Algae production pilot open ponds Lelystad

Results 2013 - 2015

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Summary

In 2012 two open microalgae ponds (one indoor and one outdoor, both 250 m^2) were built at the ACRRES pilot site in Lelystad. Both ponds are connected to an anaerobic digester and utilise excess heat and flue gas (CO_2) from the Combined Heat and Power unit (CHP). In this report the results of the algae production monitoring and the additional experiments are given for the period 2013-2015.

In 2013 for both ponds the annual production was monitored. In 2014 and 2015 monitoring was only done for the outdoor pond while the indoor pond was used for experiments in order to test adjustments of the system. Initially, in 2013 both ponds were harvested with a combination of the coalescer, a sedimentation tank, and the centrifuge. In July 2013 the harvesting system of the outdoor pond was changed to direct centrifugation of the algae culture as the harvest efficiency of the coalescer system was too low.

The annual biomass production of the outdoor pond was 90, 175 and 140 kg in 2013, 2014 and 2015, respectively, corresponding with 3.6, 7.0 and 5.6 ton dry matter per ha. The lower production in 2013 is due to the fact that in the first half of the year the pond was harvested with the coalescer. Although the change of the harvest system improved the production the annual production as realized in 2014 and 2015 was relatively low (5.5 – 7 ton dry matter per ha). Model calculations predicted an annual production of about 17 ton dry matter per ha. This yield gap is mainly due to periods with low or no production after crashes. Additionally, suboptimal algae growth may also be caused by suboptimal CO_2 availability and a suboptimal harvest efficiency. In periods with stable production the harvested biomass yield corresponded with annual production levels of 11-17 ton dry matter per ha.

In 2014 and 2015 alternative harvest systems were tested: dissolved air flotation (DAF) and sedimentation both in combination with flocculants. For the DAF unit best results were obtained for the feed grade flocculant BC floc, a polyacrylamide cationic polymer. Harvest efficiency was > 90% for a flocculant dose of 1-4 g/m^3 algae water. Unless the flocculant is feed grade, a drawback of the system is the contamination of the harvested biomass by the flocculant. Furthermore, when the effluent of the DAF unit is returned to the pond, in some tests problems arose with flocculation of algae in the pond. Sedimentation in combination with an increased pH level (10-11) and the addition of a flocculant also resulted in a harvest efficiency > 90%.

Currently, the CO_2 addition in the ponds is done by injection of flue gas in the continuous airstream that is sparged into the pond via perforated tubes at the bottom of the pond. Measurements of the CO_2 concentration in the bubbles leaving the pond indicate that the recovery of CO_2 in the water was relatively low and dissolved CO_2-concentration in the pond was fluctuating. Restricting the air sparging to periods when flue gas was added, resulted in a more stable CO_2-concentration in the pond and a 65-80% decreased energy demand for air sparging without affecting the algae growth.

Nutrient supply of the ponds is done with chemical fertilisers, but preferably side streams should be used, like digestate from the digester. A test with addition of liquid fraction of the digestate from the Acrres pilot site (0.2 m^3 added on a total pond volume of 100 m^3) showed that although the algae growth seemed not to be negatively affected, the culture got more coloured and the harvested biomass was contaminated with the organic matter that was present in the added liquid fraction. Therefore, further tests are necessary with manure products lower in organic solids.

Calculations on energy demand based on operation time and electric power of the devices showed that when harvest was done by centrifuge, the total energy demand of the system is 1.7 times higher than for harvest by DAF unit. The energy demand for the harvesting devices (pumps, centrifuge, DAF unit) is about 4 times higher. The contribution of the energy demand for harvesting to the total energy demand

of the algae system is about 55% for harvest by centrifuge and 25% for harvest by DAF unit.
When air sparging is restricted to periods with CO_2 and harvest efficiency is 90%, the energy demand of the total system is decreased with 20 and 35 % when harvesting was done by centrifuge and DAF unit, respectively.

1 Introduction

In 2012 two open microalgae ponds (one indoor and one outdoor) were built at the ACRRES pilot site in Lelystad. Both ponds are connected to an anaerobic digester and utilize excess heat and flue gas (CO_2) from the Combined Heat and Power unit (CHP) unit that burns the biogas produced in the anaerobic co-digesters in order to produce electricity (Figure 1). The use of digestate as nutrient source for the microalgae is not realized yet. First, more research is required with regard to pretreatment of the digestate as the dark colour and the presence of solids interact negatively with the algae production. Till now nutrient supply is done with chemical fertilisers.

Besides the two open ponds also two open LED light assisted pre-culture basins were built. The basins are mainly used for upscaling the inoculum for the open ponds.

Both the open ponds as well as the LED basins are providing data for algal growth under different conditions.

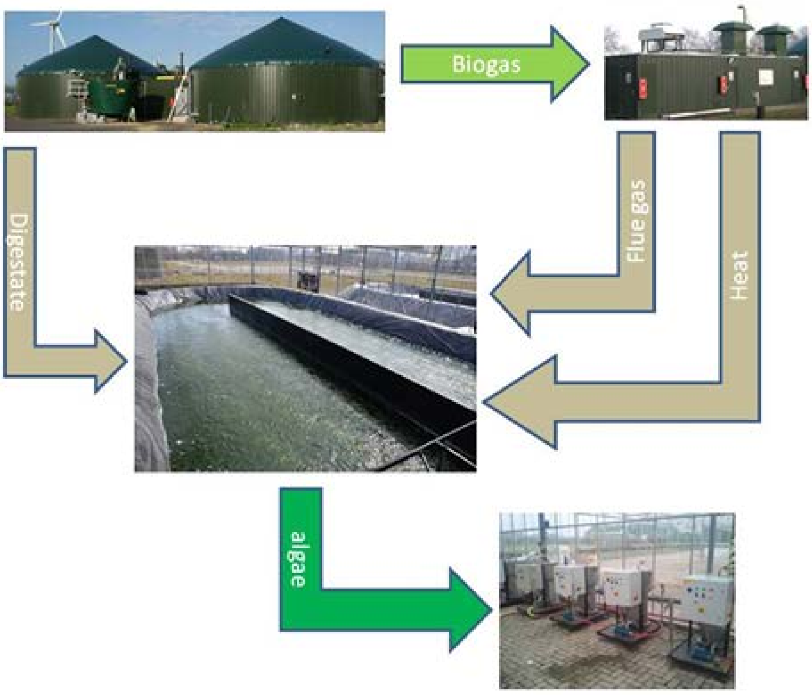

Figure 1. The open pond system connected to the biodigester plant at the ACRRES pilot site in Lelystad (the use of digestate as nutrient source is not realized yet).

Both ponds are constructed with an earth wall that is covered with black plastic (polypropylene) (Figure 2). The pond allows for a maximum water table depth of 100 cm. The average water area of both ponds is 250 m^2. The water in the ponds is stirred with a propeller mixer. Infrastructure was laid down to transport the flue gas and the cooling water from the CHP to the open ponds. Air or flue gas is sparged into the culture via perforated tubes at the bottom of the pond. The flue gas addition is regulated based on the pH level of the ponds. The water in the ponds is heated with the cooling water of the CHP that is pumped through tubes at the bottom of each of the ponds. A more detailed description of the both pond systems is given in paragraph 2.1.

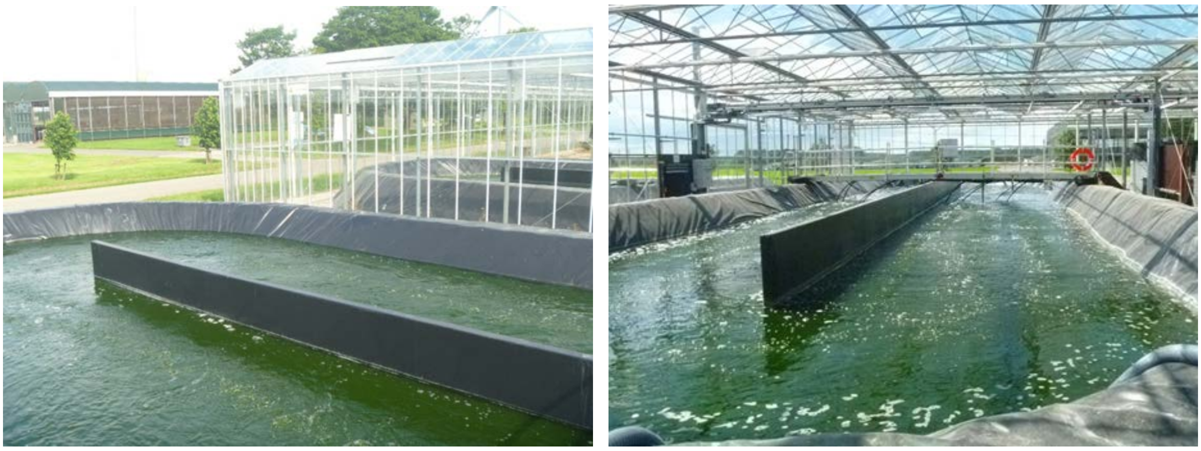

Figure 2. The outdoor and indoor open micro-algae pond at the ACRRES pilot site in Lelystad (The Netherlands).

The monitoring and supporting research with regard to the algae ponds is funded by the Interreg project Energetic Algae (EnAlgae) and the PPP project Small Scale Biorefinery.

Within the PPP Small Scale Biorefinery in Work Package 5, different companies work together to enhance the cultivation and refinery of aquatic biomass including microalgae. It includes applied research on optimizing microalgae cultivation on a practical scale, with special attention to growth factors ((LED) light, nutrients (amounts and timing), temperature, harvest rate and product quality. From various algae cultivation pilots indicators are collected for benchmarking and exploring of the economic perspectives.

The Enalgae project is aimed at the development of sustainable technologies for algal biomass production for bioenergy and greenhouse gas (GHG) mitigation. This is achieved by developing and sharing nine pilot-scale facilities across North Western Europe. The algae pilot site in Lelystad is one of them. The combined experience built up on these facilities will provide a basis for the best practice for the development and operation of commercial scale algae production. Economic models have been developed for three common types of algae reactors: open ponds, tubal reactors and flat panel airlift reactors.

This report focusses on the algae production in the open pond systems in Lelystad and describes the results obtained in 2013-2015. In chapter 2 the results are summarized while in chapter 3-5 the results of 2013, 2014 and 2015 are given, respectively.

2 Summary results 2013-2015

2.1 Biomass production

In 2013 the indoor as well as the outdoor pond were used to monitor the algae biomass production while in 2014 and 2015 the continuous yield monitoring was only done for the outdoor pond whereas the indoor pond was used for additional experiments to optimise the pond system.

Initially, in 2013 both ponds were harvested with a combination of the coalescer and the centrifuge. In the coalescer, a sedimentation tank, a preconcentration was realized. Subsequently, the collected slurry of the coalescer (about 1 m^3/day) was centrifuged to get an algae paste of 10-15% dry matter. The centrifuge water was discharged to the sewage system. In July 2013 the harvesting system of the outdoor pond was changed to direct centrifugation of the algae culture because tests had shown that the coalescer harvesting system limited the productivity of the ponds due to a too low biomass recovery. Extra centrifuge capacity was installed to be able to centrifuge sufficiently large volumes of algae culture. To minimize the amount of waste water the effluent from the centrifuges was recycled to the algae culture, as was the case with the coalescer system. For both ponds harvesting was only done on working days.

In Table 1 the dry matter production is summarized. The annual biomass production of the outdoor pond ranged from 90 to 175 kg corresponding with 3.6 to 7.0 ton dry matter per ha. The lower production in 2013 is due to the fact that in the first half of the year the pond was harvested with the coalescer. In this period 33 kg dry matter was harvested. After the change to direct centrifugation the productivity increased and 57 kg per ha was harvested.

In the first half of 2013 both ponds were harvested with the same harvest system, a combination of the coalescer and the centrifuge. In this period the harvest was 42 and 33 kg dry matter for the indoor and outdoor pond, respectively. This difference is mainly due to periods with low or no production of the outdoor pond caused by a precautionary shut down of the harvesting system in a frost period (week 3-4) and a period with two culture crashes (week 20-26). In periods without problems the biomass production of the outdoor pond was slightly higher than for the indoor pond: in week 1-19 (excluding the frost period in January) the cumulative dry matter production was 16 and 19 kg for the indoor and outdoor pond, respectively.

Table 1. Annual dry matter yield of the open ponds in 2013, 2014 and 2015.

Year	Pond	Harvest system	Annual dry matter production	
			Kg/pond	Ton/ha
2013	Indoor	Coalescer (week 1-30)	42	1.7
		Coalescer (week 31-52)	20	0.8
		Total	*62*	*2.5*
	Outdoor	Coalescer (week 1-30)	33	1.3
		Centrifuge (week 31-52)	57	2.3
		Total	*90*	*3.6*
2014	Outdoor	Centrifuge	176	7.0
2015	Outdoor	Centrifuge	141	5.6

Although the change of the harvest system improved the production the annual production as realized in 2014 and 2015 was relatively low (5.5 – 7 ton dry matter per ha). With the model AlgaeEconomics based on the actual radiation and temperature an annual production of about 17 ton dry matter per ha was predicted. This yield gap is mainly due to periods with low or no production after crashes mostly characterized by a browning of the culture. Most problems occurred in the summer time and may be caused by the presence of predators as highest populations were observed in that period. In order to control the predator populations a 75 µm drum filter was installed with a flow of 3 m^3/h. However, based on predator counts of the infow and outflow of the filter, it became clear that the effectiveness of the filter was low. Decreasing the pore size did not show out to be an option to improve the effectiveness as the water use of the filter due to backflushing increased strongly and microalgae were also filtered from the culture.
Besides high predator populations suboptimal algae growth may also be caused by suboptimal CO_2 availability and a suboptimal harvest efficiency. Both factors are discussed later on.
In periods with stable production the harvested biomass yield corresponded with annual production levels of 11-17 ton dry matter per ha.

2.2 Harvest system

A significant part of the costs for microalgae production can be ascribed to the harvest. Currently, harvesting is mostly done by centrifugation being the most reliable harvesting method, but energy demand is relatively high. Therefore, in the algae pilot also alternative harvest systems were tested like dissolved air flotation and sedimentation both in combination with flocculants. Additionally, tests were done on the harvest efficiency of the centrifuges of the pilot.

Dissolved air flotation

In 2013 a harvesting installation based on dissolved air flotation (DAF) was installed. This system is based on flocculation of the algae cells stimulated by the addition of a flocculant. After mixing the algae culture with the flocculant the mixture flows through a pressure vessel in which the mixture is aerated and put under pressure (5-6 bar). In the flotation tank the pressure is released and micro air bubbles are created causing algae flocs to float to the surface where a skimmer collects the concentrated algae slurry. The effluent water of the DAF unit is returned to the pond.

For an efficient harvest a flocculant is needed. The flocculant should be feed grade as a main destination of the algae is animal feed and recycling of the effluent water should be possible in order to reduce the water demand. After first lab tests two flocculants were selected for larger scale tests in the DAF unit: BC Floc and Greenfloc 120. BC Floc is a cationic polyacrylamide and is feed grade certified, but is not easily biologically degraded. Greenfloc 120 is a cationic starch based flocculant and is expected to degrade faster, but has no feed grade certification.

In 2014 tests were done with the two flocculants. The best results were found for BC floc. At a relatively low dose (1 g/m^3) 90-95% of the algae were removed. For Greenfloc 120 the maximum algae recovery, 70-80%, was lower than for BC floc while the dose to reach this recovery was considerably higher (40 g/m^3) than for BC floc. Moreover, the variation in achieved algae recovery was higher for Greenfloc than for BC floc. A second test with BC floc in 2015 showed that a higher dose (4 g/m^3) was needed to realize an algae recovery > 90% while at a dose of 1-2 mg/m^3 the algae recovery was 50-60%. Apparently, the species composition or the condition of the algae culture affect the flocculation.

In the DAF tests with BC floc for a dose of 1 g/m^3 the effects of returning the effluent to the pond was tested. In the test in 2014 after three days flocculation in the algae pond was observed, a possible indication that the remains of the flocculant were affecting the algae culture. However, also in the weeks before the culture showed some autoflocculation. In the second test in 2015 no disrupting effect of returning the effluent was observed.

In 2016 additional experiments with the DAF unit will be done using ozone as a flocculant. In literature positive effects are mentioned and, unlike the use of cationic polymer flocculants, contamination of the algae biomass and the algae culture in the pond will not occur.

Sedimentation

Initially, the ponds were harvested with the coalescer based on sedimentation. As mentioned before it turned out that harvest efficiency was too low. However, sedimentation may be improved by an increased pH and/or the addition of a flocculant. In 2015 test were done with the culture of the indoor pond.

Firstly, in lab scale experiments the harvest efficiency was assessed in relation to the culture pH and the addition of the flocculant BC floc. Increasing the pH improved the harvest efficiency from 25% (no pH increase) to about 70% at a pH of 12. Adding a flocculant improved the harvest efficiency further especially al lower pH levels.
Additionally, larger scale tests were done with a settler tank of 360 L. The tests were done at a pH of 11. At the lowest flocculant dose (1 mg BC floc/l) the harvest efficiency was 35-45%. Increasing the dose to 2 and 4 mg BC floc/l increased the efficiency to 50-70% and 85%, respectively.

The results show that increasing the pH improved the harvest efficiency. However, a relatively high pH (11-12) is necessary to realize a high harvest efficiency. This means that high amounts of caustic soda are necessary while after the harvest acid is necessary to decrease the pH to a level of about7- 8. A part of the required pH decrease is expected to be realized by the CO_2 sparging of the pond.

Centrifuges pilot

In 2015 the outdoor pond was harvested with three manually unloaded centrifuges (hereafter indicated as MAN, unloading was first done once a day, runtime was 24 h/day and flow rate was 750 L/h) and from September onwards a self-unloading centrifuge (hereafter indicated as AUT, runtime 24 h/day, flow rate 500 L/h) was added.

For the MAN centrifuges a test was done to assess the harvest efficiency as affected by frequency of unloading (once or twice a day) and flow rate of the algae water (350 and 750 L/hour). When unloaded once a day the harvest efficiency decreased from 40-55% (based on decrease of optical density) just after unloading to zero or even negative values after 24 hours of operation indicating that the already harvested biomass was beginning to wash out. When unloaded twice a day this washing out of biomass was not observed. Decreasing the flow of the centrifuge increased the harvest efficiency with about 10-20% (absolute). The improved harvest efficiency could, however, not compensate for the lower flow rate (50%).

Based on these results it was decided to unload twice a day for certain periods and that for a situation that unloading was done once a day, starting up was done at the end of the afternoon instead of directly after the harvest in the morning resulting in an operation period of about 16 hours instead of 24 hours.

In the period the AUT centrifuge was in operation a comparison with the MAN centrifuges could be made. The harvested algae biomass was 1.5 and 2.2 kg dry matter/week for the MAN and AUT centrifuge, respectively. For the MAN centrifuges an algae paste of 19% dry matter was harvested while the AUT centrifuge produced a slurry of 2%. In a commercial situation an additional dewatering step (e.g. centrifuge) is expected to be necessary in order to produce a paste of 15-20%.

2.3 CO2 addition

Currently, the CO_2 addition in the ponds is done by injection of flue gas in the continuous airstream that is sparged into the pond via perforated tubes at the bottom of the pond. The flue gas addition is regulated based on the pH level of the pond. In order to assess the efficiency of the CO_2 addition measurements were done of the CO_2 concentration in the air bubbles that leave the pond and the CO_2 concentration of the pond culture.

Measurements were first done in the common situation of continuous air sparging and flue gas addition when the pH was above the setpoint of pH 8. The high CO_2 concentration in the bubbles leaving the pond indicate that the recovery of CO_2 in the water was relatively low. At a depth of 40 cm the CO_2-concentration was higher than at a depth of 60 and 80 cm. This may be an indication that the CO_2-recovery was better at a higher depth due to a longer contact of bubbles and water. The low recovery is expected to be due to the relatively big air bubbles that are created resulting in a low gas exchange contact area with the water.
The dissolved CO_2 concentration in the water fluctuated and sometimes (but not always) corresponded with the period of CO_2-production (higher in night time) and CO_2-consumption (lower in day time). Maximum measured values ranged from 4.5-8 mg dissolved CO_2/l.

In a second test period air sparging was restricted to the periods when flue gas was added. This resulted in an more stable CO_2-concentration in the pond of about 6 mg dissolved CO_2/l. This sparging regime decreased energy demand with about 65% when a mix of flue gas and air was applied and 80% when 100% flue gas was applied. It must be emphasized that the tests were done in October. It may be expected that in periods with higher algae production and thus higher CO_2 demand the energy saving compared to continuous air sparging will be lower.
As the reduced air sparging did not result in significant effects on the algae growth or other problems as sedimentation of algae in the pond, this intermittent sparging regime was applied for both ponds.

In 2016 further experiments will be done on alternative methods of CO_2 addition in order to improve the CO_2 recovery in the pond which is low with the currently used method.

2.4 Nutrient supply

Nutrient supply of the ponds is done with chemical fertilisers, but preferably side streams should be used, like digestate from the digester. A drawback of digestate is the dark colour and the presence of solids that decrease the light transmission in the culture. A pre-treatment by a separation step resulting in a liquid and solid fraction and using the liquid fraction for the algae production may help to decrease this problem. Earlier lab experiments showed that a 0.5-1% diluted liquid fraction did not affect algae growth significantly. At these dilution levels nutrient supply is sufficient for algae growth. In order to assess the effect of digestate use on a bigger scale in December 2014 a test was done in the outdoor pond.

About 200 L liquid fraction was added to the pond, on a total pond volume of the pond of about 10 m^3 this means a dilution of about 500x. The measured increased NH_4-N and PO_4-P-concentration in the pond corresponded with the added amount in the liquid fraction. After the addition a significant increase of the OD was observed due to the colour of the liquid fraction. A day after the addition the harvest almost doubled. This is probably due to the simultaneous harvest of organic matter of the liquid fraction. Per addition of 200 L an amount of 7 kg organic matter is added to the pond corresponding with about 70 mg/L algae culture. For comparison the algae dry matter content in the pond is estimated at 100-150 mg/L at that moment. The cell counts showed that algae growth was not affected.

Although algae growth seemed not to be negatively affected by the addition of the liquid fraction, the contamination of the algae biomass with organic matter is a problem. Therefore, further tests are necessary with manure products lower in organic solids.

