

# Task 2 Task sheets

## **Smart cucumber**



#### Introduction to the task:

A circular economy is a model of production and consumption, which involves sharing, leasing, reusing, repairing, refurbishing and recycling existing materials and products as long as possible. Example of a circular economy would be using waste produced by everyday activities as a source of energy.

In Latvia one of the waste management stations uses biological waste to produce gas that is used for electricity and heat production. This heat is used to heat greenhouses allowing to grow vegetables all year long, even in coldest winters. Plants are grown in a hydroponic system allowing better control on growing conditions. This uses a greenhouse that is a smart greenhouse where environmental conditions are monitored, i.e. temperature, light, humidity and the amount of CO2 in the air are measured and these conditions are adjusted if necessary.

In this task you will explore principles of growing plants in a hydroponic system in a smart greenhouse. Let's imagine that your team is asked by a local waste management plant to help to design a smart greenhouse. Together you will have to build a hydroponics system for growing cucumbers in the most efficient manner.

Here are approximate times you will need to spend on each task. Some tasks do not require subject specific knowledge at the beginning so decide how to distribute the tasks among yourselves

Problem 1 - Water hardness - 2 hours

Problem 2 - Preparation of hydroponics feeding solution - 2 hours

Problem 3 - Water transport in plants - 2 hours

Problem 4 - Water demand by plants - 3 hours

Problem 5 - Exploring hydroponics - 2 hours

Problem 6 - Designing greenhouse - 30 min

## Problem 1 - Water hardness (31 p)

#### Materials and equipment

- 25 mL burette in a stand
- a small funnel
- Beaker for waste
- Two 20.00 mL volumetric pipettes
- Three 100 mL conical (Erlenmeyer) flasks
- 5 mL measuring pipette
- 10 mL measuring pipette
- One micro spatula (one end of spatula for each indicator)
- Bottle with water sample marked with blue tape
- Bottle with EDTA solution marked with green tape
- Bottle with MgSO<sub>4</sub> solution ( $c \approx 0.005$  mol/L, accurate concentration on the bottle), marked with red tape
- Bottle with ammonia buffer pH10 NH<sub>3</sub>/NH<sub>4</sub>Cl (marked with black tape)
- Sodium hydroxide solution (1 mol/L, marked with yellow tape)
- Plastic tube with *eriochrome black T* indicator mixture (1:100 mixture of eriochrome black T and NaCl), marked as ET and blue dot
- Plastic tube with murexide indicator mixture (1:100 mixture of murexide and NaCl), marked as MRX and red dot
- Wash bottle with deionised water (to clean the conical flasks),
- Pipette filler

Warning! Be aware that you are working with dangerous substances. Wear eye and hand protection. In the case of spilling them on your body (hands, face, eyes) wash under cold water and inform the lab assistant.

#### Introduction

For growth and development plants require light, water and some minerals. In our greenhouse heat and light can be obtained from gas, but for watering it is most convenient to use groundwater. Due to bedrock composition of Latvian soils frequently the water that can be obtained will be described as "hard". Using hard water in hydroponic systems is inconvenient as it will cause limestone buildup. Thus the first task is to assess the hardness of available water.

Hardness can be quantified by instrumental analysis. The total water hardness is the sum of the molar concentrations of Ca<sup>2+</sup> and Mg<sup>2+</sup>, in mol/L or mmol/L units, that is usually expressed in CaCO<sub>3</sub> equivalents, see the table below.

Water hardness	mmol/L Ca <sup>2+</sup> and Mg <sup>2+</sup> ions	mg/L CaCO₃
Soft	0 - 0.6	0 - 60
Moderately hard	0.6 - 1.2	60 - 120
Hard	1.2 - 1.8	120 - 180
Very hard	> 1.8	>180

Concentration of Ca<sup>2+</sup> and Mg<sup>2+</sup> are usually determined by complexometric titration. The titration reaction is the complexation reaction of these ions with the compound complexone III (EDTA sodium salt). The water hardness can be determined by determining the total content of calcium and magnesium ions.

The titration is carried out in the presence of an ammonium buffer solution which ensures that the pH of the solution is around 10. Under these conditions, both calcium and magnesium ions react with EDTA. To determine the stoichiometric point, an indicator eriochrome black T (azo dye) is used, which in a free form is blue, but forms red or violet coordination compounds with metal ions. Before the titration, eriochrome black T (HEr²-) forms a red complex with some of the magnesium ions, which are in excess w.r.t. the indicator:

$$Mg^{2\scriptscriptstyle +} + HEr^{2\scriptscriptstyle -} + NH_3 \rightarrow MgEr^- + NH_4{}^{\scriptscriptstyle +}$$

During the titration, EDTA (present in the form of  $H_2Y^{2-}$ ) first reacts with the remaining free magnesium and calcium ions in the solution, forming complexonates  $MgY^{2-}$  and  $CaY^{2-}$ :

$$Mg^{2+} + H_2Y^{2-} + 2 NH_3 \rightarrow MgY^{2-} + 2 NH_4^+$$

$$Ca^{2+} + H_2Y^{2-} + 2 NH_3 \rightarrow CaY^{2-} + 2 NH_4^+$$

At the end of the titration, the reaction between the complex formed by magnesium and the indicator and EDTA takes place, since the complex between EDTA and magnesium ions is much more stable:

$$MgEr^- + H_2Y^{2-} + NH_3 \rightarrow MgY^{2-} + HEr^{2-} + NH_4^+$$

Therefore, at the endpoint of the titration, the colour of the solution is determined by the free indicator which is observed as the colour change of the solution from red or violet to blue. Consider that near the stoichiometric point the reactions are not very fast and some time is required for transformations between the complexes.

For separate determination of concentration of calcium and magnesium ions in the solution, conditions in which only calcium ions react with EDTA are chosen. In strongly basic solution (pH 12-13) calcium ions react with EDTA, forming a stable complexonate. The end point of the titration under these conditions is determined using another indicator (murexide, MX<sup>-</sup>). This indicator acts identically to eriochrome black T, by forming a complex with calcium ions instead of magnesium, which then reacts with EDTA and releases free murexide at the titration endpoint changing the colour from pink to violet. In such a basic environment magnesium ions are precipitated in the form of hydroxide and do not interfere with the determination of calcium ions.

