Colorimeter

(Order Code COL-BTA)

The Vernier Colorimeter is designed to determine the concentration of a solution by analyzing its color intensity. The color of a solution may be inherent or derived by adding another reagent to it. The Colorimeter measures the amount of light transmitted through a sample at a user-selectable wavelength. Using the front panel arrow keys, you may choose from four wavelengths: 430 nm, 470 nm, 565 nm, and 635 nm. Features such as automatic sensor identification and one-step calibration make this sensor easy to use.

How the Colorimeter Works

Light from a LED light source passes through a cuvette containing a solution sample, as shown in Figure 1. Some of the incoming light is absorbed by the solution. As a result, light of a lower intensity strikes a photodiode.

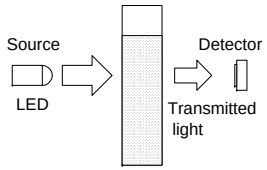


Figure 1

Important Software and LabPro/CBL 2 OS Updates!

1. **If you are using Logger *Pro*® computer software,** you will want to update your Logger *Pro* Colorimeter experiment files. The easiest way to obtain these is to update Logger *Pro* to Logger *Pro* 3[1].
2. **If you are using a calculator *and* the firmware version on your LabPro or CBL 2™ is older than 6.23 (dated 08/22/02),** you will need to update the operating system. You can do this by going to our web site, www.vernier.com, select "Downloads," and select "LabPro and CBL 2 operating system updates."

Transmittance and Absorbance

The amount of light that passes through a solution is known as transmittance. Transmittance can be expressed as the ratio of the intensity of the transmitted light, It, and the initial intensity of the light beam, Io, as expressed by the formula

$$T = I_t / I_o$$

The Colorimeter produces an output voltage which varies in a linear way with transmittance, allowing a computer, calculator, or handheld to monitor transmittance data for a solution. The reciprocal of transmittance of the sample varies logarithmically (base ten) with the product of three factors: ε, the molar absorptivity of the solution, b, the cell or cuvette width, and C, the molar concentration

$$\log(1/T) = \varepsilon bC$$

[1] If you are using Logger *Pro* 2, update to version 2.2.1. You can do this for free on our web site at www.vernier.com.

In addition, many experiments designed to use a Colorimeter require a related measurement, *absorbance.* At first glance, the relationship between transmittance and absorbance would appear to be a simple inverse relationship; that is, as the amount of light transmitted by a solution increases, the amount of light absorbed might be expected to decrease proportionally. But the true relationship between these two variables is inverse *and* logarithmic (base 10). It can be expressed as

$$A = \log(1/T)$$

Combining the two previous equations, the following expression is obtained:

$$A = \varepsilon b C$$

In effect, this formula implies that the light absorbed by a solution depends on the absorbing ability of the solute, the distance traveled by the light through the solution, and the concentration of the solution. For a given solution contained in a cuvette with a constant cell width, one can assume ε and *b* to be constant. This leads to the equation:

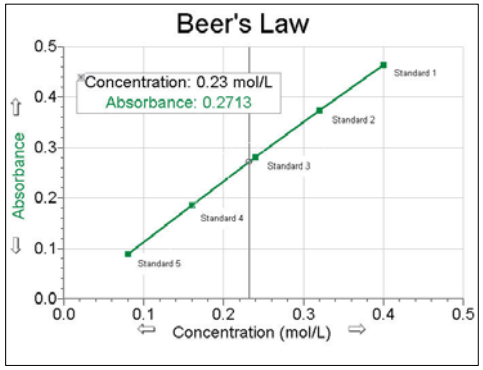
$$A = k \bullet C \quad \text{(Beer's law)}$$

where *k* is a proportionality constant. This equation shows absorbance to be related directly to concentration and represents a mathematical statement of Beer's law. Beer's law is discussed in more detail below.

