Biomass Refinery and Process Dynamics Group

Thesis Biomass Refinery and Process Dynamics

A model based analysis of the downstream processing of microalgae biomass into proteins

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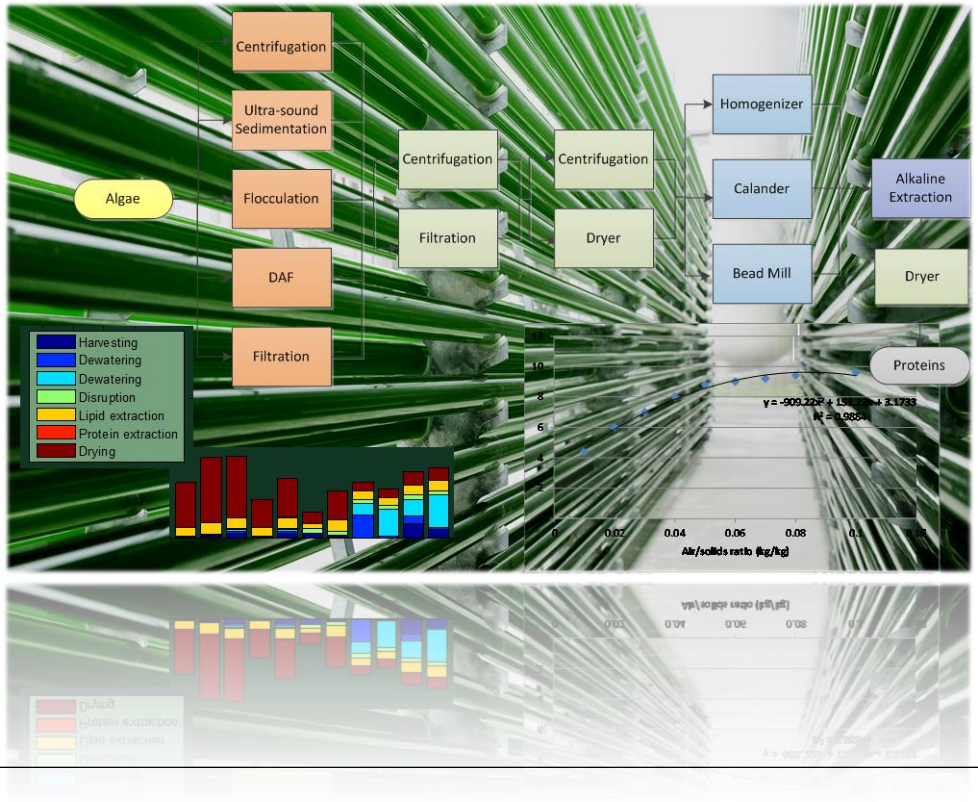

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A model based analysis of the downstream processing of microalgae biomass into proteins

Master Thesis

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Abstract

Extracting proteins from microalgae is a way to provide the world with a new protein source and to prevent insufficient protein supply in the future. Unfortunately the downstream processing of a diluted microalgae biomass is energy intensive. Also it is not known which combination of process units for the downstream processing requires the lowest ratio of energy requirements to protein yield.

In this project four steps are considered: harvesting, dewatering, disruption and extraction. Each of the four steps consists of a selection of process units. The process units are modelled by using overall mass and energy balances. The process models are organised in a superstructure to evaluate all possible combinations of process units. A model-based combinatorial approach is used to derive the energy requirements and protein yield during the downstream processing of all routes. The models used consist of flexible operating conditions, when these models are optimized, it provides insight about optimal process conditions. In this work a distinction is made between 'wet' and 'dry' processing. During 'wet' processing the microalgae stream concentration has a maximum concentration of 150 $^{kg}/_{m3}$ when it enters the disruption step. 'Dry' processing occurs when the concentration of the microalgae stream when it is being disrupted is at least 200 $^{kg}/_{m3}$. The final product has in all cases an end concentration of 700 $^{kg}/_{m}$, which is reached due to a drying step. The results show that dry processing is energetically more favourable than wet processing. During dry processing the harvesting and dewatering steps are more extensive, and less drying is needed. Drying requires a lot of energy. Furthermore, a distinction is made between the extraction of proteins and the extraction of proteins and lipids together. The results show that it is favourable to extract both proteins and lipids. By extracting lipids as well, the protein recovery is lowered, however the combined recovery of proteins and lipids is higher. Lipid recovery requires energy, which increase the total energy use. By extracting both proteins and lipids, the ratio of energy use to product recovery is lower.

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1 Introduction

The earth's population is growing exponentially and the conventional protein sources, like meat fish and soy, cannot be produced at this speed. It is predicted that in the future there will be an insufficient protein supply due to this massive increase in population (Becker 2007). An average adult male needs around 50 g of protein each day, and insufficient protein uptake causes severe malnutrition. Therefore, in the early 1950's the search for alternative and unconventional protein sources began.

Algae biomass appears as a good alternative to the conventional protein sources (Spolaore, Joannis-Cassan et al. 2006). The United Nations World Food Conference of 1974 declared the algae *Spirulina* as 'the best food for the future' and a promising tool to prevent future malnutrition due to protein deficiency (Chacón-Lee and González-Mariño 2010). Algae are a very large group of microscopic, photosynthetic organisms. Most species require little input to grow: they only need sunlight, carbon dioxide, water and nitrogen rich nutrients. During growth algae are capable to fix the carbon dioxide in organic compounds using the energy from the sun. Due to the simple form of organization, microalgae can reach very high growth rates (Williams and Laurens 2010). The organic compounds, like proteins, can be up to 70% of the total dry weight of some algae species (Becker 2007).

Becker found that the amino acid pattern of almost all algae is favourable to the conventional plant proteins (Becker 2007). Since algae are capable of synthesizing all amino acids, algae are a very good source for essential amino acids. Algae are also a favourable crop, since more biomass per unit of time is produced than with any other food crop known (Kay 1991).

The process to obtain proteins from microalgae biomass consist of a few steps: harvesting, dewatering, disruption and extraction. These steps are called the downstream processing (DSP) chain from algae to protein. The DSP begins with harvesting algae from a diluted ($5^{kg}/_{m3}$) microalgae broth. Harvesting is needed to separate the algae cells from the cultivation broth, thereby also concentrating the algae stream. This is followed by a dewatering step to concentrate the algae stream further. During the third step, disruption takes place to destroy the cell structure. After disruption the cell content (including proteins) is released into the medium. Extraction is needed to separate the proteins from the broth and cell debris. Each of the four DSP steps consist of a selection of available process units, given in Table 1.

Table 1: Overview of some possible units for the four different DSP steps

Harvesting	Dewatering	Disruption	Extraction
Centrifugation	Centrifugation	Homogenizer	Hexane extraction
Pressure filtration	Pressure filtration	Bead mill	Alkaline extraction
Vacuum filtration	Vacuum filtration	Calander	
DAF	Dryer		
Flocculation			
Ultrasound sedimentation			

Since fuel sources are getting scarce, it is not only important to search for new protein sources, but also to obtain these proteins as sustainably as possible. It is therefore important to look not only at the protein yield, but also the energy use needed to obtain these proteins. The combination of process units which produces proteins with the highest protein yield versus lowest energy ratio, is considered most suitable. This ratio of protein yield to energy requirements is called the 'y- value'. The main problem of this project is:" *Which of the process unit combinations at what conditions results in the lowest y-value, for the downstream processing of microalgae biomass?*"

In the report there is a distinction made between 'wet' and 'dry' downstream processing. During 'wet' downstream processing the algae concentration is relatively diluted when it enters the disruption step, the maximum concentration will be in this route 150 $^{kg}/_{m3.}$ Since the stream is diluted when extraction takes place, it has an effect on the recovery of the proteins. During 'dry' downstream processing the algae concentration is much more concentrated (up to 800 $^{kg}/_{m3}$) when the algae stream is being disrupted. A research question in this project is: "*Which of the two methods, 'wet' or ' dry' downstream processing, is energetically more favourable?".*

In this project the main focus lays in the extraction of proteins to produce a new protein source for food and feed. However, to obtain the proteins from the microalgae broth, it is interesting to look also into the extraction of lipids as well. Lipids from algae can be converted into biofuels (Wijffels and Barbosa 2010). By producing two end products, proteins and lipids, the DSP becomes more feasible and sustainable. However, the effect of lipid extraction may have a negative effect on the protein recovery yield. Therefore two different scenarios are determined: one scenario where only protein is recovered from microalgae and the other scenario where both proteins and lipids are recovered from the microalgae stream. A question in this project is: *"What provides the lowest y-value, extraction of proteins or extraction of both proteins and lipids?"*

The overall purpose of this project is to find the best combination of process units, with the most optimal process conditions for the different scenarios. A model based analysis is done in this project, which is based on mass and energy balances made from the process units.

2 Method

For the assessment of the downstream processing of microalgae biomass, models are made from the process units. The models and the method to answer the questions described in the introduction are described in this chapter. Section 2.1 describes how the routes are defined and gives the different superstructures. In section 2.2 there is a description of the models and the overall mass and energy balances are given. The different harvestings process units are described in section 2.3. In section 2.4 the dewatering step is explained. Section 2.5 is used to describe the drying step. Disruption techniques are introduced in section 2. 6 which is followed by the extraction step in section 2.7. In section 2.8 the pump is given. This chapter ends with section 2.9, where the optimisation method is described. This chapter ends with section 2.10, here a table is given which shows all the process conditions of the process units.

2.1 The routes

The downstream processing of microalgae consist of the four steps: harvesting, dewatering, disruption and extraction. For each of the four steps several unit operations are possible. Many combinations of process units, given in Table 1, can be made, see Figure 2-1 to Figure 2-4. These combinations are called routes. A superstructure shows all potential routes possible to obtain proteins from a microalgae stream. The connection lines in the superstructure indicate that two unit operations can be coupled. By combining unit operations starting with 'algae' and ending with 'proteins' gives a route.

A distinction is made between 'wet' and 'dry' routes. A 'wet' route describes the DSP when a relative dilute microalgae broth enters the disruption step. 'Dry' routes have more extensive harvesting and dewatering step. Therefore, the solution is more concentrated before it reaches the disruption step. A differentiation has been made between dry and wet routes, because the microalgae concentration has a big influence on the disruption steps and therefore automatically on the steps before and after the disruption. Also, a distinction is made the extraction of solely proteins and the extraction of both proteins and lipids. Together this provides four different scenarios and consequently in four different superstructures. The four different scenarios are described in Table 2.

