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B Universitat de Barcelona

HOW MUCH DYE IS IN DRINK?

Spectroscopic quantitative analysis

Charles and Michael, they often go to restaurant to have a drink. Once, they had a sweet peppermint liqueur, which has a typical green color. Suddenly, Michel, looking at the liqueur, got angry, saying: "that is terrible, everywhere are substitutes. I bet that this liqueur, this is the same case. Look at the color. It must be something poisonous inside. Surely, there is more dye inside than allowed. And, it is killing us slowly. We should take some legal action against manufacturer of this killing drink, should not we? Charles faintly smiled and told: "It seems to be, probably, you are right. Actually, I have a friend who is working in chemistry lab. He can help us to analyze this drink. And, if there is more dye than allowed ... Bad new for the manufacturer. Both of them, they ordered a one more drink and, tomorrow, Charles took a small volume of the drink (they drunk majority of the liqueur with Michael) to his friend (you) for analysis. Can you help him?

Some dyes used for coloring of foods and drinks (and it is not important whether they are synthetic or natural) can cause serious health problems (for example allergic reactions ...) if their concentration in foods and drinks and their consumption is too high. To prevent food and drink producers to add dangerous concentrations of the dyes into their products, the limits for the concentration have been established. Nevertheless, it is necessary to control the concentrations of dyes in foods and drinks to achieve the quality of the products and, to inspect whether the limits are abided by the producers, because the products must not threaten consumers. One possibility of control of dye and color compounds concentration is spectroscopy. Find more information about quantitative spectroscopic analysis to find answer to following question.

How many mg of dye is in 1 liter of sample of peppermint liqueur? Did producer fulfill a rule which says that the concentration of dyes in liqueur must not exceed 100 mg per liter of the drink?

What you should to know:



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The color of dyes is very often caused by synthetic (or natural) dyes added into the drink. In our case, to achieve a typical green color of the peppermint sweet liqueur, two dyes are used – a blue and a yellow – giving green color. The first one is Tartrazin (Fig. 1), yellow dye, which belongs to group of azo dyes and a Brilliant blue (Fig. 2), which is attributed to group of dyes derived from triphenylmethane. Absorption maximum of Tartrazin is at 427 nm wavelength and absorption maximum of Brilliant Blue can be found at 629 nm wavelength.



Fig. 1: Tartrazin dye.

Fig. 2: Brilliant blue dye.

Procedure:

If you carefully studied the basis of quantitative spectroscopic analysis, you can help to Charles and Michael. You can use the following procedure, nevertheless, you should know at any step of the procedure, what you are actually doing.

Equipment and chemicals: sample of liqueur containing two dyes – peppermint sweet liqueur (dyes E-133 – Brilliant Blue and E-102 – Tartrazin), dye standards – food dyes Brilliant blue (E-133) and Tartrazin (E-102), water.

Equipment: balances (at least 0,01 g), small beaker, spatula, 2x 100 mL beakers, glass rod, wash bottle, 4x 100 mL volumetric flask or 100 mL volumetric cylinder, test tube holder, marker, 2 mL and 10 mL pipette, pipette adapter, spectrophotometer, cuvette.

Procedure – how to do it:

1A1) Preparation of 100 mL of the solution of Brilliant blue with concentration ca 50 mg:

At packet (container) of a standard of Brilliant blue dye (E133) read the concentration of the dye in the mixture. Calculate amount (in mg) of mixture necessary for preparation of the solution of given concentration. Weight the calculated amount of the mixture (with dye) and





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add into a beaker. Pour 50 mL of water and let the dye to dissolve. Transfer quantitatively the dye solution into the volumetric flask. Add water to achieve 100 mL mark.

Calculation:

Calculate how many mg of mixture is necessary to weight to get the solution with given concentration (50 mg/100 mL). Which is concentration of your solution?

Concentration of Brilliant blue dye in solution is mg/ mL. (fill in blank fields)

<u>1B1) Dissolution of the dye solution to measurable concentration:</u>

The concentration of the solution just prepared is still too high to make the spectroscopic measurement. To get to measurable concentration, we have to dissolve the solution 100x.

