amplifier during change in position of the Lead switch. It also include speed control ckt for driver motor.

Lead configuration

To record the ECG, 12 electrode are connected to the body of the patient. Electrodes connected to ECG machine using wire called Leads.

Leads are electrode which measure the difference in electrical potential between either.

Bipolar Leads → two different point on the body

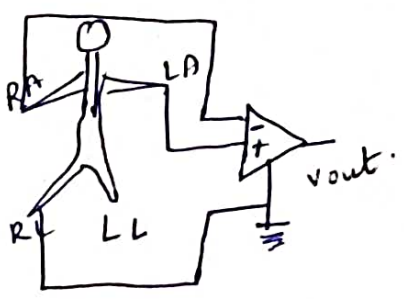
unipolar Lead →. one point on the body.

classification:

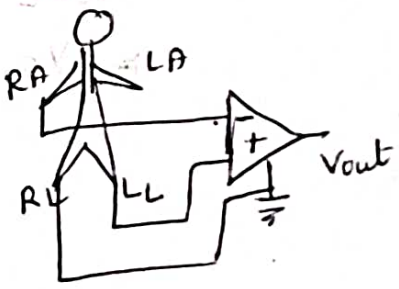
1) Standard Limb Leads (OR) Bipolar Leads (OR) Einthoven Leads.
   - Lead I
   - Lead II
   - Lead III
2) Augumented Limb Lead (OR) unipolar Lead.
   - aVR
   - aVL
   - aVF
3) precordial Lead (OR) chest Lead : $V_1 - V_6$

:) Standard Limb Leads.

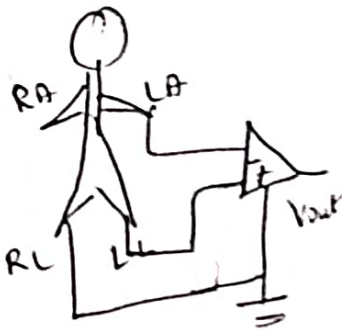
Lead I.



Lead II

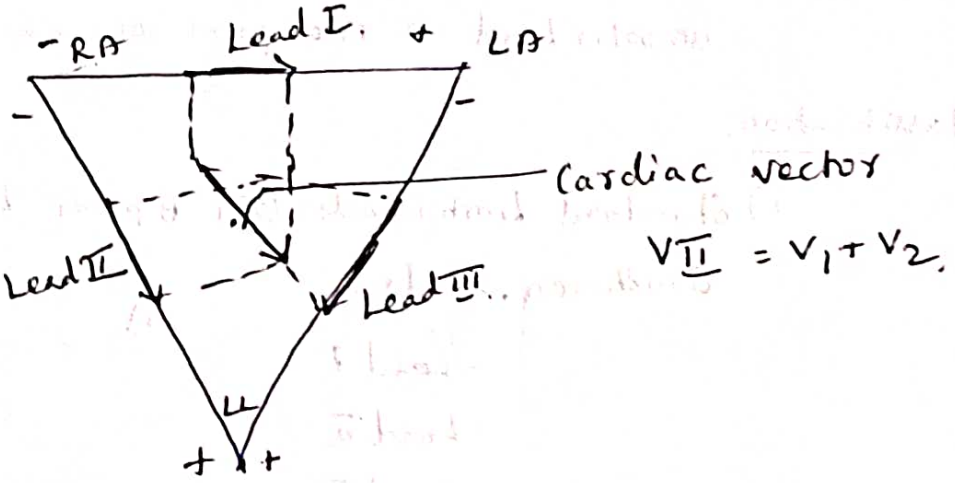


Lead III.



	+	−
Lead I	LA	RA
Lead II	LL	RA
Lead III	LL	LA

All the Lead reference point is connected to Right Leg.



The output voltage is V_{out}.

e) Augumented unipolar Limb Leads.

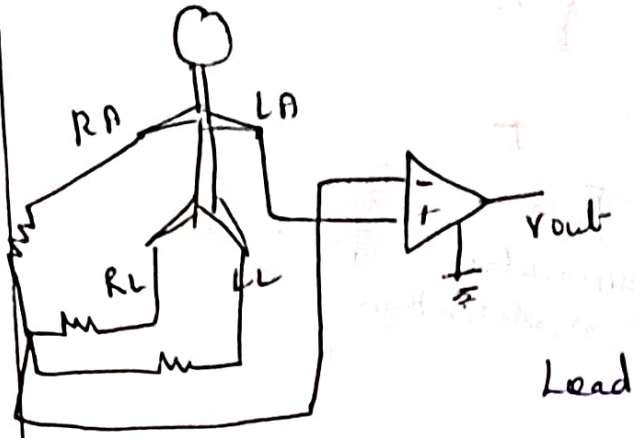
Two equal Resistors & Large resistors are connected to a pair of a Limb electrodes & the centre. Resistive network acts as a central terminal and remaining Limb electrode acts as a exploratory electrodes.

→ aVR

→ aVL

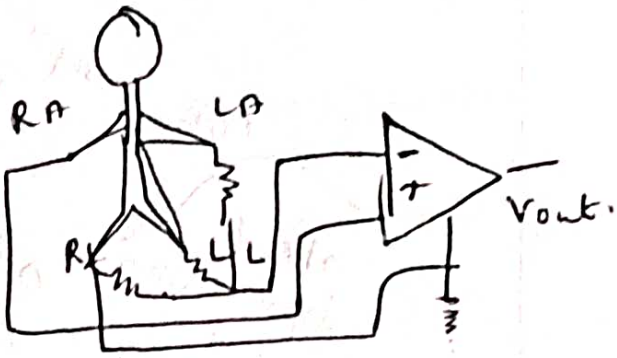
→ aV_F

Lead aVL



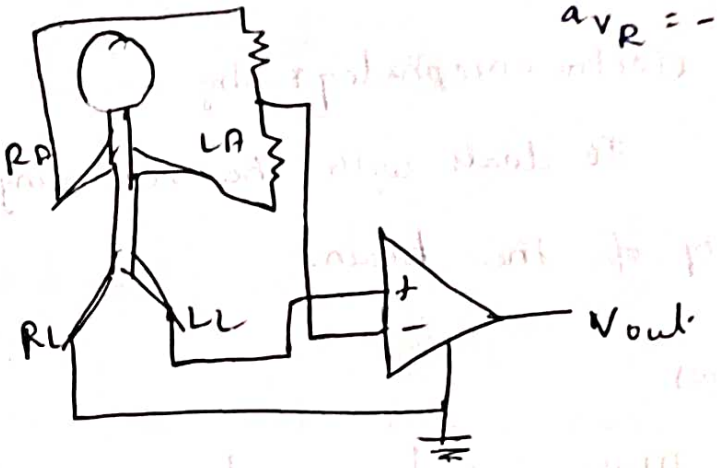
$$aV_L = V_I - \frac{V_{II}}{2}$$

Lead aV_R



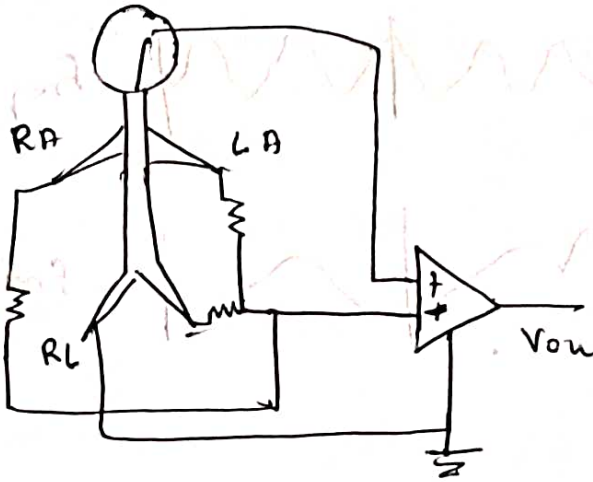
$$aV_R = -V_I - \frac{V_{III}}{2}.$$

Lead aV_F

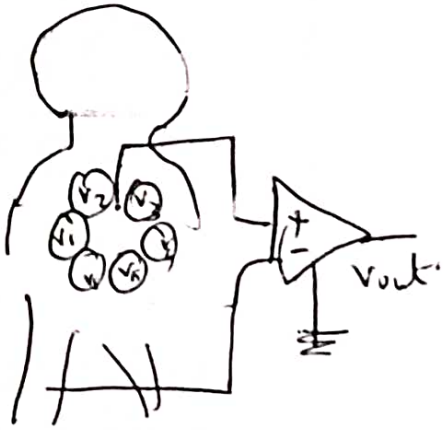


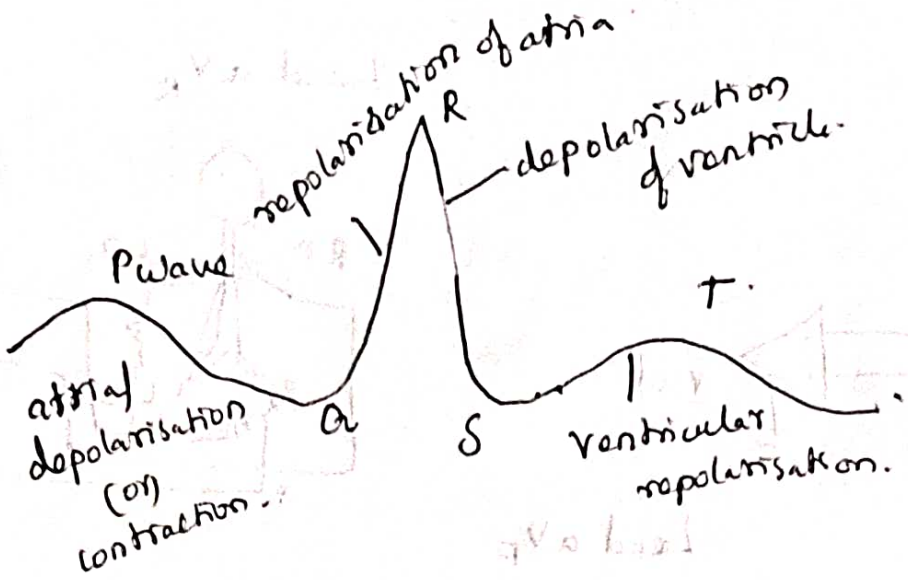
$$aV_F = V_{II} - \frac{V_I}{2}.$$

3) Chest Lead.



6 leads are connected to the chest.



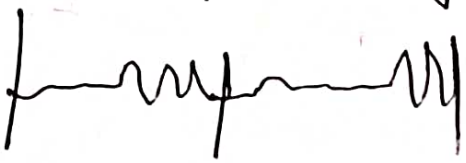


EEG electro encephalography

It deals with the recording & study of electric activity of the brain.

Waves.

Alpha  Freq - 8 - 13 Hz

Beta.  Freq - 13 - 30 Hz

theta  Freq - 4 - 8 Hz.

Delta  Freq - 0.5 - 4 Hz.

cement of electrode (10-20 electrode)

In EEG, electrodes are placed in standard position on the skull in an arrangement called 10-20 system.

→ draw a line on the skull from Nasion the root of the nose, to the inion on the occupital lobe.

C - Centripidal Lobe

O - occuipital Lobe

F - frontal Lobe.

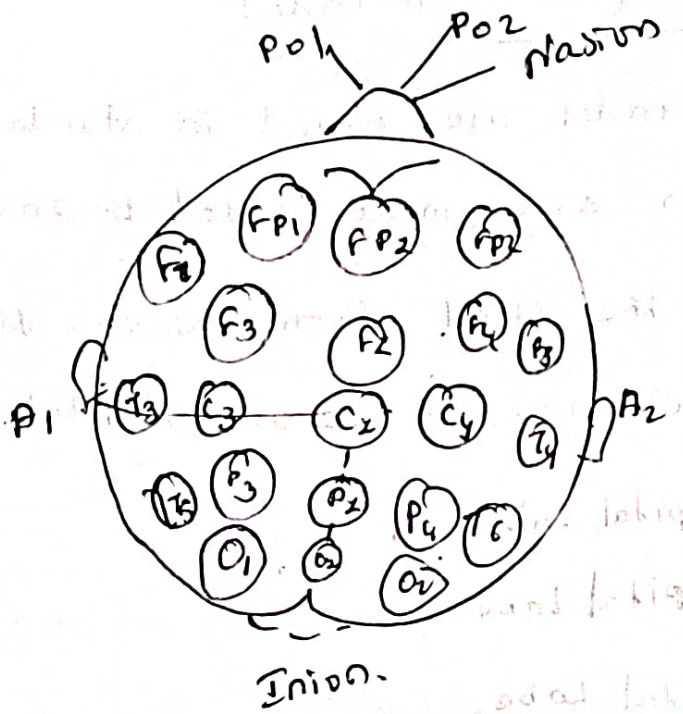
P - parental Lobe.

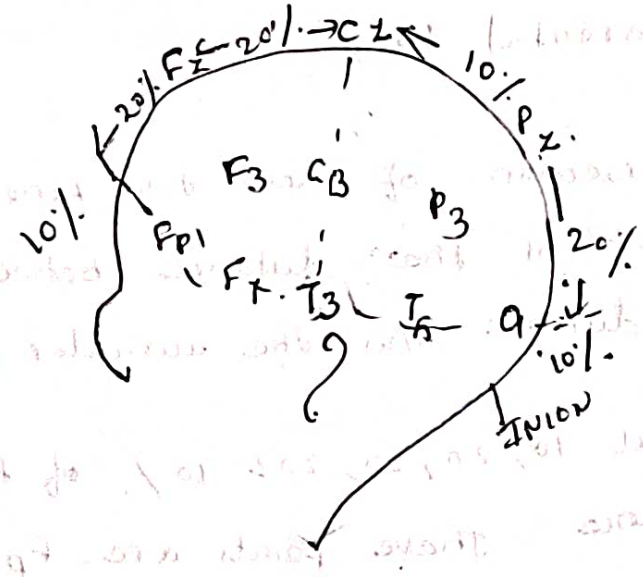
→ Mark the intersection of these two lines as C_z which is the midpoint of the distance between the Nasion & inion (or) distance b/w the auricular points.

→ Mark point at 10, 20, 20, 20 & 10 % of the total Nasion-ionion distance. These points are Fp_z, F_z, C_z, P_z & O_z.

→ Mark points at 10, 20, 20, 20, 20 & 10% of the total distance b/w the preauricular points.

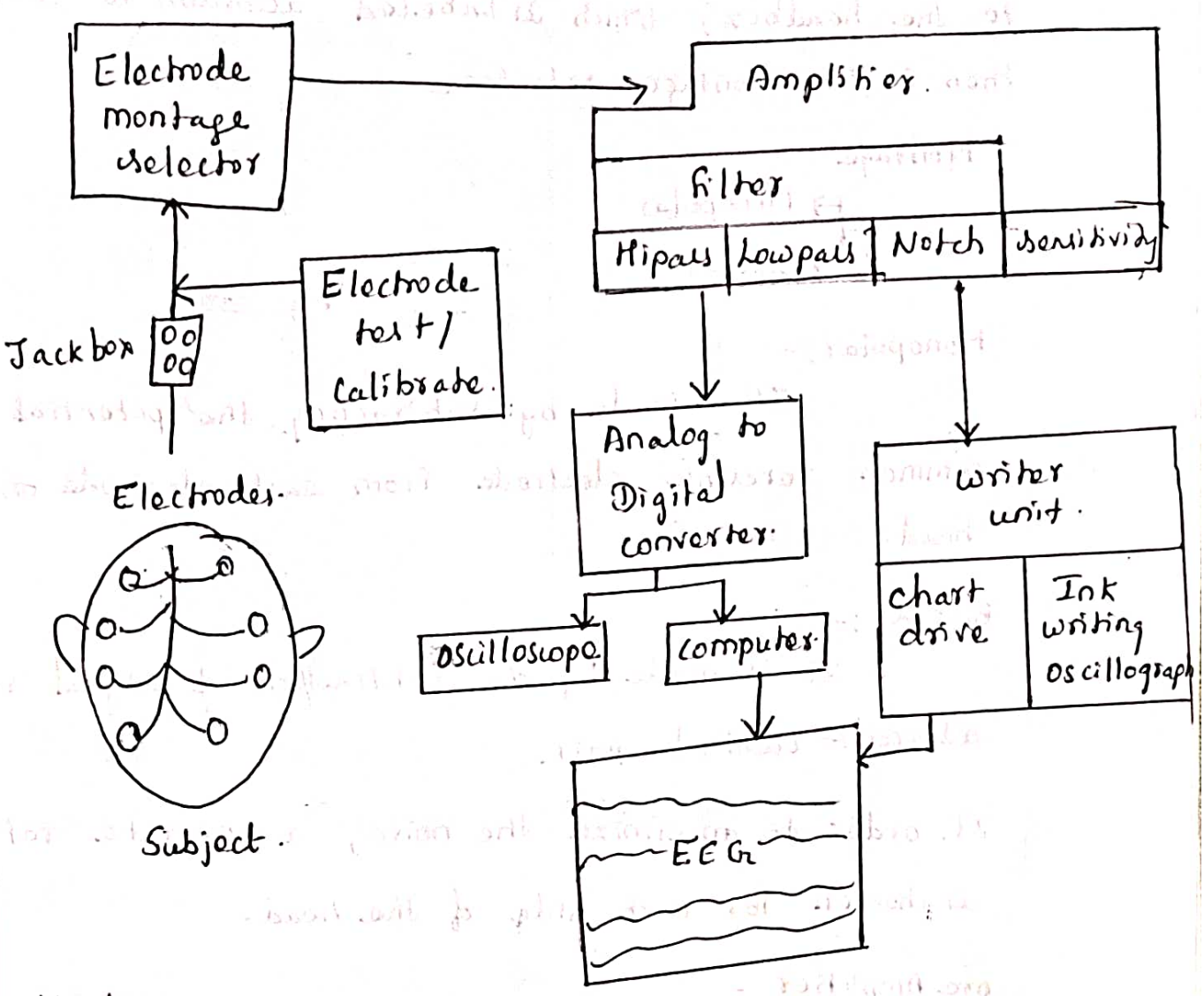
→ These points are T_3 C_3, C_z & C_4 & T_4. In these odd numbered point T_3 & C_3 are on the left & even No. of points C_4 & T_4 on the right.





EEG electrode position.

EEG Machine:



Montage:-

A pattern of electrode on the head and the channels they are connected to it is called montage.

→ Montage is always symmetrical.

Reference electrode → placed on Forehead.

EEG electrodes are arranged on the scalp. according to a standard known as the 10/20 system.

Electrode montage selector:-

EEG signals are transmitted from the elec to the headbox, which is labeled according to 10-20 syste then to the montage selector.

Montage
- Monopolar
- Bipolar.

Monopolar:-

It is made by subtracting the potential of a common reference electrode from each electrode on the head.

Bipolar:-

It is made by the subtraction of signal from adjacent electrode pair.

In order to minimize the noise, a separate reference is choosen for each side of the head.

pre Amplifier:-

It is used in EEG because it have high gain & low noise characteristics. Amplifier must have very high common mode rejection to minimize the stray interference signal from power lines & other electrical equipment.

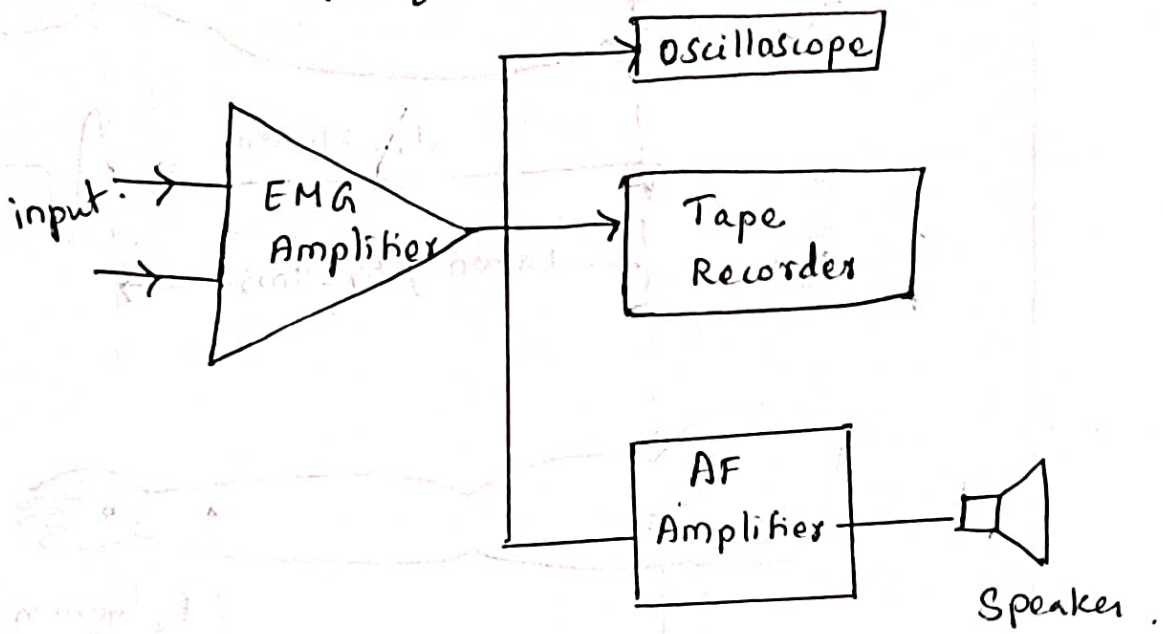
Filter:-

upper cutoff frequency controlled by the high frequency filter.

Notch filter → eliminate the interference.

EMG: Electromyography.

It is the Science of recording and interpreting the electrical activity of muscle's action potential.



In Bipolar electrode, the potential difference between two surface electrode resting on the skin is measured.

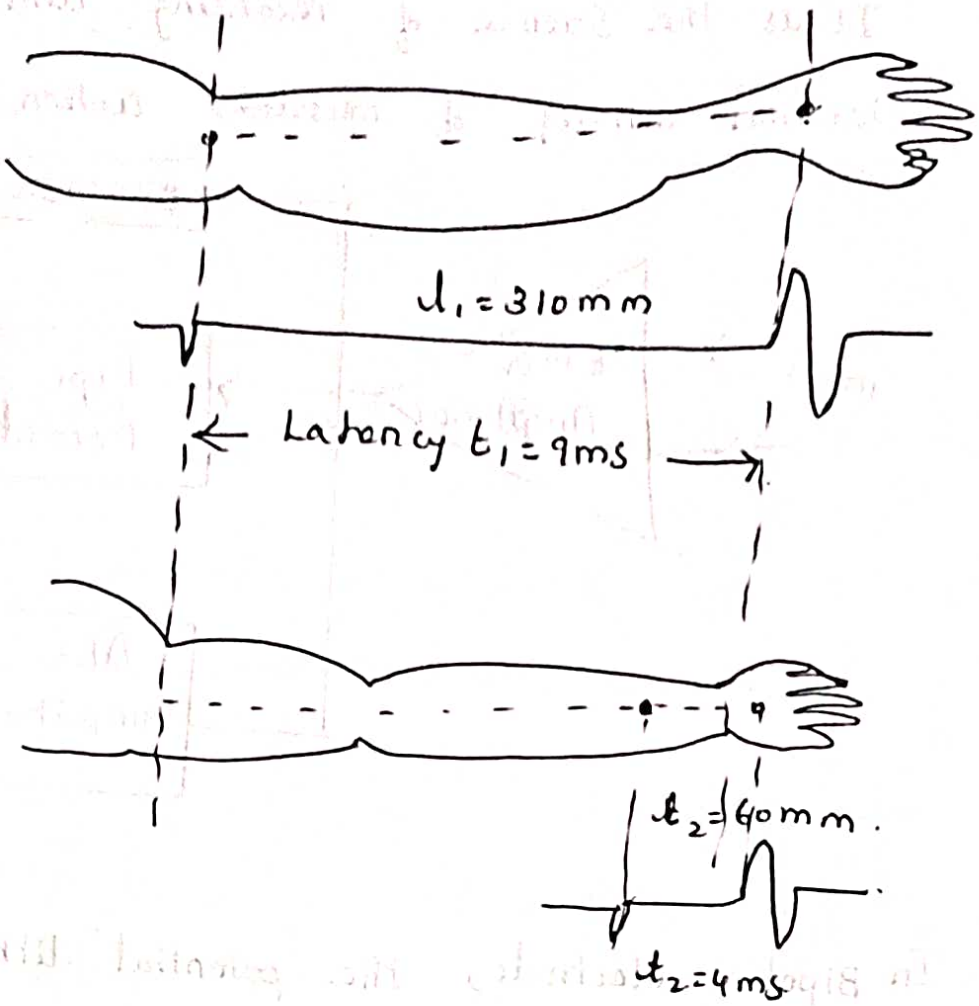
In unipolar electrode, the reference electrode is placed on the skin.

