

GAS liquid CHROMATOGRAPHY (GLC)

Dr. Rashmi. R
Asst. Professor
PG Dept. of Biochemistry

- ▶ In this technique, **components of a vapourized sample are** separated on the basis of their **partitioning between** mobile phase and stationary phase.
- ▶ In this case, **a gas is used as mobile phase**, therefore, it is termed as gas chromatography.

I. Principle

*Gas chromatography is based on the principle of **partition of a volatile compound** between a liquid/solid stationary phase and a gaseous mobile phase in a fixed set of parameters.*

*In gas chromatography, sample is a gas or in vapour phase, **eluent (M.P) is a gas and stationary phase is non-volatile liquid coated on a solid surface or a solid.***

The analyte in vapor phase distributes between the stationary and mobile phase establishing equilibrium between the two phases.

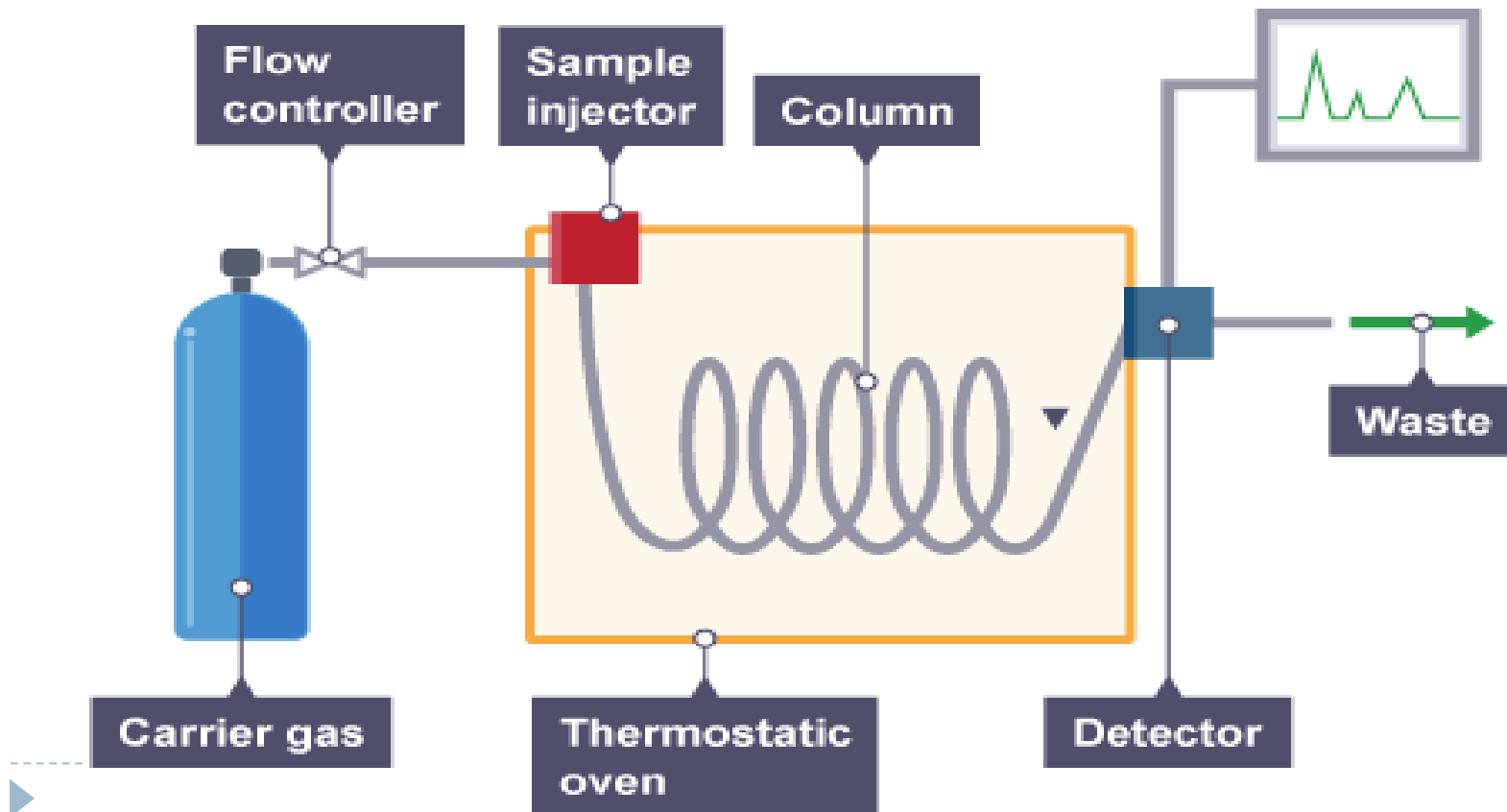
The separation mechanism is based on partitioning—components of the vaporized sample dissolve into and out of the liquid stationary phase based on their individual solubilities and polarities.

