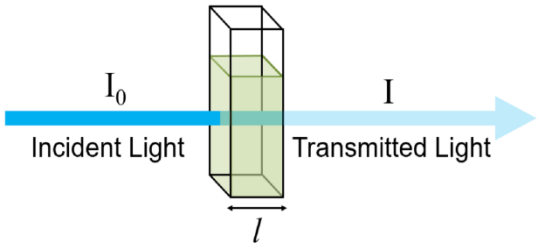
EXPERIMENT 1

AIM: To verify Lambert-beer's law for $KMnO_4$ colorimetrically.

THEORY: The Beer-Lambert law states that the absorbance of a solution is directly proportional to the concentration of the absorbing species in the solution and the path length. Thus, for a fixed path length (cuvette length), UV/Vis spectroscopy can be used to determine the concentration of the absorber in a solution. The absorbance changes with concentration, A higher concentration of the colored solution absorbs more light (and transmits less) than a solution of lower concentration.

According to Beer–Lambert law,



$$\log (I_o / I_t) = A = \varepsilon c l$$

where

I_o and I_t are the incident and transmitted intensities,

A = **absorbance** and

ε is a constant *i.e.* **absorptivity** (formerly called the *extinction coefficient*). If the concentration is measured in $molL^{-1}$, the absorptivity is called the *molar absorptivity* .

$$\mathbf{A = \varepsilon c l}$$

At constant length

$$\mathbf{A \alpha c}$$

REQUIREMENTS

Colorimeter, cuvette, six test tubes

Two Burettes or graduated cylinders two 100 mL beakers

0.01M $KMnO_4$ solution

distilled water, test tube rack, stirring rod, tissues (preferably lint-free)

PROCEDURE

(a) Determination of λ_{max}

(b) Absorbance of different concentration solution at λ_{max}

(a) Calculation of λ_{max}

This can be taken from references (tables of molar extinction coefficients), or more accurately, determined from a calibration curve.

i. Prepare 100 ml, 0.01M **$KMnO_4$** (**Molecular weight 158.03 gm/mol**) solution (stock solution), fill it in a burette

ii. Switch on the computer **and/or** the instrument powers; wait for 30 minutes for 'warm-up' of the instrument.

iii. In the instrument one can choose % transmittance or absorbance display, wavelength range of interest, etc.

iv. Take one clean and dry glass (only for visible range scan) or quartz cuvette with a given path length (say, 1 cm). Prepare a *blank* by filling a cuvette 3/4 full with distilled water

Note: To correctly use cuvettes, remember:

- Wipe the outside of each cuvette with a lint-free tissue.
- Handle cuvettes only by the top edge of the ribbed sides.
- Dislodge any bubbles by gently tapping the cuvette on a hard surface.
- Always position the cuvette so the light passes through the clear sides.

v. By using blank cuvette (with distilled water) calibrate the colorimeter (absorbance = 0, transmittance =100 %) at **filter 1** and putout the blank cuvette.

vi. Then fill another cuvette with the stock solution and measure absorbance, note the reading in table.

vii. Repeat the step v and vi for all the filters (**wave length range**)

viii. From table we can see the **λ_{max}** *i.e.* wave length at which solution show maximum absorbance/ O.D.

Observation Table for calculating the λ_{max}

S.No	Filter	Wavelength/ Range	O.D./ Absorbance
1	1	420	
2	2	440	
3	3	490	
4	4	520	
5	5	540	
6	6	570	
7	7	600	
8	8	650	
9	9	700	

(b) Absorbance of different concentration solutions

- ✓ Obtain small volumes 0.01M $KMnO_4$ (**Molecular weight 158.03 gm/mol**) of solution and distilled water in separate beakers, fill in the separate graduated burettes
- ✓ Label five clean, dry, test tubes 1–5. Use Burettes to prepare five standard solutions according to the chart below. Thoroughly mix each solution with a stirring rod. Clean and dry the stirring rod between uses.

