How Blue is Your Sports Drink?

https://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p075/chemistry/measure-blue-dye-sports-drinks (http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p075/chemistry/measure-blue-dye-sports-drinks)

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Experimental Procedure

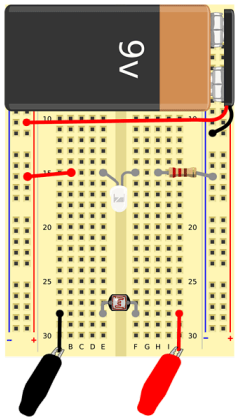
The following procedure can be broken into three parts:

1. Build and test the spectrophotometer that converts the concentration of dye in a solution into electrical resistance, which you can read off a multimeter;
2. Make a set of **standard solutions**, so that you know how to convert between the data you have (resistance) and the data you want (concentration); and
3. Determine the amount of dye in your sports drink samples with unknown concentrations of Blue 1.

Part 1: Building and Testing the Spectrophotometer

In this section, you will assemble a circuit on a **breadboard**. If you have never used a breadboard before, you should refer to the the Science Buddies resource How to Use a Breadboard (http://www.sciencebuddies.org/science-fair-projects/references/how-to-use-a-breadboard) before you proceed. You can follow a step-by-step slideshow that will show you how to put components in the breadboard one at a time. Alternatively, Table 1 lists each component and its location on the breadboard. **Important:** Read these notes before you proceed:

- Resistors are marked with colored bands. These colors *do* matter. Make sure you pick the right resistors for each step according to the markings.
- It matters in which direction some of the components are facing. Make sure you read the slideshow captions for any special notes about inserting each part.
- This section only shows you how to assemble the circuit. For a detailed explanation of how the circuit works, see the Help (#help) section.



Slideshow with step-by-step instructions viewable online.

Part name	Picture	Breadboard Symbol	Location
9 V battery			Red wire to (+) bus Black wire to (-) bus
Photoresistor			E28, F28
White LED			Long lead to E15 Short lead to F15
Jumper wires (3)			A28 to multimeter J28 to multimeter B15 to (+) bus
220 Ω resistor			H15 to (-) bus

Table 1. Components for the spectrophotometer circuit (Image credits: Jameco and Fritzing).

After you have finished building your circuit, testing the spectrophotometer is necessary to ensure that all the electronic components are connected correctly and your device works as expected.

1. Place one empty cuvette, upside-down, over the LED; and another empty, upside-down cuvette over the photoresistor. If the cuvettes are not clear on all sides, but have two grooved or frosted sides, make sure that you put the clear side facing toward the LED as well as the photoresistor. Bend the LED and photoresistor as needed to fit underneath the cuvettes.
2. Place two empty cuvettes between the LED and the photoresistor. Again, make sure that you always face the clear sides of the cuvette toward the LED and the photoresistor. The four cuvettes should touch each other and form a straight line. You can use clear tape to hold the cuvettes over the LED and the photoresistor in place. But do *not* block the light path!
3. The light from the LED should now shine directly onto the photoresistor, as shown in Figure 4. Bend the wires on the LED and photoresistor for adjustment, if needed.

