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Theme 13. Basic terms and concepts of electromagnetic radiation wave theory. Physics model of optical radiation interaction with matter. Optical methods for solutions substances concentrations measuring. Measurement of the concentration of substances in transparent fluids via optical colorimeter

Problem
Solutions concentrations definitive diagnostic test

Attendance prerequisite
Note! Answer in writing to perform

1. Define or explain physics terms and phenomena: light spectrum; light intensity; substances light absorbance

Information resources

№	Author(s)	Name of the source (textbook, manual, monograph, etc)	City, publishing house
1	R. M. Berne, M. N. Levy	Physiology	St Louis: Mosby Company, 1983
2	Vander, Sherman, Luciano	Human Physiology The Mechanisms of Body Function	New York: McGraw-Hill Book Company, 1980
3	Vander, Sherman, Luciano	Human Physiology The Mechanisms of Body Function	New York: McGraw-Hill Book Company, 1985
4	N. V. Pronina	Biological Physics The Second Module Lectures	Simferopol, 2006
5	Douglas C. Giancoli	Physics Principles with Applications	Pearson Education Limited; 7th Edition, 2016
6	Martin Hollins	Medical Physics	Tomas Nelson & Sons, 1992
7	I. Tarjan	An Introduction to Physics with Medical Orientation	Akademiai Kiado, Budapest, 1987
8	Joseph W. Kane, Morton M. Sternheim	Physics	John Wiley & Sons Third Edition, 1988
9	John Bullock, Joseph Boyle, Michael Wang	Physiology	Williams & Wilkins Third Edition, 1994

Introduction

When light passes through a medium it can be absorbed and scattered by suspended particles and molecules. The intensity of incident light I_0 is shared by absorbed light intensity I_1, scattered light intensity I_2 and passed light intensity I_3 (fig.1).

According to the law of conservation of energy: $I_0 = I_1 + I_2 + I_3$.

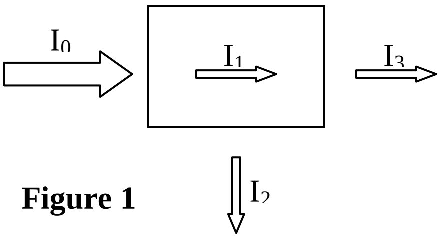


Figure 1

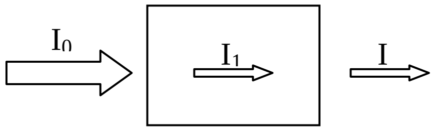


Figure 2

Light absorption. For dilute solutions, collimated incident illumination, negligible scattering the intensity of light passed through a layer of substance of thickness ***l*** being absorbed by the medium is found $\mathbf{I = I_0 \cdot e^{-k \cdot \mathit{l}}}$, where $\mathbf{I_0}$ is the incident light intensity, **k** is the absorption coefficient.

This expression is called the **Bouguer law**. It describes the reduction of light intensity by absorption when passing through a layer of substance of thickness ***l***. The absorption coefficient **k** depends on the wavelength of radiation and the nature of absorbing medium.

It was found by Beer, that the absorption coefficient of solutions is proportional to the concentration **C** of dissolved substance $\mathbf{k = \alpha' \cdot C}$, where α' is molar absorption coefficient of the absorbing species.

Thus, the expression of absorption is rewritten $\mathbf{I = I_0\, e^{-\alpha' \cdot C \cdot \mathit{l}}}$ or$\mathbf{I = I_0\, 10^{-\alpha \cdot C \cdot \mathit{l}}}$, $\mathbf{\alpha = 0.43\, \alpha'}$.

This expression is called the **Bouguer-Lambert-Beer law**.

The molar absorption coefficient varies with the wavelength of radiation.

The ratio between the absorbed light intensity $I_1 = I_0 - I$ and the incident light intensity I_0 is defined as **absorptance** $\mathbf{A = I_1/I_0 = (I_0 - I)/I_0}$.

The ratio between the transmitted intensity I and the incident light intensity I_0 is defined as **transmittance** $\mathbf{T = I/I_0}$**.**

The decimal logarithm of the ratio between the incident light intensity I_0 and the passed light intensity I is called **absorbance**

$$\mathbf{D = \log (I_0/I) = \log (1/T) = \alpha \cdot C \cdot \mathit{l}}$$

Thus, molar extinction coefficient equals $\mathbf{\alpha = D/C \cdot \mathit{l}}$, where **D** is absorbance of the sample.