Concentration can be calculate by $M_1V_1 = M_2V_2$

1 2 3 4 5

Test Tube	0.01M KMnO4. (mL)	Distilled H2O (mL)	Concentration(M)
1	2	8	0.002
2	4	6	0.004
3	6	4	0.006
4	8	2	0.008
5	~10	0	0.0100

- ✓ You are now ready to collect absorbance-concentration data for the five standard solutions.
- ✓ Switch on the computer **and/or** the instrument powers; wait for 30 minutes for 'warm-up' of the instrument.
- ✓ In the instrument one can select light sources (UV and visible), choose the slit width, scan speed and % transmittance or absorbance display, wavelength range of interest, etc.
- ✓ Take two clean and dry glass (only for visible range scan) or quartz cuvettes with a given path length (say, 1 cm). Prepare a *blank* by filling a cuvette 3/4 full with distilled water and the other cuvette with aqueous KMnO4 solution with lowest concentration.
- ✓ Read the absorbance value displayed in the meter. When the displayed absorbance value stabilizes, record its value in your data table.
- ✓ Repeat the procedure for Test Tubes 2 to 5.Similarly spectral runs are done for all the other samples starting from the lowest concentrations to next higher concentrations of $KMnO_4$. Every time one should rinse the cuvette taking a small portion of the solution to be analyzed next.
- ✓ Plot a curve between Absorbance v/s concentrations. Check whether it is a liner plot or not

Data Table/ Observation Table

Test Tube	0.01M KMnO4. (mL)	Distilled H_2O (mL)	Concentration(M)	Absorbance
	0	**10**	**0.00**	
1	2	8	0.002	
2	4	6	0.004	
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4	8	2	0.008	
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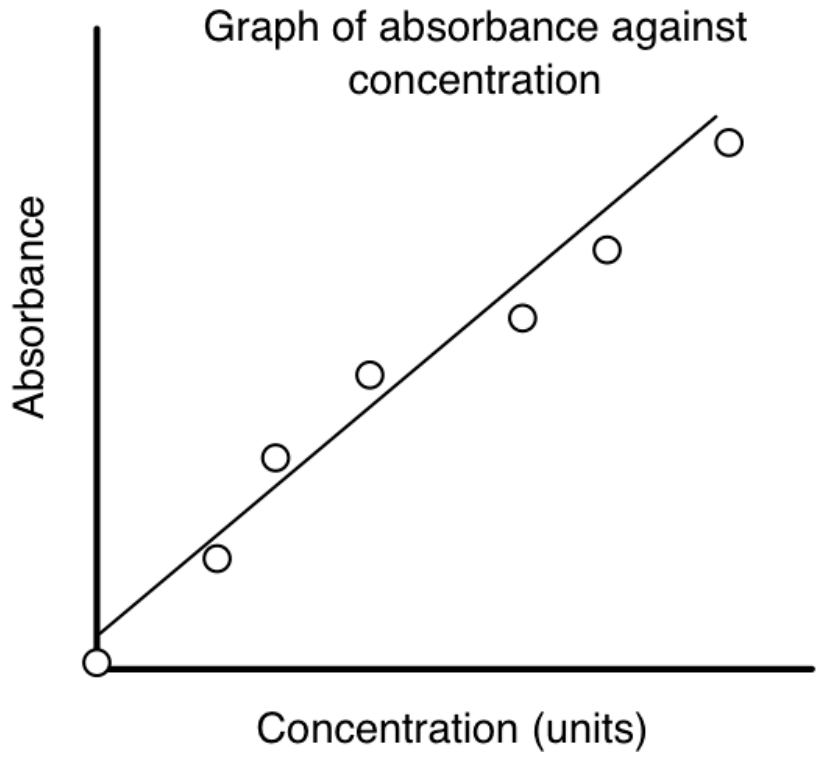
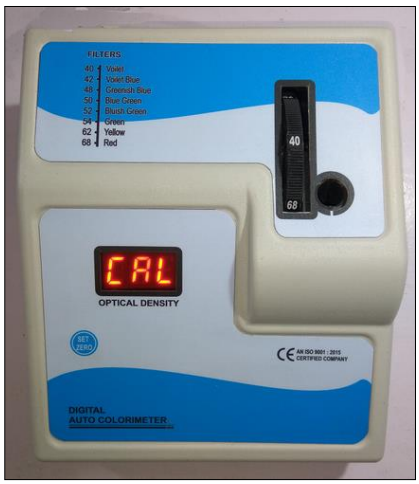


Result A linear curve is obtained between Absorbance v/s concentrations that prove the existence of Lambert-Beer law.

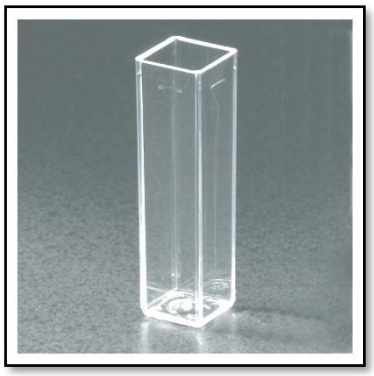
PRECAUTIONS

One should note that the Beer–Lambert law is obeyed by many substances mainly at low to moderate concentrations; therefore, dilute concentrations of the absorbing species should be measured. In practice it is advisable to measure absorbances in the range 0.1< A <1.0. Care must be taken to avoid any kind of chemical associations/dissociations of the absorbing species.