- ▶ The components distribute between the gaseous mobile phase and the liquid stationary phase and establish equilibrium.
- ▶ **This equilibrium is very rapid** and therefore, GLC has been adopted as the most useful method. It is generally termed as **Gas Chromatography (GC)**.



2. Instrumentation of GLC-PROCEDURE

- ▶ The main components of GC are **carrier gas**, **sample injection port**, **column**, **thermostatic oven** and **detector**.



3.1 Carrier gas

- ▶ **The mobile phase used in gas chromatography (i.e. gas) is called as carrier gas.**
 - ▶ It delivers the sample to the column and then, to the detector.
 - ▶ The **carrier gas must be chemically inert.** Therefore, the gases like **Argon, helium, nitrogen and carbon dioxide are the most commonly used carrier gases.**
 - ▶
 - ▶ The gases are available in pressurized tanks, therefore, their release is controlled by pressure regulators.
-



3.2 Sample injection system

- ▶ This component is used to apply a sample into the system via injection port.
- ▶ The sample is injected by a microsyringe directly into a flash vapourizer port.



3.3 Column

- ▶ Column is filled with the stationary phase, so equilibration between stationary phase and mobile phase takes place in the column.
 - ▶ The column used in GC should be rigid and able to withstand moderate pressures (up to 50 psi).
 - ▶ It should be stable at high temperature and should be chemically un-reactive.
 - ▶ These are generally made up of stainless steel, fused silica or Teflon.
-



In gas-liquid chromatography (GLC), the **stationary phase** is a high-boiling-point, non-volatile liquid film coated onto either an inert solid support packed inside a column or directly onto the inner wall of a capillary column.

Examples of stationary phases:

- ▶ Silicone oils
- ▶ Polyethylene glycol (PEG)
- ▶ Greases
- ▶ Cyanopropyl silicone phases- Can be used as stationary phases
- ▶ The mobile phase comes in contact with the liquid stationary phase and the components undergo equilibrium.



3.4 Column thermostat oven

- ▶ The column thermostat oven is **used to control the column's temperature.**
- ▶ In case of isothermal separation, column is maintained at a constant temperature.
- ▶ The temperature chosen for the separation should be below the boiling point of the lowest boiling solute.
- ▶ In case of temperature programming, **the temperature of the column is increased slowly either uniformly or in a series of steps.**



3.5 Detection system

- ▶ **The separation of components is detected with the help of a detector.** The mobile phase coming out from the column is directly delivered to the detector, where the components are detected on the basis of their physical properties.