Frequency → 10 Hz - 1kHz.

CMRR → greater than 10 MHz.

Latency:

Elapsed time between the Stimulating impulse & the muscle's action potential.



Conduction velocity $v = \frac{d_1 - d_2}{t_1 - t_2}$.

Conduction velocity $= 50 m/s$

UNIT - III

SIGNAL CONDITIONING CIRCUITS:

Need for Bio Amplifier :-

→ It must have high input impedance.
Bio Amplifier potential with input impedance is 2MΩ.
other Applications = 10MΩ.

→ It must have isolation & protection circuit.

→ To protect the patient from micro shock & macro shock.

→ Voltage gain of bio Amplifier should be more than 100db.

→ constant gain should be maintained.

→ output impedance of Bio Amplifier is small.

→ Drift free Amplifier.

→ CMRR of Bio Amplifier should be more than 80db.

→ gain of the Amplifier must be correctly calibrated.

Bio Amplifier:

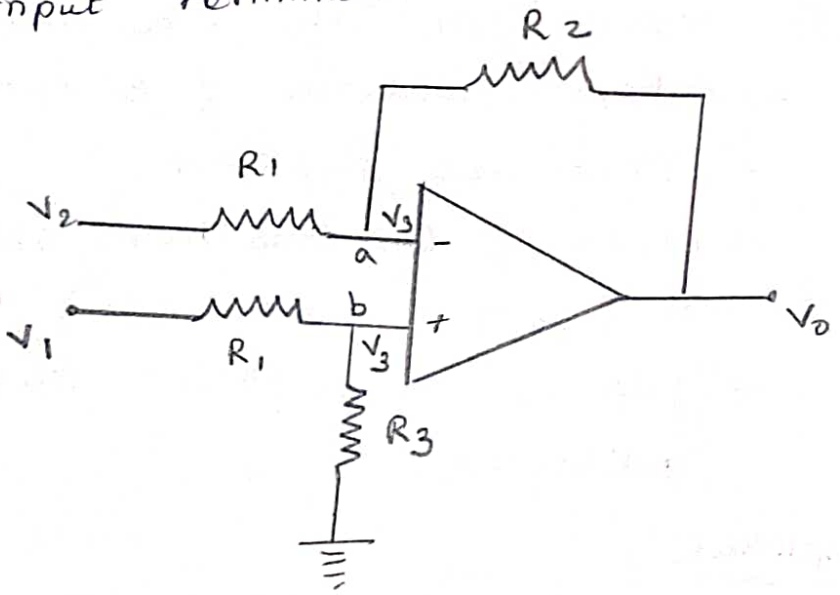
Bio signals are having low amplitude & Low frequency. so Amplifiers are needed to boost the amplitude level of the bio signals. the output of this amplifier is displayed as EEG (or) ECG waveform. These Amplifier are known as Bio Amplifiers (or) Bio-Medical Amplifier.

Types of Amplifiers:

- → Differential Amplifier
- → Operational Amplifier
- → Instrumentation Amplifier
- → chopper Amplifier
- → Isolation Amplifier.

Differential Bio Amplifier:-

A differential Amplifier produces an output voltage that is proportional to the difference between the voltage applied to the two input terminals.



There are three mode of operations.

- → single ended mode
- → Differential mode
- → common mode.

* Single ended mode :-

When either V_1 or V_2 is equal to zero, the operation of the differential Amplifier is known as single ended mode of operation.

subtracting (4) from (3) we get

$$\frac{+}{R_1}(V_1 - V_2) = \frac{V_o}{R_2}$$

$$\boxed{V_o = \frac{R_2}{R_1}(V_1 - V_2)}$$

* **Common Mode:-**

The input voltages appearing at the input terminal 1 & 2 are identical both in Amplitud and phase at every instant of time. & the circuit is said to be operating in the common mode.

$$V_1 = V_2 = V_{CM}$$

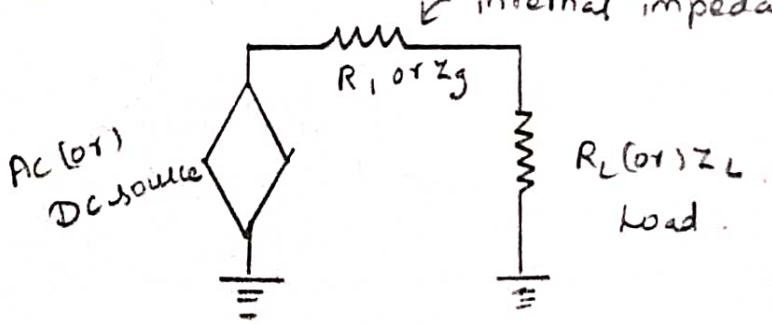
$$V_o = 0.$$

These input signals are called common mode signal. Thus the common mode input signals produce no voltage at the output of the ideal amplifier.

Impedance Matching circuits:

Impedance Matching is defined as the process of making one impedance Look like another.

Load impedance should match to the source (or) internal impedance of a driving source.



When, $V_1 = 0$, Differential Amplifier is in the ~~non~~ inverting mode.

$V_2 = 0$, It is operating in the non inverting mode.

* **Differential mode:-**

If two input signals are equal but have opposite polarity.

$$V_1 = -V_2 = V_o$$

$$V_o = \frac{R_2}{R_1}(V_2 - V_1)$$

$$V_o = \frac{2R_2}{R_1} V_o$$

The input signals are called differential mode signals.

At nodal equation at 'a' is

$$\frac{V_3 - V_2}{R_1} + \frac{V_3 - V_o}{R_2} = 0 \quad \rightarrow (1)$$

At 'b' is

$$\frac{V_3 - V_1}{R_1} + \frac{V_3}{R_2} = 0 \quad \rightarrow (2)$$

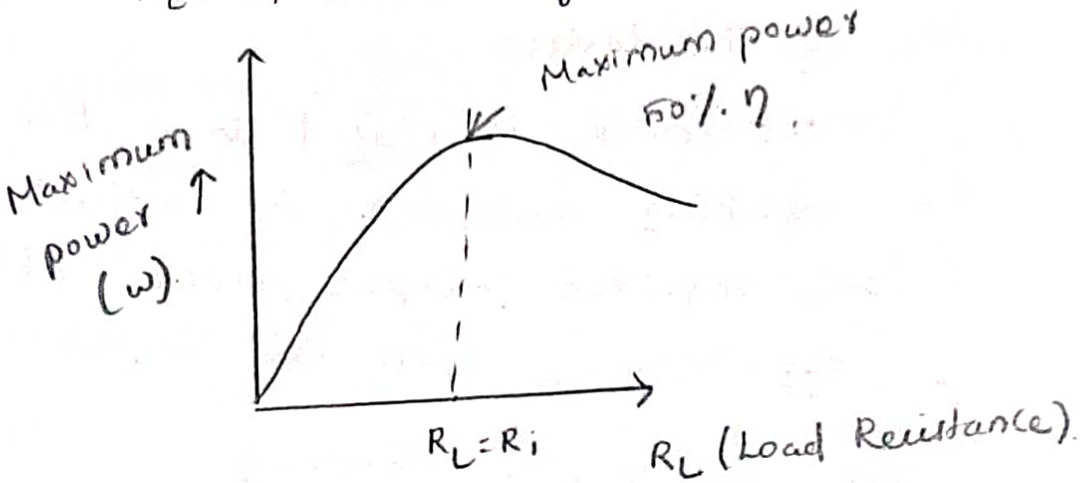
Rearranging (1) & (2)

$$\left(\frac{1}{R_1} + \frac{1}{R_2}\right)V_3 - \frac{V_2}{R_1} = \frac{V_o}{R_2} \quad \rightarrow (3)$$

$$\left(\frac{1}{R_1} + \frac{1}{R_2}\right)V_3 - \frac{V_1}{R_1} = 0 \quad \rightarrow (4)$$

The Maximum power transfer theorem says that to transfer the maximum amount of power from source to Load, the Load impedance should match the source impedance.

$$R_L = R_i \text{ (or) } Z_L = Z_g$$



Types of impedance Matching

→ Transformer Matching
→ LC Matching
→ Transmission Line Matching.

Transformer Matching:

RF transformer can be used to produce very wideband impedance Matching.

Limitation:

→ Frequency on transformer.
→ Restricted range of available impedance.

LC Matching:

→ Inductor & capacitor are used to obtain the required conjugate impedance Match.

Frequency → 30-300MHz.

→ It is used for power Amplifier.

→ good indication of the suitability of LC Matching for a transistor.

→ To determine the input & output impedance of the devices.

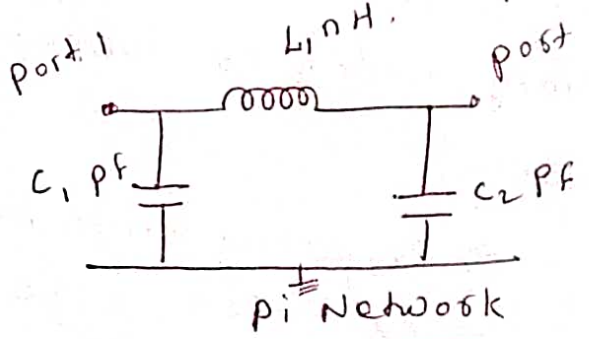
→ the device is placed in a test & the matching network is adjusted to obtain the required output power at a good efficiency & with low input return loss.

Transmission Line Matching:

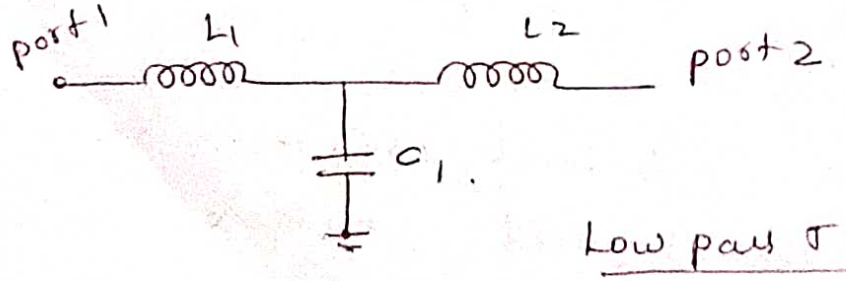
→ Length
→ Characteristic impedance should be Match.

Freq = above 150MHz.

→ π Network
→ Low Pass T Network.



pi Network



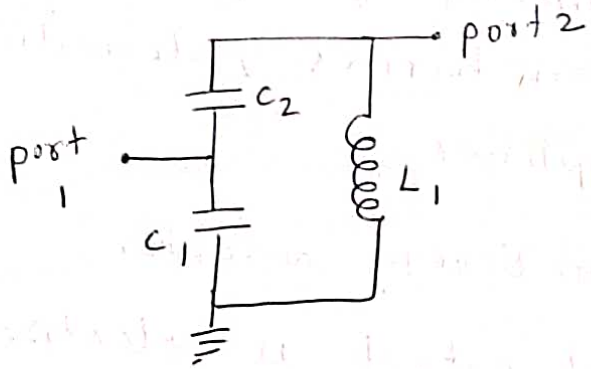
Low pass T Network

Capacitive impedance Matching

→ It is quite useful

→ very Reliable component in inter stage Matching Network.

The Network is used for Matching the output impedance of one transistor to the input Z of other transistor. This is related to Low pass pi Network except the ground & input port are changed.



For Non reactive device, the components values are same as pi Network.

port 1 → impedance is 50Ω = R_i - R_{device}

the value can be analysed by computer optimization

To Match the impedance, C_2 & L_1 are get adjusted.

Isolation Amplifier:

→ It is used in ECG for to prevent accidental internal cardiac shock.

→ It provide insulation between the patient connector & the ac power mains line cord.

→ to protect hospital patient, susceptible to electric shock.

→ 10^{12} Ω of Insulation.

→ It composed of input Amplifier, Modulator an. isolation barrier, demodulator & an output Amplifier.

→ It is an Energy converter.

→ Input & output is electrically isolated from each other.

→ Isolation barrier may be
- optical
- Magnetic transformer
- capacitive or even heat transfer.

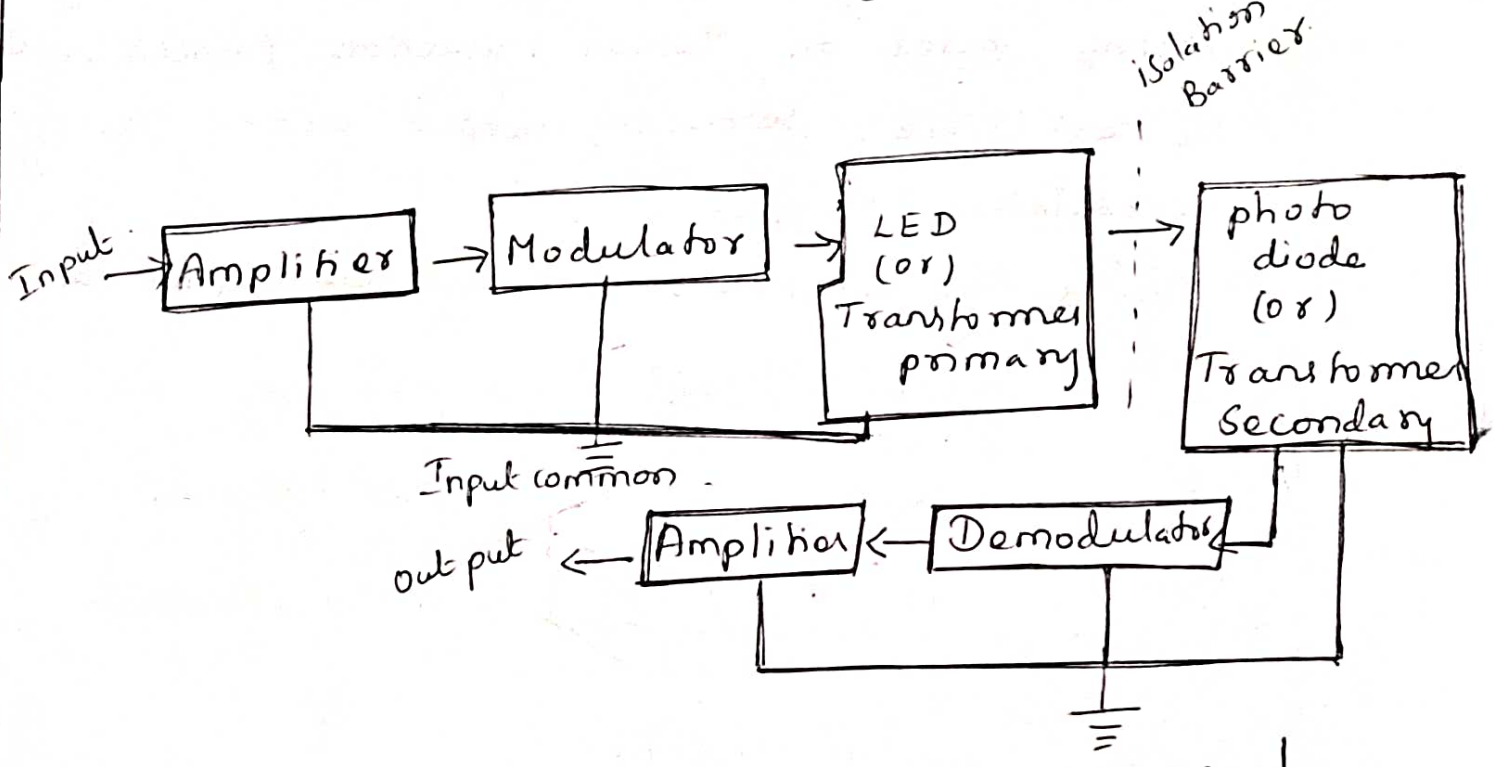
→ Isolation Amplifier → electrical energy on the modulation side is converted to some non electrically conductive energy in barrier & then converted back to electrical energy on the demodulator.

→ It operates on the principle of attenuation.

→ A high barrier impedance acts in series between input & output.

→ Ground connection to prevent noise in the circuit

→ Isolation Amplifier are classified as
- → Battery powered carrier
- → optically coupled.
- → current loading.



Isolation Amplifier are known as Pre Amplifier isolation circuit.

→ Isolation Amplifier increase the input impedance of a patient monitoring system.

→ It provide up to $10^{12}\Omega$ insulation between the patient & the power line in hospital.

→ It isolate the patient from device.

the electrical signal are obtained with elec

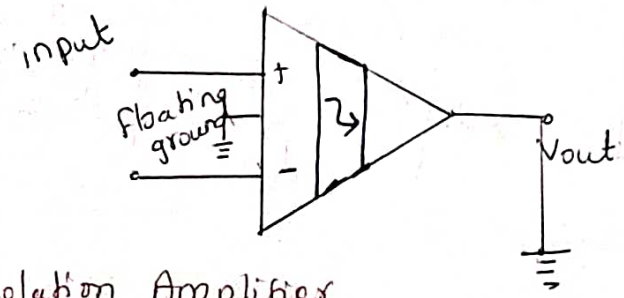
the signal received goes to the amplifier block (1)

then either it goes to the isolation barrier, optical cable or transformer can be used. If in case of an optical cable, Modulator output travels to LED. The LED converts electrical signal into light energy.

→ If transformer act as isolation barrier, the modulator output connect the primary winding of the transformer.

→ Energy from primary transfer to secondary winding based on Mutual induction principle. At the Next stage, secondary output enters the demodulation block.

→ Finally the amplified demodulated signal is obtained.



Types of Isolation Amplifier

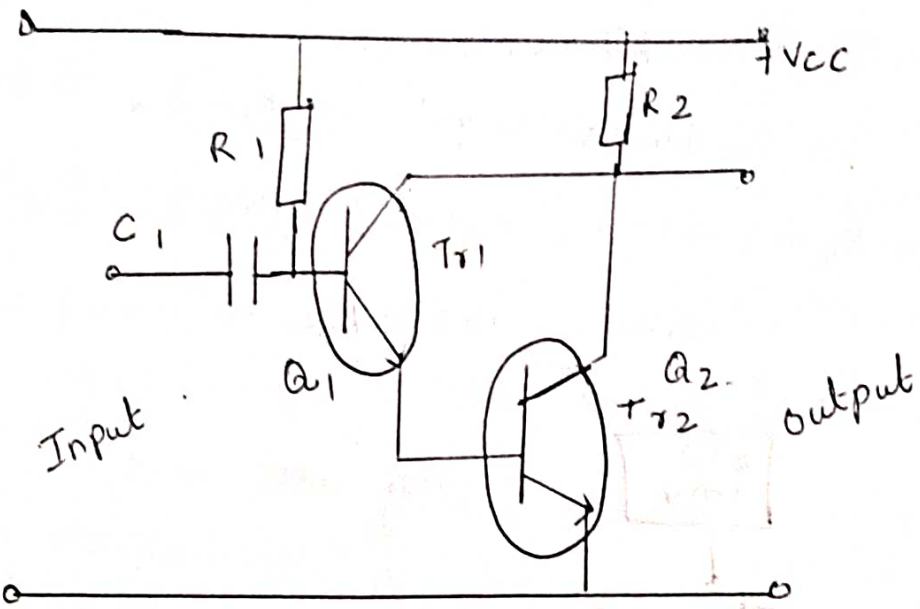
There are two types of Isolation Amplifier.

→ Darlington Pair

→ Boost tapping circuit

Darlington Pair

→ Darlington pair is an Isolation Amplifier which provides high input impedance with high current gain.



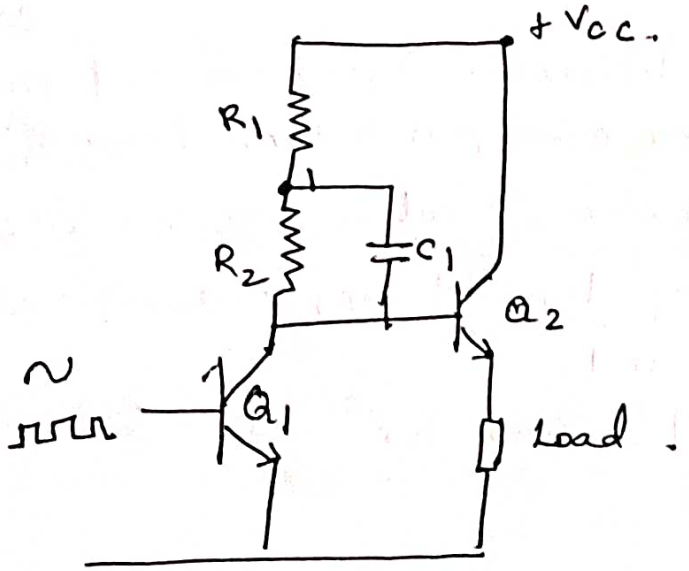
→ Two transistor are connected in common emitter component.

$$\frac{\text{input impedance}}{\text{output impedance}} = \text{common emitted current Amplification factor}$$

$$z_i = \beta^2 z_o$$

Q_1 emitter is connected directly to the base of the transistor Q_2.

(ii) Boosts Traping circuit.

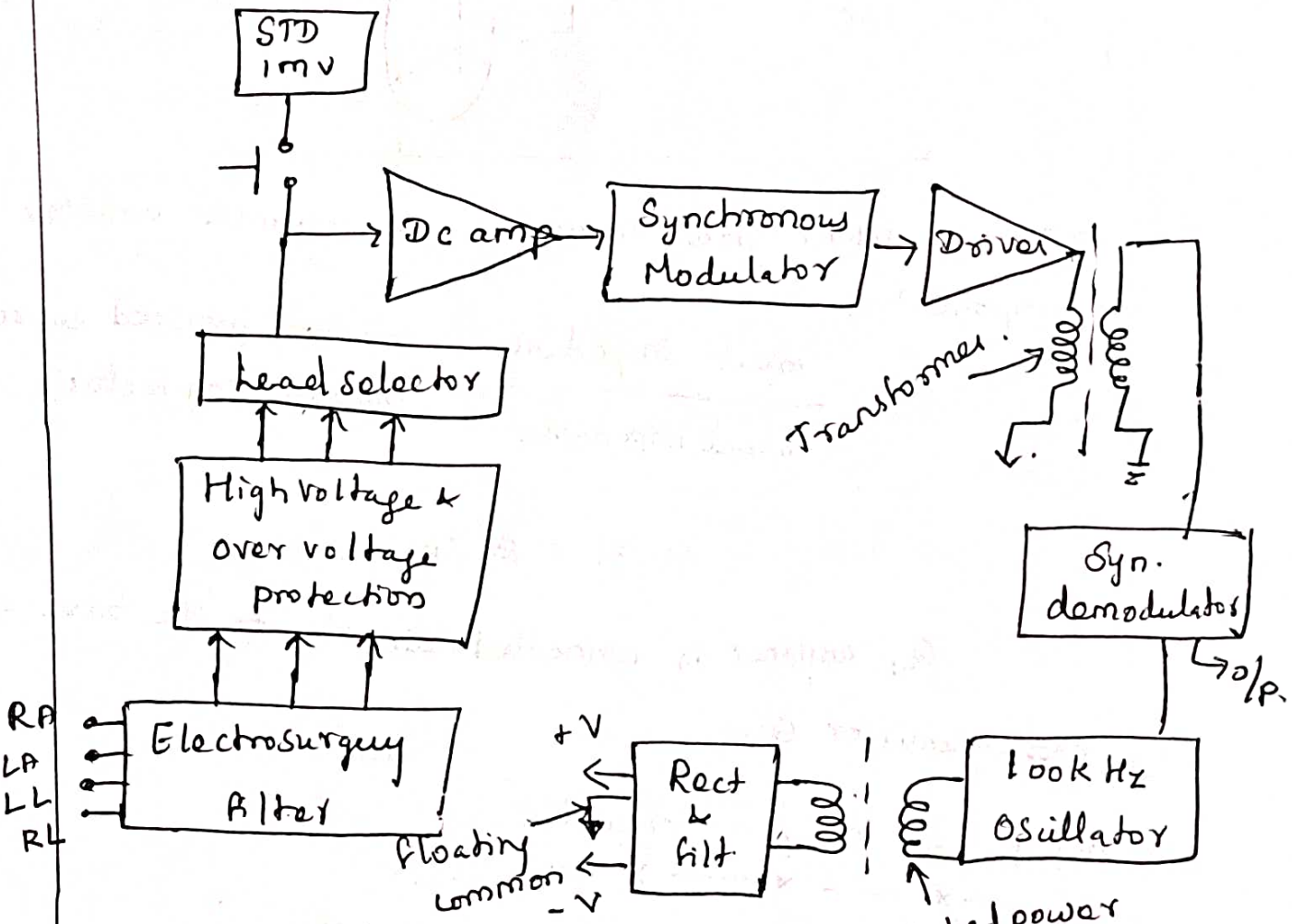


Boost Trapping circuit is also used as isolation amplifier.

feedback & value of R_1 & R_2 in series

R_4 → Limit the current flowing through Q_2.

