DEPTH OF BIOLOGY

UNIT-V

- **Electrochemical methods of analysis**
  - **Conductometry**- Introduction, Conductivity cell, Conductometric titrations, applications.
  - **Potentiometry** - Electrochemical cell, construction and working of reference (Standard hydrogen, silver chloride electrode and calomel electrode) and indicator electrodes (metal electrodes and glass electrode), methods to determine end point of potentiometric titration and applications.
  - **Polarography** - Principle, Ilkovic equation, construction and working of dropping mercury electrode and rotating platinum electrode, applications

Polarography

- It is a electrochemical method of analysis.
- Polarography involves measurement of current-voltage curves.
- When voltage is applied to electrode (immersed in solution)
- Indicator electrode → Dropping mercury electrode
- Reference electrode → Its potential remain constant during measurement.

Polarography

- The potential of the indicator electrode varies because of change of voltage during measurement of current-voltage curves.
- When voltage is applied across the electrode ions of electrolyte get polarized and carry current gives current voltage curve.
- Magnitude of current flowing is measured by using Polarograph or Polarographic cell.

Ilkovic Equation

Ilkovic equation is a relation used in polarography relating the diffusion current (id) and the concentration of the non-polarisable electrode, i.e., the substance reduced or oxidised at the dropping mercury electrode (polarisable electrode).

$$\mathbf{i_d = 607\ nD^{1/2}\ m^{2/3}\ t^{1/6}\ C}$$

Where,

- i_d = Diffusion current in microamperes
- 607 = Constant of various numerical factors including: Faraday constant (Π),density of Hg, etc.,
- n = Number of electrons duly involved in the electrode reaction,

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- D = Diffusion coefficient in $cm^2.sec^{-1}$
- m = Weight of Hg flowing via the capillary in $mg.sec^{-1}$
- t = Drop time in seconds,
- C = Concentration in mol/L.

The Ilkovic Equation holds good for the ‘drop-time’ to vary between 2 to 8 seconds. In order to accomplish this aim and objective the following two critical adjustments may be done carefully:

 Length of capillary

 Manouvering the Hg-pressure to bring the drop time very much within the range (i.e., 2-8 sec)

Factors affecting Ilkovic Equation

1. Concentration of ion
2. Temperature :- we maintain constant temperature.
3. Viscosity :- rate of diffusion of electroactive species depends upon viscosity of med.
4. Height of Hg of column :- maintain constant

Dropping Mercury Electrode (DME)

- Also known as working electrode. Here mercury drop form continuously (comes from reservoir through capillary) interval diameter = 0.03 to 0.05
- Interval bet^{n} drop from 1-5 sec

Construction:-

1. The electrode consists of Hg reservoir
2. The capillary is connected to reservoir (Length = 10-15cm long)
3. Height of mercury reservoir is adjusted such that drop time is 1-5sec.

Working

- Dropping mercury electrode (DME) is a polarisable electrode and can act as both anode and cathode.
- The pool of mercury acts as counter electrode,

