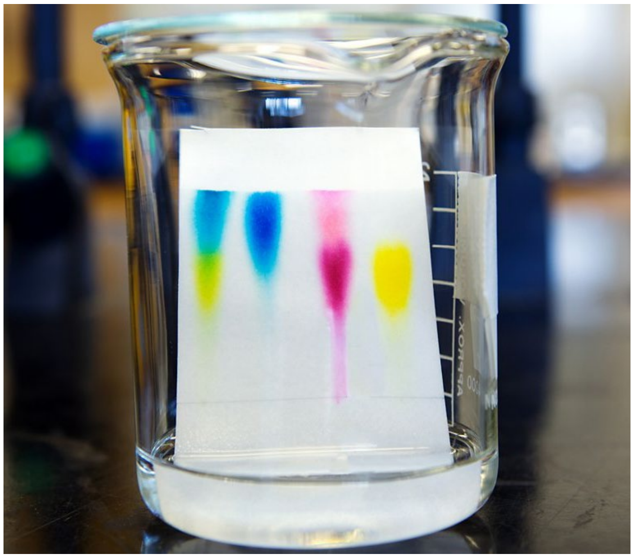
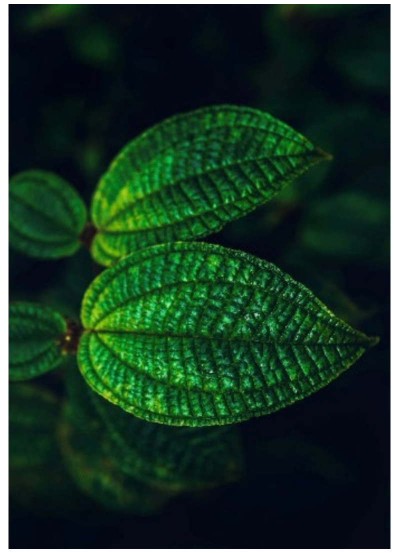
Plant Pigment Forensic Chromatography

By Diedre Young, Soybean Science Challenge





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Arkansas NGSS Standards Suggestions:

Integrated Biology:

Topic two: Structure and function

BI-LS1-2: Develop and use a model to illustrate the hierarchical organization of interacting systems that provide specific functions within multicellular organisms.

Topic three: Biodiversity and Population Dynamics

BI3-ETS1-3: Evaluate a solution to a complex real-world problem based on prioritized criteria and trade-offs that account for a range of constraints, including cost, safety, reliability, and aesthetics, as well as possible social, cultural, and environmental impacts.

Topic seven: Human impact on Earth Systems.

BI-ESS3-3: Create a computational simulation to illustrate the relationships among management of natural resources, the sustainability of human populations, and biodiversity.

Science and Engineering Practices:

Developing and using models (BI-LS-1-2)
Constructing Explanations and Designing Solutions (BI-ETS1-3)

Using Mathematics and Computational Thinking (BI-ESS3-3)

Disciplinary Core Ideas:

LS1.A: Structure and Function (BI-LS1-2)

ETS1.B: Developing Possible Solutions (BI3-ETS1-3)

ESS3.C: Human Impacts on Earth Systems (BI-ESS3-3)

Crosscutting Concepts:

Systems and System Models (BI-LS1-2, BI3-ETS1-3)

Stability and Change (BI-3SS3-3)

Integrated Chemistry:

Topic one: Matter and Chemical Reactions

CI-PS1-3: Plan and investigate to gather evidence to compare the structure of substances at the bulk scale to infer the strength of electrical forces between particles.

Science and Engineering Practices:

Planning and Carrying Out Investigations

Disciplinary Core Ideas:

PS1.A: Structure and Properties of Matter

Crosscutting Concepts:

Patterns

Chemistry II:

Topic two: Properties of matter

CII-PS1-3: Plan and investigate to gather evidence to compare the structure of substances at the bulk scale to infer the strength of electrical forces between particles.

CII-PS2-4AR: Develop and use a model of two particles interacting through electric fields to illustrate forces between particles and the changes in energy due to the interaction.