Voltage gain = 0.9.



→ The difference signal obtained from RA, LA, LL, RL is given to Low pass filter. filter to reduce the interference. Cutoff freq → 10 kHz.

→ During defibrillator, high vtg & over vtg protection is used.

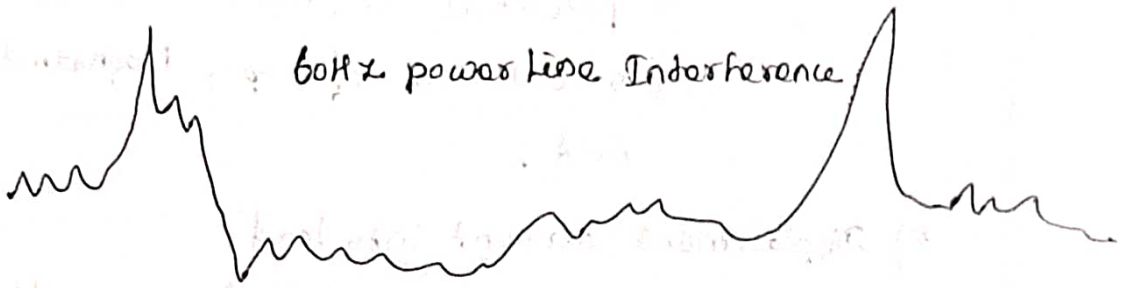
→ Lead selector for lead configuration, dc Level = 1mV.

Power Line Interference:

The power Line Interference of 50/60 Hz is the source of interference. It interrupt the recording of electrocardiogram (ECG).

The interference is caused by.

→ Electromagnetic interference by powerline
→ EMF by Machinery
→ Stray effect
→ Improper grounding.



Noise:

When frequency increases, its is high frequency, it affects the ECG signal & Mobile phone

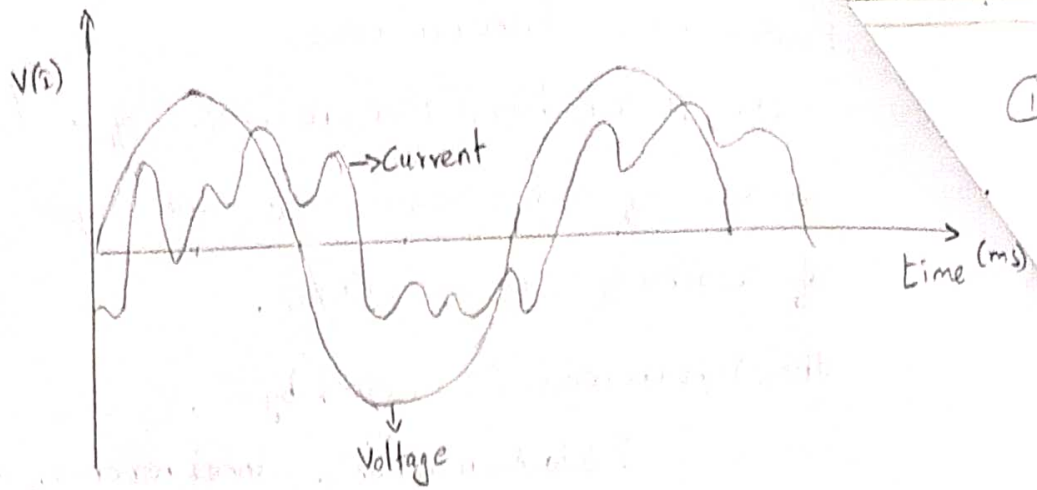
When frequency decreases, it is low frequency, it affects the ECG signal.

If there is no change in frequency → zero frequency.

Sources of the power Line Interfeence & Harmonics.

→ Non Linear Load connected to the grid.
→ Non sinusoidal.

According to IEEE 519-2014, the quantity having a frequency is the integral multiple of fundamental frequency.



Different ways:

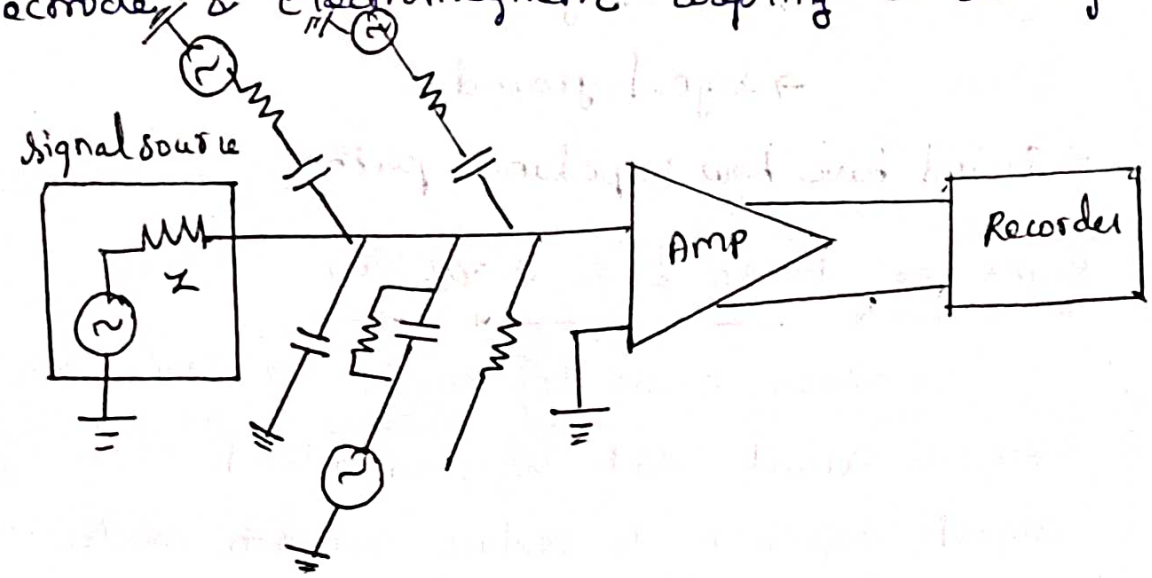
1) Magnetic Induction
   - → Cable is used
   - → Magnetic field is present.
   - → potential is proportional to the area, orientation of Loop, Magnitude of the field.

2) Displacement current into lead.
   - → Capacitive coupling between the power line and lead
   - → common mode Interference is converted into Differential mode interference.
   - → High CMRR (Common mode Rejection Ratio).

3) Displacement current into body.
   - → parasitic capacitance between the body & power line
   - → voltage drop is present.
   - → It will cause the Power Line Interference.

Sources of Noise in low level measurement.

i) Electrode & Electromagnetic coupling to ac signal.



Amplifier input signal = $E + IZ$

Where,

$E \rightarrow$ signal Amplitude

$Z \rightarrow$ impedance.

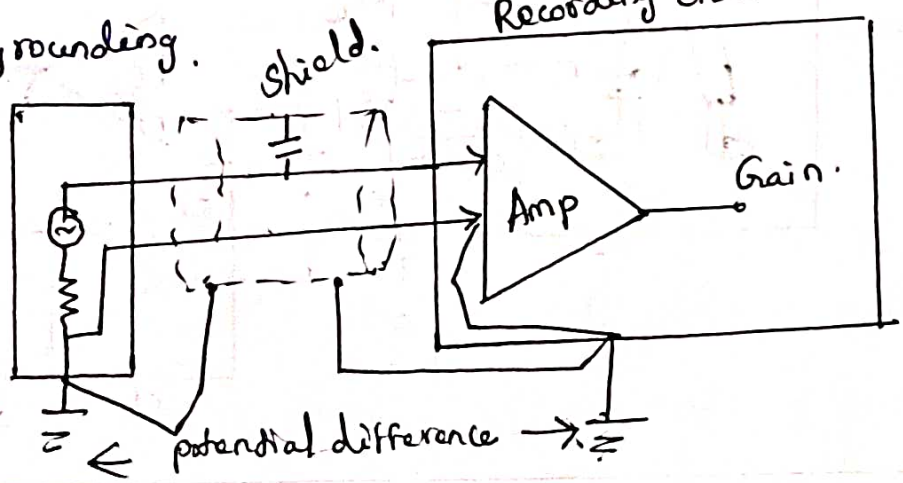
$I \rightarrow$ current generated by Noise.

→ Low level signals are sensitive to external contamination
→ Noise pick up & electromagnetic pick up. is produced.

It is avoided by.

→ Shielding
→ proper grounding
→ Shorter the twist, greater noise Rejection.

proper grounding.



There are two ground

→ Source ground (System)

→ Signal ground.

→ Ground have low impedance path.

Right Leg driven ECG Amplifier.

A Driven Right Leg circuit (or) DRL circuit is an electric circuit that is often added to biological signal Amplifier to reduce common mode interference.

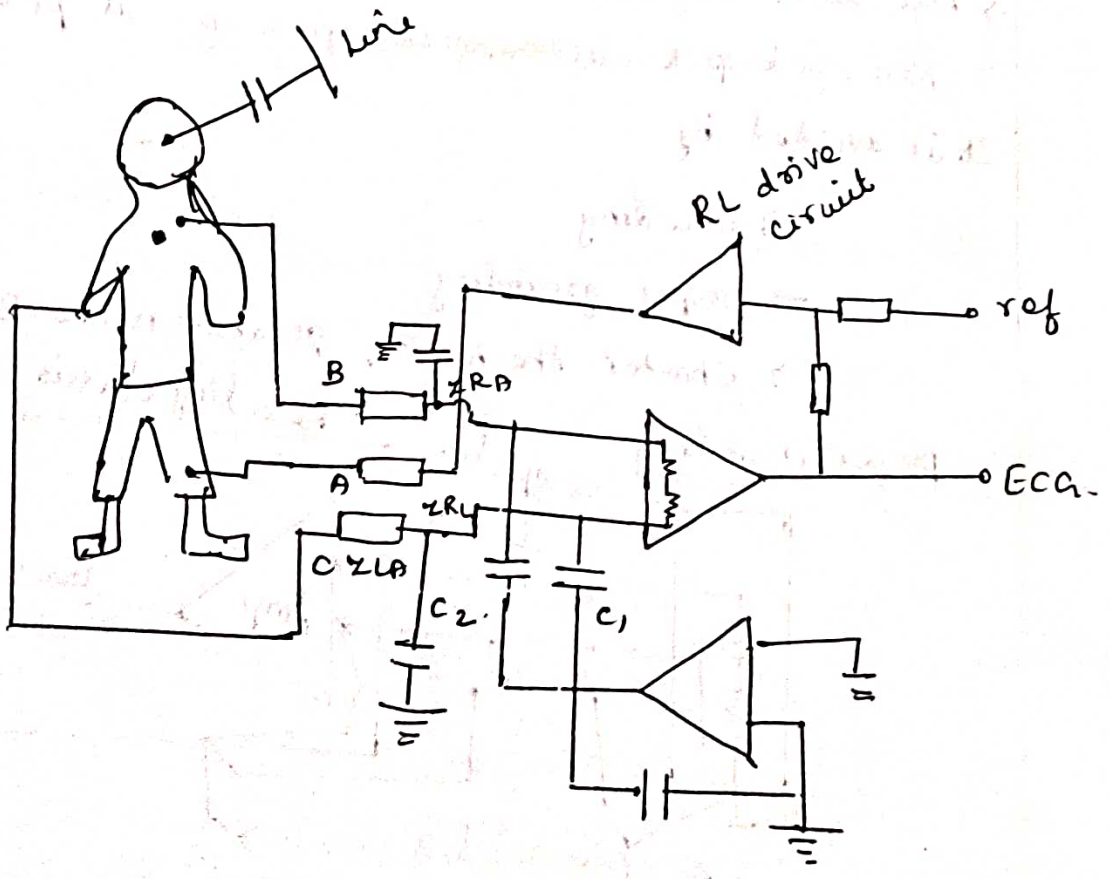
→ Right Leg driver circuitry is used to eliminate interference noise by actively cancelling the interference. Other methods of noise control include.

→ Faraday cage

→ Twisting wires

→ High Gain Instrumentation Amplifier

→ filtering.



Isolation of the patient preamplifier can also be obtained using an optical isolator. The high common-mode rejection of the amplifier is obtained by proper shielding.

→ The effective capacitance from the input leaded to the earth is made negligible.

→ Preamplifier circuitry should be preferably be shielded in a separate case.

→ To minimize the common mode signal between the body of the patient & the floating ground a right leg drive circuit is used.

→ The common mode signal after amplification in a preamplifier are inverted & fedback to the right leg electrode, reducing the common mode voltage on the input w.r.t. floating ground.

→ The presence of stray capacitance at the input of the preamplifier causes common mode current to flow in LA & RA resulting in a voltage drop at the electrode resistor.

Imbalance of stray capacitance (or) electrode resistor causes a difference signal.

For mordern ECG machines,

completely shielded patient cable & lead wire & their high common-mode voltage rejected.

Interference can be avoided by filter.

Amplifier patient safety current = 1.2 mA.

Vtg. = 115 Vac 60 Hz.

offset noise = 5 mV pp.

Optimized signal frequency = 0.05 to 100 Hz.

Band pass filter:

Filter is a device or process that removes some unwanted components or features from a signal.

Types of filter:-

→ Low pass filter.

→ High pass filter

→ Band Pass filter

Band Pass filter:

A band pass filter is an electronic device (or) circuit that allows signal between two specific frequency to pass. but that discriminates against signal at other frequencies.

Band pass filter

→ Active band pass filter

→ passive band pass filter.

Active band pass filter:

Band pass filter require an external source of power & employ active component such as transistor & integrated circuit. It is known as Active band pass filter.

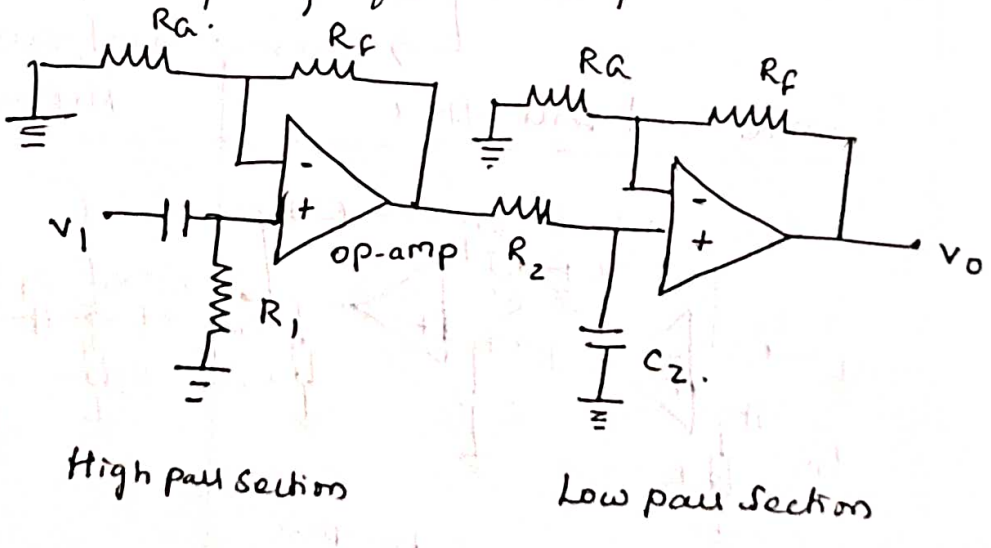
passive band pass filter:

Band pass filter use no external source of power & consist only of passive component such as capacitor & inductor. these are called passive band pass filter.

→ A Band Pass filter is the combination of a High pass filter & Low pass filter.

→ It allows only a select range of frequency to pass through.

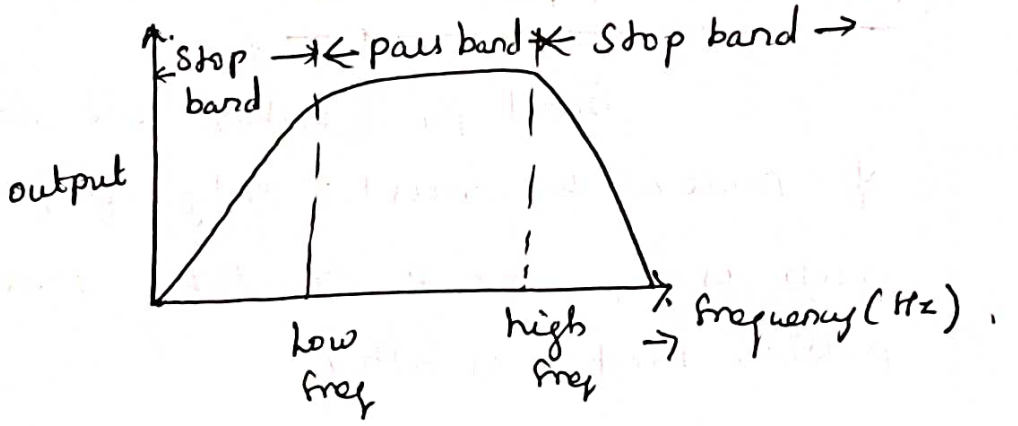
→ cut off frequency of the LPF is higher than the cut off frequency of the HPF.



filter bandwidth $= f_2 - f_1$

the range of frequency b/w f_1 & f_2 is called filter pass band.

Band pass filter → used in wireless transmitter & receiver.



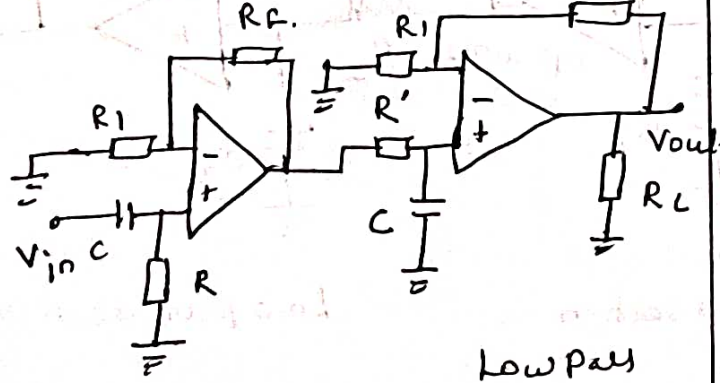
$$\text{Resonant frequency} = \sqrt{(f\text{-low} * f_{high})}$$

The distance between the Lower cut-off frequency & higher cut off frequency is called as Bandwidth.

Types of Band Pass filter

- wide band Pass filter
- Narrow Band Pass filter.

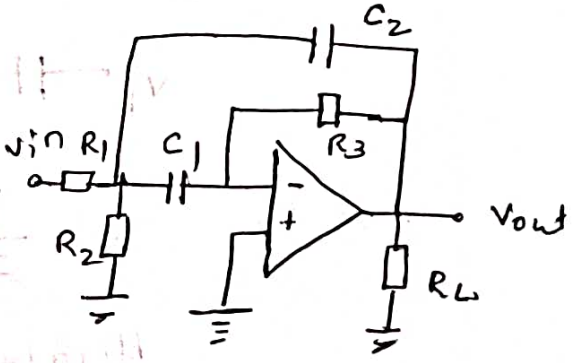
wide Band pass filter



HP section

Low Pass section.

Narrow Band Pass filter



→ Isolation of the patient circuit is obtained using low capacitance transformer whose primary winding is driven from 100 kHz oscillator.

→ Transformer secondary is used to obtain an isolated power supply of 6V for operating device.

→ Syn. Modulator is modulated at a frequency of 100KHz. (modulates the ECG signal.

→ Syn. demodulator is chosen to give a low noise performance switching FET.

→ Isolation of the patient preamplifier is obtained using an optical isolator.

→ High common mode rejection of the amplifier is obtained by proper shielding.

→ the output of the demodulator is used at the input of the power amplifier.

→ perfect shielding for isolated preamplifier circuit.

→ Line frequency interference is eliminated.

→ CMRR of 115 dB.

→ $Z = 10^{11}$ ohm.

Unit - IV

Measurement of Non electrical Parameter.

Temperature:

Temperature is one of the indicator. Two type of temperature measurement.

→ Systemic Temperature → Temperature of internal region of body

→ Surface Temperature → Temperature of external region.

Normal mouth temperature - 37°C.

Underarm temperature = 1°C Lower than above temperature.

Types of Temperature Measurement:

There are 3 type of temperature Measurement

→ Thermometer
→ Thermocouple
→ Thermistor.

Thermometer:

Thermometer instrument for measuring the temperature of the system.

Thermometer has two important element.

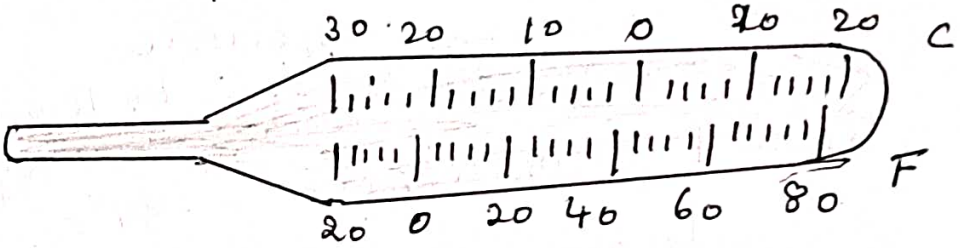
→ A temperature sensor in which some change occurs with change in temperature.

→ Some means of converting this change into a numerical value.

Working:

→ A thermometer has a glass tube sealed at both ends & it partly filled with a liquid like mercury or alcohol.

→ As the temperature around the thermometer's bulb heats up the liquid arises in the glass tube. When it is hot. the liquid inside the thermometer will expand & rise in the tube.

