

Paper Chromatography and Steam Distillation

EVERY STUDENT MUST BRING AT LEAST 3 ORANGES TO LAB FOR THIS EXPERIMENT!

Equipment

You will need a 600 mL beaker, a 50 mL graduated cylinder, 4 Expo Wet Erase markers, each a different color, three different food colors, three capillary tubes, a pencil, a ruler, a piece of chromatography paper, a piece of plastic wrap, the glassware for a distillation, a blender, a zester, a separatory funnel, 2 small test tubes, and a powder funnel.

Chemicals

You will need about 25 mL of ~70% isopropanol and a small amount of anhydrous calcium chloride.

Introduction – Paper Chromatography

In the paper chromatography portion of this experiment we will use the fact that “like dissolves like”. What this means is that a polar substance would rather dissolve in a polar substance than in a non-polar substance, and a non-polar substance would rather dissolve in another non-polar substance than in a polar substance.

For example, alcohol (ethanol) which is polar would rather dissolve in water, which is also polar, than in oil which is non-polar.

The markers and food coloring that you will use in the paper chromatography portion of this experiment get their colors from one or more pigments (molecules). These pigments generally have different polarities.

We will use this fact to separate these pigments from each other, and to determine which color pigments are in each marker and food coloring.

In paper chromatography one or more samples is spotted on special paper, chromatography paper.

The paper is then placed in a developing chamber (a beaker) with a

solvent in it.

The solvent is pulled up the paper, just as water is pulled into a paper towel.

As the solvent passes the sample it tries to pull the sample with it. However, the paper is trying to hang on to the sample. There is a tug-of-war for the sample.

If the solvent has a stronger attraction for a pigment in the sample it will pull the pigment with it.

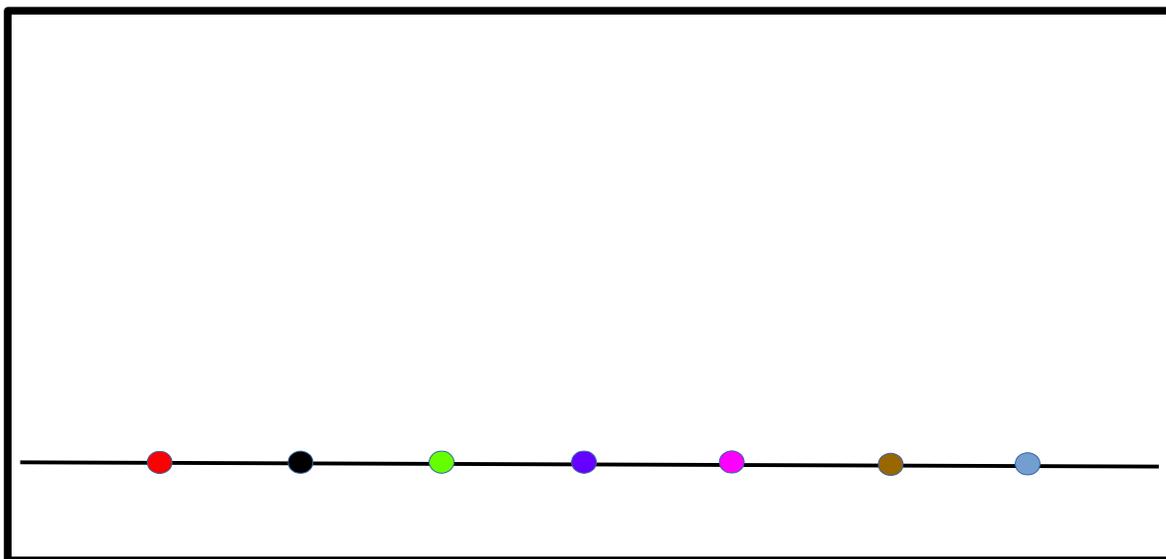
If the paper has a stronger attraction for a pigment it will hold onto that pigment.

The closer the polarity of the solvent is to the polarity of a pigment the stronger an attraction the solvent has for that pigment.