Colorimetry. Colorimetry involves the measurement of the amount of light absorbed by a colored sample. In the kinetics experiments colorimetry is used to monitor the change in concentration of a colored reactant with time.

For a colored solution, some wavelengths are absorbed more strongly than others. This wavelength dependence of absorbance enters Beer's law through the molar absorption coefficient, which is a function of wavelength. The molar absorption coefficient, **α**, is a proportionality constant between absorbance and concentration, for a given path length. A high **α** value means that a substance absorbs light strongly at the particular wavelength. A plot of **α** versus wavelength **λ** is called the **absorption spectrum** of a substance. This spectrum shows which wavelengths of light are absorbed and which are not.

Beer's law is useful in the quantitative analysis of solutions containing colored compounds. At a particular wavelength, Beer's law can be used to relate absorbance and concentration. A typical colorimetric analysis uses a series of solutions, of known concentrations, to produce a Beer's law calibration plot, whose slope can be used to calculate **α**. Then an unknown sample is analyzed by measuring its absorbance. The **α** (or slope) from the Beer's law plot is then used to relate the absorbance of the unknown to its concentration in solution. Colorimetric analyses are normally carried out at the wavelength of an absorption **maximum** since the molar absorption coefficient is least sensitive to changes in wavelength near a maximum.

Lab test protocol

NOTE! Make Lab test protocol copy and bring

Lab test

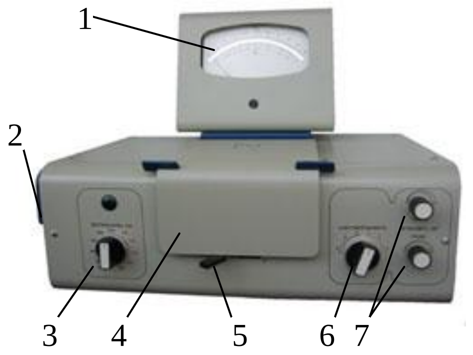
Measurement of coloured solutions concentration colorimeter.

Problem

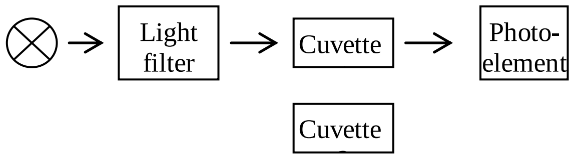
What is the effect of coloured solution concentration on absorbance?

Equipment

Colorimeter КФК-2, a set of cuvettes with distilled water and the solutions of copper sulfate of different concentrations



1 - the colorimeter scale calibrated in transmittance and absorbance units
2 - the colorimeter lamp
3 - the light filters handle
4 - the cuvette camera cover
5 - the lever to shift cuvettes
6 - the sensitivity handle
7 - the scale settings handles



3rocedure

1. Switch on the colorimeter and let it warm up (for 15 min). Set the light filter of 364 nm.

2. Place the cuvette filled with distilled water into the cuvette camera and using handles 6, 7 set the colorimeter absorbance scale on 0. Place the cuvette filled with the copper sulfate solution of concentration C_x and note the recording of absorbance. Repeat the experiment for different wavelengths and write down data in table 1.

3. Chose the wavelength corresponding to the maximum of absorbance: **λ =**

4. Measure the absorbance of copper sulfate solutions of different concentrations at the wavelength corresponding to the maximum of absorbance and write down data in table 2.

5. Plot the graph of absorbance D vs concentration C: D(C).

6. Using the graph and table 2 data find the unknown concentration C_x.

7. Find the absolute error of measurement using the graph. Find the tangent of the slope angle at point (D_x; C_x). The absolute error is $\Delta C_x = 0.001/(tg\alpha_x)$.

8. Give the result according to the expression $C = C_x \pm \Delta C_x$.

Table 1

Wavelength λ, nm	364	400	440	490	540	590	670	750
Absorbance D								

Table 2

Concentration C, %	0	0.2	0.4	0.6	0.8	1.0	C_x
Absorbance D							

Analysis and conclusions

1. Describe a wavelength effect on a color solutions light absorption (absorbance, intensity).

2. Describe a color solution concentration effect on light absorption (absorbance, intensity). Sketch the graph of intensity I of light passed through a color solution versus the solution concentration.

3. Give examples of the application of colorimetry in medicine.