Task 2

OCEAN

11th of May 2017

EUSO COPENHAGEN 2017

COUNTRY:

TEAM:

General Instructions

You have 4 hours to complete the tasks.

Wear laboratory coat and protective goggles at all times in the laboratory.
Eating and drinking is prohibited in the laboratory.

It is highly advisable to wear disposable gloves and protective eyewear when handling chemicals.

All paper used, including rough work paper, must be handed in at the end of the experiment.

All results must be entered into your blue answer sheet.

Your graphs must be handed in along with the blue answer sheet.

Printing from LoggerPro: Be sure to write country and team in the footer before you print, so you can get your print from the lab assistants.
Afterwards, you may write by hand on the printouts to show readings etc.

Only the final answer sheet, and the attached graphs, will be marked.

The Task consists of 3 experiments.

Experiment 1: 34 marks
Experiment 2: 32 marks
Experiment 3: 34 marks

Total: 100 marks

Introduction

Njord is a fish farmer who grows fish in the Kattegat. He has busy days running a family business, so Njord gets a lot of help from his smart 16-year-old daughter Freja.

Recently Njord decided that instead of buying the small creatures his fish eat, he should produce it locally, starting with fresh microalgae. Photosynthetic algae need water, carbon dioxide, light and nutrients to grow. Nutrients can be costly. It takes a lot of energy to remove nitrogen from the atmosphere and phosphorous has to be mined and will run out someday. An alternative to using these resources is wastewater, which needs to have these things removed before it's discharged in the environment. Anaerobically treated wastewater will have high nitrogen (N) and phosphorus (P) content, but not much organic carbon left, and has been shown to be a good algae growth medium.

One day, Freja went to help her father and she saw he had bought a fancy set of experimental photo bioreactors, filled them with wastewater and a panel of LED's were shining on them.

NOTICE: Note that part B has some time-consuming steps and you may want to read ahead and have one team member start them first.

Kattegat, the sea between Denmark and Sweden.

1. Algae production and wastewater treatment measurements using spectrophotometry.

Njord started pumping wastewater into his reactor at a steady rate to his algae photo bioreactors, left the LEDs on and forgot about them while he went out in the Kattegat to fix some fish cages. When he came back, he noticed that some algae had grown in the reactor and that they were waiting for him in the big tank he set up to collect the effluent. Not knowing how to analyse what had happened, he called Freja. Njord asked:

A. **How much algae am I producing?**

B. **How much carotenoids are the algae producing?**

C. **How much nutrients are they removing from the wastewater? Will we get credit for that?**

Freja dutifully came by and observed the system. She also set up a large plastic container on a big scale and waited an hour to see that 21.5 L of algae suspension had come out of the reactor. (She assumed a density of 1 kg/L.) Then she collected a few samples to take to measure at her high school. She labelled them:

- Sample 1: “Influent” - the wastewater flowing into the photo bioreactor system.
- Sample 2: “Effluent” - the liquid with algae flowing out of the photo bioreactor system.

Now you have to help Freja figure out how to answer Njord’s questions using the tools she had, especially spectrophotometry.

A spectrophotometer is a device where light of a chosen wavelength is detected after it passes through a cuvette containing a sample. The amount of light absorbed by the sample is called Absorbance (A).

$$A = -\log_{10}\frac{l}{l_0}$$

l_0 is the incident light intensity and l is the intensity of the transmitted light.

Absorbance is proportional to the concentration of the sample (c) and the distance the light travels (d) and a substance specific constant (ε). Most commonly in spectrophotometers the cuvette thickness (d) is 1 cm.

$$A = \varepsilon \cdot c \cdot d$$

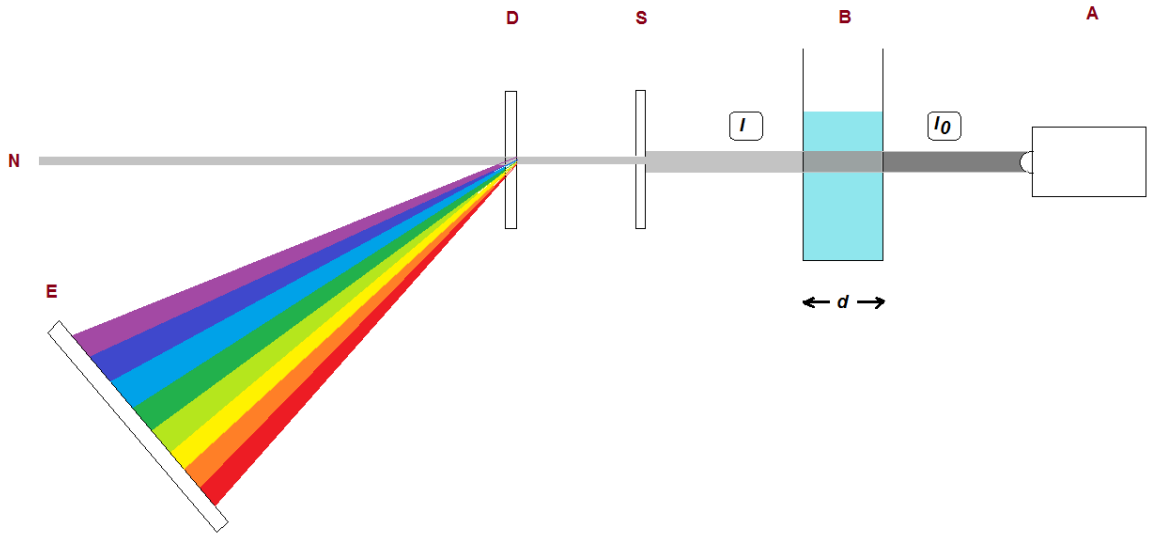


Figure 1.1. Spectrophotometer principle. From right: Lamp (A), Cuvette (B), Slit (S), Diffraction grating (D), Direct beam (N, not used), Detector array (E).
A photo of the actual inside is shown in Appendix B.
There, the diffraction grating is mounted askew to increase resolution and the slit is not visible.