#### **Description of experimental procedure:**

#### 1.1. Standardisation of EDTA solution

- 1. Fill in the burette with the solution of EDTA
- 2. Use a volumetric pipette and transfer 20,0 mL of magnesium sulphate MgSO<sub>4</sub> standard solution (c  $\approx$  0.005 mol/L, exact concentration given on the label) into a 100 mL conical flask
- 3. Using pipette add 5 mL of ammonium buffer solution (pH=10),
- 4. Use a micro spatula spoon side and add 1 pinch around 2-3 mm wide of *eriochrome black T* indicator and lightly mix the solution.
- 5. Titrate with ~xx mol/L EDTA solution until the pink colour changes to blue.
- 6. Repeat the standardisation as many times as you think is necessary to obtain three EDTA volumes required for accurate calculation of the concentration of EDTA solution.
- 7. Use the volume of EDTA to calculate the exact concentration of EDTA solution.
- **1.1.1** Write the chemical reactions occurring during Step 5 of the standardisation of EDTA solution in the answer sheet. (2 p)
- **1.1.2.** Write the volume of EDTA solution you used in the answer sheet. Circle the ones you will use for calculation (4 p)
- **1.1.3.** Calculate exact concentration of your EDTA solution. Show your calculation. (3 p)

#### 1.2. Determination of water hardness

- 1. Use a volumetric pipette and transfer 20 mL of the water sample into a 100 mL conical flask.
- 2. Add 5 mL of ammonium buffer solution (pH=10),
- 3. Use micro spatula spoon side and add 1 portion of eriochrome black T indicator,
- 4. Lightly mix the solution and titrate with EDTA solution until the pink colour changes to blue.

Repeat the titration as many times as you think is necessary to determine volume of the EDTA solution accurately. Use the titration results and calculate the water hardness (in mmol/L) of the analysed water sample.

- **1.2.1** Write the chemical reactions occurring during the Step 4 of the determination of water hardness in the answer sheet. (2 p)
- **1.2.2.** Write the volumes of EDTA solution you used in the answer sheet. Circle the ones you will use for calculation (4 p)
- 1.2.3. Calculate exact water hardness. Show your calculation in the answer sheet. (3 p)
- **1.2.4.** Classify your water sample according to the water hardness, mark your answer in the answer sheet. (1 p)

#### 1.3. Determination of calcium ion concentration

- 1. Use a volumetric pipette and transfer 20 mL of the water sample into a 100 mL conical flask.
- 2. Add 5 mL of 1 mol/L sodium hydroxide solution
- 3. Use a micro spatula <u>flat side</u> and add 1 portion of *murexide* and lightly mix the solution.
- 4. Titrate with EDTA solution until the pink colour changes to violet.

Repeat the titration as many times as you think is necessary to determine volume of the EDTA solution accurately. Use the titration results and calculate the calcium ion concentration (in mmol/L) of the analysed water sample.

- **1.3.1** Write the chemical reactions occurring during steps 2 and 4 of the determination of calcium ion concentration in the answer sheet. (3 p)
- **1.3.2.** Write the volume of EDTA solution you used in the answer sheet. Circle the ones you will use for calculation. (4 p)
- **1.3.3.** Calculate exact calcium ion concentration in your water sample. Show your calculation in the answer sheets. (3 p)

#### 1.4. Calculation of magnesium ion concentration

Use the determined water hardness and concentration of calcium ions in the analysed water sample and calculate the concentration of magnesium ions (in mmol/L) in the water

**1.4.1.** Show your calculation and the final result in the answer sheet (2 p)

## Problem 2 Preparation of hydroponics feeding solution (42 p)

#### Materials and equipment

- Laboratory scales (d=0.01 g)
- Spatula
- Weighing dish
- 100.00 mL volumetric flask
- 15 mL tube for salt mix
- Mortar
- Various salts for feeding solution preparation, listed in text
- Wash bottle with water for rinsing tools
- Paper towels
- Deionised water for preparing solution

MAKE SURE TO WASH SPATULA AND WEIGHING DISH BEFORE USING IT FOR NEXT SALT

#### Introduction

When plants are grown hydroponically, it is particularly important to provide all the microelements, and the feeding solution is the only way these elements can be provided. In this experiment you will prepare the feeding solution for cucumbers from inorganic salts. For calculation of the amount of each salt to be used you will use the information of the plant nutrient elements in a commercial cucumber fertiliser and instructions for its use.

The three essential mineral nutrients needed for cucumbers are potassium, nitrogen, and phosphorus, but also micronutrients such as magnesium are important. Traditionally the nitrogen content in the fertiliser is expressed in the form of nitrogen mass fraction (in %), whereas the content of phosphorus, potassium, and magnesium is expressed in the form of the mass fraction (in % of the total solid mass) of their oxides representing the content of these elements regardless of the chemical form they are present in the actual solution.

The composition of a commercially available cucumber fertiliser salt mix is given as: total nitrogen (N) – 15% (including 10% in form of nitrates  $NO_3^-$  and 5% in form of ammonium  $NH_4^+$ ),

soluble phosphorus pentoxide  $(P_2O_5)$  – 8%, soluble potassium oxide  $(K_2O)$  – 24%, and soluble magnesium oxide (MgO) – 3.0%.

To prepare the feeding solution, 2.0 g of fertiliser salt mix should be dissolved in 1.0 L of water.

Your task is to use the information of the fertiliser composition and create a feeding solution for cucumbers. To do that you will have to calculate what salts and in what amount are present in a fertiliser salt mix with the given nutrient content ensuring that all of the elements are in the solution when dissolved in the water.

Salts available to you are: KCl, KNO<sub>3</sub>,  $K_2CO_3$ ,  $Na_2CO_3 \cdot 10H_2O$ ,  $CaCl_2 \cdot 2H_2O$ ,  $Mg(NO_3)_2 \cdot 6H_2O$ ,  $MgCl_2 \cdot 6H_2O$ ,  $K_3PO_4$ ,  $K_2HPO_4 \cdot 3H_2O$ ,  $Na_3PO_4 \cdot 12H_2O$ ,  $Na_2HPO_4 \cdot 6H_2O$ ,  $NH_4NO_3$ ,  $NH_4CI$ .

You also have been given a salt solubility table and Periodic table at the end of the task sheets.

