1. Separation of a Mixture by Paper Chromatography

Introduction

The separation, detection, and identification of the components of a mixture can be accomplished by several techniques. Each of these techniques depends on the differing chemical or physical properties of the components of the mixture. *Chromatography* (Ebbing/Gammon, Section 1.4) is one such technique. *Paper chromatography*, which is used here, is just one of several chromatographic methods available.

Purpose

In this experiment you will learn to separate, detect, and identify the components of a mixture by using paper chromatography. The components will be cobalt(II) chloride, nickel(II) chloride, copper(II) chloride, and possibly iron(III) chloride.

A simple explanation of paper chromatography

In paper chromatography, a drop of solution containing a mixture of substances is placed near one end of a rectangular piece of filter paper. The paper serves as the *stationary phase*. The end of the paper is immersed in a liquid to a point that is just below the spot where the drop was placed on the paper. The liquid is the *mobile phase*. Capillary action (the same phenomenon that causes water to travel up a bath towel when an edge of the towel is immersed) causes the liquid to flow up the filter paper. When the liquid reaches the spot, the components of the mixture will begin to migrate upward with the mobile phase. Each component will have a characteristic chemical affinity for the paper and a characteristic chemical affinity for the liquid. These affinities are competitive: The component's affinity for the paper tends to hold the component in one place, but its affinity for the liquid tends to make the component follow the liquid as it moves upward. A component with a strong affinity for the paper and a weak affinity for the liquid will move more slowly than a component with a weaker affinity for the paper and a stronger affinity for the liquid.

A substance's affinities for the stationary and mobile phases are entirely characteristic of that substance. Different substances have different competitive affinities. Because each component of a mixture has its own characteristic affinities, each component will travel up the paper at its own characteristic rate. If the paper is sufficiently large, all the components can be separated by the time the liquid has reached the top of the paper.

Each component will now appear as a separate spot. If the components are highly colored, the spots will be visible. You can convert weakly colored or colorless spots to highly colored spots by spraying them with substances that react with the components in the spots. The filter paper will now contain a vertical array of colored spots arranged according to their characteristic rates of ascent. The word *chromatography*, which is derived from two Greek words and literally means "written in color," was coined to describe this phenomenon.

The distance traveled by a component of a spot with respect to the distance traveled by the pure liquid is a measure of that component's competitive affinities for the stationary and mobile phases. We define the component's R_F (retention factor) value in those terms:

$$R_F = \frac{\text{distance traveled by spot}}{\text{distance traveled by liquid}}$$

The R_F value of a substance is characteristic of that substance and should be a constant under invariant experimental conditions.

Concept of the experiment

You will examine the paper chromatography of $CoCl_2$ [cobalt(II) chloride], $NiCl_2$ [nickel(II) chloride], and $CuCl_2$ [copper(II) chloride]. In addition, your laboratory instructor may elect to include $FeCl_3$ [iron(III) chloride] if your distilled water and reagents are not already so contaminated with this substance that the detection of a small quantity will be virtually impossible.

The mobile phase will be a mixture of an aqueous solution of HCl (hydrochloric acid) and either acetone or 2-butanone.

CAUTION: Because of the volatility and flammability of acetone and 2-butanone, no flames will be allowed in the laboratory.

At the completion of the experiment, you will spray the paper successively with solutions of NH_3 (ammonia) and Na_2S (sodium sulfide). The former will react with the HCl in the mobile phase to form NH_4Cl (ammonium chloride) according to the equation

$$NH_3 + HCl \rightarrow NH_4Cl$$

Sodium sulfide will react with a component of a spot to give a darkly colored spot containing one of the sulfides (Fe_2S_3, CoS, NiS, or CuS) and colorless sodium chloride (NaCl).

CAUTION: Hydrochloric acid and ammonia can cause chemical burns in addition to ruining your clothing. If you spill one of these substances on you, wash the contaminated area thoroughly and report the incident to your laboratory instructor. You may require further treatment.

