

## Chromatography of Inks

### Introduction:

One of the main jobs of biochemists is to unravel the complexities of chemical compounds and reduce them to their individual components. The term chromatography comes from two Greek words, "chromate" meaning color and the word "graphon" meaning to write. Separation of the components of chemical compounds can be done by using several methods. Liquids can be separated by High Performance liquid Chromatography (HPLC), while the components of gases are separated by Gas Chromatography. Chromatography is a method for analyzing complex mixtures (such as ink) by separating them into the chemicals from which they are made. Chromatography is used to separate and identify all sorts of substances in police work. Drugs from narcotics to aspirin can be identified in urine and blood samples, often with the aid of chromatography.

Chromatography was first used to separate pigments (colors) in leaves, berries, and natural dyes. Paper chromatography is a technique used to separate, isolate, and identify chemical components of a compound. In paper chromatography, the solid surface is the cellulose fibers in the chromatography paper. A solvent or developer (water, alcohol, or acetone) is placed in the bottom of the chromatography chamber. The paper acts as a wick to pull the solvent up the paper. The solvent front will "wick" up the chromatography paper by capillary action. A minute drop of the ink or chemical mixture to be separated is placed near the bottom of the strip of chromatography paper, but slightly above the level of the solvent in the chamber. As the solvent passes over the drop of ink, the components of the ink dissolve in the solvent. Because the components of the ink do not all dissolve at the same rate, as the components of the mixture move upward, they show up as colored streaks. The separated substances on the chromatography paper form a color pattern called a chromatogram.

To determine the rate of migration for each pigment or component of the ink, the  $R_f$  value for each pigment must be calculated. The  $R_f$  value represents the ratio of the distance a pigment moved on the chromatogram relative to the distance the solvent front moved. Each pigment or compound will have a unique  $R_f$  value that scientists can use to identify the substance. The  $R_f$  value is calculated using the following formula:

$$R_f = \text{distance traveled by the compound} / \text{distance traveled by the solvent}$$

### Objective:

Use the process of paper chromatography to separate the pigments in various markers and then determine the  $R_f$  value for each color on your chromatogram.

**Materials:** The materials used in this experiment were:

1. Plastic vials
2. paper clips
3. markers in assorted colors
4. chromatography paper
5. scissors
6. pencil

**Procedure:** The procedures used in this experiment were:

1. Obtain chromatography vials and chromatography strips, and different color markers so that each person in the group will have two chromatograms.
2. Cut one end of the chromatography strip to a point. The bottom of the point will mark the starting point for movement of the solvent (H<sub>2</sub>O).
3. About 2.0 centimeters from the bottom of the strip, draw a faint horizontal line with pencil. This will mark the starting point for measuring the migration distance of each color.
4. Using a different color marker for each strip, drop a dot of ink on the center of the horizontal pencil line. Let this dry a moment & then add more ink to the dot.
5. Add a small amount of water to the bottom of the chromatography chamber. (The ink dot should be ABOVE the surface of the water.)
6. Straighten a paper clip and poke a hole through the top of your chromatography strip
7. Use the paper clip to hang the strip in your chamber. (The straighten paper clip will lay across the top of the chamber.)
8. MAKE SURE THE TIP OF THE STRIP BUT NOT THE INK IS IMMERSSED IN THE WATER!
9. Notice the separation of the ink as both the solvent and ink travel up the chromatography strip.
10. Once the solvent front has neared the top of the strip, remove the strip from the chamber and lay it on a piece of paper towel.
11. Immediately mark the solvent front with a faint pencil line.
12. Immediately mark the leading edge of each color with an "x".
13. Measure, in millimeters, the distance the solvent migrated from the tip of the strip to your solvent front pencil line.
14. Measure, in millimeters, the distance each color migrated from the point of origin (pencil line where the ink dot was placed) to the leading edge of the color (marked with an "x").
15. Record all data in Data table 1.
16. Calculate and record the R<sub>f</sub> value for each color using the formula below.

$$R_f = \text{distance traveled by the compound} / \text{distance traveled by the solvent}$$



**Analysis:**

1. Which color of marker did you use?
2. Which color separated out first from your ink dot?
3. Why did the inks separate?
4. What was your solvent?
5. If you had used markers that weren't water-soluble, how would you have had to change this lab?
6. Why did some inks move a greater distance than others?
7. How do scientists use paper chromatography in their investigations?