Materials

Shared between teams

- Heat blocks set at 60 °C
- Ice container
- Freezer
- Shovel for ice

At each station

- Laptop with LoggerPro
- Calculator
- Spectrophotometer with SpectroVis optical fibre for light emission
- Black cloth for spectrophotometer
- Centrifuge for 2 mL tubes with instruction
- Vortex mixer
- White light source
- 4 Racks for 50/15/2 mL tubes
- Lens paper for cuvette cleaning
- 2 Timers
- Dispenser with deionized water
- Ice bowl
- Waste bin for solid waste
- Waste container for organic+inorganic waste
- Plastic beaker 250 mL
- Dispenser with paper towels
- Soap
- Safety glasses

For each team

- A box with:
    - Visocolor school kit for Ammonium and Phosphate determination
    - 3 glass vials with lid
    - Plastic spoon
- 50 mL influent wastewater in a glass bottle with a screw lid
- 50 mL effluent water with algae in a glass bottle with a screw lid
- Centrifuge tubes
    - 2 x 50 mL graded tubes (notice where the 50 mL mark is)
    - 6 x 15 mL graded tubes
    - 16 x 2 mL graded tubes
- Cuvettes of thickness of 1.00 cm
    - 16 x 4,5 mL
    - 4 x 1.5 mL
- 20 disposable pipettes of 1.0 mL with 0.25 mL graduation
- 25 mL of 96% ethanol in a pipette bottle
- LED box
- Plastic ruler 20 cm
- 3 Pencils
- List book
- Black waterproof marker pen
- Safety gloves

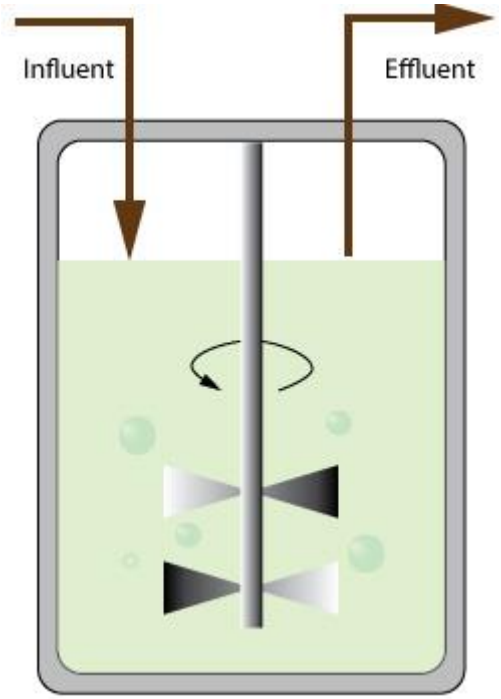


Figure 1.2. Simplified flow diagram.
For these exercises, we can think mostly about the feed (influent) and the effluent.
A detailed diagram of a lab scale algae reactor is shown in the Appendix A1.

A. How much algae am I producing?

When algae technologists ask this question, they want a result in terms of mass of algae dry weight per volume of photobioreactor per unit of time. Often $g \cdot L^{-1}$ day^{-1} is used. We call this volumetric productivity (*VP*). In a continuous reactor, volumetric productivity can be easily calculated by multiplying the Dilution rate (*D*) with the concentration of algae.

$$VP = D_w \cdot D ,$$

where D_w is the dry-weight concentration in the units g dry-weight per L, and the dilution rate is in the units day^{-1}.

Dilution rate can be measured like this, Freja found on Wikipedia:

Dilution rate

At steady state, the specific growth rate (μ) of the microorganism is equal to the dilution rate (D). The dilution rate is defined as the flow of medium per time (F) over the volume of culture (V) in the bioreactor

$$D = \frac{\text{Medium flow rate}}{\text{Culture volume}} = \frac{F}{V}$$

Question 1.1

Given Freja collected 21.5 L/hour of effluent and the reactor has a volume of 400 L, what is the dilution rate in units day^{-1}?

➢ *Write your calculations and answer in the answer sheet, box 1.1.*

Spectrophotometry is commonly used by biotechnologists as a quick way to measure the dry weight of cell biomass in a system. Since cells both absorb and scatter light, it is not a perfect measure, so you have to be sure to use it in a limited range of cell density. Still it is much faster than filtering, drying and weighing biomass. The first step is to calculate the Dry Weight concentration (D_w) in g/L - from the measured absorbance at 750 nm (A_{750}), with a formula, which Freja has provided for you:

$$D_w = 0.4561 \frac{\text{g}}{\text{L}} \cdot (\text{Absorbance at } 750\text{nm})$$

Measure absorbance

Turn on and calibrate the spectrophotometer with deionized water.

Question 1.2

Measure a complete absorbance spectrum for all wavelengths with the effluent algae suspension in the cuvette of 1 cm thickness; see **Figure 1.1**. Save the spectrum from the File menu – your team will need it in Question 2.3.

Measure the effluent algae absorbance at 750 nm **in a 1.00 cm cuvette**.

Remember to mix before measurement, as it settles very fast.

➢ *Write your measurements in the answer sheet, box 1.2.*

Use the formula to calculate the dry weight concentration in g/L.

➢ *Write your calculations and answer in the answer sheet, box 1.2.*

Question 1.3

How much algae are we making? In other words, what is the volumetric productivity in $g \cdot L^{-1}$ day^{-1}?

➢ *Write your calculations and answer in the answer sheet, box 1.3.*

Question 1.4

How much would the reactor produce if operated at the same rate for one year? (Show your work.)

