

Recording of absorption spectra with a spectrometer

Aims of the experiment

- To observe and understand light spectra.
- To recognise the connection between absorption, reflection and transmission.
- To appreciate colour as light of characteristic wavelengths.
- To identify the interaction of light with coloured liquids.

Principles

Light is that part of electromagnetic radiation that is visible to the eye. This radiation is found in the wavelength range of about 380 nm to 780 nm and consists of light particles, so-called photons. Violet light with a wavelength of 400 nm is the most energetic radiation in the visible spectrum and red light with a wavelength of 700 nm the least energetic.

If a cuvette filled with a dye solution is placed between a lamp and a compact spectrometer, the light will interact with the absorbing substances in the liquid medium. There are three possibilities for interaction: Reflection, absorption and transmission. In the case of reflection, the light is reflected at the surface of the medium with the angle of incidence being equal to the angle of reflection. In the case of absorption, the light will be partially or completely absorbed through interaction with the absorbing substance and less light will be transmitted. Transmission is the passage of the light through the solution without interaction with the medium. In the spectroscopic measurement of intensity for the determination of interactions, the reflection neglected, as it is accounted for through reference measurements.

The simplest to calculate is the transmission T . For this, the quotient of the reference intensity I_0 and the transmitted radiation I_1 is formed as follows:

$$T = \frac{I_1}{I_0}$$

The extinction E (optical density) is a measure of absorption. This is determined by calculating the negative logarithm to the base 10 of the transmission. If, instead of the transmission T , the quotient of the incident light I_0 and the transmitted light I_1 is used, the following formula results:

$$E_\lambda = -\log T = -\log \frac{I_1}{I_0}$$

Therefore by measuring the light intensities, both the transmission as well as the extinction for a dye solution can be determined. The dye solutions always absorb light at characteristic wavelengths. The wavelengths can also be important for the characterisation of components in solutions.

In this experiment, the transmission and extinction spectra of various solutions will be recorded.

Risk assessment

Some of the chemicals used are hazardous to health and should not be swallowed. The iron(III) chloride hexahydrate is corrosive and should only be handled with suitable protective gloves.



Fig. 1: Experimental apparatus for recording the light spectra of dyes.

Methylene blue	
 Signal word: Caution	Hazard statements H302 Harmful if swallowed.
Iron(III) chloride hexahydrate	
 Signal word: Hazard	Hazard statements H302 Harmful if swallowed. H315 Causes skin irritation. H318 Causes serious eye damage. Precautionary statements P280 Wear protective gloves/eye protection/face protection. P302+P352 IF ON SKIN: Wash with soap and water. P305+P351+P338 IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do. Continue rinsing. P313 Get medical advice/attention.
Universal indicator, contains ethanol	
 Signal word: Hazard	Hazard statements H225 Highly flammable liquid and vapour. Safety statements P210 Keep away from heat/sparks/open flames/hot surfaces. No smoking.

Equipment and chemicals

1 Optical glass cell (cuvette), 10 mm	477 20
1 Prism table on a rod.....	460 25
1 Lens in frame, f = +100 mm	460 03
1 Lamp housing with cable	450 60
1 Bulb 12 V/30 W, E14, set of 2.....	450 521
1 Condenser with diaphragm holder	460 20
1 Transformer 6/12 V, 30 W.....	521 210
1 Compact spectrometer USB, physics	467 251
1 Optical fibre holder.....	460 251
1 Optical bench, S1 Profile, 1 m	460 310
4 Optical carriage with clamp.....	460 311
1 Graduated pipette 10 mL	665 997
1 Pipetting ball (Peleus ball)	666 003
1 Double microspatula, steel, 150 mm.....	604 5672
1 Compact balance 200 g : 0.01 g	667 7977
1 Beaker, DURAN, 250 mL, squat	664 103
6 Beaker, DURAN, 150 mL, squat	602 043
1 Measuring cylinder 100 mL, with plastic base	665 754
1 Dye, red, 10 g	309 42
1 Iron(III) chloride hexahydrate, 50 g	671 8700
1 Methylene blue, 1 g	673 2900
1 Universal indicator, liquid, 50 mL	675 2550
also required:	
1 PC with Windows XP, 7 or 8	