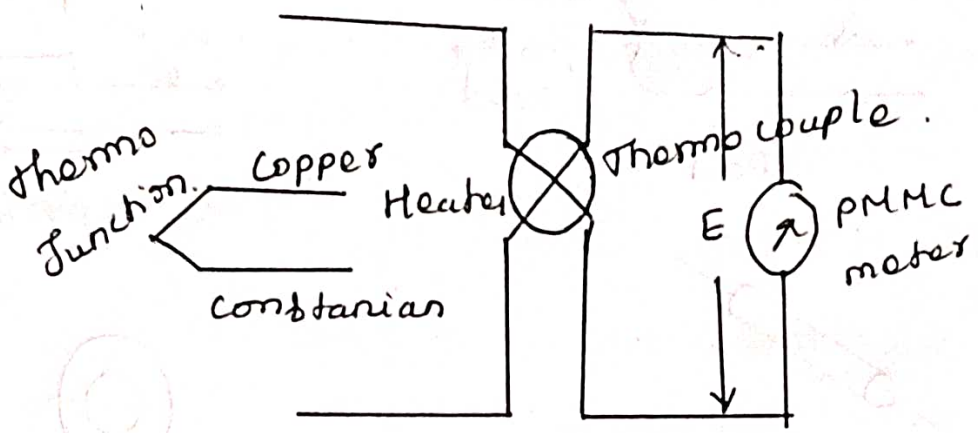


Thermocouple:

When two metal having different work function are placed together than heated by using a heater, a voltage is generated at the junction which is proportional to the temperature. This junction is called thermo couple.

principle:

Heat energy is converted into electrical energy. The heat at the junction is produced by the electrical current flowing in the heater element while the thermocouple produces an emf at its output terminal which can be measured with the help of PMMC instrument. The scale of PMMC instrument can be calibrated to read the current passing through the heater.



Construction & working:

→ A thermo couple can be formed by joining the two dissimilar metals such as platinum - Thodium, Chromel - Alumel, copper - constantian & ion.

→ The end forming a two junctions called thermo Junction there are two method

One method → to weld the wires together.

this produces brittle joint.

→ Another method of forming thermo Junction is by soldering the wire together.

→ drawback of introducing 3rd dissimilar metal.

the metal used for thermocouples are divided into two classes namely base metal & rare metals.

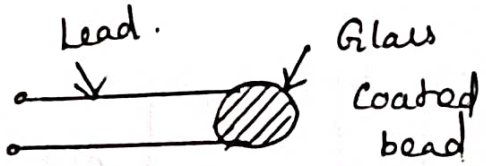
Thermistor: (thermal resistor).

→ It is composed of semi conductor material.

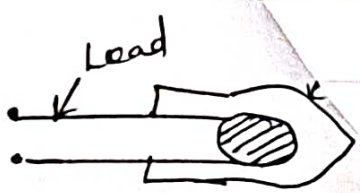
→ thermistor have negative temp. coefficient, ie) R ↓ T ↑.

→ To detect very small change in temp. which couldn't affect in RTD or a thermocouple.

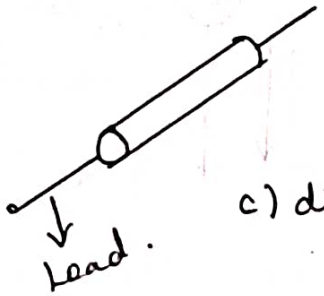
→ high sensitivity.



a) Bead

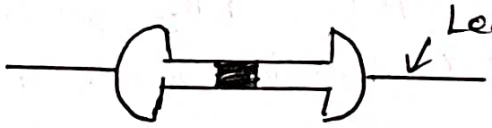


b) probe.



c) disc

d) washer.



e) Rod -

→ Thermistor are manufactured from the oxides of metals like manganese, nickel, cobalt, copper, iron, zinc, aluminium, titanium, Magnesium & uranium.

→ These oxides of their sulphides & silicates and milled mixed in appropriate proportion, pressed into desired shape with binders & finally sintered.

→ the electrical terminals are embedded before sintering (or) backed after wards.

→ Thermistor may be in the form of beads, rod, disc

→ Beads can be smallest size of 0.1mm to 1.25mm.

→ Fast response.

→ Sealed in the tip of a solid glass, ceramic (or) metal sheath to form a probe.

→ Glass probe - diameter 2.5 mm
length - 6 mm - 50 mm.

→ Thermistor disc → 2.5 mm - 25 mm → diameter
0.5 - 12.5 mm → thickness.

→ Rod type → 1.3 mm - 4.4 mm → diameter.
6.4 mm - 51 mm → Length.

washer → diameter - 19mm.

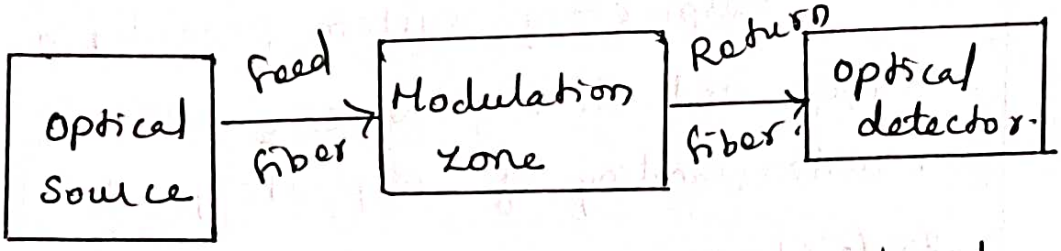
→ placed in series or parallel for increased power dissipation.

Glass probes:

→ temperature measurement where as discs & rods are employed more as temperature.

→ high power dissipation is a primary requirement.

Fibre optic temperature sensor:



→ fiber optic communication can be employed easily for monitoring & telemetry in industry environment.

→ they are also provide safety in the transmission to & form the transducer.

→ fiber sensor is that the light beam can be modulated either directly (or) indirectly without any interfacing of electrical system.

→ this modulation of light beam is done in the modulation zone of the generalized optical fiber sensor system.

→ optical fiber sensors are classified as intrinsic & extrinsic device.

The intrinsic device are also called as incoh. sensing system.

→ The intrinsic device the physical parametric to be sensed act on the fiber itself causing changes in transmission characteristics.

Advantage:

→ compatible
→ useful in HV equipment & explosive environment.
→ free from EM noise of any type.

Respiration Rate Measurement:

Respiratory system provides a means of acquiring oxygen and eliminating CO_2 various Laws are involved. in understanding of respiratory functions.

Boyle's Law: At const temp, Volume of gas $\propto \frac{1}{\text{pressure}}$

$$\frac{V_2}{V_1} = \frac{P_1}{P_2}.$$

Where,
V_i - initial volume
V_2 - final volume
P_1 - Original pressure.
P_2 - final pressure.

Charle's Law: At const pressure, Volume of gas $\propto$ temp.

$$\frac{V_2}{V_1} = \frac{T_2}{T_1}$$

$V_1, V_2 \rightarrow$ Initial, final volume.
$T_1, T_2 \rightarrow$ Initial, final temperature

Henry's Law:

At const temperature, quantity of gas $\propto$ partial pressure of that gas.

Dalton's Law:

Total pressure is the sum of the partial pressure of various gases

$$P_T = P_1 + P_2 + \ldots P_n.$$

Types of Respiration Rate Measurement.

Function: to supply oxygen & to remove carbondioxide from the tissue.

Techniques:

→ Displacement Method
→ Thermistor "
→ Impedance pneumography.
→ Co_2 method.

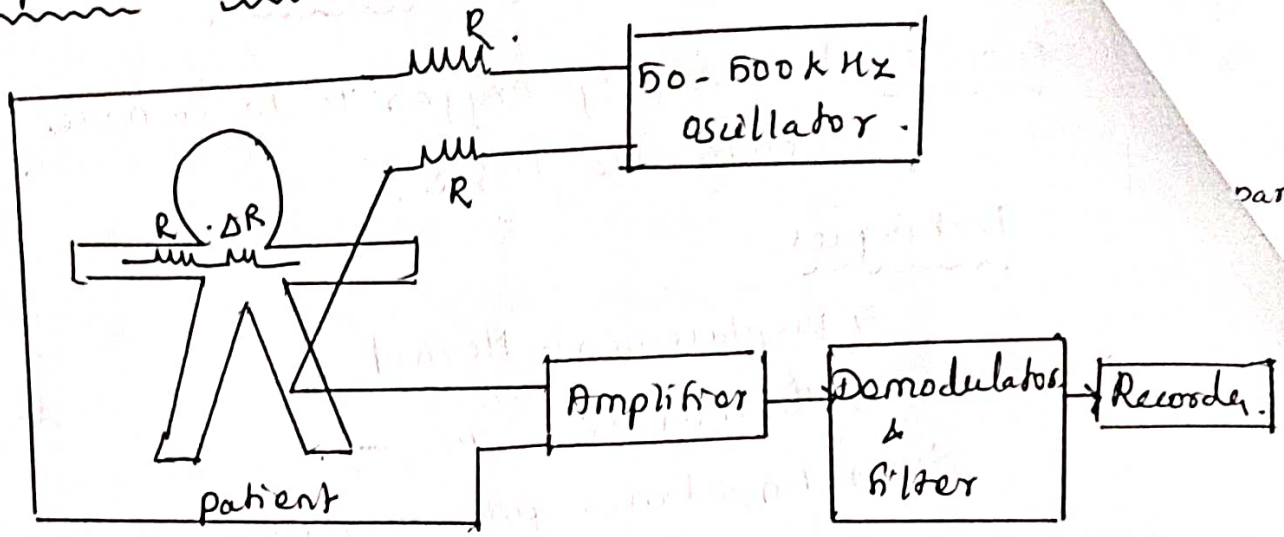
Displacement Method.

→ In this method, the transducer is held by an elastic band which goes around the chest.
→ The respiratory movements result in a corresponding resistance changes of the strain gauge.
→ It is connected as one arm of a Wheatstone bridge circuit. Its output varies with chest expansion
→ This output corresponds to the respiration activity.

Thermistor method.

→ There is a temperature difference between expired & inspired.
→ This temperature is sensed by placing thermistor in front of nostrils.
→ Thermistor is placed by using suitable stand.
→ Thermistor is connected with the bridge circuit so unbalance signal is amplified to get the respiratory activity.

Impedance pneumography:



→ indirect method.

→ It is based on the fact that the ac impedance across the chest of a patient changes as respiration occurs.

→ Ac signal is produced by oscillator circuit & is given to the chest of the patient through electrodes.

Voltage drop across the Resistance

$$V = I\,(R \pm \Delta R)$$

Where,

V → output voltage

I → current through the chest.

R → chest impedance

ΔR → change of chest impedance.

→ the output of the Amplifier is given to the demodulator & filter block.

→ Low pass filter is used to remove the residual carrier signal.

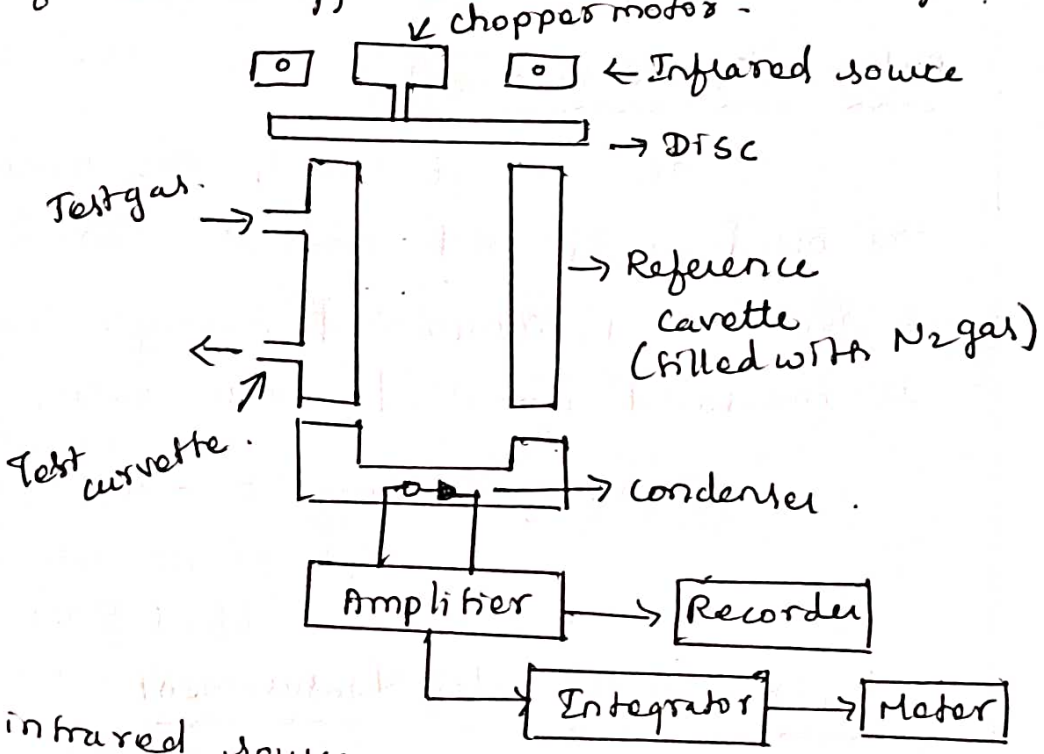
→ the output of the impedance pneumograph contain respirating rate data.

CO_2 Method:

→ Respiration rate can be measured by measuring CO_2 in expired air.

→ It is based on the absorption property of IR rays by certain gases.

→ When IR rays are passed through the expired air which contain certain amount of CO_2. some of the radiations are absorbed by it. so there is a loss of heat energy associated with the rays.



→ Two infrared sources are available. in this setup, the beam from one infrared source falls on the test cuvette side.

→ the beam from the infrared source falls on the reference cuvette. the detector is filled with a sample of pure CO_2. The detector has two identical portion. these portions are separated by a thin flexible metal diaphragm. Because of the absorption of CO_2 in the test cuvette, the beam falling on the test side of the detector

is weaker than the reference side. The gas in reference side is heated more than on the test so diaphragm is pushed slightly to the test side of the detector. the diaphragm forms one plate of a capacitor. the ac signal appears across the detector is amplified & recorded using recorder. the amplified output is integrated & meter display the reading. continuous monitoring of the respiration rate is used.

pulse rate Measurement:

The rate at which the heart muscle contra the blood is ejected from the ventricles & a pulse of pressure is transmitted through circulatory system is measured is called pulse rate.

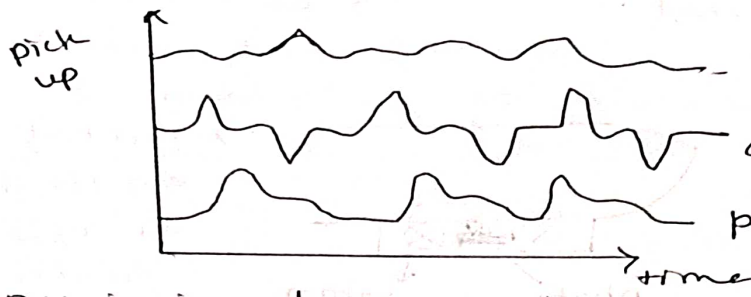
pulse wave transmit - 5 - 15 m/s.
velocity - 10 - 15 times faster than blood flow.

Types of pulse rate Measurement.

(1) Electrical impedance changes
(2) Strain gauge (or) microphone (Mechanical)
(3) optical changes (change in density).

(1) Electrical impedance Method:

→ It is used to measure the impedance change between two electrode caused by the change in blood volume between them.



Change in impedance = 0.1Ω

→ The impedance is measured by applying an AC between electrodes attached to the body

→ AC (10-100 kHz) used to prevent polarization of the electrode.

(ii) Mechanical Method :-

Mechanical method involves the use of a strain gauge connected to the rubber band placed on around the limb (or) Finger. expansion in the band due to change in blood volume causes a change in Resistance of the strain gauge.

other technique

→ A sensitive microphone is placed on the skin's surface to pick up the pulsation.

photo electric method :

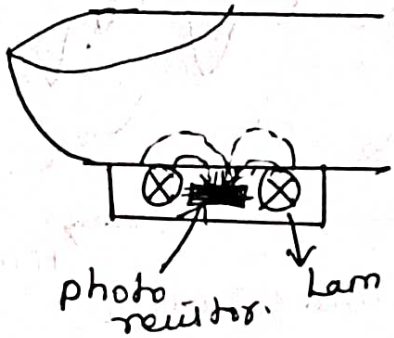
It is used to measure pulsatile blood volume change is by the photoelectric method Two methods are common.

→ Reflection method

→ Transmittance method.

Reflection method :

→ photo resistor is pla
adjacent to the exc
Lamp

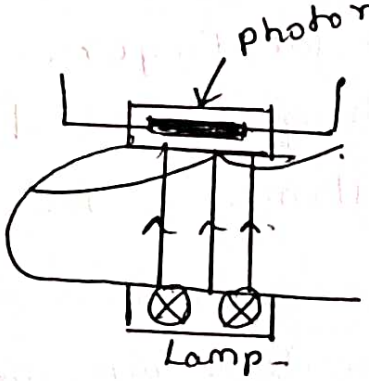


→ part of the light emitted by the LED is reflected & scattered from the skin and the tissue and falls on the photo resistor.

→ the quantity of light reflected is determined by the blood saturation of the capillaries.

→ voltage drop across the photo resistor, connected as a voltage divider will vary in proportion to the volume changes of the blood vessels.

Transmittance method :



→ Light emitting diode (LED) and photo resistor are mounted in an enclosure that fits over the tips of the patient finger.

→ Light is transmitted through the finger tip of the subject finger and the resistance of the photoresistor is determined by the amount of light reaching it.

→ with each contraction of the heart, blood is forced to the extremities & the amount of blood in the finger increases.

change optical density → Finger Light transmission reduces

↓

Resistance increases

→ photo resistor is connected as a part of a voltage divider circuit & produces a voltage that varies with the amount of the blood in finger increases

→ If alter the optical density with the result that the light transmission through the finger reduces & the resistance of the photo resistor increases.

→ photo resistor is connected as a part of a voltage divider circuit & produces a voltage that varies with the amount of blood in the finger.

→ This voltage ie) follows the pressure pulse & waveform is displayed on the oscilloscope (or) recorded on the Strip chart Recorder.

3) Optical changes:

LED photo transistor photo plethysmograph transducer consist of a Ga-As infrared Emitting diode.

→ compact package (6.25 × 4.5 × 4.75 mm).

peak spectral emission → 0.94 μm.

peak bandwidth → 0.04 μm.

phototransistor is sensitive to radiation between 0.4 & 1.1 μm

→ The signal from the photocell is amplified & filtered (0.5-5 Hz) & the time interval between two successive pulse is measured.

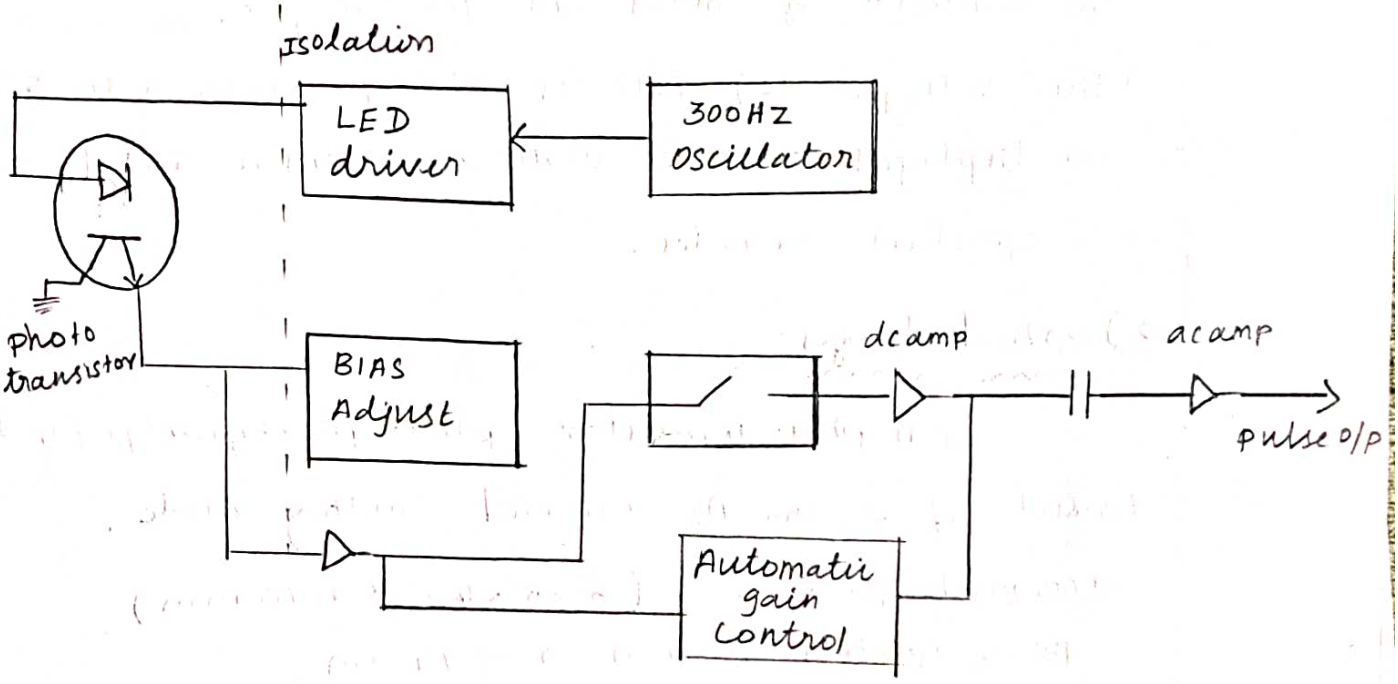
Measuring range :- 20-50 bpm.

The circuit consist of two parts, a LED oscillator & driver, which produce 300 Hz, 50 μs infrared light pulses to the finger probe attached to the patient & the phototransistor that pickup the

the attenuated light. the electrical signal obtained the photo transistor is amplified & its peak value is sampled & filtered. An automatic gain control circuit adjust the amplifier gain to yield a constant average pulse height at the output.

freq = 0.8 - 5 Hz.

→ This signal is transmitted across the isolation barrier, demodulated, Low pass filtered & transmitted to the analog multiplexer resident on the CPU board.



Blood Pressure Measurement:

The pressure measured during the cardiac cycle process is called Blood pressure. Measurement. the maximum pressure reached during cardiac ejection is called Systolic pressure. the minimum pressure occuring at the end of the ventricular relaxation is called diastolic pressure.

Types of Blood pressure Measurement:-

→ Direct Method

→ Indirect method

→ Auscultatory Method.

Indirect Method of BP Measurement.

Sphygmomanometer is used to measure blood pressure indirectly.

→ Sphygmomanometer consist of inflatable rubber bladder which is known as cuff, rubber squeeze ball pump & valve assembly.

→ pressure is measured using manometer with mercury column.

procedure:

→ The cuff is wrapped around the patient's upper arm at a point about midway between the elbow and shoulder

→ the stethoscope is placed over an artery distal to the cuff. The cuff is inflated so the pressure inside the inflated bladder is increased to a point greater than the anticipated systolic pressure.

