Chromatography

- **Chromatography** (from Greek χρῶμα *chroma* "color" and γράφειν *graphein* "to write") is the collective term for a set of laboratory techniques for the separation of mixtures.

- Thin Layer Chromatography (TLC)
- Column Chromatography
- Gas Chromatography (GC)
- High Pressure Liquid Chromatography (HPLC)

Thin Layer Chromatography

Please read Unit 8.

Purpose

Thin layer chromatography (TLC) is a chromatography technique used to separate mixtures and identify unknown organic compounds. Thin layer chromatography is performed on a sheet of glass, plastic, or aluminum foil, which is coated with a thin layer of adsorbent material, usually silica gel. This layer of adsorbent is known as the stationary phase.

Principle

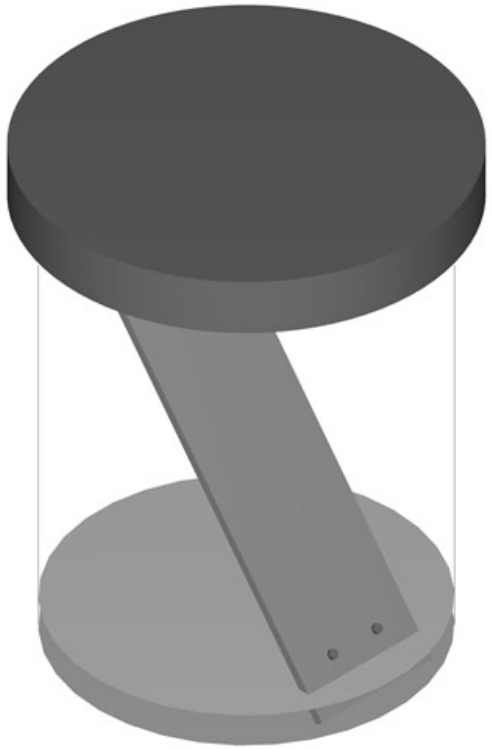
- Solid-liquid technique
  - Stationary phase – solid (silica gel)
  - Mobile phase – liquid (ethyl acetate)

- Competition between stationary and mobile phase in their attraction for organic compounds.
  - stationary phase is very polar
  - the more polar the compound, the more it interacts with the stationary phase
    - more interaction = shorter distance traveled

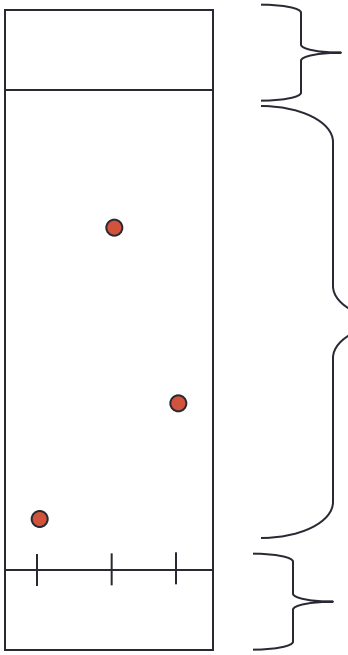
1. A line is drawn approximately one cm from the bottom of the silica plate. A small amount of each sample is placed on the line.

2. A small amount of the solvent is placed in the bottom of a beaker and the silica plate is placed inside. A watch glass is placed on the top and the solvent is allowed to travel up the strip.

3. When the solvent reaches about 1 cm from the top of the strip, the strip is removed from the chamber and allowed to dry. Using your pencil, mark the level reached by the solvent on the strip.



