The Green Gold vs. Salt

Taking the first step to sustainable desalination by using halophilic algae

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Acknowledgments

We have not done this research all by ourselves. So we want to thank the people who have helped, supported and given us advice during our research.

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Summary

This report will serve as a summary of the *technasium meesterproef* (a final project for the technasium: a subject at our school where you have to research different technical topics and design solutions for the problems), with the title *'The green gold vs. salt'*. This project takes a look at the possibility if halophilic algae can absorb sodium from water from greenhouse horticulture. This way desalination can possibly be achieved in a sustainable and biological manner. A distinctive feature of this method with algae, is that sodium will be filtered selectively. Because of this, other elements present in the water will remain and the desalinated water becomes potable and widely usable. The algae also emit oxygen instead of carbon dioxide, which makes it a long-term and environmentally friendly production method.

This report consists of the following compartments:

Cause & relevance

In this chapter the reason and the relevance of this research are panned out. Besides that, the thesis question is explained and the hypothesis is formulated.

Outline of the situation

The tomatoes on your sandwich, the flowers you buy for a birthday and the strawberries you eat during summer; all these products are produced by the greenhouse horticulture! The Dutch greenhouse horticulture affects a big market and reaches many people from a large number of countries.

Because of the current developments surrounding sustainability and circularity, the greenhouse horticulture industry has made agreements in cooperation with the government and other parties '*to work towards emission-free cultivation (i.e. no waste of nutrients and pesticides) in 2027*' (Eindrapport Emissieloos telen, 2017).

One of the specific objectives is to reach zero emission of water. In other words, to work with water in a completely circular manner and to reuse all the wastewater. This is hard to realise, because of one nutrient which builds up in the wastewater: sodium. This happens because this ion is absorbed by the plants to a lesser extent than other elements and secondly because it is difficult to filter sodium selectively. Almost all existing methods not only filter the sodium, but also other elements and substances from the water, while these other elements and substances can actually be useful for the growth of the plant. This is why it is relevant to look for a filter method which can singly remove sodium from water, and which preferably is sustainable too.

Thesis question

This research looks at the possibility to selectively filter and separate sodium-ions from greenhouse horticultural water using algae. With the goal to enable reuse of this water. The thesis question is:

Can algae selectively remove sodium from greenhouse horticultural water?

The hypothesis is that the research will show that halophilic algae are capable of doing this (scientific substantiation for this can be found in the theoretical framework).

FIGURE 1: GREENHOUSE HORTICULTURE (SOURCE: AGRO & CHEMIE)

Theoretical framework

This chapter contains the summarised theoretical background, which is used for the orientation on the thesis question.

Greenhouse horticulture

The greenhouse horticulture produces many of our daily products such as flowers, vegetables and fruits. The horticultural industry strives for usage of the most efficient systems and techniques for cultivation.

A lot of water is needed for the cultivation of crops. In substrate cultivation, the plant is rooted in a mat of rockwool, with water with nutrients being added on a regular basis (see figure 2). The excess water (drainwater) drips down out of the mat, is caught and reused. At a given moment this water is not usable anymore, so it has to get discharged.

The government and the greenhouse horticultural sector have made agreements about decreasing the amount of discharge of drainwater, aiming at the greenhouse horticulture being (close to) emission-free in 2027. The main reason why there is emission at all, is sodium accumulating in drainwater, which is bad for the crops (Glastuinbouw Waterproof, 2017).

FIGURE 2: SUBSTRATE CULTIVATION OF TOMATOES (SOURCE: FLORANEWS)

Reusing the greenhouse horticulture water means using the drainwater as irrigation water. Irrigation water is very important for a good yield, so there are some strict requirements for it. Irrigation water of good quality has a low concentration of sodium. The exact allowed sodium concentration differs per crop, but in the greenhouse horticulture they generally use a standard composition for testing (see table 1: Composition standard water). However, drainwater has a significantly higher sodium concentration than irrigation water and existing desalination techniques remain insufficient. For example, there is reverse osmosis; a very expensive desalination technique, with the biggest disadvantage being the fact that it removes almost all minerals from the water (Aquasana, 2017). This way, only dead water remains, which isn't usable anymore. In summary: sodium accumulates in greenhouse horticulture water, but existing desalination techniques are still not sufficient enough to solve this problem. Thus a zero emission isn't possible yet.

Bepaling	Eenheid	Streefcijfer	Grenswaarden
EC	mS/cm	3.0	2.5 – 3.5
pH		5.5	5 – 6
NH_4	mmol/l	0.5	0.1 – 0.5
K	mmol/l	7.0	5 – 8
Na	mmol/l	6.0	4 – 8
Ca	mmol/l	8.0	5 – 8
Mg	mmol/l	3.5	2.5 – 4.5
NO_3	mmol/l	17.0	13 – 21
Cl	mmol/l	6.0	4 – 8
SO_4	mmol/l	6.0	3.5 – 6.5
HCO_3	mmol/l	1.0	0.1 – 1.0
P (H_2PO_4)	mmol/l	0.7	0.5 – 1.5
Fe (DTPA)	µmol/l	37.5	30 – 45
Mn	µmol/l	20	15 – 25
Zn	µmol/l	5	3 – 10
B	µmol/l	50	35 – 65

TABLE 1: COMPOSITION STANDARD WATER (SOURCE: GLASTUINBOUW WATERPROOF)

Algae

'Algae' is actually a collective name for a huge group of different organisms. Species of algae have in common that they, just like plants, get their energy from photosynthesis. Algae are everywhere; in fresh and salt water. In a research was proven that there are salt tolerant algae. A halotolerant or halophilic organism is an organism which can survive and grow in extremely high salt concentrations. Some of this species of algae seem to absorb salt to keep up with the osmotic pressure of their surroundings (University of Nebraska-Lincoln, 2016). This phenomenon lays at the foundation of this research into the possibility if algae can absorb enough salt to desalinate greenhouse horticulture water.

Halotolerant organisms survive by adapting to the fluctuating concentrations of sodium in their environment. They do this via accumulation of organic compounds, called compatible solute or osmoprotectants ("*organic-osmolyte*"-strategy). If the salt concentration rises in the vicinity, the cells create osmotically active organic matter (some carbohydrates for example). This way the osmotic value in the cell increases and thus the water stays in the cell. (Wikipedia, *unknown*).

Halophiles are extremophilic organisms which live in very saline environments and which need the salt concentration to survive. Although most halophiles fall under the domain Archaea, there also are some bacterial halophiles and some eukaryote species, like the algae *Dunaliella Salina*. These halophilic organisms need sodium-chloride for growth and actually absorb the salts. They do this via accumulation of anorganic salts in their cytoplasm ("salt-in"-strategy). Their life processes, mainly their enzymes and their cellular structure (charged amino acids on their surface) are adapted to the high salt concentrations and lose their function in lowering salt concentrations. Many halophilic organisms immediately rupture because of the alteration of osmotic circumstances when placed in distilled water. They are very vulnerable to those alterations. (Wikipedia, *unknown*).

Algae offer a chance to function as a sustainable, biological desalination method for greenhouse horticultural water. After all, multiple investigations pointed to the fact that halophilic algae need sodium. For this research the following algae will be used for the experiment (see figure 3 from left to right: Chlorella Vulgaris, Dunaliella Salina & Scenedesmus Sp.).