2.5 Energy consumption

The energy consumption of the ponds was not measured separately. Therefore, based on electric power of the devices and operation time, the energy demand was calculated for a situation with harvest by centrifuge and harvest by dissolved air flotation (Table 2). In the first scenario (basis scenario) a common pond operation was assumed i.e. the blower, mixer and grazer filter are 24 h/day in operation. The operation time of the harvest devices (harvest pumps, centrifuges and DAF unit) depend on the amount of biomass to be removed daily and the harvest efficiency of the harvest device. For the basis calculation a daily removal of the biomass from the pond of 25% and a harvest efficiency of 90% for the centrifuge as well as DAF unit is assumed. Further assumptions are an algae concentration of the culture of 100 g/m^3 and a pond volume of 100 m^3. For the system with the DAF unit it is assumed that the harvested slurry by the DAF unit is subsequently centrifuged.

The results show that for harvesting by centrifuge the total energy demand of the system is 1.7 times higher than for harvest by DAF unit. The energy demand for harvesting for the harvest devices is about 4 times higher. The contribution of the energy demand for harvesting to the total energy demand is about 55% for harvest by centrifuge and 25% for harvest by DAF unit. When the harvest efficiency is decreased to 60% these percentages are about 65% and 30 % respectively.

Table 2 shows that the energy demand of the blower is relatively high when they are 24 h/day in operation. As mentioned before restricting the air sparging to periods with CO_2 demand did not affect the algae growth while energy demand could be decreased with 65%. This reduction decreased total energy demand with 20 and 35 % when harvesting was done by centrifuge and DAF unit, respectively.

Table 2. Energy demand (kWh/day) of the algae pilot (based on operation time and electric power of the devices) for a situation with harvest by centrifuge and harvest by dissolved air flotation. Basic assumptions: algae concentration pond: 100 g/m^3, daily biomass removal: 25%.

Scenario	Device	Harvest system	
		Centrifuges	DAF system
Common pond operation	Blower	65	65
Harvest efficiency 90%	Mixer	22	22
	Grazer filter	7	7
	Harvest pumps	6	6
	Centrifuges	111	7
	Dissolved air flotation unit		16
	Total	*211*	*123*
Common pond operation	Blower	65	65
Harvest efficiency 60%	Mixer	22	22
	Grazer filter	7	7
	Harvest pumps	8	8
	Centrifuges	167	10
	Dissolved air flotation unit		24
	Total	*269*	*136*
Blowers 35% of time in operation	Blower	23	23
Harvest efficiency 90%	Mixer	22	22
	Grazer filter	7	7
	Harvest pumps	6	6
	Centrifuges	111	7
	Dissolved air flotation unit		16
	Total	*169*	*81*

2.6 Heat and water consumption

The use of excess heat of the CHP of the total pilot (2 ponds and incidently the LED basins) ranged from 1115 to 1450 GJ per annum (Table 3). For all three years the heat consumption in the summer time was the half of the consumption in the other seasons. This is due to the fact that the indoor pond reached its setpoint of 25 ºC in the summer while the outdoor pond was heated almost throughout the whole year. The use of the excess heat resulted in an increase of the culture temperature of about 5 and 10 ºC for the outdoor and indoor pond, respectively. The stronger effect of the indoor pond is due to the effect of the glasshouse.

The total water consumption ranged from 2090 to 2280 m^3 per annum. This includes the water used for cleaning the harvest equipment and the water used for operating the LED basins. About 250-350 m^3/pond is used for maintaining the setpoint water level (compensation for evaporation). The main part was used for refilling after cleanings and increases of the setpoint water level. With regard to the latter, it must be emphasized that increasing the setpoint was not always necessary from the point of view of the pond management, but was done for additional experiments. Therefore, in a situation of a commercial algae production system water use is expected to be lower.

Table 3. Heat and water consumption of the total algae pilot.

Year	Heat consumption (GJ/a)	Water consumption (m^3/a)
2013	1220	2100
2014	1115	2280
2015	1450	2090

3 Pilot results 2013

3.1 Description ponds

Indoor pond

The indoor pond is operated as a continuously growing culture that is harvested by continuous circulation of the algae culture over a sedimentation tank, the coalescer (Figure 3). By recirculating water from the coalescer the use of fresh water or media is minimized. On working days (5 days a week) the content of the coalescer is collected yielding approximately 1 m^3 of slightly concentrated algae culture per day that is, subsequently, centrifuged (Figure 7) resulting in an algae paste of 10-15% dry solids. The water leaving the centrifuge is not reused but discharged to the sewage.

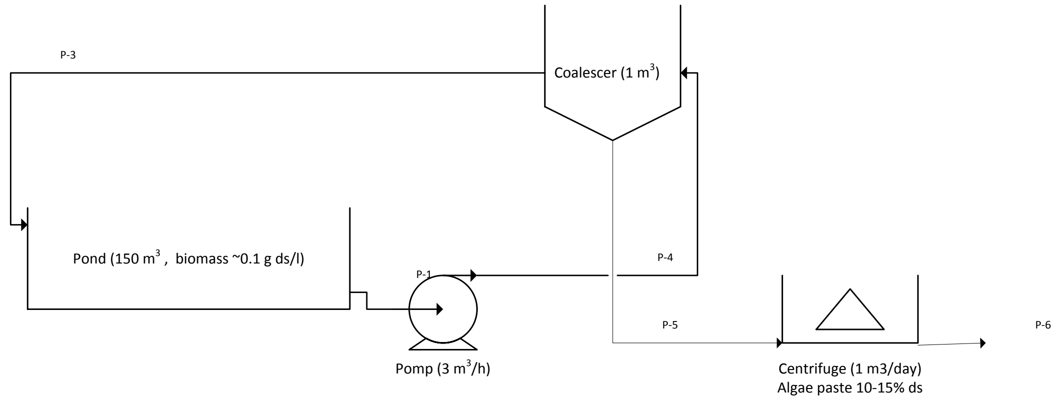

Figure 3. Schematic overview of the harvesting system as used for the indoor pond .

Figure 4. The coalescer (sedimentation tank, left) and the centrifuge (right).

Outdoor pond
Until July 2013 the harvesting system of the outdoor pond was also based on a coalescer for preconcentration and, subsequently, centrifugation of the collected slurry. However, in July 2013 the harvesting system was changed to direct centrifugation of the algae culture because tests had shown that the coalescer harvesting system causes limitations to the productivity of the ponds due to a too low biomass recovery.

In the new harvest system the algae culture is harvested by centrifugation. Extra centrifuge capacity was installed to be able to centrifuge sufficiently large volumes of algae culture (Figure 5 and 6). To minimize the amount of waste water the supernatant from the centrifuges is recycled to the algae cultures, as was the case with the old coalescer system.

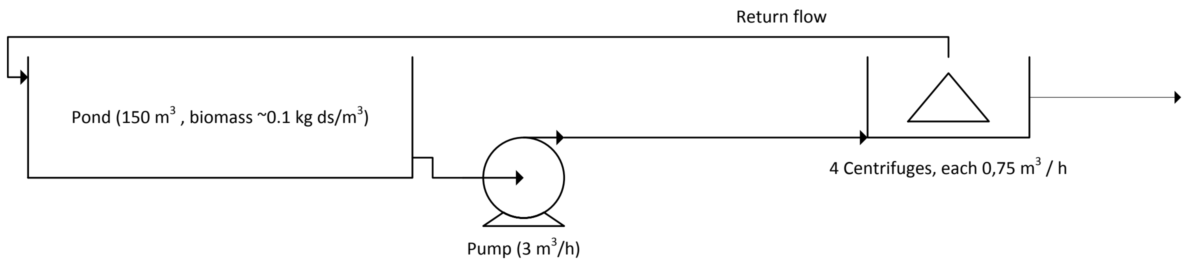

Figure 5. Schematic overview of the centrifuge harvesting system.

Figure 6. The four centrifuges for harvesting the outside pond.

3.2 Growing conditions

The algae productivity is strongly determined by light and temperature conditions. In Figure 7 the monthly sum of global radiation (obtained from a nearby weather station (KNMI, 2013)) is shown. In May the radiation level was lower than the long term average while in July and August radiation levels were higher than the long term average. The total annual radiation was comparable with the long term average. Figure 7 (right) gives the culture temperature of the both ponds and the average air temperature. In January-March average air temperature in 2013 was lower than the long term average. Due to the use of excess heat of the digester the culture temperature of the outdoor pond is about 4.5 °C higher than the average air temperature. For the indoor pond the difference in temperature between the culture and the air temperature is twice as high, about 10 °C. This is due to the additional effect of the greenhouse.

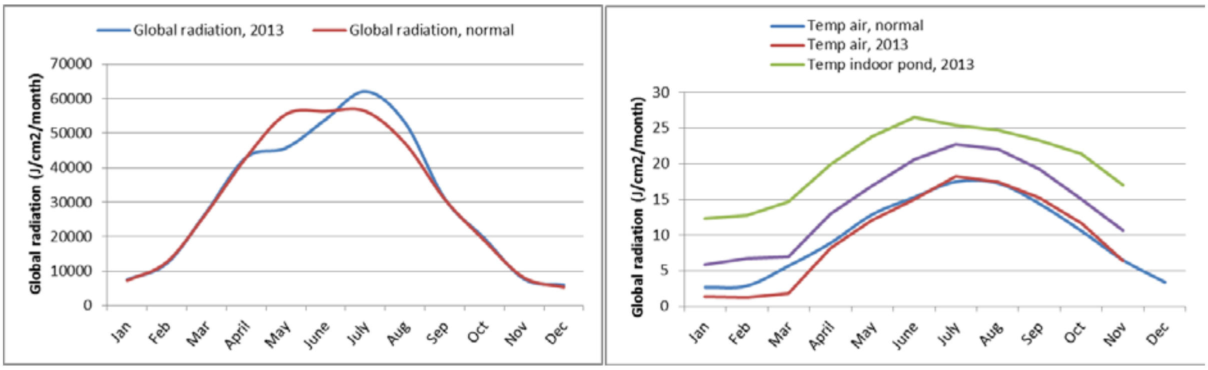

Figure 7. Global radiation and air and culture temperature in 2013 and the long term average (normal).

3.3 Biomass production

Figure 8 and Table 4 show the biomass production of the both ponds. The algae culture in the indoor pond continued to produce biomass throughout 2013 without crashing. The total accumulated amount of dry matter in 2013 amounts to 62 kg. This corresponds with a dry matter production of 2.5 ton per ha (water area pond = 250 m^2). Almost 80% of the total production is realized in the period April-September.

In order to compare the pilot results to the yield predictions of the open pond model (Spruijt et al., 2014), the model needs some adjustments to allow for a correct comparison. Firstly, the normalized global radiation data (average of last 30 years) in the model are substituted by actual data from 2013. Secondly, the actual global radiation is reduced to 60% intensity in order to compensate for the influence of the greenhouse. Thirdly, actual culture temperature data are used instead of a fixed ideal temperature of 25 °C (realized by heating). These adjustments result in a strong decrease of the production from 20 ton dm/ha/year in the original model to 10 ton dm/ha/year for the indoor pond in 2013 (Figure 9). The realized annual production (2.5 ton/ha) was 25% of the level predicted by the model.

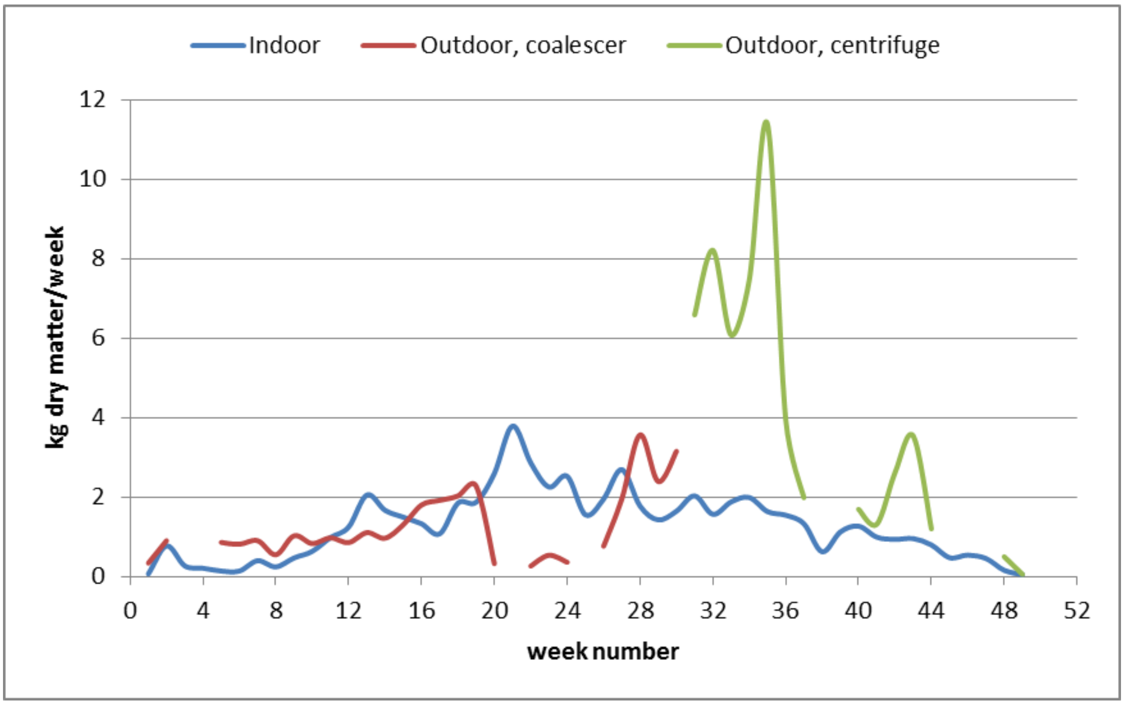

Figure 8. *Harvest data from both the indoor (T1) and the outdoor pond (T2), the harvesting system for the outdoor pond was changed in week 31 by substituting the coalescer by direct centrifugation.*

Table 4. *The total harvested biomass (kg dry matter) in different periods in 2013 in the indoor and outdoor pond.*

Period	Indoor pond	Outdoor pond
Week 1-30	42	33
Week 31-52	20	57
Week 1-52	62	90
Week 1-19	17	19
Week 1-2, 5-19	16	19
Week 31-36	12	46

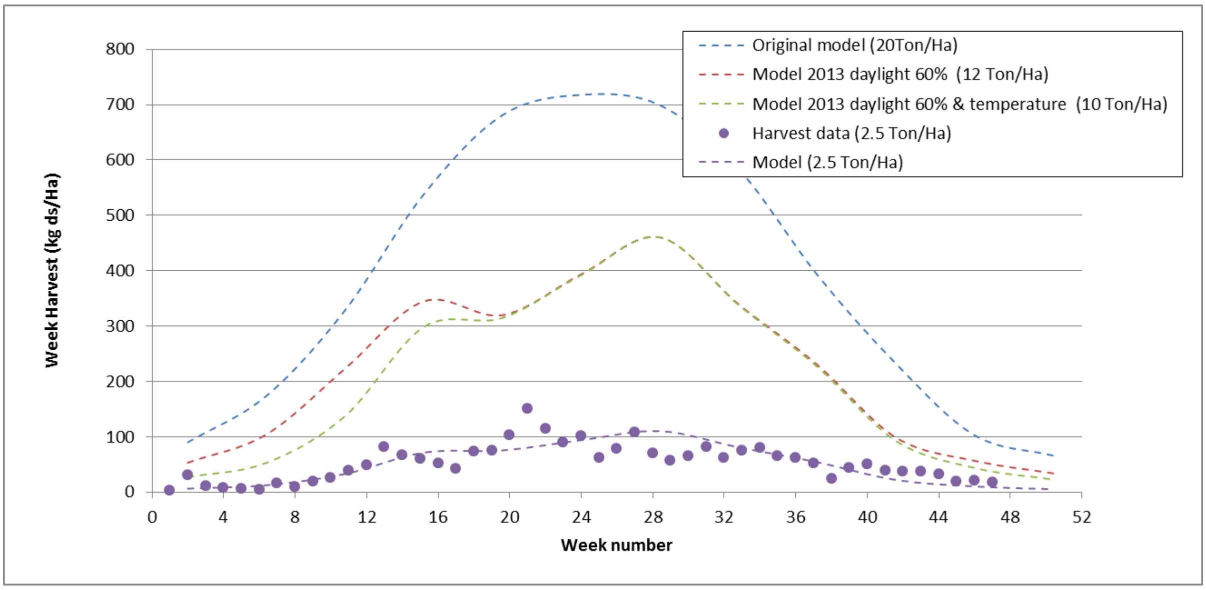

Figure 9. Harvest data from the indoor pond transformed to commercial production and compared to the algae production model in both original settings and with adjustments to light and temperature data from the pilot facility.

As mentioned before the outdoor pond was harvested with two systems. In the period that the pond is harvested with the coalescer (week 1-30) the total accumulated amount of harvested biomass was 33 kg (Table 4). In the same period in the indoor pond 42 kg was harvested. This difference is mainly due to periods with low or no production of the outdoor pond (see Figure 8). Firstly, in January a frost period caused a precautionary shut down of the harvesting system of the outdoor pond (week 3-4) while for the indoor pond production and harvesting could be continued in this period. The second period of low production of the outdoor pond was in week 20-26 when the culture crashed twice (see paragraph 3.5 for more details). In periods without problems the biomass production of the outdoor pond was somewhat higher than for the indoor pond, in week 1-19 (excluding the frost period in January) the cumulative dry matter production was 16 and 19 kg for the indoor and outdoor pond, respectively (Table 4).

After changing the harvesting system of the outdoor pond to direct centrifugation (week 31), the amount of weekly harvested biomass strongly increased (Figure 8). The direct centrifugation caused an increase in the weekly harvested biomass from about 2.5 kg dry weight to about 8 kg dry weight. In the period from week 31-36 the outdoor pond produced 46 kg dry matter while the production in the indoor pond was 12 kg dry matter. However, the initial difference rapidly decreased later in the year partly due to interruptions in the harvesting. The first interruption in week 37-39 is caused by an attempt to restart the algae culture with a fresh inoculum. In week 45-47 the harvesting by centrifuging was temporarily stopped because of a low biomass density in the pond.

The total accumulated dry matter production in 2013 for the outdoor pond was 90 kg dry matter (33 kg in the period harvested with the coalescer and 57 kg in the period harvested with the centrifuges) corresponding with annual production of 3.6 ton dry matter per ha.

As for the indoor pond the harvest data of the outdoor pond were compared with the results of the growth model that was fed with the actual data for light and temperature (Figure 10). Based on the actual global radiation and culture temperature an annual yield of 14 ton dry matter per ha is predicted. This is higher than the yield level predicted for the indoor pond (10 ton dry matter per ha). This is due to the higher light intensity (the light intensity of the indoor pond is reduced to 60% by the glass in the greenhouse) although the lower culture temperature compensates a part of the light effect. The yield data in the period that the pond is harvested with the coalescer correspond with an annual production

level of 3.0 ton per ha (excluding the periods with no production). For the centrifuge the found data correspond with an annual yield level of 9 ton dm/ha which is about three times than for the coalescer harvest.

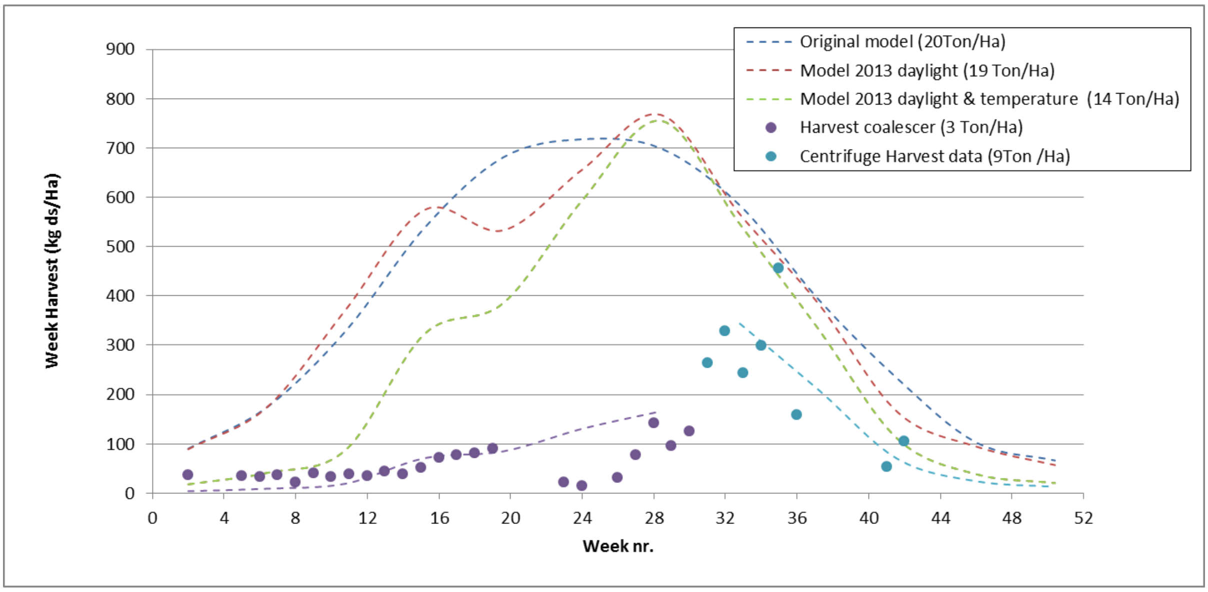

Figure 10. Model prediction and pilot results of biomass production for the outdoor pond.

The dry matter production is related to the global radiation and the culture temperature as shown for the indoor pond in Figure 11. However, both growing factors are also related. From the model calculations (Figure 9 and 10) it can be seen that the effect of temperature is smaller than the effect of global radiation (compare predicted yield levels with and without adjusting for actual temperature).

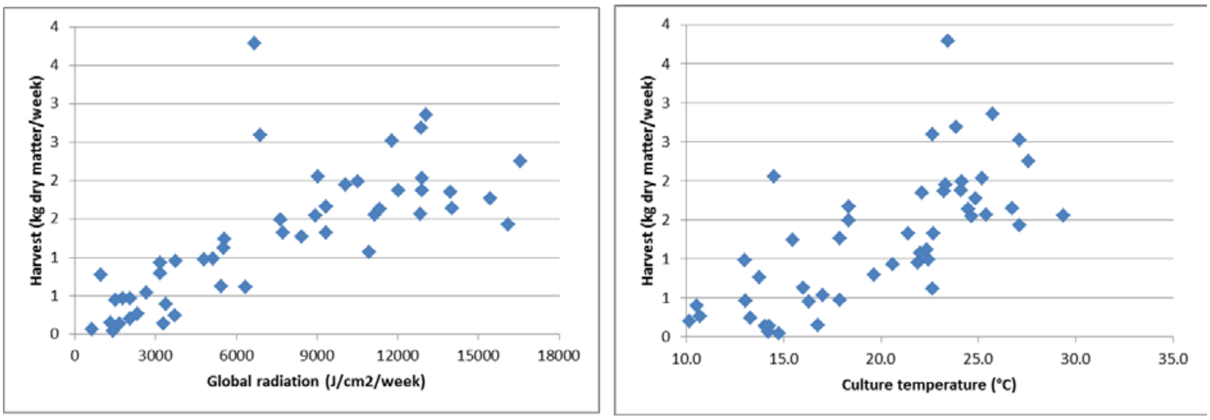

Figure 11. Harvested algae dry matter as affected by global radiation and culture temperature of the indoor pond.