Set-up and preparation of the experiment

Preparing the solutions

General note: The solutions prepared in this way might need to be diluted further, as only a small amount of dye can influence the colour strength of the solution. For the transmission

spectra, the solutions should be diluted so that the extinction is between 1 and 2 within the measurement range of the compact spectrometer.

Preparing a red dye solution: For a red solution, weigh 0.05 g of dye powder (red, non-toxic) into a beaker and add 100 mL of water using a 100 mL measuring cylinder. Stir the solution well until the dye has completely dissolved. Then remove 10 mL of solution and dilute it with 90 mL of water. This produces 100 mL of dye solution.

Preparing a yellow dye solution: For a yellow solution, weigh 0.49 g of iron(III) chloride hexahydrate into a beaker and add 75 mL of water using a 100 mL measuring cylinder. Then stir the solution well until the substance has completely dissolved. This produces 75 mL of dye solution.

Preparing a blue dye solution: For a blue solution, weigh 0.05 g of methylene blue into a beaker and add 50 mL of water using the 100 mL measuring cylinder. Stir the solution well until the dye has completely dissolved. Then remove 1 mL and fill this up to 150 mL using a measuring cylinder (100 mL).

Preparing a violet dye solution: Dilute about 2 mL of universal indicator with 77 mL of water then add about 1 mL of 2 M sodium hydroxide solution. The solution should have a pH value of about 10. This produces 100 mL of dye solution.

Set-up of the apparatus

On the optical bench, set up the apparatus consisting of a transformer, a lamp, a lens, a glass cuvette, a prism table, an optical fibre and a compact spectrometer to record the light spectra of dyes in Spectra Lab (see Fig. 1).

Note: When setting up the apparatus, make sure that the optical fibre is well illuminated by the lamp. If this is not the case, the light beam can be focussed by moving the lens.

Performing the experiment

1. Insert the lamp connection leads into the transformer and switch the transformer on. Then check the alignment of the optical fibre and move the lens to refocus, if required.

2. Start SpectraLab. Adjust the integration time with the minus  and plus  buttons so that the maximum intensity is about 75 %.

Note: In order to ensure comparability of the measurement values, the integration time chosen should not be changed during the experiment.

3. Switch the lamp off to record the background spectrum and open the Offset 10 display screen.

Note: The program automatically stores this spectrum when the Offset 10 display option is opened, it is therefore advisable not to open this again.

4. Change to the Reference I2 display screen, switch the lamp on again and place the cuvette filled with water onto the prism table. Stop recording the spectrum by pressing Pause . This spectrum will now be used as the reference spectrum.

Note: This display should not be opened again during a measurement, otherwise it will be overwritten when the measurement is stopped again.

5. Then place a cuvette containing the dye solution onto the prism table.

6. Change to the display "Intensity I1". Here the spectrum intensity after the light passes through the cuvette can be seen. The water reference spectrum is also displayed in grey.

7. Change to the display "Transmission T". For the display "Transmission T", the ratio of the spectrum of a dye solution to the reference curve is calculated and displayed.

8. For the display "Extinction E", the extinction (optical density) is calculated and displayed.

Note: The solutions should not be too strong, as extinction values over 2 cannot be adequately displayed.

9. Under "Save file", the measurements can be stored for each dye solution separately. It is also possible to save several measurements made with "Record"  in one file. In this way, further spectra for other dye solutions can be measured and compared.

Observation

The reference spectrum of water

Initially a reference spectrum for water is recorded. For this, the intensity I_2 [%] is measured in dependence upon the wavelength λ [nm]. This produces a continuous colour spectrum of a colourless medium (see Fig. 2).

