Paper Chromatography of Food Dyes

Introduction:

Chromatography is an experimental technique which can be used to separate mixtures. There are a wide variety of methods including column, thin-layer, and paper chromatography. All of these methods use the fact that different substances have different solubilities in a given solvent. In this experiment we will use paper chromatography. A dye mixture will be deposited on a strip of paper. The solvent will flow through the adsorbent material (the paper) by capillary action. As the solvent passes the sample, the substances in the dye which are most soluble will be swept along with the solvent. With the proper choice of solvent, the mixture can be resolved into its components.

There are seven dyes which the Food and Drug Administration (FDA) have approved for use in our foods. They are blue1, blue2, yellow5, yellow6, red3, red40, and green3. Commercial samples of five of these food dyes have been purchased for this experiment. Green3 and blue2 are rarely used and are not included in our experiment.

The first objective in this experiment is to find a solvent which is best able to separate the five FDA dyes. To accomplish this, each table will run their chromatogram with one of 3 possible solvent systems. Each group will spot on their chromatogram the five commercial dyes and a prepared mixture of the five dyes. As a group, we will determine the best solvent system.

Each group a) will use the class-selected solvent to analyze three prepared unknown mixtures and b) will attempt to identify the FDA dyes in three different food products.
Procedure:

1. Prepare approximately 10 mL of the eluting solution as directed by your instructor. Different tables will be assigned different eluting solutions. Pour the solution into the developing chamber and close the jar with the cap.

2. Wash and dry your hands. Using a ruler to tear against, obtain a piece of chromatography paper approximately 21 cm long. Hold the paper near the edges and try to handle the paper as little as possible. Using a PENCIL, draw a light line approximately 2.0 cm from the bottom of the paper as illustrated below. Beginning approximately 3 cm from the edge, place a small (X) at intervals of 3 cm. Place the following labels below each (X): B1, Y5, Y6, R3, R40, and mixture.

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 X  X  X  X  X  X
  B1  Y5  Y6  R3  R40  Mix
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3. A closed end capillary tube will be used for spotting. Practice the spotting procedure on a piece of filter paper. Once you are confident that your spotting procedure results in a small spot, spot each dye onto the chromatography paper at the center of the (X).

4. Carefully roll the paper forming a tube-like shape. Fasten the top edges together with a small piece of tape. The edges should be close together, but NOT overlapping.

5. Carefully put the paper into the developing solution. Recap the jar and allow the solvent to move up the paper. **DO NOT DISTURB** the system until the solvent front is approximately 1.5 cm from the top of the paper. When the chromatogram is complete, remove the paper and carefully unroll the paper. Mark the solvent front and allow the chromatogram to completely dry. (Regardless of the position of the solvent, remove your chromatogram after 30 minutes. If your solvent is not near the top of the paper, perhaps you should find a group of students who have better results and use their solvent for the next chromatogram.)
6. Compare your chromatogram with those obtained by your classmates. As a class determine which of the solvents can be used to distinguish between the FDA dyes. The group of students who used the selected solvent system will need to carefully label their chromatogram and make it available for all the students to use for the second part of the experiment.

7. Each pair of students will prepare another chromatogram. Prepare the chromatogram as follows:

   a) Spot the prepared mixture of all five dyes.
   b) Select three different prepared unknown mixtures and spot each one Be sure to label the chromatogram with the unknown numbers. These unknowns have been prepared by mixing two or more of these commercial samples. Each group will attempt to identify the dyes used in the prepared unknowns.
   d) Select three kool-aid samples. To prepare your kool-aid sample, place a small amount of the kool-aid sample into a spot plate. Dissolve the sample by adding several drops of warm distilled water.

8. Prepare the chromatogram and develop it in the class-selected solvent system. Compare your chromatogram with the standard and attempt to determine the FDA dyes used in the samples.

9. Complete the data page and turn in your labeled chromatograms.
Name ___________________________________________ Section ________________________

Data:

Solvent system used: ____________________________________________________________

Did your solvent system allow for good separation of the prepared mixture?

Which solvent system does the best job of separating the commercial food dyes? ______ Why do you think it is the best solvent?

Sketch the chromatogram selected by the class as the standard.
Did this class selected solvent system allow you to identify the composition of your unknown mixtures?

If so, which FDA dyes were mixed to prepare your unknowns?

Unknown # _______ contains ________________________________

Unknown # _______ contains ________________________________

Unknown # _______ contains ________________________________

If you experienced difficulty determining the composition of the unknowns, please discuss those difficulties. How might you modify the experiment to improve your results?

Kool-aid sample(s)

The food sample ______________ contains ________________________________

The food sample ______________ contains ________________________________

The food sample ______________ contains ________________________________

If you experienced difficulty determining the composition of your kool-aid samples, please discuss those difficulties. How might you modify the experiment to improve your results?