3.4 Culture density

The culture density was monitored by daily measurement of the optical density (OD). This is a quick method that provides a good indication of the performance of the culture. In Figure 12 the course of the OD at 440 nm is shown for the two ponds. In this figure the average is shown of 5 working days in order to minimize measuring errors and to visualize general trends. In Figure 13 also the culture depth is given.

Generally, there seems to be a seasonal effect in the culture density, with a low density in the winter months and a higher density between April and October. However, there are some other factors to be mentioned. Firstly, in the summertime the culture depth is decreased from about 70-80 cm to about 40 cm. This decrease in culture depth coincides with an increased culture density. However, in the beginning of the year no effect of changing the culture depth (from 40 to 80 cm) on culture density was observed.

Furthermore, the culture density can also be affected by predators. The OD of the indoor pond remained low throughout the first half of the year. This may be due to predatorial pressure as in the first half of 2013 large amounts of predators such as Daphnia and Brachionus were observed. Moreover, the sudden increase of OD in June coincides with the disappearance of these algae consuming predators. Around that time we have attempted to remove the predators by giving a heat treatment and shortly afterwards (July 2013) a drum filter with a pore size of 70 micron was installed to remove predators from the algae culture.

In the outdoor pond the sudden drops in OD are due to culture crashes (see 3.5).

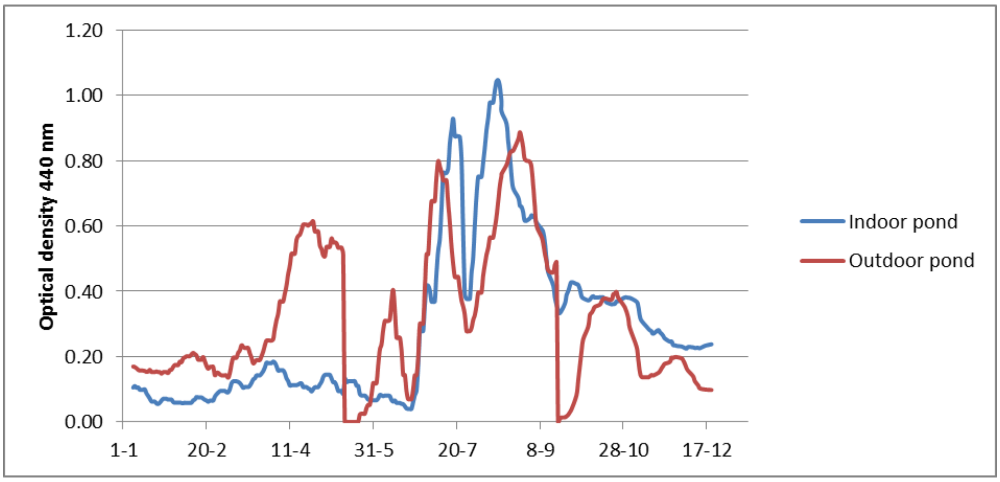

Figure 12. Optical density a7 440 nm of the indoor and outdoor pond in 2013.

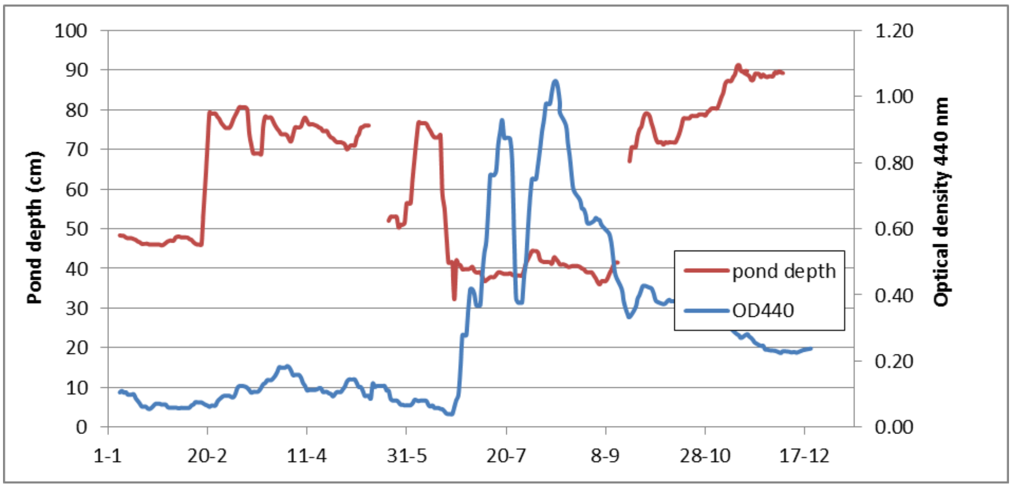

Figure 13A. Pond depth and optical density at 440 nm of the indoor pond

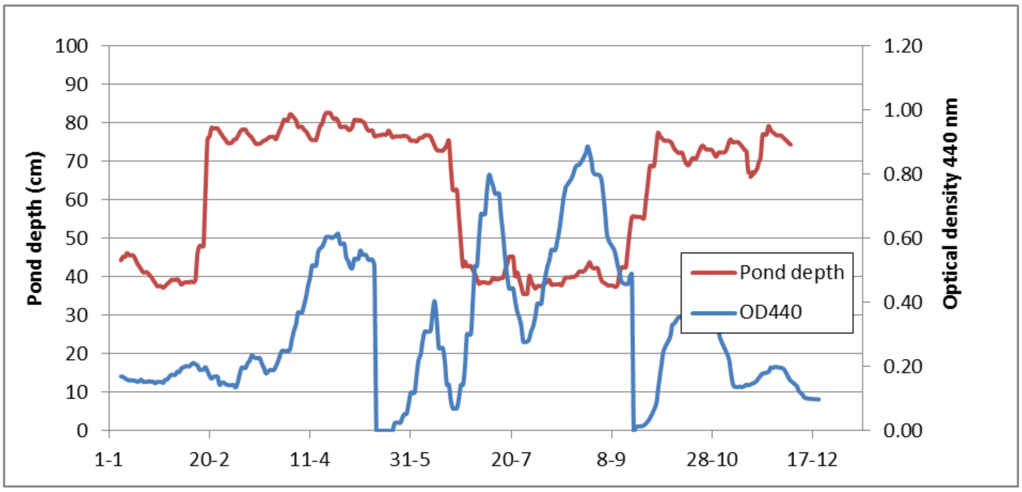

Figure 13B. Pond depth and optical density at 440 nm of the outdoor pond

3.5 Culture crashes

From literature it is known that culture crashes can occur at unexpected moments. Surprisingly, the indoor pond kept growing throughout 2013 without a serious crash. The outdoor pond, on the other hand, has been emptied and restarted twice because of a culture crash. The reason behind the two crashes remains unclear. However, from observations of the ponds we can nowadays recognise the first signs of a culture crash.

The first crash of 2013 occurred in the second week of May, when without a clear reason the algae culture in the outdoor pond suddenly turned brown and the harvested algae paste started smelling of ammonia. A few days later the culture density reduced drastically because the algae present in the culture started to flocculate. Eventually, the culture density reduced to approximately zero, with all biomass harvested or settled at the bottom of the pond. After cleaning and refilling the pond with a fresh inoculum the fast growing culture collapsed again within 3 weeks.

As described above the first signs of a crash are a change of colour and a bad odour culminating in the final crash which is clear by the sudden and dramatic drop in culture density. For the second crash we first noted that the green culture starts to turn brownish with a hint of yellow on the 6th of June. The change in colour was accompanied by a rotting odour that is most distinct when handling the harvested biomass but also noticeable around the algae culture in the pond itself. Several days later, between 11th and the 14th of June the crash manifests and optical density reduced dramatically (Figure 14).

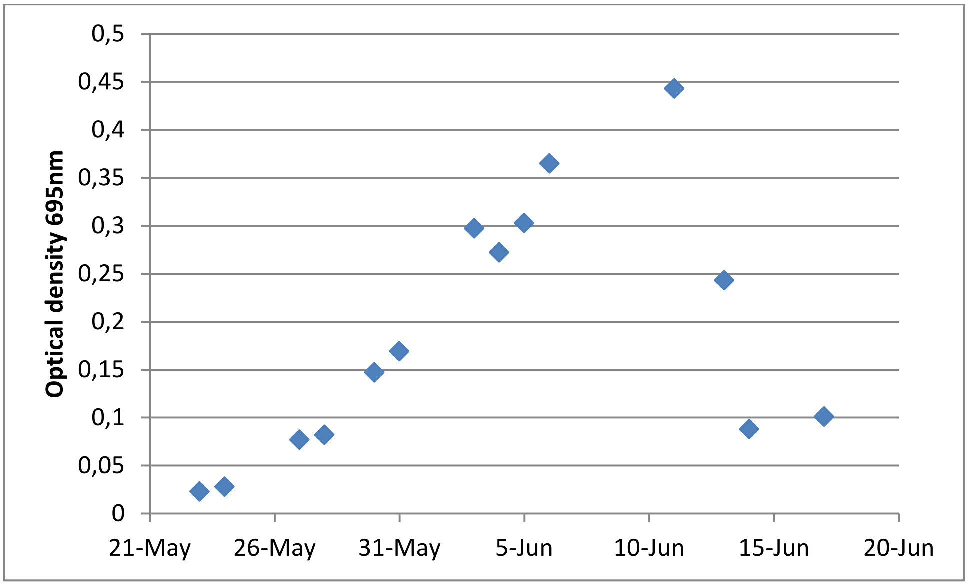

Figure 14. The optical density before and after the second crash of the algae culture of the outdoor pond in June.

3.6 Heat consumption

Optimising the use of excess heat from the biogas fired CHP-engine present at the Acrres pilot site has been an important reason for the construction of the algae ponds. Firstly, it will enhance the algae production, In addition, due to subsidies for using excess heat extra revenues can be obtained.
The heat consumption of the total algae pilot (two ponds and two pre-culture basins) is calculated based on the difference in temperature between the ingoing and outgoing water (cooling water from the CHP) and the water flow through the heating system. The distribution of excess heat within the algae pilot, across the production ponds and preculture basins, is not measured. Figure 15 shows the calculated heat consumption of the total algae pilot.
From the start of measurements in March until middle of June and from the beginning of September onwards, the heat consumption is 3.7-3.9 GJ per day. In the summer (Middle of June till beginning of September) the heat consumption is about 50% lower (1.8 GJ per day). This corresponds well with the temperature of the algae ponds (Figure 7). From the middle of June to the beginning of September the indoor algae pond had reached its setpoint temperature of 25°C and needed no additional heating. The outdoor pond was heated throughout the whole year.
From Figure 15 it can be seen that the heat consumption in the spring and autumn is constant while average temperatures are quite different within these periods. This means that independent of the temperature a fixed maximum amount of heat is daily added to the algae pilot per day. For the outdoor pond this results in a culture temperature that is approximately 5°C above the average environmental temperature (Figure 7). For the indoor pond the difference in temperature between the culture and the

environment is twice as high, about 10°C, due to the additional effect of the glasshouse.

The exact effect of the raised temperature on the algae production cannot be derived from the harvest data. In general, an increased productivity would be the expected result as long as the algae cultures are not overheated.

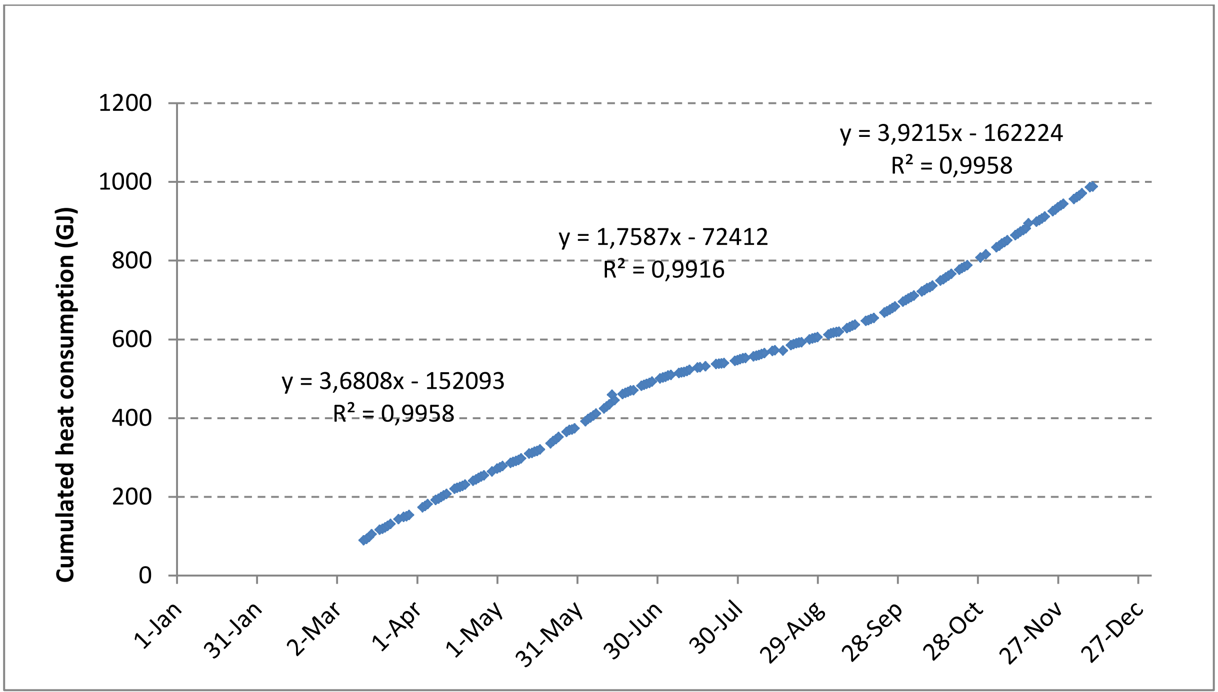

Figure 15. Cumulative heat consumption by the algae pilot facility in 2013.

3.7 Water consumption

Water consumption of the combined algae pilot is measured at the intake point for tap water. For the last 9 months of 2013 the total water consumption has been 1600 m^3, which gives an extrapolated overall consumption of about 2100 m^3 per year. This includes water used for cleaning the harvest equipment and the water used for operating the LED supported photobioreactors.

A better insight in the water consumption of the separate algae ponds can be derived from the course of the water level of the ponds. For the indoor pond, any increase of the water level (or culture depth) is a direct result of water intake. Based on these increases a cumulative water addition of 800 m^3 can be calculated. This is partly caused by increasing the set point for the water level from 40 to 80 cm (in February and in September, Figure 13A), this accounts for about 200 m^3 in 2013. The remaining 600 m^3 is used to compensate for water losses due to harvesting and evaporation. Harvesting was performed on 240 days with a loss of approximately 1 m^3 per day accumulating to 240 m^3 for 2013 which leaves thus about 350m^3 of water that must have been lost by evaporation throughout the year.

A similar calculation can be made for the outdoor pond. Compared to the indoor pond, additionally, also the rainfall has to be taken into account. The calculated cumulative water addition in 2013 amounted to

1075 m^3. About 655 m^3 is explained by crashes, equipment failure, planned pond cleaning and water needed to increase the pond depth from 40 to 80 cm (in February and September, Figure 13B). On 115 days the pond was harvested with the coalescer resulting in a water loss of 115 m^3. In the period the pond was harvested with the centrifuges no water loss at harvest was calculated as all water is returned to the pond. The remaining amount of water, 305 m^3, must have been the sum of water addition due to maintain the setpoint (compensation for evaporation) and rainfall (775 mm corresponding with 200 m^3).

3.8 Conclusions

- The indoor pond kept growing continuously in 2013 without serious culture crashes. The outdoor pond, however, has been emptied and restarted twice because of culture crashes. No clear explanation could be given for the observed crashes.
- The total annual algae production of the indoor pond, as harvested by the coalescer, was 2.5 ton dry matter per ha. From January till the end of July the outdoor pond was also harvested by the coalescer. Excluding the periods with crashes the harvested dry matter yield was about 20% higher than for the indoor pond corresponding with an annual production of 3 ton dry matter per ha. After changing the harvest method to direct centrifugating end of July, the harvested yield increased strongly with a factor 3 corresponding with an annual production of about 9 ton dry matter per ha. However, from the mid of September onwards the yield decreased due to problems with the growth after a restart with a new inoculum.
- The heat consumption of the total algae production system was 3.7-3.9 GJ per day in the periods January-May and October-December. In the summertime (June-September) the heat consumption decreased to 1.8 GJ per day. The available excess heat of the CHP-unit allowed an increase of the culture temperature of about 5 °C above the average air temperature for the outdoor pond. For the indoor pond temperature was about 10 °C above the air temperature. The stronger effect for the indoor pond is due to the effect of the glasshouse.
- The total calculated annual water consumption (based on changes in water depth) was about 800 and 1100 m^3 for the indoor and outdoor pond, respectively. The higher water consumption of the outdoor pond was mainly due to refilling the ponds after crashes that did not occur in the indoor pond.

4 Results pilot 2014

In this chapter the algae production in the open pond systems in Lelystad is described. First, the results of the outdoor pond, being the production pond, are discussed (paragraph 4.1). Unlike 2013, the indoor pond is used for experiments in order to test adjustments of the system. For this pond in 2014 a new harvest method based on dissolved air flotation was tested. The results of these experiments are described in paragraph 4.2.

4.1 Outdoor pond

4.1.1 Description pond

In July 2013 the harvesting system of the outdoor pond was changed to direct centrifugation, test results showed an increased biomass productivity compared to harvesting with the coalescer (sedimentation tank). The year 2014 was used for continued monitoring of the algae production with the aim to get an indication of the production potential results for all seasons.

The process design for the algae production as tested in 2014 was based on the following guidelines.

- The algae culture is kept until it crashes,
- Nutrients are added in order to keep a target level of 25 mg N/l and 3.5 mg P/l,
- Continuous sparging of air,
- Flue gas is added to the sparged air when the pH increases above the setpoint level (pH 8),
- The algae culture is heated (with excess heat of the CHP) until the setpoint is reached (25 °C),
- Water is not refreshed, but only added when water level decreases due to evaporation and after cleanings,
- The harvest is done by direct centrifugation (Figure 16),
- A drum filter (mesh 75 µm) added to remove predators (Figure 17).

The design is similar to the previous years except for the changed harvesting system and the addition of a predator filter.

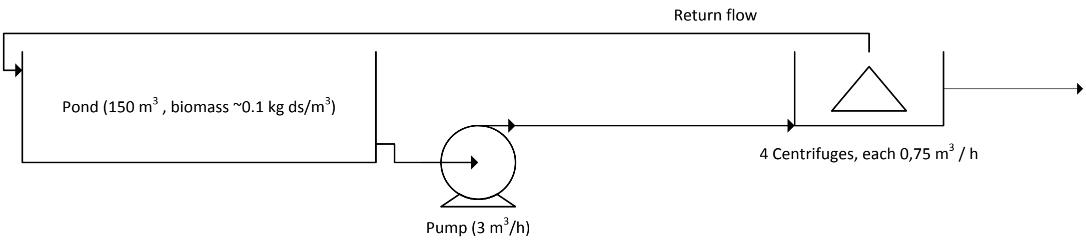

Figure 16. Schematic overview of centrifuge harvesting system.

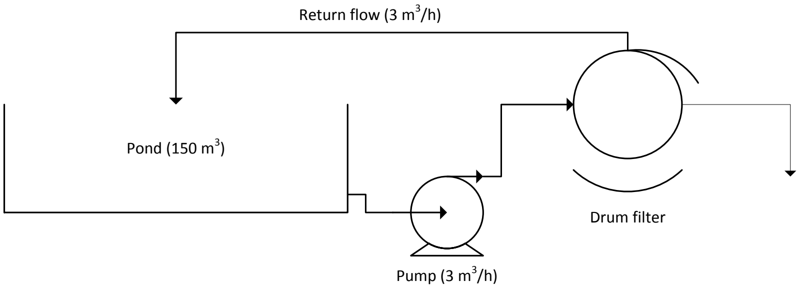

Figure 17. Schematic overview of predator control by drum filter

4.1.2 Growing conditions

The algae productivity is strongly determined by light and temperature conditions. In Figure 18 the monthly sum of the global radiation is shown. In March and June the radiation levels were higher than the long term average and in May levels were lower than the long term average. The total annual sum of the global radiation was 2% higher than the long term average.

Figure 18 also shows the average air temperature and the culture temperature of the outdoor pond. The average air temperature was 1.6 °C higher than the long term average. Temperatures were especially higher in the winter, spring and autumn. The average culture temperature was 4.7 °C higher than the average air temperature in 2014 due to the addition of excess heat of the CHP unit.

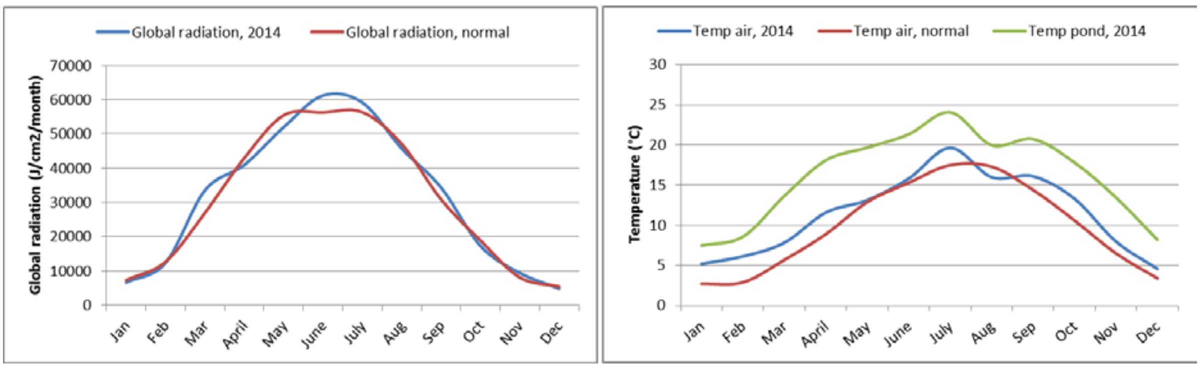

Figure 18. The global radiation and the air and culture temperature in 2014.