The intensity spectrum of all dye solutions

In the next step, an intensity spectrum of every dye solution was recorded. For this, SpectraLab calculates the intensity I_1 [%] in dependence upon the wavelength λ [nm]. It can be seen from this that for every dye there is a loss of intensity at characteristic wavelengths (see Fig. 3). After recording a transmission and an extinction spectrum for each dye solution, this property will be examined more closely in the further course.

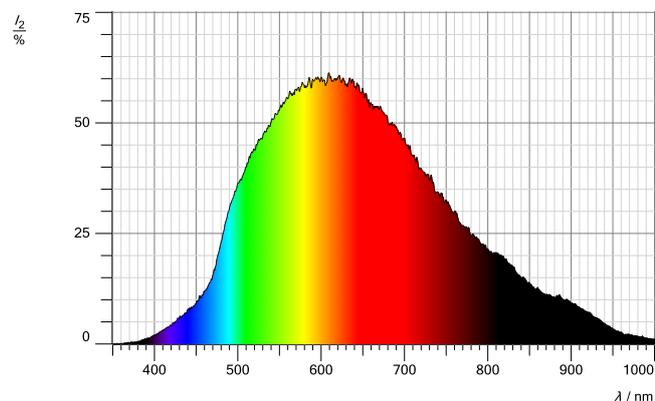


Fig. 2: Reference spectrum for a cuvette filled with water.

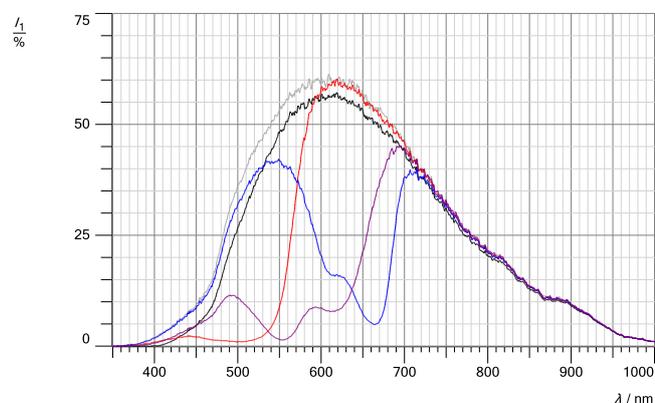


Fig. 3: Intensity spectrum of every dye solution. Here, the intensity of the reference spectrum of water is displayed in grey, of a yellow solution in black, a red solution in red, a blue solution in blue and a violet solution in violet.

The transmission and extinction spectra of dye solutions

For recording transmission spectra in SpectraLab, the transmission T [%] was calculated and shown plotted against the

wavelength λ . For displaying an extinction spectrum, the extinction E was calculated and plotted against the wavelength λ .

If the cuvette is filled with a red solution, all light wavelengths from 400 nm to 600 nm are absorbed. In contrast, the red light in the range 600 nm to 730 nm is transmitted. The perceived colour of the solution is therefore red (see Fig. 4). It can be seen in the extinction spectrum that colours in the range 400 nm to 600 nm are weakened in passing through the solution. The optical density in the colours violet to yellow has therefore increased in comparison to white light (see Fig. 5).

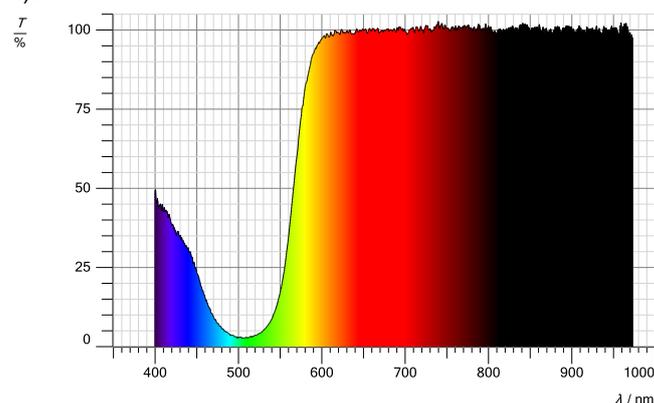


Fig. 4: Transmission spectrum of a red dye solution.