Table 2: Properties of the four different scenarios

Scenario	Wet/dry disruption	Extracted	Figure
1	Wet	Proteins	Figure 2-1
2	Wet	Proteins + Lipids	Figure 2-2
3	Dry	Proteins	Figure 2-3
4	Dry	Proteins +Lipids	Figure 2-4

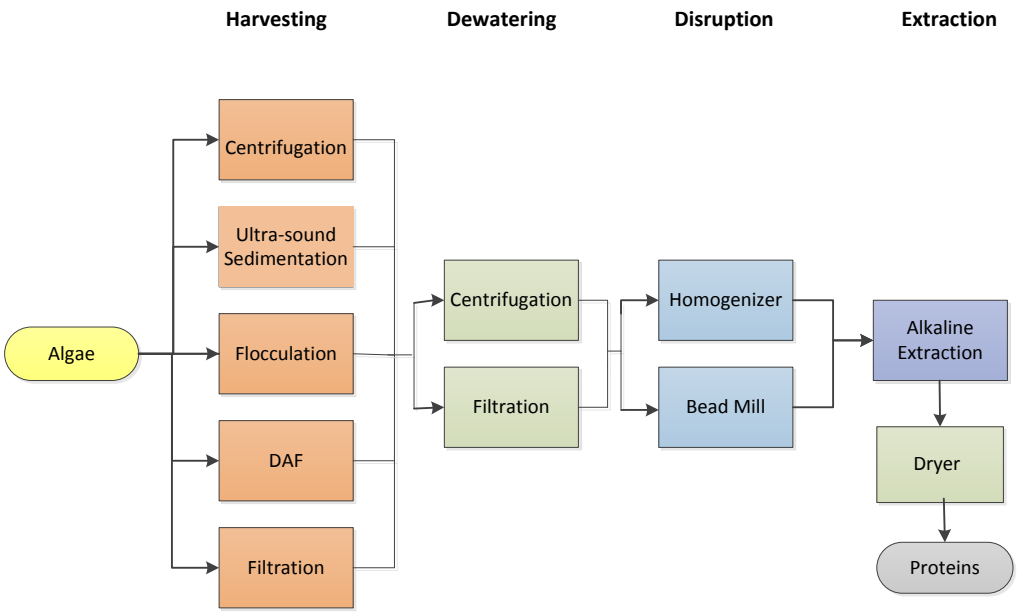


Figure 2-1: DSP from microalgae to protein. This process scheme shows 'wet' routes, because the disruption step takes place at a rather low algae concentration. The concentrations when disruption takes place are 50 $^{kg}/_{m3}$, 100 $^{kg}/_{m3}$ and 150 $^{kg}/_{m3}$

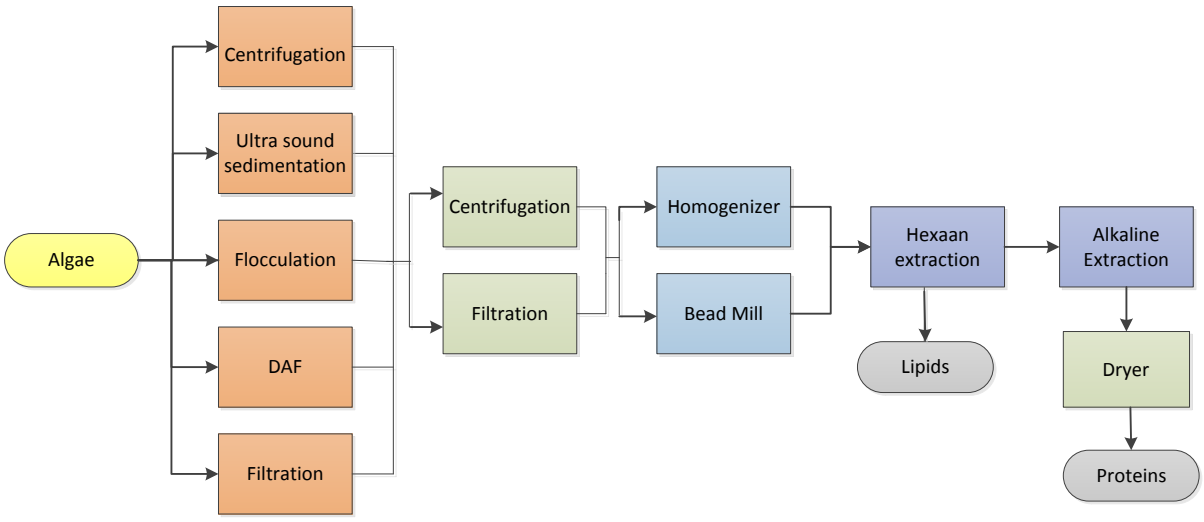


Figure 2-2: DSP from microalgae to proteins + lipids. This process scheme contains 'wet' routes, since the disruption takes place at a relative low microalgae concentration. The concentrations when disruption takes place are 50 kg/m3, 100 kg/m3 and 150 kg/m3

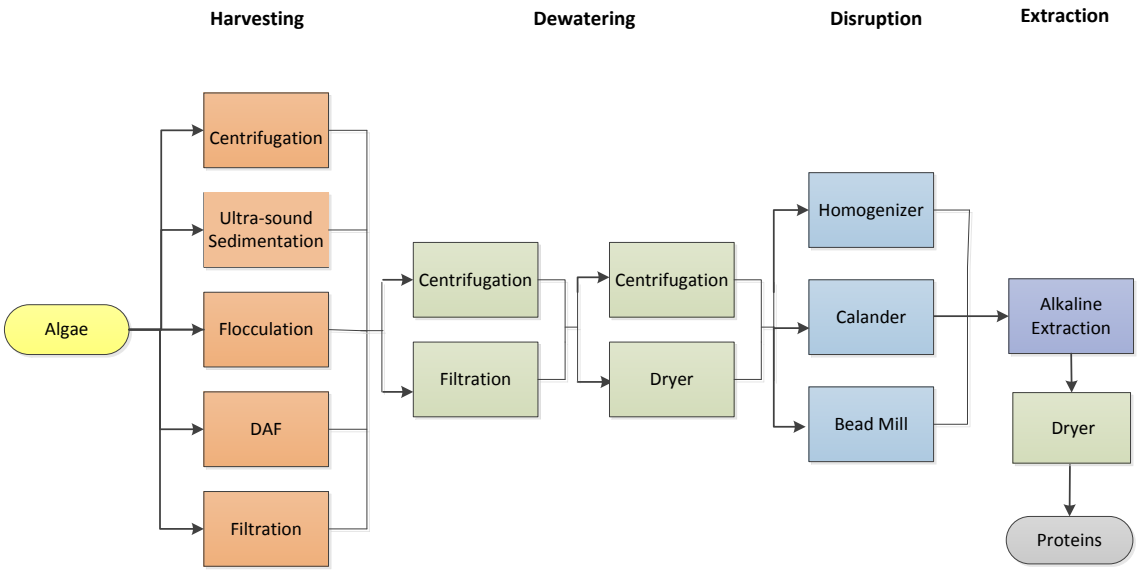


Figure 2-3: : DSP from microalgae to protein. This process scheme shows 'dry' routes, because the disruption step takes place at a higher microalgae concentration. The concentrations when disruption takes place are 200 $^{kg}/_{m3}$, 300 $^{kg}/_{m3}$, 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3}$

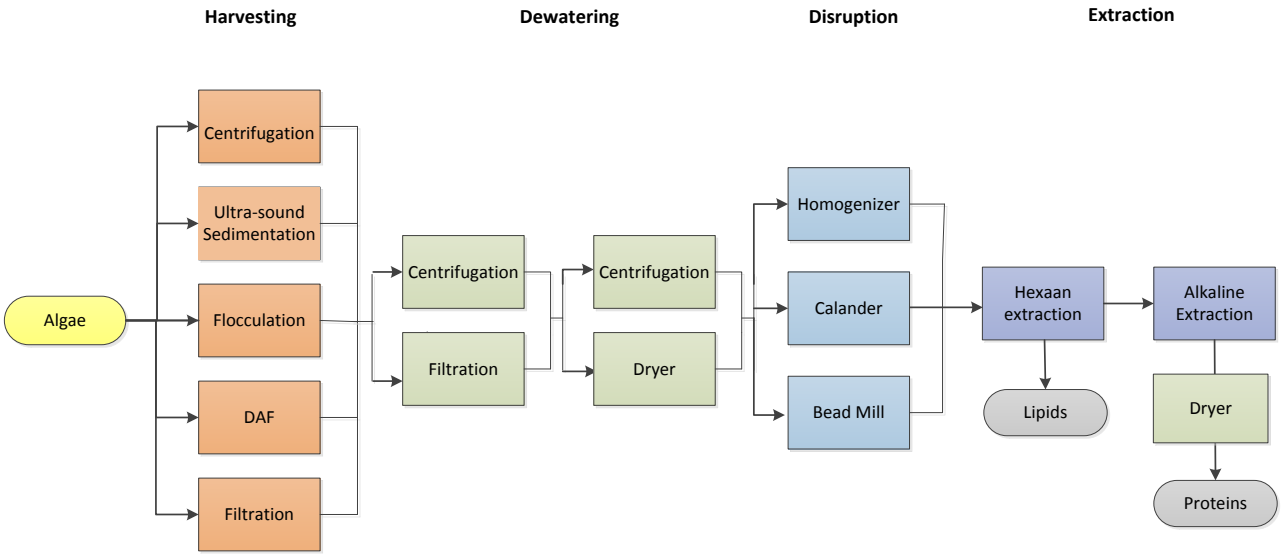


Figure 2-4: DSP from microalgae to proteins + lipids. This process scheme shows 'dry' routes, because the disruption step takes place at a higher microalgae concentration. The concentrations when disruption takes place are 200 $^{kg}/_{m3}$, 300 $^{kg}/_{m3}$, 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3}$

2.2 Model description

The answer to the project question is tackled via a model-based approach. In this chapter the different operation units are described. Mass and energy balances are made for each unit to give an relationship between the process conditions and the incoming and leaving stream of a processing unit.

The mass balance is described as:

$$F_{main,in} * C_{X,main,in} + F_{co,in} * C_{X,co,in} = F_{main,out} * C_{X,main,out} - F_{co,out} * C_{X,co,out} \quad \text{Eq. 2.1}$$

Where F is the flow rate in $^{m^3}/_s$ for the *main*stream and the *co*stream entering (*in)* and leaving (*out*) the processing unit and where C_x is the concentration of component X ($^{kg}/_{m^3}$) . X can stand for either algae, protein, lipid, depending on the mass balance.