In this guide and in some of our computer programs, transmittance is expressed as percent transmittance or %T. Since T=%T/100, the formula can be rewritten as

$$A = \log(100/\%T) \text{ or } A = 2 - \log \%T$$

Beer's Law



In general, absorbance is important because of its direct relationship with concentration according to Beer's law. Many experiments in chemistry and biology are based on this concept. To obtain a Beer's law curve, several standards (solutions of known concentration) are prepared and their absorbance values are determined using a Colorimeter. A graph of absorbance *vs.* concentration is then plotted. A solution of unknown concentration is placed in the colorimeter and its absorbance measured. When the absorbance of this solution is interpolated on the Beer's law curve, as shown on the previous page, its concentration is determined on the horizontal axis. Alternatively, its concentration may be found using the slope of the Beer's law curve.

Using the Colorimeter

The Colorimeter is easy to use and maintain. Simply connect it to your data collection interface, configure your software, and you are ready to make measurements. For best results, let the system stabilize at the desired wavelength for 5 minutes prior to calibration or data collection.

Wavelength Selection

You can select one of four LED light wavelengths with the Vernier Colorimeter; violet (430 nm), blue (470 nm), green (565 nm), and red (635 nm). You can select one of these nearly monochromatic colors using the wavelength selection arrows on the top of the Colorimeter (shown here).

There are several ways you can decide which of the four wavelengths to use.

- Look at the color of the solution. Remember that the color of a solution is the color of light that passes through it. You want to use a different color of light that will be absorbed, rather than transmitted; for example, with a blue copper (II) sulfate ($CuSO_4$) solution, use the red LED (635 nm).
- Another easy method is to place a cuvette containing the solution in question in the Colorimeter and check to see which of the wavelengths yields the highest absorbance.
- Directions for most colorimetry experiments express a recommended wavelength. Use the wavelength closest in value to the recommended wavelength. Even if the LED wavelength is somewhat different, a Beer's law curve can usually be obtained at almost any wavelength in the vicinity of the recommended wavelength.

Collecting Data with the Colorimeter

This sensor can be used with the following interfaces to collect data:

- Vernier LabQuest® as a standalone device or with a computer
- Vernier LabQuest® Mini with a computer
- Vernier LabPro® with a computer, TI graphing calculator, or Palm® handheld
- Vernier Go!®Link
- Vernier EasyLink®
- Vernier SensorDAQ®
- CBL 2™

Here is the general procedure to follow when using the Colorimeter:

1. Connect the Colorimeter to the interface.
2. Start the data-collection software[2].
3. The software will identify the Colorimeter. Proceed to Step 4 to calibrate the Colorimeter

The Colorimeter needs to be powered about 5 minutes before calibrating in Step 4. One of the four green wavelength indicator lights will be turned on when it is powered.

4. Calibrate the Colorimeter.
   a. Press the < or > button on the Colorimeter to select the correct wavelength setting for your experiment (430 nm, 470 nm, 565 nm, or 635 nm).
   b. Open the Colorimeter lid.
   c. Insert a cuvette, usually filled with distilled water, for your blank cuvette (100% transmittance or 0 absorbance). **Important:** Line up one of the *clear* sides of the cuvette with the arrow at the *top* of the cuvette slot. Close the Colorimeter lid.
   d. Next, press the CAL button to begin the calibration process. Release the CAL button when the red LED begins to flash. The absorbance should now be 0.000 or 0.001
   e. When the LED stops flashing, the calibration is complete and your unit is ready to collect data.

Arrow

Important: Unlike older versions of the Colorimeter, with this model you do not need to go to a special calibration menu in our data-collection programs.

5. Collecting data.
   a. There are two common modes for Colorimeter data collection:
      - Absorbance *vs.* concentration (Beer's law) – If you want to collect data in Events with Entry mode, you can open a different Logger *Pro* Colorimeter file (with a computer). With calculators or handhelds, you will need to change from Time Graph to Events with Entry mode.
      - Absorbance *vs*. time – When the Colorimeter is automatically identified by Logger *Pro*, it will already be set up to collect in this mode. With calculators or handhelds, auto-ID will set the mode to Time Graph.
   b. Place the cuvette with a sample into the Colorimeter cuvette slot. **Important:** Line up one of the clear sides of the cuvette with the arrow at the *top* of the cuvette slot.
   c. Begin collecting data (choose Collect or Start in the program).

[2] If you are using Logger *Pro* 2 with either a ULI or SBI, the sensor will not auto-ID. Open an experiment file for the Colorimeter in the Probes & Sensors folder.