Calculation:

Calculate, how many mL of the prepared solution we need to transfer to 100 mL volumetric flask to get 100 mL of the measurable solution diluted 100 times with respect to original solution.

Concentration of Brilliant blue dye in solution is mg/ mL. (fill in blank fields)

1A2) Preparation of 100 mL of the solution of Tartrazin with concentration ca 45 mg:

At packet (container) of a standard of Tartrazin dye (E102), read the concentration of the dye in the mixture. Calculate amount (in mg) of mixture necessary for preparation of the solution of given concentration. Weight the calculated amount of the mixture (with dye) and add into a beaker. Pour 50 mL of water and let the dye to dissolve. Transfer quantitatively the dye solution into the volumetric flask. Add water to fill the flask up to 100 mL mark.



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Calculation:

Calculate how many mg of mixture is necessary to weight to get the solution with given concentration (45 mg/100 mL). Which is concentration of your solution?

Concentration of Tartrazine dye in solution is mg/ mL. (fill in blank fields)

<u>1B2</u>) Dissolution of the dye solution to measurable concentration:

The concentration of the solution just prepared is still too high to make the spectroscopic measurement. To get to measurable concentration, we have to dissolve the solution 25x.

Calculation:

Calculate, how many mL of the prepared solution we need to transfer to 100 mL volumetric flask to get 100 mL of the measurable solution diluted 25 times with respect to original solution.

Concentration of Tartrazine dye in solution is mg/ mL. (fill in blank fields)

2A) Preparation of calibration solution of the blue dye:

Prepare 5 clean test tubes and insert them into test tube holder. Mark the test tubes by marker 1A up to 5A. Prepare the calibration solutions by dissolution of the solution in 1B1 according to the table below. Do not forget to calculate the missing data – concentrations of calibration solutions.



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Test tube No.	Dye solution (1B1)	water [mL]	Dye concentration
	[mL]		[mg/100 mL]
1A	1	4	
2A	2	3	
3A	3	2	
4A	4	1	
5A	5	0	

2B) Preparation of calibration solutions for yellow dye:

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Prepare 5 clean test tubes and insert them into test tube holder. Mark the test tubes by marker 1B up to 5B. Prepare the calibration solutions by dissolution of the solution in 1B2 according to the table below. Do not forget to calculate the missing data - concentrations of calibration solutions.

Test tube No.	Dye solution (1B2) [mL]	water [mL]	Dye concentration [mg/100 mL]
1B	1	4	
2B	2	3	
3B	3	2	
4B	4	1	
5B	5	0	



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3) Measurement and preparation of calibration:

Measure the absorption spectra of the calibration solutions of the both of the dyes. First, measure the solution with the lowest concentration and continue, step by step, to the most concentrated solution. It is not necessary to rinse the cuvette among the individual calibration standards. Read the value of absorbance at 629 nm for blue dye and 427 nm for yellow dye. Write down the measured values into the table below.

Test tube No.	absorbance (blue dye solution)	absorbance (yellow dye solution)
1		
2		
3		
4		
5		

Prepare the calibration dependences (absorbance vs. concentration) for both the dyes.

4) <u>Sample preparation:</u>

Fill in the cuvette with sample and record its absorption spectrum. Read the value of absorbance at 629 nm (blue dye) and at 427 nm (yellow dye). In the case that absorbance of sample is higher than the absorbance of the most concentrated standard, dilute the sample accordingly.

vzorek	absorbance při 629 nm	absorbance při 427 nm

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5) <u>Calculation – dye concentrations in sample:</u>

Using calibration curves or the equations of these curves, calculate the concentration of blue and yellow dyes in the sample of peppermint liqueur.

Conclusion:

Communicate your results:

Write to Charles and Michael an e-mail about the results of analysis of the sample of peppermint liqueur with all the necessary information and comments.

Questions:

1. What is the name of law which we used for determination of dyes concentrations in the sample?



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2. Why the producer of the peppermint liqueur did not use for coloring of the beverage chlorophyll instead of the two synthetic dyes? Speculate ...





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