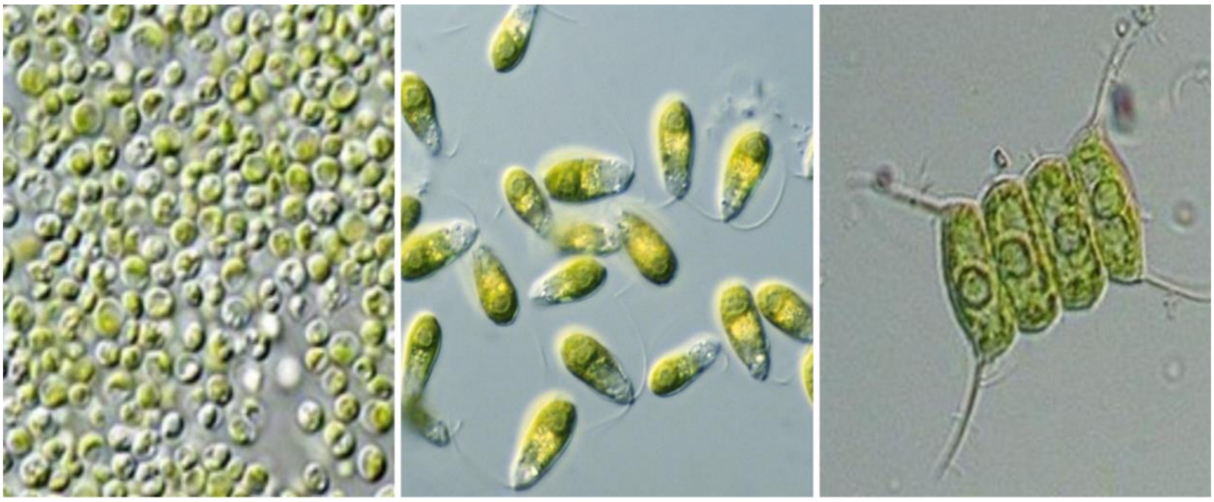


FIGURE 3: SCENDESMUS SP. (SOURCE: WIKIPEDIA), DUNALIELLA & SCENDESMUS (SOURCE: ALGAE RESEARCH & SUPPLY)

Method of research

This research consisted of a series of experiments with a communal purpose: answering the thesis question. In this chapter the way all three experiments were executed is elaborated with a step-by-stap plan. At the end there is an explanation of the method for the calculations.

The first experiment is the main experiment (*algae and sodium experiment*). Simultaneously the second experiment is carried out (*algal growth experiment*). Upon completion of the main experiment, the third and last experiment is performed (*microscopic research*). These experiments were carried out over a period of five weeks starting January 10th of 2022.

Algae and sodium experiment

The main experiment of this research was worked out as follows:

Goal of the research: determining the amount of sodium every algae species can absorb from the greenhouse horticultural water with a specific molar concentration sodium.

Necessities:

- Algae species (available online in 50 ml solution):
  - 1st Algae species: Scenedesmus Sp.
  - 2nd Algae species: Chlorella Vulgaris
  - 3rd Algae species: Dunaliella Salina
- Standard water for greenhouse horticulture (10 mmol Na+/L): 20 litres
- Standard water for greenhouse horticulture (20 mmol Na+/L): 20 litres
- 5-litre tanks (8 pieces)
- Aquarium pumps with air hoses (8 pieces)
- UV Lamp (4 pieces)
- Time switch
- Power strip (2 pieces)
- Light intensity meter (Arduino)
  - Measurement program for the Arduino
  - Glass jar
  - Phone lamp
  - Glass stirrer
- Sample bottles (36 pieces)

FIGURE 4: SETUP PREPARATORY PERIOD

Preparatory period:
For four weeks the algae had been located in a specially organised setup (see figure 4). The delivery from the United States was quicker than expected. The setup was as follows; the solutions with algae were put into PET bottles with one aquarium pump per bottle connected to it and a lamp illuminating the bottles. Both the lamp and the pumps were turned on 16 hours a day. The algae are halophilic, thus needing minerals and salt to survive, which is why every week 100 ml tap water was added. The Dunaliella Salina got a different solution: the F2-medium, because the supplier advised this. The Technical Teaching Assistants made this solution.

Setup for the main experiment

Firstly, at the start of the experiment, the testing setup was prepared. The setup looked as follows (figure 5): All eight tanks were provided with their own pump. Per two tanks there was a lamp hovering above them. Every species of algae was distributed between two tanks; with one tank containing standard water with a sodium concentration of 10 mmol/L and the other containing standard water with a sodium concentration of 20 mmol/L. Besides that, there also were two tanks only containing standard water (of 10 and 20 mmol Na+/L) without algae, which were used as blank/control tests.

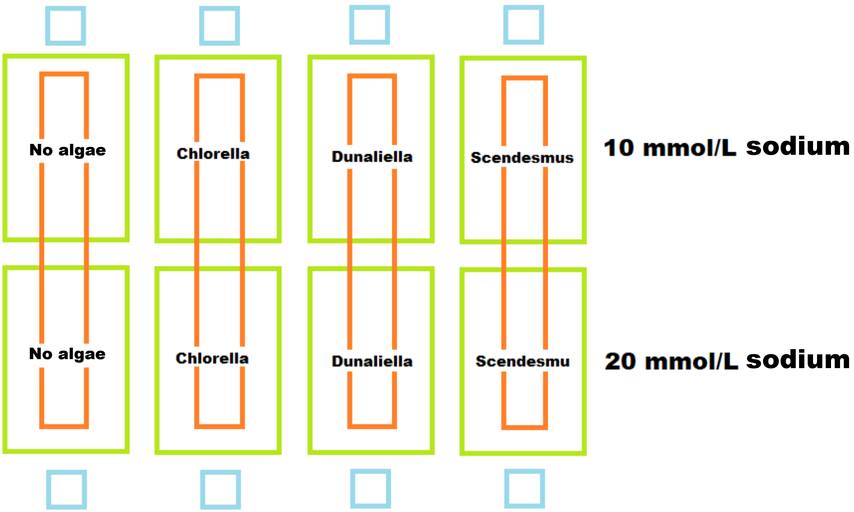


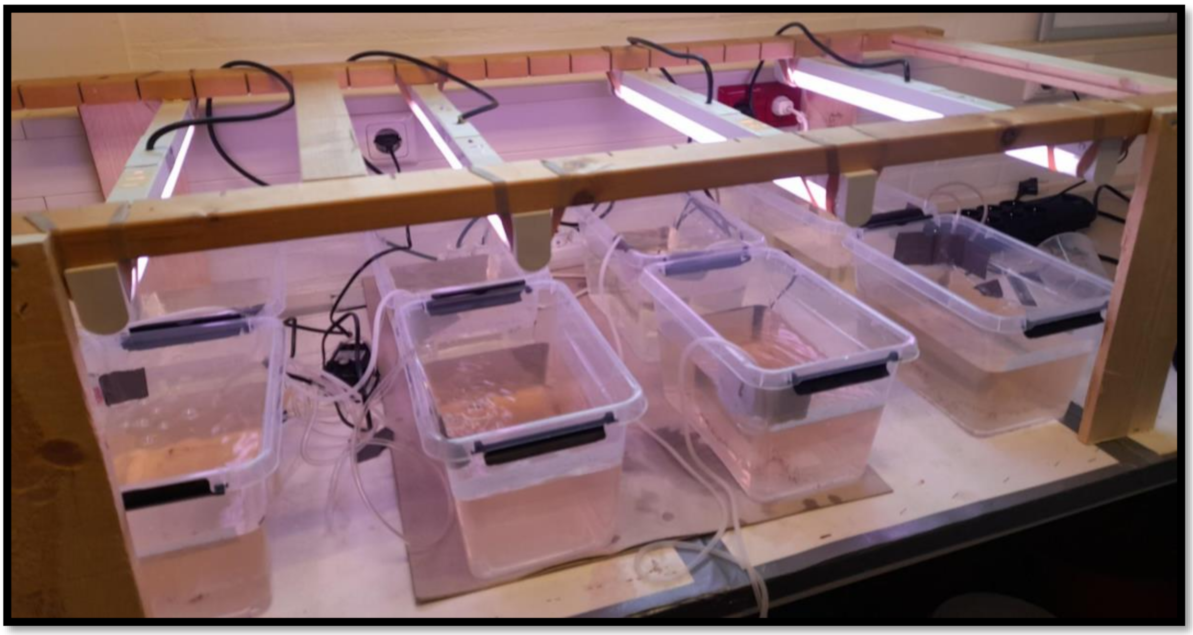
FIGURE 5: THE SETUP OF THE EXPERIMENT SCHEMATICALLY DISPLAYED;
GREEN = TANK
ORANGE = LAMP
BLUE = PUMP

During the experiment, every week a sample of 175 mL was taken from every tank (see figure 7). These samples were packaged and sent to the research lab *Eurofins Scientific*. There the concentrations of particles in the water were measured. The results of their measurements were e-mailed back and with those results conclusions for this research could be made.