You will be able to determine the R_F value for each substance by observing its ascent in the absence of the other substances. You will also subject a known mixture of all the substances to chromatography so that you can see that the same R_F values are obtained with a mixture. Finally, you will be able to identify the components in an unknown mixture on the basis of its chromatography and derived R_F values. This unknown will contain one or more of the substances whose behavior you have studied in this experiment.

Procedure

Getting started

1. Obtain your unknown mixture.
2. Obtain directions for discarding the solution that you will use as the mobile phase in this experiment.
3. Obtain a 5 × 9-inch piece of filter paper, a smaller scrap of the same paper, 5 capillary tubes, 4 test tubes, a piece of clear plastic film, a rubber band, and an 800-mL beaker. If your laboratory instructor elects to use $FeCl_3$, obtain 6 capillary tubes and 5 test tubes.
4. Add 35 mL of either acetone or 2-butanone to the 800-mL beaker, followed by 10 mL of the HCl solution. Swirl the beaker gently to mix the solution. Cover the beaker with the plastic film, and hold the film in place with the rubber band.
5. Place the large piece of filter paper on a clean paper towel. Use a pencil (not a pen) and a ruler to draw a line 2 cm from one of the longer edges.
6. Fold the paper in half so that the line you have drawn is bisected. In the same manner, fold the paper in half a second and then a third time. The line will have been divided into 8 equal segments. Refold the paper so that it looks like an accordion, as shown in Figure 1.1.

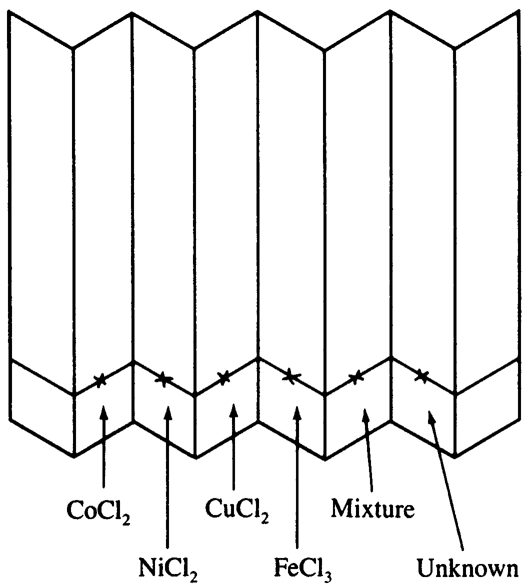

FIGURE 1.1
A piece of chromatography paper that has been properly folded for use in this experiment.

7. Mark the approximate center of each of the six inner segments of the line with the pencil. There will be no marks in the first segment on the left and the first segment on the right.
8. Write "$CoCl_2$" under the first mark on the left, "$NiCl_2$" under the next mark, "$CuCl_2$" under the third mark, "$FeCl_3$" under the fourth mark (if this substance is included in the experiment; leave it blank otherwise), "Mixture" under the fifth mark, and "Unknown" under the last mark.
9. Add some distilled water to one of the test tubes. Use one of the capillary tubes to practice spotting the scrap of filter paper. The maximum diameter of an acceptable spot is no larger than about 0.5 cm. When your procedure is satisfactory, empty the test tube and dry it. Dry the capillary tube by touching its wet tip to a piece of paper towel.

Doing the experiment

1. Mark each of the test tubes for recognition. Place a small amount (about 1/4 inch) of the appropriate solution in the corresponding test tube.
2. Open the large piece of filter paper and place it once again on the paper towel. Use a clean capillary tube to spot the paper, placing the appropriate solution on the corresponding mark on the 2-cm line.
3. Dry the paper by holding it *briefly* over a heat lamp or by waving it gently in the air. Refold the paper like an accordion.
4. Open the 800-mL beaker and gently place the folded paper inside. The 2-cm line should be above the surface of the liquid but close to it. Do not splash. Replace the plastic film. The result is shown in Figure 1.2.