The energy balances are used to calculate the power consumption for each unit operation. The energy balances consist of the energy needed for heating , cooling, pressurising, specific energy needed for unit operation , mixing and pumping. The total energy input for each unit operation H_{total} ($^J/_s$) is described as:

$$H_{total} = H_h + H_c + H_{pr} + H_s + H_m + H_p \quad \text{Eq. 2.2}$$

With H_h ($^J/_s$) the energy needed for heating, H_c ($^J/_s$) represents the energy needed for cooling, H_{pr} ($^J/_s$) stands for the energy needed to pressurize the ingoing stream , H_s ($^J/_s$) is the mechanical energy needed for the individual unit operation, H_m ($^J/_s$) stands for the energy needed for mixing and finally H_p ($^J/_s$) is the energy needed to pump the outgoing stream to the next unit.

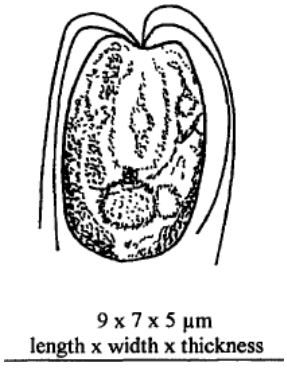


Figure 2-5: Morphology of *tetraselmis* species. Source: Jaouen *et al*, 1998

For this project the microalgae species *Tetraselmis* is used, because this species is most commonly investigated at the Wageningen University and Research Centre and at AlgaePARC to examine microalgae as biofuel and food source. *Tetraselmis* sp. possess well known nutritional qualities and is easily grown on industrial scale (Jaouen, Vandanjon et al. 1999). The morphology of the microalgae species can be seen in Figure 2-5. The protein content is assumed to be 36% w/w % dry matter and the lipid of percentage of 22 w/w % (Schwenzfeier, Wierenga et al. 2011). Data derived from other microalgae species were used when data was not known for the T*etraselmis* species.

2.3 Harvesting

The recovery of microalgae biomass from the culture media can be achieved with different kind of solid-liquid separation steps. Harvesting is mainly done to recover the microalgae from the culture medium and as a direct result the stream is concentrated.

During the harvesting there is only one stream going into the system and two streams are coming out, as can be seen in Figure 2-6. The waste stream consist of the water which is removed from the ingoing algae stream.

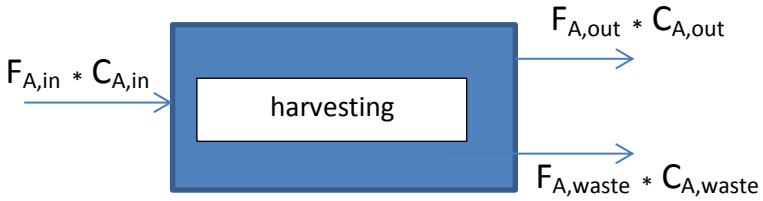


Figure 2-6: Illustration of flows during harvesting

Harvesting is described by the following mass balance:

$$0 = F_{A,in} * C_{A,in} - F_{A,out} * C_{A,out} - F_{A,waste} * C_{A,waste} \quad \text{Eq. 2.3}$$

$$C_{A,out} = C_{A,in} * Cf \quad \text{Eq. 2.4}$$

$$F_{A,out} = \frac{F_{A,in} * C_{A,in} * R}{C_{A,out}} \quad \text{Eq. 2.5}$$

$$F_{A,waste} = F_{A,in} - F_{A,out} \quad \text{Eq. 2.6}$$

$$C_{A,Waste} = \frac{F_{A,in} * C_{A,in} * (1-R)}{F_{A,waste}} \quad \text{Eq. 2.7}$$

Here *F* is the volumetric flow rate in $^{m3}/s$. *C* is the concentration of the stream in $^{kg}/_{m3}$. *R* represents the microalgae recovery in $^{kg}/_{kg}$ and *Cf* is the concentration factor, which is dimensionless.

The basic equation describing the energy needed for harvesting is:

$$H_{harvesting} = H_s + H_p \quad \text{Eq. 2.8}$$

With H_s ($^{J}/_{s}$) as the mechanical energy needed for the individual unit operation and H_p ($^{J}/_{s}$) stands for the energy needed to pump the microalgae stream to the next operation unit. The pumping distance is assumed to be 25 meters.

2.3.1 Centrifuge

During centrifugation cells are separated from the liquid by the difference in density and of the microalgae cells which is the density of the culture medium (assumed in the models to be similar to water). Different types of industrial centrifuges can be used for continuous flows. For the harvesting of microalgae with a size of around 10 µm the nozzle type centrifuge is the most suitable. This centrifuge contains internally stacked discs, as can be seen in Figure 2-7. This system needs minimal manual intervention and is most suitable for harvesting the microalgae compared to multi-chamber and solid bowl centrifuges (Williams and Laurens 2010).

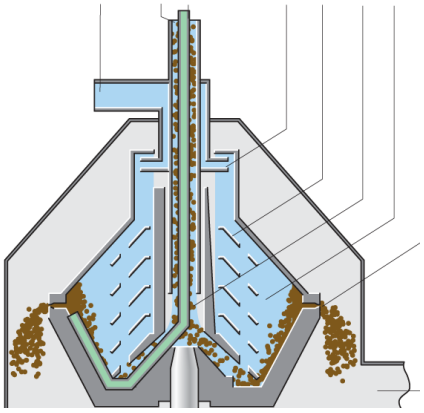


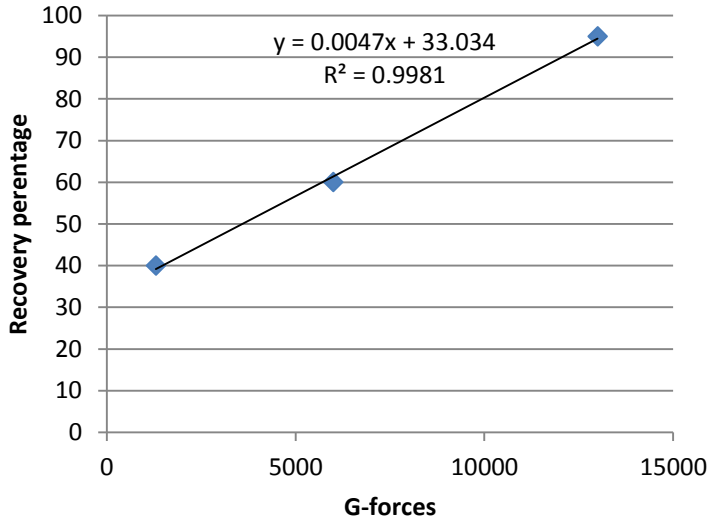
Figure 2-7: Nozzle type centrifuge
Source: (Molina Grima, Belarbi et al. 2003)

The energy consumption according to Wileman *et al.* is:

$$H_s = E * F_{A,in} * Cf \qquad \text{Eq. 2.9}$$

Where H_s ($^{J}/_{s}$) is the mechanical energy consumption for the centrifuge, E is the energy requirement for the centrifuge is shown in Table 3. This number is given by Wileman *et al*. F is the volumetric flow rate ($^{m^3}/_{s}$) and Cf is the concentration factor which is dimensionless.

The recovery of the biomass depends on several factors, like the cells' characteristics and the residence time of the cell slurry. The efficiency of the cell harvest was assessed by Heasman *et al.* for nine different algae species (Heasman, Diemar et al. 2000). It was concluded that over 95% could be recovered at 13.000 x g. The recovery was significantly smaller at 6.000 x g (60%) and at 1300 x g the efficiency declined to 40%. graph 1 shows the correlation between g-forces and microalgae recovery percentage. The graph shows that solids recovery is inversely proportional to the relative centrifugal force.



graph 1: G-forces versus recovery percentage. Source: (Heasman, Diemar et al. 2000)

The g-forces are related to the rotational speed (RPM) as follows:

$$RCF = 1.118 * 10^{-3} * r * S_{rpm}^2 \qquad \text{Eq. 2.10}$$

Where *RCF* represents the relative centrifugal force measured in g; *r* is the rotational radius (m) and S_{rpm} is the rotational speed measured in revolutions per minute (RPM).

For the process model a relation between RPM and energy is necessary to relate the recovery with energy consumption in $^{J}/_{m3}$ or in $^{kWh}/_{m3.}$ No data from literature, manufacturers or lab is known about this correlation, so the effect of g-force is neglected in this work.

Table 3: Energy use of centrifuge, pressure and vacuum filtration in $^{J}/_{m3}$. Source: (Wileman, Ozkan et al. 2012)

Centrifuge	Pressure filtration	Vacuum filtration
E = 1.188.000 J/m^3	**E = 1.692.000 J/m^3**	**E = 7.245.000 J/m^3**

The process conditions of the centrifuge are given in chapter 2.10. In this section Table 4 shows the process conditions of all process units.

2.3.2 Pressure and vacuum filtration

A common separation method in biotechnology processes are filter presses operating under pressure or vacuum . Filtration is simple, efficient, recovers large quantities of biomass and works with a continues flow (Kim, Yoo et al. 2013).

Filtration is a mechanical method to separate particles based on size. The algae cells are separated from the fluid by pushing the stream at high pressure through a filter. The fluid passes the filter, but the oversized solids, like microalgae cells, are retained. The difference between the two filtration techniques is based on the pressure difference in the system. With pressure filtration the pressure difference is achieved by creating a higher pressure at the retentate side, which results in the pushing of the stream through the membrane. With vacuum filtration the pressure difference is accomplished by creating a vacuum on the filtrate side, the liquid is sucked through the membrane (Borowitzka, Moheimani et al. 2013).

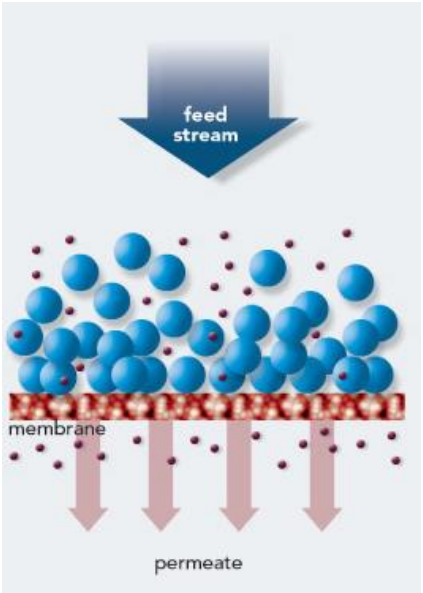


Figure 2-8: Basic overview pressure and vacuum filtration

Microalgae recovery using pressure and vacuum filtration can be up to 95%, which is the same recovery when a centrifuge is used (Brentner, Eckelman et al. 2011).

Eq. 2.9 describes the energy requirements for vacuum filtration and pressure filtration.

Table 3 shows the energy use (the *E* value) of both filtration systems. This table shows that the energy requirements for pressure filtration is much lower than for vacuum filtration. Because of this big difference in energy use, only pressure filtration is considered in his project.