Topic seven: Organic Chemistry

CII7-ETS1-1: Analyze a major global challenge to specify qualitative and quantitative criteria and constraints for solutions that account for societal needs and wants.

Science and Engineering Practices:

Planning and Carrying Out Investigations (CII-PS1-3)

Developing and Using Models (CII-PS2-4AR)

Analyzing and Interpreting Data (CII7-ETS1-1)

Disciplinary Core Ideas:

PS2.B: Types of Interactions (CII-PS1-3)

PS1.A: Structure and Properties of Matter (CII-PS2-4AR)

ETS1.C: Optimizing the Design Solution (CII7-ETS1-1)

Crosscutting Concepts:

Scale, Proportion, and Quantity (CII-PS1-3)

Energy and Matter (CII-PS2-4AR)

Interdependence of Science, Engineering, and Technology (CII7-ETS1-1)

Soybean science challenge

Plant Pigment Forensic Chromatography

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Objective: Students will learn about various plant pigments and their function, that pigments have polarity, and how to calculate the Rf value (distance pigment traveled/distance solvent traveled) to identify unknown pigments. Students will also learn how separating plant pigments can help solve a crime!

Assessment: Students will prepare a written report summarizing their findings, and each group will prepare an oral presentation to share their findings with their classmates.

Key points: Rf, plant pigments and function, polarity/solubility of pigments, identification of pigments

Materials:

2 pieces of filter paper (chromatography paper or a coffee filter can be used)

1 - 150 mL beaker (any glass jar can be used if it has a lid, and make sure the jars are the same for the whole class) or a large test tube apparatus

1 beaker cover or stopper

1 Ruler

scissors (one per group)

pencil (one per group)

1 coin

A paper clip

fresh spinach or soybean leaf (one per group)

red leaf such as coleus leaf (one per group)

70% isopropyl alcohol (rubbing alcohol). A 9:1 ratio of petroleum-based ether to acetone can also be used. Keep in mind that these solvents are flammable and volatile and should be used in a fume hood.

Preparation: About 10-20 minutes depending on class size.

Time duration: One to two class periods depending on class size.

Elicit: Create a KWL chart by asking students to brainstorm and share anything they already know about plants and colors. Then guide them with open-ended questions about what they want to learn about pigments, plants, and forensic science.

Engage: Each of these videos is approximately 3-5 minutes. You can pick and choose.

https://www.youtube.com/watch?v=eqMv0pnyqkU

https://www.youtube.com/watch?v=wi9ubY76yZY

https://www.youtube.com/watch?v=YHKWCM5HaQU

https://www.youtube.com/watch?v=3VxeqgXa02c

https://www.youtube.com/shorts/24iU4jBJpK0

Explain:

As primary producers in the food chain with some bacteria and algae, plants produce their own food by using the sun's energy to transform carbon dioxide and water into glucose. In this process of photosynthesis, plants convert the sun's energy into chemical energy that is stored in the bonds of the glucose molecule. This energy fuels the metabolic processes of cells and is essential for life on earth. For photosynthesis to transform light energy from the sun into chemical energy (bond energy) in plants, the pigment molecules absorb light to power the chemical reactions. Plant pigments are macromolecules produced by the plant, and these pigments absorb specified wavelengths of visible light to provide the energy required for photosynthesis. Chlorophyll is necessary for photosynthesis, but accessory pigments collect and transfer energy to chlorophyll. Although pigments absorb light, the wavelengths of light that are not absorbed by the plant pigments are reflected in the eye. The reflected wavelengths are the colors we see in observing the plant. (Example: green pigments reflect green light) Plants contain different pigments, and some of the pigments observed include chlorophylls (greens), carotenoids (yellow, orange red), anthocyanins (red to blue, depending on pH), and betalains (red or yellow). The process of chromatography separates molecules because of the different solubilities of the molecules in a selected solvent. In paper chromatography, paper marked with an unknown, such as plant extract, is placed in a developing chamber with a specified solvent. The solvent carries the dissolved pigments as it moves up the paper.

The pigments are carried at different rates because they are not equally soluble (and of different polarities). A pigment that is more polar will travel the greatest distance, and a pigment that is less polar will move a shorter distance.