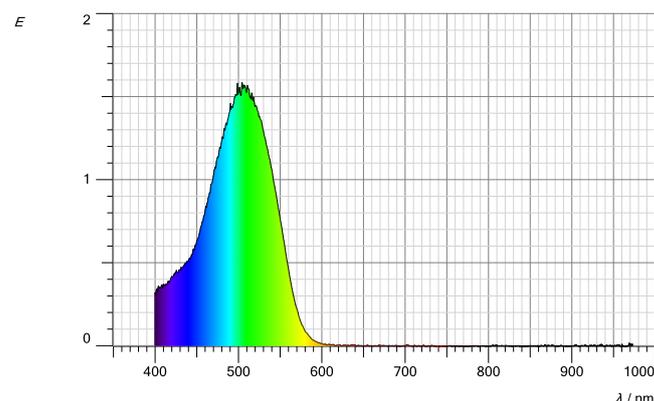


Fig. 5: Extinction spectrum of a red dye solution.

In the case of the yellow dye solution, the violet-blue light at 400 nm – 450 nm is absorbed and all other colours of visible light are transmitted. As violet-blue is the complementary colour of yellow, the solution appears yellow (see Fig. 6). In the case of a yellow dye solution, the optical density at a wavelength of 400 nm - 450 nm is increased. This can be observed in the following extinction spectrum (see Fig. 7).

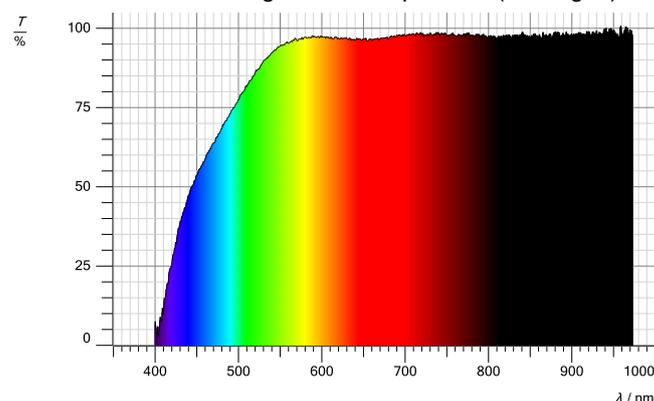


Fig. 6: Transmission spectrum of a yellow dye solution.

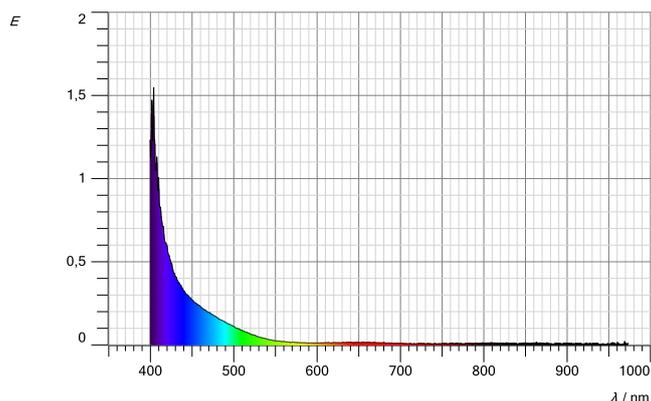


Fig. 7: Extinction spectrum of a yellow dye solution.

In the case of the blue dye solution, only the orange-red light at 590 nm – 700 nm is absorbed and all other colours of visible light are transmitted. The solution appears blue, as the complementary colour red-orange is absorbed (see Fig. 8). If the extinction is now considered, it can be seen above all in the red-orange range of 590 nm - 700 nm that the radiation of white light has been weakened (see Fig. 9).