2.3.3 Dissolved air flotation

Dissolved Air Flotation (DAF) is a method to separate a solid phase (algae) from a liquid phase (medium). This is done by injecting a gaseous phase(air) into the liquid phase (Bondelind, Sasic et al. 2013). Dissolved air flotation is found in several industrial application and can also be used as a method to recover microalgae biomass from a diluted stream (Rawat, Ranjith Kumar et al. 2013).

In DAF the microalgae stream is supersaturated with air at a very high pressure in a saturation tank (Sim, Goh et al. 1988). After supersaturating the stream with air, the stream enters the separation tank and depressurizes back to atmospheric pressure in a separation tank. This depressurization of the stream results in the formation of very fine bubbles of air with a range in size from 10-3000 µm in diameter, depending on the method used (Uduman, Qi et al. 2010). These bubbles float to the top of separation tank. Microalgae cells attach to the fine bubbles of air and float to the water surface in the tank (Borowitzka, Moheimani et al. 2013). The top layer of the broth in the separation tank consists of a slurry with a relative high concentration microalgae attached to the foam (air + solids). The algal foam that is formed stays on the surface for a period of time and can be removed. Harvesting of the cells occurs when the foam layer is removed from the separation tank(Chen, Yeh et al. 2011). In Figure 2-9 a typical DAF system is illustrated.

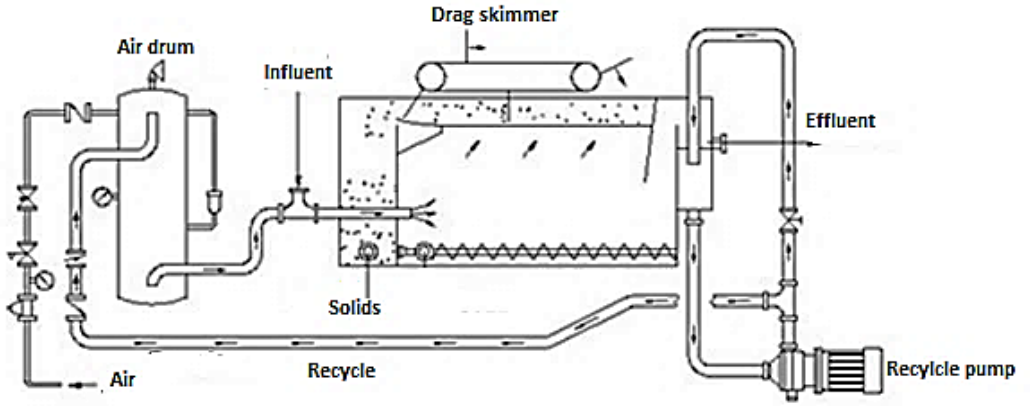
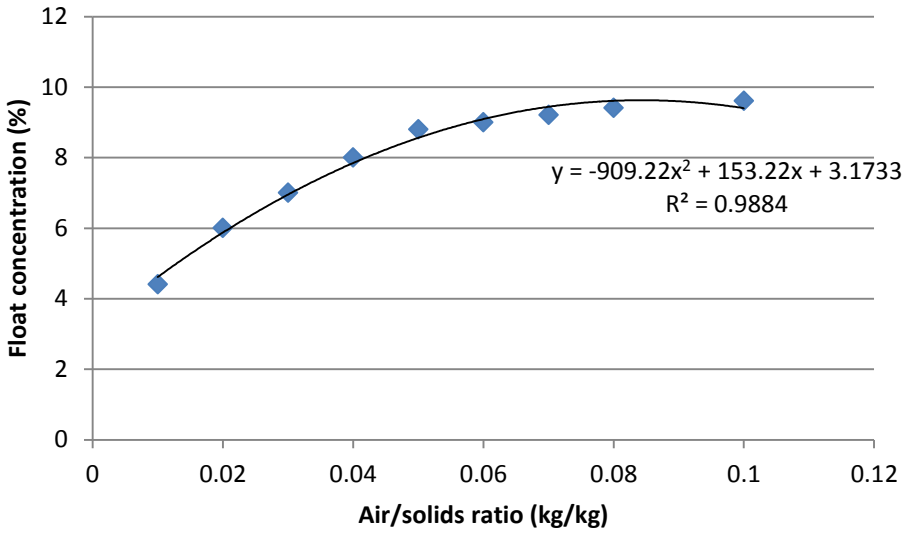


Figure 2-9: Typical Dissolved Air Flotation system with recycle flow. In the DAF the foam layer, which contains the concentrated algae, is skimmed off. At the bottom of the tank algae-free medium (the subnatant) is removed. A part of the subnatant is recycled. In this recycle flow the pressurizing of the air takes place. The recycle flow moves back to the inlet of the saturation tank and is supersaturated with air. When leaving the separation tank, the recycle flow with air is conjoined with a new ingoing stream of microalgae broth (Edzwald 2010). The total amount of air delivered to the system depends on the pressure in the saturator and the recycle flow.

The foam can contain up to 10% of dry weight microalgae cells (Rawat, Ranjith Kumar et al. 2013). To achieve such a concentrated microalgae biomass concentration in the foam, it is

important to add chemicals to neutralize the negatively charged cells to achieve proper flotation (Phoochinda and White 2003). Addition of flocculants to the medium helps to increase the size of the flocs, which results in higher cell concentrations in the foam. Sim *et al.* proposed Chitosan as flocculant during DAF harvesting. Chitosan is manufactured by the hydrolysis of chitin and produces very large stable flocs and results in 95% algae removal.

A typical recycle flow rate is 8-12% of the total ingoing flow. In the model, a recycle flow of 12% is assumed(Wang, Hung et al. 2005, Edzwald 2010). The amount of air added to the recycle flow depends on the amount of solids going into the DAF System. This ratio is called the Air/Solids mass ratio and is defined as kg air per kg microalgae cells.



graph 2: Air solids ratio (kg/kg) versus float concentration (5); the amount of air needed to achieve a certain concentration (in weight percentage) of algae in the foam which floats on the surface of the separation tank. Source: Wang *et al*, 2005.

graph 2 illustrates that increasing the A/S ratio beyond an optimum value results in an insignificant increase of solid concentration in the foam (Wang, Hung et al. 2005). According to Wang *et al* the retention time of the microalgae solids in the DAF system should be at least 3 minutes. The separation tank must be at least 5% of the size of the total inflow. It is assumed that the volume of the separation tank is big enough to achieve this retention time.

The energy needed to harvest microalgae cells in a DAF system is described as:

$$H_{harvesting} = H_{pr} + H_s + H_p + H_{rcfl} \qquad \text{Eq. 2.11}$$

With H_{pr} ($^{J}/_{s}$) the energy need to pressurize the air into the recycle flow , H_s ($^{J}/_{s}$) represents the mechanical energy needed for the individual unit operation, H_p ($^{J}/_{s}$) is the energy needed to pump the outgoing stream to the next unit and H_{rcfl} ($^{J}/_{s}$) to pump the recycle flow back to the separation tank.

The specific work required for the DAF to compress the air can be described in the following equation:

$$W = \frac{RT}{\Upsilon - 1}\left[\left(\frac{P_2}{P_1}\right)^{\frac{\Upsilon-1}{\Upsilon}} - 1\right] \qquad \text{Eq. 2.12}$$

Here W is the work in $^{J}/_{mol}$ of air; R represents the gas constant; T is the temperature in Kelvin; P is the pressure used in the model and Υ is the ratio specific heat of air and has a value of 1.4.

$$H_{pr} = W * S_a * F_{air} * \frac{1}{\eta} \qquad \text{Eq.2.13}$$

F_{air} in $^{m^3}/_{s}$ is the airflow and is based on the A/S ratio; this ratio is set between 0.02 and 0.05. S_a describes the air solubility in water at room temperature and has a value of 42.0. The stream is assumed to have the same properties as water. η is the efficiency of the air compressor. The efficiency of an air compressor is never 100% efficient due to leaks and the conversion of energy into heat. Therefore, it is assumed that the compressor is 50% efficient (Coward, Lee et al. 2013).

The mechanical energy is assumed to be 10% of the total energy use of a DAF system as described by Coward *et al*. This is according to Coward 7.6 $^{kWh}/_{m^3}$. H_s has a value of $3.8*10^4$ $^{J}/_{s}$.

2.3.4 Flocculation by chitosan

During flocculation a flocculant is added to the algae stream. The flocculant interacts with the surface of the algae cells resulting in coagulation of algae cells, which creates large particles. The larger particles (aggregate) coalesce into larger flocs. These flocs are separated from the medium due to sedimentation (Riano, Molinuevo et al. 2012). The process scheme of flocculation is shown in Figure 2-10.

The pH of the solution influences the size of the flocs. In neutral solution chitosan is able to produce larger and denser flocs, while in acidic solution the flocs are much smaller and looser. Adjusting the pH to a final of 8.0 increases the viscosity of chitosan and improves precipitation of the flocs (Morales, Delanoue et al. 1985).

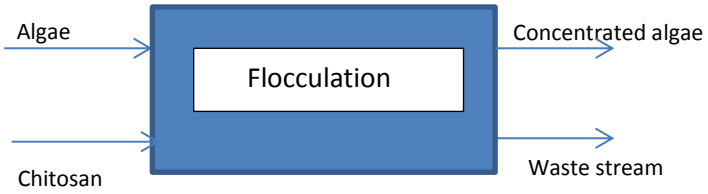


Figure 2-10: Schematic overview of flocculation with chitosan

Chitosan is used as flocculant, it is natural, bio-degradable, non-toxic, safe to handle and used in several food, agricultural and chemical industries (Ahmad, Yasin et al. 2011). Ahmad

et al concluded that chitosan has no negative effect on human health and is suitable to harvest microalgae cells which are used for protein recovery.

The energy requirements for flocculation is described as:

$$H_{harvesting} = H_m + H_p \quad \text{Eq.2.14}$$

Next to energy for pumping H_p, also energy for mixing , H_m, is needed. The energy consumption of mixing is defined as:

$$H_m = E * (F_{A,in} + F_{co,in}) \quad \text{Eq.2.15}$$

Where *E* is the energy requirement for mixing in $^{J}/_{m^3}$, *F* is the volumetric flow rate $\left(^{m^3}/_{s}\right)$ and *in* and *co* represent the ingoing algae stream and the flocculant stream respectively. The energy requirements *E* for mixing is estimated to be 360 $^{J}/_{m^3}$ (Brentner, Eckelman et al. 2011).