The final setup looked as follows: see figure 6.

FIGURE 7: SAMPLE BOTTLE

FIGURE 6: FINAL SETUP



Algal growth experiment

This sub-study was carried out as follows:

Goal of the research: determine how much the algae grow during a period of five weeks using light transmission (translucence).

Necessities: noted in *Algae and sodium experiment*.

Setup sub-study algae growth:
For this sub-study the following preparations were done: a box was constructed with on one side two small holes (for wires from the Arduino) and on the opposite side a bigger hole (for the light source) which could cover a glass jar. Besides that an Arduino was programmed to measure the light transmission using an LDR (Light-Dependant Resistor) (see figure 8).

When the preparation was done, twice every week some water from the tanks with algae was removed using the glass jar. This jar was covered with the box. On the side the wires from the Arduino were attached and the other side was lit up for 30 seconds using a phone's flashlight. On a computer the measurement of the Arduino were displayed and stored. These measurements were triplicate measurements (repeated three times).

```
#define LDRpin A0 // pin where we connected the LDR and the resistor

int LDRValue = 0;    // result of reading the analog pin

void setup() {
 Serial.begin(9600); // sets serial port for communication
}

void loop() {
 LDRValue = analogRead(LDRpin); // read the value from the LDR
 Serial.println(LDRValue);     // print the value to the serial port
 delay(1000);              // wait a little
}
```

FIGURE 8: PROGRAM CODE ARDUINO (BRON: TWEAKERS)

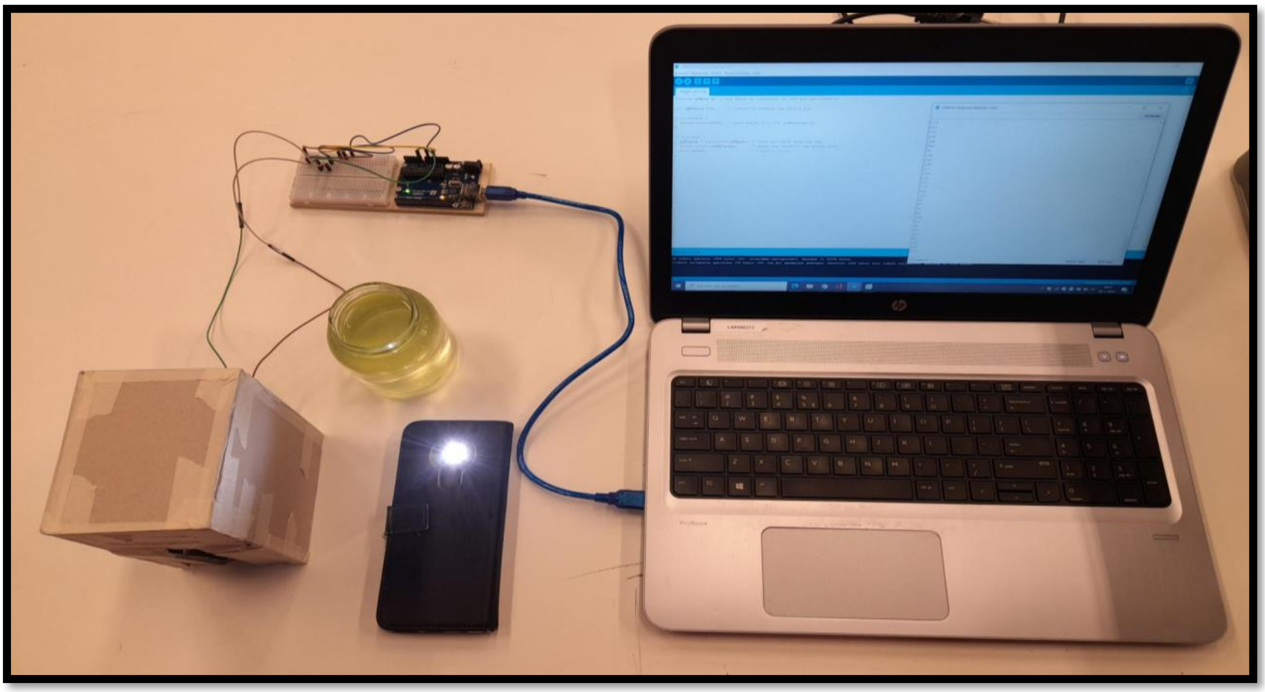


FIGURE 9: MEASURING ARRANGEMENT TRANSLUCENCE

Microscopic research

At the end of the five weeks one last experiment was carried out. This sub-study was as follows:

Goal of the research: checking if the original algae are still in the intended tanks and seeing if other algae may have entered the tanks using microscopic research.

Necessities

- Microscope
- Slides (minimum 24 pieces)
- Cover slips (minimum 24 pieces)
- Pipette
- Distilled water
- Needle-shaped object (for pressing cover slips)

Step-by-stap plan:

Firstly, for this research all necessities were prepared. After which, samples were taken from every tank and were put under an optical microscope. Of every tank two to three samples were analysed to get a better understanding of all the (different) organisms in the tank (figure 10). Besides that, pictures were taken of the samples under the microscope (figure 11). This research was mostly empirical and therefore the acquired informatiom is so too.

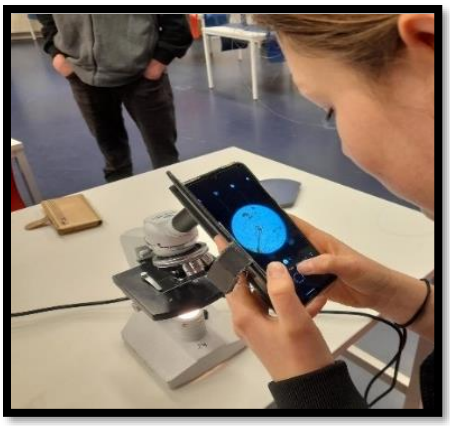


FIGURE 10: MICROSCOPIC RESEARCH IN PRACTICE

FIGURE 11: MAKING PICTURES OF THE SAMPLES THROUGH THE MICROSCOPE

Results

In this chapter the results of all three studies are elaborated by graphs and further written explanation.

Sodium intake

In the lab results a rise in sodium concentration can be seen (figure 12). This can be explained by the fact that there was a strong decrease in water volume in the tanks, because of the high intensity of evaporation. (figure 13 & 14). So, this explains the rise in sodium concentration.