Molar masses of ions, ionic compounds and anhydrous salts (in g/mol) and their approximate solubilities in g / 100 g (indicated by shading)

E°	°, V	-2.93	-2.91	-2.87	-2.71	-2.37	-1.66	-1.19	-0.76	-0.74	-0.45	-0.26	-0.14	-0.13	-0.04	0.00	+0.34	+0.80	+0.85	
Catio	ONS molar mass	K <sup>+</sup>	Ba <sup>2+</sup>	Ca <sup>2+</sup>	Na <sup>⁺</sup>	Mg <sup>2+</sup>	Al <sup>3+</sup>	Mn <sup>2+</sup>	Zn <sup>2+</sup>	Сг <sup>3+</sup>	Fe <sup>2+</sup>	Ni <sup>2+</sup>	Sn <sup>2+</sup>	Pb <sup>2+</sup>	Fe <sup>3+</sup>	H <sup>⁺</sup>	Cu <sup>2+</sup>	Ag <sup>⁺</sup>	Hg <sup>2+</sup>	NH <sub>4</sub>
Anic and thier	ONS molar mass	39.098		40.078		24.305	26.982	54.938	l			58.693	118.710	207.2	55.845	1.008	1		200.59	18.038
SO <sub>4</sub> <sup>2-</sup>	96.063	174.26	233.39	136.14	142.04	120.37	342.15	151.00	161.44	392.18	151.91	154.76	214.77	303.26	399.88	98.08	159.61	311.80	296.65	132.14
NO <sub>3</sub>	62.005	101.10	261.34	164.09	84.50	148.32	213.00	178.95	189.39	238.01	179.85	182.70	242.72	331.21	241.86	63.01	187.56	169.87	324.60	80.04
SO <sub>3</sub> <sup>2-</sup>	80.063	158.26	217.39	120.14	126.04	104.37	$\times$	135.00	145.44	$\times$	135.91	138.76	X	287.26	$\times$	82.08	$\times$	295.80	$\times$	116.14
PO <sub>4</sub> <sup>3-</sup>	94.971	212.27	601.92	310.18	163.94	262.86	121.95	354.76	386.08	146.97	357.48	366.02	546.07	811.54	150.82	98.00	380.58	418.58	791.71	149.09
НРО	2- 95.979	174.18	233.31	136.06	141.96	120.28	317.94	150.92	161.36	391.93	151.82	154.67	214.69	303.18	399.63	98.00	159.53	311.72	296.57	132.06
CO <sub>3</sub> -	60.009	138.21	197.34	100.09	105.99	84.31	$\times$	114.95	125.39	$\times$	115.85	118.70	X	267.21	$\times$	62.03	$\times$	275.75	$\times$	96.09
SiO <sub>3</sub> <sup>2</sup>	76.084	154.28	$\times$	116.16	122.06	100.39	282.21	131.02	141.46	332.24	131.93	134.78	194.79	283.28	339.94	78.10	139.63	$\times$	$\times$	$\times$
AcO <sup>-</sup>	59.052	98.15	256.43	159.18	82.04	143.41	206.14	174.04	184.48	231.15	174.95	171.80	$\times$	326.30	235.00	60.06	182.65	166.92	319.69	77.09
O <sup>2-</sup>	15.999	94.20	153.33	56.08	61.98	40.30	101.96	70.94	81.38	151.99	71.84	74.69	134.71	223.20	159.69	18.02	79.55	231.74	216.59	$\times$
OH	17.007	56.11	171.34	74.09	40.00	58.32	78.00	88.95	99.40	103.02	89.86	92.71	152.73	241.22	106.87	18.02	97.56	$\times$	$\times$	$\times$
Cl	35.453	74.55	208.23	110.98	58.44	95.21	133.34	125.84	136.29	158.36	126.75	129.60	189.62	278.11	162.20	36.46	134.45	143.32	271.50	53.49
Br	79.904	119.00	297.14	199.89	102.89	184.11	266.69	214.75	225.19	291.71	215.96	218.50	278.52	367.01	295.56	80.91	223.35	187.77	360.40	97.94
1	126.905	166.00	391.14	293.88	149.89	278.11	407.70	308.75	319.19	432.71	309.65	312.50	372.52	461.01	$\times$	127.91	$\boxtimes$	234.77	454.40	144.94
S <sup>2-</sup>	32.065	110.26	169.39	72.14	78.05	56.37	150.16	87.00	97.45	200.19	87.91	90.76	150.78	239.27	$\times$	34.08	95.61	247.80	232.66	68.14
For all	compoui	nds mo	lar mass	ses are o	niven wi	th 2 de	rimal ol	aces	اطبياه ه	a (aalubilib	i a wataa .	10/1000)		Пь	drolysis					

For all compounds, molar masses are given with 2 decimal places.

| soluble (solubility in water > 1g/100g) | hydrolysis |
| slightly soluble (solubility in water 0.1- 1g/100g) | oxide reacts with water |
| insoluble (solubility in water < 0,1g/100g) |

#### 2.1. Determination of ions present in the fertiliser

**2.1.1.** Use the composition information of the fertiliser and write down what ions will be present in the fertiliser solution, if it consists only of part of the given salts and can be fully dissolved in the water! Fill out the table in the answer sheet(4 p).

When you feel confident about your answer, call the lab assistant, they will take the answer sheet page containing your answer and give you a table containing the correct answer.

We suggest proceeding by calculating the amount of each ion in the fertiliser, but you are free to use any other approach leading to the final result. If the final mass of all salts will be correct, you will gain full points for this part of the task. If you are using your own methods to approach the result, proceed to 2.2.1. Submit your calculations on a separate sheet, **label the sheet as P2.1.2** 

**2.1.2.** Calculate the mass fraction of each ion and use the charge balance to determine what other ion has to be present (7 p)

#### 2.2. Calculation of the composition of the fertiliser

Determine what salts the fertiliser is made of! Calculate the mass fraction of each salt in the fertiliser! Use the composition information of the fertiliser. You should use as small a number of salts as possible and as low a number of chemical elements not mentioned among the contents as possible.

- **2.2.1.** In the answer sheet indicate which salts you will use by circling the correct ones. (5 p) You may need to select additional salts after performing calculations
- **2.2.2.** In the answer sheet indicate the mass of each salt you need to create 100g of fertiliser salt mix (10 p).

#### 2.3. Preparation of the feeding solution

At this point you should exchange the answer sheets to questions 2.1. and 2.2. to table with the correct salt composition.