Riano *et al* derived a second order polynomial equation describing microalgae recovery due to flocculation with chitosan (Riano, Molinuevo et al. 2012).

$$R = 84.3 + 17.5\, C_F^* - 1.3S^* - 11.1(C_F^*)^2 - 3.7(S^*)^2 - 2.6C_F^*S^* \quad \text{Eq.2.16}$$

Where

$$C_F^* = \frac{C_F - 0.128}{0.086} \quad \text{Eq.2.17}$$

$$S^* = \frac{S - 325}{194} \quad \text{Eq.2.18}$$

With C_F the flocculant concentration ($^{kg}/_{m^3}$) where *S* is the agitation speed for mixing (rpm) and *R* is the microalgae recovery percentage (%).

The biomass recovery can reach up to 92% at a chitosan concentration of 214 $^{mg}/_{l}$ and a agitation speed of 131 rpm (Riano, Molinuevo et al. 2012). This is based on laboratory scale and is assumed to be linear on larger scale.

2.3.5 Ultra sound sedimentation

Ultrasound sedimentation is based on the separation of algae cells from the broth by acoustic induced aggregation of the algae cells (Bosma, van Spronsen et al. 2003). The algae broth is continuously pumped into a resonator chamber. The chamber contains a transducer and a reflector. When turned on it creates a standing wave (when chamber size and frequency are well defined). The standing wave created places of high potential energy (bellies) and low potential energy (nodes). The algae cells move to the nodes of the standing wave and aggregate due to acoustic interaction forces and algae-algae interaction forces. The algae aggregates stay in the nodes of the standing wave when the field is on. When the electric field in the chamber is removed, the algae aggregates (flocs) sediment to the

bottom of the chamber and can be removed from the broth. In Figure 2-11 the principal of ultra sound harvesting is illustrated.

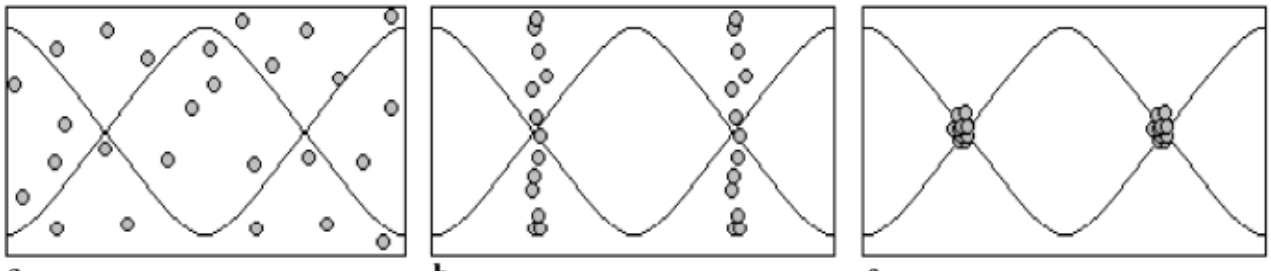


Figure 2-11: Principal of ultrasound sedimentation harvesting: a.) No ultrasonic field b.) Field is turned on and cells migrate to pressure node planes c.) Cells aggregate into the knots of the ultrasonic field. Source: Bosma *et al.*

The recovery of the algae from the broth and the concentration factor were separately determine by the use of an experimental design (Bosma, van Spronsen et al. 2003). Bosma *et al* concluded that both recovery and CF depend on biomass concentration, flow rate, time frequency, power input and ratio F_{out} and $F_{in.}$ Two polynomials were developed by to describe the *R* and *Cf*.

$$R = 0.75 + 1.2 * 10^{-9} * C_{A,in}^{*} + 0.052 * F_{A,in}^{*} - 3.3 * 10^{-3} * \tau^{*} + 3.2 * 10^{-3} * H^{*} - 4.1 * 10^{-3} * {F_{A,in}^{*}}^{2} + 8.5 * 10^{-6} * \tau^{*2} - 6.6 * 10^{-11} * C_{A,in}^{*} * F_{A,in}^{*} + 2.7 * 10^{-3} * F_{A,in}^{*} * H^{*}$$

Eq.2.19

$$Cf = 18 - 2.7 * 10^{-8} * C_{A,in}^{*} + 1.3 * F_{A,in}^{*} - 1.6\left(\frac{F_{A,out}}{F_{A,in}}\right)^{*} - 0.77 * {F_{A,in}^{*}}^{2} + 3.1 * 10^{-9} * C_{A,in}^{*} * \left(\frac{F_{A,out}}{F_{A,in}}\right)^{*}$$

Eq.2.20

$C^{*}_{A,in}$, $F^{*}_{A,in}$, τ^{*} and H^{*} are coded values used by Bosma *et al* and stand for algae concentration, flow rate, time frequency (s) , power input ($^{J}/_{s}$) and ratio F_{out} and F_{in} , respectively.

$$C_{A,in}^{*} = \frac{C_{A,in} - 17}{16}$$ Eq.2.21

$$F_{A,in}^{*} = \frac{F_{A,in} - 10}{8}$$ Eq.2.22

$$\tau^{*} = \frac{\tau - 180}{120}$$ Eq.2.23

$$H^{*} = \frac{H_{S} - 6}{2}$$ Eq.2.24

$$\left(\frac{F_{A,out}}{F_{A,in}}\right)^{*} = \frac{\frac{F_{A,out}}{F_{A,in}} - 6}{4}$$ Eq.2.25

An ultrasound sedimentation processing unit described by Bosma *et al* can process up to 18 $^{l}/_{day}$ algae broth. In this project it is assumed that a processing unit can process up to 432 $^{l}/_{day}$

(0.018 $^{m^3}/_{h}$)(Cappon 2013). If the ingoing flow exceeds this number, more parallel process units are used.

The energy required for ultrasound sedimentation is:

$$H_{harvesting} = H_s + H_p \quad \text{Eq.2.26}$$

$$H_s = P_{in} * N_{units} \quad \text{Eq.2.27}$$

In this equation P_{in} is the power input ($^{J}/_{s/unit}$) and N_{units} stands for the number of units needed (-).

On lab scale ultrasound sedimentation is a very good method to harvest algae from the medium. On industrial scale harvesting with ultrasound sedimentation is more difficult, since the efficiency is not optimal due to small density difference between algae and medium. On the other hand, it does not create shear stress and the occupation space of the unit operation is relatively small (Bosma, van Spronsen et al. 2003).

2.4 Dewatering

The algae biomass slurry is further concentrated during the dewatering step. Centrifuging, filtration (both mechanical) and drying (thermal dewatering) are all suitable methods to dewater the algae stream to a more concentrated stream. In Figure 2-12 the flow scheme of dewatering is shown.

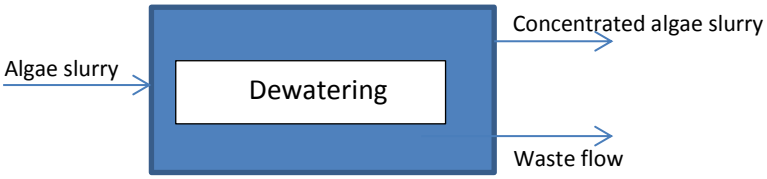


Figure 2-12: Flows during dewatering

Both centrifuging and filtration are used as dewatering method. These two process units are described in chapter 2.3.1 and 0.

Drying is used both as dewatering step (before disruption) and also after protein extraction to concentrate the protein flow to the end concentration of 700 $^{kg}/_{m3}$. The dryer is seen twice in superstructure Figure 2-3 and Figure 2-4, which display the 'dry' routes.

2.5 Drying

During drying, heat is used to evaporate water from the algae broth. During this step no algae cells are lost to a waste stream. Thermal dewatering takes place when hot air of 373 Kelvin is used to evaporate the water in the algae stream. The algae solution itself however

should not reach temperature's above 316 K. Above 316 K the temperature has a negative effect on the protein stability and solubility in the algae cells.

Thermal dewatering is described by the following algae mass balance:

$$0 = F_{A,in} * C_{A,in} - F_{A,out} * C_{A,out} \quad \text{Eq. 2.28}$$

$$C_{A,out} = C_{A,in} * Cf \quad \text{Eq. 2.29}$$

$$F_{A,out} = F_{A,in} / Cf \quad \text{Eq. 2.30}$$

The evaporated water flow is described as:

$$F_{waste} = F_{A,in} * \left(1 - \frac{C_{A,in}}{\rho_{algae}}\right) - F_{A,out} * \left(1 - \frac{C_{A,out}}{\rho_{algae}}\right) \quad \text{Eq.2.31}$$

The energy requirements for drying with heat is:

$$H_{dewatering} = H_h + H_s + H_p \quad \text{Eq.2.32}$$

$$H_h = F_{A,in} * \rho_A * cp_A * (T_{A,out} - T_{A,in}) \quad \text{Eq.2.33}$$

$$H_s = \Delta H_{vap} * V_w * \rho_A * \left(\frac{(T_{air} - T_{A,out}}{T_{air} - T_{A,in}}\right) \quad \text{Eq.2.34}$$

Here, ρ_A represents the density of the algae stream ($^{kg}/_{m3}$), cp_A stands for the heat capacity of the algae stream ($^{J}/_{K}$), ΔH_{vap} is the heat of evaporation for water ($^{J}/_{kg}$), V_w is the amount of evaporated water (m^3), ρ_A is the density of the algae stream ($^{kg}/_{m3}$), T_{air} is the temperature of the heated air (K), $T_{A,in}$ is the temperature of the ingoing algae slurry (K) and $T_{A,out}$ stands for the temperature of the outgoing stream and in the dryer (K).

2.6 Disruption

To extract proteins from the microalgae cells it is necessary to disrupt the cells first. By subjecting a stream of microalgae to a force, the cell wall structure will be destroyed. By disrupting the cells, the cell content is released into the suspension and can be further processed in the extraction step. Disruption of microalgae cells is energy intensive, since the algae cells are very small (average of 10µm). Also the cell wall of microalgae is difficult to disrupt: most microalgae species have a tri-layered cell wall structure which results in a though and pliable cell wall. The rigidity of the cell wall differs a lot per species (Lee, Lewis et al. 2012).

The different disruption methods can be broadly divided into two big categories, namely mechanical and non-mechanical disruption. Non-mechanical disruption can be further divided into physical, chemical and enzymatic disruption. Mechanical methods are mostly preferred for the disruption of microalgae cells, since mechanical disruption depends less on which species is used. Also chances of contamination are smaller compared to non-

mechanical methods. A big drawback when using mechanical disruption is the generation of heat, so cooling is needed (Lee, Lewis et al. 2012).

