Introductory Chemistry Lab: Paper Chromatography

Outcomes

As a result of today's laboratory you will have:

Prepared samples for chromatographic analysis.
Applied samples to a filter paper stationary phase.
Developed a chromatogram.
Calculated the R_f values for substances separated by chromatography.
Interpreted chromatographic results to identify the number of dyes present in different colors of candy.

Prelab

Prepare a Title (can use the lab handout for this), Purpose (a concise statement) and a Procedure (short "to do" list … see "Writing a Procedure" in the lab handouts folder), and Data Tables.

Purpose

To investigate the number of dyes present in different colors of selected M & M candies.

Safety Note: ***Eating and drinking in the lab is never permitted. No candy is to be eaten in the lab.***

Background Information

Chromatography is a method of separating and analyzing mixtures based on differences in solubility, the ability of a substance to dissolve in a solvent. The sample mixture is applied to a stationary solid material called the stationary phase. A liquid or a gas is then moved through the stationary phase. The moving liquid or gas is called the mobile phase. Substances that are more soluble in the mobile phase are more attracted to the moving liquid or gas than to the stationary phase. The more soluble components of the mixture will move faster through the stationary phase. On the other hand, components of the mixture that are less soluble in the moving liquid or gas are more attracted to the stationary phase. These substances will move slowly through the stationary phase. Since the components of the mixture move through the stationary phase at different rates, they will separate from each other.

A model that illustrates how chromatography works is a marathon. At the start of the race, all the runners are on the starting line, just like the molecules in a mixture at the when they are applied to the stationary phase. When the race begins the runners take off, each running at a different rate. As the race progresses, the fastest runners begin to pull ahead into a "front pack" leaving the slower runners behind. The slower runners also begin to separate into groups: medium-fast, medium, medium-slow, slowest. Just like the runners separate, the molecules in a mixture will begin to separate as they move through the stationary phase at different rates. The molecules more attracted to the mobile phase will move ahead in the race through the solid, leaving behind the molecules that are more attracted to the stationary phase..

There are many different types of chromatography used by chemists. The types vary in the stationary and mobile phases. There are also differences in how the components of the mixture are detected. In simple systems the various colored components are visually detected. Other types of chromatography use instruments to monitor physical properties such as refractive index, light absorbance at specified wavelengths, electrical conductivity, etc.

The goal of preparative chromatography is to collect (isolate) individual components present in mixtures. In this case the moving liquid is collected at the end of the chromatography process. Each separated component is collected in a different container as it arrives at the end of the chromatography apparatus.

In analytical chromatography the goal is to determine both the identity and the relative amount of the individual components present in the sample. The components are not collected. These chromatographic systems are usually more sophisticated and use instruments to monitor the mobile phase as it leaves the stationary phase. The identity of the component is determined from the time it takes to move through the

stationary phase. The relative amounts of each component can be determined from the intensity of the signal measured by the detecting instrument.

In this lab session you will use simple paper chromatography to analyze the mixtures of dyes present in M&Ms. Filter paper will be used as the stationary phase. The mobile phase is 1.0 % NaCl solution. You will determine how many dyes are found in a color mixture and if any dyes are common to more than one color.

Procedure Work in pairs

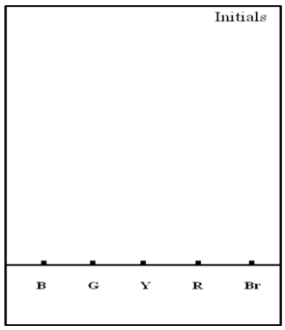
Part I Preparing the Chromatogram
Keep the filter paper clean. Hold by the edges only.
Place the filter paper on paper towels, not directly on lab bench

1 .Lay a pre-cut piece of filter paper on a paper towel. Use a pencil and a ruler to draw a line along one of the shorter sides, ~2 cm from the edge of the paper. This is the bottom of the chromatogram. Make five dots with the pencil along the line, starting at least 2.0 cm from the side edge of the paper and leaving ~ 2.0 cm between dots. Label each of the five dots on the filter paper *below the line using a pencil* for the colors you are going to test. Write your initials in pencil at the very top right of the paper.

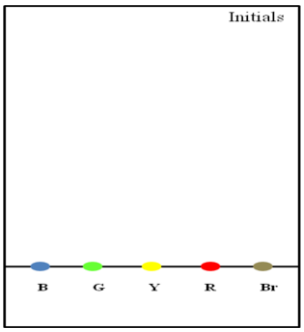
2. Place each of the five M&M's on separate watch glasses. Add a drop of deionized water in the center of each one. Wait about a minute for the color to dissolve.

3. Apply a sample of each candy color to the appropriate dot on the filter paper. Dip the small straw in one of the colored solutions and lightly touch (keep the spot from spreading) it to one of the pencil marks. Use a different straw for each color,

4. After all the color spots on the filter paper have dried, repeat step 3 three times.



Start Spots



Finished Spotting

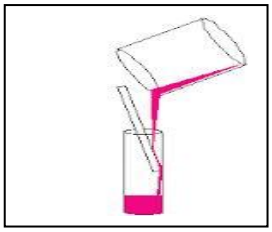
5. When the paper is dry, thread the furnished wire through the filter paper about 1 cm from the top.

6. Bend the wire to suspend the filter paper inside a dry 2-L beaker. The colored dots from the M&M samples should be at least 1 cm above the 200-mL line at the bottom of the beaker.