4.1.3 Biomass production

In Figure 19 the weekly harvest of the outdoor pilot pond is shown for 2014 together with the global radiation (weekly sum). At the start of the year the pond still contains the algae culture that was inoculated in September 2013. Until mid-February the weekly harvest (with a single centrifuge in operation, only on working days) is low, approximately 1 kg algae biomass per week. From mid-February onwards we started harvesting with three centrifuges, as a result the harvested biomass immediately increased to about 4 kg per week. Then from mid-March onwards harvesting was done with four centrifuges. Together with the increased radiation in the spring the daily harvest increased to 8-12 kg dry matter per week.

Around mid-June the culture suddenly crashed. The restart at the end of June is done with another algae mixture (Chlorella protothecoides and Scenedesmus obliquus). The fresh inoculum (that was growing perfectly well in the preculture basins) showed a very slow growth with a lag-phase of approximately a month. Only from the beginning of August the culture picked up and the harvesting was restarted. From August onwards the daily harvest started decreasing with the seasonal reduction of the global radiation.

At the end of 2014 the outdoor pond had produced a total of 176 kg dry weight algae biomass corresponding with about 7 ton dry matter per ha.

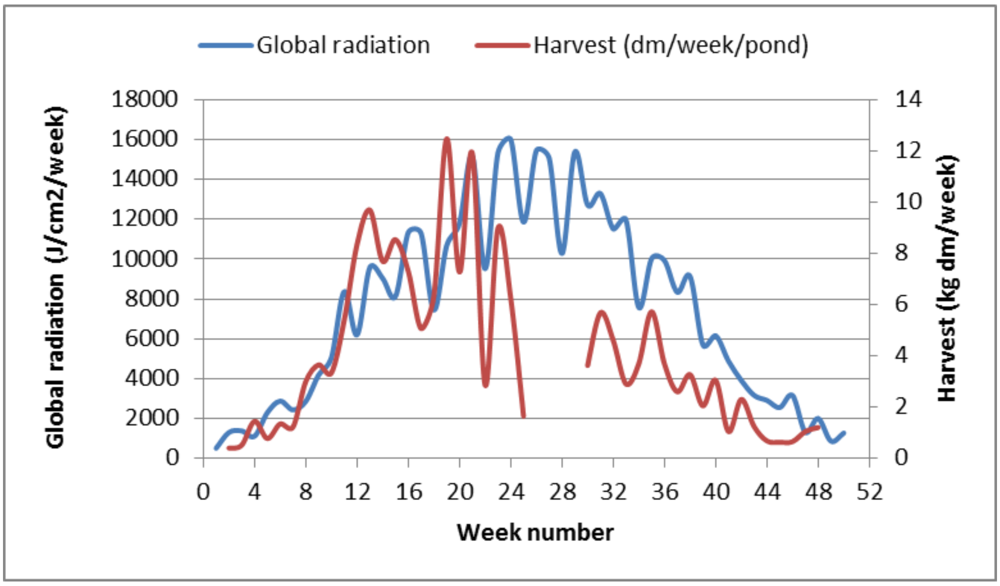

Figure 19. The weekly algae biomass yield and the global radiation (weekly sum, obtained from KNMI weather station Lelystad) of the outdoor pond in Lelystad in 2014.

As for 2013, we compared the harvest data of 2014 with yield predictions of the open pond model. Assuming a photosynthetic efficiency (conversion of solar radiation energy in glucose energy) of 1.5% and an optimal culture temperature of 25 °C a biomass production of 20.6 ton per ha was predicted (Figure 20). The predicted biomass production using the actual culture temperature was 17.4 ton per ha. Until week 13 the actual harvest corresponds with the predicted yield levels. After week 13 the actual harvest data are lower than the predicted yield level. This is partly due to the crash in June, but also in the second part of the year, after the pond recovered from the crash, the actual harvest was lower than in the spring corresponding with an annual yield level of about 6 ton biomass per ha.

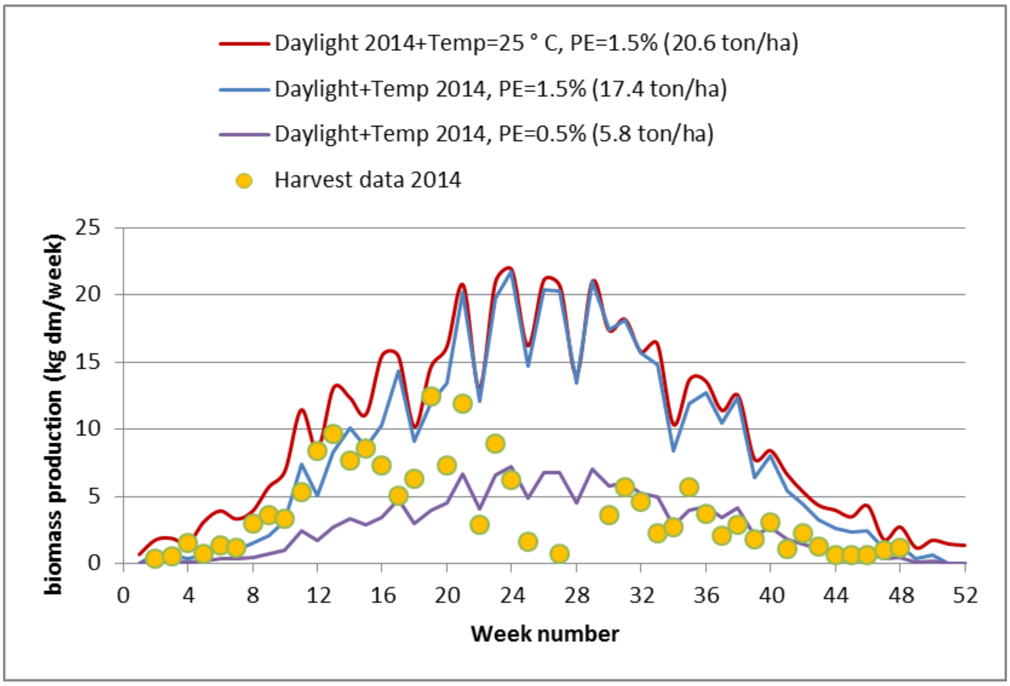

Figure 20. Harvest data from the outdoor pond compared to model calculations (PE = photosynthetic efficiency, Temp =culture temperature).

4.1.4 Culture density / species

The culture density of the ponds is checked daily by measuring the optical density. In addition, the cell density and species composition is assessed once a week by microscope. Figure 21 shows the optical density combined with the depth of the culture (left) and combined with the biomass harvest (right). In the spring an increase of the optical density is observed coinciding with the increased harvest. However, after the crash in June, initially, in week 32-38 the optical density of the culture increased to a comparable level as was observed in the spring while the harvest was considerably lower in that period.

The decrease in culture depth from 95 cm to 40 cm in week 13 coincided with an increase in optical density. This may be due to a higher amount of light per unit volume. However, in the week before the decrease in culture depth the OD already increased from 0.27 to 0.40. Additionally, also the global radiation increased in the weeks after the culture depth was decreased. Therefore, it is difficult to relate the increase in optical density directly to the decrease in culture depth.

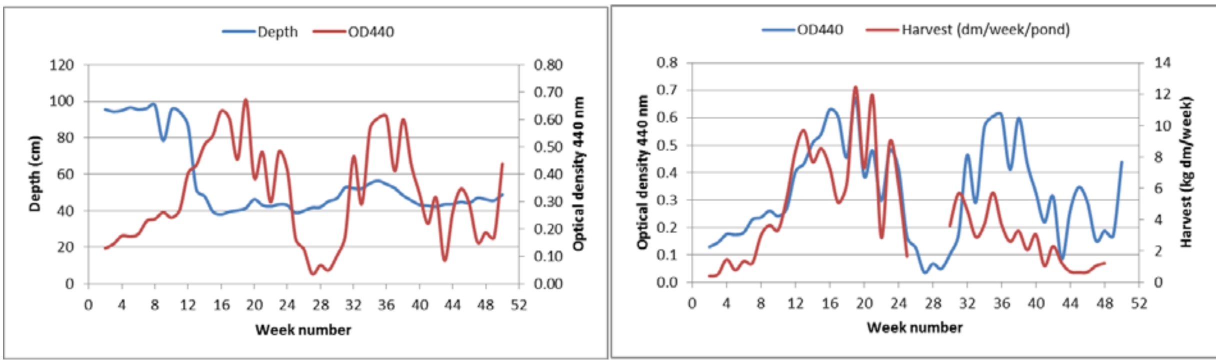

Figure 21. The depth and the optical density of the culture at 440 nm (left, weekly average) and the biomass harvest and the optical density of the culture (right, weekly average) of the outdoor pond in

Lelystad in 2014.

In Figure 22A the cell density is given combined with the optical density of the culture, in Figure 22B the relationship between optical density and cell density is shown. The latter is done for two periods, before and after the crash in June. In the winter and spring (before the crash) at the same optical density the cell density is lower than in the period after the crash. This difference is probably the result of a changed species composition of the algae culture.

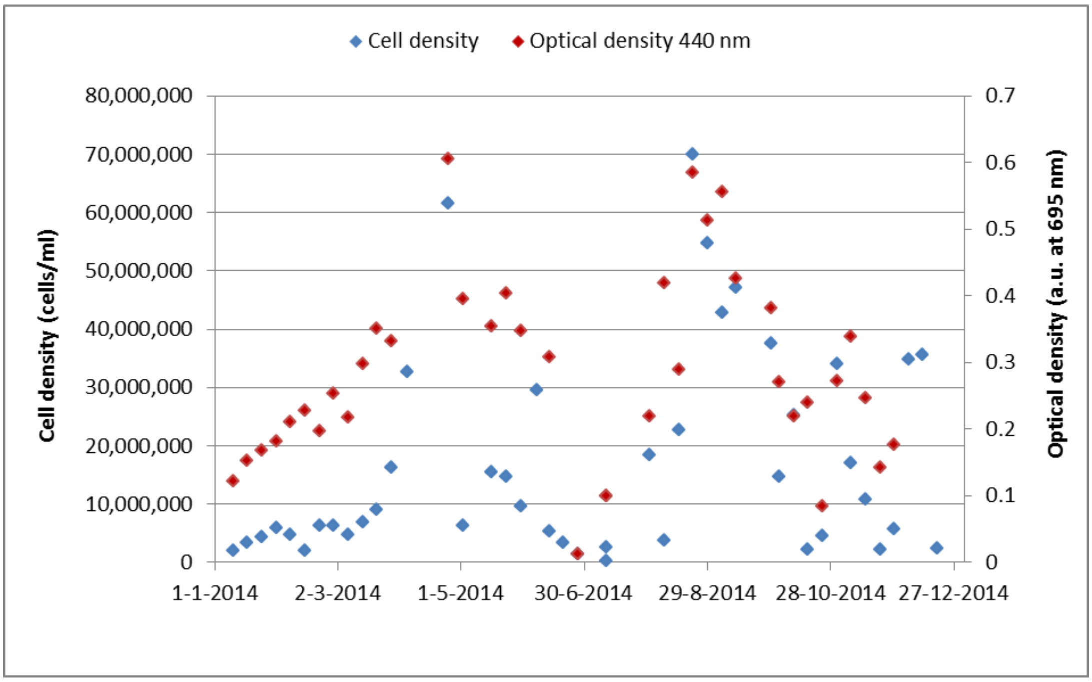

Figure 22A. Collected data for optical density at 440 nm and cell density of the outdoor pond in 2014.

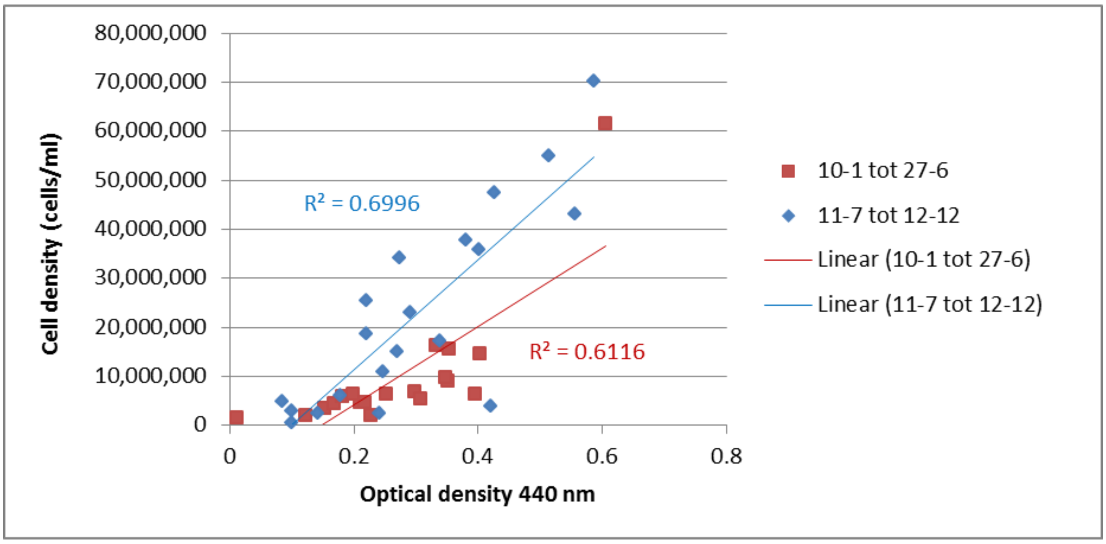

Figure 22B. Relationship between optical density at 440 nm and cell density of the outdoor pond in 2014.

In Figure 23 the microscopy counts are shown. First of all the results show that at the start of the year the pond contains little of the original algae mixture that was used for the inoculation in September 2013, the original mixture contained *Chlorella sp,. Scenedesmus sp,* and *Pheaodactylum sp.* which make up for only 25% of the total cell count. Approximately 75% of the algae mixture consists of other (contaminating) species. The exact identity of those species was not determined.

The inoculum used to restart the ponds in June consisted of a mixture of both *Chlorella protothecoides* and *Scenedesmus obliquus*. As mentioned before, the precultures of the inoculum grew well, but when the 20 m^3 preculture was transferred to the outdoor pond, initially, the culture did not grow well. Only 4 weeks after inoculation the density of the culture started to increase, by then predominantly consisting of *Chlorella spp*. It is not clear whether this determined *Chlorella* species is the species used for inoculation, as the microscope does not allow to distinguish in that detail. In October

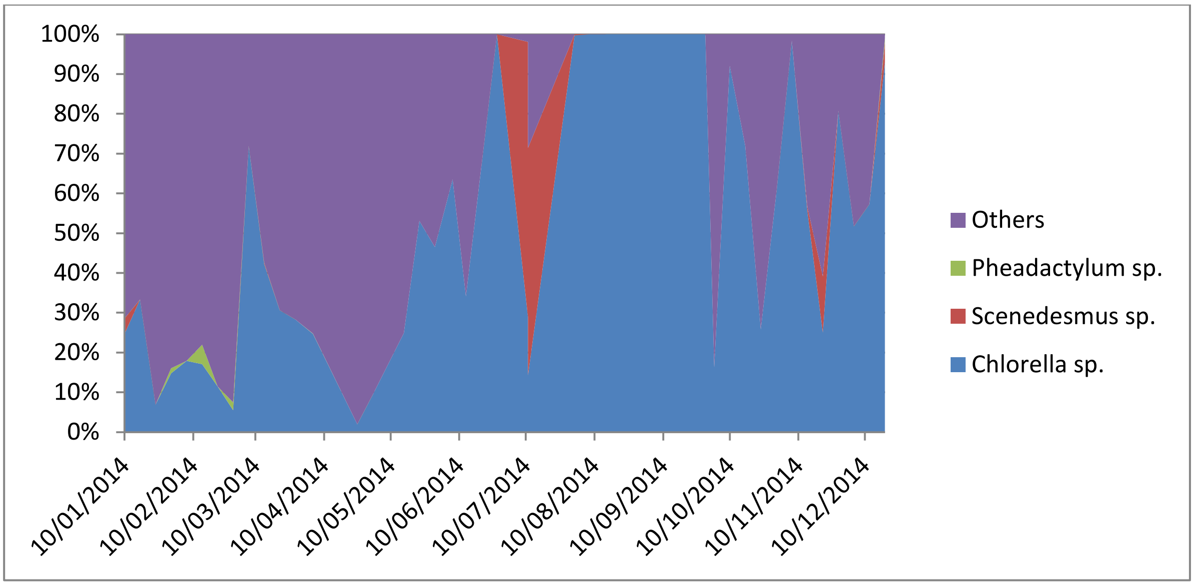

Figure 23. Species composition of the algae mix throughout 2014 as assessed by microscopy.

4.1.5 Predators

In 2013 a drum filter was installed in order to remove a part of the grazers. Figure 24 shows the total number of grazers in the inflow and outflow of the grazer filter. The course of the number of grazers in the pond during the year is characterized by large fluctuations The grazer pressure was highest in August with total number increasing to about 12,000/l. Generally, the grazer pressure in the summertime was higher than in winter and spring.

The efficacy of the grazer filter was low (Figure 25). Figure 10 shows that the size distribution of the found grazers did not change that much. It is remarkable that the outflow of the grazerfilter still contains grazers > 75 µm while the grazer filter had a mesh of 70 µm.

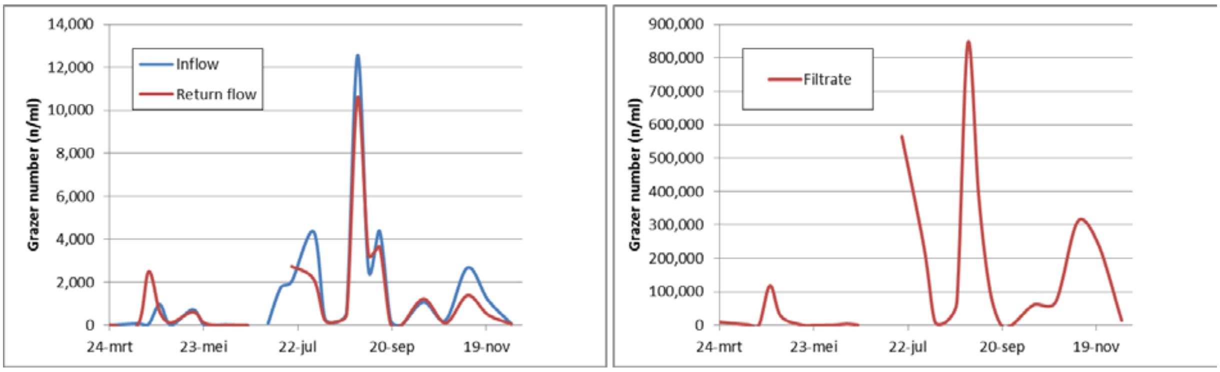

Figure 24. *Total number of grazers in the inflow and return flow of the grazer filter (left) and the total number of grazers in the filtrate of the grazer filter (right) of the outdoor pond in 2014.*

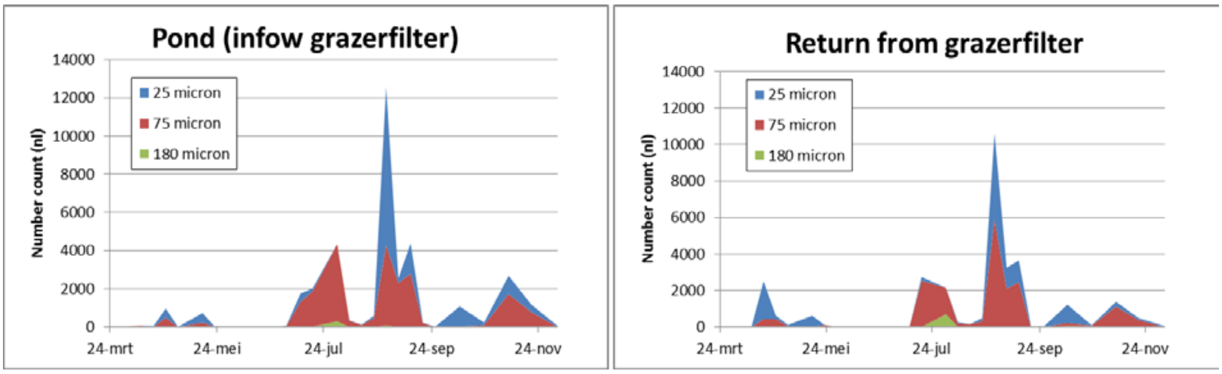

Figure 25. *Total number of grazers in the different size classes for the inflow (left) and the return flow of the grazer filter (right) of the outdoor pond in 2014.*

4.1.6 Test liquid fraction digestate as nutrient source

As mentioned before nutrient supply is done with chemical fertilisers, but preferably side streams should be used, like digestate from the digester. A drawback of digestate is the dark colour and the presence of solids that decrease the light transmission in the culture. A pre-treatment by a separation step resulting in a liquid and solid fraction and using the liquid fraction for the algae production may help to decrease this problem. Earlier lab experiments showed that a 0.5-1% diluted liquid fraction did not affect algae growth significantly. At these dilution levels nutrient supply is sufficient for algae growth. In order to assess the effect of digestate use on a bigger scale in December 2014 a test was done in the outdoor pond

Nutrient content digestate and liquid fraction

Before the test the digestate of the Acrres-digester was separated with a screw press. Table 5 gives the nutrient content of the digestate and the liquid fraction. The liquid fraction still contains organic solids. Based on total N and P, the N/P-ratio of the digestate and the liquid fraction was 5.8 and 6.8, respectively. Microalgae are only able to take up mineral N and P (NH_4-N and PO_4-P). The PPO-analysis shows that about 40% of the total P is present in the form of PO_4-P. The ration NH_4-N/PO_4-P is about 10 (based on the PPO-analysis). The N/P-ratio of microalgae biomass is about 5-10, so, the liquid fraction is expected to provide sufficient N and P.

It must be emphasized that before the test the pond is not emptied, cleaned and restarted with new inoculum. As the nutrient supply in the pond was sufficient at that moment, the possible nutrient effects of the liquid fraction (e.g. NH_4-N instead of NO_3-N) cannot be assessed.

Table 5. Nutrient content (kg/ton) digestate and liquid fraction of the digestate.

	Dry matter	Organic matter	Total N	NH_4-N	Total P	PO_4-P	K	Mg	Na
Digestate, Blgg[1]	73	48	4.31	2.15	0.74		5.15	0.75	0.55
Liquid fraction, Blgg[2]	56	34	3.96	1.93	0.58		5.00	0.60	0.50
Liquid fraction, PPO[3]			3.98	2.30	0.56	0.23			

1 analysis by Blgg-lab, average of 2 samples

2 analysis by Blgg-lab, average of 3 samples

3 analysis by PPO-lab, 1 sample

Results addition of liquid fraction

At the start of the experiment 200 L liquid fraction was added to the pond. As the pond contained about 100 m^3, the liquid fraction was diluted 500x. Initially, it was planned to add more liquid fraction, however, as the pond colour changed significantly after adding 200 L, it was decided to be careful and first monitor the effects on the algae. After one week, another 200 L was added.