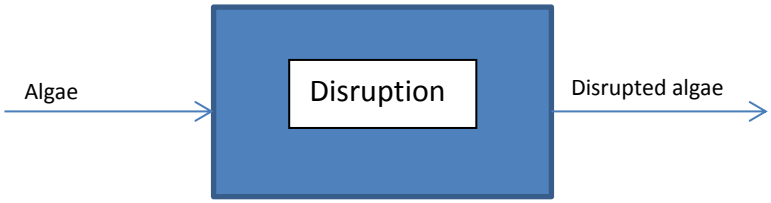


Figure 2-13: Schematic representation of mechanical disruption of microalgae

However a certain percentage of the cells is disrupted and their cell content, including the proteins and lipids, is released to the medium. The protein content and lipid content of *tetraselmis* sp. is 36% and 22% respectively (Jaouen, Vandanjon et al. 1999). In this work it is assumed when a cell is disrupted the whole cell content (proteins and lipids) is released to the medium and available for extraction.

The mass balance is:

$$0 = F_A * C_{A,in} * f_p + F_A * C_{A,in} * f_l - F_A * C_{P,release} - F_A * C_{l,release} - F_A * C_{P,algae} - F_A * C_{l,algae} \quad \text{Eq.2.35}$$

$$C_{P,release} = C_{A,in} * D * f_p \quad \text{Eq.2.36}$$

$$C_{l,released} = C_{A,in} * D * f_l \quad \text{Eq.2.37}$$

$$C_{P,algae} = C_{A,in} * (1 - D) * f_p \quad \text{Eq.2.38}$$

$$C_{L,algae} = C_{A,in} * (1 - D) * f_l \quad \text{Eq.2.39}$$

Where f_p is the algal protein content ($^{kg}/_{kg}$) and f_l is the algal lipid content ($^{kg}/_{kg}$); *F* is the volumetric flow rate in $^{m3}/s$. *C* is the concentration of the stream in $^{kg}/_{m3}$. *D* is the disruption efficiency in $^{kg}/_{kg}$. The subscripts *A*, *P* and *L* stands for algae, proteins and lipids respectively. The subscript *release* stands for the proteins released into the medium due to disruption of algae cells. $C_{p,algae}$ indicates the lipids remained inside the intact cells.

2.6.1 High pressure homogenizer

A high pressure homogenizer (HPH) can mechanically disrupt microalgae cells. In an HPH the cell suspension is forced under high pressure through an opening (orifice). The fluid flows through the orifice, spreads across the seat surface and collides on an impact ring (Middelberg 1995). There are various valve designs to maximize the disruption. In Figure 2-14 a typical HPH valve seat is shown (Lee, Lewis et al. 2012).

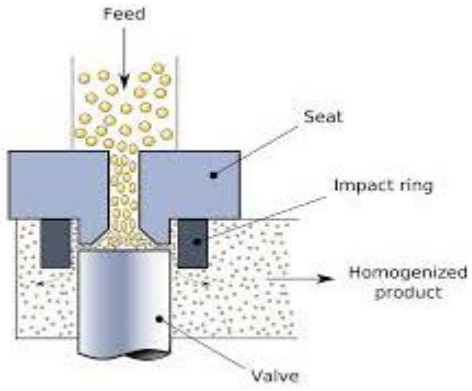
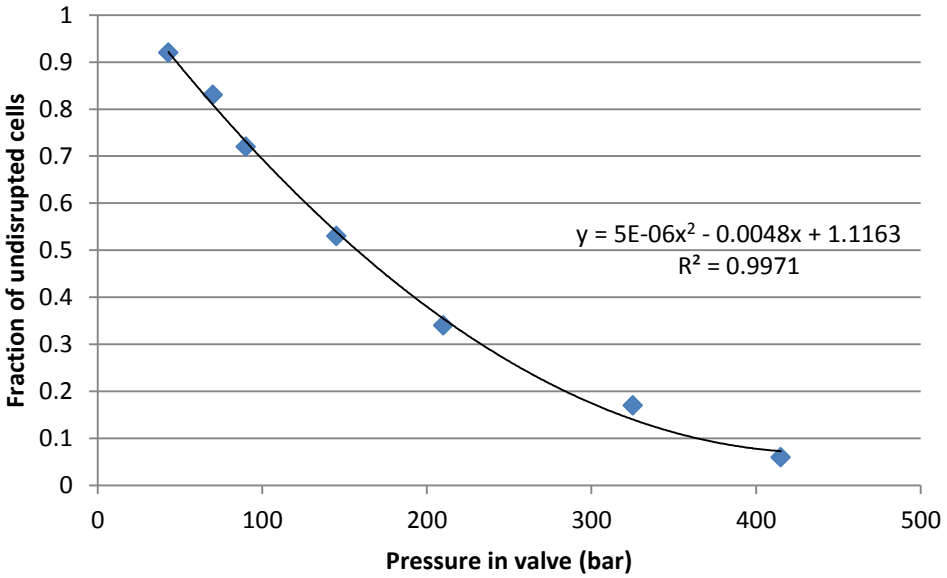


Figure 2-14: A typical HPH valve seat.
Source: Lee, Lewis *et al. 2012*

The disruption mechanism is thought to be the result of the sudden decrease of pressure which results in the release of gas bubbles. These bubbles burst inside the cells (Clarke, Prescott et al. 2010). However, it is not known how exactly the disruption exactly works; it is only known that the shear stress of the process disintegrates the cells.

High pressure is a promising technique, since it is effective for aqueous solutions. HPH eliminates the need for drying the solutions to high concentrations, which is an energy intensive step. Furthermore, HPH is suitable for us on an industrial scale, because it is relatively easy to scale up to larger volumes (Spiden, Yap et al. 2013).

There are a few parameters that have a major impact on the disruption. One important controllable parameter is the pressure applied on the medium, and its accessory pressure drop to atmospheric pressure across the valve, orifice and impact ring. Other important parameters controlling the extent of the disruption are the number of passes through the HPH, the flow rate and the temperature (Lee, Lewis et al. 2012). The amount of passes in the HPH is between one and ten times. Spiden *et al* describes a relation between the pressure used in the HPH and the fraction of (un)disrupted cells. graph 3 shows the fraction of undisrupted cells versus the pressure used in the HPH.



graph 3: Relation between amount of disrupted cells and pressure in valve of a HPH. Date used from Tetraselmis sp. Source: Spiden et al, 2013.

From this graph the decay constant *K* is determined. This constant is determined for the *Tetraselmis* sp. (in Spiden *et al* this constant is defined as A).

$$K = 5 * 10^{-6} * P^2 - 0.0048 * P + 1.163 \quad \text{Eq.2.40}$$

According to Spiden *et al* the disruption is determined with the equation:

$$D = 1 - K^{N_{passes}} \quad \text{Eq.2.41}$$

Where *K* is the decay constant (-) ; *P* is the pressure inside homogenizer and varies between 35-415 (bar), N_{passes} is the amount of times that the flow passes the homogenizer (-) and *D* describes the disruption efficiency ($^{kg}/_{kg}$).

The energy requirements for the HPH is:

$$H_{disruption} = H_c + H_{pr} + H_p \quad \text{Eq.2.42}$$

with H_c ($^{j}/_{s}$) the energy needed for cooling, H_{pr} ($^{j}/_{s}$) is the energy needed for disruption with pressure and H_p ($^{j}/_{s}$) is the energy needed to pump the outgoing stream to the next unit.

The temperature of the medium increases 2 K for every 100 bar and per pass applied on the microalgae inflow (Lee, Lewis et al. 2012). Cooling is needed to keep the algae flow temperature constant at room temperature as to prevent protein degradation. By multiplying the ingoing flow with the heat capacity of the broth (similar to water) and with the pressure and the amount of passes used with the HPH, it is possible to determine the amount of $^{J}/_{s}$ needed to keep the broth at the same temperature.

H_h is described as:

$$H_h = F_{A,in} * \rho_A * cp_A * \Delta K * N_{passes} \quad \text{Eq.2.43}$$

Where $F_{A,in}$ is the ingoing algae stream ($^{m3}/_{s}$), cp_A stands for the heat capacity of the algae stream ($^{J}/_{K}$) and ρ_A is the density of the algae stream ($^{kg}/_{m3}$) and ΔK is the temperature increase in the system (K).The work done by the piston, $H_{pr,}$ can be calculated by multiplying the operating pressure with the amount of passes and the flow of the algal broth processed (Samarasinghe, Fernando et al. 2012).

H_{pr} is described as:

$$H_{pr} = F_{A,in} * P * N_{passes} \quad \text{Eq.2.44}$$

With $F_{A,in}$ as the ingoing algae flow ($^{m3}/_{s}$) and cp_w represents the heat capacity of water in $^{J}/_{kg/K.}$

2.6.2 Bead milling

Bead milling is a mechanical method to disrupt cells in a gentle way. Very small solid bead are added to a suspension in a vessel. This vessel is either rotated around its axis or can be shaken. Due to the rotation of the vessel the beads start rolling away from the direction of the rotation. Also some beads move up against the curved wall of the vessel and cascade back on the suspension. Cell disruption occurs due to the grinding and the collision of the beads against the cells and the impact of the cascading beads onto the cells in the suspension. In Figure 2-15 the setup of a bead mill with a rotating vessel around its axes is illustrated. When the vessel is shaken, the cells gets disrupted due to cascading beats.

There are two types of bead milling. The first type disrupts cells by shaking the entire vessel, this set-up is suitable for laboratory scale and is shown in Figure 2-15. The second type of bead milling, contains a rotating agitator and is more suitable for industrial scale and can be scaled up to a few m^3. The vessel itself is fixed and is filled with the beads and the microalgae suspension. Grinding of the cells takes place due to the rotating agitator in the solution. The vessel contains a cooling jackets to prevent the proteins from denaturation (Lee, Lewis et al. 2012).

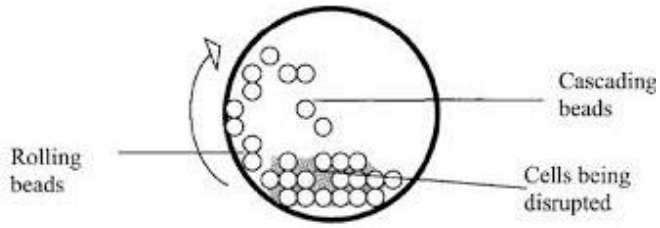


Figure 2-15: Rotating vessel in bead milling system

The efficiency of the disruption depends on a several parameters, i.e. the size and composition of the beads, the amount of beads compared to the suspension, the residence time and the design and the speed of the internal agitator. The efficiency also depends on some characteristics of the microalgae suspension, such as the temperature of the broth, the viscosity and the concentration.