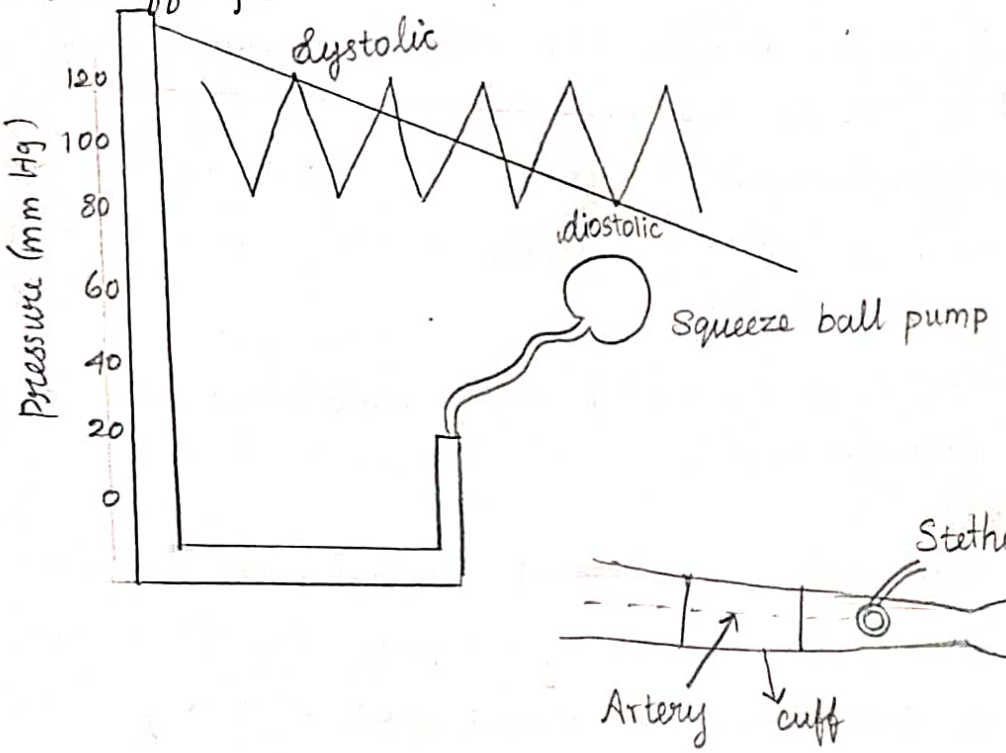
→ the pressure compresses the artery against the underlying bone. so blood flow is stopped in the vessel.

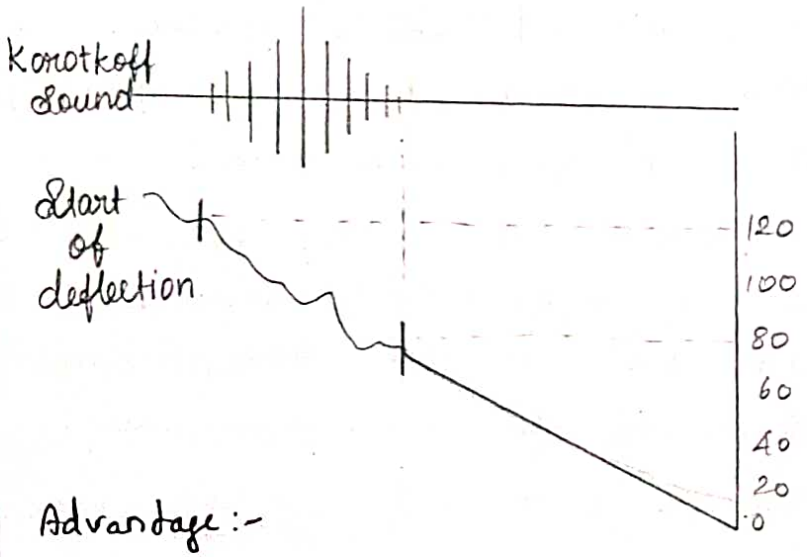
→ then the doctor slowly reduces the pressure in the cuff and he watches the mercury column, when the Systolic pressure exceed the cuff pressure, then the doctor can hear the some crashing, snapping sound through the stethoscope. This sound is known as korotkoff sound.

→ This sound is vanished when pressure drops below the diastolic pressure.

→ The pressure reading in the mercury column which korotkoff sound is disappeared is noted.
diastolic pressure — 80mm/Hg for normal person.

→ this sound is disappeared at some point. this is known as muffing.





Advantage :-

→ very simple

→ painless technique.

→ No Hazardous.

disadvantage :-

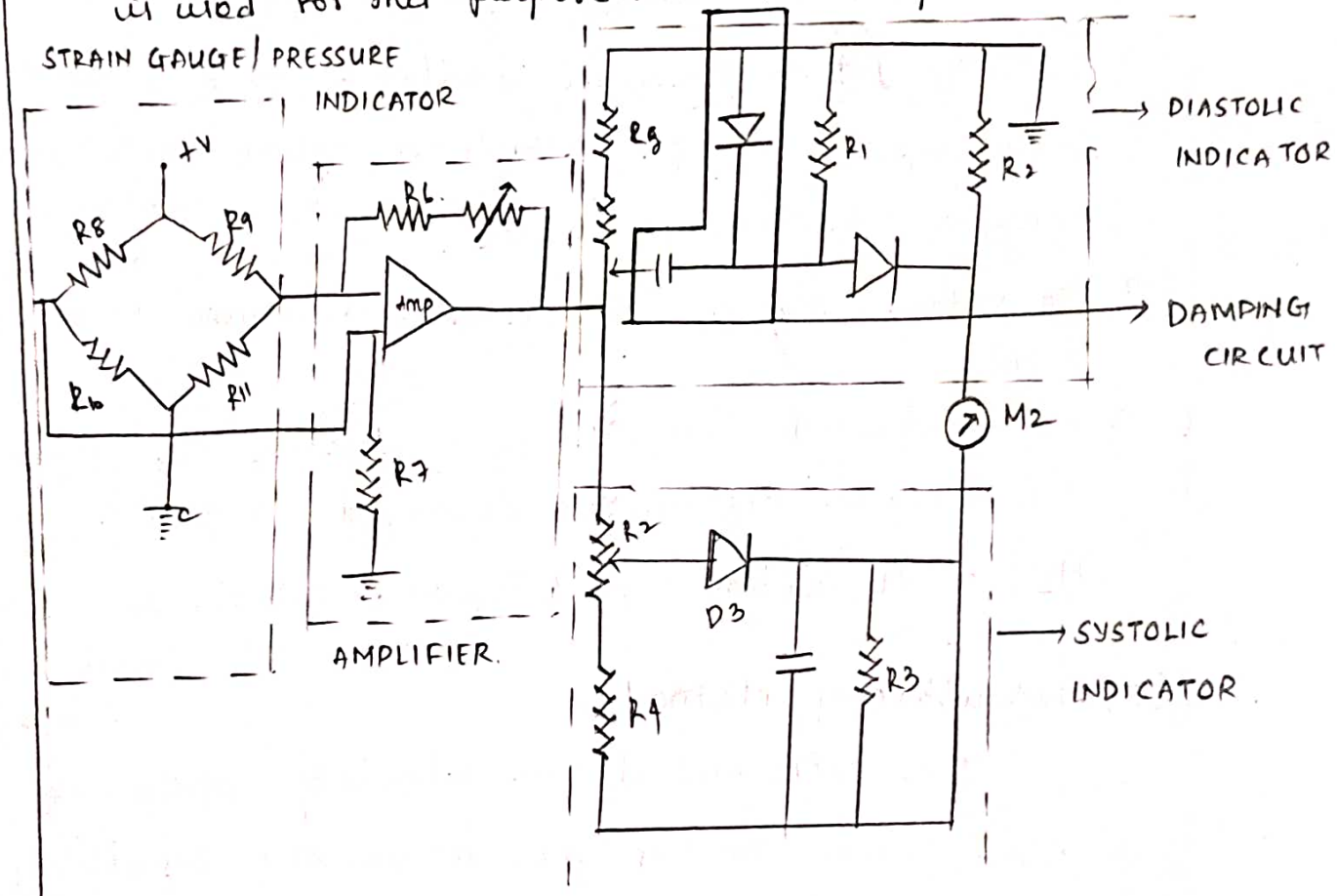
→ Accuracy is less depend upon the doctor.

Direct Method of BP Measurement

→ Accurate blood pressure reading, direct BP measurement is used.

→ Blood pressure in deep region, direct method is used.

→ Fluid filled catheter is used. Before inserting the catheter into the blood vessel, fluid filled system should be completely flushed out. Usually sterile saline is used for this purpose. Blood clotting is avoided.



Working :-

→ Blood is taken from the vessel using the catheter tip probe. pressure exerted is transmitted to the pressure transducer.

→ the output of the transducer is given to pressure monitor Transducer converts pressure into electrical signals. It is displayed in the monitor.

→ Strain guage pressure transducer is used. the change in pressure is given to the amplifier circuit.

Isolation Amplifier as in ECG system can be

→ Two indicator are available for systole display & diastole display.

→ If output of the amplifier is positive going pulse then D_3 will be ON. capacitor C_3 is charging upto the peak value. R_3 & C_3 combination is used to get sometime constant value which is used for stable display.

→ diastole circuit shows reading in indirect way

→ Initially clamping circuit available C_1 & D_1 are used to develop the voltage which is equal to the peak-peak value of the pressure pulse.

→ The voltage is appeared across R_1 resistor. Then D_2 diode is ON.

→ C_2 is charged upto the peak value of the pulse signal. The diastolic pressure is displayed using the indicator. M_2. M_2 reading = peak systolic value - peak to peak pulse pressure value.

(iii) Auscultatory Method :-

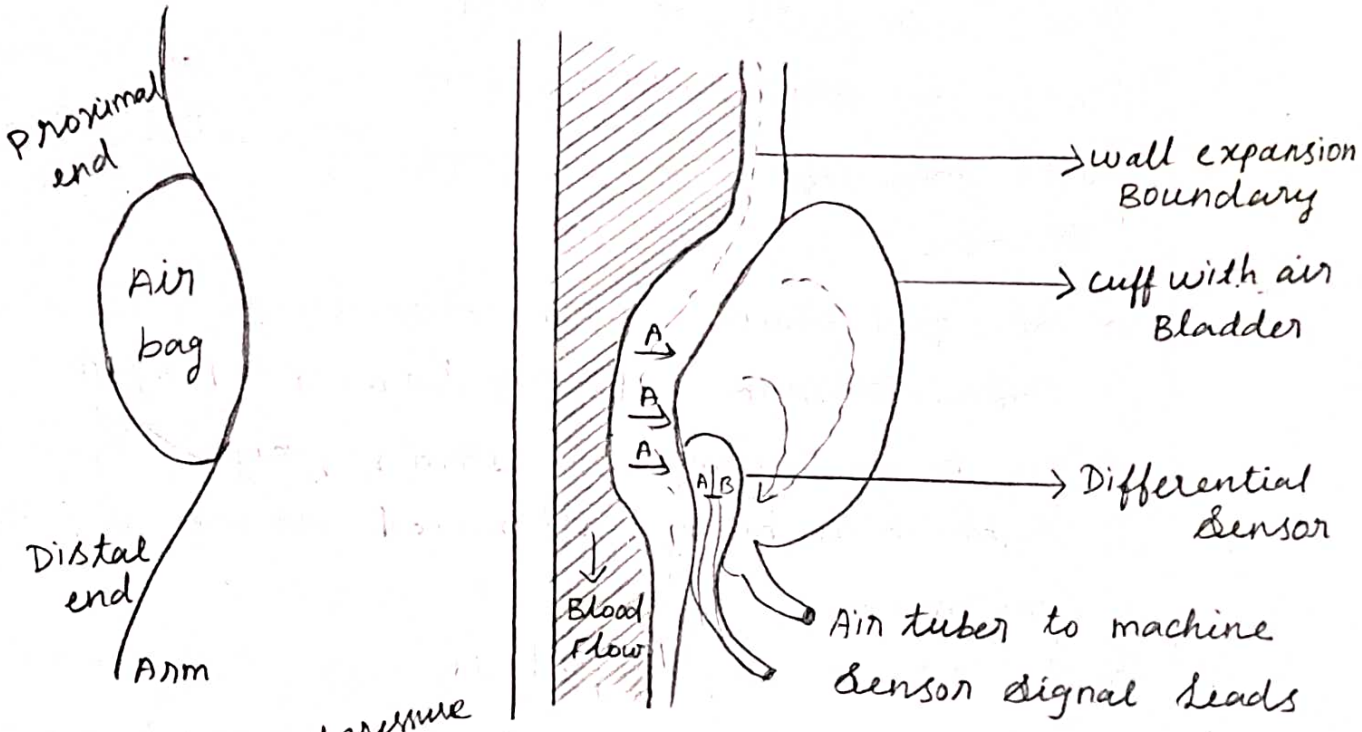
The differential auscultatory technique is a non invasive method for accurately measuring BP.

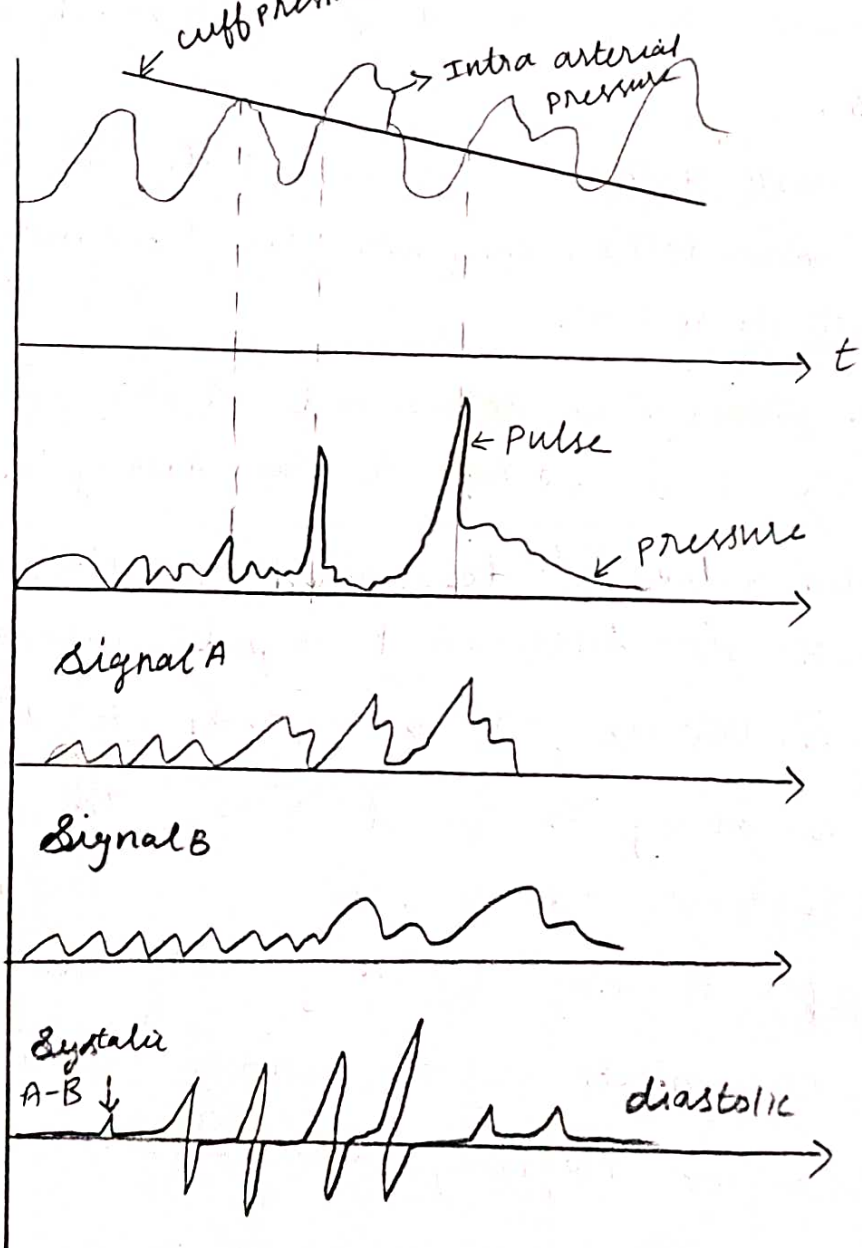
→ A special cuff mounted sensor consisting of a pair of pressure, sensitive element, isolates the signal created each time the artery is forced open.

→ As long the cuff pressure exceed the pressure in the artery, the artery is held closed & no pulse is generated.

→ As soon as the intra arterial pressure rise to a value, which momentarily exceed the cuff pressure the artery snaps open & pulse is created.

isolated.


Proximal end
Air bag
Distal end
Arm
wall expansion Boundary
cuff with air Bladder
A
A
A
A/B
Differential Sensor
Blood Flow
Air tuber to machine
Sensor Signal Leads


cuff pressure
Intra arterial pressure
t
Pulse
pressure
Signal A
Signal B
Systolic
A-B
diastolic

once the artery is open, Blood flows through it giving rise to the low frequency pressure wave which lasts until the arterial pressure again drops below the cuff pressure. this process is repeated until the cuff pressure drops to a value below the diastolic.

→ The signal consist of a slowly rising, low frequency component with a fast pulse as denoted by A.

→ Due to the air bag characteristics, high frequency component is highly attenuated leaving only the low frequency signal.

→ The low frequency signal is transmitted to the side of the sensor facing the air bag as denoted by the arrow marked B.

→ since most artefacts signals falls in the frequency range below 10Hz. they are also transmitted to both sides of the sensor.

→ Systolic pressure is determined as the pressure at which the First opening of the artery occurs.

→ Diastolic value is determined as the pressure at which the differential signals essentially disappears. because this corresponds to the last time the artery is forced open. The differential sensor subtracts the side 'B' signal from the side "A signal. thereby cancelling out the pressure wave components & the motion artifact signals and the higher frequency korotkoff signals are

Blood Flow Measurements:

The Blood flow rate can be determined with the help of blood flowmeter. The rate of flow of blood in a vessel is given as the volume of the blood that passes through the vessel in a given unit of time.

Types of Blood Flow Measurement

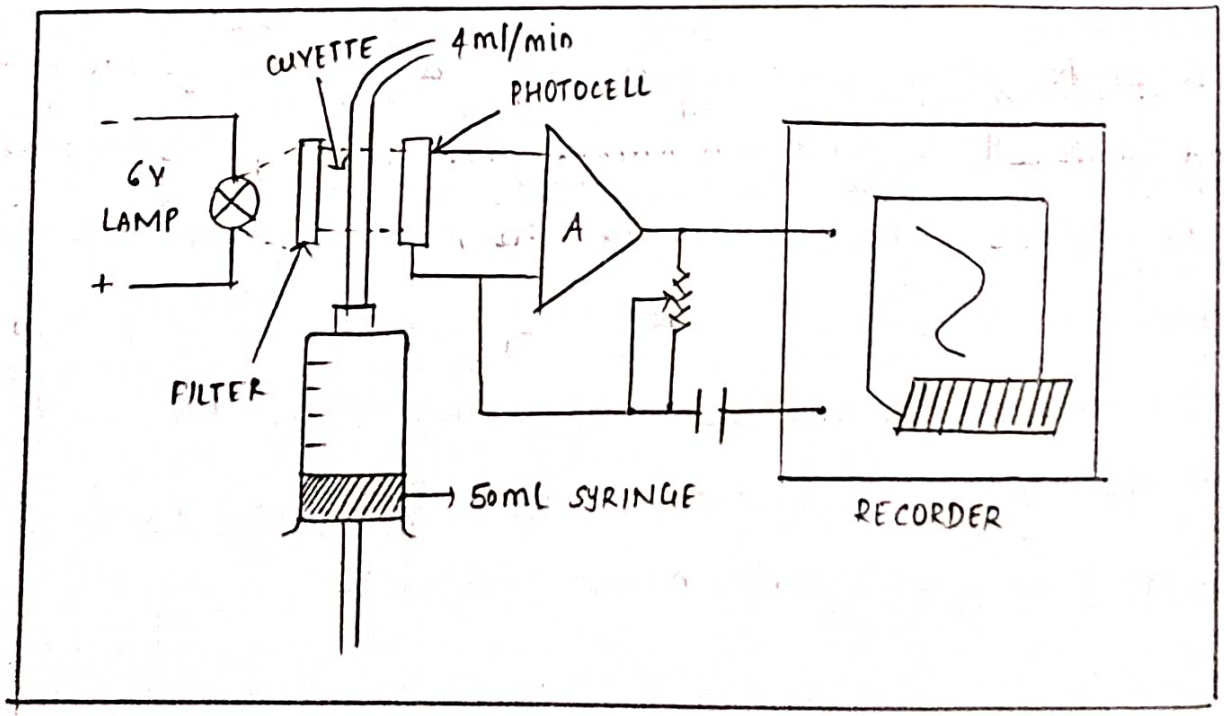
1) Electromagnetic blood flow meter
2) Thermal convection method
3) Radiographic method.
4) Indicator Dilution method
5) ultrasonic blood flow meter.

Dye Dilution Method:

Indicator substance is dye.

Indocyanine green (cardiogreen) dye → uses Fox & Wood for recording dilution curve.

→ dye is preferred because of its property of absorbing light in 800nm region of the spectrum



concentration of cardiogreen can be measured with the of infra red photocell transducer. Dry curvettes of a v volume = 0.01 ml are available

procedure:

→ It consist injecting the dye into right atrium by means of a venous catheter. usually 5 mg of cardiogree dye is injected in a 1ml volume. the quantity used may be 2.5 mg in the case of children.

→ A motor driven syringe constantly draws blood from the radial to femoral artery through a cuvette. the curve is traced by a recorder attached to the densitometer. After the curve is drawn, an injection of saline is given to flush out the dye from the circulating blood.

problems:

→ dye concentration 20 mg/ml of blood, optical density rises less with an increase in dye concentration than below this level.

→ peak concentration is less than 20mg/ml. → for optimum accuracy.

→ the photometric part consist of a source of radiation and photocell & an arrangement for holding the disposable polyethylene tube constituting the cuvette. An interference filter with a peak transmission of 805 nm is used to permit only infrared radiation to be transmitted. the wavelength is the isobestic wavelength for haemoglobin at various level of oxygen saturation.

In order to avoid the formation of bubbles, the curette tubing should be flushed with a solution of silicone in either. A flow rate of 40ml/min is preferred in order to get as short a response time as possible for the sampling catheter.

→ the sampling syringe has a volume of 50ml/min. the output of the photocell is connected to a Low drift amplifier.

→ High input impedance & Low output impedance.

Amplification ∝ Resistance value of potentiometer

Recording paper wide ⇒ 200m

paper speed - 10mm/s.

→ In the recording of dye dilution curves. A catheter is used to transport the blood containing dye from the sampling site, inside the cardiovascular system, to the densitometer located outside the body.

1) velocity of the flow within the catheter is not uniform, which cause the dye to mix within the tubes as it travel downstream. the mixing is a function of the flow rate & volume of the sampling system. the viscosity of the sampled fluid & the shape of the connector configuration of the sampling tube.

2) the second source is the measuring instrument itself. which may not have response characteristics fast enough to record instantaneous dye concentration as it actually occurs in lumen.

→ Distortion is very important, when the indicator method is used to measure the volume, since it is the measurement of the mean transit time of an indicator from the point of injection to the point of sampling.

→ To reduce distortion, computer software based corrections have been used.

2) Indicator Dilution Method:

It helps in the determination of rate of blood flow and not the velocity of blood.

Indicator → Any substance having No toxic side effect can be used.

It is readily mix with the blood & its concentration can be easily determined after mixing.

principle:

The substance used should be stable but should not be retained in the body.

Indicator types

→ Indocyanine dye

→ cardiogreen are widely used.

Radioisotopes are employed for this purpose.

Frequent indicator → isotonic saline.

There are two types of measurement.

(1) open circulation method

(2) closed circulation method.