- In absorbance *vs*. concentration experiments, you will be prompted to *keep* the absorbance value (when it stabilizes), and *enter* the concentration of the standard solution. Repeat the process for the remaining standards.
- In absorbance *vs*. time experiments, readings will be taken in real time for the amount of time set up in the data-collection program.

d. Data collection will end when you choose Stop, or when the pre-set experiment length has been reached.

e. After data collection is completed, you may use some of the tools in our data collection programs to analyze the collected data. For example, in absorbance *vs*. concentration (Beer's law) experiments, you can perform a linear fit on the data, then interpolate along the resulting linear fit to determine the concentration of an unknown.

Data-Collection Software

This sensor can be used with an interface and the following data-collection software.

- **Logger *Pro* 3** This computer program is used with LabQuest, LabQuest Mini, LabPro, or Go!Link.
- **Logger *Pro* 2** This computer program is used with ULI or Serial Box Interface.
- **Logger Lite** This computer program is used with LabQuest, LabQuest Mini, LabPro, or Go!Link.
- **LabQuest App** This program is used when LabQuest is used as a standalone device.
- **EasyData App** This calculator application for the TI-83 Plus and TI-84 Plus can be used with CBL 2, LabPro, Vernier EasyLink. We recommend version 2.0 or newer, which can be downloaded from the Vernier web site, www.vernier.com/easy/easydata.html, and then transferred to the calculator. See the Vernier web site, www.vernier.com/calc/software/index.html for more information on the App and Program Transfer Guidebook.
- **DataMate program** Use DataMate with LabPro or CBL 2 and TI-73, TI-83, TI-84, TI-86, TI-89, and Voyage 200 calculators. See the LabPro and CBL 2 Guidebooks for instructions on transferring DataMate to the calculator.
- **Data Pro** This program is used with LabPro and a Palm handheld.
- **LabVIEW** National Instruments LabVIEW™ software is a graphical programming language sold by National Instruments. It is used with SensorDAQ and can be used with a number of other Vernier interfaces. See www.vernier.com/labview for more information.

Absorbance and Transmittance Ranges for the Colorimeter

For best results, our laboratory testing of the colorimeter indicates that absorbance or transmittance values should fall within these ranges:

percent transmittance:	10% – 90%
absorbance:	0.05 – 1.0

We have found that Beer's law experiment results begin to lose their linearity at absorbance values above 1.0 (percent transmittance values less than 10%). If you have a solution that transmits such a low level of light, consider diluting the solution so that it falls within this range.

Using cuvettes with the Colorimeter

The Colorimeter is designed to use polystyrene cuvettes. Fifteen of these cuvettes and lids are supplied with the Colorimeter. The cuvettes have a volume of approximately 4 mL. Two opposite sides of the cuvette are ribbed and are not intended to transmit the light from the LED. The two smooth surfaces are intended to transmit light. It is important to position the cuvette correctly in the Colorimeter, with a smooth side facing the arrow at the back of the slot, and with the ribbed edges facing left and right. The light travels from the LED at the top, through the cuvette, to the detector below the slot.

Just like most spectrophotometer sample tubes, individual plastic cuvettes vary slightly in the amount of light they absorb. You may choose to ignore these differences. For most lab exercises, this variation will not have a noticeable effect on experimental results.

For best results, variation in light absorbed by individual cuvettes can be controlled either by using the same cuvette for all trials of a particular experiment or by *matching* a set of cuvettes. The easiest and most reliable is the first method. If a student plans to use five trials for a Beer's law experiment, the five standard solutions can be transferred to the same cuvette for each trial. This requires that the cuvette be clean and dry after each trial *or* rinsed several times with the solution that will be added to it.

This method takes very little time and successfully controls a potential variable. It also eliminates concerns over possible scratches that may eventually develop on a cuvette. The effect of the same small scratch is eliminated using the 100% calibration.

As an alternative, you may choose to match cuvettes. Matched cuvettes are a set of cuvettes that all absorb light (when empty) at approximately the same level. This involves more work on the part of the teacher, but saves time in student procedures. If students have 5 or 6 cuvettes with similar absorbance levels, then each sample can be added to a different cuvette, eliminating the drying or rinsing step described in the previous paragraph.