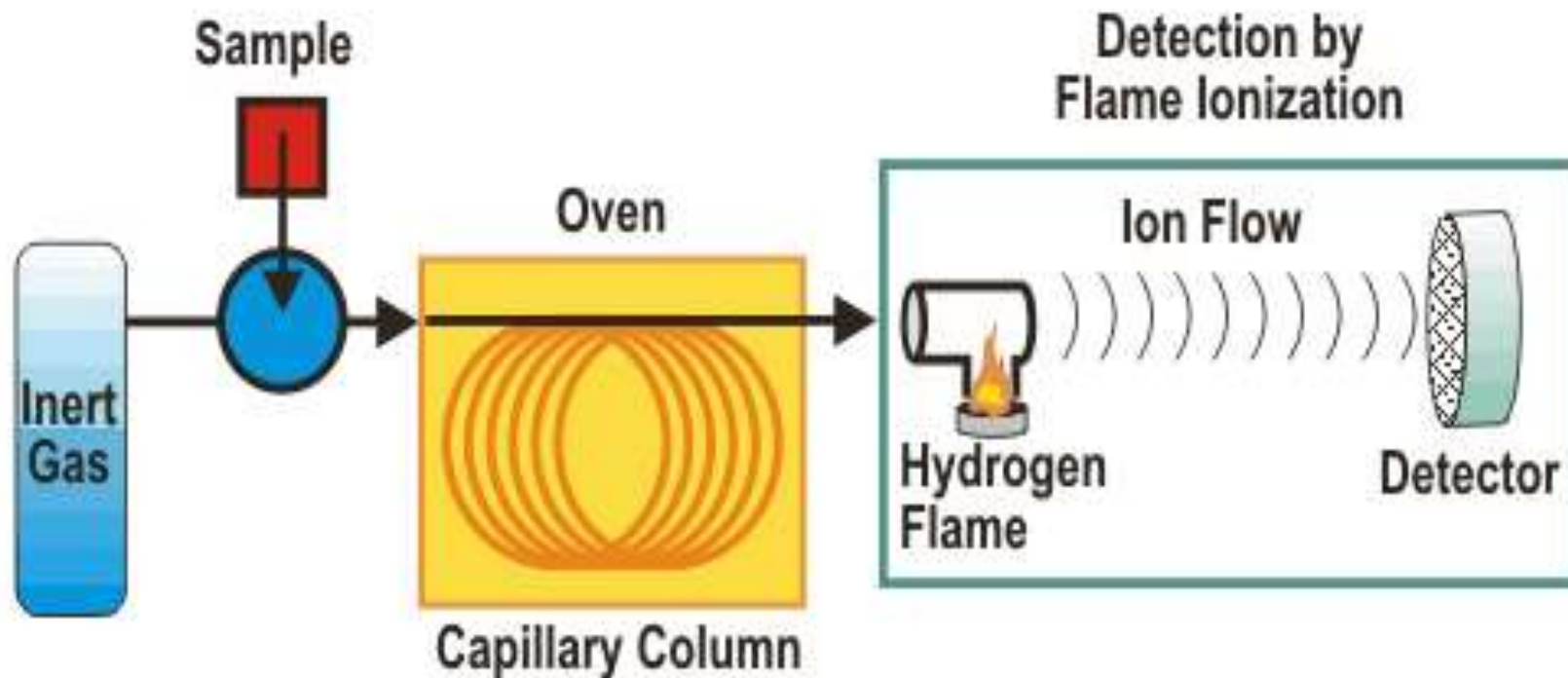


3.5.1. Flame ionization detector

- ▶ The sample coming out from the column is delivered to the detector.
- ▶ In case of FID, **the sample enters into the small air/hydrogen flame.**
- ▶ **The components present in the sample are burnt in the flame and produce ions and electrons.**
- ▶ **The ions are detected with the help of a metal collector biased with high DC voltage.**
- ▶ **The detector monitors the current produced by the charge carrying ions, which is proportional to the rate of ionization and in turn mass of the ion producing component in the sample.**



Thermal conductivity detector



Advantages and disadvantages of GC

Advantages	Disadvantages
This method provides a high resolution power.	It can be used for volatile samples, therefore, an additional step is required to make the sample volatile for non-volatile samples It deals with the gases at very high pressure, therefore, careful handling is required The compounds should be thermally stable, therefore, sometimes data due to thermally unstable components is lost under GC conditions
It is highly sensitive.	
It provides good accuracy and precision.	
The technique is fast.	
It can work with fewer amounts of the components.	



I. **Forensic Science:**

- ▶ Drug Detection: GLC, often combined with mass spectrometry (GC/MS), is used to identify illicit substances in bodily fluids and other samples.
- ▶ Fire Investigation: GLC can analyze volatile compounds at fire scenes to determine the cause and origin of the fire.
- ▶ Explosives Investigation: GLC can detect traces of explosive residues in crime scenes.
- ▶ **Environmental Analysis:** GLC helps in identifying and quantifying pollutants in water, soil, and air samples.
- ▶ **Pesticide Analysis:** GLC is used to detect and quantify pesticides in food, water, and soil samples.
- ▶ **Pollutant Identification:** GLC helps identify and quantify various pollutants in environmental samples.



▶ **3. Pharmaceutical Industry:**

- ▶ Drug Analysis: GLC is used to analyze the purity and composition of pharmaceutical drugs.
- ▶ Metabolite Identification: GLC can be used to identify metabolites in biological fluids.

▶ **4. Food Industry:**

- ▶ Food Quality Control: GLC helps in the quality control of food products by identifying and quantifying various components, additives, and contaminants.
- ▶ Flavor and Aroma Analysis: GLC is used to identify and quantify the volatile compounds responsible for the flavor and aroma of food products.



Thank YOU