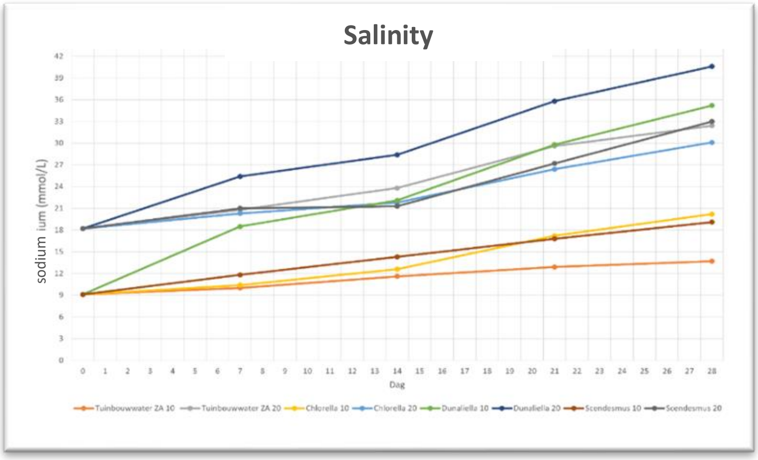


FIGURE 12: SODIUM VALUES OF ALL TANKS DURING FOUR WEEKS

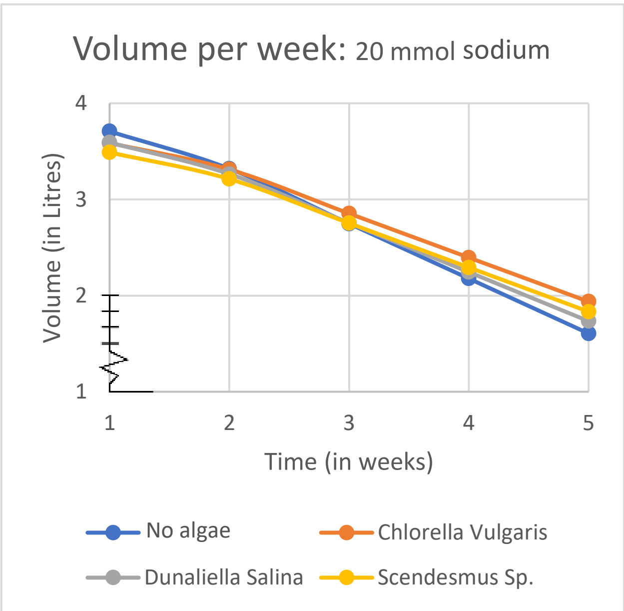


FIGURE 13: VOLUME PER WEEK (20 MMOL)

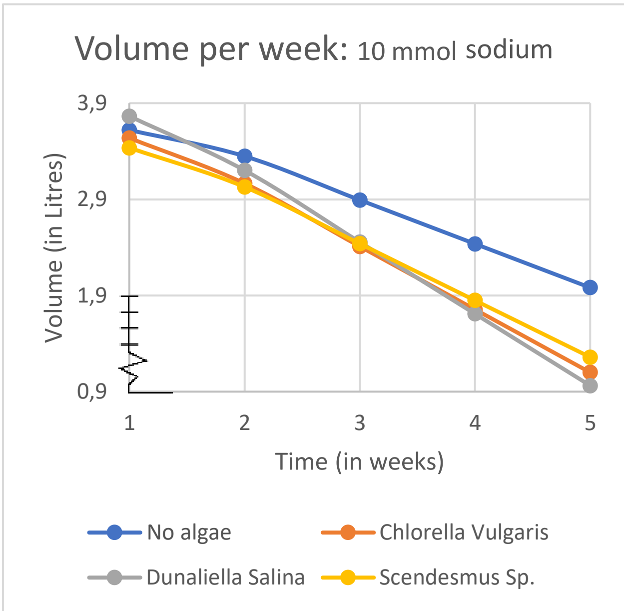


FIGURE 14: VOLUME PER WEEK (10 MMOL)

To be able to make a correct conclusion a mass balance was set up. A mass balance is based on the idea that there's always the same amount of (sodium) particles in the tanks (only taking out samples decreases the amount). This is why the theoretical sodium concentrations were calculated (the supposed amount of sodium in the water without interference). With these theoretical values, it could be determined if there was an actual decrease of sodium (because the decrease in sodium from taking samples was taken into account). If there was a decrease, this would mean that sodium was taken in by algae. The mass balance is as follows:

$$Total\ mass\ sodium = \ sodium_{in\ tank} + \ sodium_{in\ algae} + sodium_{in\ samples}$$

The mass balance showed that *Chlorella 10, Dunaliella 10, Scenedesmus 10, Horticultural water 10* & *Horticulutral water 20* had taken in sodium. With respectively (rounded) intakes of 2,34 mmol, 12,33 mmol, 3,95 mmol, 0,14 mmol & 3,45 mmol (see figure 15).

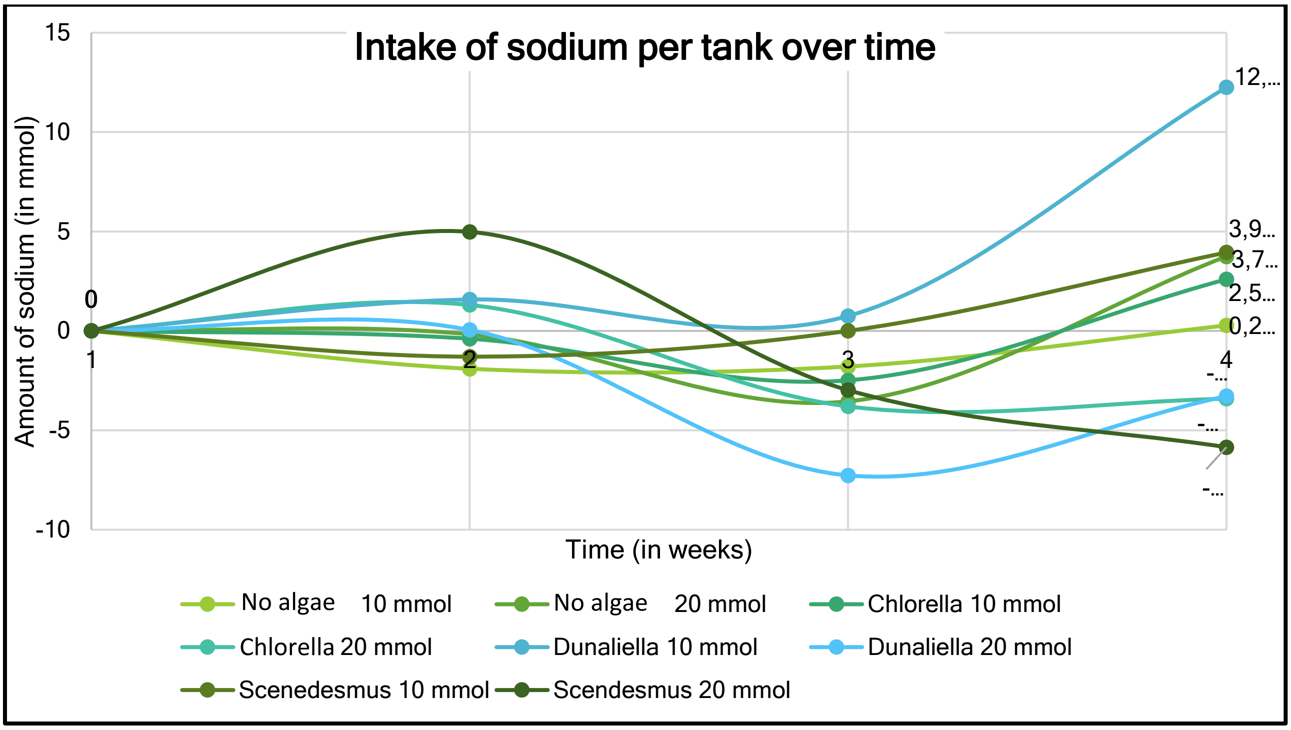


FIGURE 15: INTAKE OF SODIUM BY ALGAE OVER TIME

Based on these first results the hypothesis is confirmed. However, there is some nuance to this, because the timing of de sodium intake gives extra information to specify how the algae do it. All algae which have taken up sodium, did so from week 3 to week 4. Besides that almost all of these were 10 mmol tanks. From the results it becomes clear that the 10 mmol tanks all showed strong evaporation, with a final volume around half a litre less than the 20 mmol tanks. This indicates the concentrations of sodium being high in these tanks.