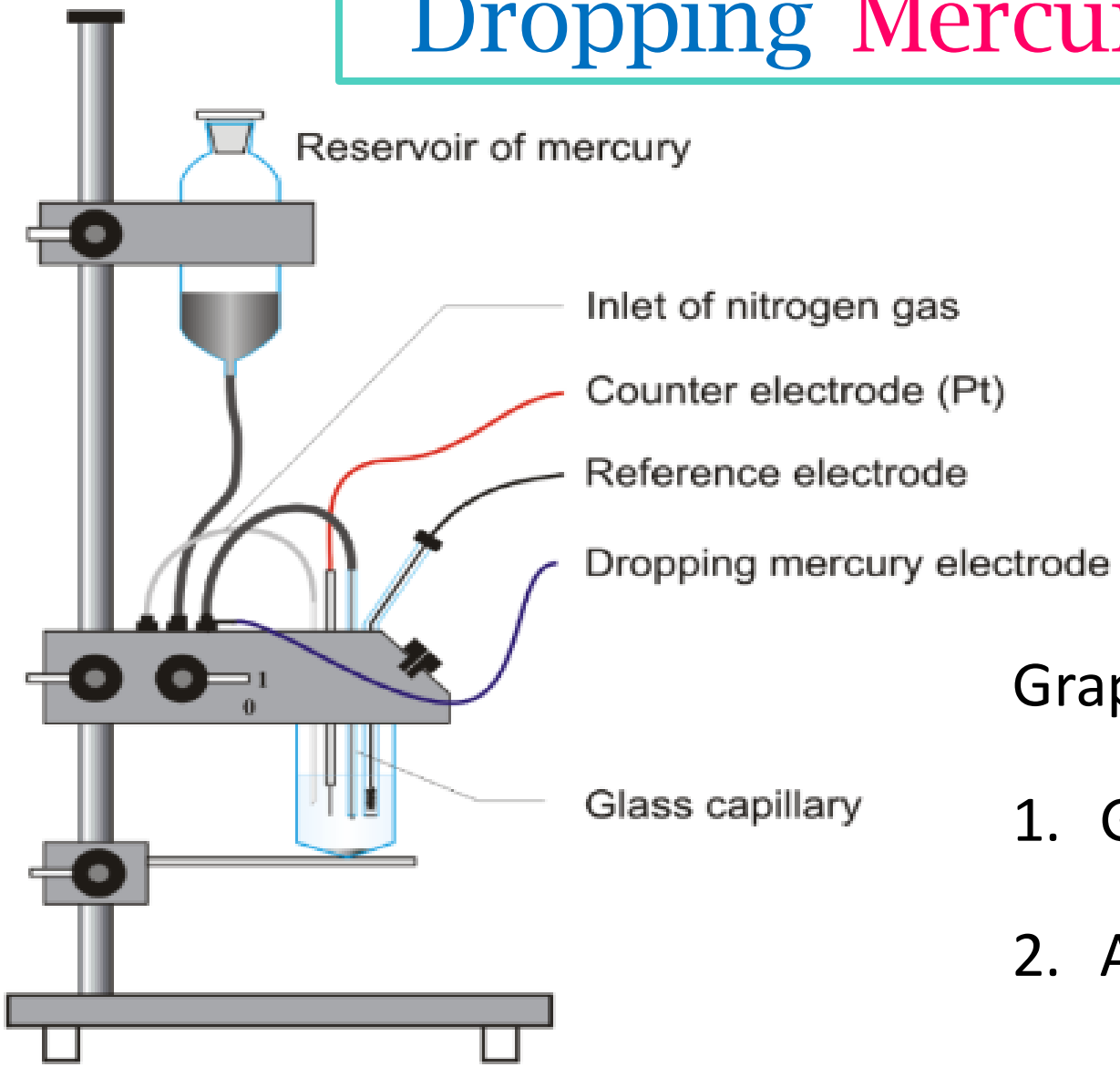
i.e., anode if DME is cathode or cathode if DME is anode.

Analyte solution + KCl electrolyte

To the analyte solution KCl is added 50-100 times of sample concentration.

We create bubbles of pure N or H to expel out oxygen.

Dropping Mercury Electrode (DME)



Graph plot between voltage applied & current.

1. Graph is called polarograph
2. Apparatus is called polarogram

Dropping Mercury Electrode (DME)

Advantage :- Surface area is reproducible [Fresh Hg drop]

- Electrode can be renewed so eliminate poisoning effect.
- Hg form amalgam with many metals.