Figure 26 shows that even strong diluted (≤ 0.5%) a colouring is observed.

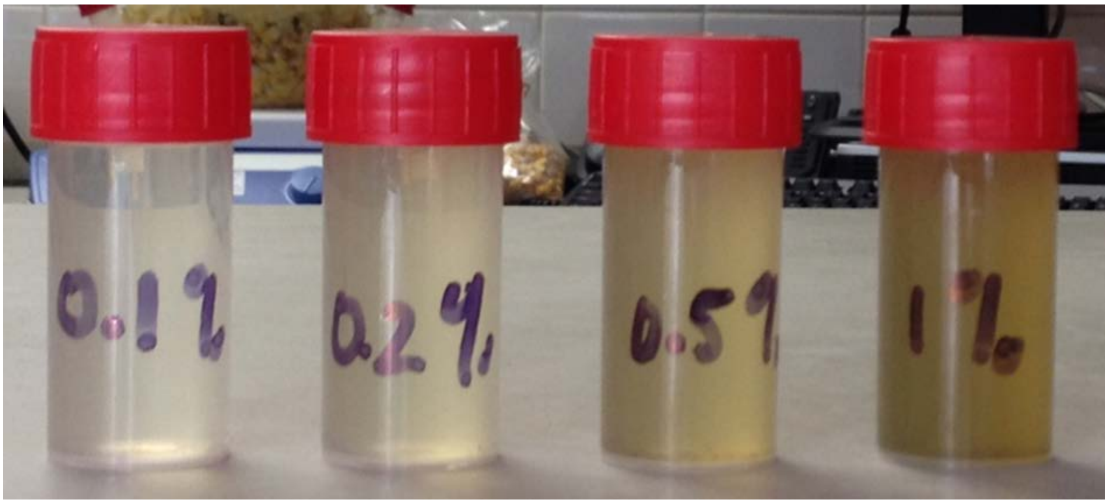

Figure 26. Dilution of the liquid fraction with water (0.1-1 vol-%).

In Figure 27 the course of the NO_3-N, NH_4-N and PO_4-P concentration in the pond is shown. Before the start of the experiment the pond already contained 43 mg NO_3-N/L and 5.8 mg PO_4-P/L. No NH_4-N was found as the pond was only fed with NO_3-N.

After the first addition of the digestate the NH_4-N concentration increased to about 5 mg/L. This increase corresponded quite well with the amount of added NH_4-N to the pond. In the week after the first addition the NH_4-N content decreased with about 1 mg/L. After the second addition the NH_4-N concentration increased to about 10 mg N/L after which it decreased again with 1 mg/L after one week.

The PO_4-P concentration increased slightly from 5.8 to 6.3 mg/L after the first addition and, subsequently, to 6.8 mg/L after the second addition. The observed increase corresponded with the added amount of PO_4-P to the pond.

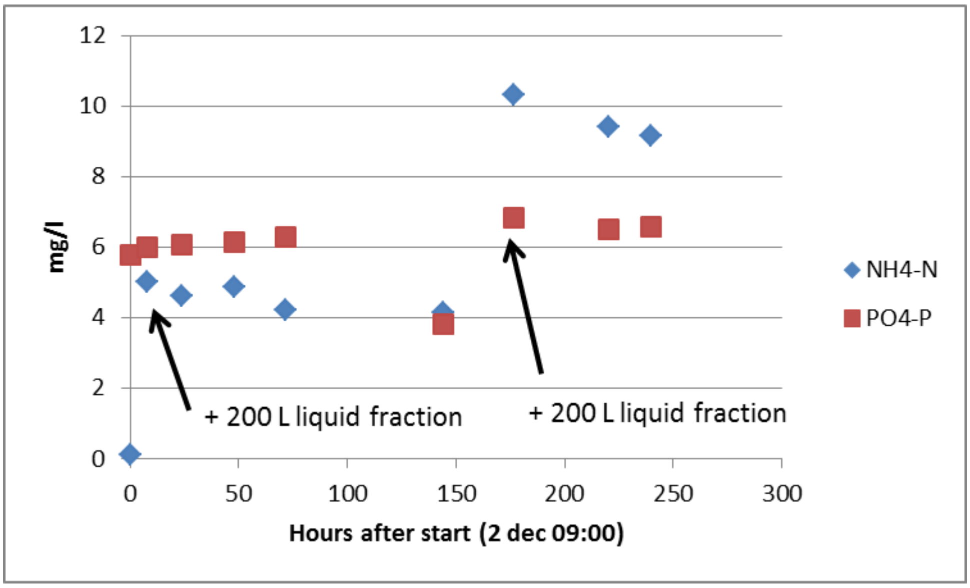

Figure 27. NH₄-N and PO₄-P concentration (mg/L) in the pond after the addition of liquid fraction.

Figure 28 shows the optical density of the pond after the addition of liquid fraction. After both additions, a significant increase of the OD was observed. After the addition the OD decreased, which will probably partly be due to the simultaneous harvest of organic matter and microalgae with the centrifuge. Mineralisation in the pond may also play a role, however, the temperature was relatively low beginning of December.

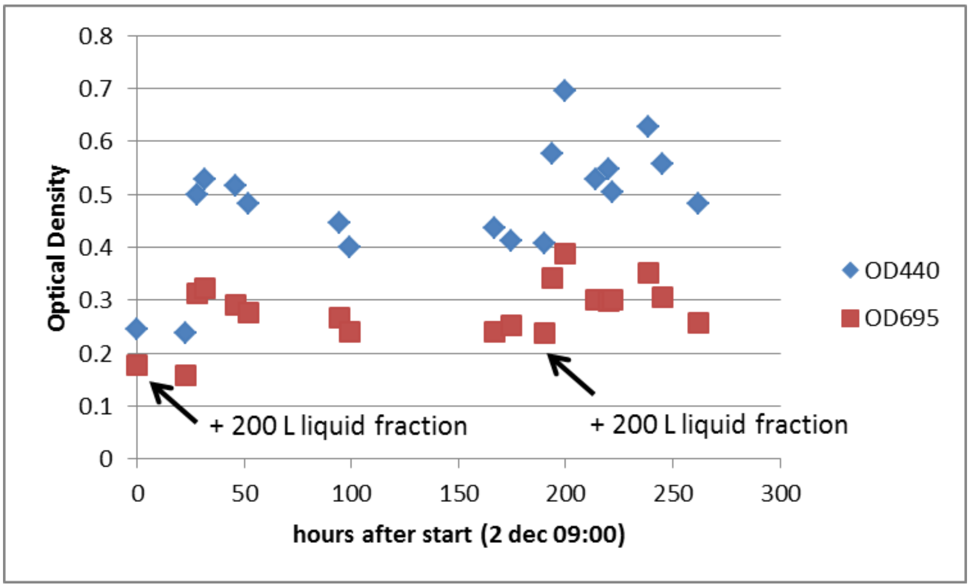

Figure 28. Optical density (440 and 695 nm) of the pond in the pond after the addition of liquid fraction.

In Figure 29 the daily harvest (wet paste) of the outdoor pond is shown. A day after the addition of liquid fraction the harvest almost doubled. This is probably due to the simultaneous harvest of organic matter in the liquid fraction. Per addition of 200 L an amount of 7 kg organic matter is added to the pond corresponding with about 70 mg/L algae culture. For comparison the algae dry matter content in the pond is estimated at 100-150 mg/L at that moment. After the first day the harvest decreases, but remains higher than in the period preceding the experiment.

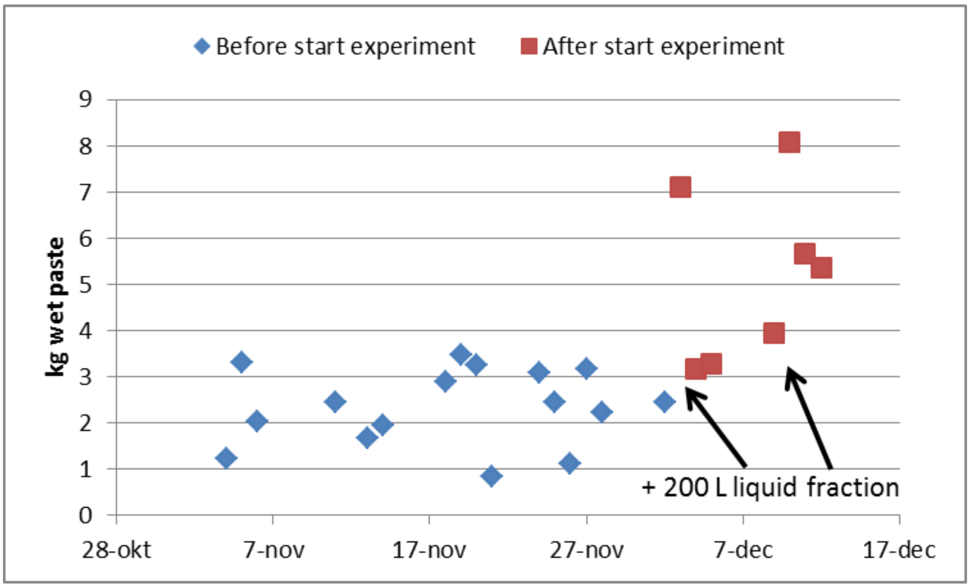

Figure 29. *Daily harvest (wet paste, kg) of the pond before and after the addition of the liquid fraction.*

The cell counts (Figure 30) show that after the addition of the liquid fraction a relatively strong increase of cell number was observed, however comparable fluctuations were also observed in the period before the addition of the liquid fraction.

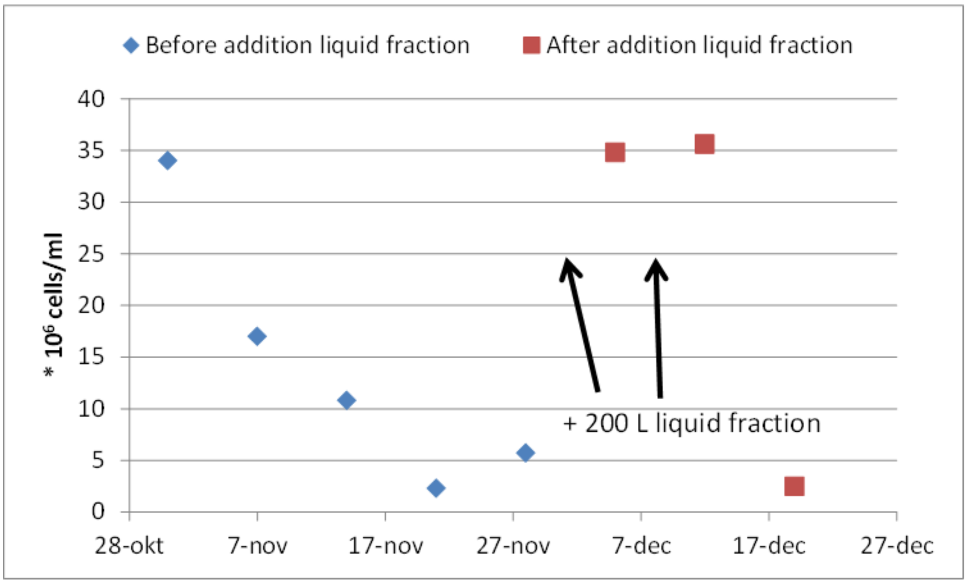

Figure 30. *Cell density (number/ml) of the outdoor pond before and after addition of the liquid fraction of digstate.*

Several studies have shown that microalgae can be grown on (liquid fractions of) manure or digestate (Van der Weide et al., 2014; Park et al. (2010); Uggetti et al., 2014). However, the contamination of the algae biomass with organic matter is a problem. Therefore, further tests are necessary with manure products lower in organic solids. If these products are available, a risk analysis has to be done in order to ensure that the concentrations of heavy metals and pathogens are below threshold levels, especially when the algae biomass is used as feed additive,

4.2 The indoor pond

4.2.1 Description harvesting system

In 2013 a harvesting installation based on dissolved air flotation (DAF) was installed (Figure 31). With this method it is expected that harvest costs can be decreased (Baros et al., 2015). In 2014 first tests were done using the algae culture of the indoor pond. Figure 32 gives a schematic overview of the harvesting system.

The algal culture was pumped to the DAF unit using a 0.6 kW pump with a flow of 3 m^3/h. To stimulate the flocculation a flocculant is used. Before the test a stock solution of the flocculant is made and this stock solution is injected into the algae culture with a pump of 0.25 kW. Subsequently, the algae-flocculant mixture flows through a pressure vessel (pvc pipe) in which the mixture is aerated and put under pressure (5-6 bar) by a 1.75 kW pump with an air intake of 3 L/min resulting in 0.2 m^3 of air injected per hour into the algal solution. In the flotation tank the pressure is released and micro air bubbles are created. Inside the flotation tank these bubbles cause algae flocs to float to the surface where a skimmer with a 0.25 kW motor collects the concentrated algae slurry. Subsequently, the collected slurry is centrifuged. The effluent water is returned to the algae pond.

Figure 31. Dissolved air flotation unit.

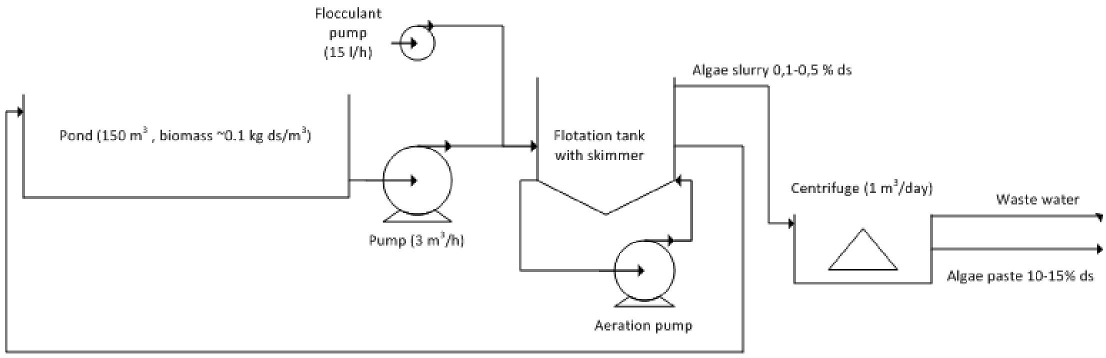

Figure 32. Schematic overview of harvesting system with dissolved air flotation unit.

4.2.2 Results harvest DAF unit

For an efficient harvest of the algae a flocculant is needed. For the selection of the flocculant two preconditions are important. Firstly, the flocculant should be feed grade as a main destination of our algae biomass is animal feed. Secondly, recycling of the effluent water should be possible in order to limit water use. The preconditions limit the choice of the flocculant. The following flocculants were selected for lab-scale testing:

1. BC-floc cationic	cationic polyacrylamide	Breustedt Chemie B.V.
2. Greenfloc 120	cationic starch	Hydra 2002 Ltd.
3. C-Bond HR35849	cationic starch	Cargill

From the above flocculants only BC-Floc is supplied with a GRAS (Generally Recognized As Safe) certification for use in feed production. However, this polyacrylamide may affect the algae growth when the effluent water is returned to the pond as it is not easily degraded biologically. That means it may accumulate in the growth culture and cause flocculation in the algae pond itself.

The two starch based flocculants are both biologically degraded easily. However, they are not GRAS certified for animal feed. The best prospects are for Greenfloc as it is intended and approved for drinking water purification. However, this does not necessarily mean that the products is safe for feed production. In the case of drinking water purification the flocculant ends up in the solid waste, while in the case of algae harvesting the flocculant ends up in the feed. C-bond has the least prospect for feed applications. The flocculant is intended for use in the paper industry and the MSDS (Material Safety Data Sheet) states: “Product which is not food compatible, intended only for technical use and which, if ingested, might present health risk for humans and animals “.

First, lab experiments were done in beaker glasses to get an indication of the algae recovery. The latter is based on the difference in optical density of the algae culture and the supernatant after sedimentation of the flocs. Figure 33 shows that BC floc gave the highest recovery, about 90% at a dose of 5 mg/l. The two starch based flocculants resulted in a lower recovery with Greenfloc 120 giving the best results, a recovery of circa 55% at a dose of 30 mg/l and higher. The flocculant C-Bond gave the lowest recovery, about 35% at 125 mg/l. For the starch based flocculants the required dose was considerably higher than for BC floc.

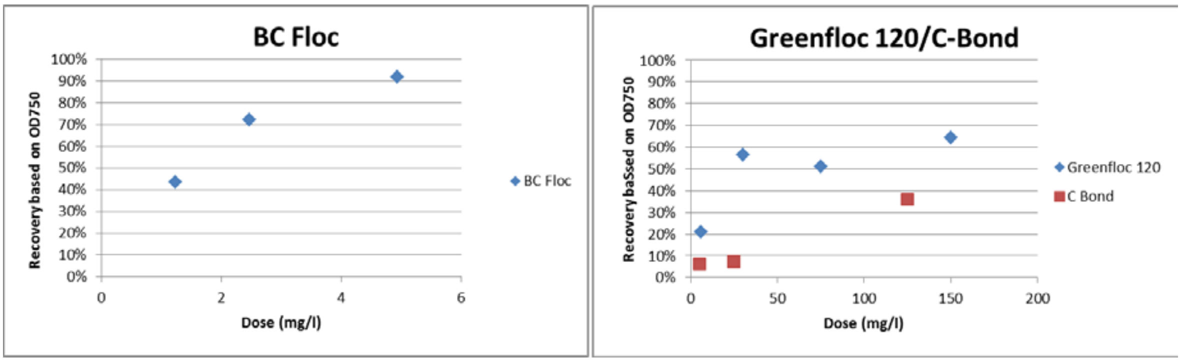

Figure 33. *Algae recovery (%, based on difference in optical density of the algae culture before and after addition of the flocculant) in relation to the dose of the flocculant BC floc (left) and Greenfloc/C-Bond (right).*

Based on the results of the lab experiments in August 2014 DAF unit tests were done for BC floc and Greenfloc 120. Figure 34 shows the results of tests done with BC floc. For different doses the algae recovery was assessed. This was done by measuring the OD of the influx of the DAF unit (algae culture of the indoor pond) and the outflow of the DAF unit (return flow to the algae pond). The algae culture flow through the DAF unit was 4.5 m^3/h for the test on August 20th. As there were some problems to maintain the water level in the flotation tank, for the tests on August 21th the flow was decreased. After the tests it turned out that the flow was lower than expected (1.8 m^3/h) and, therefore, the flocculant dose was higher than planned. The results show that the recovery is maximal for a dose of 1 g/m^3 onwards. In the lab experiments the algae recovery was lower, this is possibly due to the flotation effect.

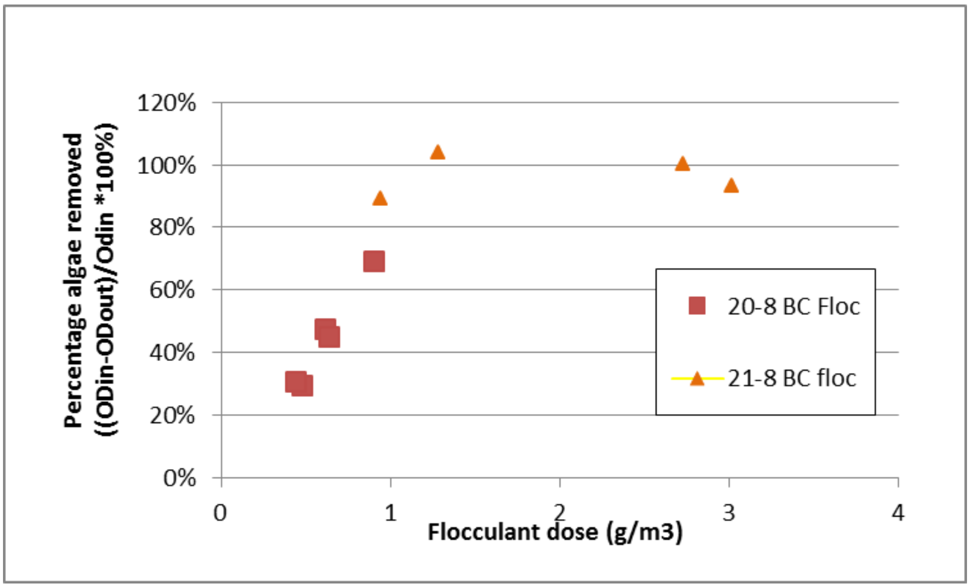

Figure 34. *Algae recovery (%, based on difference in optical density of the algae culture of the inflow and outflow of the DAF-unit) in relation to the dose of the flocculant BC floc.*

In October further tests are done with the flocculant Greenfloc 120. The results are shown in Figure 35. The maximum algae recovery, 70-80 %, is lower than for BC floc. Moreover, the variation in achieved recovery is higher for Greenfloc than for BC floc. The relationship between flocculant dose and algae recovery is less strong than for BC floc.

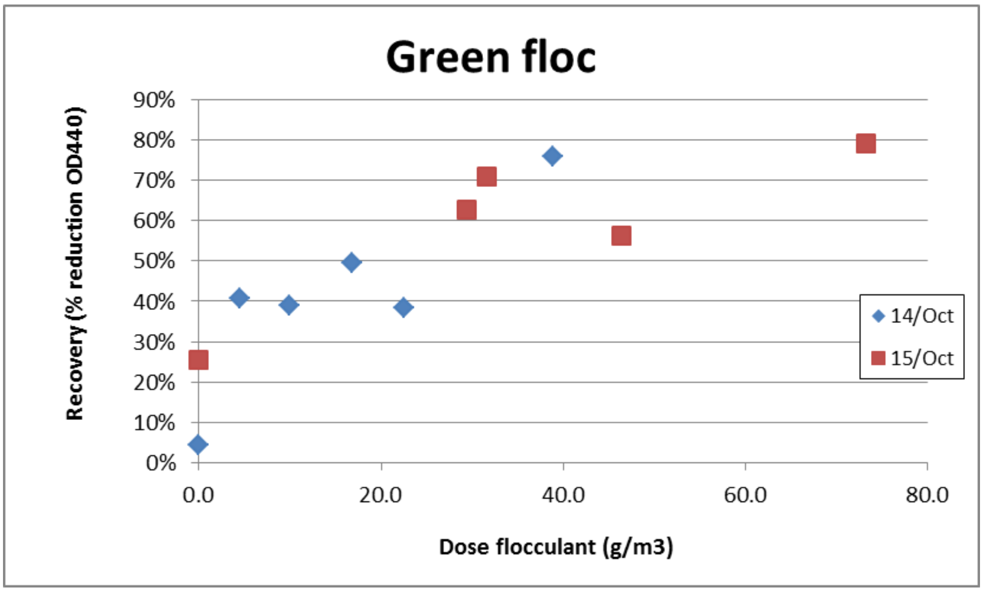

Figure 35. Algae recovery (%, based on difference in optical density of the algae culture of the inflow and outflow of the DAF-unit) in relation to the dose of the flocculant Green floc.