Dunaliella Salina took in the most sodium and had a concentration of 30.2 mmol/L in week 3, which rose to 47.9 mmol/L in week 4. The remaining tanks all displayed an average rise in molar concentration of 6.5 mmol/L from week 3 to week 4. The previous weeks the rise in concentration was lower. Only *Horticultural water 10 mmol* differed with an increase in concentration of 1.6 mmol/L. This tank took up the least sodium. From this correlation between the amount of sodium in the water and the intake of sodium a causal connection can be made: the more sodium in the water, the more intake of sodium by the algae.

However, the fact is that even the 20 mmol tanks took in sodium. This happened in week 2 instead of week 4. The fact that the algae of these tanks did not take in any sodium after that, can be caused by the concentration in those tanks getting too high. This indicates the presence of an optimal concentration for sodium intake. This fits with the theory from the theoretical framework. After all, processes such as osmosis are based on the differences between concentration inside and outside of the organism. When intake of sodium by algae happened, the average concentration of sodium was 26.6 mmol/L

Algal growth

The results in regards to translucence show a causal link between the amount of transmitted light and the algal growth; the less light is permitted, the more the algae have grown. This information indicates to what extent the water with a specific salinity offers a supportive and beneficial living condition for the algae (more information to be found in theoretical framework section *Algae*).

The results show that all algae-species have grown. This can be concluded form the exponential decline in translucence, which matches the exponential growth algae normally exhibit (figure 16).
The *Dunaliella Salina* showed the most growth and took in the most sodium as well. This shows the species can function properly in the saline environment. In contrast to Dunaliella Salina, all of the 20-mmol tanks (with the exception of *Horticulture water 20*) show little algal growth.

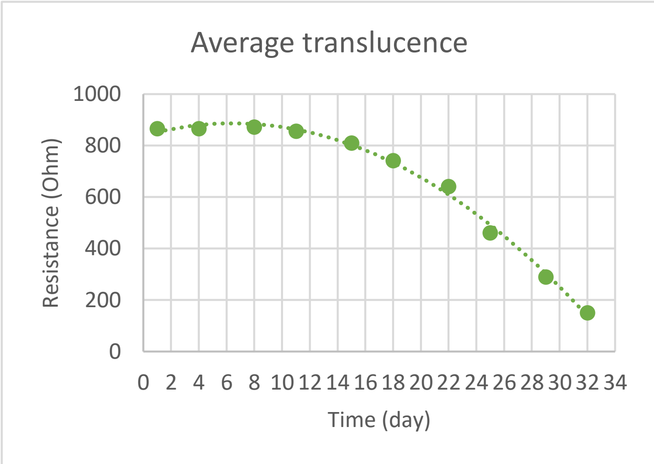


FIGURE 16: AVERAGE TRANSLUCENCE

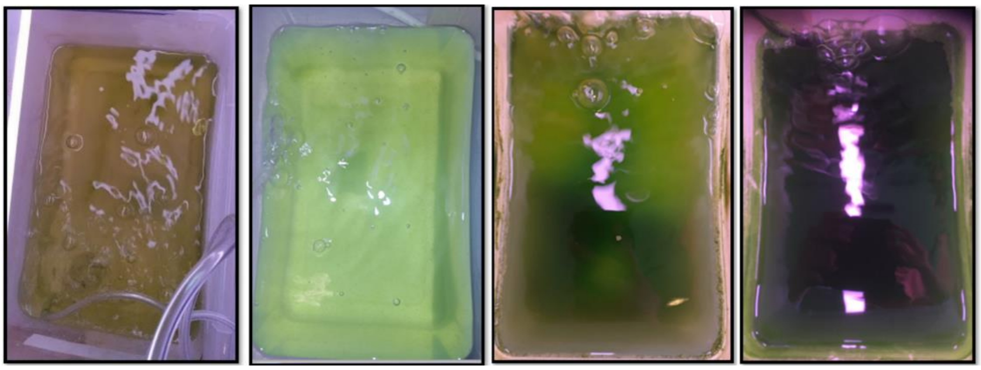


FIGURE 17: ALGAL GROWTH DURING FOUR WEEKS

In addition, the results showed that all algae grew the most during the same period they took in the most sodium.

Table 2 shows that in the period from day 22 to day 29, all the algae that took in sodium (marked yellow) show a sharp drop in translucence (minus 400-592 Ohms). This means that these algae grew a lot. A specification can be made, based on the fact that the algae that took in the most sodium (Chlorella 10 & Dunaliella 10), grew the most. A causal link could be derived from this correlation: the more algae, the more sodium is absorbed. The paragraph *Sodium intake* concludes that the concentration also plays a role. So, sodium intake most likely depends on a combination of these factors.

	NA 10 mmol	NA 20 mmol	Chl. 10 mmol	Chl. 20 mmol	Dun. 10 mmol	Dun. 20 mmol	Scen.10 mmol	Scen. 20 mmol
Day 1	873	889	870	839	878	855	866	863
Day 8	864	878	876	879	882	873	857	866
Day 15	791	826	859	823	774	810	821	782
Day 22	589	680	683	679	592	595	674	642
Day 25	421	473	427	582	186	551	592	457
Day 29	244	405	167	453	45	322	373	313
Day 32	142	183	40	254	25	153	279	133

TABLE 2 AVERAGE RESISTANCE PER TANK PER DAY (IN OHM)

Microscopic research

At the end of the five weeks of the experiment, a microscopic research was performed to see which algae were in the water in the tanks. The algae have been identified on the basis of their characteristics described in the paragraph *Algae*. This research is largely empirical and so are the results. The results of this research are classified according to the types of algae that were originally placed in the 10- and 20-mmol-sodium tanks. The pictures were taken on different microscopes and with different phones, therefore the quality and color differ.

Chlorella Vulgaris

The first thing that stands out from the results of the microscopic research is that some tanks are contaminated with different algae. This influences the conclusions that can be drawn.
With the Chlorella algae you can clearly see that this algae is still alone in the tank. This species has also taken in sodium. So it can be concluded that Chlorella Vulgaris is a species of algae that can absorb sodium.

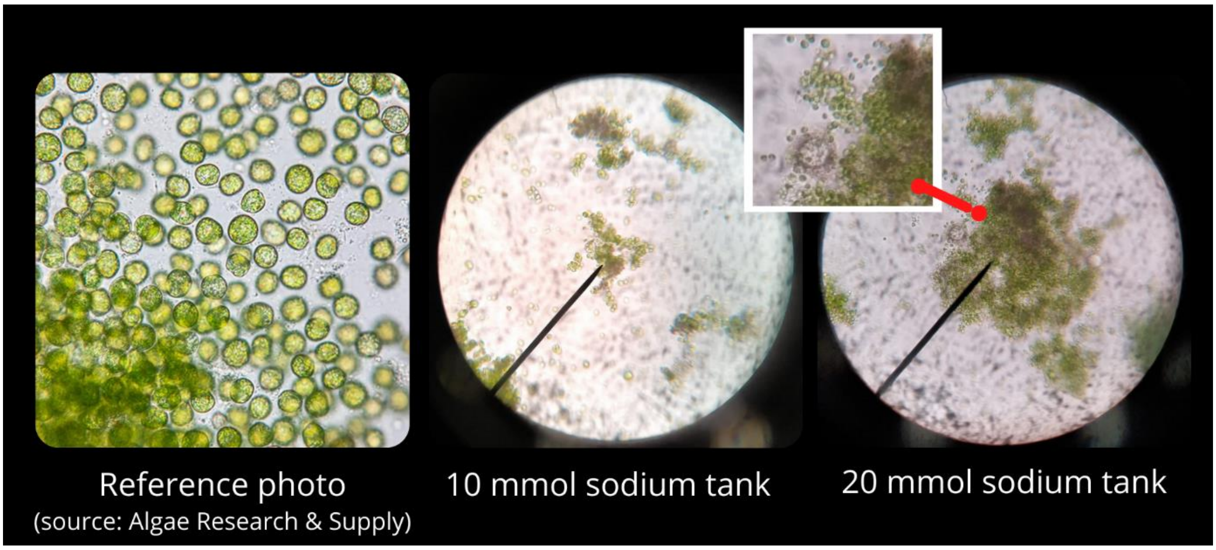


FIGURE 18 CHLORELLA VULGARIS TANKS UNDER THE MICROSCOPE

Dunaliella Salina

The Dunaliella tank was completely taken over by a different algae. This algae species seems to be Chlorella. This means, it cannot be concluded that Dunaliella has absorbed sodium, rather it seems that Chlorella has done this.

