

Plant Pigments/Chromatography

Introduction

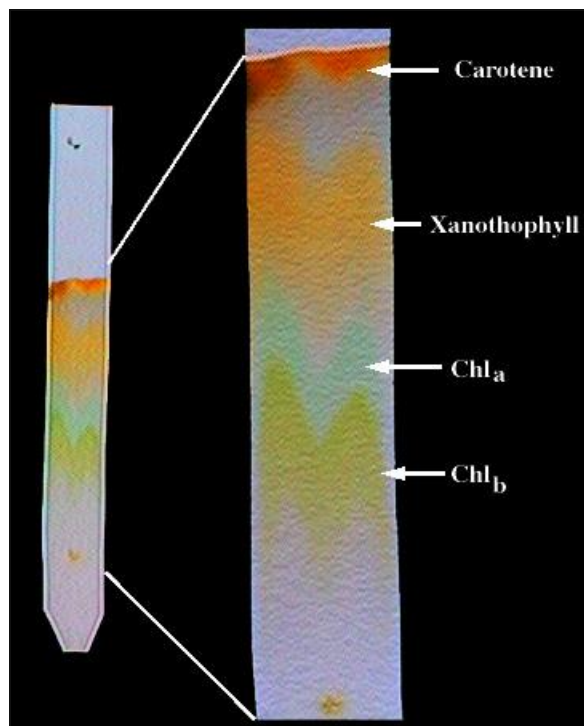
Paper chromatography is a useful technique for separating and identifying pigment and other molecules from cell extracts that contain a complex mixture of molecules. The solvent moves up the paper by capillary action, which occurs as a result of the attraction of solvent molecules to the paper and the attraction of the solvent molecules to one another. As the solvent moves up the paper, it carries along any substances dissolved in it. The pigments are carried along at different rates because they are not equally soluble in the solvent and because they are attracted, to different degrees, to the fibers of the paper through the formation of intermolecular bonds, such as hydrogen bonds.

Beta carotene, the most abundant carotene in plants, is carried along near the solvent front because it is very soluble in the solvent being used and because it forms no hydrogen bonds with cellulose. Another pigment, **Xanthophyll** differs from carotene in that it contains oxygen. Xanthophyll is found further from the solvent front because it is less soluble in the solvent and has been slowed down by hydrogen bonding to the cellulose. **Chlorophyll's** contain oxygen and nitrogen and are bound more tightly to the paper than the other pigments. **Chlorophyll a** is the primary photosynthetic pigment in plants. A molecule of chlorophyll a is located at the reaction center of the photo systems. The pigments collect light energy and send it to the reaction center. Carotenoids also protect the photosynthetic systems from damaging effects of ultraviolet light.

Distance moved by Pigment Band (millimeters)

| Band Number | Distance (mm) | Band Color |
|-------------|---------------|------------|
| 1 | | |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |

Distance Solvent Front Moved _____



Analysis of Results

The relationship of the distance moved by a pigment to the distance moved by the solvent is a constant called R_f . It can be calculated for each of the pigments using the formula:

| | |
|---------|----------------------------------|
| $R_f =$ | <u>distance pigment migrated</u> |
| | <u>(mm)</u> |
| | distance solvent front migrated |
| | (mm) |

Record your R_f values in the table below.

| | |
|--|---|
| | = R_f for carotene (yellow to yellow -orange) |
| | = R_f for xanthophyll (yellow) |
| | = R_f for Chlorophyll a (bright green to blue green) |
| | = R_f for Chlorophyll b (yellow green to olive green) |

Topics for Discussion

1. What factors are involved in the separation of the pigments?

2. Would you expect the R_f value of a pigment to be the same if a different solvent were used? Explain.

3. What type of chlorophyll does the reaction center contain? What are the roles of the other pigments?
