

# Crime Scene Investigation Lab: Identification of Inks in Felt Tip Pens Using Paper Chromatography



## Background

**A crime has been committed!**

Baby Blue Ivy has been kidnapped! At the scene of the crime was a handwritten ransom note to Beyonce and Jay Z. In it the kidnapper demands \$10 million dollars or

**Baby Blue Ivy will be looking blue forever!**

The celebrities believe that a disgruntled former employee is behind the kidnapping.

Due to your well known expertise in such matters, they have hired your services to find out who the culprit is. You sneak into the three suspects' offices late at night. You then 'borrow' pens/markers from these three suspects' desks. You will compare the ink in these markers to that of the ransom-note's. You will use paper chromatography to determine who wrote the note.

## Theory

During its development over the last 40 years, chromatography has revolutionized modern analytical chemistry. Chromatography is a group of techniques used to separate colored mixtures into their component parts. Many types of chromatography are now routinely used in laboratories around the world to separate and identify components in mixtures. The analyses of blood and urine samples for drugs and the analyses of drinking and ground water for hazardous chemicals are two common procedures involving chromatographic separations.

All chromatography techniques involve a **stationary phase** and a **mobile phase**. The stationary phase can be either liquid or solid. The mixture to be separated is usually placed on the stationary phase. The mobile phase can be either liquid or gas. The mobile phase moves along the stationary phase, carrying some or all of the mixture with it, resulting in the separation of the mixture components.

In liquid chromatography, separation is based on the preferential attraction of each component in the mixture to either the mobile or stationary phase. The attraction is due to particular intermolecular interactions.

Paper chromatography is the simplest form of chromatography. Although paper chromatography is not used for drug or hazardous chemical analyses, it is extremely useful for separation and identification of food dyes. In paper chromatography, a sample of the mixture to be separated is placed on a piece of chromatography paper, which acts as the stationary phase. One edge of the paper is placed in a solvent, such as water, alcohol, or a mixture of both, which acts as the mobile phase. Many different solvent systems are possible, depending on the components to be separated.

The chromatography paper acts like a wick, drawing the solvent up the paper by capillary action. The wicking occurs because the solvent is attracted to water molecules that are permanently bound to cellulose fibers of the paper. The water molecules bound to the paper and the paper itself forms the stationary phase.

A sample is applied, or spotted, on an area of the paper near the bottom edge, known as the origin line. The bottom edge of the paper is then placed in a solvent, and solvent is drawn up the paper. When the leading edge of the mobile phase, the solvent front, reaches the sample, the sample components are preferentially attracted to either the stationary or mobile phase.

This attraction depends on the relative polarities of the sample components and the two phases. Recall that like solvents dissolve like solutes. However, the attraction is seldom an all-or-nothing situation. Most compounds, whether they are ionic or molecular, are somewhat attracted to both phases. Equilibrium is established for each component between the two phases, as shown by Equation 1.



As the solvent front moves up the paper, fresh solvent passes the spotted sample, and new equilibria are continually established. At the same time, any components that have dissolved in the mobile phase encounter fresh stationary phase, and new equilibria are established. Thus, the components of a mixture move up the paper at different rates and separate, producing a pattern called a chromatogram.

The overall effect of these equilibria is that the movement of the components depends directly on their relative attractions for the mobile and stationary phases. We characterize this movement in terms of a retention factor ( $R_f$ ) defined by

Equation 2.

$$R_f = \frac{\text{distance traveled by component, cm}}{\text{distance traveled by solvent front, cm}} \quad (\text{Eq. 2})$$

If a component moves with the solvent front, its  $R_f$  value can be as high as 1.0. If the component does not move at all, its  $R_f$  value can be as low as 0.0. The  $R_f$  value for a component is reproducible for a particular component-solvent system, if the experimental conditions are closely controlled.

