Gas chromatography

It is a process of separating component(s) from the given crude drug by using a gaseous mobile phase.

- It involves a sample being vaporized and injected onto the head of the chromatographic column.
- The sample is transported through the column by the flow of inert, gaseous mobile phase.
- The column itself contains a liquid stationary phase which is adsorbed onto the surface of an inert solid.

Principles:

- The principle of separation in GC is "partition." The mixture of component to be separated is converted to vapour and mixed with gaseous mobile phase.
- The component which is more soluble in stationary phase travel slower and eluted later. The component which is less soluble in stationary phase travels faster and eluted out first.
- No two components has same partition coefficient conditions. So the components are separated according to their partition coefficient.
- Partition coefficient is "the ratio of solubility of a substance distributed between two immiscible liquids at a constant temperature.'

Instrumentation

- Carrier gas - He (common), N_2, H_2, Ar
- Sample injection port- micro syringe
- Columns
- Detectors
- Add-ons : Mass Spectroscopy (MS)

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Types of Gas Chromatography

1) Gas liquid Chromatography (GLC): When stationary phase is non volatile liquid coated on inner support and the mobile phase is gas, then the technique is called as gas liquid chromatography GLC is a partition type of chromatography

2) Gas Solid Chromatography (GSC): When stationary phase is solid of a large surface area and the mobile phase is gas, then the technique is called as gas solid chromatography Silica gel, alumina, charcoal, molecular sieve etc are used as a stationary phase.

Instrumentation

Carrier Gas

Inert gas like argon, helium, nitrogen may be used as carrier gas. Hydrogen gas is less preferred because of its explosion hazards. Selection of Selection of carrier gas depends on the nature of the mixture to be separated, purity required and detector used for the analysis. The main purpose of the gas in GC is to move the solutes along the column. Hence; mobile phase is often referred to as carrier gas.

Carrier gas should be

1) Inert 2) Suitable for detector 3) Readily available 4) Have good flow rate 5) Free from fire and explosion hazard

Injection port

Liquid sample is injected by means of a calibrated micro syringe. The sample is injected through a rubber septum at the head of the column. At the temperature of the injection port liquid sample is readily converted to vapors without decomposition.

Chromatographic Column

This is the backbone of chromatography. Column is made up of stainless still or glass and is 2 to 3 meter long and have internal diameter 2 to 4 mm.

Types of column

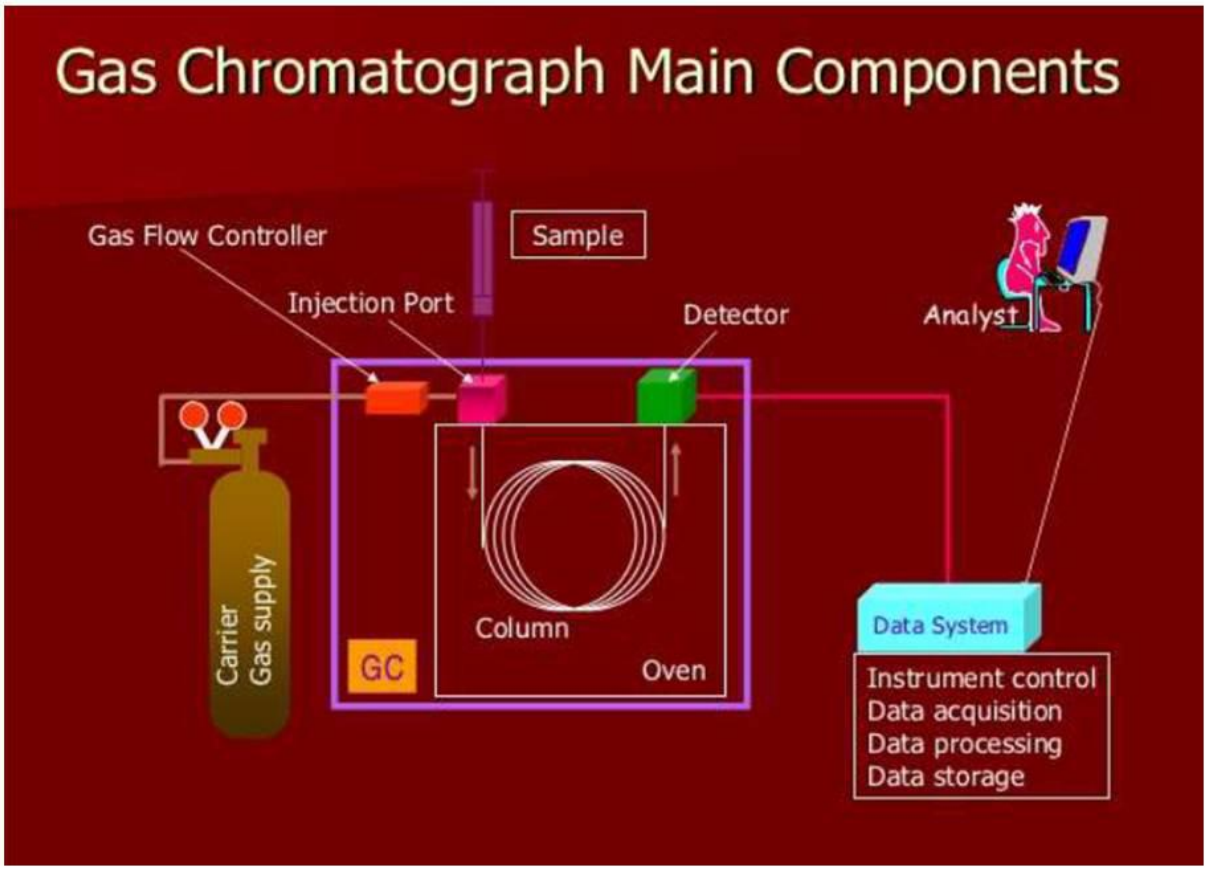
a) Packed column: It is made up of Teflon having internal diameter 2 to 4 mm and length 5 meter. Column is packed with finely divided solid as absorbent in gas solid chromatography.