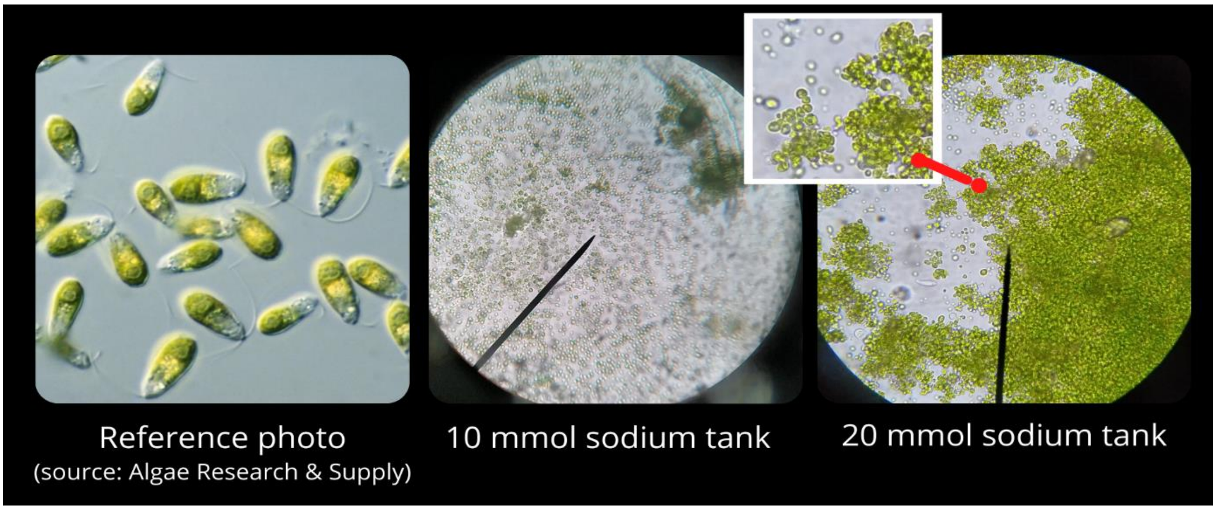


FIGURE 19 DUNALIELLA SALINA TANKS UNDER THE MICROSCOPE

What stands out, is that there are a lot of similarities between the behavioral patterns of the algae in the Dunaliella tanks and the Chlorella algae: firstly they both take up sodium simultaneously (from week 3 to week 4), secondly they both show extremely strong growth in the period from day 22 to 29 with a final resistance of 25 Ohm for Dunaliella and 40 Ohm for Chlorella. The only difference between the two is the sodium intake. Chlorella took up 2.34 mmol and Dunaliella took up 12.33 mmol. This gap can be explained by a clear difference between the two tanks, which is the sodium concentration of the water. The concentration in the tank of Dunaliella in week five was 47.9 mmol/L and for Chlorella 22.6 mmol/L. This supports the claim that the concentration is an important factor for the degree of sodium absorption, as read in *Sodium absorption*. The sodium absorption in the Dunaliella tank is therefore most likely due to contamination with Chlorella.

Scenedesmus Sp.

Scenedesmus Sp. was present in the destined tanks, but there was also contamination from Chlorella in the tanks. The behavioral patterns of Chlorella are similar to the Scenedesmus tanks, firstly for the period in which the sodium was absorbed and secondly for the concentration at which that absorption took place. The concentration in Scenedesmus was 22.2 mmol/L and Chlorella had 22.6 mmol/L. A difference is that Scenedesmus grew less than Chlorella, but it did absorb more. It is therefore not possible to conclude that Scenedesmus was the species that absorbed sodium. It is of course possible that Chlorella and Scenedesmus either symbiotically or mutualistically lived together in that tank, but more research will have to be done to be able to conclude that.

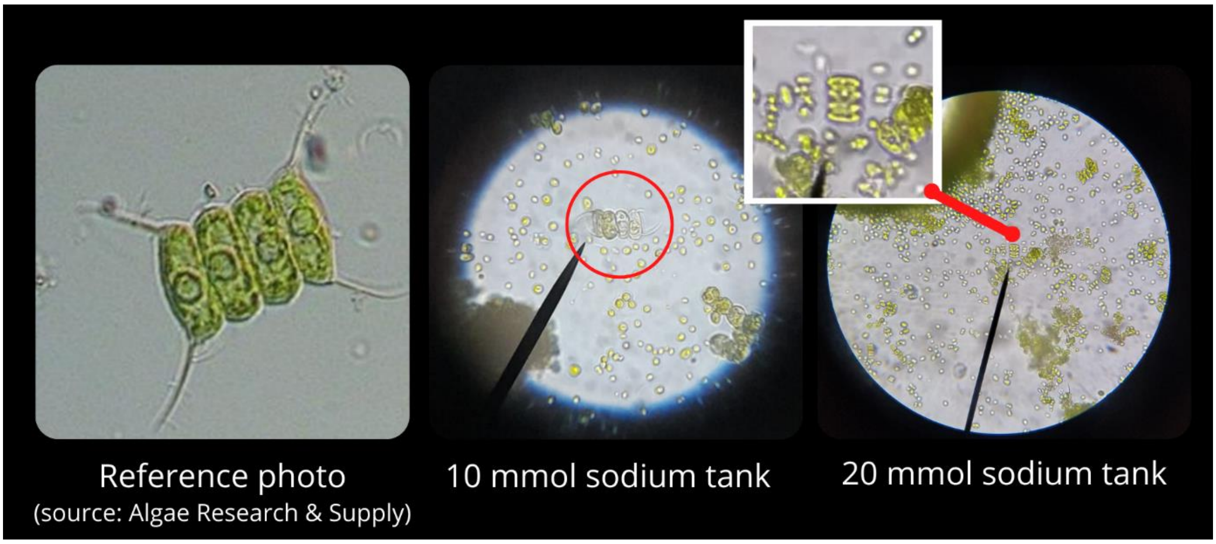


FIGURE 20 SCENEDESMUS SP. TANKS UNDER THE MICROSCOPE

Horticultural water

The tanks with just horticultural water were meant to function as blank/control tests. Yet in both tanks sodium was absorbed. This can be attested to the fact that Chlorella algae are found in both tanks. The 10-mmol tank took in 0.14 mmol sodium and 20-mmol tray 3.45 mmol. Chlorella is clearly visible in the microscopic photos of the 10-mmol tank. Many similarities can also be found between the Chlorella algae and the measurement results of the horticultural tanks. The moments of absorption are the same and the concentration of the horticultural 20-mmol tank is 34.7 mmol/L, making it the second highest concentration of all tanks. This tank therefore has the closest concentration to that of the Dunaliella tank (which had the highest concentration and also contained Chlorella vulgaris).

For the factor algal growth, it is clear to see that a lot less algae have grown in the horticultural tanks with final resistances of 244 Ohm for 10-mmol and 183 Ohm for 20-mmol. The fact that the 20-mmol tank absorbed more than the 10-mmol tank and also had a higher concentration and more algae growth supports the aforementioned causal relationships between these variables and the final sodium uptake.