➢ *Write your calculations and answer in the answer sheet, box 1.4.*

B. How much carotenoids are the algae producing?

A lot of the reason that fish are considered a healthy food is because they eat organisms that eat algae. Many algae contain carotenoids, auxiliary photo pigments. Carotenoids, especially Astaxanthin, are a small part of the salmon diet, but they account for the nice pink-red colour and a significant portion of the cost. As strong anti-oxidants, these molecules are also considered a healthy part of the human diet. Because both chlorophylls and carotenoids absorb light at around 450-500 nm (**Figure 1.3**), it can be tricky to measure carotenoids directly in cell extracts. Therefore, scientists have developed ways of estimating the carotenoid content by comparing the peaks at different wavelengths.

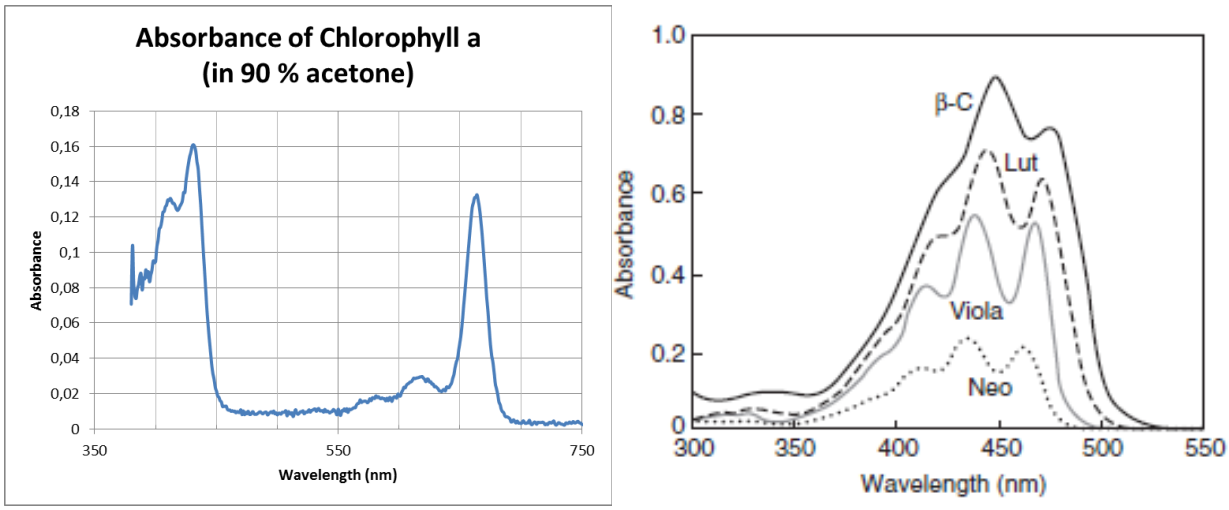
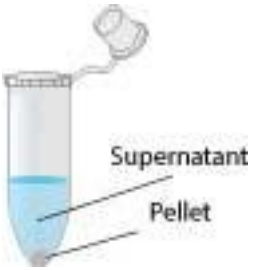


Figure 1.3. Absorbance spectra of chlorophyll a in acetone compared to the carotenoids beta-carotein, lutein, violaxanthin, and neolaxanthin (From Lichtenhaler and Buschmann 2001).

Follow the protocol below to extract and measure the pigments:

<u>Be aware that this experimental step is a bottleneck and time consuming.</u>

Protocol



1. Balance the centrifuge – look at the instructions placed by the centrifuge. Each tube has to have a tube placed opposite with the same volume, so the weight is equally distributed.
2. 2 mL culture is centrifuged at 6000 rpm for 5 minutes.
3. Remove and discard the supernatant. Take care – the pellet can be on the tube side.
4. 2 mL 96% Ethanol is added to the pellet. Shake and mix the tube to begin suspending the pellet.
5. Hold the tube on the Vortex mixer for 5 minutes to fully disrupt the pellet.
6. Incubate at 60 °C for 40 minutes. Then in the ice bath for 15 minutes.
7. Centrifuge again.
8. Transfer the supernatant to the cuvette, being careful not to disturb the pellet. Insert the cuvette in the spectrophotometer in an orientation to let the light travel 1.00 cm through the cuvette. Measure the absorbance spectrum of the supernatant.

Use the equations below to calculate the concentrations of chlorophyll a (c_a), chlorophyll b (c_b) and total carotenoids ($c_{(x+c)}$):

In a solution with Ethanol 96%:

$$c_a = (13.36\ \text{mg/L}) \cdot A_{664} - (5.19\ \text{mg/L}) \cdot A_{649}$$

$$c_b = (27.43\ \text{mg/L}) \cdot A_{649} - (8.12\ \text{mg/L}) \cdot A_{664}$$

$$c_{(x+c)} = \big((1000\ \text{mg/L}) \cdot A_{470} - 2.13\, c_a - 97.64\, c_b\big)/209$$

where A_{470} is absorbance measured at 470 nm, A_{649} absorbance measured at 649 nm, and A_{664} absorbance measured at 664 nm.

Question 1.5

What values did you measure for A_{470}, A_{649}, and A_{664}?

- *Write your answer in the answer sheet, box 1.5.*

Question 1.6

What did you calculate for c_a, c_b, and $c_{(x+c)}$?

- *Write your calculations and answer in the answer sheet, box 1.6.*

Question 1.7

How much chlorophyll (a+b) and carotenoids do the algae contain in mg per gram dry weight?

- *Write your calculations and answer in the answer sheet, box 1.7.*

Question 1.8

What are the production rates of these molecules?

- *Write your calculations and answer in the answer sheet, box 1.8.*

C. How much nutrients are the algae removing from the wastewater?

To answer this, Freja gets two simple colorimetric test kits that the high school had in the closet and follows the instructions. Measure N and P in both the influent and the effluent waters.