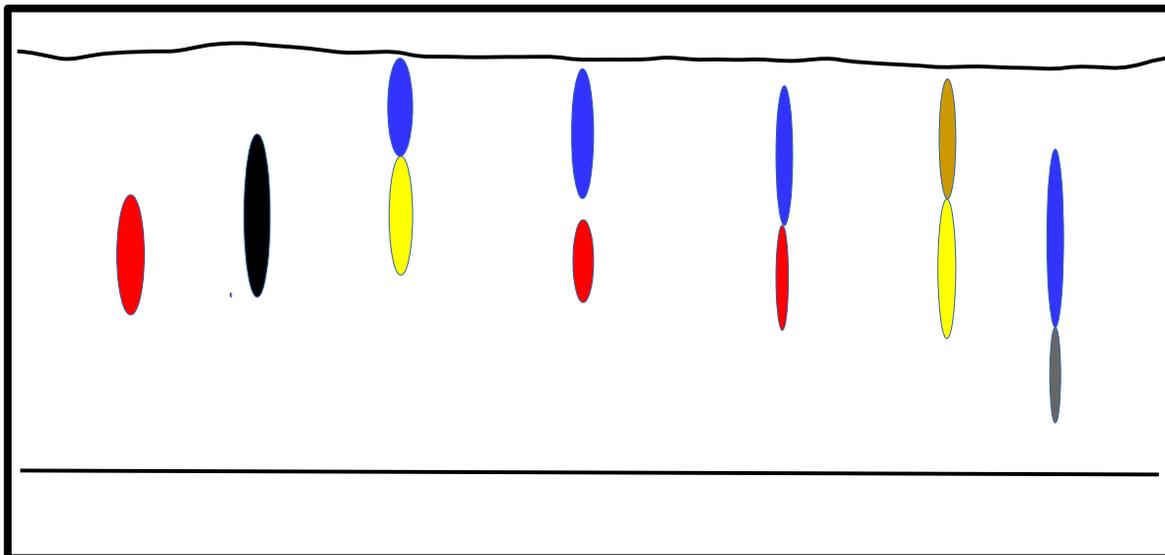
The closer the polarity of the paper is to the polarity of a pigment the stronger an attraction the paper has for that pigment.

As the solvent travels up the paper the sample is separated into it's individual pigments because each pigment has a different polarity, and will be more or less attracted to the paper (an solvent) than another pigment.

The paper would look something like this before it is placed in the developing chamber.



And after the solvent has traveled up the paper most of the way it might look like this.



This is just an example, the colors are not accurate. Each color represents a pigment. The pigments are separated from each other due to their different polarities.

The line at the top is the solvent front. That is, how far the solvent traveled up the paper before it was removed from the developing chamber.

The ratio of how far a pigment travels up a paper to how far the solvent traveled is called its R_f number.

$$R_f = \frac{\text{Distance to middle of spot}}{\text{Distance to solvent front}} \quad (\text{Equation 1})$$

Each distance is measure from the line on which the samples were spotted, not the bottom of the paper.

Introduction – Steam Distillation

In steam distillation a nonpolar molecule, with a high boiling point, is isolated by boiling a mixture of water and the molecule.

Steam distillation is commonly used to collect essential oils, among other uses. In the steam distillation portion of this experiment you will collect an essential oil (limonene) from orange peels.

You will boil a mixture of orange peel and water, collecting the distillate. The distillate will contain both limonene and water.

The reason that you can distill limonene, which boils at a temperature of 176 °C at a temperature of about 100 °C is that the mixture (water and limonene) will boil when the sum of their partial pressures is equal to the atmospheric pressure:

$$P_{\text{atm}}^{\circ} = P_{\text{water}}^{\circ} + P_{\text{limonene}}^{\circ} \quad \text{Equation 2}$$

Here P_{atm}° is the atmospheric pressure, P_{water}° is the partial pressure of the water, and $P_{\text{limonene}}^{\circ}$ is the partial pressure of the limonene. Because we have a mixture, the partial pressure of water and the partial pressure of the limonene will both be less than that of the pure substance. However, the mixture will boil when the temperature of the mixture is such that **Equation 2** is true.