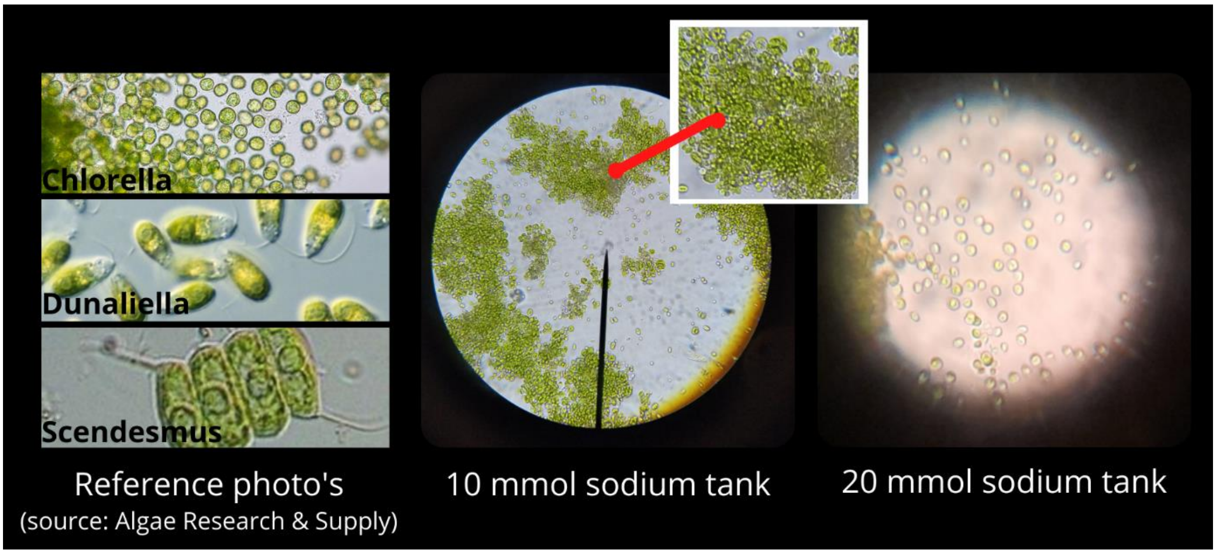


FIGURE 21 HORTICULTURAL WATER TANKS UNDER THE MICROSCOPE

Conclusion

This chapter answers the research question and examines whether the hypothesis is correct.

The answer to the research question '*Can algae selectively remove sodium from greenhouse horticultural water?*' can be divided into two parts. The answer is partly: yes, the Chlorella algae can selectively absorb sodium. This confirms the hypothesis. However, the concentration at which they do this is so much higher (26.6 mmol/L) than the concentration of standard horticultural water (max. 20 mmol/L) that it is not a suitable desalination method for greenhouse horticulture.

This conclusion can be drawn from the two causal relationships that the research results showed: 1) the higher the salinity of the water, the higher the sodium-intake by the algae & 2) the higher the amount of algae, the higher the intake. This is based on the fact that the results firstly showed that sodium was only absorbed when the salinity in the tanks increased and secondly, that most sodium was absorbed in the tanks with the highest concentrations.

Therefore it can be concluded that at higher salinities the Chlorella Vulgaris algae can absorb sodium.

In other words; this green gold defeats salt!

Discussion & recommendations

This chapter discusses the validity and reliability of the research and recommendations are made for possible follow-up research.

For this research there are four influential factors distinguishable:

1. Start-up situation
   The following adjustments can be done to improve the start-up situation and with that the validity and reliability of future research:
   - The first recommendation is to expect that delivery from the United States from the company *Algae Research Supply* takes approximately one week. Based on that information, the planning can be made and the logistics for the algae care can be arranged on time.
2. Starting values
   The starting values can be specified by the following adjustments and with that the validity of future research:
   - The second recommendation is to either separate the algae completely from the water before putting the algae in the tanks or, if full separation is not possible, to take samples from the tanks after adding the algae and let those be examined by a research lab. Both are possible, but the second option is probably better since it results in the highest validity.
     During this research practice showed that coffee filters work well to filter algae form liquid, but it is time-consuming and not all of the algae can be removed properly from the filter (i.e. it will leave residue).
   - The third recommendation to improve the starting measurements is to use scientifically approved measuring cups or to weigh the amount water. As a result, the chance of getting measurement uncertainties is small(er) and the validity of the research improves.
3. Measurement setup
   The following adjustments can be done to improve the measurement setup and with that the validity of future research:
   - The fourth recommendation is to take into account that there is a high likelihood that contamination will take place and to try to create a start-up situation that prevents this. What appears from the results/conclusion of this research is that the tanks quickly become contaminated with unwanted and/or unknown algae, so for a follow-up research it should be attempted to create a sterile environment. This can be done by adjusting the measurement set-up:
     1) by placing the tanks further apart or
     2) by closing the tanks completely from the outside air (photobioreactor).
     These adjustments reduce the chance that the bins will be contaminated by splashes from the air-pumps.
   - The fifth recommendation is to prevent unwanted evaporation by closing the tank off with a lid. Since for algae growth circulation by pump is necessary and a pump can cause high evaporation. When using a lid, sufficient air supply can still be realized by using separate tubes for air and CO_2 (comparable to a photobioreactor).
4. Microscopic research
   - The sixth recommendation is to use a special microscope camera when taking pictures of the images on the microscope. This is more convenient and faster in a practical sense, but also gives the research higher validity.

Future prospects & opportunity's

Now that the results showed that halophilic algae can absorb sodium, it becomes interesting to look at the future prospects for this desalination method. The conclusion states that it is not a suitable method for greenhouse horticulture due to the fact that the Chlorella algae only start absorbing sodium at a higher salinity. So, with these facts in mind; what else can this desalination method be used for?

A high demand for innovative and sustainable desalination methods…

The United Nations' *World Meteorological Organization* (WMO) has been warning the world for a global water crisis in their reports for a while. Mostly due to climate change, but also due to population growth; less and less people have access to enough clean and drinkable water (NU.nl, 2021).

71% of the earth's surface is made out of water, so it may sound strange that we are running short. The problem is that the freshwater supply that humans can use only makes up 2.5% of all the water on earth. Saltwater, on the other hand, makes up 97.5% (Lenntech, unknown). This water cannot yet be used to solve the water shortage because of one thing: the high sodium concentration. Existing desalination techniques are not yet optimal, are too expensive and consume a lot of fossil fuels (can be read in Theoretical Framework). They cause damage to the environment and the desalinated water is often 'dead' water (i.e. no elements and minerals in the water, making it unusable for drinking or for horticulture).

This leaves a lot of room for innovation. Halophilic algae can offer a promising, alternative, biological desalination method, since seawater has a much higher sodium concentration than greenhouse water and this research shows that the Chlorella algae absorb sodium at high salt concentrations.

On first hand there are two possible practical applications for algae as a desalination method:

Start small, go big: biomimicry research with algae as the inspiration for a new and effective desalination method…

The first opportunity is to analyze halophilic algae through the lens of biomimicry. Biomimicry aims to translate evolutionary solutions from nature into practical applications for humans. In this situation, that would mean looking at how the algae absorb the sodium and artificially imitating this mechanism. This mechanism could be developed and then applied on a larger, more industrial scale.

FIGURE 22: EXAMPLE OF BIOMIMICRY: BEAK OF A BIRD AS THE INSPIRATION FOR THE STREAMLINED FRONT OF A EXPRESS TRAIN (SOURCE: THE SUSTAINABLE MAG)

Going green, going clean: a large scale and circular desalination process with oxygen-emitting algae to counter climate change…

A second practical application is to start using algae in a similar way as the Dutch Water Authorities do with aerobic bacteria. Their biological water-purification process works as follows: first the waste water is collected. This water, along with oxygen, is pumped into a tank. This tank contains aerobic bacteria. The water is pumped around and the bacteria eat the waste. When this has been done sufficiently, the bacterial water flows into a settler: here the bacteria sink to the bottom. The purified water flows through to its destination and the bacterial remains (sludge) go to a fermenter where it is made into biogas. The formed biogas is used to supply the whole process which makes it circular. The excess biogas supplies nearby households (Union of Water Boards, 2013). This process is shown in figure 23.