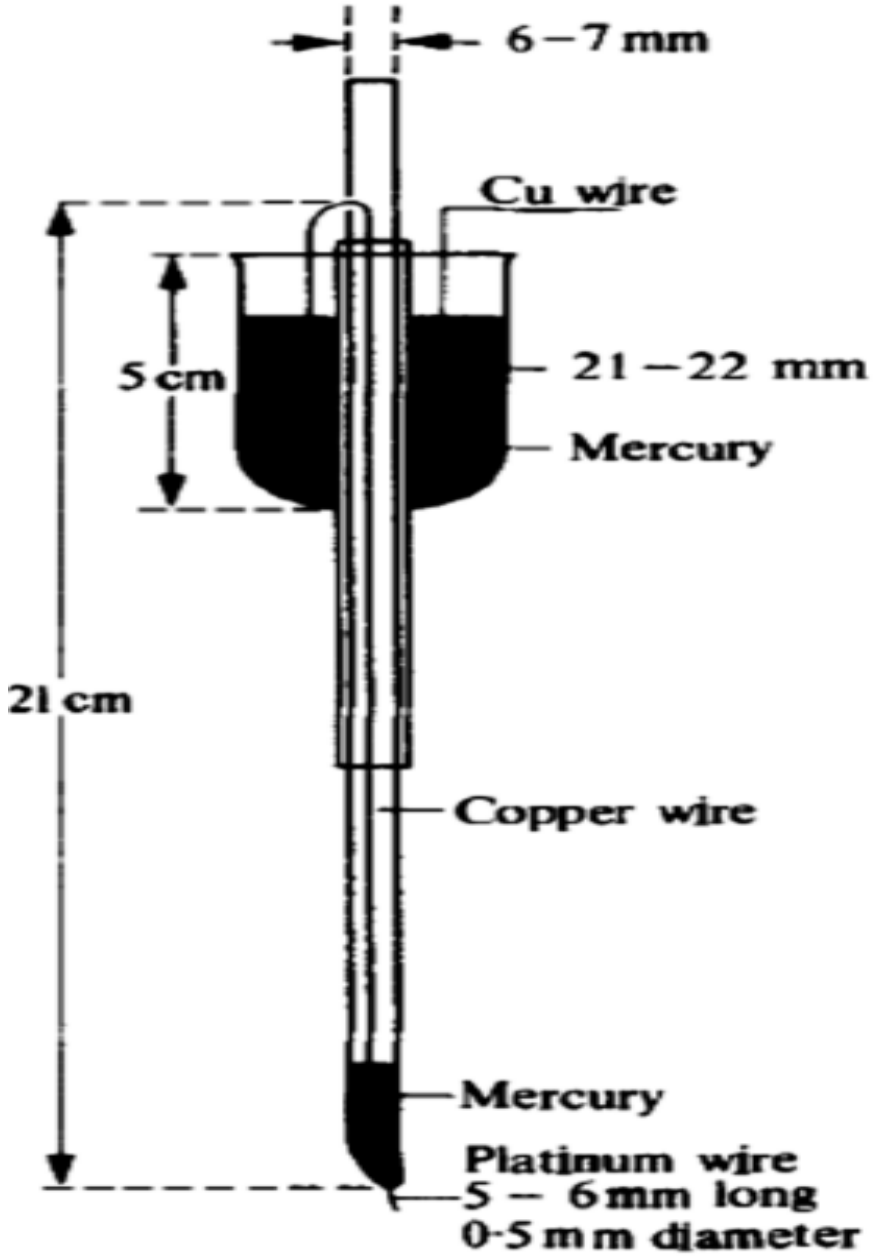
Disadvantage :- Capillary is very small → so easily blocked.

- Hg is very toxic
- Voltage applied may change surface tension → lead to change in drop size.

Rotating Platinum Electrode

Construction :-

- It consists of about 5mm point wire having 0.5mm diameter
- Below standard Hg seal by passing through small hole.
- A wire from mercury seal is connected to the source that applied potential.



Working :-

1. R.P.E is used a indicator electrode.
2. Analyte solution → KCl add [50-100 times of sample concentration]
3. Pure N_2 gas or H gas is pass to eliminate/ remove O_2
4. Potential is applied across the electrode & titration start.
5. Graph plot betn → Vol. of solution added v/s diffusion current
6. End point is detected

Application of Polarography

1. **Qualitative Analysis :-** Helps in characterisation of organic matters and various metal interaction.
2. **Quantitative Analysis :-** Used in determination of concentration of drug, metal ion etc. in given sample.
3. **Inorganic Application :-** In polarography

- Half wave potential gives identity of element.
- Diffusion quantity (element)
- In polarography used in determination of concentration of cation and anion.

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4. Organic Application :-

Polarography → used in the determination of structure of organic compound.

5. Estimation of Dissolved oxygen :- Helps in calculation of oxygen dissolved in aqueous solution/ organic solvent. (amount of O2 is estimated)

6. Pharmaceutical Application :- Estimation of trace element in pharmaceutical

7. Alkaloid like quinine, quinidine, morphine can be determined.

8. Analysis of volatile oil constituent.

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9. Fat soluble and H_2O soluble Vitamin undergo electro oxidation/ reduction. So, estimated by polarography.
10. Hormone like Insulin, Sex hormone, Thyroxine can be determined by Polarography.
11. Antibiotics

a. Penicillin

b. Streptomycin

These can be determined by Polarography.