

COLORIMETRY



EiMAS
Institut Alam Sekitar Malaysia
Environment Institute of Malaysia



Colorimetry - Take Home Messages

- ◆ Colorimetry is the measurement of color.
- ◆ The intensity of the color relates to the concentration in the sample, based on the instrument's calibration.
- ◆ In a comparison measurement, consistency is key.



Colorimetry - Outline

- ◆ Colorimetry Theory
- ◆ How Does the Spectrophotometer Read a Sample?



What is Colorimetry?

- ◆ Colorimetry is using color intensity measurements to determine the amount of a substance dissolved in solution.
- ◆ Measure samples with a colorimeter or spectrophotometer



Running the Test

- ◆ Pour sample into a clean sample cell
 - Rinse cell with sample before filling to volume





Running the Test

- ◆ Add reagents to the sample, as indicated in the procedure





What do the Reagents Do?

Reagents + Sample



Colored Complex





Running the Test

◆ Reaction time

- Wait the amount of time specified in the procedure for reagents to react with sample and form color





Using the Instrument

◆ Zero the instrument

➤ Sample Blank

- Cell filled with sample, no reagents added

OR

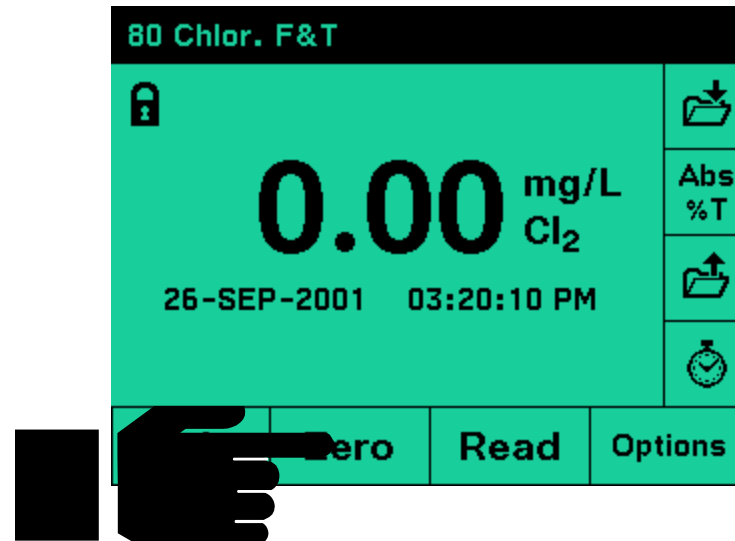
➤ Reagent Blank

- Cell filled with DI water to which reagents are added



Zero the Instrument

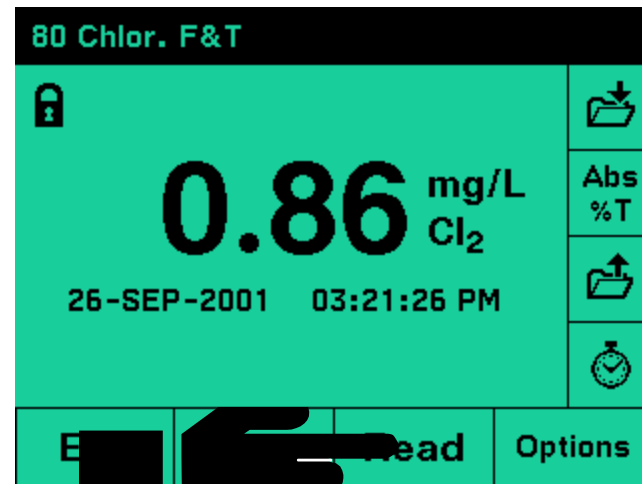
- ◆ Wipe sample cell clean, place into instrument, and touch the Zero key





Read the Sample

- ◆ After the reaction period, wipe sample cell clean, place into instrument, and touch the Read key