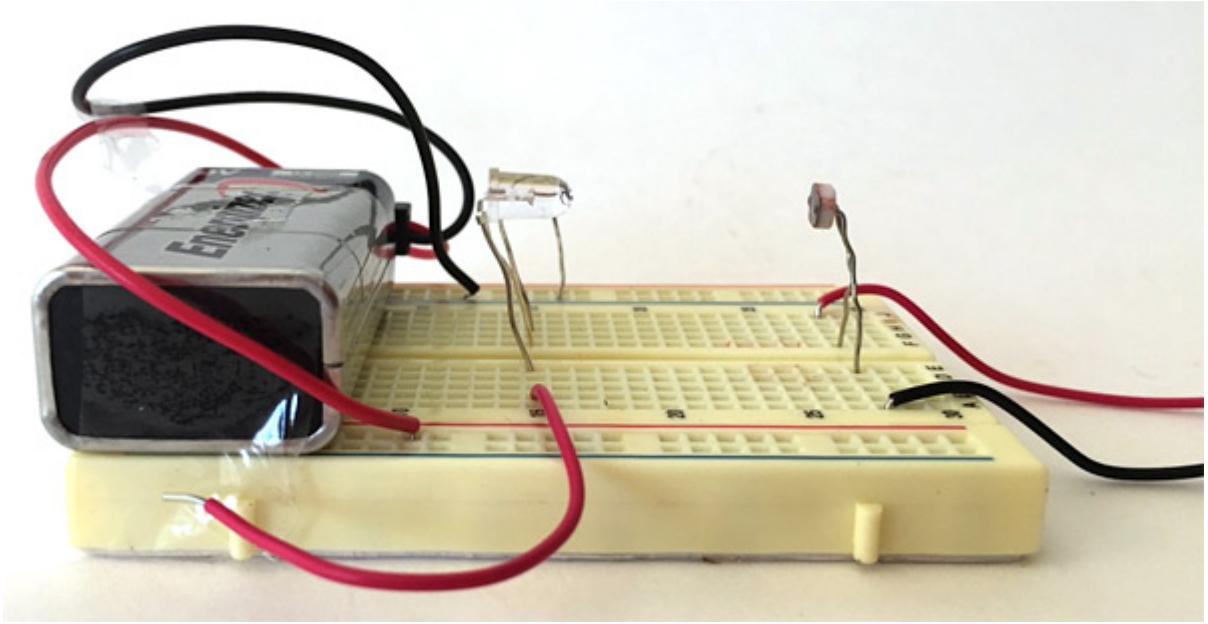


Figure 4. Make sure that the LED and the photoresistor are properly aligned. Note that in this picture, the cuvettes are not yet placed on top of the photoresistor and LED.

4. Set up the multimeter to measure the resistance of the photoresistor.
    a. Plug the black multimeter probe into the port labeled COM.
    b. Plug the red multimeter probe into the port labeled VΩmA.
    c. Turn the dial setting to 200 ohms (Ω).
    d. Turn the power switch to ON.
    e. Use alligator clips to attach the red and black multimeter probes to the red and black jumper wires connected to the photoresistor coming from A28 and J28.
5. Turn on the LED by connecting the jumper wire from B15 to the power (+) bus.
6. Cover the circuit (but not the multimeter) with the cardboard box to block ambient light.
7. Read the resistance across the photoresistor and record it in your lab notebook.
    a. Note the units of the resistance. A "k" indicates kilo-ohms (kΩ) and an "M" indicates mega-ohms (MΩ).
    b. If your multimeter screen displays a "1 .", that means the resistance is too high for the dial setting. Turn the dial up to the next highest range (for example, from 200 to 2000) and check again.
    c. If this is your first time using a multimeter, refer to the Science Buddies resource How to Use a Multimeter (http://www.sciencebuddies.org/science-fair-projects/references/how-to-use-a-multimeter), specifically the section How do I measure resistance? (http://www.sciencebuddies.org/science-fair-projects/references/how-to-use-a-multimeter#qmultimetermeasureresistance), to learn more.
8. Remove the box and turn off the LED by removing the jumper wire from the power (+) bus.

9. Cover the circuit with the box again. In the dark, the resistance should be in the mega-ohm range. Remember that you may need to adjust the dial setting to get a measurement. Record the resistance in your lab notebook. *Note*: Stray light will cause problems with the data. Perform the readings in a dimly lit room if stray light is a problem and/or use a black permanent marker to shield the photoresistor from light from the sides and back of the cuvette.
10. Remove the box and turn off the multimeter to conserve battery power.

Part 2: Calibrating the Spectrophotometer

Now that you know that your spectrophotometer is working, the next step is to make the standard solutions to calibrate it. You will make a series of dilutions of blue dye, as shown in Figure 5, with known concentrations, and measure them with your spectrophotometer to create a **calibration curve**. Each dilution is made by consecutively diluting your solution by half. It is essential to use dye-free utensils and cups to get an accurate set of standards.

Figure 5. Standard solutions of Blue 1 for creating the calibration curve.