Paper Chromatography Procedure

- 1.) Cut a piece of chromatography in half. You should end up with a piece that is about 4 or 5 inches high and about twice that in width.
- 2.) Using a pencil (NOT A PEN!) and a ruler draw a straight line horizontally about 2 cm from the bottom of the paper.
- 3.) Using the pencil make 7 marks on the line, spaced out approximately evenly. No mark should be closer than about 2 centimeters to an edge of the paper.
- 4.) Using pencil, label the spots beneath the line you drew with their color. For example (you need not use these colors):

1. Brown Expo
2. Blue Expo
3. Green Expo
4. Black Expo
5. Pink Food Color
6. Black Food Color
7. Green Food Color

5.) With the Expo markers make a small mark on the line where you made the pencil mark. The mark should be 1 – 2 millimeters in diameter.

6.) Using a different capillary tube (you can do a different color on each end of a capillary tube) for each different color make a similar mark at the appropriate places with each food color.

7.) Staple the ends of the chromatography paper together so that it forms a cylinder with the spots on the outside of the cylinder. **MAKE SURE THE ENDS ARE NOT TOUCHING!**

8.) Measure about 25 mL of ~70% isopropanol with your graduated cylinder and pour it into a clean 600 mL beaker.

9.) Place the spotted chromatography paper into the beaker with the isopropanol in it, the end closest to the spots down. **MAKE SURE THE PAPER DOES NOT TOUCH THE SIDES OF THE BEAKER!**

MAKE SURE THE SOLVENT DOES NOT TOUCH THE SPOTS!

10.) Cover the beaker with plastic wrap, sealing it so that the vapors can not get out.

11.) When the solvent front (the top of the wet area) gets to within 1 or 2 cm from the top of the paper remove the paper, with pencil

trace the top of the solvent front, and let the paper dry. This will likely take about 1 hour.

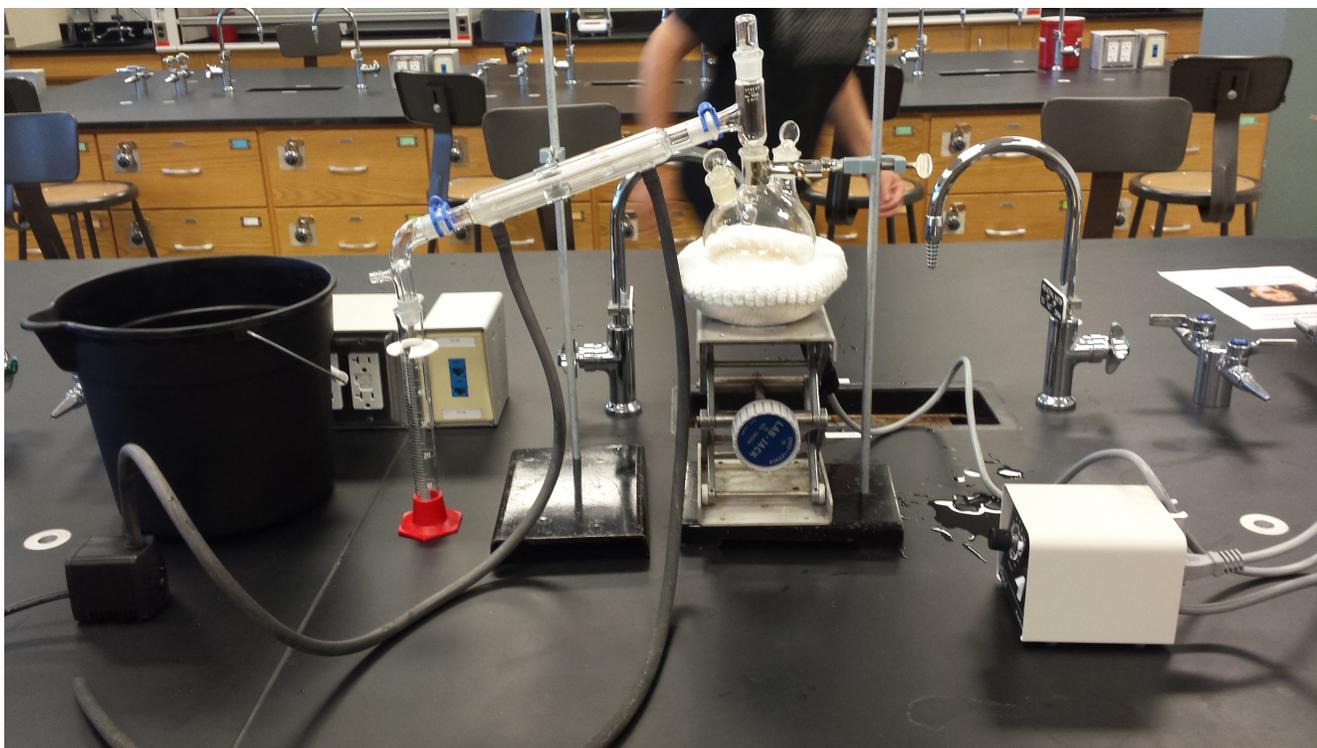
12.) When the paper is dry, remove the staples and lay the paper flat. Measure the distance from the bottom line to the solvent front. Record this in your data section (**A1**). Make sure to use units and the correct number of significant figures.