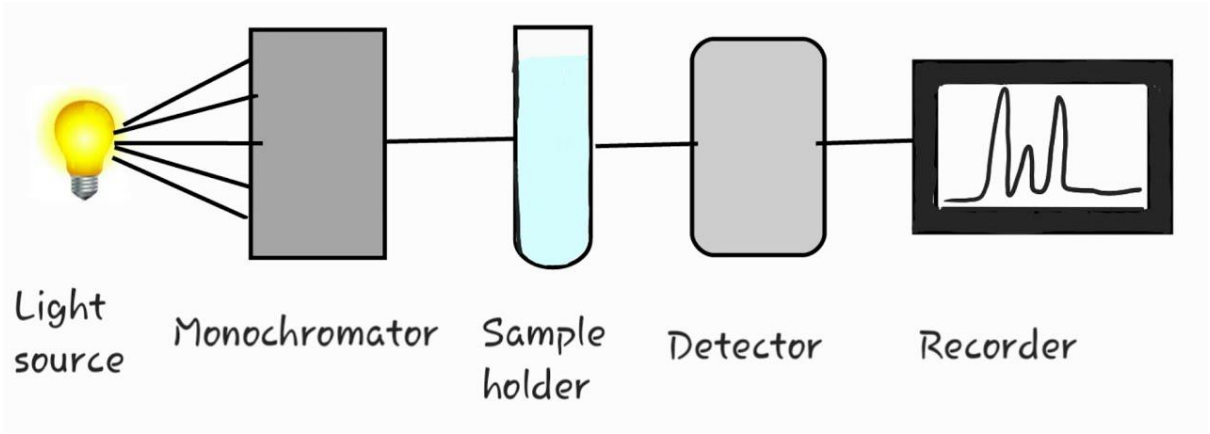


Colorimeter

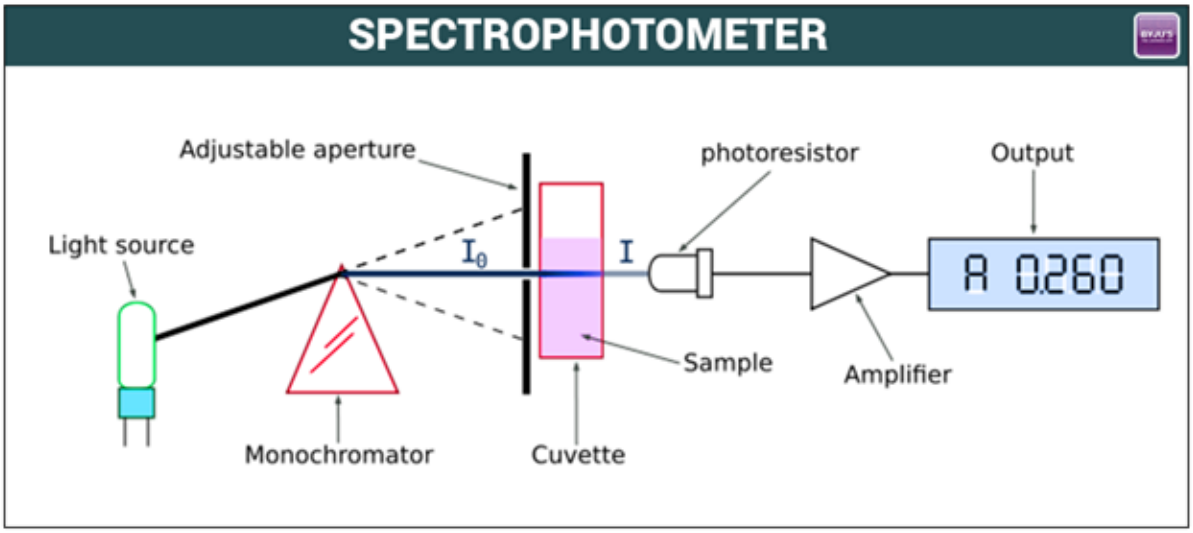
UV-Visible Spectrophotometers



Cuvette



Instrumentation


SPECTROPHOTOMETER
Adjustable aperture
photoresistor
Output
Light source
I_0
I
A 0.260
Sample
Amplifier
Monochromator
Cuvette