b) Capillary column: These columns are 15 meters to 100 meter long and have internal diameter less than 1 mm (i.e., 0.25 to 0.30 mm). This column does not contain packing but contain stationary phase coated on their inner wall.

Detectors used in gas chromatography

Most commonly used detectors

1) Thermal conductivity detector (2) Flame ionization detector (3) Electron capture detector



Working of Gas Chromatography

1) When sample is solid it is converted to liquid form by dissolving in suitable solvent

2) Sample is injected on hot metal block placed at injection system

3) The sample, injection port, column and detector are heated to suitable temperature to prevent condensation of sample

4) The separated components coming out along with mobile gas phase are directly passed in to the detector system for their analysis

5) Detector response is recorded in the form of chromatogram.

Chromatogram

Plot of the detector response against time is called chromatogram Number of peaks represents the number of components presents in the sample (Separation of component is based on their partition coefficient The separated component exit along with the mobile phase at the end of the column These components are then passed through the detector and detector give response to read out device Magnitude of the response depends upon the concentration of the component.

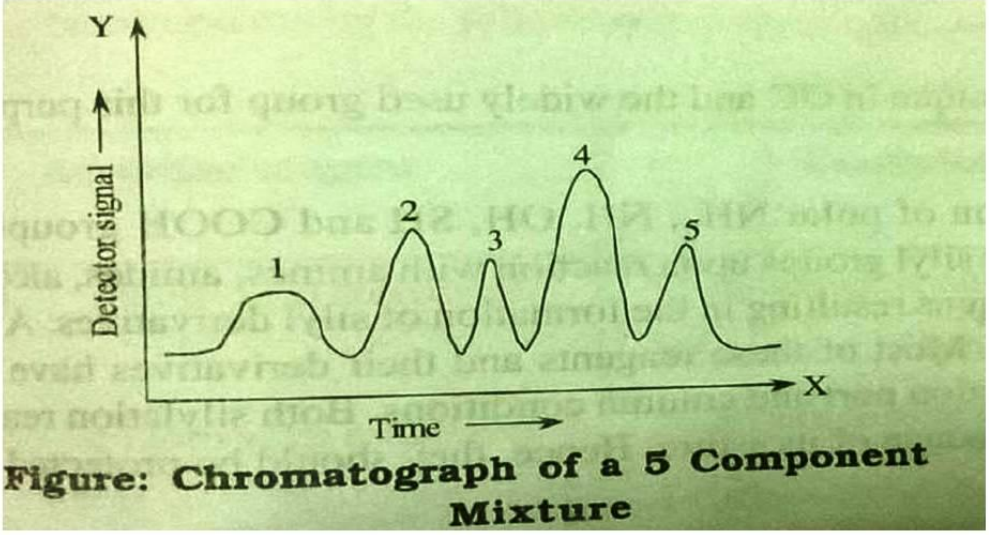


Figure: Chromatograph of a 5 Component Mixture

Area under the peak is proportional to the concentration of component

Applications:-

- Qualitative Analysis – by comparing the retention time or volume of the sample to the standard / by collecting the individual components as they emerge from the chromatograph and identifying these compounds by other methods like UV, IR, NMR.
- Quantitative Analysis- area under a single component elution peak is proportional to the quantity of the detected component/response factor of the detectors.

(Check class notes for the facts not included in this note.)