Doucha *et al* describes a power function which describes the degree of disruption in microalgae cells in a specific bead mill (KDL-Pilot A, Dyno-Mill) (Doucha and Lívanský 2008):

$$D = 17.48 * Q^{n1} * BD^{n2} * BF^{n3} * PV^{n4} * DW^{n5} \quad \text{Eq.2.45}$$

With the constant n_1 = -0.0356, n_2 = 0.326, n_3 = 0.0768, n_4 = and n_5 =-0.763 and *D* describes the degree of disruption.

The optimal diameter for the beads (*BD*) is 0.5 mm, and this will stay constant in the model. The beads can best be made of zirconia-silica, zirconium oxide or titanium carbide. This is due to their greater hardness and density. After the bead milling treatment, the beads are separated from the suspension by a sieve. Denser beads can be more easily separated from agitated solution (Hopkins 1991).

A typical beadmill unit can process up to 450 $^{l}/_{h}$ (Doucha and Lívanský 2008). The total amount of process units (N_{units}) is therefore based on the ingoing flow rate.

The energy requirement for the bead mill is:

$$H_{disruption} = H_c + H_s + H_p \quad \text{Eq.2.46}$$

The energy consumption per bead mill unit is 3.3 kW (Doucha and Lívanský 2008) and is described as:

$$H_s = 3300 * N_{units} \quad \text{Eq.2.47}$$

The temperature of the suspension going in is pre-cooled to 277 K. This pre-cooling is needed to prevent a high temperature rise during the grinding. The bead mill used in the model can cool during the grinding by using cooling jackets. The temperature of the outflow is approximately 308 K and must be cooled back to room temperature.

$$H_{c,1} = F_{A,in} * \rho_A * cp_w * (T_{A,in} - T_{BM,in}) \quad \text{Eq.2.48}$$

$$H_{c,2} = F_{A,out} * \rho_A * cp_w * (T_{BM,out} - T_{A,out}) \quad \text{Eq.2.49}$$

The sum of $H_{c,1}$ and $H_{c,2}$ gives the total energy needed for cooling, H_c. In these equations cp_w is the heat capacity of water ($^{J}/_{kg/K}$), $T_{A,in}$ is the temperature of the ingoing flow (K), $T_{A,out}$ is the temperature of the outgoing flow and is the same as the ingoing flow. $T_{BM,in}$ is 277 K and $T_{BM,out}$ is 308 K.

2.6.3 Calander

The calander consist of two walls in the shape of big cylinders which are placed very closely next to each other. Between the cylinders is a small gap, the algae stream is moved through this. The distance between the cylinders is adjustable. For the disruption of *tetraselmis* sp. a gap distance between the cylinders of 20µm is used. The cylinders, made of steel, move in the opposite direction from each other, with adjustable speed. The highest disruption of the *tetraselmis* sp. is achieved when one cylinder moves with 15 RPM and the other one with 18 RPM. At this speed the algae cells are disrupted due to high forces on the cell wall. The cylinders can be heated up to 513 K, in this project a temperature of 323K is chosen. In this case, it is possible to heat the cylinders above 316 K, because the time the cylinders are in direct contact with the algae cells negligible and has no effect on the protein solubility and stability.

Figure 2-16: The calander, Collin WP110. FBR, Wageningen UR

In Figure 2-16 and Figure 2-17 the calander is shown. The calander is a new innovative method to disrupt microalgae cells. Information is provided by Food and Bio based Research Centre, Wageningen UR, the Netherlands.

Figure 2-17: Close up Calander. One Cylinder is visible in this picture.

It is assumed that 90% of the cells can be disrupted using this method. The ingoing algae concentration should be between 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3}$, which is relatively dry. Because of this, in this project the calander is only used for the 'dry' routes.

The energy requirement for the calander is:

$$H_{disruption} = H_h + H_s + H_p \quad \text{Eq.2.50}$$

$H_h = 3.3 * 10^3$ $^{J}/_{s}$.

H_h is the energy for heating and is 1.65 kW/cylinder.

$H_s = 3.0 * 10^3$ $^{J}/_{s.}$ H_s is the mechanical energy with a value of 1.5 kW/cylinder.

2.7 Extraction

After the disruption step the algae slurry contains cell debris and components and some undisrupted cells. During the extraction step the stream is mixed with a solvent. Components are separated from each other based on difference in solubility.

2.7.1 Hexane extraction

Hexane extraction is used to extract lipids from the algae slurry. In Figure 2-18 the flow scheme of lipid extraction with hexane is illustrated. By adding hexane, two distinct phases appear, separated by polarity: one phase is the ingoing algae flow which is polar. The other phase (hexane) is nonpolar. Lipids dissolve in the nonpolar phase.

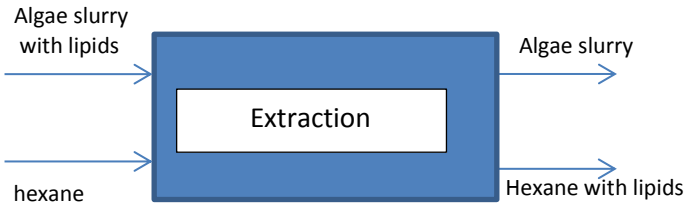


Figure 2-18: Flow scheme lipid extraction with hexane

The lipid mass balance for extraction is:

$$0 = F_{A,in} * C_{L,A,in} - F_{A,out} * C_{L,A,out} - F_{hex,out} * C_{L,hex,out} \quad \text{Eq.2.51}$$

$$C_{L,hex,out} = \frac{F_{A,in} * C_{L,in} * Y_l}{F_{hex,out}} \quad \text{Eq.2.52}$$

$$C_{L,A,out} = \frac{F_{A,in} * C_{L,A,in} * (1-Y_l)}{F_{A,out}} \quad \text{Eq.2.53}$$

Here, $F_{A,in}$ is the ingoing algae slurry flow in $^{m3}/s$, $C_{L,A,in}$ is the concentration of lipids in the ingoing algae slurry stream ($^{kg}/_{m3}$) , $F_{A,out}$ is the outgoing algae slurry containing a small amount of lipids ($^{m3}/s$), $C_{L,A,out}$ represents the concentration of lipids in this outgoing algae slurry, which is very low ($^{kg}/_{m3}$). $F_{hex,\ out}$ is the outgoing hexane flow in $^{m3}/_{h}$ and $C_{L,hex,out}$ is the concentration of lipids in the hexane flow.

Process models on algae lipid extraction yields are not given in literature, so a fixed yield is assumed: for the wet route a yield of 0.7 is assumed and for the dry route a yield of 0.91 is taken (Brentner, Eckelman et al. 2011). The hexane flow is assumed to be 15 v/v % of the ingoing algae stream (Kleinegris, Janssen et al. 2010) .

The energy input is given by:

$$H_{hex,extraction} = H_c + H_m + H_p + H_s \qquad \text{Eq. 2.54}$$

$$H_c = cp_A * \rho_A * \left(T_{A,in} - T_{react}\right) + cp_H * \rho_H * \left(T_{hex,in} - T_{react}\right) \qquad \text{Eq. 2.55}$$

$$H_m = F_{A,in} * \rho_A + F_{hex,in} * \rho_h * K_{power} * \rho_{av} * {u_m}^3 * \left(\frac{1}{2}\sqrt[3]{4 * \frac{\tau}{\pi} * F_{A,in} * F_{hex,in}}\right)^5$$

Eq.2.56

Where $F_{A,in}$ is the ingoing algae flow and $F_{hex,in}$ is the ingoing hexane flow ($^{m3}/_{s}$), T_{react} is the temperature at which the hexane extraction takes place which is at 293 K. $T_{hex,in}$ is the temperature of the hexane (K), which is 293 K, $T_{A,in}$ is the temperature of the ingoing algae stream (K). The energy need for mixing, H_m is given by Wesselingh and Krijgsman. This equation contains K_{power}, which is the power constant equal to 0.4, u_m is the stirrer speed ($^{1}/_{s}$) , ρ_{av} is the average density ($^{kg}/_{m3}$) and τ is the residence time (s).

The mechanical energy (H_s) in this model, is assumed to be 0.1% of the total energy needed for hexane extraction estimated by Brentner *et al.* A value of 0.1% is chosen because this provides an energy use which is than in the same order of magnitude as other process units used in this project. The energy needed for hexane extraction, including mixing and regeneration, was estimated to be 1200 MJ for 294 litre of algae oil (Brentner, Eckelman et al. 2011).

It is assumed that extraction extracted does not have an impact on the protein quality. It does have an effect on the protein extraction. The fraction of the proteins which are extracted with the alkaline solution is lower. The protein fraction which is extracted from the stream during the alkaline extraction step (chapter 2.7.2) is 0.64 instead of 0.75 when hexane extraction has taken place.

2.7.2 Alkaline extraction

Proteins can be extracted with an alkaline solution. When the cells are disrupted during the disruption step, proteins partly dissolve into the stream. When proteins dissolve into a liquid they can be extracted. For the extraction step it is favourable to obtain the highest possible amount of proteins in the liquid, by increasing the solubility of the proteins. Microalgae contain two types of proteins: water-soluble and water-insoluble proteins. When an alkaline solution (water with NaOH) is added to the algae stream, the solubility of the water-insoluble proteins increases and become (more) soluble, resulting in more proteins being dissolved into the liquid (Gerde, Wang et al. 2013).

When NaOH is added the pH increase and enhances the protein solubility. The pH cannot exceed the value of 12,5 otherwise the tertiary structure of the proteins will change and this has an negative effect on the proteins (Chronakis, Galatanu et al. 2000).

Figure 2-19 shows the flow scheme when an alkaline solution is added to the algae stream.

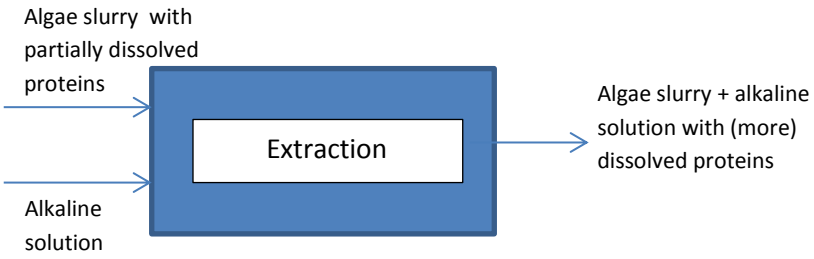


Figure 2-19: Flow scheme protein extraction with alkaline solution

The proteins dissolve in the water phase, other components, like lipids, cannot dissolve in the liquid phase and can therefore be easily removed by centrifuging the liquid. When centrifuging with a concentration factor of 1, the debris can be removed from the liquid. For the centrifuge Eq. 2.9 is used.