Open circulation method:

Assume,

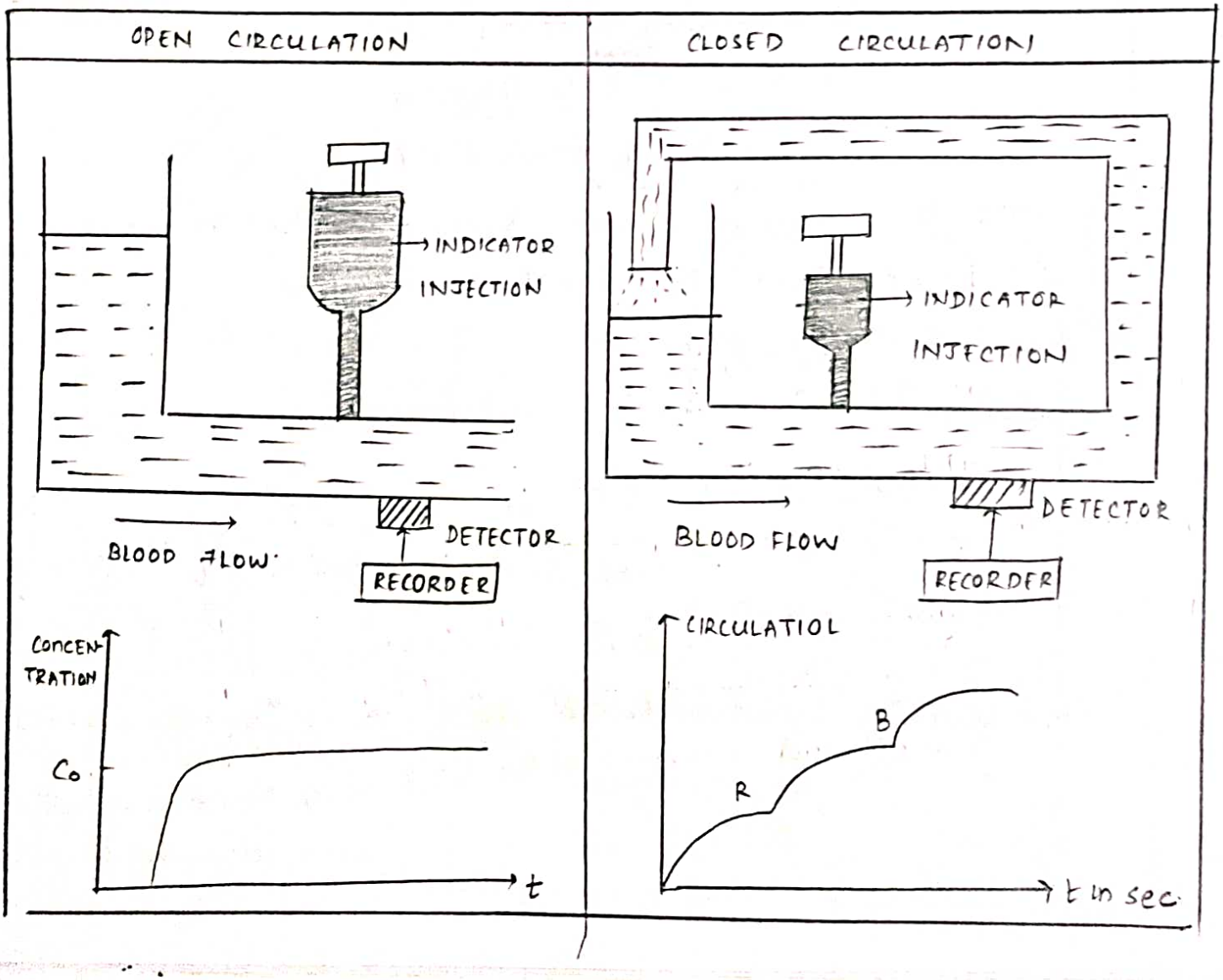
Blood is not recirculated.

The indicator is injected into the blood flow continuously at the beginning time 't' with a constant infusion rate of I gram / minute. A detector measures the concentration of the downstream from the injection point.

The output of the detector is connected to the recorder

$$\text{Rate of flow (Litres/minute)} = \frac{I \text{ (milligram/minutes)}}{C_o \text{ (milligram/Litres)}}$$

The flow can be measured with the help of injection rate (I), and measured concentration (C_o).



(ii) closed circulation method:

Dye (or) Isotope is used as an indicator.

Assume,

Blood is recirculated.

Here at first, the indicator is injected and its concentration is measured with the help of detector and when the indicator is recirculated the concentration increases step by step. The output of the detector is connected to the recorder & the flow can be determined.

$$f_1 = \frac{(c - v\cos\theta)}{c}$$

Where

f = transmitted frequency

c → velocity of sound in blood.

θ → angle of inclination.

v → velocity of blood cells.

Assume the incident & scattered radiation are both inclined at θ to the direction of flow.

$$f_2 = f_1 \left[\frac{c}{c + v\cos\theta}\right]$$

resultant Doppler shift $\Delta f = f - f_2 = f - f_1 \left[\frac{c}{c + v\cos\theta}\right]$.

$$\therefore \Delta f = \frac{2fv\cos\theta}{c}$$

Separation of transmitter & receiver → Doppler shift is zero

Right angles → minimum doppler shift frequency is present.

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Ultrasound Blood Flow Measurement.

In ultrasonic blood flow meter, the velocity of the flowing blood can be determined with a beam of ultrasonic flow meter ∟ Doppler type ultrasonic flow meter.

There are two type

→ Transit time velocity meter type.

→ Doppler shift velocity type.

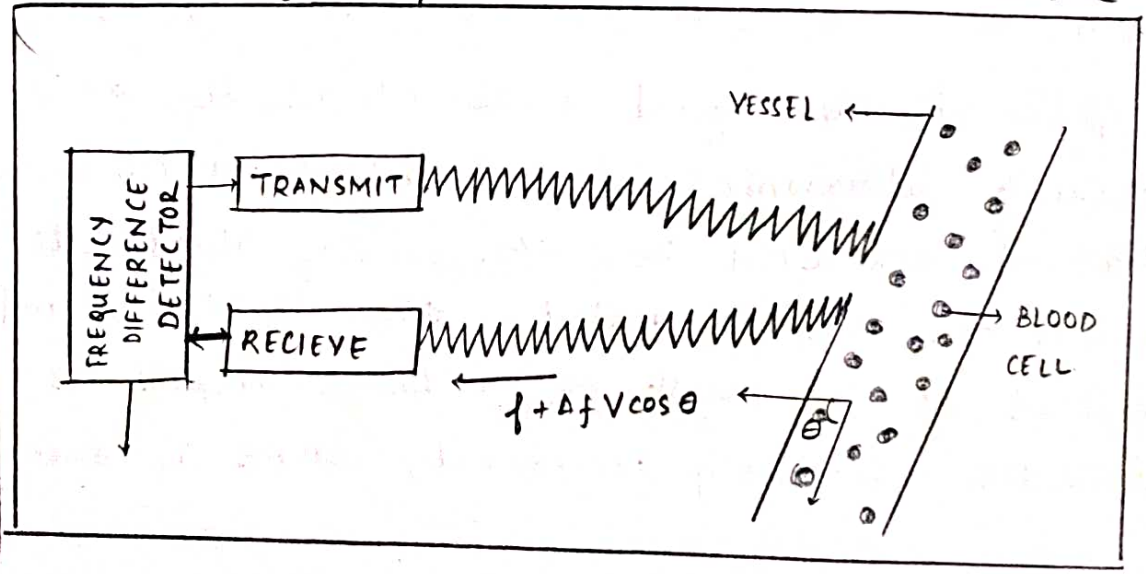
(i) Transit time type:

A pulsed ultrasonic beam is directed at a shallow angle through a blood vessel & its transit time is then measured. When the blood flow is in the direction of energy transmission, the transit time short ∟ When the blood flow is in opposite direction the time value is greater.

(ii) Doppler shift flow velocity type

→ Non invasive technique to measure blood velocity in a particular vessel from the surface of the body.

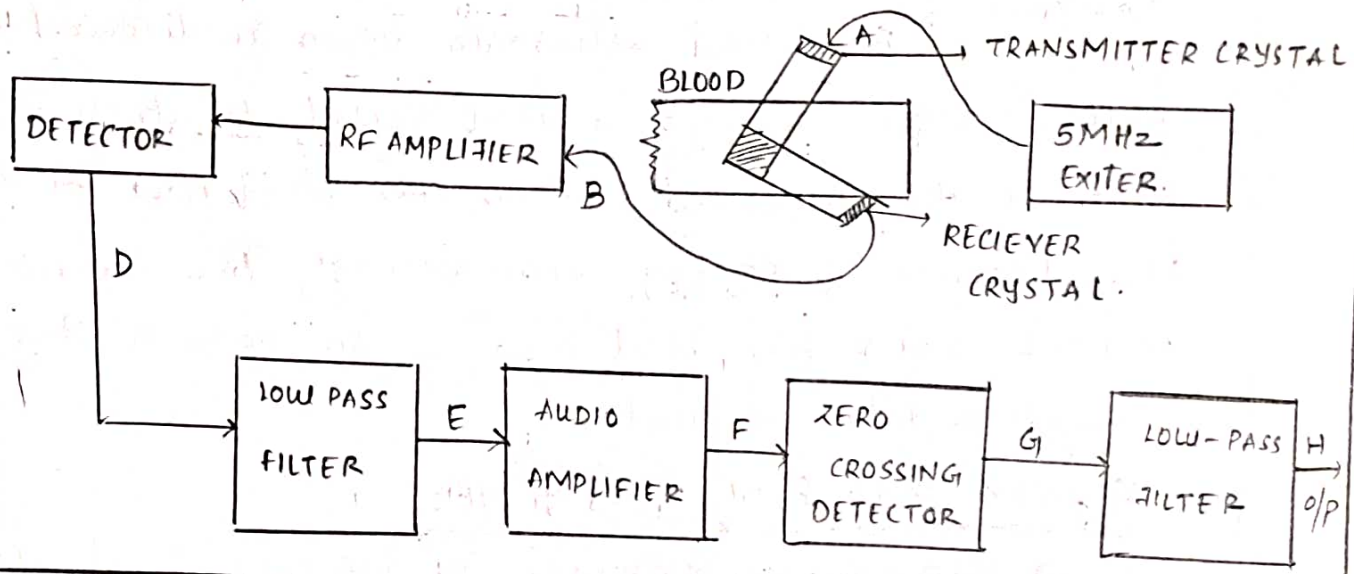
→ It is based on the analysis of echo signal from the erythrocytes in the valvular structure.



→ Because of the doppler effect, the frequency of the signals changes relative to the frequency which the p transmit.

→ Doppler freq shift is a measure of the size & direction of the flow velocity.

→ The incident ultrasound is scattered by the blood cells & the scattered wave is received by the second transducer. Frequency of moving scatterers & velocity of the scatterers.



→ Alteration in frequency occurs first as the ultrasound arrives at the scatterer & second as it leaves the scatterer. If the blood is moving towards the Transmitter, the apparent freq is given by f_1

→ The piezo electric crystal A is electrically excited to generate ultrasonic waves, which enter the blood. ultrasound scattered from the moving blood cells excites the receiver crystal. The electrical signal received at B consist of a large amplitude excitation frequency component, which is directly

coupled from the transmitter to the receiver, plus a very small amplitude doppler-shifted component scattered from the blood cells.

→ The detector produces a sum of the difference of the frequencies at D. The low pass filter selects the difference frequency, resulting in audio frequency at E.

→ Each time the audio wave crosses the zero axis, a pulse appear at G.

→ The filtered output level at H will be proportional to the blood velocity.

→ Low freq gain is too high, resulting in wall motion artifacts.

Max. Doppler shift = 15 kHz.

Wall motion signal = 100 Hz (below).

pulsed ultrasonic doppler flow detector:-

Instrument is based on the CW Doppler. The angle between the sound beam & velocity vector should be known.

→ If the ultrasonic source is pulsed & the Doppler shift of the returning echo is determined. If the return signal is range gated, then the distance to the moving interface as its velocity w.r.t. the beam can be measured.

The system consist of

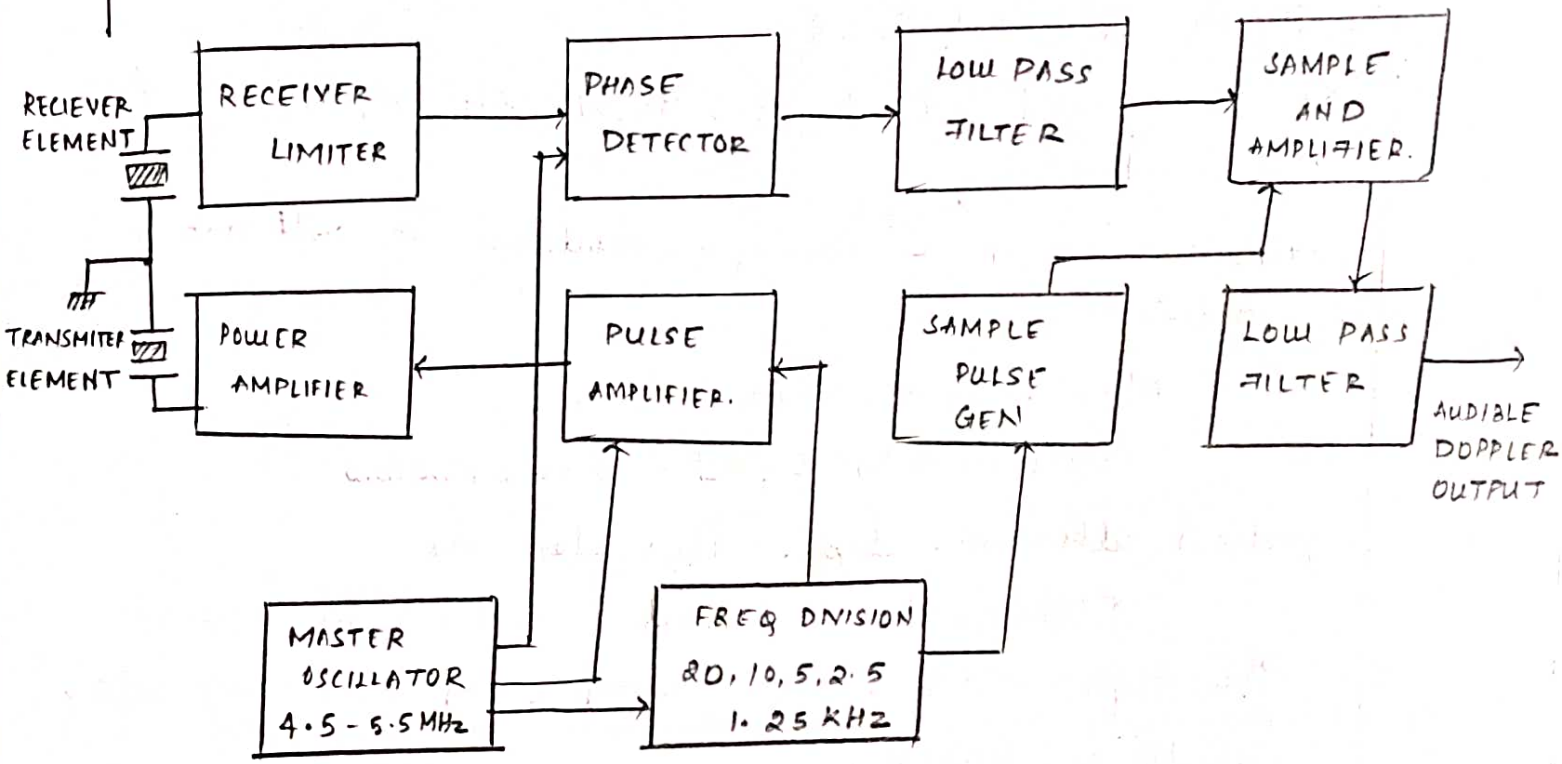
→ Transmitter
→ Receiver
→ zero crossing Detector
→ Spectrum Analyzer.

Transmitter:

It comprises a master oscillator whose frequency can vary from 2 - 10 MHz.

5 MHz → good compromise to detecting the blood flow in vessels 4-6 cm in depth.

→ Master oscillator also drives the pulse repetition frequency (PRF) ripple counter & provides a



continuous reference signal to the receiver phase detector for the detection of the doppler-shifted echoes.

→ The selection of PRF depends on the
- → depth of the vessel
- → expected Doppler shift
- → Attenuation characteristics of the tissue.

PRF (kHz)	Depth range	Max. detectable kHz.
25	3	12.5
18	4.3	9
12.5	6	6.25.

when gate opens, n cycles of the Master Oscillator pass through to the power Amplifier.

P = 10-30w during 1ms excitation burst.

pulse echo transducer → use tungsten epoxy Q-1.B-2.6.

Q of pulsed doppler transducer = 5 to 15.

Receiver:

the back scattered Doppler shifted signals from the blood vessel range in intensity from 50dB to more than 120dB.

Bandwidth - 3MHz

Gain - 80dB.

→ Receiver is followed by a single side band type quadrature phase detector which separate the upper & Lower doppler side band for sensing Flow direction.

→ the detector consist of a phase shift network, which split the carrier into two components that are in quadrature which means they are 90°.

→ the doppler shift of the received echo, back scattered by the moving blood is detected by sensing the instantaneous phase difference between the echo & reference signal from the master oscillator.

→ the envelope frequency from the phase detector is the Doppler difference frequency

Zero crossing Detector:

In order to measure blood velocity, frequency meter is needed to analyze the frequency component of the Doppler signal.

Doppler velocity meter uses zero crossing detector for this purpose.

Function:

To convert audio frequency amplifier output to a proportional analog output signal.

→ It is a series of pulses.

→ These pulses are passed through a Low pass filter to remove the high frequency component.

filter pass frequency = 0 – 25 Hz.

The detector gives not only the velocity with which the blood flow but also its direction.

Spectrum Analyzer:

→ It is used to derive blood flow velocity information from the Doppler signal.

→ A short length of audio signal to produce spectral display.

→ frequency as the abscissa
time as the ordinate.
Spectral intensity represented by record darkening.

Cardiac output Measurement:

Cardiac output is the amount of blood delivered by the heart to aorta per minutes.

Normal adult cardiac output - 4-6 Liters/min

Types of cardiac output Measurement.

The cardiac output is measured by using three methods.

→ Fick's method
→ Indicator dilution method
→ Measurement of cardiac output by impedance change
→ Thermodilution method.

Fick's method:

Cardiac output is determined by the analysis of gas keeping of the organism.

It can be calculated by continuously infusing oxygen into the blood (or) removing it from blood & measuring the amount of O_2 in the blood before & after its passage.

$$I = C_A Q - C_V Q.$$

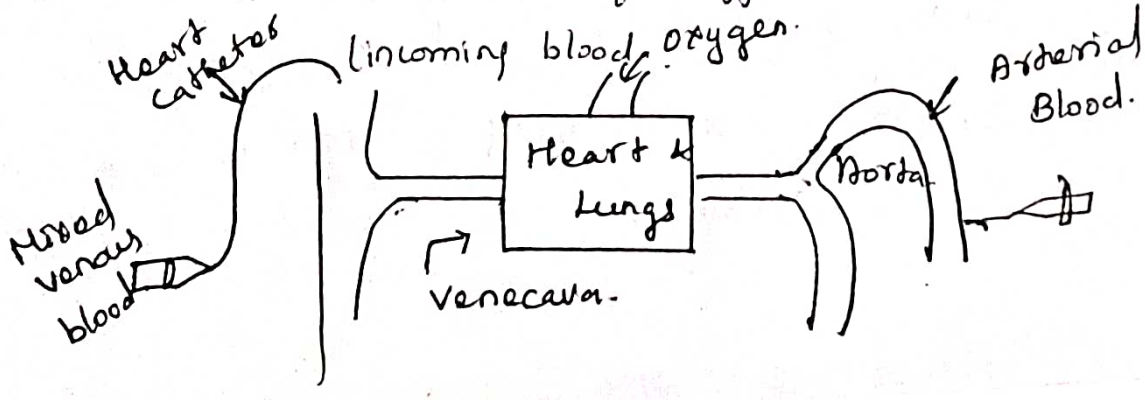
I → amount of infused or removed oxygen per unit time

$$Q = \frac{I}{C_A - C_V}.$$

Q - cardiac output in Litre/min

C_A → concentration of oxygen in arterial blood. (outgoing blood)

C_V → concentration of oxygen in venous blood (incoming blood)



(ii) Indicator dilution method:

Amount of dye or radioisotope is used as an indicator in the blood circulation & then measuring the concentration of the indicator w.r.t. time.

The concentration of the indicator $C = \frac{dM}{dV}$.

$$C\frac{dv}{dt} = \frac{dM}{dt}$$

Where, $\frac{dv}{dt} = Q$. cardiac output

$$CQ = \frac{dM}{dt}$$

$$\int dM = CQ \int_0^t dt$$

$$M = Q \int_0^t C\,dt$$

$$\therefore \boxed{Q = \frac{M}{\int_0^t C\,dt}}$$

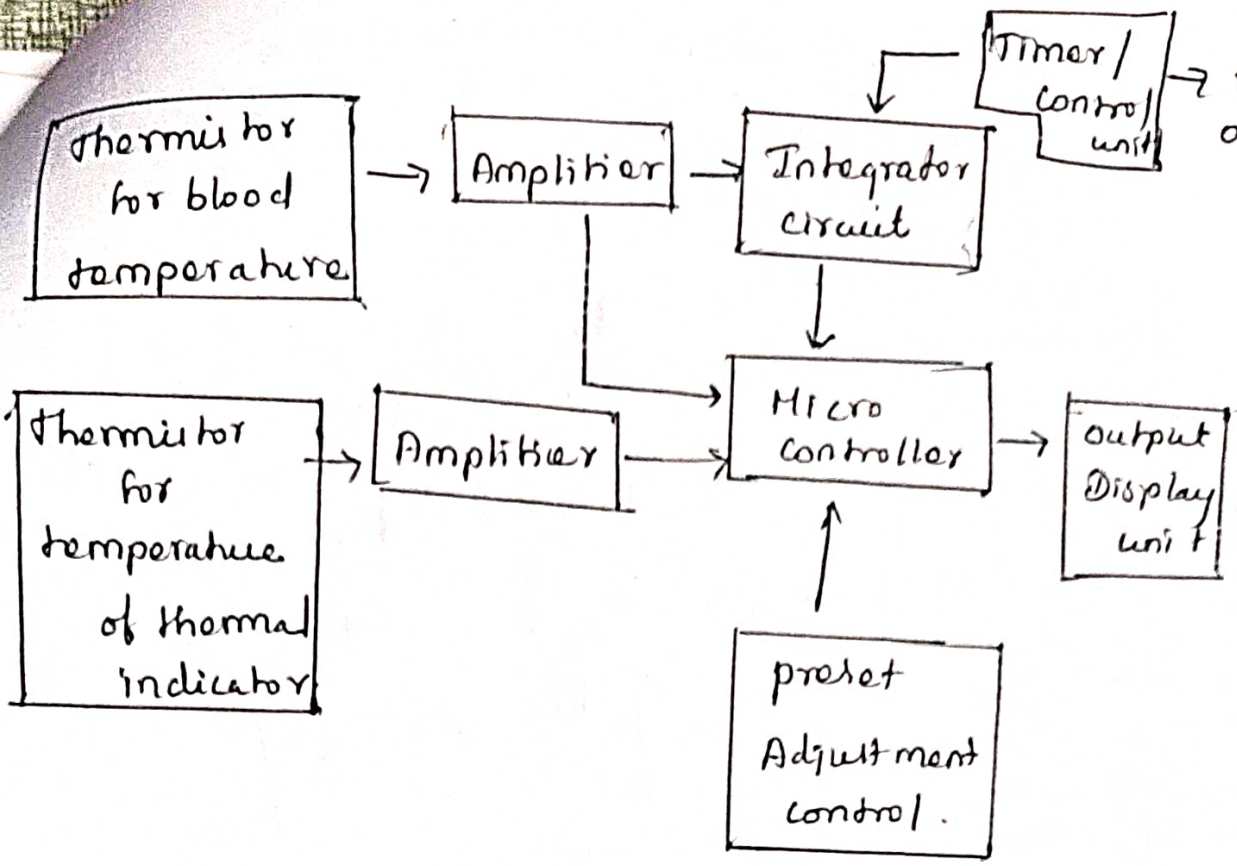
(iii) thermo dilution method:

→ 10ml of 5% dextrose in water of room temperature is injected as the thermal indicator into right atrium.

→ After mixing, it is detected in the pulmonary artery by a thermistor.

→ The temperature difference between the injected temperature and the circulating blood temperature is measured.

→ Amplifier block is used to remove non linearity of the thermistor.



Measurement of cardiac output by impedance change

The cardiac output can be determined by the impedance method.

Four electrode are placed surrounding the thorax.