1. Set out eight clean cups and label them 1–8.
2. The solutions will be diluted as follows:
    - 1 (most concentrated)
    - 1/2
    - 1/4
    - 1/8
    - 1/16
    - 1/32
    - 1/64
    - Water only
3. Pour 8 oz. of water into the first cup (#1) and 4 oz. of water into the remaining cups (2–8).
4. Mix 1/8 teaspoon (tsp.) of blue dye with the 8 oz. of water in cup #1. *Note*: The concentration of blue dye in the commercial package is approximately 0.026 M (mol/L). After dilution (1/8 tsp in 1 cup = 1:384), the concentration is 68 µM (µmol/L).
5. Stir the contents of cup #1 with a clean spoon.
6. Using the measuring cup, pour 4 oz. from cup #1 into cup #2 and mix with a clean spoon.
7. Thoroughly rinse the measuring cup and spoon and mix 4 oz. from cup #2 with the water in cup #3.
8. Repeat the two-fold dilutions for cups 4–7. Cup #8 will be your "blank," and should not contain any dye.
9. Transfer the blank and the standard solutions into eight clean and labeled cuvettes. Use the eyedropper, a transfer pipette, or pour carefully. *Note*: The cuvettes hold approximately 3 mL of solution.
10. Prepare your orange absorption filter by adding 120 mL or 1/2 cup of water into a clean cup. Add two drops of red and two drops of yellow liquid food coloring and mix the solution well with a clean spoon.
11. Transfer the orange solution into a clean cuvette and place the cuvette next to the LED so that the clear sides face the LED and the photoresistor.

12. Attach the red and black multimeter probes to the red and black wires in contact with the photoresistor (coming from A28 and J28) using the cables with the alligator clips, if they are not yet connected.
13. Set the multimeter to read resistance again. Remember that you might have to adjust the range as you take different readings.
14. First, place your blank sample without dye in between the orange cuvette and the photoresistor, as shown in Figure 6. Again, the clear sides of the cuvettes should face toward the LED and the photoresistor.