Use the mass fractions of salts and prepare **1.00** g of fertiliser salt mix. Feel free to use the mortar to make the mixture homogeneous.

2.3.1. In the answer sheet indicate the mass of each salt that you have weighed (1p)

Use the prepared fertiliser salt mix and pure deionized water and prepare 100.00 mL of the hydroponics feeding solution. Use a volumetric flask and label it with YOURCOUNTRY\_A/B. Pour the rest of the salt mix in a plastic tube and label it YOURCOUNTRY\_A/B.

**2.3.2.** Label the flask and tube. After completion of work the lab assistant will measure conductivity of the solution to assess the quality of your work. They will also inspect your prepared salt mix. (6 p)

#### 2.4. Impact of water hardness

If we would like to use groundwater for a feeding solution, we have to take into account that it is not deionised. In this part let's consider a groundwater sample with  $Ca^{2+}$  concentration of 1.2 mmol/L and  $Mg^{2+}$  concentration of 0.3 mmol/L

Calculate the concentration of magnesium ions in the hydroponics feeding solution prepared according to the instructions for the use of the fertiliser using pure deionized water.

**2.4.1.** Enter your calculated concentrations of Mg<sup>2+</sup> in answer sheet (1 p)

The magnesium ion concentration will be significantly affected by using such ground water. Therefore, the chemical composition of the salt mix required to prepare the feeding solution should be adjusted.

- **2.4.2.** In the answer sheet complete the table by writing the adjusted magnesium ion amounts in the feeding solution (1 p)
- **2.4.3.** Mark the salts (with Y/N in the table in the answer sheet) the mass of which have to be changed to prepare a salt mix for feeding solution made in groundwater. (5 p)

In fact, such groundwater can not be used for preparing the feeding solution.

**2.4.4.** Write down the chemical formula of the compound the formation of which does not allow to use the groundwater for preparation of the feeding solution. (2 p)

## Problem 3 Water transport in plants

#### Materials and equipment

- Pre-cut cucumber stem samples in a Petri dish filled with water
- Light microscope
- Glass slides
- Cover slips
- Forceps
- Dropper
- Stain containing Astra blue and Safranin
- Water
- Filter paper
- Scalpel and needle for manipulating cross sections
- Cucumber leaf shared with problem 4
- Other materials (sharpeners, erasers, ruler, calculator)

After water together with minerals is taken up by roots, water will move through the whole plant.

**3.1.** Fill in the blanks with the given terms. (8 p)

Not all terms will be used and some may be used several times. We suggest filling out the terms here, in the task sheet to ease filling out table in the answer sheet later.

#### Terms:

a)	Active transport	f)	Negative pressure	k)	Ring
b)	Breathing	g)	organic	l)	Stomata
c)	Dicot		substances	m)	Transpiration
	(eudicotyledons)	h)	Phloem	n)	Vascular bundles
d)	Evaporation	i)	Pith	o)	Water
e)	Monocot	j)	Positive pressure	p)	Xylem

Vascular t	issues in plar	nts are compo	sed of two	types of tissue	s, 1.[	and	], which are
responsib	le for the tran	sport of 2. [	and	] throughou	the plar	it. In non-	-woody
organs the	ese tissues ai	e organized ir	nto structur	es called vasc	ular bund	dle which	are found in
various pa	arts of the pla	nt such as roo	its, stems, a	and leaves. 3.	[ ] t	issue trar	nsports water
and miner	als from the r	oots to the res	st of the pla	nt, while 4. [	] tis	sue trans	sports
organic co	mpounds su	ch as sugars a	and amino a	acids to where	they are	needed.	ı
The struct	ure of vascul	ar tissues can	vary deper	nding on the ty	pe of pla	ant and th	ie
environme	ental condition	ns. In 5.[	] plants,	vascular bund	lles are t	ypically a	rranged in a
6.[	] in the ste	m or root, whi	le in 7. [	] plants	vascular	bundles	are scattered
throughou	t the stem. W	ithin most cor	nmonly fou	nd vascular bi	undles, th	1е 8. [	]
tissue is ty	pically locate	ed towards the	centre of t	he bundle, wh	ile the 9.	[	] tissue is
located to	wards the pe	riphery.					
The move	ment of wate	r and minerals	s through th	ne xylem tissue	is drive	n by a pro	ocess called
10. [	], which in	volves the 11.	.[	] of water from	the leav	es through	gh
	12.[	1. This crea	ates a 13. [	1.	that pulls	water ar	nd minerals

up from the roots and thro	ugh the 14. [	] tissue. The flow of	organic compounds
through the 15.[	] tissue, on the other	r hand, is driven by a	process called pressure
flow, which involves the 10	3.[ ] of suç	gars and other compo	unds from sources in the
plants to the places where	they are needed in the	e plants (areas of activ	e growth (for example
apical and lateral merister	ns and fruits)) or are	as of sugar storage (t	ubers, and bulbs).

#### 3.2. Cross section of cucumber stem

You have been provided with already cut cross-sections of the cucumber stem, which are stored in 70% alcohol solution. Carefully follow the instructions below for proper microscopy preparation and staining.

#### Objective:

To observe, measure, and draw a vascular bundle in a pre-cut cucumber stem using microscopy and staining techniques.

#### Procedure:

- Obtain a pre-cut cucumber stem sample.
   Choose a good quality, thin, uniform-thickness sample.
- 2) Place the sample onto a clean glass slide.
- **3.2.1.** Call the lab assistant to evaluate your selection. They will mark your answer sheet (1 p)
  - 3) Add a drop of stain onto the sample.
  - 4) Let the sample sit in the stain for one minute.
  - 5) Rinse the sample with water.
  - 6) Blot the excess water from the sample with a filter paper. Repeat rinsing till the water is clear
  - 7) Place a cover slip over the dyed sample.
  - 8) View the sample under the microscope.
  - 9) Adjust the focus and choose the appropriate magnification to view the vascular bundle.
  - 10) Locate a clear vascular bundle with all parts visible.
- **3.2.2.** Choose a good quality vascular bundle and make a scientific drawing of the vascular bundle with 2 to 3 layers of surrounding cells in answer sheet (8 p)(see the criteria provided).