Caps are supplied for the original 15 cuvettes. A cuvette may or may not have a cap on it when placed in the Colorimeter. The purpose of the cap is to prevent evaporation of solvent when an experiment is run over a period of several days. You may find it convenient to store standard solutions in capped cuvettes. If you purchase a replacement set of 100 cuvettes, 20 caps will be included. We felt teachers would probably not need to have one cap per cuvette. The caps can certainly be reused as cuvettes are replaced. Replacement cuvettes may be purchased using the order code CUV. This package includes 100 cuvettes and 20 caps.

This sensor is equipped with circuitry that supports auto-ID. When used with LabQuest, LabQuest Mini, LabPro, Go! Link, SensorDAQ, EasyLink, or CBL 2, the data-collection software identifies the sensor and uses pre-defined parameters to configure an experiment appropriate to the recognized sensor.

Specifications

Colorimeter range:	0 to 3 (absorbance)
Useful Range:	0.05 to 1.0 absorbance (90% to 10% T)
Wavelengths:	430 nm, 470 nm, 565 nm, 635 nm
13-bit resolution (SensorDAQ)	0.018 %T
12-bit resolution (LabQuest, LabQuest Mini, LabPro, ULI II, SBI):	0.035 %T
10-bit resolution (CBL 2):	0.14%T
Supply voltage:	5VDC ±25 mV
Supply current (typical):	40 mA
Power up time:	700 ms (maximum)
Output voltage range:	0–4 V
Transfer function;	$V_{out} = 0.035*(\%T) + 0$
Stored Calibration Values:	Slope: 28.571
	Intercept: 0

Using Vernier Sensors with Other Interfaces

Our sensors may be used with interfaces from manufacturers other than TI or Vernier Software & Technology. Some interfaces may not use the same connectors. Please contact the interface manufacturer for an adapter or go to www.vernier.com/probes/specs/pinout.html for pin assignments and other information. Interfaces that have power saving features (typically battery powered units) may cause problems with sensors that require a longer warm-up period (>1 sec). If available, select a constant power option for these sensors.

Extending the Length of the Cable

The cable length may be increased by using an extension cable (order code EXT-BTA). These cables are 2 m in length and allow you to extend the sensor farther from the interface. If longer cabling is needed, contact Vernier for assistance.

Ordering Information

Replacement Cuvettes (pkg of 100 with 20 lids)……CUV

Suggested Experiments

Beer's Law

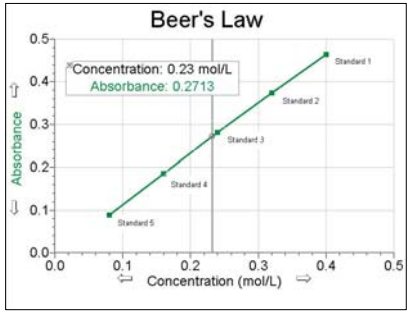
- **Crystal Violet:** Dilute solutions of crystal violet yield a good Beer's law curve using the green LED (565 nm). A stock solution of 2.5×10^{-5} M crystal violet is prepared by adding 0.020 g of solid crystal violet to enough water to yield 2 liters of solution. Dilute to obtain standard solutions.
- **Copper Sulfate:** Standard solutions that are 0.1, 0.2, 0.3 and 0.4 M $CuSO_4$ will yield a good Beer's law curve at 635 nm (red LED). Prepare a stock solution by adding 10 g of NH_4NO_3 to 10 mL of 0.1 M $CuSO_4$ and 90 mL of 0.20 M NH_3 (forms the $Cu(NH_3)_4^{2+}$ complex ion) and dilute to obtain standard solutions.
- **Food coloring Solutions: Red, Blue, Green:** A less expensive alternative to using the solutions above is to prepare solutions using food coloring. We have obtained very good Beer's law curves using these solutions. We added about 6 drops of red, blue or green McCormick brand food coloring to 1 liter of water The red solution can be analyzed using the blue LED (470 nm), the green solution with the blue LED (470 nm) or the red LED (635 nm), and the blue solution with the red LED (635 nm). Since the actual concentration of the solutions will not be known, refer to the original solution as "100%" and then dilute to 80, 60, 40, and 20%. Check the original solution to see that its absorbance is not greater than 1.0.