For the test with BC floc the effluent water of the DAF unit was returned to the algae pond. After three days of testing flocculation of the algae in the pond was observed indicating that the flocculant was affecting the algae culture. However, the algae culture also showed some autoflocculation. This was observed before the tests with Greenfloc were started. During the tests with Greenfloc the effluent water was not returned to the algae pond.

Table 6 shows a calculation for the costs of the flocculant based on the observed maximum algae recovery and the corresponding flocculant dose. The results show that the costs for BC floc are considerably lower than for Greenfloc.

Table 6. Cost calculation for the use of flocculants.

Flocculant	Dose (g/m^3)	Price (€/kg)	Algae density culture (g/m^3)	Recovery (%)	Costs €/m^3	Costs €/kg biomass
BC Floc	1	4.5	100	90	0.0045	0.05
Greenfloc	40	3.2	100	75	0.128	1.71

4.3 Heat consumption

The total heat consumption for the whole algae pilot in 2014 was 1115 GJ (Figure 36). In the periods January-beginning of June and beginning of September-December the daily heat consumption was 3.8 and 4.0 GJ/day, respectively. During summertime heat consumption was about 40% lower (2.3 GJ/day) mainly due to the fact that the indoor pond had reached its setpoint temperature of 25°C and needed no additional heating. The outdoor pond was heated almost throughout the whole year.

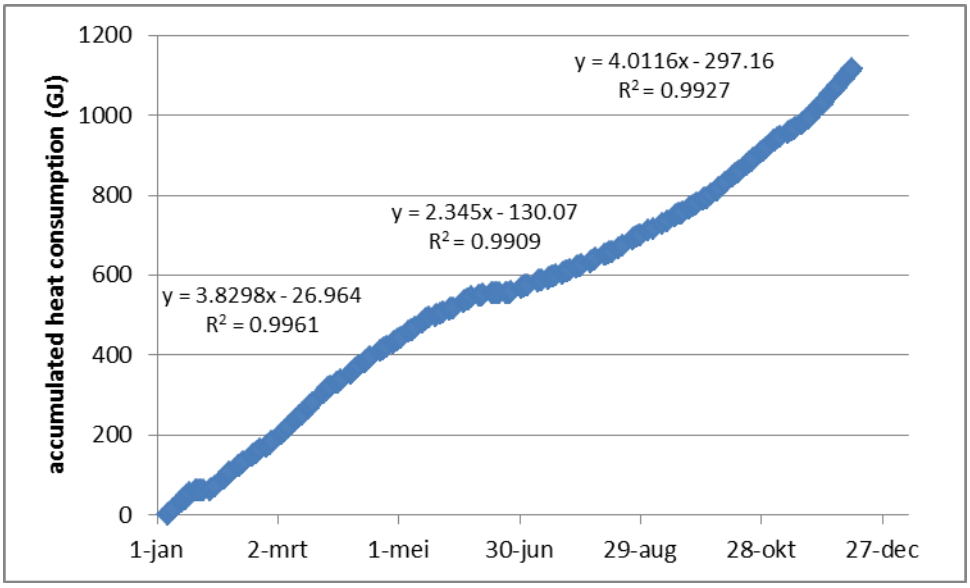

Figure 36. Cumulative heat consumption by the algae pilot facility in 2014.

4.4 Water consumption

The total water consumption of the whole algae pilot in 2014 was 2282 m^3. This includes water used for cleaning the harvest equipment and the water used for operating the LED supported photobioreactors.

A better insight in the water consumption of the separate algae ponds can be derived from the course of the water level of the ponds. For the indoor pond, any increase of the water level (or culture depth) is a direct result of water intake. Based on these increases a cumulative water addition of 1200 m^3 can be calculated. This is for a major part caused by cleanings and refilling (6 times) consuming about 950 m^3. The remaining 250 m^3 is used to compensate for water losses predominantly due to evaporation.

A similar calculation can be made for the outdoor pond. Compared to the indoor pond, additionally, also the rainfall has to be taken into account. The calculated cumulative water addition in 2014 amounted to 450 m^3. About 100 m^3 is explained by pond cleaning and refilling (1 time). The remaining amount of water, approximately 350 m^3, must have been the sum of water addition due to maintain the setpoint (compensation for evaporation) and rainfall (690 mm corresponding with 175 m^3).

In addition to the above calculated use of water for the both ponds, the grazer filter also used water for backflushing (this water is discharged to the sewage). This amount is, however, not recorded separately.

4.5 Conclusions

- The monitoring of the algae biomass production was only done in the outdoor pond. In the summertime a couple of periods with low production were observed. One serious crash occurred after which the pond was cleaned and refilled.
- The total annual algae production of the outdoor pond, harvested with centrifuges, was 7 ton dry matter per ha. In the first three months the harvest corresponded with an annual production of 15-17 ton dry matter per ha, thereafter the production decreased to al level corresponding with an annual production of 6 ton dry matter per ha.
- The grazer population fluctuated throughout the year but was highest in the summertime. The effectiveness of the installed grazer filter was low.
- Tests with the DAF unit showed that the algae biomass recovery ranged from 70% (flocculant Greenfloc) to 95% (flocculant BC floc). Returning the effluent of the DAF unit to the pond resulted in negative effects on the algae production (only tested with BC floc).
- Addition of liquid fraction to the outdoor pond resulted in a colouring of the culture already at dilutions <0.5%. Based on cell counts it could be concluded that algae growth was not negatively affected by the liquid fraction. However, the harvest data showed that the organic matter in the liquid fraction is simultaneously harvested with the algae biomass.
- The heat consumption of the total algae production system was 3.1 GJ/day, ranging from 3.8-4.0 GJ per day in the winter, spring and autumn to 2.3 GJ per day in the summer. The available excess heat of the CHP-unit allowed an increase of the culture temperature of about 5 °C above the average air temperature for the outdoor pond. For the indoor pond temperature was about 10 °C above the air temperature. The stronger effect for the indoor pond is due to the effect of the glasshouse.
- The total water consumption for the whole algae pilot was 2280 m^3. Based on changes in water level, the annual water consumption was estimated on about 1200 and 450 m^3 for the indoor and outdoor pond, respectively. The higher water consumption of the indoor pond was mainly due to a higher cleaning and refilling frequency.

5 Results pilot 2015

As in 2014 the outdoor pond was used for continued monitoring of the algae production (paragraph 5.1) while the indoor pond was used for additional experiments to test adjustments of the system (paragraph 5.2).

5.1 Outdoor pond

5.1.1 Description pond

A detailed description of the outdoor pond is given in paragraph 3.1. The following changes were made:

- Until September harvesting was done with three centrifuges instead of four centrifuges in 2014. One of the available four centrifuges was used for harvesting the indoor pond. These centrifuges were not self-unloading but are unloaded manually after 16 hours operation four times a week. From the beginning of September onwards the centrifuge from the bioethanol plant was added. Unlike the other three centrifuges this was a self-unloading centrifuge that was harvesting for 24 h/day five days a week.
- Till the end of October the pond was continuously sparged with air and flue gas was added to the sparged air when the pH of the culture increases above the setpoint level. Thereafter sparging was restricted to the periods with CO_2-demand (pH > setpoint).

5.1.2 Growing conditions

In Figure 37 the monthly sum of the global radiation is shown. In 2015 the total annual sum of the global radiation was 2.5% higher than the long term average due to higher radiation levels in April and June.

Figure 37 also shows the average air temperature and the culture temperature of the outdoor pond. The average air temperature was 0.7 °C higher than the long term average. Temperatures were especially higher in the months November and December. The average culture temperature was 5.8 °C higher than the average air temperature in 2015 due to the addition of excess heat of the CHP unit.

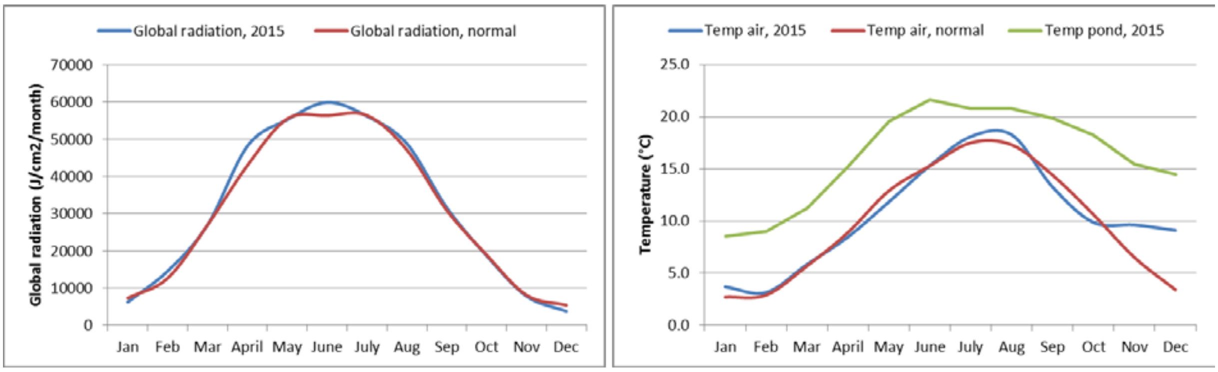

Figure 37. The global radiation and the air and culture temperature in 2015.

5.1.3 Biomass production

In Figure 38 the weekly harvest from the outdoor pilot pond is shown for 2015 together with the global radiation (weekly sum). Until the end of February the weekly harvest (with a single centrifuge in operation, only on working days) is low, approximately 1 kg algae biomass per week. The low production in week 7-8 was due to a technical problem with the pond which made a restart necessary. From beginning of March onwards we started harvesting with three centrifuges. In April the weekly harvested biomass increased to about 8-12 kg per week in April. In this period the centrifuges were harvested twice a day as tests showed that this improved the daily harvest (see also paragraph 5.1.7). In May and June problems arose with grazers affecting the production negatively. In the end it was necessary to empty and clean the pond. End of June the pond was restarted with new inoculum. However, beginning of July the pond crashed again, possibly as a result of high temperatures combined with a high grazer pressure. From the middle of July to the middle of August the pond was out of operation due to problems with the scaling up of the inoculum in the 350 L and 20 m^3 basins. However, in September and October the production was stable and on a relatively high level (6-8 kg dm/week). As mentioned before in this period the harvest was done with an extra centrifuge (from the bioethanol plant).

At the end of 2015 the outdoor pond produced a total of 141 kg dry weight algae biomass corresponding with about 5.6 ton dry matter per ha.

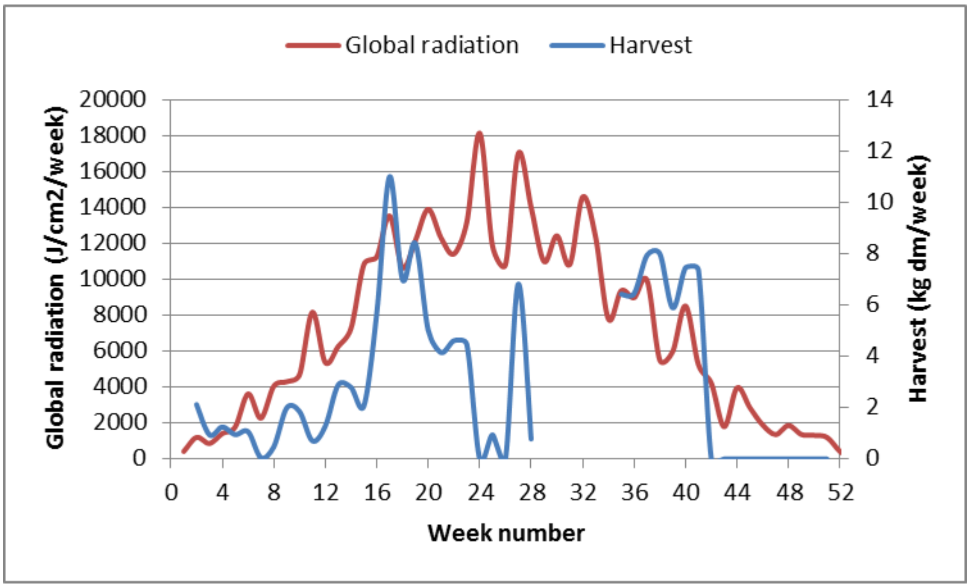

Figure 38. The weekly algae biomass yield and the global radiation (weekly sum, obtained from KNMI weather station Lelystad) of the outdoor pond in Lelystad in 2015).

As for 2013 and 2014, we compared the harvest data of 2015 with yield predictions of the open pond model. Assuming a photosynthetic efficiency (conversion of solar radiation energy in glucose energy) of 1.5% and an optimal culture temperature of 25 °C a biomass production of 20.7 ton per ha was predicted (Figure 39). The predicted biomass production using the actual culture temperature was 17.1 ton per ha. The actual yield level was lower. Until week 19 and from week 34 onwards the actual harvest corresponds with an annual yield level of about 11-12 ton dm/ha.

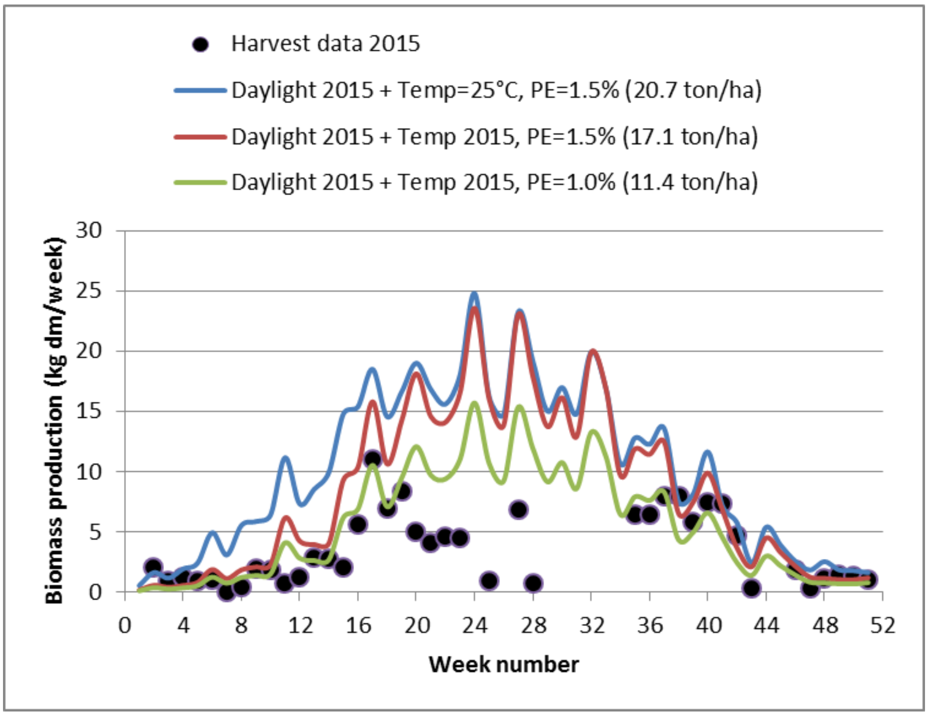

Figure 39. Harvest data from the outdoor pond in 2015 compared to model calculations (PE = photosynthetic efficiency, Temp =culture temperature).

5.1.4 Culture density / species

The culture density of the ponds is checked daily by measuring the optical density. In addition, the cell density and species composition is assessed once a week by microscope. Figure 40 shows the optical density combined with the depth of the culture (left) and combined with the biomass harvest (right). As observed in the other years in the spring the optical density increased, but this increase was earlier than the increase in dry matter production. This may be due to the fact that the harvesting was done twice a day from middle of April onwards. This resulted in a higher harvest compared to emptying the centrifuges once a day (see also 5.1.7). In May-July the optical density regularly decreased due to problems with grazers. After the restart in August the OD increased to levels also observed in the spring and then decreased from October onwards.

Till beginning of May the culture depth was kept at 40-50 cm except for the period May-June. In this period tests were planned with regard to monitoring the production at different water levels. However, the problems with the algae growth in this period confused the experiment. After a restart end of June the water level was increased to 70 cm in order to prevent too high culture temperatures as in the first week of July the temperatures were extremely high. After the restart in August a water level of 40-50 cm was maintained.

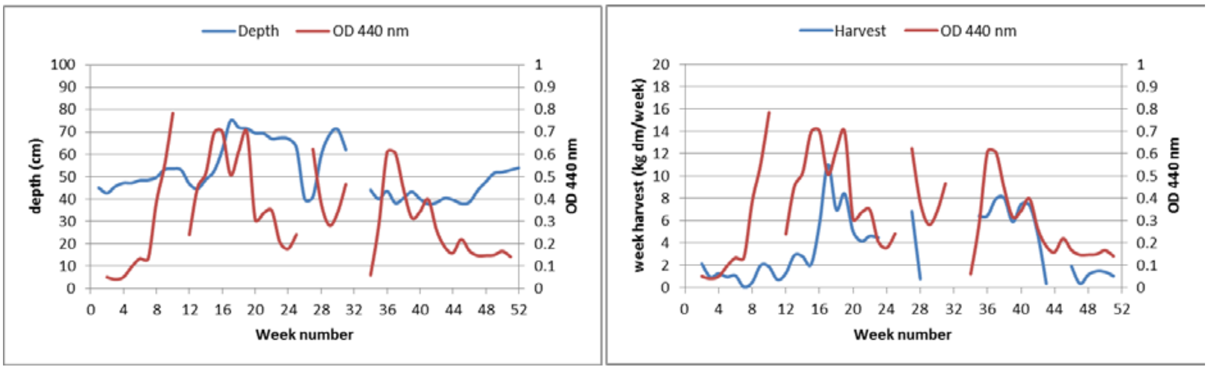

Figure 40. The depth and the optical density of the culture at 440 nm (left, weekly average) and the biomass harvest and the optical density of the culture (right, weekly average) of the outdoor pond in Lelystad in 2015.

In Figure 41A the cell density is given combined with the optical density of the culture, in Figure 41B the relationship between optical density and cell density is shown. The latter is done for two periods, January-June and July-December. In July-December the relationship between OD and cell density was weak, however, this was mainly due to one outlier point (OD = 0.6 while cell density is very low). If this point is excluded the slope of the regression lines of the two distinguished periods is comparable.

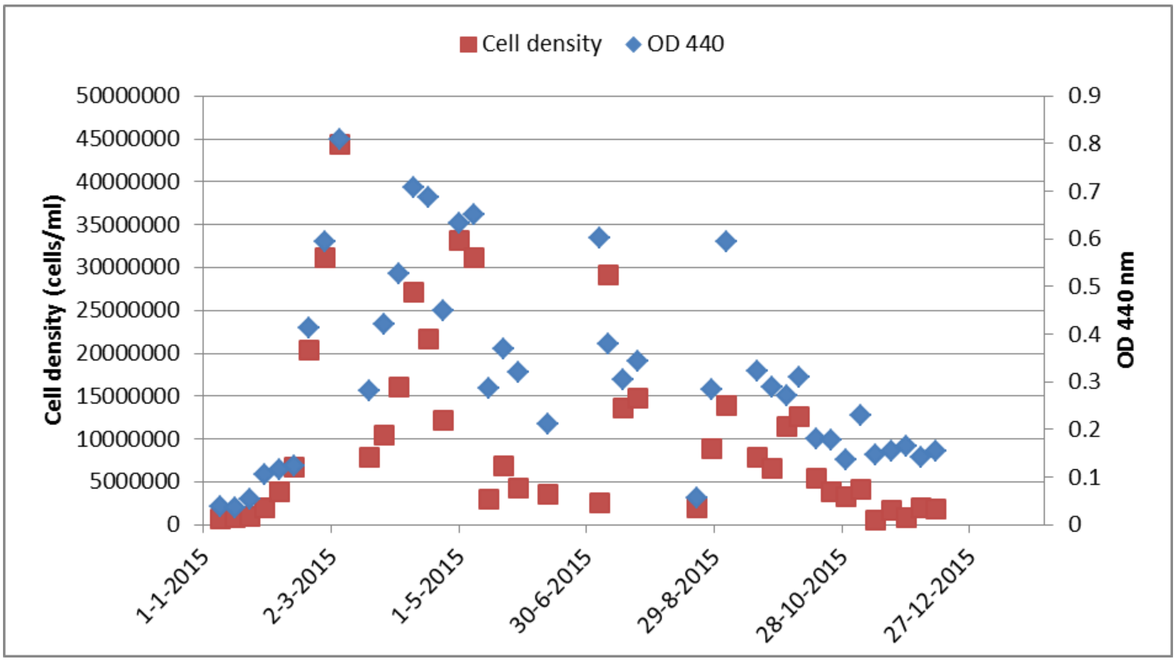

Figure 41A. Collected data on optical density at 440 nm and cell density for the outdoor pond in 2015.