A biological drawing shows the ability to make observations. It is an accurate representation of the object to be studied in a drawing, respecting the proportions of all structures. Only what is observed is drawn, and the amount of detail is appropriate to it. When drawing the cells visible in a microscope, only a few cells are drawn to represent the overall picture.

Evaluation criteria for biological drawing.

• The image is drawn with a pencil with clear lines.

- The drawing shows the shape of the cells, the shape and size of the components as seen under the microscope.
- The image has labels to indicate what is being drawn. The labels should be written horizontally, outside the drawing. The lines of the markings should be drawn straight and not intersect.
- The scale or magnification used for the observation is indicated.

You do not need to add a title for this task/drawing. Use the letters from problem 3.1 to identify the drawing elements.

Observe the whole stem cross section. Answer questions concerning cross section. Mark the letter of the correct answer in the answer sheet

- **3.2.3.** How are vascular bundles in cucumber positioned? (1 p)
  - a) In one ring
  - b) Scattered throughout whole stem
  - c) In cross shape
  - d) In two concentric rings
- **3.2.4.** Safranin binds to lignified tissues such as xylem. Where else in the cucumber stem you would find lignin? (1 p)
  - a) Center of the stem
  - b) A layer of cells below epidermis
  - c) In parenchyma
  - d) In cambium
- **3.2.5.** What is the purpose of lignin in cell walls? (1 p)
  - a) To add mechanical strength
  - b) To transport water
  - c) To perform photosynthesis
  - d) To ensure storage of sugars
- **3.2.6.** Astra blue stain will bind to (1 p)
  - a) Cell walls
  - b) Vacuole
  - c) Cytoplasm
  - d) Nucleus

#### Measuring cells

When looking at objects in a microscope, we can also assess their size using stage micrometre and ocular scales. In this task you will not have to measure the exact size of a bundle cell, but to assess it in the order of magnitude.

Choose a magnification in which bundle cells are nicely discernable and assess the approximate size of the largest cell - what proportion of your field of vision they take. Repeat that with two other vascular bundles. Then compare them to pictures that are obtained by photographing stage micrometre under the same microscope (Figure 3.2).

Assessing measurements of bundle cells



Figure 3.2. View of stage micrometre in 40x, 100x and 400x magnification. The whole scale is 1 mm long.

**3.2.7.** Pick a correct approximate dimension of a vascular cell diameter (2 p)

- a) 50-500 mm
- b) 5-50 mm
- c) 500-5000 µm
- d) 50-500 µm
- e) 5-50 µm
- f) 50-500 nm
- g) 5-50 nm

When you are finished observing the cross section, switch off the light and clean the microscope after your work.

**3.2.8.** Read following statements and mark which are applicable to the phloem (P), which to xylem (X) which to both (PX) and which to none (0). Write your answers in the answer sheet. (5 p)

Move substances from roots to leaves.

Are made from elongated, dead cells with thick cell walls that provide support and durability.

Can contract to facilitate movement of liquid.

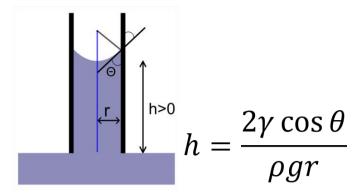
Will transport molecules against concentration gradient.

Will be tapped by aphids to obtain nourishment.

#### 3.3. Importance of transpiration.

Some of the water transport in xylem can be explained by capillary action. Capillary action is the process of a liquid flowing in a narrow space without the assistance of, or even in opposition to, any external forces like gravity. The effect can be seen in the drawing up of liquids between the hairs of a paint-brush or in a thin tube. It occurs because of intermolecular forces between the liquid and surrounding solid surfaces. If the diameter of the tube is sufficiently small, then the combination of surface tension (which is caused by cohesion within the liquid) and adhesive forces between the liquid and container wall act to propel the liquid.

The height h of a liquid column in capillary is given by Jurin's law



#### where

 $\gamma$  is the liquid-air surface tension (force/unit length),  $\theta$  is the contact angle,  $\rho$  is the density of liquid (mass/volume), g is the local acceleration due to gravity (length/square of time), and r is the radius of the tube.

Lets evaluate if capillary action is sufficient to ensure water transport in plants. Cucumber plants can be grown up to 15 metres long, but tallest redwoods may reach the heights of 100 metres.

**3.3.1.** Answer the question concerning capillary action. Mark the letter of the correct answer in the answer sheet (1 p)

To have the liquid reaching higher, one should

- a) Increase the density of the liquid
- b) Decrease the radius of the capillary
- c) Increase the radius of the capillary
- d) Decrease surface tension of the liquid

Imagine that a student decided to model redwoods xylems vascular action in the lab with water and glass capillaries they should consider that

For a water-filled glass tube in air at standard laboratory conditions,  $\gamma = 0.0728$  N/m at 20 °C,  $\rho = 1000$  kg/m³, and g = 9.81 m/s². Because water spreads on clean glass, assume for the following the contact angle between water and glass is 0°

**3.3.2.** Write an equation to calculate the height of the water column in a glass capillary under the given conditions. Write your equation in the answer sheet. (1 p)

You may use the symbol of the unknown physical quantities

**3.3.3.** Diameter of the redwoods xylem cell is 30 micrometres. Calculate the height of the water column if a student performs simulation in the lab with water and glass capillary with the same diameter. Give your result expressed in cm and round it as integer. (1 p)

The process of transpiration is identified as a crucial mechanism for the transport of water and nutrients through the vascular system of plants as capillary action is not sufficient alone. Based on this knowledge, the next part will focus on the counting of stomata, which are small openings on the surface of leaves that allow for gas exchange during transpiration. Different plants exhibit variation in the number, distribution, and type of stomata. Within a single plant, the distribution and number of stomata may vary between the upper and lower surfaces of the leaves.

Objective: remove the surface layer of the back and front of the cucumber leaf (peel off the thin surface layer of the leaf) and observe the stomata and epidermal cells on the back and front of the cucumber leaf.