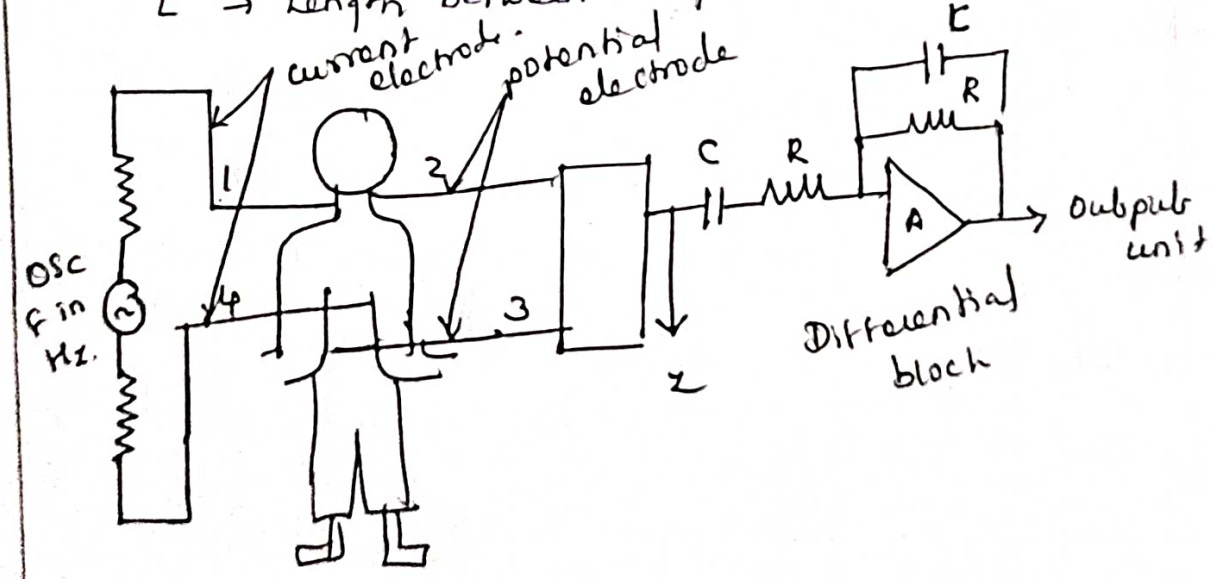
current electrode - 1, 4

Voltage electrode - 2, 3.

ρ - Resistivity of the patient's hemacrit.

A → Area of thorax.

L → Length between the potential electrode 2 & 3.



Resistance of thorax $R = \rho \frac{l}{A} = \frac{\rho L^2}{AL} = \frac{\rho L^2}{V}$ ∴

$$\therefore V = \frac{\rho L^2}{R}$$

$$\frac{dv}{dR} = -\rho \frac{L^2}{R^2} dv$$

$$dv = -\frac{\rho L^2}{R^2} dR.$$

for ac excitation, z is used

$$\boxed{dv = -\frac{\rho L^2}{z^2} dz}$$

Cardiac output can be measured by multiplying dv with heart beat rate per minute.

11

Unit- V

Biochemical Measurement

Blood gas Analyzer

Blood gas analyzer are used to measure the pH, partial pressure of carbondioxide (PCO_2) & partial pressure of oxygen (PO_2) of the body fluids with special reference to the human blood.

Types of blood gas Measurement

- → Acidbase balance
- → Blood pH measurement
- → Blood PCO_2 measurement
- → Blood PO_2 measurement.

(i) Acid-base Balance:

* pH of extracellular fluid → 7.35 to 7.45 → body fluid is alkaline
* pH exceed 7.45 → body in state of alkalosis.
* pH below 7.35 → acidosis.

Alkalosis & Acidosis are diseases condition.

Three physiological Mechanism

- → buffering by chemical means
- → Respiration
- → excretion in to urine by kidney.

* Blood & tissues fluid contain chemical buffers which react with added acid and bases and minimize the resultant change in hydrogen ion.
* Carbondioxide can be removed by increased breathing.

∴ Hydrogen concentration of blood can be effectively modified.

(ii) Blood pH measurement:

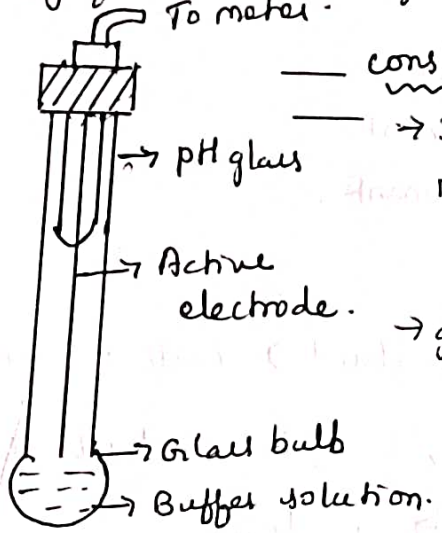
The chemical balance in the body can be determined by the pH value of blood & other body fluid.

→ pH is defined as the hydrogen ion concentration of
→ It is the logarithm of the reciprocal value of H^+ ion concentration.

$$pH = -\log_{10}[H^+] = \log_{10}\frac{1}{[H^+]}$$

pH = 7 → Neutral solution
pH Less than 7 → acidic
pH above 7 → Basic.

→ Body fluids are slightly basics in nature



— construction & working:

→ It is made up of a thin glass membrane and allow only the hydrogen ion to pass through it.
→ glass electrode provide a membrane interface for H^+ ions.
→ the glass bulb at the lower end of the pH meter contains highly acidic buffer solution.

→ the glass tube consist of Ag-AgCl electrode & the reference electrode which is made of calomel (or) Ag-AgCl is placed in the solution in which pH is measured. The potential is measured across the two electrode.
→ electrochemical measurement, which should be obtained by each electrode → Half cell. The electrode potential is called half cell potential.

→ The glass electrode only to measure pH values around 7.

pO_2 measurement:

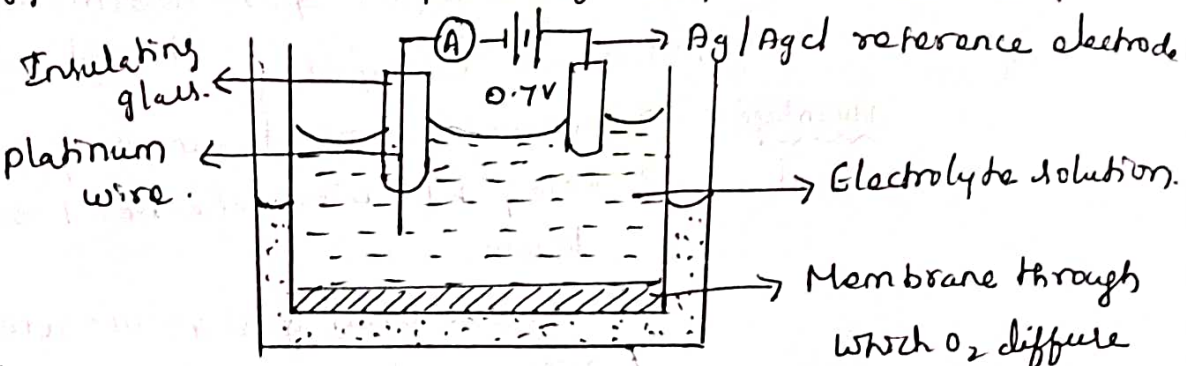
→ The platinum wire which is as active electrode is embedded in glass for insulation and only its tip is exposed.
→ It is kept in the electrolyte solution in which the oxygen is allowed to diffuse.

Reference electrode – Ag/AgCl

→ A voltage of 0.7V is applied between the platinum wire and the reference electrode.

→ the negative terminal is connected to the active electrode through a micro ammeter & positive terminal is given to the reference electrode.

→ Oxygen reduction take place of the platinum cathode.



The electrolyte is generally sealed in the electrode chamber by means of the membrane through which the oxygen can diffuse from the blood or sample solution.

There are two type of PO_2 measurement.

In-vitro Measurement

→ Blood sample is taken & the measurement for O_2 in Laboratory.

→ electrode is placed in the sample blood solution & PO_2 is determined

In vivo measurement.

→ O_2 saturation is determined while blood is flowing in circulatory system.

disadv:

→ reduction process in platinum cathode.

Blood PCO_2 measurement:

There is a Linear relationship between the logarithm of PCO_2 & pH of a solution.

→ It is made by surrounding a pH electrode with a membrane selectivity permeable to CO_2.

improved PCO_2 electrode → severinghous electrode.

→ In this electrode, the membrane permeable to CO_2 is made of Teflon & glass contain matrix layer

which allows only the CO_2 gas molecules to diffuse it.

→ CO_2 can diffuse rapidly through the membrane and the measurement can be done easily.

Non invasive Blood gas monitoring:-

Blood gas monitoring has two techniques.

→ Invasive techniques → to determine arterial blood gases.

→ Non-Invasive techniques

Advantage: ↳ intermittent blood sampling.

→ delay b/w when the blood sample is drawn.

→ when the blood gas values are reported average about 30 min.

Disadvantage:

→ painful

→ irreversible cell damage occurs.

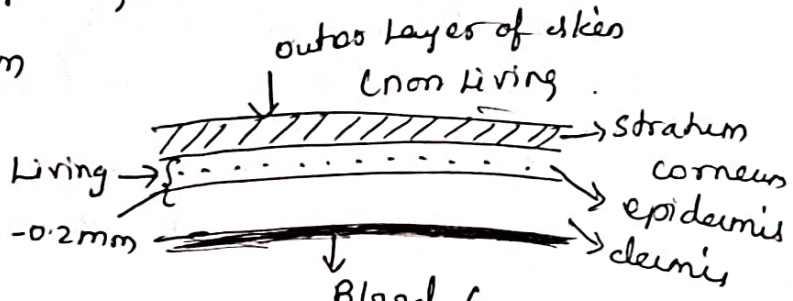
Skin characteristics:

The skin has 3 principal layer.

(i) Stratum corneum

(2) epidermis

(3) dermis.



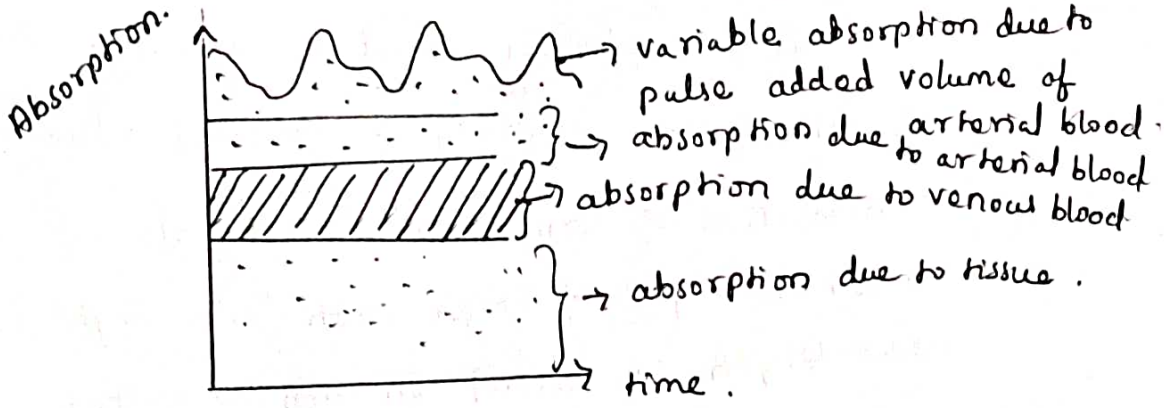
Normal gas diffusion → skin is low → heat is increased 40°C.

→ skin become more permeable to gases.

pulse oximetry:

pulse oximeter → 1980 → introduced by yoshiya.

Instrument determines SO_2 by Analyzing the time varying (or) ac, components of the light transmitted through the skin during Systolic phase of the blood flow in the tissue.



Other Instrument:

Accuracy: 2rr%.

→ Ear → toe

→ finger tip oximetry.

Transcutaneous SO_2 Sensor:

Transcutaneous SO_2 sensor is used for both transmission and reflective mode, make use of light source & photodiode.

Transmission mode → two faces each other & segment of the body is interposed.

Reflection mode → Light source, photodiode are mounted adjacent to each other on the surface of the body.

These transmission sensor are placed on the finger trips, toes, ear Lobes (or) Nose.

Light source → Red & infrared LED.

↓ ↓

Wavelength → 660nm 940nm

Colorimeter:

→ colorimeter is used to determine blood protein and iron Level.

→ It is used to measure the transmitted and absorption Light as it passes through a sample.

principles:

This principle is used for Analysis the absorbance transmittance property of chemical admittance substance

A solution of concentration 'c' is placed in a curvette with a length of high path 'L', Light rays of particular wavelength is passed through a filter.

I_0 → initial intensity of the light entering the curvette.

I_1 → Light leaving the curvelte

Transmittance $T = \frac{I_1}{I_0} \times 100\%$.

for second curvette, has intensity I_2, It is lower than I_1. the light transmitted through successive curvelte decreases in the same manner.

The absorbance $A = -\text{Log} \frac{I_1}{I_0}$

$A = \text{Log} \frac{1}{T}$

Total Absorption of the two liquid column can be obtained by the sum of individual absorbance.

Amount of Absorbance $\propto$ No. of molecules.

Beer's Law Absorbance can be measured as.

$A = aCL$

L → Light path Length of the curvette.

C → concentration. of the absorbing substance

a - Absorbivity.

Absorbivity → can be obtained by measuring the absorption of the liquid with known concentration.

Concentration of unknown solution

$$C_u = C \cdot \frac{A_u}{A}$$

Where, C → concentration of Std. solution

A_u → absorption of unknown solution

A → absorption of Std. solution.

Adv:

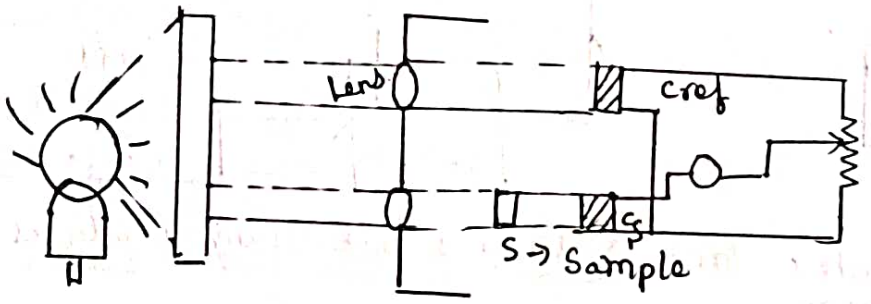
- Simple in construction & operation.
- → high accuracy

dis Adv:

→ Bandwidth is large.

(i) filter photometer (or) colorimeter.

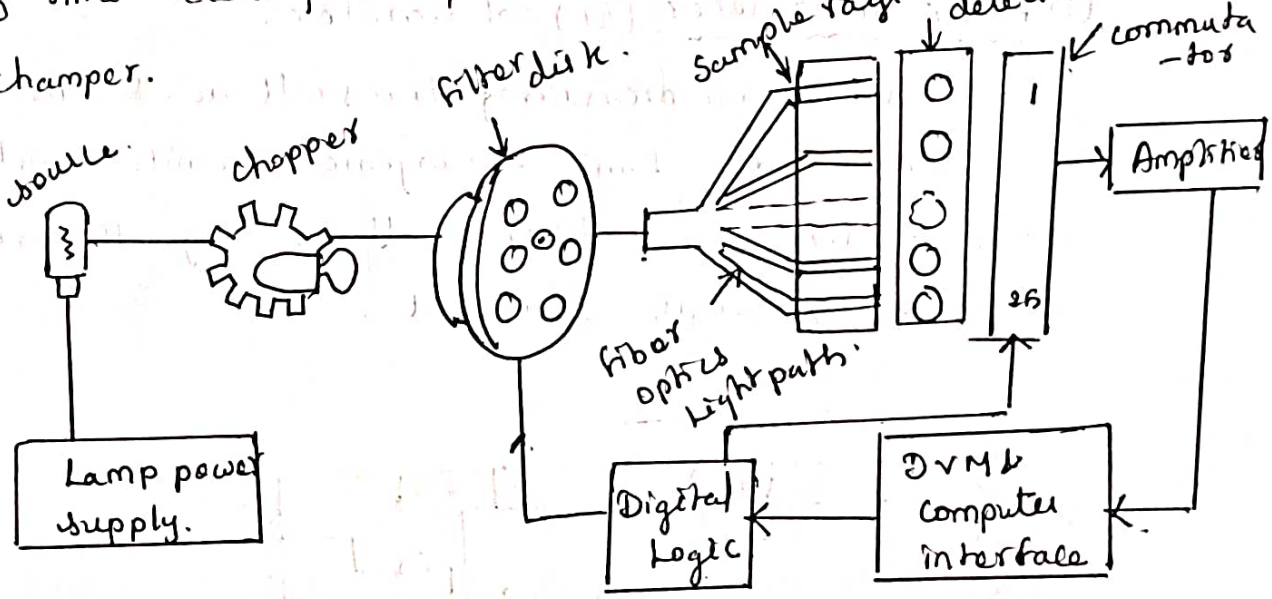
→ It is used for measuring transmittance & absorbance of solutions. The Lamp arrangement emits light of different wavelength & filter F allows only the suitable range of wavelength through it.



The light beam from the filter is converted into parallel beam by an optical arrangement. The light falls on two photoelectric cells. The reference cell (C_{ref}) and the sample cell (C_s). Without the sample, the output of the sample cell (C_s) is reduced & hence the potentiometer divides the output of C_{ref} until of, galvanometer show the balance The potentiometer can be calibrated in transmittance or absorbance unit.

Multichannel colorimeter (photometer)

In multichannel photometers, a single light path, a batch of samples is introduced. Measurement can be carried out simultaneously using a multiplicity of fiber optic light paths and detectors and then the samples are scanned electronically instead of mechanically. The 24 sample cuvettes are arranged in a rack in a 3 key eight matrix. The 25th channel serves as a reference beam & eliminates possible source & detector drifts. The time required to place the cuvette rack in to the measuring position corresponds to the amount of time necessary to put one sample into a sample chamber.



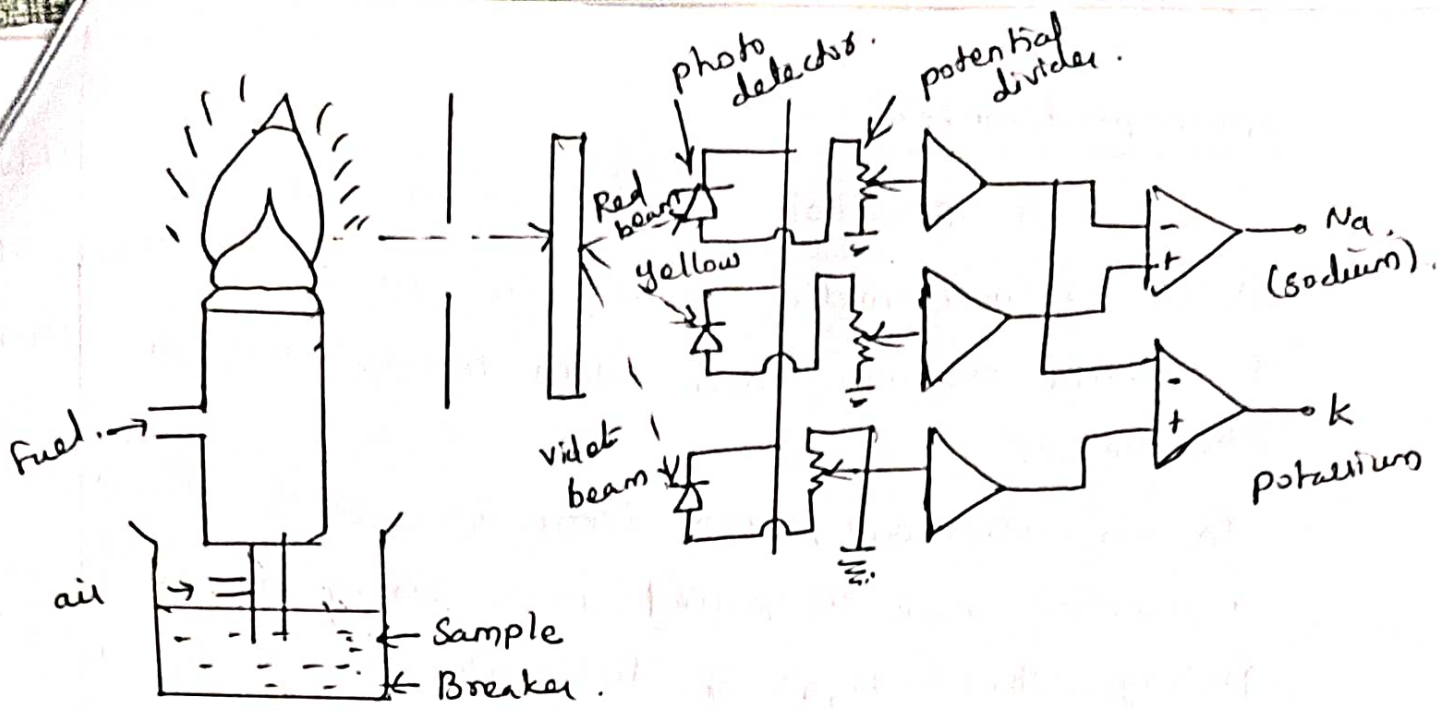
Flame photometer (or) Sodium & potassium Analyser.

A flame photometer is used in order to determine the concentration of potassium (K), sodium (Na), calcium (Ca) & Lithium. It is used in the Analysis of blood or urines.

Flame:
yellow → sodium
violet → potassium.

Lithium → used for calibration

When their solutions are aspiration into flame. This characteristics is used in flame photometer.



In this method, Fine droplets of the sample is aspirated into a gas flame that burns in a chimney. A known amount of Lithium salt is added to the sample as a reference.

As a result, → red light emitted by Lithium.

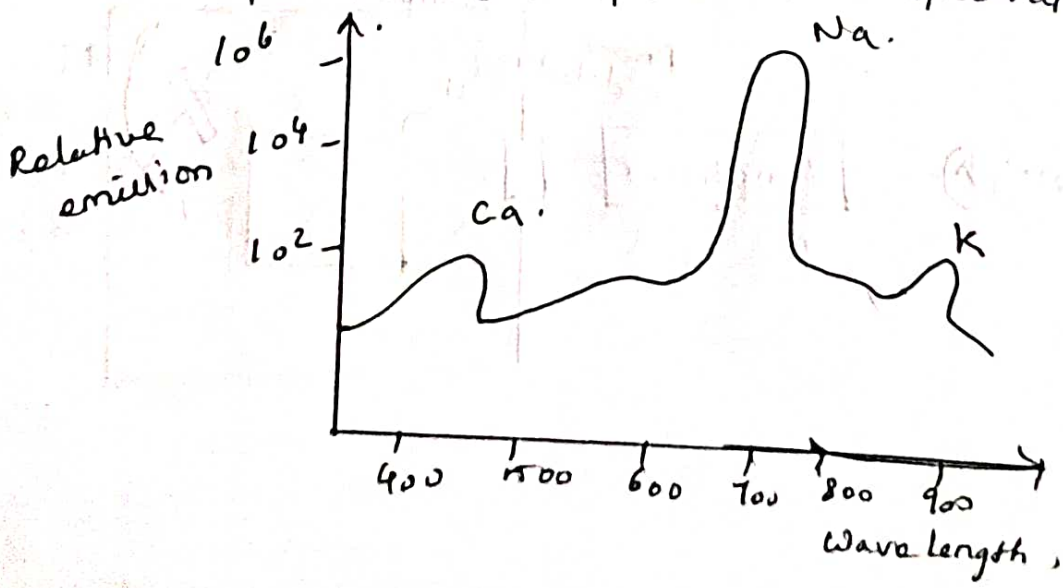
→ yellow " by Sodium

→ violet " by potassium.

these diffracted colours are made to incident on photodiodes the photodetector circuit consist of a reverse biased diode in which the current flow increases & the incident light intensity increases. A calibration potentiometer is used in every channel. O/p → Sodium & potassium.