Part II. Developing the Chromatogram

1**. Use a stirring rod** (see below) to *c*arefully add the 1.0% NaCl solution to a level ~ ½ the distance between the chromatogram origin line and the bottom of the paper chromatogram. T**he colored dots must be above the level of the mobile phase**

2. Do not disturb the beaker or the chromatogram.



Carefully
Pour solvent onto the stirring rod with
End of rod touching the beaker walls
This method is universal for pouring with minimum splashing

3. Leave the chromatogram in the beaker until the mobile phase (wet looking solvent line) stops moving.

4. Remove the chromatogram from the beaker and place on several layers of paper towel. Mark the progress of the mobile phase and the outline of each dye spot (blob) with a pencil. Note: the solvent line will very likely not be perfectly straight.

5. Dry the chromatogram with a hair dryer.

Part III. Analyzing the Chromatogram

The leading edge of the mobile phase is called the solvent front. The distance that a dye spot travels. compared to the distance that the solvent travels is called the ratio of fronts, retention factor, or R_f value. Different components in the mixture will have different R_f values. I**f two dyes have the same color and the same R_f value, they may be the same compound. If two dyes have different colors or different R_f values, then they are different compounds.**

Data

1. Measure the distance traveled by the solvent (the mobile phase) from the pencil starting line to the solvent front for each M&M color. Record the values in Table 1.

2. For each M&M color, the distance traveled by each component dye from the pencil dot on the starting line to the leading edge of the dye spot. Record the values in Table 1. Measure all visible spots (as shown below).

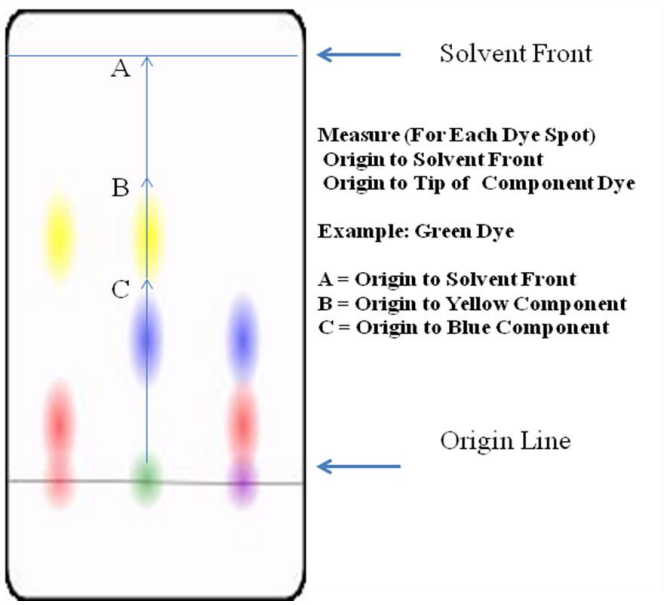


Table 1: Distance Traveled (in cm) By Solvent Front and Each Dye Spot

Replace "M&M Color n" in Table 1 & 2 below with the actual color of each M&M used

	M&M Color 1	M&M Color 2	M&M Color 3	M&M Color 4	M&M Color 5
Solvent Front					
Yellow Dye					
Red Dye					
Orange Dye					
Blue Dye					

Clean Up

1. The NaCl solution is drain-disposable. Use alcohol to remove any ink markings from your glassware. Clean all glassware with soap, rinse with tap water and then with deionized water. Towel dry and return to its storage location.
2. Wipe off your lab bench with a wet paper towel; return your lab stool to its place before leaving the lab.

Calculations

Calculate the R_f values for each component dye found in the five M&Ms.

$$R_f = \frac{\text{Distance traveled from origin to tip of component dye}}{\text{Distance traveled by the solvent}}$$

Results

Tabulate the R_f values for each dye spot found in the various M&Ms.

Table 2: R_f Values

	M&M Color 1	M&M Color 2	M&M Color 3	M&M Color 4	M&M Color 5
Yellow Dye					
Red Dye					
Orange Dye					
Blue Dye					

Sketch your chromatogram.

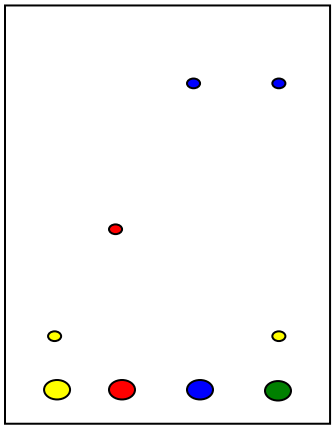
Conclusion

Describe the number of individual dyes spots present in the different candy colors you tested. Indicate if any of the individual dyes spots are present in more than one color and state your evidence for this conclusion.

Staple your chromatogram to the back of one of your group's lab report.

Questions

1) A particular mixture was separated into three components using paper chromatography. The R_f value of dye A is 0.78, the R_f value of dye B is 0.43 and the R_f value of dye C is 0.19.
   a) Which component of the mixture: A, B, or C, is the most soluble in the mobile phase? Why?
   b) Which component of the mixture: A, B, or C, is the most attracted to the stationary phase? Why?
2) Why is the starting line of the dye dots at the bottom of the chromatogram marked in pencil?
3) Explain how the chromatogram displayed below shows that the green dye contains no red component.
4) Which component dye (spot) on the chromatogram below is least attracted to the stationary phase?



Blue Component Spots

Red Component Spots

Yellow Component Spots

Above Origin Sample Spots:
Yellow Red Blue Green