#### Procedure:

- 1) Put a drop of water on a microscope slide.
- 2) To remove the surface layer of the back of the leaf, gently fold the leaf over your index finger with the back of the leaf facing up / toward you. Using forceps, peel off the thin, transparent membranous skin on the back of the cucumber leaf. A small piece of the peeled leaf surface is sufficient for the sample that you will view under the microscope. It is enough to obtain a small piece of the peeled layer.
- 3) Place the peel on a slide with a drop of water, then cover it with a coverslip, avoid air bubbles. Blot any excess water from the slide with a paper tissue or filter paper.
- 4) View the sample under the microscope.
- 5) Adjust the focus and choose the **appropriate magnification** [400x] to focus on the peel and observe the stomata, guard cells, and epidermal cells.
- 6) Count the number of stomatas in the field of the vision without moving the slide.
- 7) Move the slide and count the number of stomata again. Record your observations.
- 8) Repeat step 7
- 9) Calculate the average number of stomata in the field of the microscope.
- 10) Draw a biological drawing of a stoma and surrounding epidermal cells and label its parts (see the criteria provided before).
- 11) Clean the glass slides and coverslips for the next use.
- 12) Repeat the steps 1 11 with a peel from the upper surface.
- **3.3.4** Draw a scientific drawing of stoma from both sides of the leaf in the answer sheet. Label opening with A, guard cells with B, and epidermal cells with C. (10 p)
- **3.3.5.**Provide your measurements of stomatal number in the answer sheet. (6 p)

No.	Lower surface	Upper surface
Stomata in field 1		
Stomata in field 2		
Stomata in field 3		
Average stomata in field		

- **3.3.6.** Explain the difference in observed stomatal densities in different sides of leaves, by choosing the correct explanation. Mark the letter in the answer sheet. (1 p)
  - a) By having a higher amount of stoma on the upper side of the leaf, better efficiency of photosynthesis is ensured.
  - b) By having a lower amount of stoma on the upper side of the leaf, plants can perform photosynthesis also in the night.
  - c) By having a lower amount of stoma on the upper side of the leaf, plants protect themselves from loss of water on sunny days.
  - d) By having a higher amount of stoma on the upper side of the leaf, plants can take up extra water on rainy days.

## Problem 4 Water demand by plants

#### Materials and equipment

- Cucumber leaf shared with task 3
- Two 3ml cuvettes with lid
- 50 ml tube with 96% ethanol
- Mortar and pestle
- Funnel
- Filter paper
- 15 ml tube with a cap
- 2 plastic pipettes
- Spectrophotometer
- The solar intensity spectrum of the light source dependence on the wavelength (Graph1) on a separate page
- Millimetre paper
- Radiation power of the light source in the given spectrum:  $I_0 = 500 \text{ W/m}^2$

#### Theoretical background:

The intensity of light decreases as it passes through an object, because of the interaction between the electromagnetic field of light and electrons. As light is transmitted, part of it also can be absorbed, as part of the energy is transformed into internal, chemical, or other forms of energy. Light intensity is expressed as the power per area; hence intensity is proportional to power. As the power of light decreases, so does the intensity. Processes of transmission and absorption can be described with quantities called transmittance and absorbance. If the intensity of the light beam that enters the object is I<sub>0</sub>, transmittance T can be calculated using the formula:

$$T = \frac{I}{I_0} \cdot 100\%$$

where I is the intensity of light that is transmitted and depends on wavelength.

Absorbance A can be calculated using the formula:

$$A = \log_{10} \frac{I_0}{I}$$

An absorption spectrum  $A(\lambda)$  is sensitive to the interaction of atoms or molecules. At a given wavelength absorbance is proportional to the concentration of a solution. A device to determine absorbance and transmittance in the solutions is called a spectrophotometer. To determine the absorbance and transmittance of certain molecules, the device must be calibrated to the solvent, ethanol in our case.

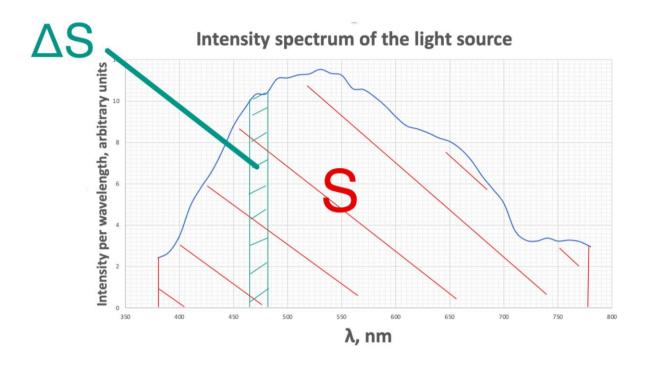


Figure 4.1. Solar intensity spectrum and area under the curve. Radiation power per square metre of the light source in the given spectrum:  $I_o = 500 \text{ W/m}^2$ 

In Figure 4.1 the total area under the curve S is the total intensity of the light source  $I_0$  and  $\Delta S$  is the area under the curve in a given range of wavelength. To calculate the intensity of the light source at the given wavelength range -  $\Delta I_0$  formula can be used:

$$\Delta I_0 = \frac{\Delta S}{S} \cdot I_0.$$

If all the area S is split into smaller areas under the curve and the intensity of each range is calculated, by knowing the percentage of absorbed light, the absorbed energy per second per square metre can be calculated.

**Objective of this task:** Determine how much energy in Joules is absorbed by a square metre of cucumber leaf every second?

#### Prepare cucumber leaf pigments for light absorbance measurements

- 1. Take approx. 4 cm<sup>2</sup> fragment of cucumber leaf, grind it in the pestle, and add 7 ml of ethanol. Continue grinding until the ethanol has extracted the pigment from the leaf (less than 1 minute)
- 2. Filter the solution through filter paper using a funnel into a 15 ml tube
- 3. Close the tube to prevent evaporation
- **4.1.** Call the lab assistant, who will record the result of your work together with your participant code. (1p).

Assistant will also show you to the spectrophotometer. You will have 15 minute time slots for measurements. If all devices are occupied, please wait. If you do not manage to acquire full spectrum, do not despair, you can continue to the next tasks with partial spectra and append data afterwards when device is free again.

#### Acquire the absorption spectrum of a cucumber pigment.

- 1. Transfer 3 ml of extracted pigment solution into a cuvette with a plastic pipette, and close the cuvette with a lid. Prepare another cuvette filled with ethanol, 3 ml.
- 2. Note that if absorbance exceeds the limits of the spectrophotometer, the device will display "1." In that case, you have to dilute the sample with ethanol. The absorbance maximum of pigment is at 440 nm.
- 3. You will have to measure the absorbance spectrum from 380 nm to 740 nm. Use 20 nm increments for taking initial measurements. The spectrum will have one to several peaks for absorbance, add additional measurements with 10 nm increments for higher resolution around maximums of the peaks.
- 4. Remember to calibrate with each wavelength

#### To measure the sample:

- 1. Switch on the spectrophotometer, and wait until it goes through the startup procedure.
- 2. Check if the Absorption is measured, indicated by ABS on the display. If not, use arrows pointing left and right to switch to the absorption mode (Figure 4.2.).