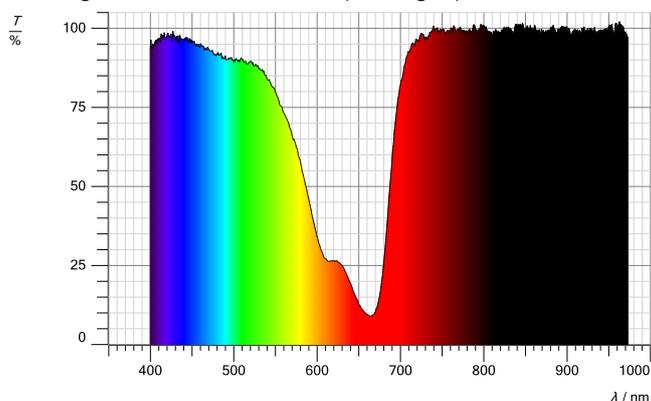


Fig. 8: Transmission spectrum of a blue dye solution.

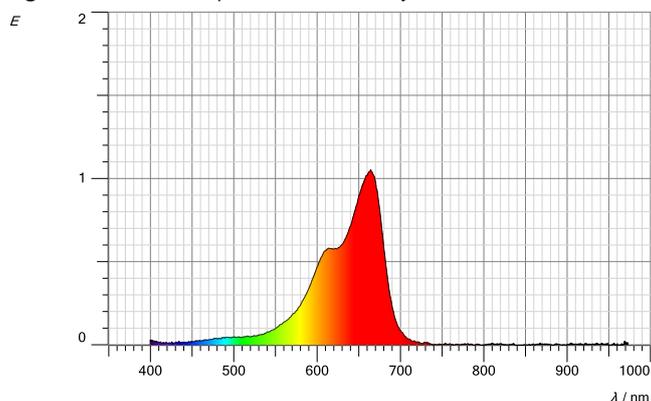


Fig. 9: Extinction spectrum of a blue dye solution.

If a violet solution is placed in the cuvette, then absorption takes place mainly in the wavelength range of green light at 530 nm, of yellow light at 550 nm and of red-orange light at 620 nm. Additionally, the intensity falls in the range of violet-blue light. As the absorption of the yellow-green and the orange-red light is predominant, the solution appears in the complementary colour violet (see Fig. 10).

In the extinction spectrum, the increase in optical density in the range 400 nm - 700 nm is displayed. The extinction maximum is in the yellow-green range at a wavelength of 540 nm (see Fig. 11).

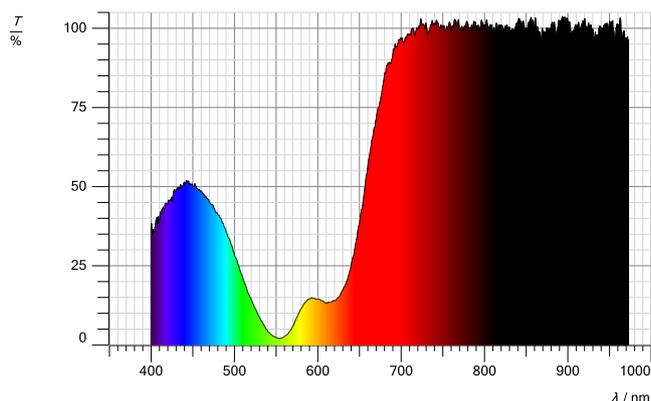


Fig. 10: Transmission spectrum of a violet dye solution.