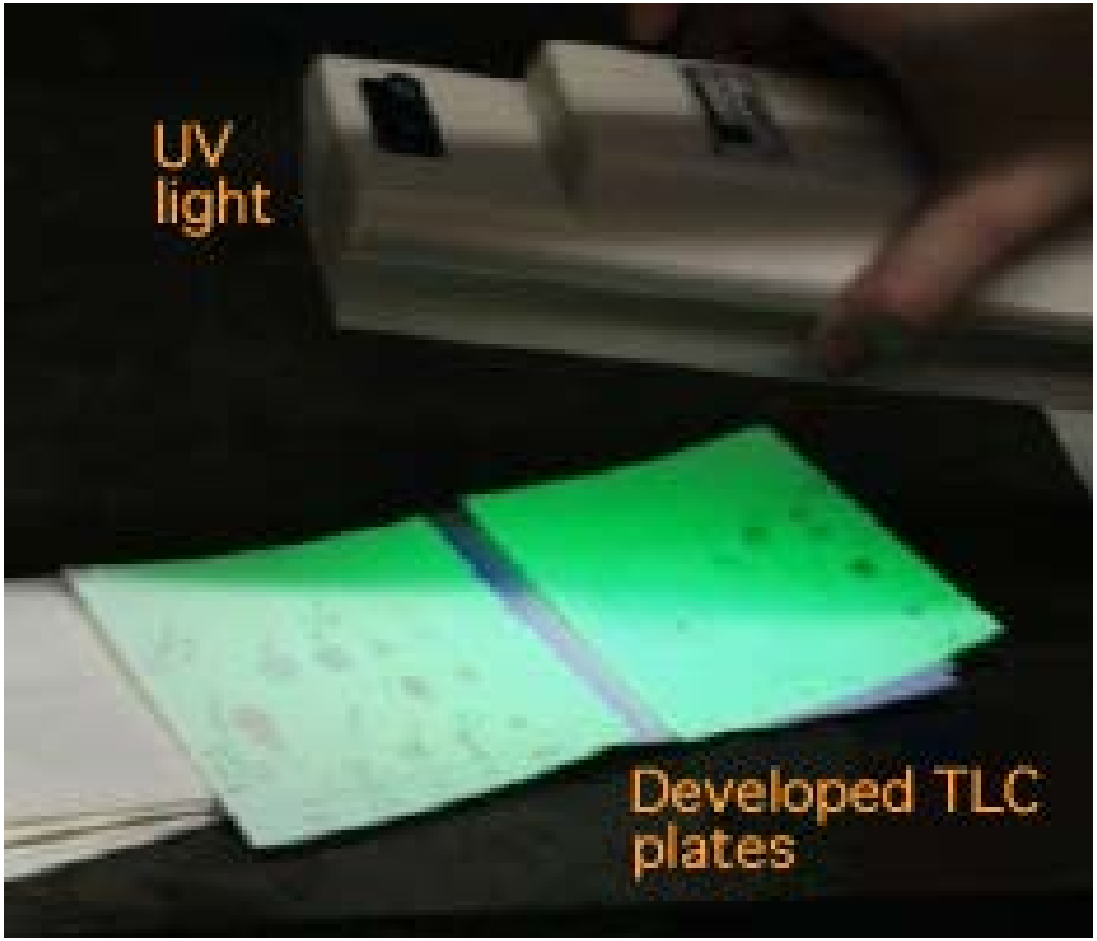
TLC plate and sample spots



Mark solvent front with pencil

When dry, place the plates under a UV lamp to see how far the spots have moved. VERY CAREFULLY draw a circle around each with your pencil

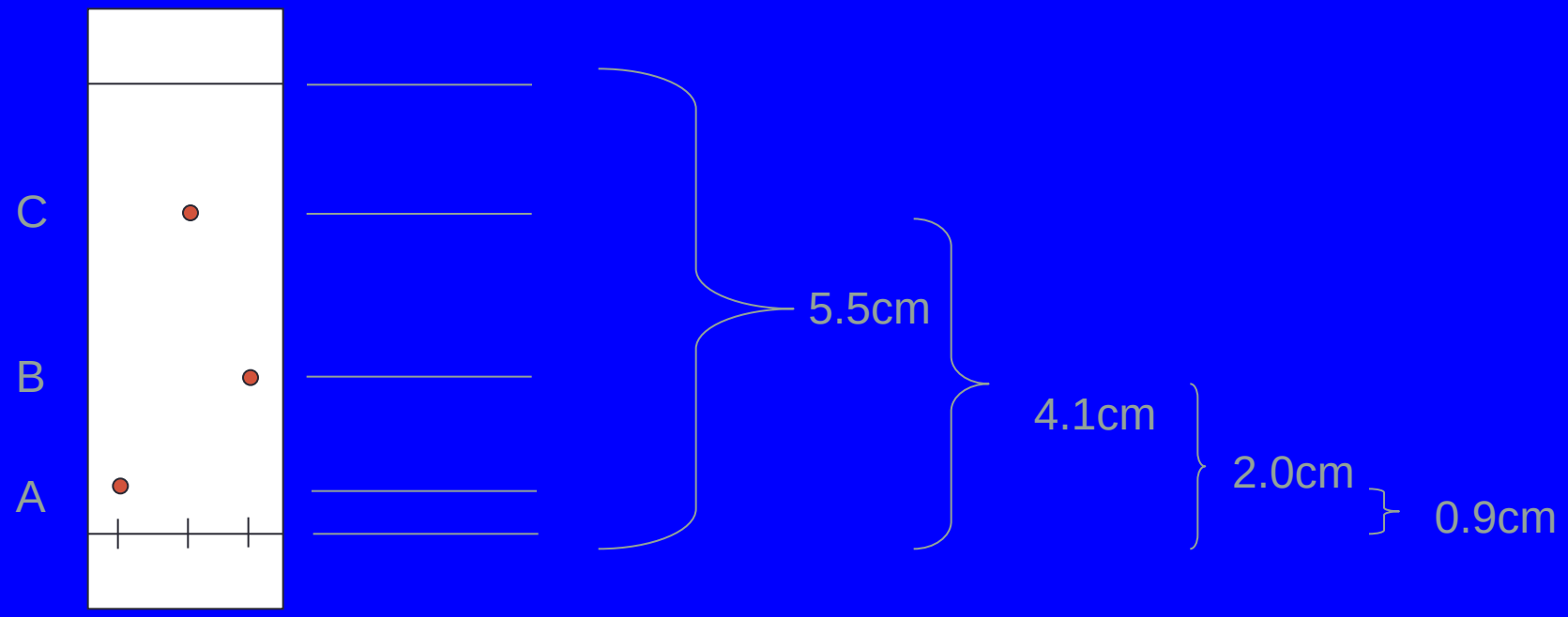
Pencil a line ~1 cm from bottom


UV
light
Developed TLC
plates

Using the Rf value to identify unknowns

- Under a definite set of experimental conditions, a given compound will always travel a fixed distance relative to the distance traveled by the solvent front (aka Rf).
- Therefore, Rf value can allow us to identify a unknown compound.

Rf Value Calculation



$$Rf = \frac{\text{distance traveled by compound}}{\text{distance traveled by solvent front}}$$