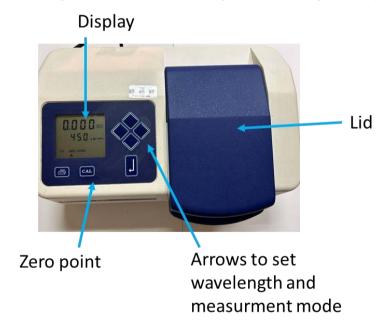


Figure 4.2. - spectrophotometer

- 3. Set the wavelength by pressing the UP and DOWN arrows.
- 4. Insert the cuvette, filled with ethanol for calibration. Be careful, observe the slit where light passes through to determine the orientation of the cuvette.
- 5. Press CAL to calibrate, and wait until the display shows 0.00(0) last digit may vary in time.

- 6. Make sure that the absorbance of the ethanol is equal to 0 and the transmittance is equal to 100%.
- 7. Take out the cuvette with ethanol and insert the cuvette with the sample.
- 8. Close the lid, take and write down the measurement.
- **4.2.** Write down measurements in the Answer sheet in Table (4 p)
- **4.3.** Plot the absorption spectrum  $A=f(\lambda)$  on the millimetre paper provided, and try to use most of the paper's area. Name the graph "Graph 2" (4 p)

Determine the amount of light energy absorbed by a square metre of cucumber leaf every second

To determine the energy absorbed by the leaf you will have to compare the areas below the curves in Graph1 (given on an extra sheet of paper) and Graph2

Split both graphs (Solar intensity spectrum and the absorption spectrum of a cucumber) into wavelength intervals. Choose the intervals yourself, the intervals can be different, but must match in both graphs

**4.4.** Mark the chosen intervals on both graphs (4 p)

Calculate the fraction of each interval area of the Solar intensity spectrum relative to the total area below the Solar intensity spectrum. You can choose a method for the area determination yourself.

**4.5.** Write the interval fractions on the Graph 1 (3 p)

Calculate the radiation power per square metre for each interval.

**4.6.** Write the radiation power per square metre of each interval on the Graph 1 (3 p)

The obtained absorbance values may differ from the actual absorbance values for a leaf depending on the concentration of the liquid and the dimensions of the cuvette hence normalising the data using coefficient k is needed. Let's assume that maximal absorbance that one could obtain is 2.

$$A=k\cdot A_0$$

where A - absorbance of a leaf, k - coefficient of normalization

$$k=\frac{2}{A_{0max}},$$

 $A_{0\text{max}}$ — the absorbance at the wavelength most absorbed by the leaf,  $A_0$  — the measured absorbance at the given wavelength.

- **4.7.** Write down your calculated k in the answer sheet, and show your calculation (2 p)
- **4.8** Determine the normalised average absorbance for each interval of the absorption spectrum. Write them on the Graph 2 (3 p)

To calculate k you used that maximal absorption of a leaf pigment at a certain wavelength is equal to 2. Using formulas provided for A and T, find how much of the light is transmitted through the sample if absorbance is 2.

**4.9.** Show your calculations and the final result in the answer sheet. (3 p)

Create a table in the answer sheet where you enter interval data from Graphs1 and 2.

**4.10.** Determine the absorbed energy per second (absorbed radiation power) for each wavelength interval, and show your calculation for one wavelength interval. (3 p)

Find the sum of all energies absorbed per second for all wavelength intervals to get the total energy absorbed per second (total absorbed radiation power).

**4.11.** Write down your calculated sum in the answer sheet (1 p)

Let us assume that the cucumber plant consumes 3·10<sup>-4</sup> ml of water for every joule of light absorbed. How much water is needed in one hour per square metre of cucumber leaf?

**4.12.** Write down your calculated value in answer sheet, show calculation (2 p)

## Problem 5 Exploring hydroponics

#### Materials and equipment

- 2 PVC tubes different length
- 90 degree elbow tube
- Measuring tape
- Adhesive tape
- A container with a connector for a PVC tube
- a stand
- Water
- Stopwatch on the screen
- Graduated cylinder
- Permanent marker
- Clamp for stopping water flow
- Milimeter paper
- scissors
- Computer with excel
- Instruction for excel use on separate page <a href="https://docs.google.com/document/d/1c5jdtoepVibED5-yWu07zIO5">https://docs.google.com/document/d/1c5jdtoepVibED5-yWu07zIO5</a> 4QJFf3l/edit

When water moves through a tube, the adjacent layer of water is slowed down by the walls of the tube. The remaining layers slide over one another and are also slowed down. Let's assume that the flow rate Q of water in a tube is proportional to the pressure difference  $\Delta p$  on both sides of the tube and the length  $\boldsymbol{l}$  of a tubes to the power of n:

$$Q = \frac{\Delta V}{\Delta t} \propto \Delta p \cdot l^n$$

where  $\Delta V$  is the volume of the water that flows through the cross-section area of a tube in the time interval  $\Delta t$ . The density of water  $\rho$  is  $1000 \text{ kg/m}^3$ , g is the acceleration due to gravity equals to 9,81 m/s², and in h is the level of water above the free end of the tube.(see figure 5.1)

**Objective of this task:** Find the value of n by measuring flow rate as a function of tube length by decreasing the length of a tube.

**5.1.** Measure the initial length of both PVC tubes. Write down the measurements in the answer sheet (1 p).

Slide the 90 degree elbow tube on the PVC tube. Connect the tube to the connector to create the experimental system given in Figure 5.1. Use adhesive tape to fix the tube on the table surface. Use adhesive tape to wrap the end of PVC tube before connecting to container to avoid leaks.