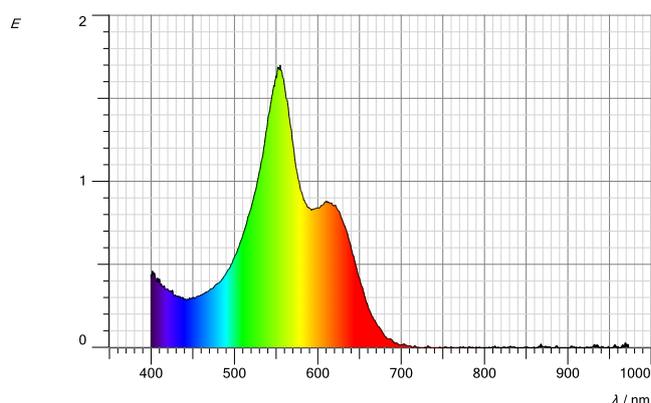


Fig. 11: Transmission spectrum of a violet dye solution.

Evaluation

Calculation of various measurement parameters in SpectraLab

Reference and intensity spectra: Intensity I_1 , which indicates the intensity after passage of the light through the solution in the cuvette, is displayed in the reference spectrum and in the intensity spectrum. For this, the offset intensity I_0 is subtracted from the measured intensity I of the emerging light:

$$I_1 = I - I_0$$

The only difference in the spectra is due to the liquids in the cuvette. For the reference spectrum, the cuvette is filled with water. In the case of the intensity spectrum, the cuvette is filled with various dye solutions.

Transmission spectrum: For displaying the transmission spectrum, the transmission T is calculated from the ratio of the intensity I_1 of the light emerging from a dye solution to the reference intensity I_2 .

$$T = \frac{I_1}{I_2}$$

Extinction spectrum: For the extinction spectrum, the extinction E (optical density) is calculated. This is calculated by taking the negative logarithm to the base 10 of the transmission T , where the transmission is the quotient of the incident light I_1 and the emerging light I_2 .

$$E = -\log T = -\log \frac{I_1}{I_2}$$

Interpretation of the spectra

As shown in the various examples, there are two possibilities for transmission and extinction which determine the perceived colour of a solution. The first case occurs with the red transmission spectrum, as an example. Here, all wavelengths of the visible colours from 400 nm – 600 nm are absorbed and only the wavelength of the red dye solution in the range 600

nm – 730 nm is transmitted. The extinction spectrum also reflects this, as here the optical density in the range 400 nm – 600 nm increases. The solution appears red because only the wavelength of red light is transmitted.

The second case is that only the complementary colour is absorbed and all other colours are transmitted. This is the case with, for example, the blue solution. It can be seen in the transmission spectrum that the wavelength of the complementary colour in the range 590 nm - 700 nm is absorbed and all other colours in the range 400 nm – 590 nm and 700 nm – 750 nm are transmitted. The colour therefore appears blue, the complementary colour of the absorbed colour red-orange.

The assignment of perceived colour to transmitted light through absorption of the complementary colour is shown in Table 1.

Tab. 1: Colours of absorbed and transmitted light.

Absorbed light		Transmitted light
Wavelength	Colour	Observed colour
730 nm	deep red	green
640 nm	red	blue-green
590 nm	orange	blue
550 nm	yellow	blue-violet
530 nm	yellow-green	violet

510 nm	green	deep red
490 nm	blue-green	red
450 nm	blue	orange
425 nm	blue-violet	yellow
400 nm	violet	yellow-green

Results

Through intensity measurements on various liquids, it can be shown that every coloured solution absorbs and transmits characteristic wavelengths. This becomes particularly clear in the images of the intensity spectra of the individual dye solutions (see Fig. 3). The absorbed wavelengths no longer show any intensity in the transmission spectra, and the transmitted wavelengths are displayed. Considering the extinction spectrum, on the other hand, this represents a measure of absorption. The intensity maxima are found at precisely the wavelengths that are not transmitted in the transmission spectrum. These are specific for any one dye.

Cleaning and disposal

The methylene blue solution should be disposed of in the container for organic halogen-free substances, the universal indicator solution and the iron(III) chloride solution in the canister for liquid organic waste with halogens. The red dye (non-toxic) can be poured into the laboratory drain.