Note: The concentrations of N and P in the synthetic influent differ from the concentrations in normal wastewater, in order to make it possible to measure with little practice.

Use the centrifuge to remove the algae from at least 4 x 2 mL of effluent sample. Save the supernatant for testing. The algae pellet is not needed.

Analyse Ammonium

Dilute the influent 50 times before analysis using plastic pipette and graded test tube. The effluent does not need dilution.

1. Fill a glass vial with the sample to the 5 mL mark, using a plastic pipette
2. Add 10 drops of the reagent labelled NH_4 - 1
3. Seal the vial and mix
4. Add 1 level measuring spoonful of the reagent labelled NH_4 - 2 (the spoon is in the kit)
5. Seal the vial and shake until the powder has dissolved
6. Wait 5 minutes
7. Open the vial and add 4 drops of the reagent labelled NH_4 - 3
8. Seal the vial and mix
9. Wait 7 minutes
10. Read the absorbance in the spectrophotometer at 700 nm
11. Use the standard curve in **Figure 1.4** to determine the concentration

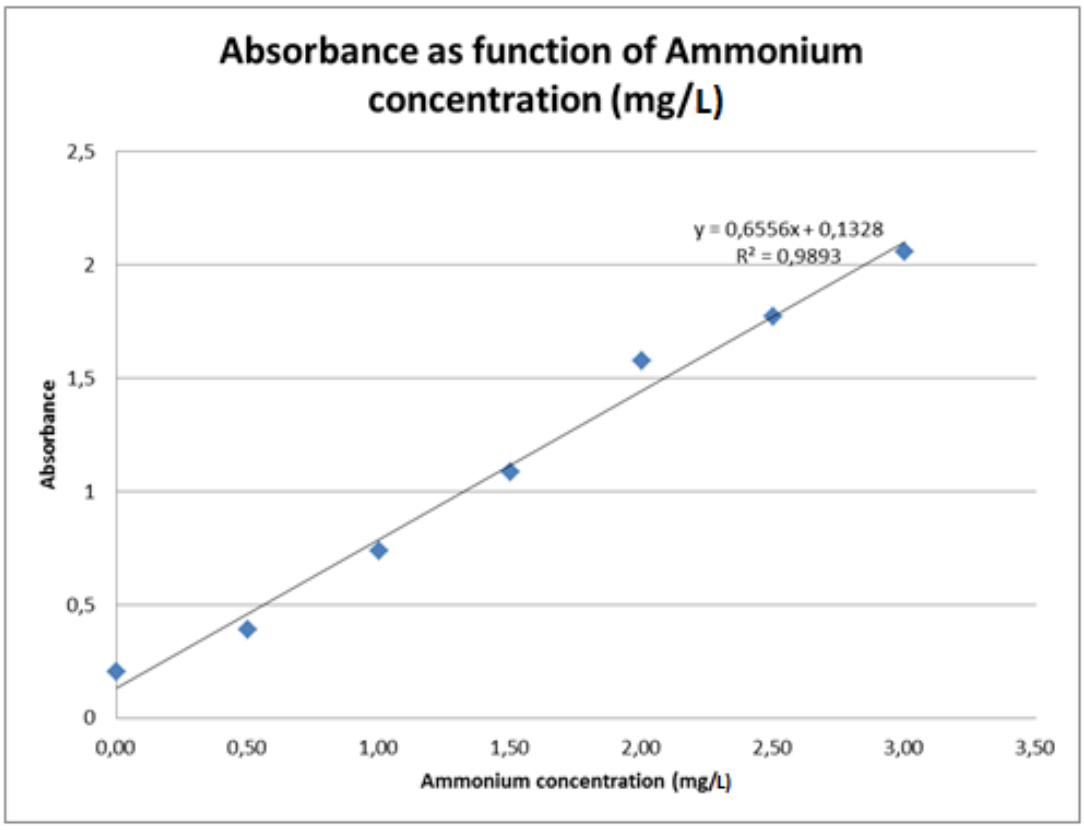


Figure 1.4. Standard curve for absorbance-ammonium concentration correlation.

Question 1.9

What values (see the answer sheet) for NH_4^+ did you determine for influent?

What values (see the answer sheet) for NH_4^+ did you determine for effluent?

What is the NH_4^+ removal percentage?

What is the NH_4^+ removal rate in $mg \cdot L^{-1} \cdot d^{-1}$?

➢ *Write your calculations and answer in the answer sheet, box 1.9.*

Analyse Phosphate

We expect much higher levels in both influent and effluent of PO_4^{3-}, so please dilute your original influent and effluent samples 10 times before starting the test, using plastic pipette and graded test tube. Make sure to rinse and dry vials that you reuse.

1. Fill a glass vial to the 5 mL mark with the sample, using a plastic pipette
2. Add 6 drops of the reagent labelled PO_4 - 1
3. Seal the vial and mix
4. Add 6 drops of the reagent labelled PO_4 - 2
5. Seal the vial and mix
6. Wait 10 minutes
7. Measure Absorbance at 700 nm
8. Use the standard curve in **Figure 1.5** to determine the concentration

