

Act. conc. =  
1071 mM

107 mM

1.226 w/ 6040/ade

**Table 1. % Transmittance of diluted solutions of known concentration**

Solution	Dilution Ratio mL stock/mL Water	Molar Concentration ( $\mu\text{M}$ )	Measured % Transmittance	Measured Transmittance as a Decimal
1. (stock solution)	10 mL/0 mL	520 mM	98.2%	.982
2.	8 mL/2 mL	416 mM	81.3%	.813
3.	6 mL/4 mL	312 mM	75.3%	.753
4.	4 mL/6 mL	209 mM	40.6%	.406
5.	3 mL/7 mL	156 mM	33%	.330
6.	2 mL/8 mL	104 mM	18.7%	.187
7.	1 mL/9 mL	52 mM	11.3%	.113
8.	0 mL/10 mL	0 mM	0.000%	0.000%

$M_1 V_1 = M_2 V_2$ ;  $(520 \text{ mM}) V_1 = M_2$

$\frac{520 \times 3}{10} =$   
 $\frac{1560}{10} =$   
156

Step 6: It would be really useful if the class got a straight line that goes through zero. Scientists try to find linear relationships because such relationships make it easier to identify unknowns and predict outcomes of investigations. It is often helpful to have the slope of the relationship between the dependent and independent variable be positive. What mathematical routine could we choose to plot to get a linear relationship between transmittance and molarity that has a positive slope, and which line goes through zero? Try out some of these techniques and report back to the whole group:

1/T versus [dye]

$1 \times 10^T$  versus [dye]

log T versus [dye]

$-\log T$  versus [dye]

$y = 0.00195x + 0.0171$

$r^2 = .977$

■ Explanation to Strengthen Student Understanding

Transmittance and Absorbance Relationship

The relationship between transmittance and absorbance is  $Abs = -\log_{10}(T)$  where T is the transmittance, rather than the %T (i.e., 0.50, not 50%). When the absorbance is 1.0, only 10% of the light beam is reaching the detector. When  $Abs = 2$ , only 1% of the light beam is reaching the detector and when  $Abs = 3$ , only 0.1% of the light is reaching the detector. It should be no surprise that the accuracy and sensitivity of low-cost instruments start to suffer at absorbance values higher than 1.5. If a sample shows an absorbance higher than 1.5, it is a good idea to dilute the sample by a factor of 5 or 10 and remeasure. The extra work is paid back in better accuracy in your measurement. A useful rule is don't accept measurements of  $>1.5Abs$ . If you get such an absorbency value, dilute the sample and measure it again.

■ Investigation

Many common sports drinks contain blue #1 dye. Students will use the relationship between transmittance, absorbance, and concentration (as well as their calibration line from the prelab) to determine the concentration of this dye in the sports drink.

Procedure

Obtain a sample of the blue-colored sports drink. Use the skills and information gained in the prelab to design a data-collection and data analysis procedure to determine the molarity or concentration of blue #1 dye in the sports drink.

Data Collection and Computation

1. Determine the molar concentration of blue #1 dye in the sports drink. Show all work.

2. Determine the mass of blue #1 dye found in 500 mL of the drink. Show all work.

$y = 0.00195x + 0.0171$  (226)  $= 0.001954 + 0.0171 = x = 107$   $x = 107$   
 $65 \times 1071 = 535.5 \text{ mol} \quad 792.85 \text{ g}$   $\neq 424571.175 \text{ g FD\&C blue \#1}$

### Argumentation and Documentation

In the conclusion of lab, have students justify the procedure they chose, the instrumentation they used, and the selection of the kind of data they decided was needed to determine the concentration of blue #1 dye in a blue-colored sports drink containing only this dye.

### Postlab Assessment

1. Suppose a solution was too concentrated for an accurate reading with the spectrophotometer. The concentrated solution was diluted by placing 1.00 mL of the concentrated solution in 4.00 mL of water. The solution was then placed in the spectrophotometer and an absorbance was obtained and after a few calculations the molar concentration was calculated to be  $3.5 \times 10^{-6} \text{ M}$ . What was the concentration of the original stock solution before dilution?

$\frac{1}{5} = \frac{3.5 \times 10^{-6}}{x}$   $x = 1.75 \times 10^{-5} \text{ M}$

2. If a 0.10 M solution of a colored substance has a maximum absorbance at 500 nm and an absorbance of 0.26 M at this wavelength, what will be the measured absorbance of a 0.20 M solution at 500 nm?

3. The spectrophotometer really measures the percent of light that is transmitted through the solution. The instrument then converts %T (transmittance) into absorbance by using the equation you determined in the prelab section. If the absorbance of a sample is 0.85, what is the percent of light transmitted through the colored sample at this collected wavelength?