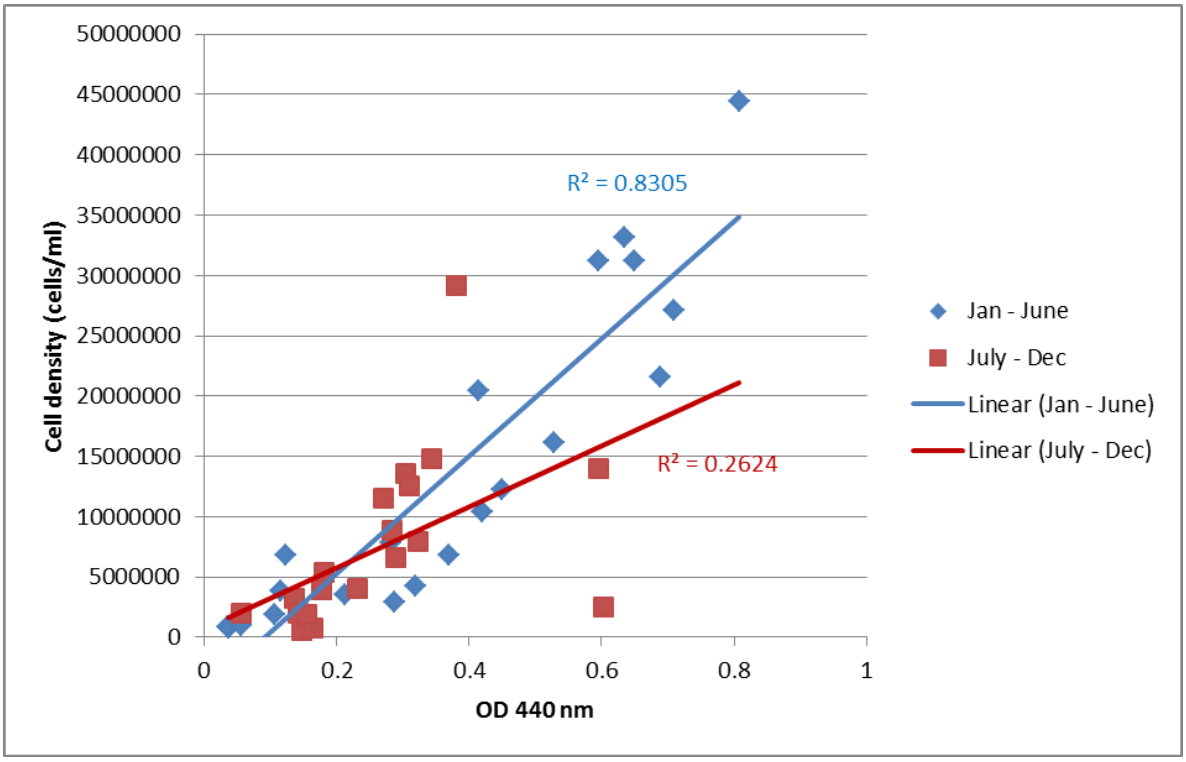

Figure 41B. Relationship between optical density at 440 nm and cell density for the outdoor pond in 2015.

In Figure 42 the microscopy counts are shown. The culture was dominated by small Chlorella species. In the spring and late summer also Scenedesmus species were observed.

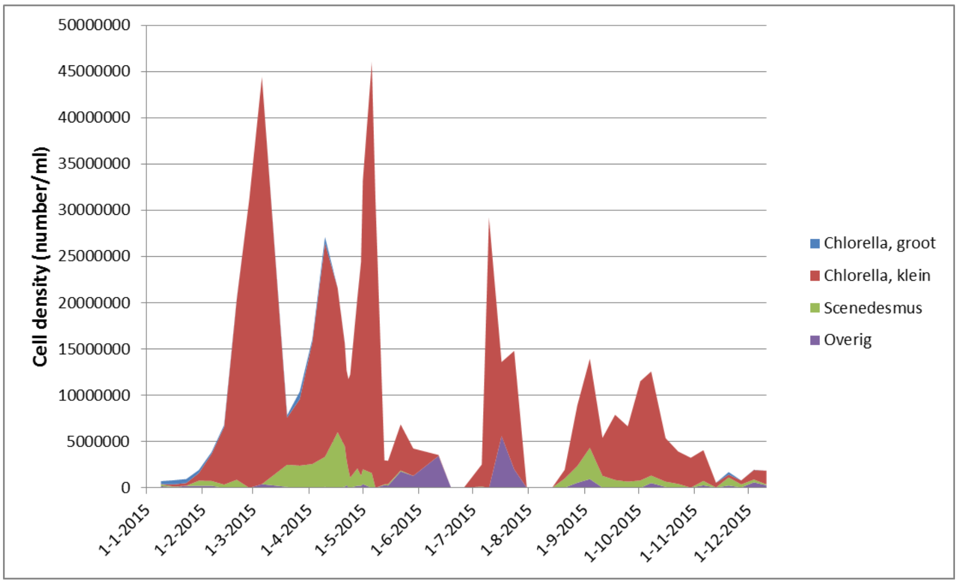

Figure 42. Species composition of the algae mix throughout 2015 as assessed by microscopy.

5.1.5 Predators

In 2013 a drum filter was installed in order to remove a part of the grazers. Figure 43 shows the total number of grazers in the inflow and outflow of the grazer filter. The course of the number of grazers in the pond during the year is characterized by large fluctuations The grazer pressure was highest in May and July with total number increasing to about 250,000/l. Generally, the grazer pressure in the summertime was higher than in autumn, winter and spring.

The efficacy of the grazer filter was low (Figure 43). Figure 44 shows that the size distribution of the found grazers did change slightly, the number of grazers found on an sieve of 75 µm was lower in the outflow.

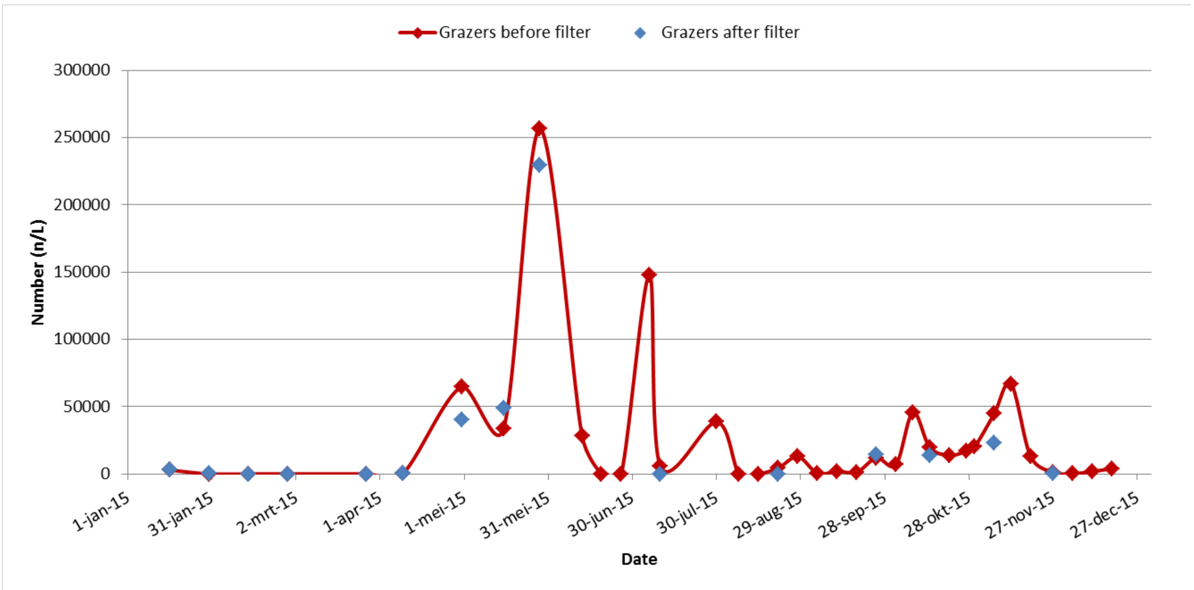

Figure 43. Total number of grazers in the inflow and return flow of the grazer filter of the outdoor pond in 2015.

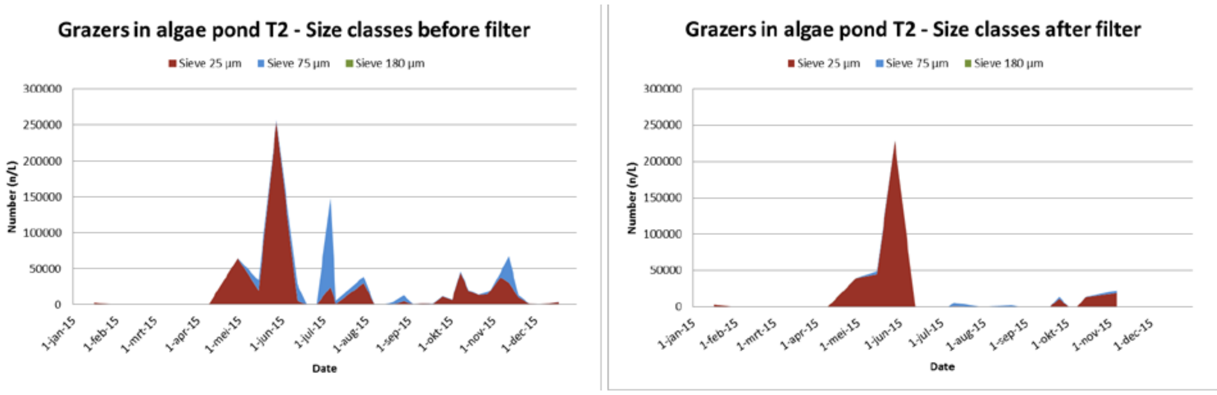

Figure 44. Total number of grazers in the different size classes for the inflow (left) and the return flow of the grazer filter (right) of the outdoor pond in 2015.

5.1.6 Harvest efficiency centrifuges

The centrifuges that are used for harvesting the algae were emptied after 24 hours of operation and were run with a flow rate of 750 L algae water/h (maximum value). In April a test was done to assess the effect of a lower flow rate and emptying the centrifuges twice instead of once a day.

The test was done with the four available centrifuges, three were used for harvesting the outdoor pond and one was used for harvesting the indoor pond. The standard harvesting settings were a flow rate of 750 L/h and emptying the centrifuge once a day after 24 hours of operation. For the outdoor pond this setting was compared with 1) emptying the centrifuge twice a day with a flow rate of 750 L/h and 2) emptying the centrifuge once a day and a flow rate of 375 L/h. For the indoor pond only one centrifuge was available and this centrifuge was run with the standard setting. The test was done for 24 hours starting in the morning after the emptying of the centrifuges. Emptying the centrifuges was done after 9 hours (only the centrifuge that was emptied twice a day) and 24 hours (all centrifuges).

As indicator of the harvest efficiency the reduction of the optical density was used. This reduction was calculated as follows:

Reduction OD (%) = $(OD_{outflow} - Od_{inflow})/OD_{inflow} * 100$

The results are shown in Figure 45. When emptied once a day the harvest efficiency decreased from around 80% to 20% for the indoor pond and from around 40-55% to zero or even negative values for the outdoor pond, especially at the lower flow rate. A negative value indicates that already recovered algae biomass in the centrifuge is washed out again. The decrease of the harvest efficiency was measured from 9 hours after the start of the experiment onwards. Initially, at the lower flow rate the harvest efficiency was higher than at the higher flow rate. When the centrifuge was emptied twice a day the harvest efficiency increased after the first emptying moment.

Generally, the harvest efficiency of the indoor pond was higher than for the outdoor pond due to differences in algae species composition. The indoor pond contained more bigger species (*Scenedesmus spp*) while the outdoor pond culture was dominated by smaller *Chlorella spp*.

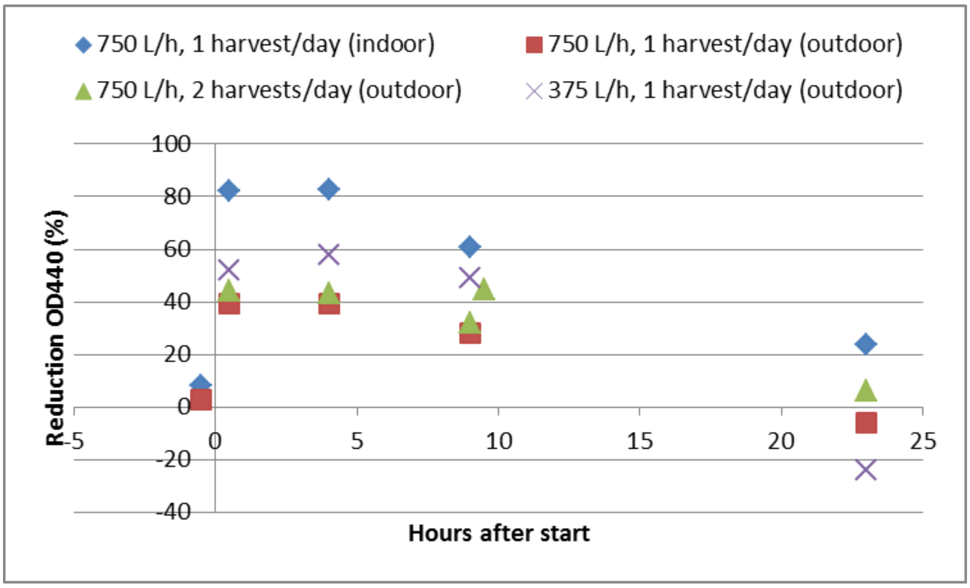

Figure 45. Effect of flow rate (L/h) and emptying frequency of the centrifuges of the indoor pond (1 centrifuge) and outdoor pond (3 centrifuges) on the reduction of the optical density (440 nm).

Based on these results it was decided that for a situation that the centrifuges were emptied once a day starting up was done at the end of the afternoon instead of directly after the harvest in the morning resulting in an operation period of about 16 hours instead of 24 hours.

As mentioned before in September an extra centrifuge was added to the outdoor pond. This was a self-unloading centrifuge (hereafter indicated as AUT) that was harvesting for 24 h/day at a flow rate of 500 L/h. The manually emptied centrifuges (hereafter indicated as MAN) were operating at a flow rate of 750 L/h for 16 hours/day.

The harvest efficiency (based on the reduction of the optical density) of the AUT centrifuge was about 95% while for the MAN centrifuges the harvest efficiency was about 70% directly after the start up. Based on earlier detailed measurements (Figure 45) it may be expected that the harvesting efficiency will decrease during the operation period of 16 hours (not measured in this period).

Table 7 shows the harvest of the centrifuges for week 40-42. This was a period of a good and stable pond performance. The MAN centrifuges produced a paste of about 19% dry matter while the AUT centrifuge produced a slurry of 1-3%. In week 40 and 41 the total dry matter harvest per centrifuge was higher for the AUT centrifuge while in week 42 the yield of the MAN centrifuge was slightly higher although differences were small. The lower yield of the AUT centrifuge in week 42 was due to al lower dry matter content of the harvested slurry.

Table 7. Harvest of the centrifuges (MAN = manually unloaded; AUT = self-unloading) of the outdoor pond in week 40-42.

Week	Wet (kg)		Dry matter-%		Dry matter (kg)		
	MAN	AUT	MAN	AUT	MAN	MAN	AUT
	n=3	n=1	n=3	n=1	n=3	n=1	n=1
40	25.2	102.6	18.5	2.7	4.66	1.55	2.80
41	25	103.8	19.3	2.4	4.82	1.61	2.54
42	19.3	106.1	18.8	1.1	3.62	1.21	1.13
Total	*23.2*	*104.2*	*18.8*	*2.1*	*4.37*	*1.46*	*2.16*

5.2 The indoor pond

In 2015 experiments were conducted with regard to the harvest system with the DAF unit (paragraph 5.2.1). Additionally, measurements were done with regard to the efficiency of the CO_2-addition to the pond (paragraph 5.2.2).

5.2.1 Results harvest DAF unit

In 2013 a harvesting installation based on dissolved air flotation (DAF) was installed. In paragraph 4.2.1 a detailed description is given. In 2014 first tests were done with two flocculants, BC floc and Greenfloc 120 with BC floc giving the best results with regard to biomass recovery. However, after recycling the effluent of the DAF-unit back to the algae pond, problems with flocculation of algae biomass in the pond were observed. In 2015 a second test was done.

The test was carried out from 9-13 November with the flocculant BC floc. The effluent from the DAF unit was returned to the indoor pond. First, on 9 November a dose response test was done in order to assess the effectiveness of the flocculation. Three doses of BC floc (1, 2 and 4 g/m^3) were tested for one hour of operation. The effectiveness of the DAF unit was measured via the optical density of the inflow and the outflow of the DAF unit at the end of each test period. Figure 46 shows the results. Unlike earlier experiences a substantial difference in efficiency between 440 and 695 nm was observed. This may be due to the presence of the algae species *Micractinum*. In the weeks before the test a bigger difference between OD 440 and 695 was observed compared to measurements earlier in the year or compared to measurements in the outdoor pond. Based on the measurement at 440 nm the harvest efficiency increased from 20% at a flocculant dose of 1 g/m^3 to about 60% at a dose of 4 g/m^3. For the measurement at 695 nm the harvest efficiency ranged from 50-100%.

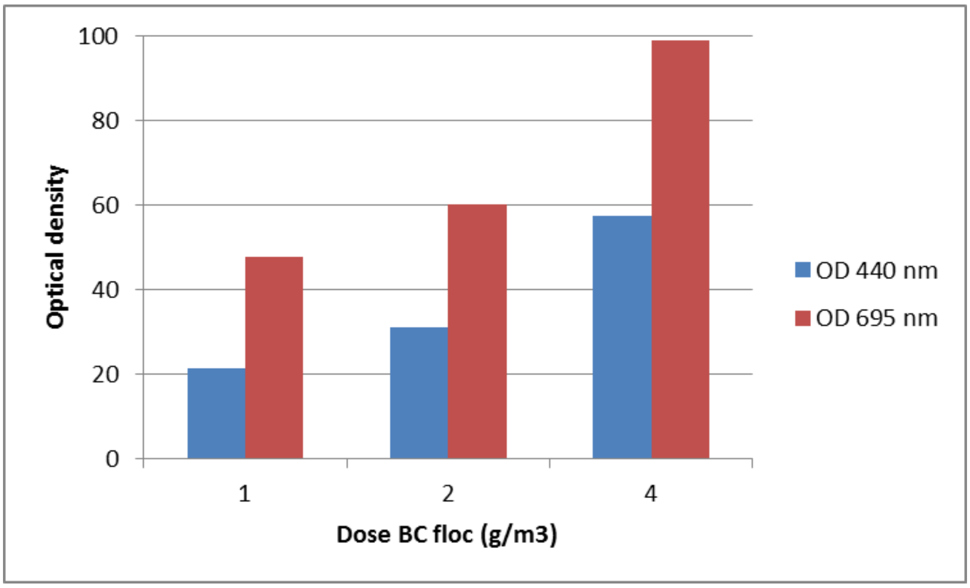

*Figure 46. The reduction of optical density (%, ODinflow-ODoutlfow/ODinflow*100%) as affected by the dose of the flocculant BC floc (g/m^3) for the indoor pond algae culture.*

Subsequently, on 10-13 November the DAF unit was run daily between 9:00 and 17:00 o'clock at a flocculant dose of 1 g/m^3 and a flow rate of 2 m^3/h. The effluent of the DAF unit was returned to the pond.

Figure 47 shows the optical density (440 and 695 nm) in November and December. The OD 440 nm decreased from the beginning of November onwards. This decrease will mainly be due to a decreased radiation level. The return of the effluent of the DAF unit did not seem to have a disrupting effect on the general pattern. The OD 695 nm was stable till 20 November and started to decrease afterwards. The difference in optical density between 440 and 695 nm decreased coinciding with a disappearing of the algae species *Micractinum*.

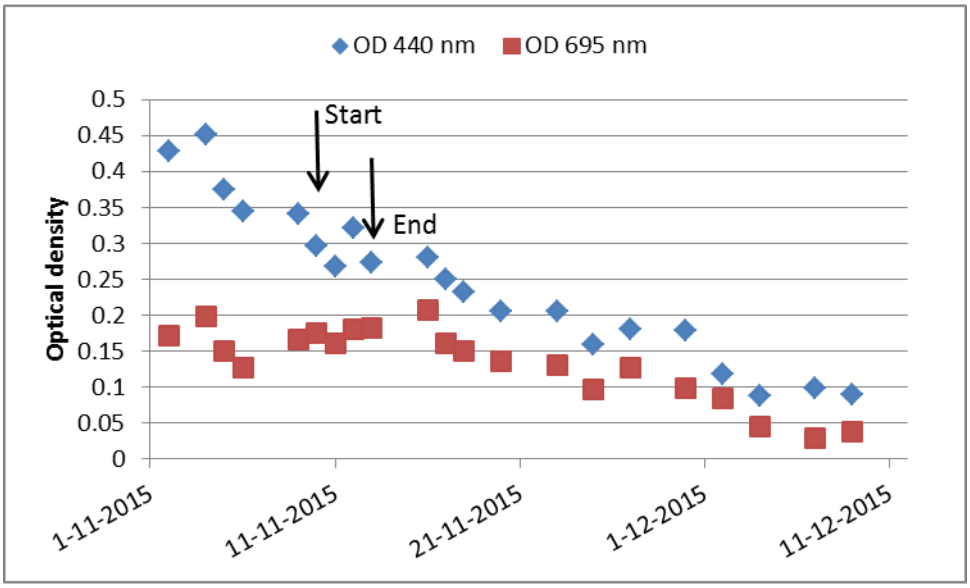

Figure 47. The optical density at 440 and 695 nm of the indoor pond in November-December 2015 (start/end = start/end of the DAF experiment).

5.2.2 Sedimentation tests

In addition to the tests with the DAF unit also the efficiency of sedimentation in combination with an increased pH and flocculant was tested on the algae culture of the indoor pond.

First on a lab scale sedimentation tests were done in order to assess the effect of pH and flocculant dose on the sedimentation. The tests were done in beakers filled with 500 ml algae culture. After increasing the pH with sodium hydroxide and/or adding the flocculant, the culture was intensively stirred for 15 min. Thereafter the sedimentation was measured after 15 min by measuring the optical density of the supernatant. Figure 48 shows the decrease in optical density for the different treatments. Without increasing the pH and without adding a flocculant, the OD decrease was about 25%. Increasing the pH improved the sedimentation to about 70% at a pH of 12. Adding a flocculant improves the sedimentation further especially al lower pH levels.

Additionally, larger scale tests were done with a settler tank of 360 L. The tests were done at a pH of 11. The results are given in Figure 49. At the lowest flocculant dose (1 mg BC floc/l) the sedimentation efficiency was 35-45%. Increasing the dose to 2 and 4 mg BC floc/l increased the sedimentation efficiency to 50-70% and 85%, respectively.

The results show that increasing the pH improved the harvest efficiency. In literature, also positive effects of increased pH on the flocculation of algae biomass are mentioned (Wu et al., 2012; Vandamme et al., 2012). Mentioned mechanism behind the increased flocculation is an interaction of the algae biomass with precipitations of magnesium and calcium salts.

In our experiments a relatively high pH (11-12) was necessary to realize a high harvest efficiency. This means that high amounts of caustic soda are necessary while after the harvest acid is necessary to de decrease the pH to a level of about 8. A part of the required pH decrease is expected to be realized by the CO_2 sparging of the pond.