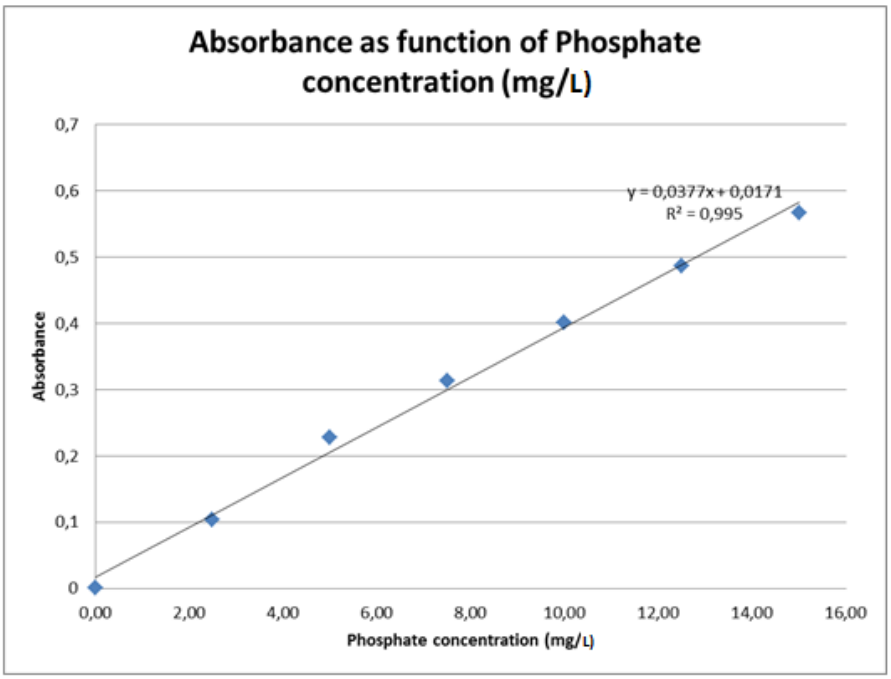


Figure 1.5. Standard curve for absorbance-phosphate concentration correlation.

Question 1.10

What values (see the answer sheet) for PO_4^{3-} did you determine for influent?

What values (see the answer sheet) for PO_4^{3-} did you determine for effluent?

What is the PO_4^{3-} removal percentage?

What is the PO_4^{3-} removal rate in $mg \cdot L^{-1} \cdot d^{-1}$?

➢ *Write your calculations and answer in the answer sheet, box 1.10.*

Question 1.11

Use nutrient removal rate and volumetric productivity to estimate the N and P content of the algae.

- *Write your calculations and answer in the answer sheet, box 1.11.*

Question 1.12

What is the annual saving in DKK/L reactor, given the Danish tax of 5 DKK per kg N and 110 DKK per kg PO_4^{3-}?

- *Write your calculations and answer in the answer sheet, box 1.12.*

2. Designing an illumination system for algae production using LEDs.

After analysing the absorbance spectra Freja asked her father, “Why do you use day-light LEDs?” And Njord answered: “Because I suppose the algae need daylight to grow!” “Yes”, Freja said, “That is true, but they only need part of it.”

Compare your absorption measurements in **Question 1.2** on algae in water with the spectrum in **Figure 1.3** of chlorophyll a in acetone.

Question 2.1

Which colours can Freja identify in **Figure 1.3** as relevant for the photosynthesis? State the wavelength intervals for chlorophyll a. (Note that in acetone these are shifted relative to those you would observe in water for your algae.)

➢ *Write your answer in the answer sheet, box 2.1.*

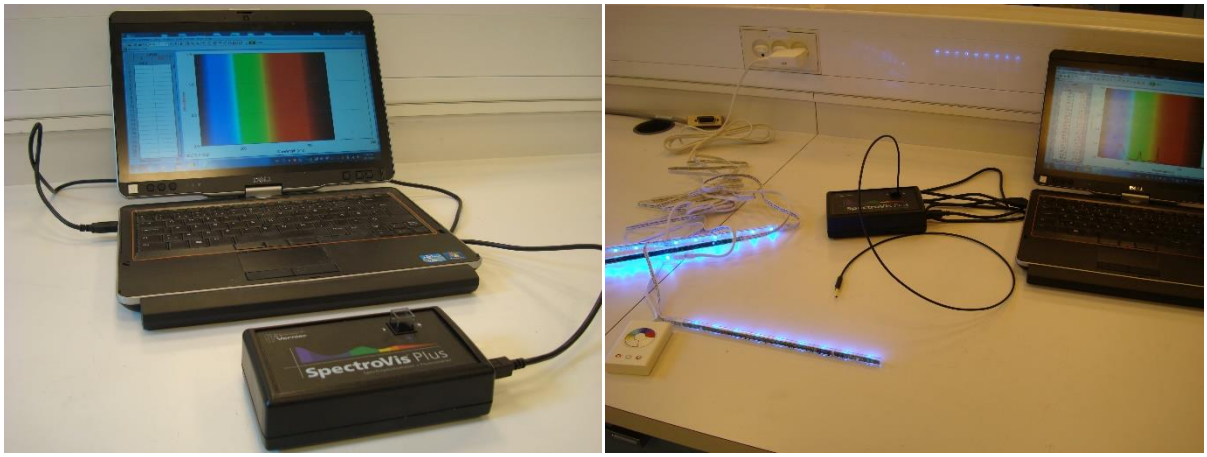
Freja continued:

“Let me illustrate how much you can save on your electricity bill by providing just the necessary light. I will take the red part of the spectrum as an example.”

Question 2.2

Which spectra does Freja have to compare for her example?

➢ *Tick your answer or answers in the answer sheet, box 2.2.*



Absorption measurement.
Spectrophotometer with cuvette and cable connected to a computer with LoggerPro.

Emission measurement.
Spectrophotometer with optical fibre unit and cable connected to a computer with LoggerPro.