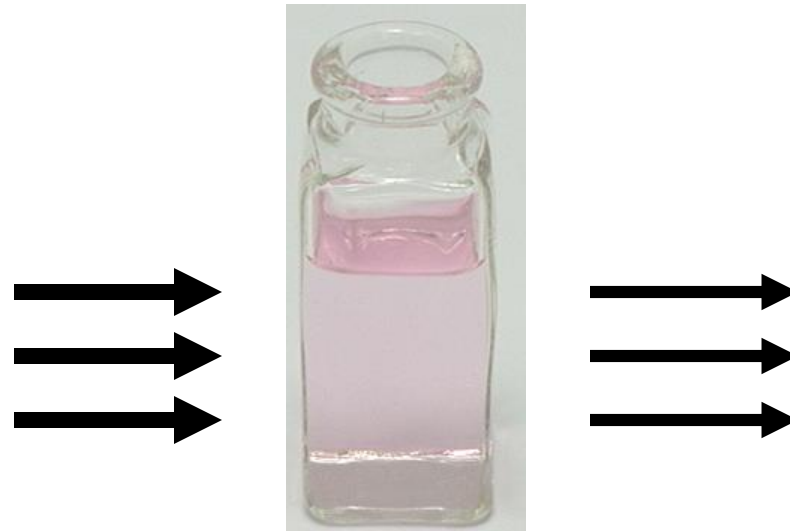


Spectrophotometers

- ◆ Measure the intensity of light passing through a colored sample
- ◆ Convert light intensity measurements to concentration



Spectrophotometers

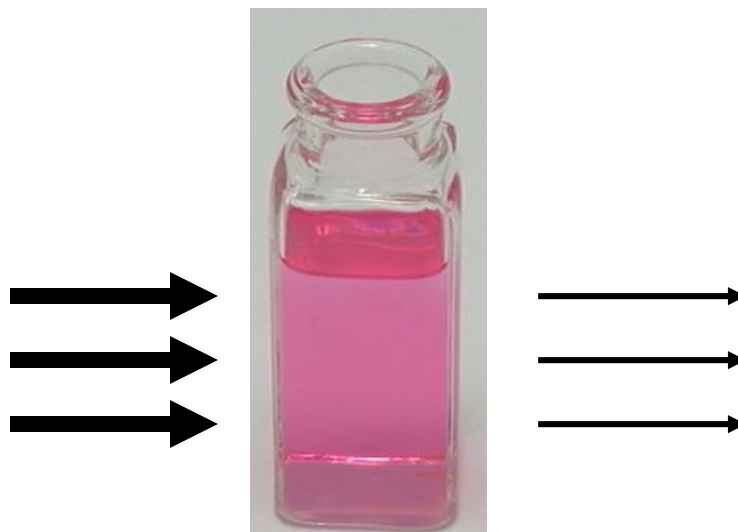


In this case, the sample absorbs very little of the incident light.

It is a low concentration sample.



Spectrophotometers



In this case, most of the incident light is absorbed by the sample.

It is a high concentration sample.