You can find detailed instructions for the following experiments in the Vernier lab books listed with each experiment.

Experiment 11, *Chemistry with Vernier*

Determining the Concentration of a Solution: Beer's Law

This experiment uses standard and unknown solutions of $NiSO_4$ (or green food coloring) using the Vernier Colorimeter. Use the red LED (635 nm). Data from this experiment is shown here using the Logger *Pro* program. Note that our programs allow you to determine the concentration of an unknown sample by interpolating its absorbance value along the regression curve.



Experiment 20, *Chemistry with Vernier*

Chemical Equilibrium: Finding a Constant, K_c

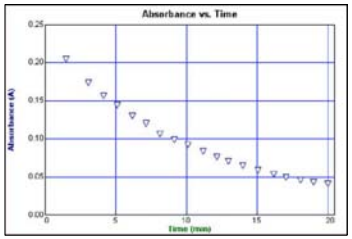
Our lab books contain an experiment for determining the equilibrium constant for this well-known reaction in chemistry.

$$Fe^{3+}(aq) + SCN^{-}(aq) \longleftrightarrow FeSCN^{2+}(aq)$$

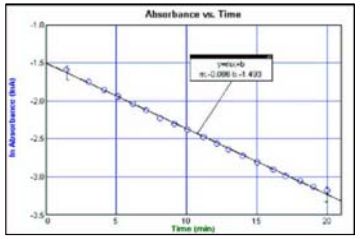
Experiment 30, *Chemistry with Vernier*

Determination of the Rate Law for Reaction of Crystal Violet

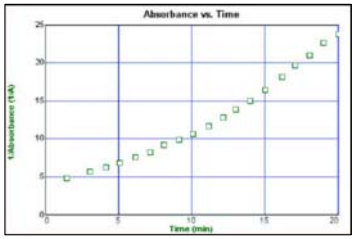
The data here were obtained by reacting 10 mL of 2.5×10^{-5} M crystal violet solution and 10.0 mL of 0.10 M NaOH. The first graph is absorbance *vs*. time. The next graph shown is the natural log of absorbance *vs*. time, showing the reaction to be first order with respect to crystal violet.



absorbance vs. time: reaction is not zero order



ln absorbance vs. time: reaction is first order



1/absorbance vs. time: reaction is not second order

Tests 7 and 8, *Water Quality with Vernier*

(Ortho Phosphates, Total Phosphates, and Nitrates)

To determine the concentration of an ion in a colorless solution using a colorimeter, an agent must be added to the solution to yield color (such as a colored complex ion) or turbidity through the formation of a precipitate The assumption is that the intensity of the color (and its resulting ability to absorb light from the LED) is proportional to the concentration of the ion in solution. Hach Company markets pre-massed *pillows* for analysis of such ions as nitrate (NO_3^-), and phosphate (PO_4^{3-}), as well as many other colorimetric tests. Water Quality tests for these ions using a Vernier Colorimeter are described in our Water Quality lab books.

Experiment 7, *Biology with Vernier*

Photosynthesis

In this experiment, students monitor the progress of photosynthesis using a blue dye (2,6-dichlorophenol-indophenol, or DPIP). As photosynthesis proceeds, the dye turns from blue to colorless when reduced.

Experiment 8, *Biology with Vernier*

The Effect of Alcohol on Biological Membranes

Students see the effect of different alcohols on beet cell membranes by examining the amount of red pigment released with the Vernier Colorimeter.

Experiment 13, *Biology with Vernier*

Biological Membranes

In this experiment, students determine the stress of various factors (osmotic balance, detergents, or pH) on biological membranes. The absorbance of light is used to monitor the extent of cellular membrane damage.

Experiment 9, *Biology with Vernier*

Population Dynamics

In this experiment, students monitor the growth in yeast populations using a Colorimeter.

Warranty

Vernier warrants this product to be free from defects in materials and workmanship for a period of five years from the date of shipment to the customer. This warranty does not cover damage to the product caused by abuse or improper use.

Measure. Analyze. Learn.™

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