Figure 2.1a

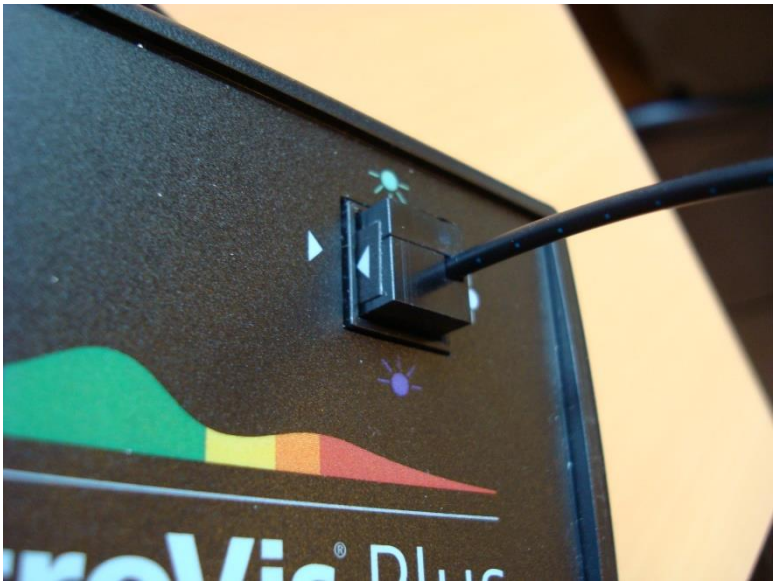


Figure 2.1b. Close-up of fibre unit mounted for emission measurements. Note the matching arrows.

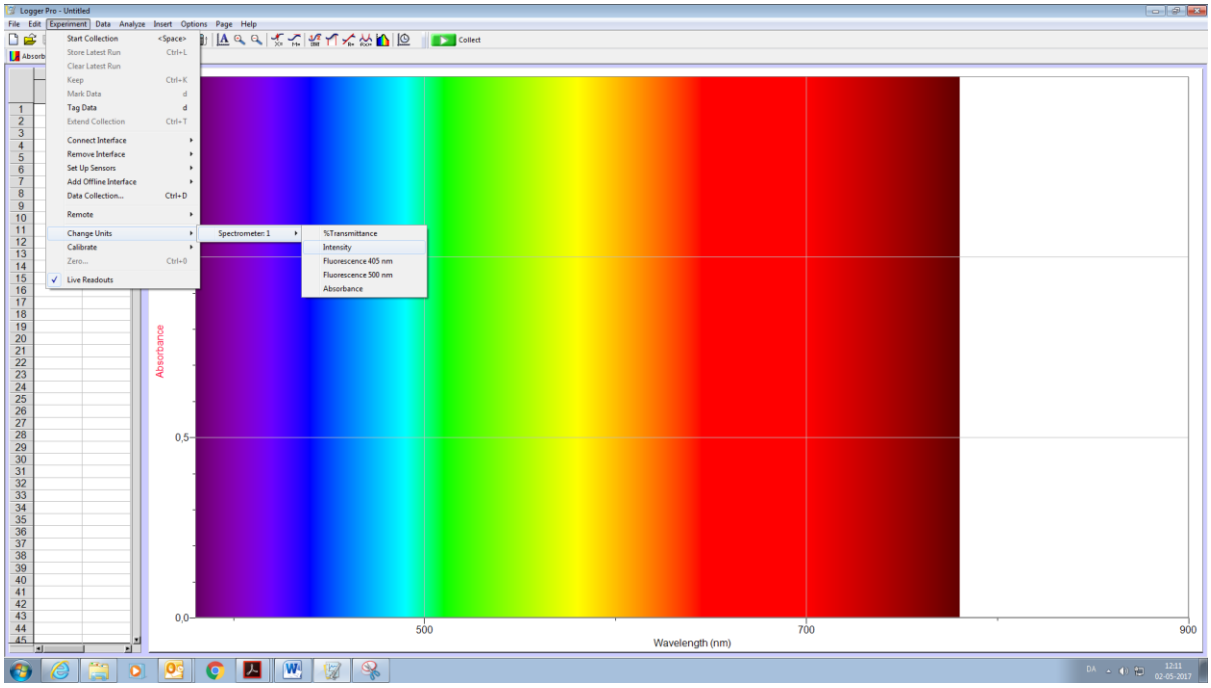


Figure 2.2. "Change units" under "Experiment" for setting of spectrophotometer mode. Choose "Absorption" for absorption and "Intensity" for emission spectra.
Look inside the spectrophotometer in Appendix B.

Measure the absorption peak of the algae in the red part of the spectrum (or retrieve the spectrum saved in Question 1.2)! Keep the graph, but change the setting to "intensity" in Logger Pro (follow the link Experiment -> Change Units ->…). Notice, that the absorption graph now shows a dip, where light is absorbed.

In the following, where you are going to investigate the emitted light spectrum from various light sources, keep this setting.

Question 2.3

Measure the emission spectrum of the red LED in the multicolour LED bar! Show this spectrum together with the spectrum from Question 2.2 in the same graph with intensity on the y-axis[1].

➢ *Print your graph with team number. Mark it with "Graph 2.3".*

Question 2.4

How many nanometres should the red LED light be shifted to match the red absorption dip for the algae?

➢ *Write your answer in the answer sheet, box 2.4.*
Add your readings of the necessary numbers to "Graph 2.3".

"OK", Freja said. "Let us study this. It does not sound right that the plants can only grow at very particular colours. Let us study the literature..."

Aha, Njord says, I think I have found it. "Plants can collect photons also with higher energy and transport it to the chlorophyll for use there. (You see, light comes in quanta called photons, and are only absorbed in full quanta.) The collection is done by carotenoids. You see the graph below that shows the CO_2 consumption of algae illuminated by light of different frequencies. It shows that the green alga, Chlorella, collects photons between 400 nm and 680 nm with nearly the same efficiency, called the quantum efficiency η_λ. More precisely: When the energy is transported to chlorophyll a, it is absorbed there with an efficiency η_λ called the quantum efficiency."

[1] When you shine light from a strong light source into the spectrophotometer, the graph may get a flat top in certain intervals, due to saturation. You can avoid this either by reducing the sample time (follow the link Experiment -> Set up sensors -> Spectrometer…), or more simply, by moving the light source a bit away from the instrument (taking care to avoid stray light from other sources).