13.) Measure the distance from the bottom line to the center of each spot. For some of the marks you made there will be more than one spot, and for some there will only be one spot. Measure all of them. Record these in (**A2**), using units and the correct number of significant figures.

14.) Calculate R_f for each spot. Show all of your work, include units and significant figures.

Steam Distillation Procedure

1.) Wait for your professor to walk you through the set-up of the distillation glassware. Follow his/her instructions very carefully! When finished, your glassware should look like this:



- 2.) Zest your oranges, being careful not to scrape your fingers! Do not allow any of the white fleshy material to combine with the orange peel.
- 3.) Tare your 150 mL beaker, then add the orange peel to the beaker. Reweigh the beaker with the peels in it and record the mass of the orange peels in **(B1)**.
- 4.) Add the orange peels and about 250 mL of D.I. water to a blender (make sure all of the orange peel is transferred to the blender) and chop to a suspended slurry.
- 5.) Add all of the orange peel slurry and a stir bar to your 500 mL round bottom flask through a powder funnel.
- 6.) Distill until you have collected about 50 mL of distillate.
- 7.) Pour the distillate into a separatory funnel. The limonene should be on the top since it is less dense than water (the density of pure limonene is 0.84110 g/mL at room temperature).
- 8.) Remove the water by opening the stopcock over a beaker. You may pour the water down the drain.
- 9.) Transfer the orange oil (the oil left in the separatory funnel) to a test tube. Add a small amount of anhydrous calcium chloride (this will absorb any water still in the oil).
- 10.) Place a second test tube in a beaker and tare them out together on a balance.
- 11.) Transfer the dry orange oil to the tared test tube and place it back in the beaker on the balance. Record the mass of the purified orange oil **(B2)**.

Data and Analysis
Paper Chromatography

Data:

Distance from bottom line to solvent front: _____ (A1)

Record the distance, in cm, from the bottom line to each spot (A2)

Distance to Each Spot (cm)

Color	1	2	3	4	5	6	7

Steam Distillation

Mass of orange peel: _____ (B1)

Mass of orange oil: _____ (B2)

Analysis (Paper Chromatography):

1.) Calculate the R_f for each spot. Record your results in the following table.

R_f 's

Color	Spot 1	Spot 2	Spot 3	Spot 4	Spot 5	Spot 6	Spot 7

2.) The more polar a pigment is, the greater it's R_f (in this experiment). For each mark that you started with (1-7), in the following table state which color pigment was most polar and which

was least polar. If there was only 1 color pigment, say "1 pigment".

POLARITY

Color	Spot 1	Spot 2	Spot 3	Spot 4	Spot 5	Spot 6	Spot 7
Most Polar							
Least Polar							

Analysis (Steam Distillation):

1.) Calculate the mass percent of limone in your orange peel.

$$\frac{B2}{B1} \times 100 = \text{Mass Percent Limonene in your orange oil}$$

Mass percent limonene: _____ (B3)

2.) Calculate the average mass of limonene per orange for your oranges:

$$\frac{B2}{\text{Number of oranges}} = \text{Average Mass of Limonene per Orange}$$

Average Mass of Limonene per Orange: _____ (B4)

TURN IN PAGES 8 - 9

NAME: _____

SECTION: _____