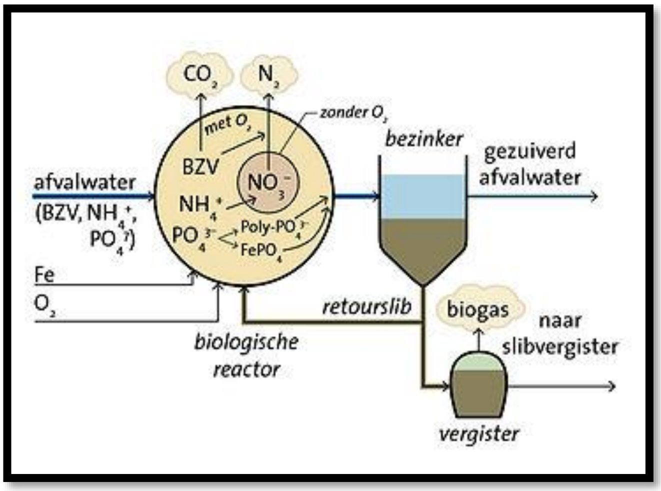


FIGURE 23: BIOLOGICAL WATER TREATMENT BY DUTCH WATER AUTHORITIES (BRON: NEMO KENNISLINK - SITTROP GRAFISCH REALISATIE BUREAU)

The fact that this biological purification process with bacteria is already being done successfully on a large scale offers perspective for a similar process with algae desalinating seawater. After all, algae and bacteria are both natural products and can absorb substances selectively: bacteria can absorb nitrogen and phosphorus and algae can absorb sodium, according to this research. In addition, both organisms grow exponentially, which makes them easier to attain in large quantities and both sink when there is a lack of circulation, which makes the first step of separating the algae from the water easier (see figure 24). One difference between algae and bacteria is that algae make oxygen and bacteria carbon dioxide. This immediately makes algae more environmentally friendly and sustainable.

FIGURE 24 ALGAE- PRECIPITATION AFTER ONE DAY WITHOUT PUMPP

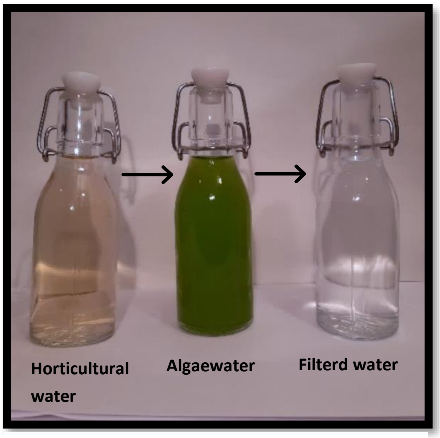


FIGURE 25 DESALINATION PROCES

So, a similar facility with algae could be possible. In this case, the wastewater is replaced with seawater. The desalinated seawater must be useable. There is a high probability that an an extra purification step will have to be added after the settler, as the requirements are very strict for water quality. If follow-up studies show that full desalination is not possible with algae alone, then algae can still serve as a pretreatment for other technologies. As a result, the ecological footprint and financial costs of desalination are reduced and the process becomes more sustainable.

All things considered, using algae as a saltwater desalination method has a lot of potential. On three aspects algae are beneficial over other existing desalination methods:

- *Ecologically beneficial*: algae produce oxygen and consume CO_2. This is a distinctive characteristic, as most filtering methods use fossil fuels and emit CO_2. Thus, this method has a **small ecological footprint**. This is a positive aspect for the long term, because legislation regarding emissions are becoming stricter, with the ultimate goal of achieving net zero. Algae as a biological desalination method therefore fit well within the ideology of the future.

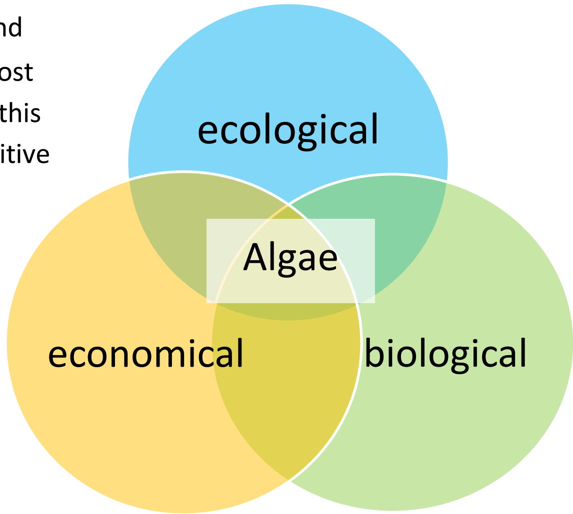


FIGURE 26 BENEFICIAL ASPECTS OF ALGAE AS A DESALINATION METHOD

- *Economically beneficial*: halophilic algae grow exponentially when consuming lots of sodium. This results in the eventual forming of algae-residue/waste (just like the bacterial sludge at the Dutch Water Authorities). This surplus does not have to function as waste, because the algae-residue can serve as biomass for food supplements and/or biodiesel.
  - Firstly, the algae residue can be used for food. The Chlorella algae are already used as a **dietary supplement**. The Information Center for Nutritional Supplements & Health says: "*Chlorella is used as a source of vitamins, especially vitamins A, B and K and minerals such as iron, zinc and iodine and essential fatty acids and proteins. Chlorella can be a good addition, especially for vegetarians*." Some of the algae residue can therefore be sold as a dietary supplement and with a declining meat consumption and a growing number of people becoming vegetarian, the market prospects for this are positive (NOS, 2020).
  - Secondly, the biomass can be converted into biodiesel. The machines can be run on the diesel. This makes it possible to realize a **circular process with biodiesel**. This is financially beneficial and enviromentally friendly.

- *Biologically beneficial*: a desalination method with algae is a method for and by nature. It is a sustainable method that contributes to a future with a clean, livable and healthy earth.

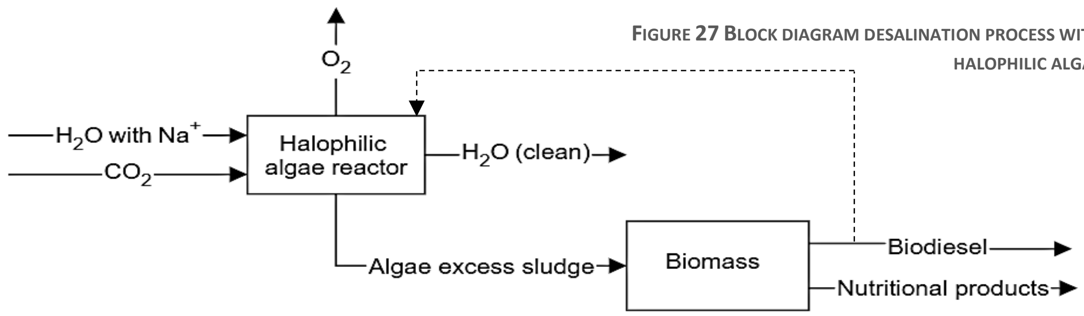


FIGURE 27 BLOCK DIAGRAM DESALINATION PROCESS WITH HALOPHILIC ALGAE

All in all, a saltwater desalination method with algae looks promising, because it is ecologically, economically and biologically responsible. It is a method that fits the prospect of the future.

So when you start working with algae, you are truly sitting on a green-goldmine!

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