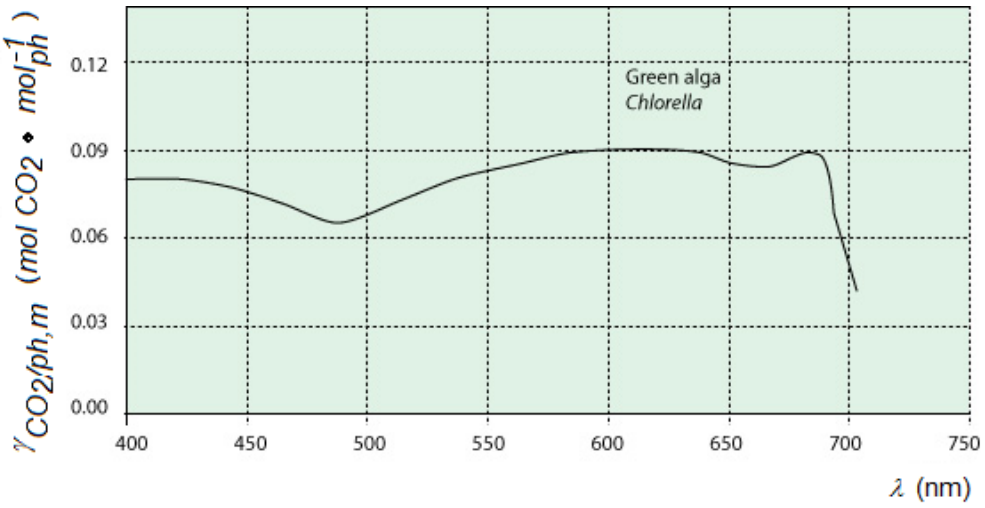


Figure 2.3. The CO2 consumption in mole/mole photons absorbed as a function of photon wavelength. This consumption can be taken to be proportional to the quantum efficiency η_λ.
After Emerson, Robert, and Charlton M. Lewis. "The dependence of the quantum yield of Chlorella photosynthesis on wave length of light." American Journal of Botany (1943): 165-178.

Question 2.5

What process in the plant does the consumption of CO_2 signify?

➢ *Tick your answer in the answer sheet, box 2.5.*

Question 2.6

Measure the emission spectrum for the green LED in the LED bar! Read off the peak wavelength. For Chlorella, in **Figure 2.3**, find the ratio of the quantum efficiency at this wavelength relative to the quantum efficiency at the wavelength of the red LED peak. Show how you read off the necessary numbers from the graph.

➢ *Print the graph and write your calculations and answer in the answer sheet, box 2.6.*

"So, it is not such a big waste after all to use my white light LEDs", said Njord, "They seem to match quite perfectly the wavelength interval which results in photosynthesis, also called "the photosynthetic active regime" (PAR). You see the spectrum for yourself!"

Question 2.7

Measure the spectrum of the white light LED bulb and compare it with the absorption spectrum of the algae.

➢ *Print the graph and write your answer in the answer sheet, box 2.7.*

"Well, it is more complicated than that. You lose more than the quantum efficiencies indicate!" replied Freja. "You see, light comes in quanta called photons, and are only absorbed in full quanta. The energy of a quantum of light at a certain frequency is proportional to the frequency." The constant *h* of proportionality is called Planck's constant and its value is $h = 6.626 \cdot 10^{-34}$ J/Hz or $h = 4.136$ meV/THz (note: 1 THz $= 10^{12}$ Hz). The latter unit contains the energy unit electronvolt

and is practical for atomic processes. One electronvolt is the energy that an electron gains when it goes through a one volt battery, $1\ \mathrm{eV} = 1.602 \cdot 10^{-19}$ J, and $1\ \mathrm{Hz}$ is $1\ \mathrm{s}^{-1}$. The speed of light c is the wavelength times the frequency.

Question 2.8

Using the table in the answer sheet, calculate the energy in eV of a photon at the peak wavelength of the green light LED, at the peak wavelength of the red-light LED and at the red absorption dip of your algae sample.

- *Write your answer in the answer sheet, box 2.8. State the wavelengths you use for your calculation and state the related frequencies of these wavelengths.*

Question 2.9

For one photon reaction step to take place, how much energy (in eV) needs to be absorbed at the red dip in your algae graph from Question 2.3? How many percent of energy is wasted if this is provided by a green, respectively red LED and the photon energy is transported by the carotenoids to the chlorophyll? What are the respective efficiencies in energy consumption?

- *Write your answer in the answer sheet, box 2.9.*

Question 2.10

How many percent light energy is lost if Njord uses green LEDs to "feed" the red absorption in the chlorophyll instead of LEDs optimized to that absorption? Remember to take into account that red, yellow and green LEDs are similar in construction and can be assumed to convert the electric input energy to light energy output with equal efficiency. Take also into account the quantum efficiencies. Finally, note the algae can also obtain energy for use in the chlorophyll through the absorption of green light by the carotenoids.

- *Write your answer in the answer sheet, box 2.10.*

3. Predator-Prey interactions.

NB: You will have to use and study the Appendix C carefully in order to solve this part.

Freja said to her father Njord, "The microalgae are very good to use as feed for copepods. The copepods can afterwards be used as feed for fish. In school, we have taken some videos of how fast different species of Copepods can swim and the size of the escape distance from a fish."

The videos are placed on the desktop. By using video analysis in "Logger Pro", find out which of the two copepods species we shall choose as feed for the fish, when the fish has a reaction distance to the copepod prey of 5 mm and attacks at a speed of 200 mm/s over a distance of 20 mm. Look in Appendix C1 for introduction to Copepods and Appendix C2 for Experimental filming-setup.

Freja asked Njord to read the following!

