

Chromatography is a separation technique in which a mobile phase carrying a mixture moves in contact with a selectively adsorbent stationary phase.

Introduction

In 1903 a Russian botanist by the name of Mikhail Tswett first applied this technique to separate coloured plant pigments. He passed a solution of these compounds through a glass column packed with finely divided Calcium Carbonate.

Modern chromatography encompasses a diverse group of separation methods, all of which are based on components partitioning between a *stationary phase* and a *mobile phase*. The mobile phase carries the sample components through the stationary phase.

Adsorption involves interactions at a surface on a normally solid stationary phase. Do not mix up the word absorption with adsorption. Think of adsorption as a 'sticking' process.

Paper Chromatography

- Stationary phase = filter paper
- Mobile phase = solution e.g. methanol + food colouring/felt-tipped markers/mixture of indicators

Thin Layer Chromatography(TLC)

- Stationary Phase = aluminium oxide/silica gel supported on glass plate
- Mobile Phase = solution e.g. methanol+ food dyes

This is a superior technique to paper Chromatography because the component mixture separate more definitively. This is due to the fact that TLC plate contains smaller particles than the chromatography paper.

Often the components to be separated are not coloured so the sample is sprayed with a UV absorbing chemical and the sample is analysed under UV light.

Column Chromatography

- Stationary phase= long glass tube containing solid adsorbing phase e.g. Silica gel dissolved in the suitable solvent
 - Mobile Phase= solution to be separated added to top of column and passes through column, its components travel at different rates through the column and they can be collected. These collected samples are called the eluted samples.
- The above Chromatographic techniques can be used to separate the colours in
1. food dyes
 2. Felt-tipped markers
 3. Mixture of indicators in Universal indicator
 4. Dyes in flowers

Gas Chromatography

- Stationary Phase =high boiling point liquid spread on solid particles
- Mobile Phase = carrier gas

How does it work?

See diagram in textbook

The sample to be analysed is vapourised as it is injected into column. The components in sample have different solubilities with the stationary phase.

This technique is used to analyse the level of alcohol or other drugs in blood or urine samples.

Can you remember how the presence of alcohol in the blood is initially recognised?

Mass Spectrometer

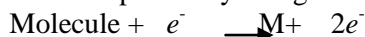
See page 32 Fig 4.12

Step 1. Vapourisation

In a mass spectrometer, molecules in the gaseous state under low pressure are bombarded with a beam of high-energy electrons.

Step 2. Ionisation

Due to the energy of this beam of electrons it removes one of the electrons of the molecule to be analysed and this molecule becomes positively charged.



M⁺ known as a free radical or radical cation

Step 3. Acceleration

The cations are then accelerated through an ionisation chamber.

Step 4. Separation

The mass spectrometer then sorts out the cations on the basis of their mass by applying a magnetic field. The lighter molecules are easier to deflect

Step 5. Detection

Mass spectrometer is used to identify

- Isotopes
- Abundance of isotopes
- Relative atomic masses
- Identify unknown compounds

GC-MS

Gas Chromatography-Mass Chromatography

The components separated by Gas Chromatography are then analysed using Mass chromatography. A mass spectrum is obtained and each individual compound can be analysed and identified by comparing the spectra from a database of spectra. This powerful and thorough technique is used in the investigation of various drugs by forensic scientists.

GC-MS can be used to detect the gases from waste dumps and organic pollutants in water.

HPLC

Chromatography in which a pump is used to force a liquid through a column is called HPLC.

See fig. 23.52

HPLC

Stationary phase= solid

Mobile phase = liquid

HPLC is used more than GC because:

- Separation of mixtures is more efficient
- HPLC is carried out at lower temperatures than GC therefore compounds that would decompose at high temps using GC can be analysed.

HPLC is used in analysing

- Growth promoters in meat
- Vitamins in food
- Concentration of caffeine in Coca Cola

Spectroscopy

Spectroscopy is the study of the interaction of electromagnetic waves and matter.

Spectrometers are used by chemists to analyse compounds.

Infra-Red Spectrometers

IR radiation has a longer wavelength than visible light. This radiation is invisible but is given off by hot objects. The human body gives off IR radiation. Often infra-red cameras are used to find people trapped in collapsed buildings.

Organic compounds absorb IR radiation. Each functional group will absorb IR radiation at different frequencies, thus every organic compound has a unique IR spectrum.

See Fig 23.54 IR spectrum of Polystyrene

UV Spectrometers

UV light has a wavelength shorter than visible light.

The main source of this radiation comes from the sun. Certain compounds absorb UV light, e.g. washing powders contain certain chemicals that absorb UV light and emit visible light. These compounds are called optical whiteners. UV spectroscopy is used in identifying compounds and their concentrations.

A universal indicator is typically composed of water, [propan-1-ol](#), [phenolphthalein](#) sodium salt, sodium hydroxide, [methyl red](#), [bromothymol blue](#) monosodium salt, and [thymol blue](#) monosodium salt.