One important variable is the composition of the solvent. If one of the solvent components is volatile, the percent composition of the solvent may change during the analysis, due to evaporation. This will affect the results. Covering the chromatogram developing container so that the air in the container remains saturated with solvent vapor can prevent evaporation.

A sample containing two or more components can be separated, or resolved, if we choose a solvent system for which the sample components have distinctly different  $R_f$  values. In order to choose the most appropriate solvent, we first determine the  $R_f$  values of the individual sample components in a variety of solvent systems. Then we choose the best solvent system for separating all of the sample components.

Figure 1 illustrates the preparation of a chromatogram. Spots of the sample to be resolved are placed on the origin line of the chromatography paper and the bottom edge of the paper is placed in the solvent. Solvent moves up the paper separating the components. The distance a component has moved is determined by measuring the distance from the origin line to the center of the component spot.

In Figure 1(c), the left-hand spot is the sample being resolved and the right-hand spot is Compound A, a component that we think is present in the mixture. The distance the solvent front moved is 4.0 cm, and the distance the middle component of the mixture moved is 2.0 cm. The middle component's  $R_f$  is  $2.0 \text{ cm} / 4.0 \text{ cm} = 0.50$ . The distance Compound A moved is 2.0 cm, so its  $R_f$  is also 0.50. On the basis of identical  $R_f$  values, we can conclude that Compound A is probably one of the mixture's components.

Frequently, other information from chromatograms supports findings based on  $R_f$  calculations. For instance, when resolving food dyes, we can compare the colors of the spots. For example, when resolving green food coloring, we observe two spots,

one yellow and one blue. These colors, in conjunction with the  $R_f$  values of the components, help identify the dyes in the green food coloring.

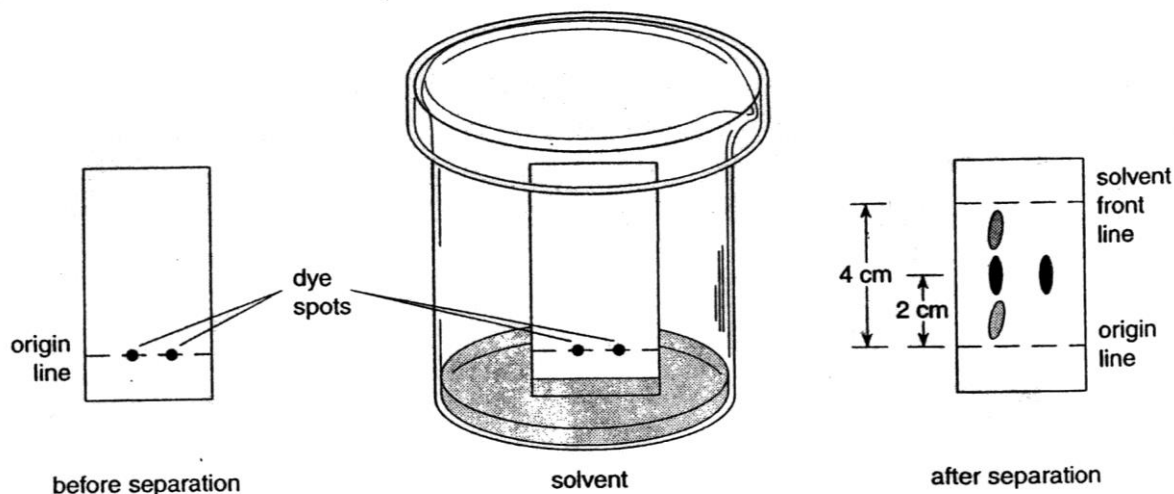


Figure 1.

Felt tip pens have a 'rod' inside them. It's made of an absorbent material and is impregnated with a **mixture** of different colored dyes. The color you get from a pen depends on the mixture of inks used. Ink dyes can be separated using paper chromatography.

There are two types of pens - permanent and non-permanent. The difference is how easily they can be washed from what they have been used to mark (whether deliberately or accidentally).