The scenario:

A crime has been committed at the edge of a soybean field next to the local park. A man was beaten unconscious and robbed on the edge of the field. A lady walking her dog reported seeing two “shady looking” characters walking briskly from the park. An elderly woman on a park bench noticed a young man with grass stains on his clothes carrying a book bag. An analysis of the crime scene revealed that the victim was found next to a soybean bush with leaves surrounding him. Crushed grass, soybean leaves, and dirt were found under his body. Stains were found on the man’s clothing and were collected by the crime lab. They also collected leaves from the soybean bush, leaves from beside the victim, and grass and dirt from under the victim. The couple seen walking briskly from the park were questioned by police. They claim to have been watching a soccer game that took place at the other end of the park. The blanket they were sitting on was found to contain bits of plant material that was collected to be analyzed at the lab. The young man with the book bag was also questioned. He is a student at the local college and claims to have been in the park collecting leaves for a biology project. The bag contained leaves collected from trees and bushes located from the entire park. A sample of what appeared to be grass stains on his clothes was collected for analysis at the lab.

The elderly woman sitting on the park bench was questioned by police and was found to have stains on her clothes and hands. Samples were collected to be analyzed at the crime lab. The crime lab analyzed the stains found on the victim’s clothes. They discovered that the stains match pigments found in the grass that were under the victim’s body and the soybean leaves that were next to him.
Should any of these people be questioned further? Is one of these people the criminal? *

**Teachers, you can set up the crime scene any way you want for forensic evidence and suspects.*

Elaborate:

Students will be placed into groups of three to four students. Each group will be given (or will need to retrieve) materials for the lab. Each group of students will work with one plant sample collected at the crime scene and one plant sample collected from a suspect*. **Your results will be shared with the class. Your data and your classmates' data will be tabulated to help you solve the crime. Your instructor will coordinate this.***

The Experiment:
1. Cut a strip of chromatography (or coffee filter) paper so that it just fits inside a large 15-cm test-tube apparatus (or 150 beaker).
Cut one end to a point and draw a faint pencil line across the tip. The 'chromatography' paper, when hung on the paper clip in the test tube or beaker, should just barely touch the bottom.
2. Tear the plant matter from the crime scene into small pieces. Take a coin and rub the coin on the plant that is placed over the faint pencil line. Do this at least three times to make the line more intense.
3. Place the strip of chromatography paper in the chromatography apparatus. The tip of the paper should just touch the bottom, and the pigment line **must** be above the solvent level. Your teacher will dispense the chromatography solvent to you in the fume hood. Watch the solvent travel up the paper strip. ***As soon as the solvent reaches the top***, remove the chromatography paper and allow it to dry. Replace the lid on the apparatus.
4. Colored bands or dots should be present on the chromatography paper. Using a pencil, circle the bands or dots. Identify and label the pigments on your chromatography paper. The following may be present (from top to bottom): carotene (orange), xanthophyll (yellow), chlorophyll *b* (yellow green), chlorophyll *a* (blue green), and anthocyanin (red).
5. Repeat steps 3–5 for the plant pigments collected from a suspect.
6. Calculate the *Rf* values for each pigment on your chromatogram. The *Rf* value is a ratio of the distance the pigment traveled to the distance the solvent traveled.

R = *<u>Distance traveled by pigment</u>*
Distance traveled by solvent f

Measure the distance traveled by each pigment (from the dotted line to the spot) and the distance traveled by the solvent (from the dotted line to the top of the paper). Calculate the Rf value for each pigment in each chromatogram.

9. Prepare a data table and tabulate your results with your classmates' results.

10. Compare plant pigments found on the suspects to those found at the crime scene.
    1. Do any of the plant pigments match? If so, which ones?
    2. What should the crime lab do next?
    3. Based on this evidence, should the crime investigators pursue any of these suspects? Who and why?

Evaluate:

Prepare a written report summarizing your findings and prepare an oral presentation to share your findings with your classmates.

Extend:

Have students collaborate to discuss other ways to determine the culprit.

Have a forensic scientist speak to the class about what they do for a living.