Paper Chromatography of a Spinach Leaf Lab

# Background Information:

Paper chromatography is a process that uses special filter paper to separate and identify the different substances in a mixture. Chromatography means, “to write with color.” The substances in the mixture dissolve in the alcohol and move up the paper. The heavier substances move up the paper more slowly. The lighter substances move up the paper more quickly. Resulting in heavy and light substances get separated from one another on the paper.

Plants contain chlorophyll, a green pigment, as well as carotenoids, pigments that range in color from red to orange to yellow.



**Question:** Do green leaves also contain other pigments?

# Materials:

Isopropyl alcohol ruler

spinach leaf scissors

chromatography paper pencil

paperclip tape

beaker colored pencils

# Procedure:

1. Obtain a strip of chromatography paper.
2. Use a ruler to measure and draw a light pencil line 2-cm above the bottom of the paper strip.
3. Here is the tricky part! Place the edge of the spinach leaf over the pencil line and using the edge of a coin gently press on the spinach leaf to create a single green line over the pencil line. You want this line to be thin and *concentrated* with the pigment from the spinach leaf. Therefore, repeat this edging process carefully about 3-4 times. Be sure not to press too hard or you will poke a hole through the paper.
4. Tape the top of the paper strip to a pencil so that the end of the strip with the green line hangs down. The pencil should be able to sit across the top of the beaker with the bottom of the paper strip just touching the bottom of the beaker. Cut off any excess paper from the TOP of the strip if it is too long.
5. Remove the pencil/paper strip contraption from the beaker for the moment. Record observations in data table.
6. Carefully add isopropyl alcohol to the beaker until it reaches a depth of 1-cm in the beaker.
7. Lay the pencil across the top of the beaker with the paper strip extending into the alcohol. MAKE SURE THAT THE LEVEL OF THE ALCOHOL IS BELOW THE GREEN LINE ON YOUR PAPER STRIP! IF THE ALCOHOL IS GOING TO COVER THE GREEN LINE, POUR OUT SOME ALCOHOL BEFORE YOU GET THE GREEN LINE WET!
8. Observe as the alcohol gets absorbed and travels up the paper by capillary action. This may take up to 20 minutes. Do not touch your experiment during this time.
9. When the alcohol has absorbed to approximately 1-cm below the pencil, you may remove the pencil/paper strip from the beaker to dry on your counter. With a pencil, mark the distance the alcohol has traveled on the paper, as well as the distance each pigment has traveled.
10. Using colored pencils, draw and label your results in the data table.

Name: Period:

# Paper Chromatography of a Spinach Leaf Lab – ANALYSIS SHEET

# Data:

|  |  |
| --- | --- |
| **Filter Paper** | **Use colored pencils to draw and label your observations** |
| Paper After Chromatography |  |

Distance Alcohol Traveled: mm

|  |  |  |  |
| --- | --- | --- | --- |
| **Band Number** | **Name of** **Pigment** | **Color of Pigment** | **Migration distance (mm)** |
| **Solvent**  | N/A |  |  |
| **1 (top)** |  |  |  |
| **2** |  |  |  |
| **3** |  |  |  |
| **4 (bottom)** |  |  |  |

# Pigment Identification:

# Chlorophyll a = blue-green

# Chlorophyll b = olive green

# Xanthophyll = yellow

# Carotene = orange/yellow

# Analysis Questions:

1. Did the leaf you tested contain different pigments? Use your results to support your claim.

2. Why did the separation of pigments in the spinach extract occur as it did?

(How does paper chromatography work?)

3. Why are leaves green even though other pigments are present?

4. Based on what you have learned in this lab, explain why leaves tend to change color in autumn.