Lithium → Standard reference.

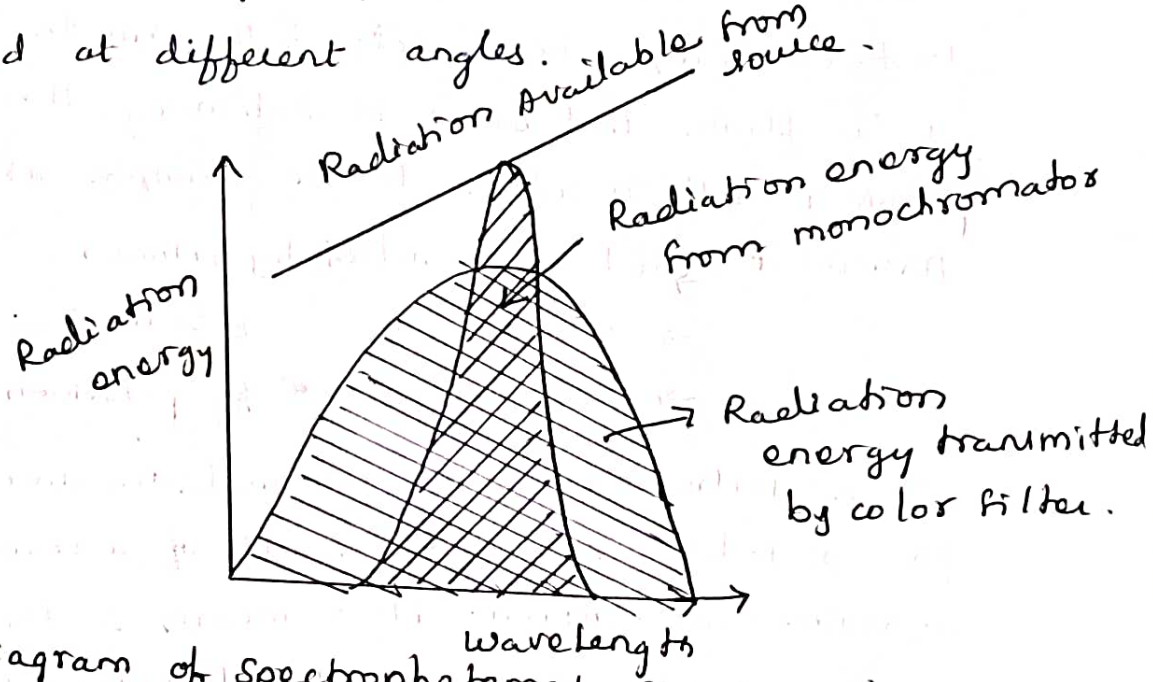
The output can be compared with spectral illustration.



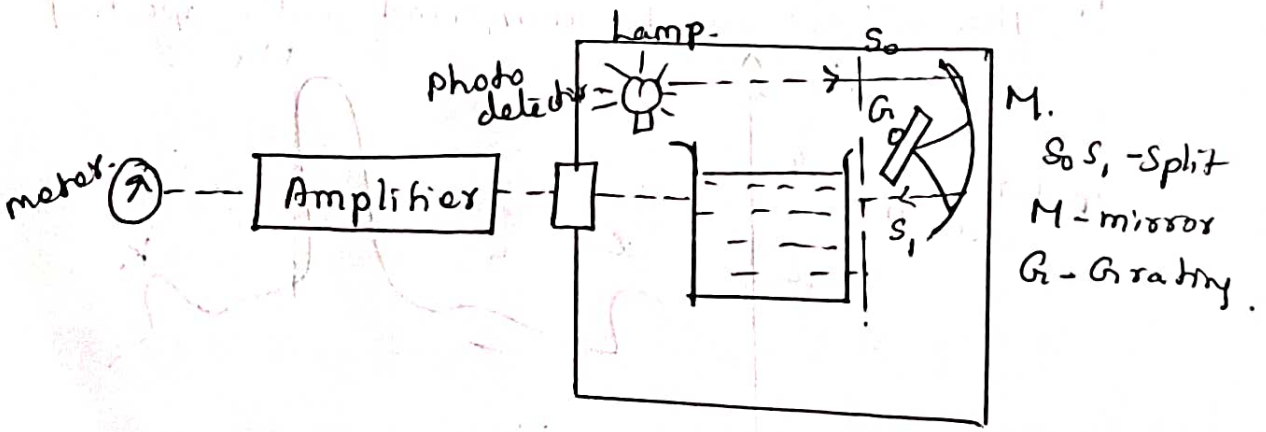
Spectrophotometer:

A spectrophotometer is an instrument which isolates monochromatic radiation in more efficient & versatile manner than colour filters used in filter photometers.

In this instrument, light from the source is made into a parallel beam & passed to a prism or diffraction grating, where light of different wavelength is dispersed at different angles.



Block diagram of Spectrophotometer (or) simple Spectrophotometer.



In spectrophotometer, selection filter of colorimeter is replaced by a monochromator. Monochromator uses diffraction grating (G) (or) a prism to disperse light from the lamp. Light falls through the slit so into its spectral components. Split S_1 is used for selecting a narrow band of the spectrum which is used to measure the absorption of the sample in cuvette. The light from the cuvette is given to photo detector. It convert light into correct only electrical signal. The electrical signal is amplified by using an amplifier. The output from the amplifier, is given to meter which shows absorption.

→ Light absorption is varied when the wavelength is varied.

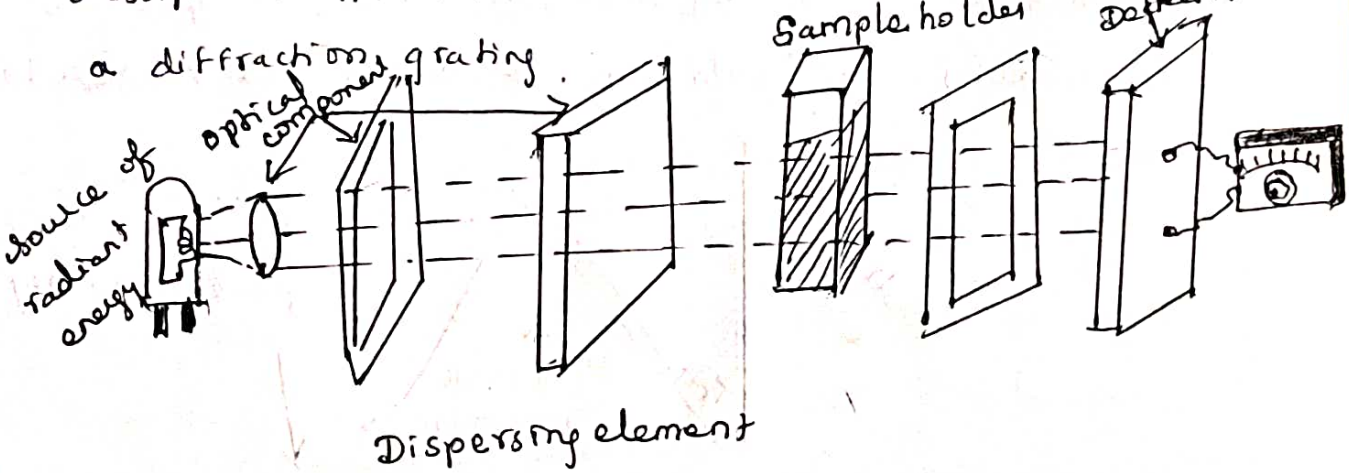
→ mirror M is reduced to reduce the size of the instrument

Spectrophotometer type instruments:-

The essential components are.

→ A source of radiant energy, which may be a tungsten lamp, a xenon-mercury arc, hydrogen (or) deuterium discharge lamp etc.

→ filtering arrangement for the selection of a narrow band of radiant energy. It could be a single wavelength absorption filter & interference filter, a prism or a diffraction grating.



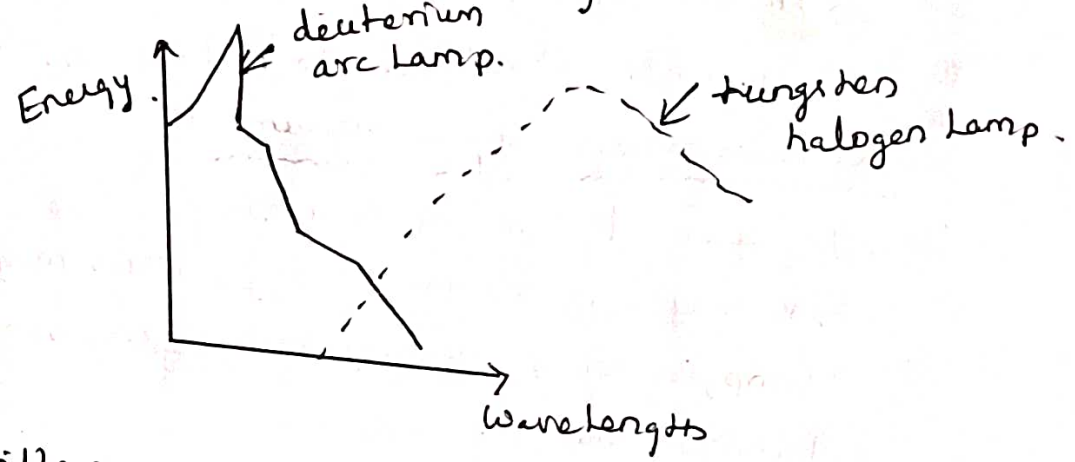
→ An optical system for producing a parallel beam of light for passage through an absorption cell. The sy may include lenses, mirrors slits, diaphragm etc.

→ A detecting system for the measurement of unabsorbed radiant energy which could be the human eye, barrier layer cell, photo tube (or) photo multiplier tube.

→ A readout system or display, which may be an indicating meter or numerical display.

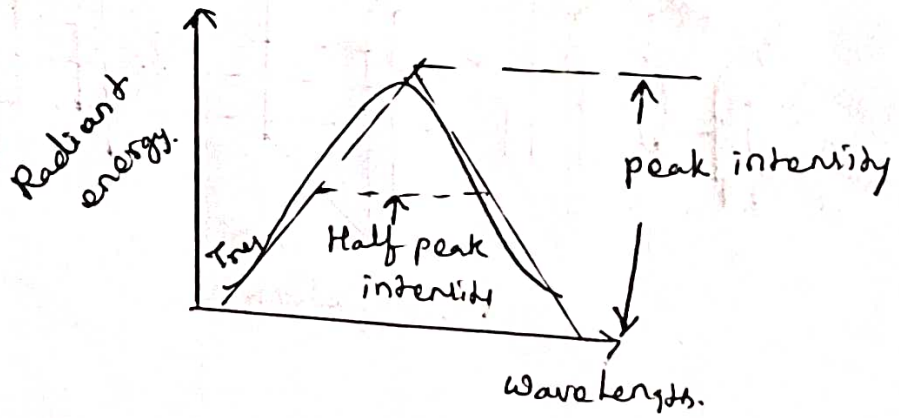
Radiation sources:

To provide a sufficient intensity of light which is suitable for making a measurement.



optical filter:

A filter may be considered as any transparent medium which by its structure, composition or colour enables the isolation of a particular wavelength.



Auto Analyzer:

Auto analyzer is used to measure blood chemistry & display on that on graphic recorder it contain various blocks.

Sampler:-

Samples are feeder into the analyzer by using the samples. upto 120 samples can be placed in the samples. Sampler contains the holes in which the sample tubes can be placed.

Mixer (or) Proportioning pump & manifold.

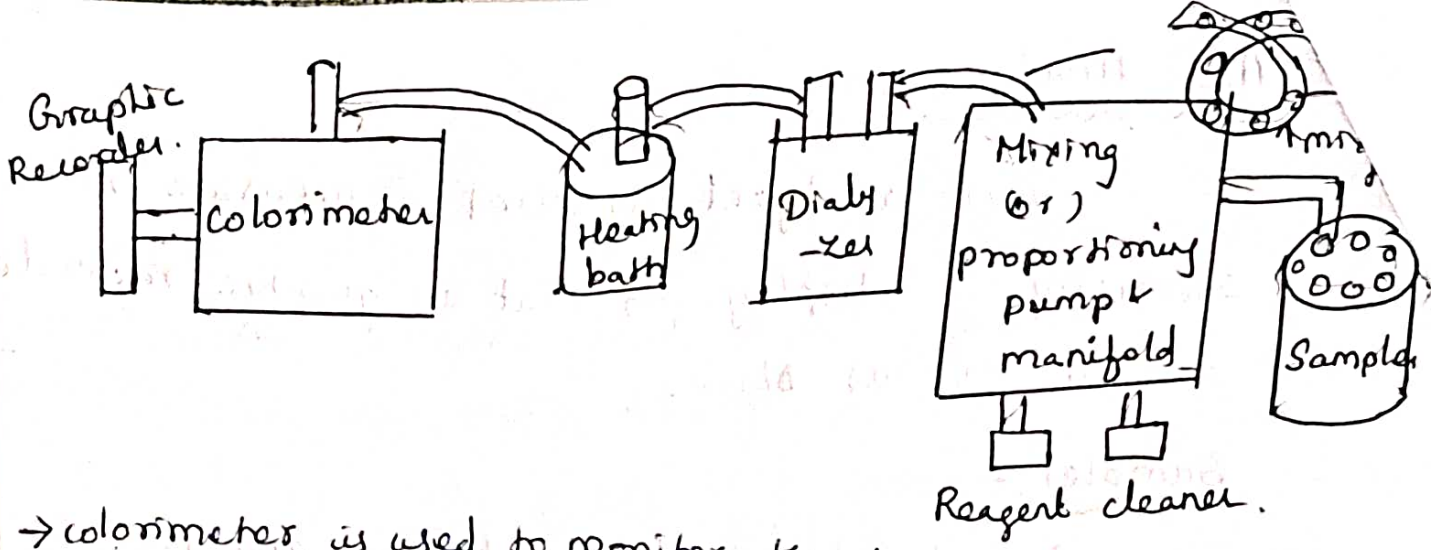
→ Least of Analyzer. proportioning pump is basically a simple peristablic pump working simultaneously on a No. of tubes with certain ratio of diameter.

→ Samples are mixed with reagents to affect the proper chemical colour reaction which can be read by the colorimeter. Air segmentation in the mixing tube separates the samples & is used to separate sample from the cleaner.

→ It also pumps fluid at precise rate to other modules. The proper odour development depends on reaction time & temperature.

→ Dialyzer separates interfacing substance from the sample by permitting selective passage of sample through a semipermeable membrane.

→ Heating bath is used to heat fluids continuously to exact temperature 37°C. This module is very important colour & Temperature.



→ colorimeter is used to monitor the changes in optical density of the fluid which flows through a tubular flow.

Colour intensity ∝ Concentration substance.
↓ converted into
electrical voltage.

→ Graphic recorder is used to convert electrical signal into a graphic display.

Blood cell counter:

Blood consist of a fluid called plasma.

plasma fluid occupies
- → 55 % blood
- → 45 % blood cell

Types of Blood cells.

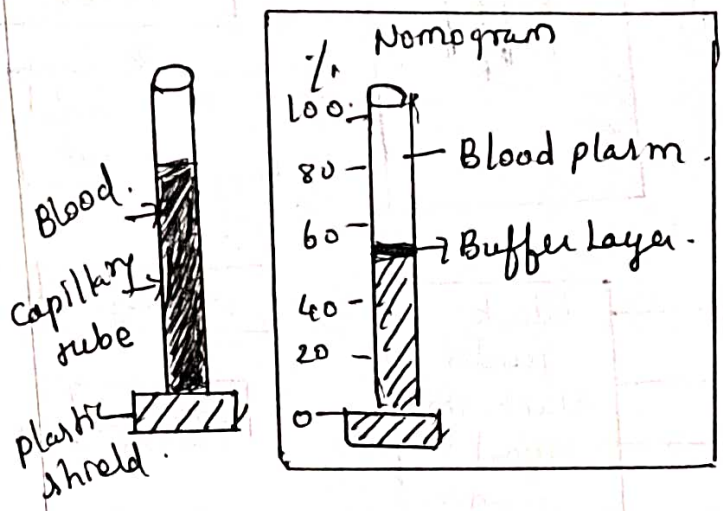
→ RBC (Red blood cell)
→ WBC (White Blood cell)
→ Blood platelets (Thrombocytes)

Red blood cells	White blood cells	Blood Platelets
→ Erythrocytes	→ Leucocytes	→ Thrombocytes.
diameter - 8 μm	diameter - 10 μm	diameter - 2 - 4 μm.
Cell nucleus is absent	cell nucleus is present	No nucleus.
1 mm^3 blood = 4.5 - 5.5 million RBC	6000 - 10000 WBC	20,000 - 80,000 No. of platelets.
Lifetime: 120 days	17 - 24 days.	
O_2 → rich → brightened O_2 → depleted → dark red.	→ help to maintain immune System	→ Blood clotting Mechanism.

Blood cell counter type:

(i) Hematocrit determination
(ii) Manual method
(iii) conductivity method
(iv) Laser based cell counter.

(i) Hematocrit determination.



→ determine the blood cell in a given volume of blood hematocrit

→ Blood sample is tracked into capillary tube & one end of tube is sealed with plastic material.

→ Blood & cell volumes are compared by measuring the length of column.

→ Nomogram gives the direct reading.

→ RBC have high electrical resistivity. settles at the bottom. thus it can be determined & extracted.

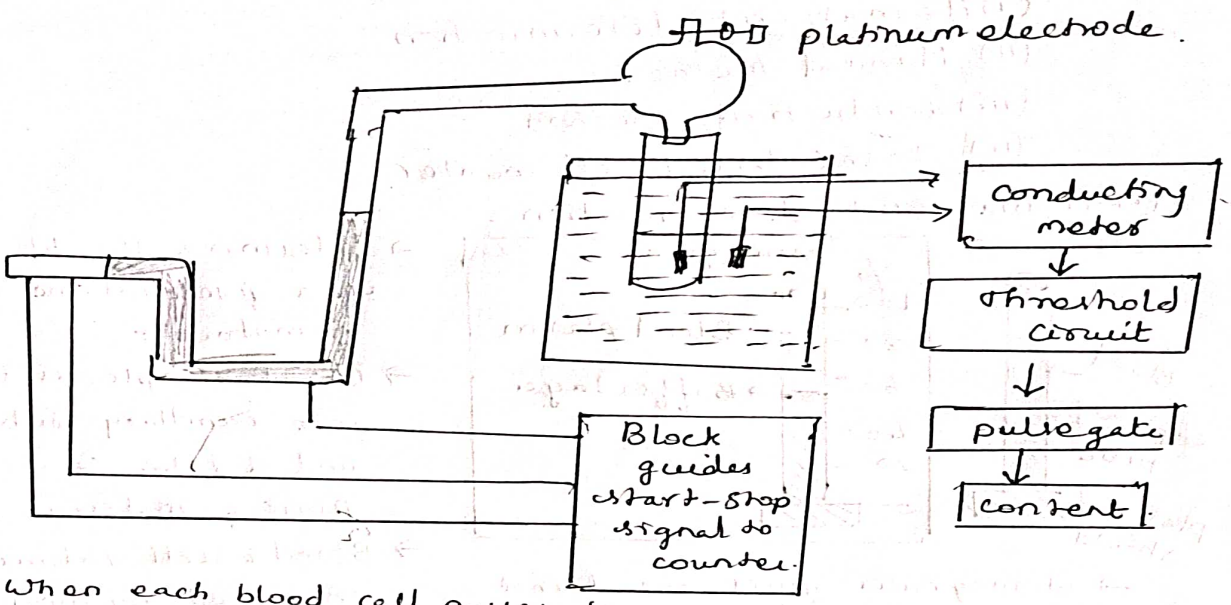
(ii) Manual method:

→ performed by microscope. At 1st the blood is diluted in the ratio of 1:100 or 1:200 for counting RBC & in the Ratio of 1:10 or 1:20 for WBC. The diluted blood is then brought to the counting chamber of 0.1mm drop which is divided into No. of squares.

→ Tedious & time consuming.

(iii) counter Method (or) conductivity method.

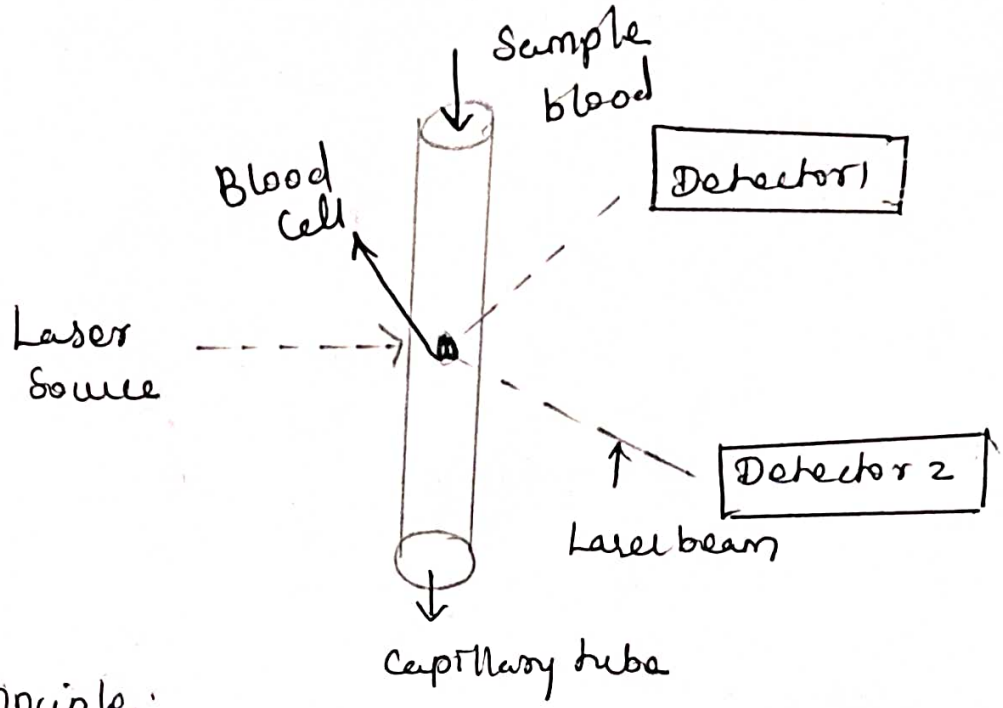
Blood cell have lower electricity then solution in which they are suspended. Here the beaker contains diluted blood solution & a closed glass tube with a very small orifice is inserted into the beaker.



→ When each blood cell passes through the orifice into glass tube it is temporarily blocks, the electrical path between the electrode. There is a drop in the conductance between the electrode. The change in the conductance gives pulse at the output of conducting meter. Amplitude of pulse & pulse of these pulse are given to the counter through the gate. Blood. The gate open when the mercury level reaches first contact point & closes when it reaches the second contact point. No. of cells present in the solution, passing through the solution can be counted. The count is completed within few seconds & care should be taken to keep the orifice from blocking. This method is used to determine the RBC, WBC counts & the haemoglobin concentration.

(iv) Laser based cell counting:

→ Modern techniques to determine the No. of RBC, WBC & platelets.



Principle:

Laser based blood cell counting is the "angle of scattered light is different for different size blood cells."

→ The blood is diluted & pulled through the capillary tube. The Laser Light is pulled through the glass tube & the blood cells in the tube scatter the light. The scattering angles of platelets & RBC are different

→ They are detected by two different photo detectors, the detectors are given the digital voltmeter which gives the density of blood cell & platelets.

→ Lysing agent is used to destroy the RBC & WBC Number can be determined.

→ The haemoglobin concentration in the RBC also be measured in this method.