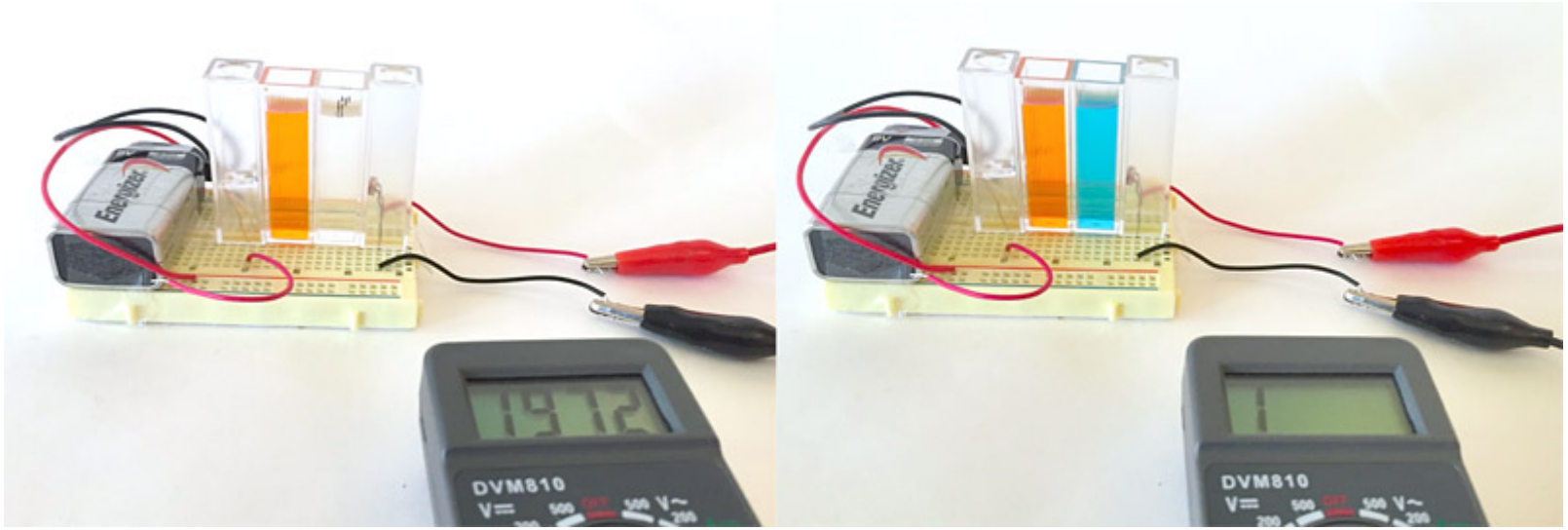


Figure 6. Spectrophotometer setup for measuring your blank (left), standard, and samples (right). Note that in these pictures, the LED is not yet switched on. For your measurements, you also have to cover the spectrophotometer with a cardboard box to block out surrounding light.

15. Plug in the wire to turn on the LED and cover the breadboard with a small cardboard box. Read the resistance on the multimeter and record the data in your lab notebook.
16. Remove the blank cuvette and replace it with the cuvette containing the next standard solution, starting with the lowest concentration. Cover the breadboard again with the cardboard box and write down the resistance for this solution. Continue the measurements for each of your seven standards.
17. Repeat steps 14–16 with the entire set of standards, including the blank, two more times.
18. Make a data table in your lab notebook, showing the dilutions and the concentrations of blue dye in all your standards (#1 = 68 µM, #2 = 34 µM, etcetera) together with all three recorded resistance measurements for each solution. The resistance should be higher as the solutions get darker.

Part 3: Measuring Your Sports Drink Samples

You are now ready to take readings from your spectrophotometer with real sports drink samples.

1. Start with a visual evaluation of each of your blue beverages. Which one do you think contains the most amount of blue dye? Write down your assumptions in your lab notebook.
2. Label as many clean cuvettes as you have sports drinks that you would like to test. Make sure the label is explicit for each drink.
3. Using the clean eyedropper or a transfer pipette, fill each clean cuvette with one of the blue-colored beverages, such as in Figure 7.

Figure 7. Several sports drink samples with unknown concentrations of Blue 1 prepared for measurement on the spectrophotometer.

4. Check that your spectrophotometer setup is still in measuring mode with the leads attached to the multimeter, and both the LED and multimeter switched on. Place the cuvette with the orange solution next to the LED if it is not already there.
5. Place one of your sports drink samples on the device in between the orange filter solution and the photoresistor. The clear sides of the cuvette need to face the LED and the photoresistor.
6. Cover the spectrophotometer with the cardboard box and record the resistance on the multimeter in your lab notebook. *Note*: If the resistance of your solution exceeds the maximum resistance of your calibration curve, dilute your sample and measure again. You can do a 1:2 dilution in a fresh cuvette (1.5 mL water + 1.5 mL sample solution) or a 1:6 dilution (2.5 mL water + 0.5 mL sample solution).
7. Measure your sample two more times.
8. Continue measuring all your sports drink samples on the spectrophotometer and record the resistance for each in your lab notebook. Be sure to measure each sample a total of three times.

Analyzing Your Results

1. Open a spreadsheet and enter the resistance data for your calibration curve. Calculate the average for your three resistance readings for each standard. Subtract the resistance that you measured for the blank from all of the readings you made for samples with dye. This step subtracts the light loss due to the plastic, the water, and other factors.
2. Graph the average resistance of your three readings on the y-axis versus the concentration of the standard solutions in µM on the x-axis. *Note*: If you are using Microsoft Excel, use the "Scatterplot" chart. Excel also has tools for adding trend lines.
3. More-advanced students can add a trend line to the data and display its equation and its correlation factor R^2.
4. Graph the average resistance of each of your blue beverages on the chart.
5. Determine the concentration of blue dye in your sports drink samples, based on where they are on the graph or use your calibration curve to calculate the concentration of blue dye in your blue beverages. Remember to account for your dilutions if a sample had to be diluted.
6. Which of the beverages had the highest concentration of blue dye? Do your results agree with your visual evaluation of the sports drinks?

Frequently Asked Questions (FAQ)

FAQ for this Project Idea available online at https://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p075/chemistry/measure-blue-dye-sports-drinks#help (http://www.sciencebuddies.org/science-fair-projects/project-ideas/Chem_p075/chemistry/measure-blue-dye-sports-drinks#help).