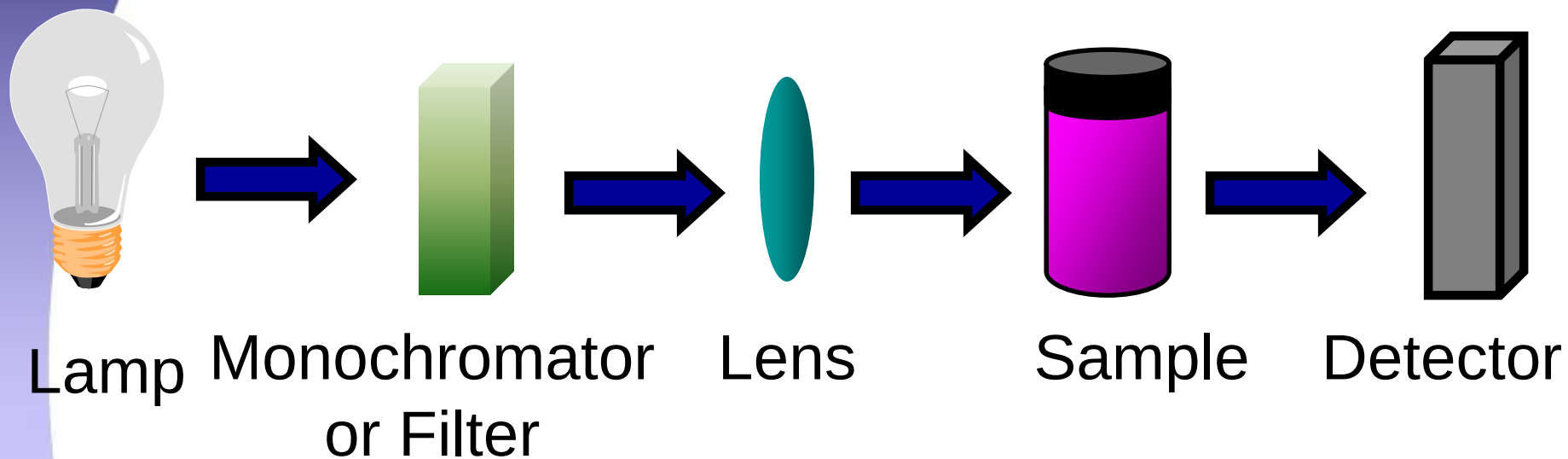


Spectrophotometers

- ◆ Generate light energy
- ◆ Select a specific wavelength of light
- ◆ Pass the light beam through a sample
- ◆ Measure the change in intensity of the light
- ◆ Convert change in light intensity to a displayed concentration



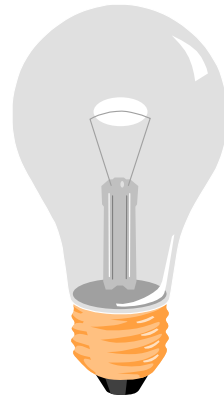
Basic Spectrophotometer





Light Source

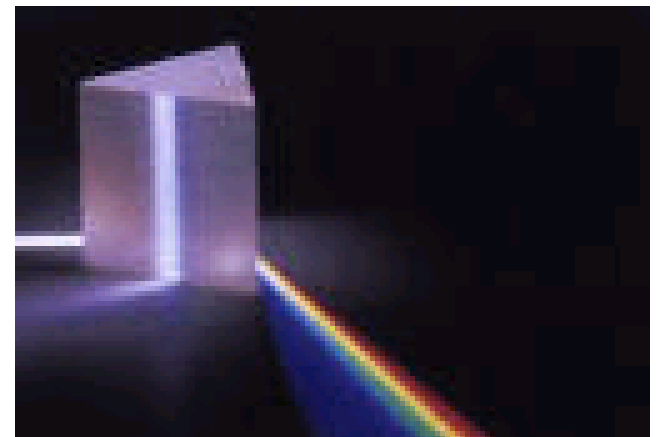
- ◆ Generates light energy
 - Typically tungsten lamp
 - Produces white light - a mixture of all colors





Monochromator/Filter

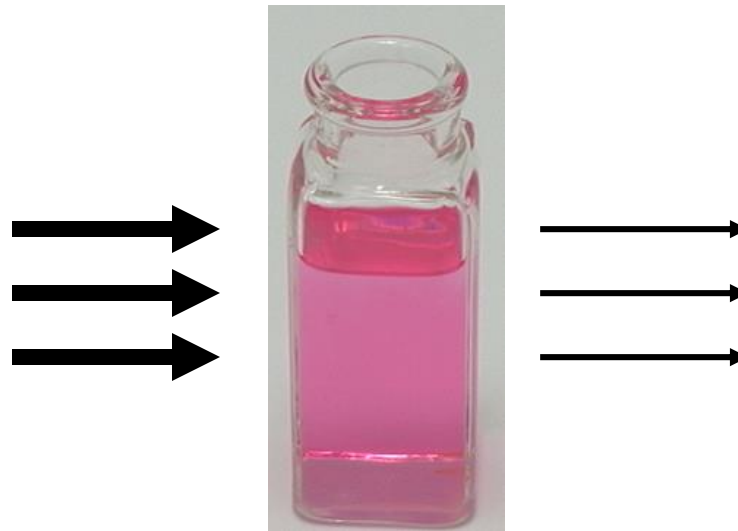
- ◆ Converts incoming white light to a single optimal wavelength for the test
 - Prism
 - Diffraction Grating
 - LED





Sample Cell

- ◆ After wavelength is selected, light passes through the sample cell





Sample Cell

- ◆ Light passes through the sample cell, so it is important that the cell is clean.
 - Clean inside - acid or reagent
 - Clean outside - lint free cloth
 - No dust, fingerprints, scratches
 - Cells are sufficiently filled





Detector

- ◆ Detector measures the amount of light absorbed by the sample
 - Converts light energy to electrical energy



Instrument Reading

- ◆ Software calculates a concentration reading from the detector measurement using an internal calibration curve
 - Calibration curve converts a measured light absorbance into concentration



COLORIMETRY