We experimentally investigate 'predator detection distance' and 'escape speed' of two different copepod species. We introduce one species at the time in the aquarium to observe their predator detection distance and escape speed. This gives us two movies to analyse: movie 1 shows the escape behaviour of the species *Centropages hamatus*, movie 2 shows the escape behaviour of the species *Temora longicornis*. Both movies are filmed at 500 frames per second, and the horizontal width of the movie frames is 39 mm.

Open the movies in Logger Pro. Get readings of time in seconds by setting the "Movie Options" – "frame rate". Use the "Set Scale" function to convert distances to mm.

Question 3.1

For each of the 2 copepods, by performing the 13 steps below, determine:

- the predator detection distance (in mm) of the two copepod species (i.e. the distance between the tip of pipette and the copepod at the time it starts escaping).
- the jump distance (in mm) of the two copepod species.
- the jump speed (in mm/s) of the two copepod species.

➢ *Write your answer in the answer sheet, box 3.1.*

1. At what time (in s) does the copepod start the jump?
2. At what time (in s) does the copepod end the jump?
3. What is the distance (in mm) in the z-direction from pipette tip to the copepod when the copepod starts the jump?
4. What is the distance (in mm) in the z-direction from pipette tip to the copepod when the copepod ends the jump?
5. Print the graph of z as a function of time for the jump, and attach it to the answer sheets.
6. What is the distance (in mm) in the x-direction from pipette tip to the copepod when the copepod starts the jump?
7. What is the distance (in mm) in the x-direction from pipette tip to the copepod when the copepod ends the jump?
8. What is the distance (in mm) in the y-direction from pipette tip to the copepod when the copepod starts the jump?

9. What is the distance (in mm) in the y-direction from pipette tip to the copepod when the copepod ends the jump?
10. Print *x* and *y* as a function of time in the same graph, and attach it to the answer sheets. Remember to write your country and team before printing.

For the following three steps, you can use the equations below the questions to calculate distances and speeds in three dimensions.

11. What is the distance (in mm) from pipette tip to the copepod when the copepod starts the jump ≈ ´Predator detection distance´?
12. What is the jump distance (in mm)?
13. What is the escape speed (in mm/s)?

Equation 1: ´Predator detection distance´

$$d_{predator} = \sqrt{(x_{start} - x_{pipette})^2 + (y_{start} - y_{pipette})^2 + (z_{start} - z_{pipette})^2}$$

Equation 2: ´Jump distance´

$$d_{jump} = \sqrt{(x_{start} - x_{end})^2 + (y_{start} - y_{end})^2 + (z_{start} - z_{end})^2}$$

Equation 3: ´Escape speed´

$$v_{escape} = \frac{d_{jump}}{\Delta t}$$

x_{start}, y_{start}, z_{start}, are respectively the x, y and z position of the copepod at the start of the jump, and x_{end}, y_{end}, z_{end}, are the x, y and z position of the copepod at the end of the jump.

Question 3.2

We know that the fish species in our aquaculture system can detect copepods at a distance of 5 mm. Furthermore, it attacks at a constant speed of 200 mm/s over a distance of 20 mm.

Can the copepods escape from an attacking fish? To answer this, assume one dimensional motion, and use the copepod escape speeds as averages for their movement. Visualize by drawing three lines on a graph for positions (on the secondary axis) of the fish and the two copepods versus time (primary axis) from the time where the copepods start jumping.

➢ *Write your answer in the answer sheet, box 3.2.*

Question 3.3

Which species is the most appropriate prey species for feeding the fish in our aquaculture system?

➢ *Write your answer in the answer sheet, box 3.3.*

Question 3.4

To which animal group do the copepods belong?

➢ *Write your answer in the answer sheet, box 3.4.*

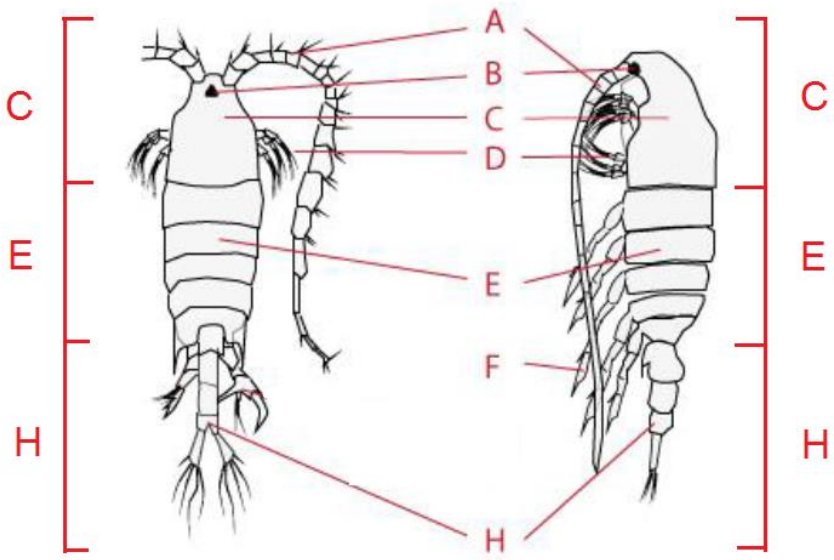


Figure 3.1. Schematic drawing of a copepod, seen from two sides.

Question 3.5

From which sides are the copepods in **Figure 3.1** depicted?

➢ *Tick the correct answers in the answer sheet, box 3.5.*

Question 3.6

Can you name the parts with the letters?

➢ *Give the relevant letters (**Figure 3.1**) to the names and write your answer in the answer sheet, box 3.6.*

Question 3.7

Many copepods contain oil. Which advantages can it give the copepods to synthesize oil?

➢ *Tick the correct answer or answers in the answer sheet, box 3.7.*

END OF TASK 2: OCEAN