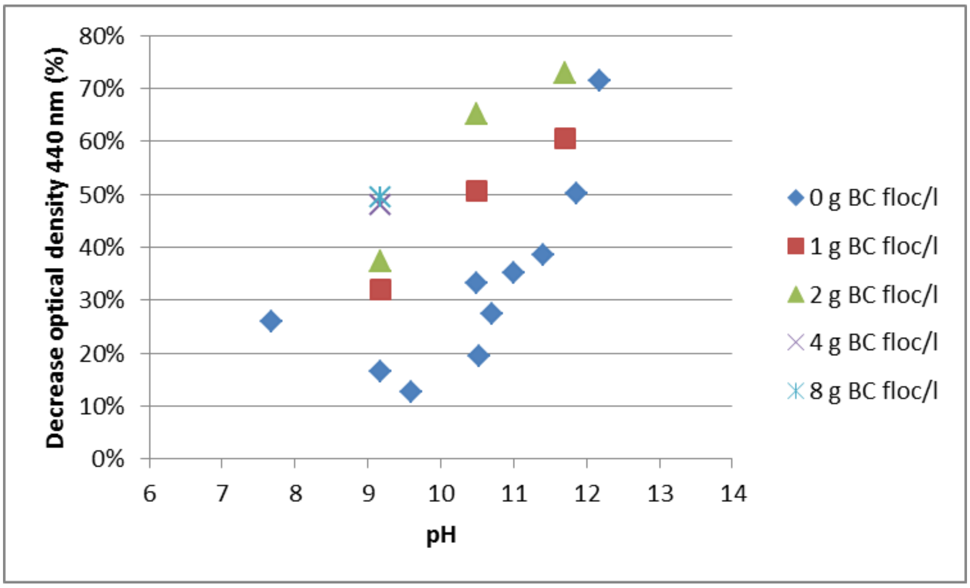

Figure 48. *Effects of pH increase and flocculant (BC floc) dose on the sedimentation efficiency(reduction of optical density 440 nm) in a lab scale experiment (beakers of 500 ml).*

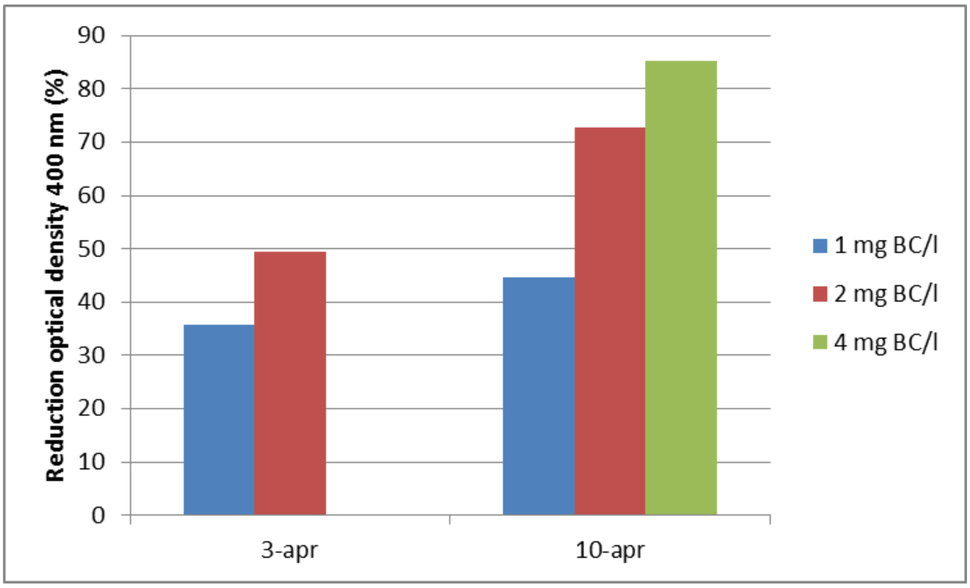

Figure 49. *Effects of flocculant (BC floc) dose on the sedimentation efficiency (reduction of optical density 440 nm) at a pH of 11 in the settler tank of 360 l.*

5.2.3 Efficiency CO_2-addition

Currently, the CO_2 addition in the ponds is done by injection of flue gas in the continuous airstream that is sparged into the pond via perforated tubes at the bottom of the pond. The flue gas addition is regulated based on the pH level of the pond. In order to assess the efficiency of the CO_2 addition measurements were done of the CO_2 concentration in the air bubbles that leave the pond and the CO_2 concentration of the pond culture.

Figure 50 shows the measurement set up. The CO_2 measurements were done in two buckets that were conversely placed with the open side just below the water level of the pond. One bucket (1) is placed above the air bubbles that leave the pond at the water surface. This is an open bucket. The air is leaving the bucket via a tube in which a CO_2 probe is placed that measures the CO_2 concentration in the air. The second bucket (2) is placed above the water surface on a position without air bubbles. In the headspace of this closed bucket the CO_2-concentration of the air is measured. Subsequently, the dissolved CO_2-concentration in the algae pond can be calculated with Henry's Law that states that the concentration of the dissolved gas in the water is proportional to the partial pressure of the gas in the air above the water. Both CO_2-probes and the temperature are continuously logged.

$$C_{water} = K * P$$

C_{water} = concentration gas in water (mol/l)

K = Henry's law constant (mol/L.atm)

P = partial pressure gas (atm)

For CO_2 at a temperature of 25 °C (298.15 K) the value for K is 0.034 mol/L.atm. For lower or higher temperatures the K can be calculated with:

$$K_{act} = K_{stand} * e^{(2400*(1/Tact-1/298.15))}$$

K_{act} = Henry's law constant at the actual temperature (K)

K_{stand} = Henry's law constant at the standard temperature (298.15 °C)

T_{act} = actual temperature (K)

The CO_2 concentrations and the temperature of the algae culture and in the headspace of the closed bucket are continuously logged.

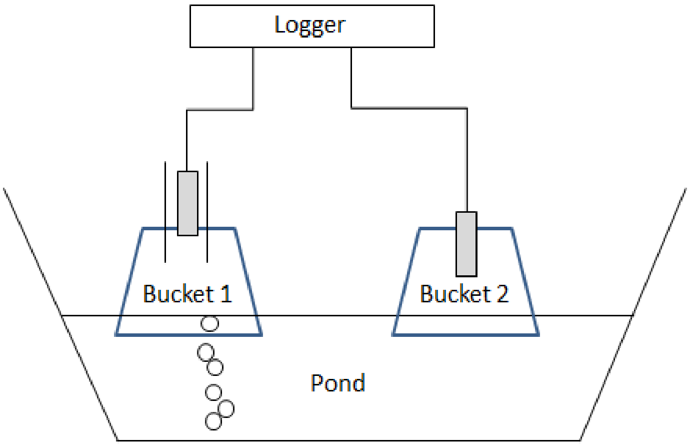

Figure 50. Setup of the CO_2 measurements.

In Figure 51 the CO_2-concentrations of the two sensors are given. In the period 8 October - 14 October air sparging was done continuously and, depending on the pH of the pond culture, flue gas was injected in the airstream. This means that the input flow to the pond consisted of a mix of air and flue gas. The measurements were started at a pond water level of 40 cm, and the level was increased to 60 and 80 cm on 9 and 12 October, respectively. At the moment flue gas was added the CO_2-concentration in the bubbles leaving the pond increased to 15000-30000 ppm. The CO_2-concentration of the inflow to the pond could not be measured as the concentration was higher than the measuring range of the available probes. Therefore, the CO_2-recovery in the pond could not be assessed. However, the high concentrations in the bubbles indicate that the recovery of CO_2 in the water was relatively low. At a depth of 40 cm the CO_2-concentration was lower than at a depth of 60 and 80 cm. This may be an indication that the CO_2-recovery may have been higher at a higher depth due to a longer contact of bubbles and water.

The CO_2-concentration in the closed headspace is an indicator of the dissolved CO_2 in the water. The figure shows that the concentration fluctuates. In the first three days the highest concentrations were measured at night time coinciding with the period of CO_2-production instead of CO_2-consumption. However, in the following days this pattern is less clear. Maximum measured values ranged from 3000-5000 ppm CO_2 in the headspace corresponding with 4.5-8 mg dissolved CO_2/l (according Henry's law).

From 14 October onwards air sparging was restricted to the periods when flue gas was added. This resulted in an more stable CO_2-concentration in the pond (Figure closed headspace). In the closed headspace the CO_2-concentration stabilized at about 4000 ppm corresponding with about 6 mg dissolved CO_2/l.

Figure 52 shows the percentage of the time that flue gas was added to the pond in the period September-November. Till the beginning of November, when a mix of air and flue gas was added, on average about 35% of the time flue gas addition was on resulting in an energy reduction of 65% for the air sparging. As the reduced air sparging did not result in significant effects on the algae growth or other.

problems as sedimentation of algae in the pond, this intermittent sparging regime was applied for both ponds.

From the beginning of November onwards 100% flue gas was added. This reduced the time flue gas addition was on to about 20% indicating that further reduction of energy consumption for air sparging may be possible assuming that no negative effects on the algae production will occur.

It may be expected that the time the flue gas addition is on, will depend on the production level of the pond. In Figure xxx the relationship is given between the daily global radiation and the percentage of the time the flue gas addition is on. This done for the period September-October when a mix of flue gas and air was added. The found relationship is weak (R2 = 0.27). This may partly be due to fluctuations in CO2-concentration in the flue gas (as indicated by fluctuations in CO2-concentration of the air in the bubbles leaving the pond).

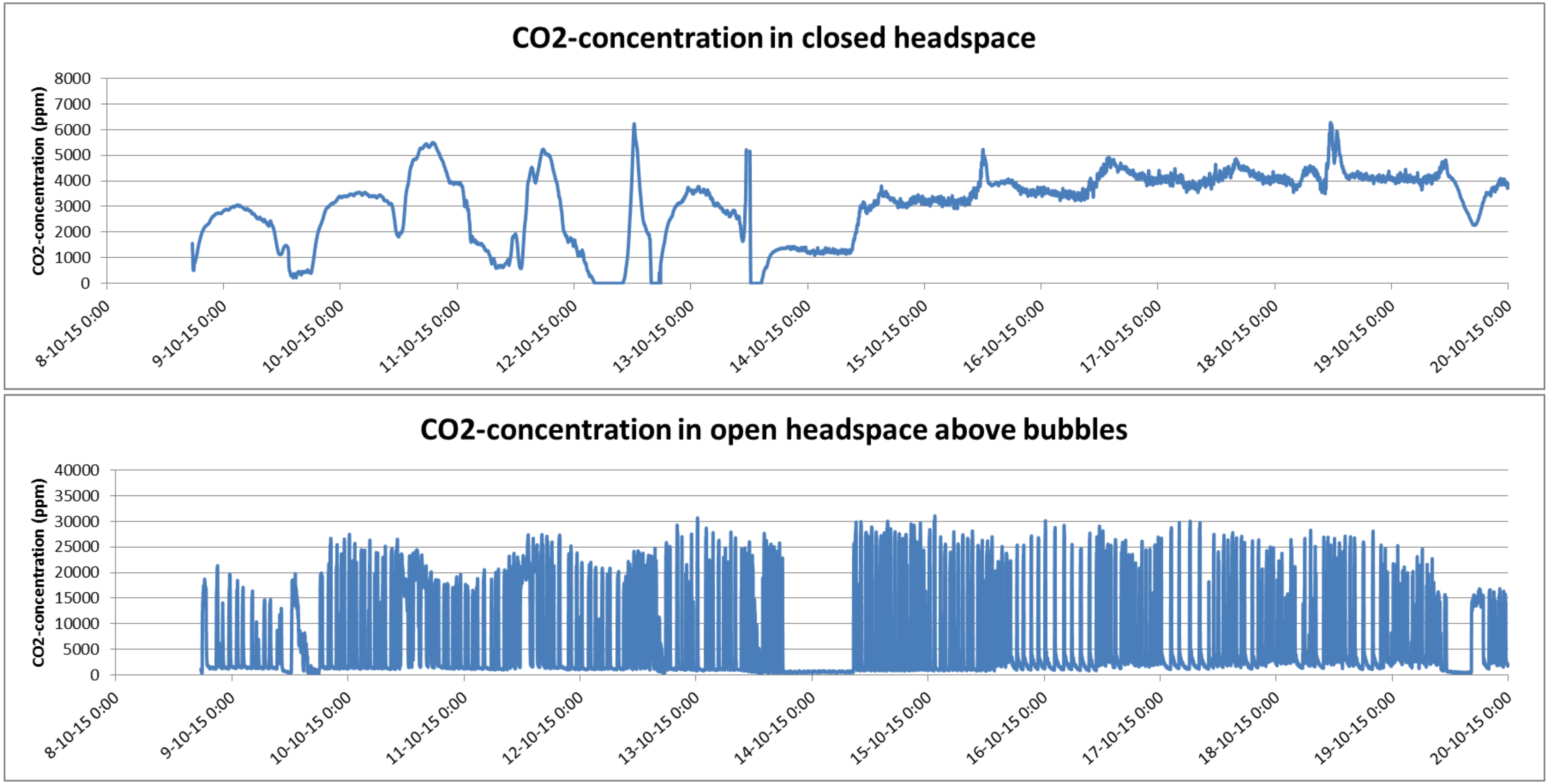

Figure 51. *CO_2-concentration in closed headspace and in open headspace above bubbles, measurements done in the indoor pond in October 2015.*

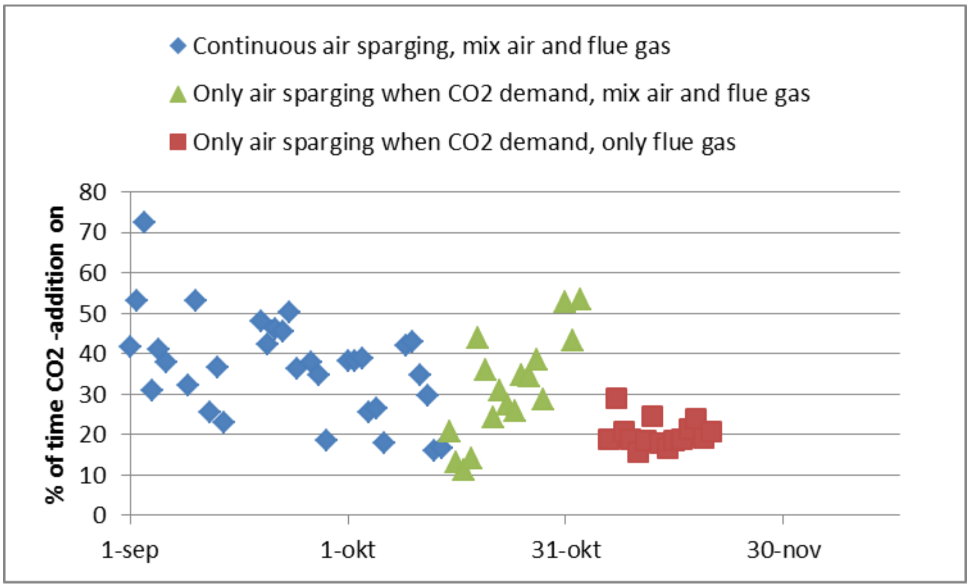

Figure 52. *The percentage of the time the flue gas addition was in operation as affected by sparging and flue gas addition regime in the period September-November 2015.*

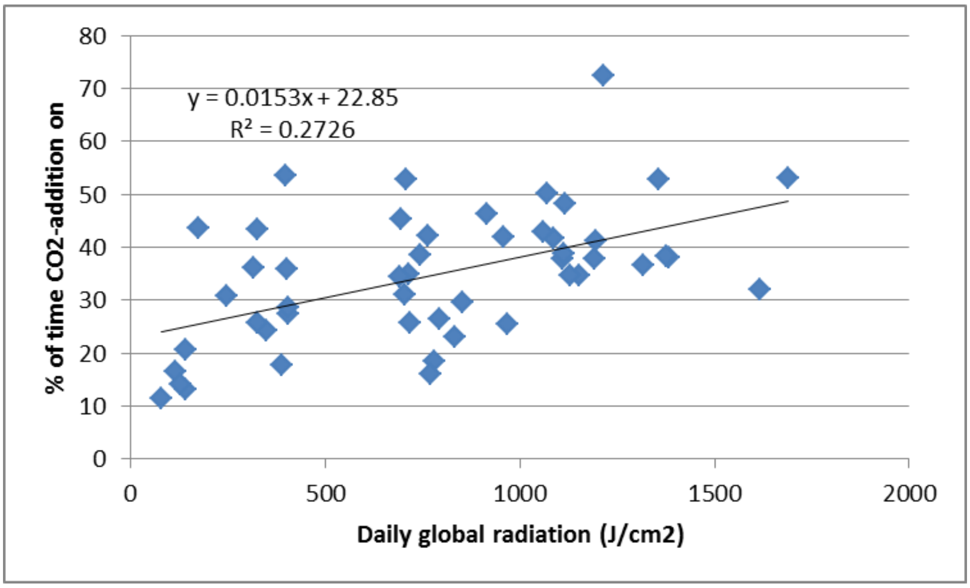

Figure 53. *The relationship between the daily global radiation (J/cm^2) and the percentage of the time the flue gas addition was in operation in the period September-October 2015 (in periods with CO_2 demand a mix of air and flue gas was added).*

5.3 Heat consumption

The total heat consumption for the whole algae pilot in 2015 was 1448 GJ (Figure 54). In the periods January-middle of June and beginning of September-December the daily heat consumption was 4.4 and 5.5 GJ/day, respectively. During summertime heat consumption was about 70-75% lower (1.3 GJ/day) mainly due to the fact that the indoor pond had reached its setpoint temperature of 25°C and needed no additional heating. The outdoor pond was heated almost throughout the whole year except for some periods with high temperatures in July.

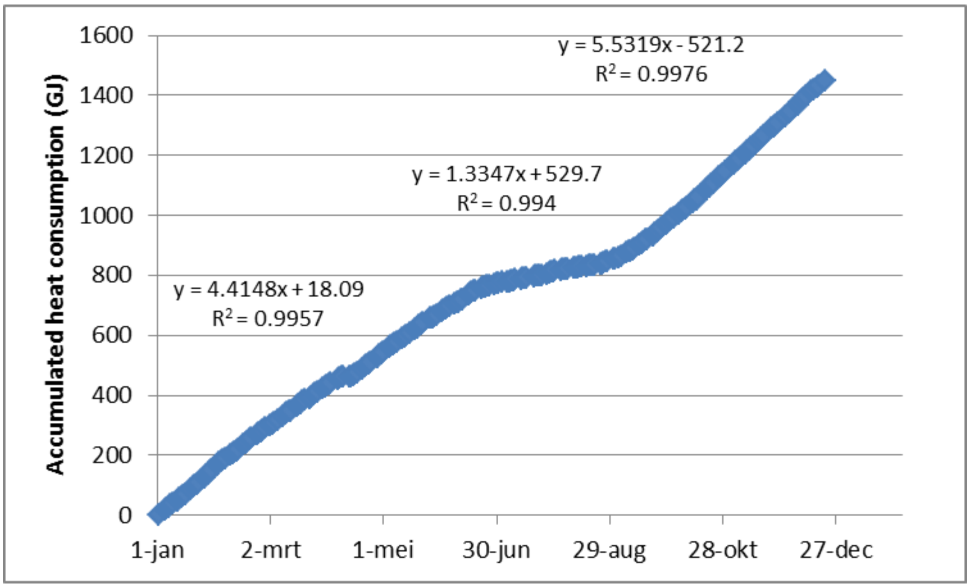

Figure 54. Cumulative heat consumption by the algae pilot facility in 2015.

5.4 Water consumption

The total water consumption of the whole algae pilot in 2015 was 2090 m^3. This includes water used for cleaning the harvest equipment and the water used for operating the LED supported photobioreactors.

A better insight in the water consumption of the separate algae ponds can be derived from the course of the water level of the ponds. For the indoor pond, any increase of the water level (or culture depth) is a direct result of water intake. Based on these increases a cumulative water addition of 755 m^3 can be calculated. About 520 m^3 is used for cleanings and refillings and increasing the setpoint for the water level (from 40 to 70/80 cm in April, July and September). The remaining 235 m^3 is used to compensate for water losses predominantly due to evaporation. In addition to the calculated use of 755 m^3, the grazer filter used 305 m^3 for backflushing (measured by a separate meter, this water is discharged to the sewage). This gives a total calculated annual use of 1060 m^3.

A similar calculation can be made for the outdoor pond. Compared to the indoor pond, additionally, also the rainfall has to be taken into account. The calculated cumulative water addition in 2015 amounted to 845 m^3. About 530 m^3 is explained by pond cleaning and refilling and water needed to increase the setpoint for the water level of the pond. The remaining amount of water, approximately 315 m^3, must have been the sum of water addition due to maintain the setpoint (compensation for evaporation) and rainfall (725 mm corresponding with 180 m^3). In addition to the calculated use of 845 m^3, the grazer filter used 145 m^3 for backflushing. This gives a total calculated annual use of 990 m^3.

5.5 Conclusions

- The monitoring of the algae biomass production was only done in the outdoor pond. A relatively stable algae production was observed in the spring and autumn. In the summertime a couple of crashes occurred.
- The total annual algae production of the outdoor pond, harvested with centrifuges, was 5.6 ton dry matter per ha. In the periods with stable production (spring, late summer and autumn) the harvest corresponded with an annual production of 11-12 ton dry matter per ha.
- The grazer population fluctuated throughout the year but was generally highest in the summertime. The effectiveness of the installed grazer filter was low.
- Repeating the test with the DAF unit with BC floc as flocculant showed that compared to the tests in 2014 a higher dose was needed to realize an algae recovery > 90% (4 g/m^3 in 2015 vs 1 g/m^3 in 2014). Apparently, the species composition or the condition of the algae culture affect the flocculation efficiency. Unlike 2014 returning the effluent of the DAF unit to the pond did not result in disrupting effects on the algae production.
- Tests with a settler tank of 360 L showed that sedimentation could be improved by increasing the pH and adding a flocculant. At a pH of 11 the harvest efficiency could be increased to 35-45%, 50-70% and 85% at a flocculant dose of 1, 2 and 4 mg BC floc/l, respectively.
- Measurement of the CO_2 concentration in the bubbles leaving the pond indicate that the recovery of CO_2 in the water was relatively low probably due to the relatively big air bubbles that are created resulting in a low gas exchange contact area with the water. When air sparging was restricted to the periods when flue gas was added, a more stable CO_2-concentration in the pond was found. Moreover, this sparging regime decreased energy demand with about 65% when a mix of flue gas and air was applied and 80% when 100% flue gas was applied. As the reduced air sparging did not result in significant effects on the algae growth or other problems as sedimentation of algae in the pond, this intermittent sparging regime was applied for both ponds.
- The heat consumption of the total algae production system was 4.0 GJ/day, ranging from 4.4-5.5 GJ per day in the winter, spring and autumn to 1.3 GJ per day in the summer. The available excess heat of the CHP-unit allowed an increase of the culture temperature of about 6 °C above the average air temperature for the outdoor pond.
- The total water consumption for the whole algae pilot was 2090 m^3. About 50% is used for refilling after cleaning and increasing the setpoint for the water level and about 20% is used for backflushing the grazerfilter.

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