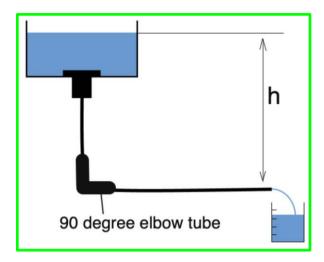


Figure 5.1. Schematic representation of experimental system

Fill the system with water, avoid air bubbles, and make sure there is no leakage. In case of severe leakage, ask the lab assistant for help. Mark the water level on the container with a small line, you will need this line to refill the tank.

Measure the time and volume of water that flows out of the tube. Choose height and volume wisely. Make sure that the water level drop in the tank is not significant so that the pressure difference doesn't change. Restore the water level, so that h is the same every time you measure. Remove the long tube and attach the shorter tube, perform same measurements.

**5.2.** Write down the measurement for each tube length () Write them in the table in the answer sheet. Calculate  $\Delta p$  for each measurement (3 p)

Decide if this setup is satisfactory, if not, choose the variable you will change and repeat initial measurements.

**5.3.** Choose the final setup parameters for the rest of the experiment fill I in the answer sheet. (2 p)

When you have obtained the setup you consider optimal, calculate the flow rate Q.

**5.4.** Show calculations and final results for Q in the answer sheet. (2 p)

Change back to the long tube. Shorten the long tube a little bit (don't forget to measure how much you have cut off). Make sure to cut just the horizontal part of the tube and restore the water level, so that h is the same every time you measure.

Make at least 8 measurements with ever decreasing tube length while keeping the height difference constant.

**5.5.** Fill in your data in answer sheet (4 p)

Plot graph  $Q=f(\ell)$  in Excel. Add trendline and equation to find n. Choose the correct function for the trendline. Instructions on how to operate Excel can be found on a separate sheet.

**5.6.** Write trendline equation in the answer sheet (2 p)

#### **5.7.** Write down your obtained n (2 p)

Let us assume that each hour water flow is turned on for 1 minute. Calculate how long should be the tube of your system (and your used water level) to provide the right flow rate according to the results in Task 4.12.

**5.8.**Show calculations and write the answer in the answer sheet. (2 p)

### Problem 6 Designing greenhouse

- **6.1.** Based on the groundwater composition analysed in the Problem 1, which is the most probable bedrock of your greenhouse site? Write the letter of the answer in the answer sheet. (1 p)
  - a) Sand
  - b) Dolomite
  - c) Clay
  - d) Granite
- **6.2.** Why should groundwater you have in the greenhouse site, not be used for feeding solution preparation? Write the letter of the answer in the answer sheet. (1 p)
  - a) It has too much magnesium that would be harmful for plants.
  - b) It would build a limescale in tubes.
  - c) It would cause all minerals to form sediments and would not be usable by plants.
  - d) Calcium in the water would form Ca(OH)<sub>2</sub> making water unsuited to plants.

You decide that you will build a greenhouse that has space for 100 cucumber plants. Leaf area index is the one-sided leaf area per unit of ground surface area. For cucumber cultivation it is assumed that the leaf area index should be 3.

**6.3.** Indicate which of the consequences mentioned below might occur if you would plant cucumbers to have larger or smaller leaf area index. The consequences that you would observe in case of larger index mark L and for those in the case of smaller, mark S, put 0 if none fits.. Do not forget to write your answer in the answer sheet. (4 p)

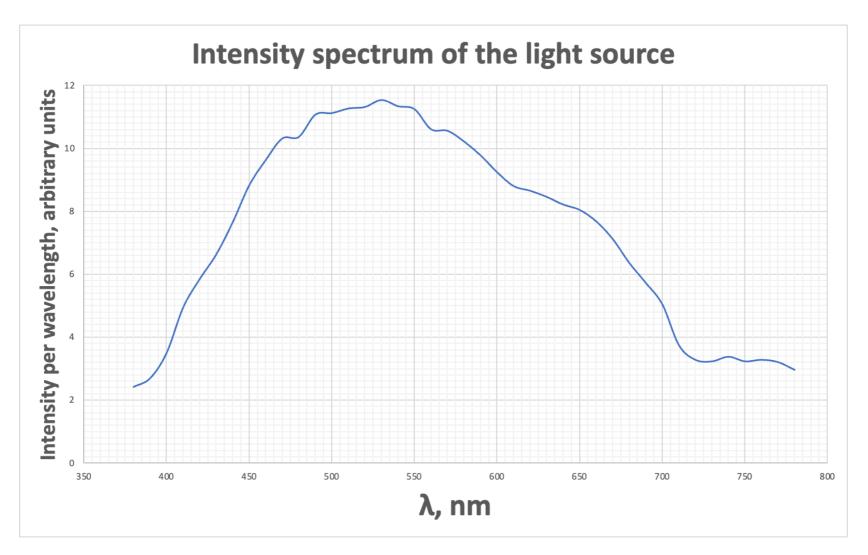
Leaves would receive too little of light

You would experience economic losses

Air in the greenhouse would be too humid

- **6.4.** Estimate the area you need to grow 100 cucumber plants based on the estimated area of a cucumber leaf you had on your table in the beginning of the task. It is known that usually in a greenhouse you have 15 leafs per plant. (2 p) If needed, a leaf will be shown to you.
- **6.5.** You decide to use glass to cover your greenhouse. Absorbance of which part of the light spectrum by glass should be avoided? Write the letter of the answer in the answer sheet. (1 p)
  - a) Green
  - b) Red and blue
  - c) Infrared
  - d) Ultraviolet
- **6.6.** Plants require light and water to grow. You observe that water demand by plants increases on a sunny day compared to a cloudy one. Pick the correct explanation, based on your observations and write the letters of the answers in the answer sheet. (2 p)
  - a) In sunny days transpiration through leaves increases
  - b) In sunny days transpiration through stem increases
  - c) In sunny days transpiration through leaves decreases
  - d) In sunny days xylem diameter decreases

- **6.7.** By measuring air humidity and assessing the amount of water you fed to your plants, you discover that  $\frac{2}{3}$  of water stays in the plant. How is this water used? *Pick the correct explanation/s, based on your knowledge and write the letter(s) of the answer in the answer sheet. (1 p)* 
  - a) It stays in plant cells as cytoplasm
  - b) It is used in photosynthesis as a source material for glucose
  - c) It is used in photosynthesis as a source material for CO<sub>2</sub>
  - d) It is used to maintain the shape of the plant by maintaining turgor pressure



Radiation power of the light source in the given spectrum:  $I_o = 500 \text{ W/m}^2$ 