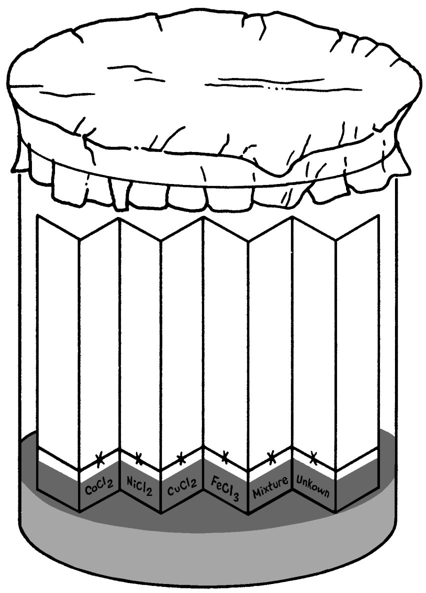

FIGURE 1.2
The experimental arrangement, showing the covered beaker containing the folded chromatography paper (the stationary phase) and the solution (the mobile phase).

5. The beaker must be absolutely stationary throughout the experiment. To view the paper in its entirety, move to another position rather than turning the beaker.
6. Allow the liquid to ascend to within 2–3 cm of the top of the paper (about 30–40 minutes will be required). You should be able to observe the progress of some of the spots during the ascent. Record your observations, including the colors of the spots that you are able to see.
7. When the liquid has reached the desired height, remove the paper from the beaker. Place the wet paper on the paper towel and mark with a pencil the position to which the pure liquid has ascended.
8. Partially dry the paper, using the drying methods described in Step 3.
9. The operation described in Step 10 must be done in a hood.

CAUTION: Sodium sulfide and its reaction product with water are toxic substances that should not be tasted or inhaled. An efficient hood must be used during this step.

10. Wearing rubber or plastic gloves, spray the paper with a solution of NH_3. The paper should be moist but not wet. Next spray the paper with a solution of Na_2S. A dark spot should now be visible for each substance. Dry the paper.

11. Circle each spot with a pencil.
12. Measure the vertical distance that the approximate center of each of these spots has traveled from the original mark on the 2-cm line. Your measurements should be to the closest tenth of a centimeter. Record your results.
13. Measure and record the vertical distance from the 2-cm line that the pure liquid has ascended. Calculate the R_F value for each spot.
14. Use the R_F values to deduce the identity of each component in the unknown mixture.
15. Attach the dry chromatography paper to your report.

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Date:	....	Student name:	....
Course:	....	Team members:	....
Section:	....		....
Instructor:	....		....

Prelaboratory assignment

1. Match the name of each compound with its formula.

copper(II) chloride	$NiCl_2$
sodium sulfide	HCl
iron(III) chloride	NH_3
nickel(II) chloride	$CuCl_2$
cobalt(II) chloride	Na_2S
ammonia	$CoCl_2$
hydrochloric acid	$FeCl_3$

2. A piece of filter paper is spotted with a solution containing a mixture of two components, A and B. The chemical affinity of A for the stationary phase is less than that of B, and the chemical affinity of A for the mobile phase is greater than that of B. Which substance will have traveled farther at the completion of the chromatography experiment? Which substance will have the larger R_F value? Explain each answer.

3. What special safety precautions must be taken during this experiment?

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Results

	Observations	**Distances**	**R_F Values**
$CoCl_2$			
$NiCl_2$			
$CuCl_2$			
$FeCl_3$			
Mixture			
Unknown No. _______			

Distance solvent traveled: .... cm

Components of the unknown mixture:

Questions

1. Why is a pencil used to draw the lines on your piece of filter paper rather than a pen (see, for example, Ebbing/Gammon, Figure 1.16)?

2. Iron(III) chloride is a common impurity in this experiment because the distilled water in the solution of HCl may have passed through iron pipes. Examine your piece of filter paper carefully. If this impurity is present, why is it a horizontal streak rather than a spot? If it is not present, why would its presence as an impurity result in a horizontal streak of color rather than a spot?

3. Write balanced equations for the reactions of $FeCl_3$, $CoCl_2$, $NiCl_2$, and $CuCl_2$ with Na_2S.