The protein mass balance for extraction is:

$$0 = F_{A,in} * C_{P,A,in} - F_{A,out} * C_{P,A,out} - F_{centrifuge,out} * C_{P,centrifuge} \qquad \text{Eq. 2.57}$$

Here, $F_{A,in}$ is the ingoing algae slurry flow in $^{m3}/s$, $C_{P,A,in}$ is the concentration of proteins in the ingoing algae slurry stream ($^{kg}/_{m3}$) both soluble and insoluble proteins. The ingoing alkaline stream does not contain proteins, so is not present in the mass balance. $F_{A,out}$ is the outgoing algae stream mixed with the alkaline stream containing solubilized proteins ($^{m3}/_{s}$), $C_{L,A,out}$ represents the concentration of proteins in this outgoing stream($^{kg}/_{m3}$). $F_{centrifuge}$ is the amount of debris which is removed from the liquid stream after centrifuging ($^{m3}/_{s}$) and $C_{P,centrifuge}$ is the concentration of insoluble proteins found in the removed debris ($^{kg}/_{m3}$).

The energy input is given by:

$$H_{alk,extraction} = H_h + H_m + H_p + H_s \quad \text{Eq. 2.58}$$

$$H_h = cp_A * \rho_A * (T_{react} - T_{A,in}) + cp_H * \rho_H * (T_{react} - T_{alk,in}) \quad \text{Eq. 2.59}$$

$$H_m = F_{A,in} * \rho_A + F_{alk,in} * \rho_w * K_{power} * \rho_{av} * u_m{}^3 * \left(\frac{1}{2}\sqrt[3]{4 * \frac{\tau}{\pi}} * F_{A,in} * F_{alk,in}\right)^5$$

Eq 2.60

$$H_s = E * (F_{A,in} + F_{alk,in}) * Cf \quad \text{Eq. 2.61}$$

Where H_s is the energy needed for the centrifuge to remove the debris from the liquid, the concentration factor (*Cf*) is fixed on one. T_{react} has an assumed value of 303 K, $T_{alk,in}$ is the temperature of the alkaline solution which is 298 K. $F_{alk,in}$ is the ingoing flow of the alkaline solution ($^{m3}/_{s}$). It is assume that the alkaline solution inflow is 0.15 fraction of the ingoing algae flow.

It is assumed that 0.75 of all the proteins is extracted during the alkaline extraction, when only proteins are extracted during the downstream processing and hexane extraction does not take place. The protein extraction is lower when first hexane extraction takes place. The fraction of proteins extracted are then assumed to be 0.64 of the total amount of proteins.

2.8 Pumping

The algae slurry has to be transported from one unit operation to the next and can be done by pumping the liquid. Since pumping requires energy, and the number of unit operation varies between routes, the energy requirements for pumping has to be taken in the energy balances.

The energy requirements for pumping the stream between unit operations is described by Wileman *et al*:

$$H_p = 2 * f * \rho_A * \frac{F_A^3 * L}{A * d} \quad \text{Eq. 2.62}$$

Where H_p is energy requirements for pumping ($^{J}/_{s}$), L is the length of the tubes (L) and is in the models 25 meters, d is the diameter of the tube (m) which is 0.1, A is the cross-sectional area (m^2) , f is the Fanning fraction and for a laminar flow of a viscous algae solution can it be calculated with:

$$f = \frac{16}{Re} \quad \text{Eq.2.63}$$

Where *Re* is the Reynolds number. A modified Reynolds number, which takes the rheological properties of the flow into account, can be calculated according to Wileman *et al* with:

$$Re = \frac{\rho_A * u^{2-n} * d^n}{8^{n-1} * K_{factor}}$$ Eq. 2.64

Where *u* is the speed of the liquid ($^{m}/_{s}$), K_{factor} is the consistency factor ($^{Poise}/_{m)}$ and *n* is the behaviour index. K_{factor} and *n* both depend on the biomass concentration, *n* has a value of one when C_A is below 50 $^{kg}/_{m3}$ and n=0.8 when the concentration of the algae slurry is above 50. *K* can be described as:

$$K = \left(7 + \frac{C_A}{\sqrt{1+0.0005(C_A-100)^2}}\right) * 10^{-4}$$ Eq. 2.65

2.9 Optimization of the models

The harvesting starts with a diluted concentration of 5 $^{kg}/_{m3}$ and will have a flow of 5 $^{m3}/_{h.}$ Therefore, the initial algae mass flow is 25 $^{kg}/_{h.}$. The models made for the different processing units are used to optimize and analyse the different routes, so a model based combinatorial optimization approach has been used. In the different models many variables are constant and based on literature results. However, some decision variables where not defined and can vary between an lower and upper border. For example in the model of Beadmill, the bead filling must have a number between 60% and 90%. The decision variables in the models are concentration factor, passes in homogenizer, pressure in homogenizer, bead filling and air flow in DAF. In order to simulate and model the DSP of algae slurry, mixed integer non-linear optimization (MINLP) is used to evaluate each route. MINLP can be used when both nonlinear (decision variables) and discrete components (constants) are used in the models. The conditions used in each process and the range of conditions for the mixed integer nonlinear optimization are shown in chapter 2.10.

During the optimization the decision variables x_d vary to determine the best process conditions. The best process conditions are based on the smallest *y-value*. When the ratio of energy requirements to product recovery (the *y-value*) is low it is considered positive. During the optimization step the best variables for each route are calculated. The best route can be determined when the best decision variables for each individual route are known. The 'best' route is determined based on the lowest *y*-value.

Minimize $y = H_{route}/X_{route}$

With $H_{route} = H_{harv} + H_{dew} + H_{disr} + H_{hex} + H_{alk} + H_{dryer}$

$X_{route} = F_{dryer} * C_{protein,dry}$ (wet routes 1-60 and dry routes 1-23)

$X_{route} = F_{hex} * C_{lipid,hex} + F_{dryer} * C_{protein,dryer}$ (wet routes 61-120 and dry 24-34)

Given equations *2.1 – 3.67*

$x_d \quad \epsilon \quad \left\{ \begin{matrix} concentration\ factor, passes\ in\ homogenizer, \\ pressure\ in\ homogenizer, beadfilling, \\ airflow\ DAF \end{matrix} \right\}$

Where H_{route} is the energy consumption of all processing units in one route in $^{J}/_{s}$ with an start concentration of 25 $^{kg\ algae}/_{h}$. H_{harv} is the energy consumption during harvesting, H_{dew} is the energy consumption during dewatering, H_{disr} is the energy consumption during disruption, H_{ex} is the energy consumption during hexane extraction (this process unit is only used for 'wet' routes 61-120 and 'dry' routes 24-34) , H_{alk} is the energy consumption during protein extraction by an alkaline solution and H_{dryer} is the energy consumption when the flow is dried by a dryer. X_{route} is the amount of product (protein and lipid) extracted during the downstream processing in $^{kg}/_{s}$. F_{dryer} is the flow after alkaline extraction consisting of the alkaline solutions and dissolved proteins and is dried in the dryer $(^{m3}/_{s})$; $C_{protein,dryer}$ is the concentration proteins dissolved in the alkaline solution after it has been dried to 700 $^{kg\ proteins}/_{m3\ alkaline\ solution}$. F_{hex} is the flow after hexane extraction consisting of hexane and dissolved lipids $(^{m3}/_{s})$; $C_{lipid,hex}$ is the concentration lipids dissolved in the hexane $(^{kg}/_{m3})$.

In total 120 wet routes and 180 dry routes are defined. These routes can be seen in appendix chapter 7.2 and 7.3. All 120 'wet' routes are calculated and conclusions are made in chapter 0. Not all 180 'dry' routes are optimized. Based on the results of the 'wet' routes, 34 'dry' routes are chosen. The process units which provided the lowest y -values in the 'wet' routes, are determined. Based on these y-values, a selection is made from the 180 'dry' routes, which results in 34 routes that are optimized. These 34 dry routes can be seen in appendix chapter 7.3.

2.10 Process conditions

The process conditions of the process units are given in Table 4.

Table 4: Process conditions of all process units

	Pressure	Flow range	Temperature	Concentration factor/End concentration	Other
Harvesting					
Centrifuge	1 bar		293 K	Up to 400 $^{kg}/_{m3}$	
Pressure filtration	1 bar		293 K	Up to 270 $^{kg}/_{m3}$	
Vacuum filtration	1 bar		293 K	Up to 370 $^{kg}/_{m3}$	
Ultrasound sedimentation	1 bar	Max flow of 0.018 $^{m,3}/_{h}$	293 K	Maximum Concentration factor of 22.5	
Flocculation	1 bar		293 K	Up to 25 $^{kg}/_{m3}$	Concentration chitosan 0.220 $^{kg}/_{m3;}$ stock pH of 8; 50-600 RPM
DAF				End concentration of 10 $^{kg}/_{m3}$	

Dewatering					
Dryer	1 bar		316 K	End concentration of 800 $^{kg}/_{m3}$	
Disruption					
Homogenizer	Between 35 bar and 415 bar		Cooled inside system, heat production of 2 K for every 100 bar	Between 10 $^{kg}/_{m3}$ and 300 $^{kg}/_{m3}$	Between 1 and 10 passes
Bead Milling		Max flow of 0.450 $^{m3}/_{h}$	Inflow cooled to 278 K	Between 10 $^{kg}/_{m3}$ and 200 $^{kg}/_{m3}$	Bead filling between 60% and 90%
Calander	1 bar		323 K	Between 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3}$	
Extraction					
Hexane extraction	1 bar		293 K		0.15 $^{m3}/_{h}$ hexane for 1 $^{m3}/_{h}$ disrupted algae stream. u_m is the stirrer speed = 10 $^{1}/_{s}$ τ is the residence time = 3600 s
Alkaline extraction	1 bar		298 K		0.1 $^{m3}/_{h}$ alkaline solution for 1 $^{m3}/_{h}$ disrupted algae stream u_m is the stirrer speed = 1.67 $^{1}/_{s}$ τ is the residence time = 3600 s

3 Results and Discussion

3.1 Energy requirements versus product recovery

3.1.1 'Wet' routes

All combinations of the available unit operations were evaluated for the 'wet' routes. These routes are defined in appendix 7.2 are also defined in superstructure Figure 2-1. In Figure 3-1 the *y-values* of the first 60 routes are shown. The *y-value* is the ratio of the energy requirements to the amount of product obtained (*y-value* is described in chapter 2.9).

Routes 1-60 are the 'wet' routes, in these routes only proteins are extracted. Routes 1-20 have a concentration of 50 $^{kg}/_{m3}$ when it enters the disruption step. For routes 21-40 the concentration of disruption is 100 $^{kg}/_{m3}$ and for routes 41-60 this is 150 $^{kg}/_{m3.}$ Route 1-4 uses a centrifuge as harvesting step, route 5-8 uses ultrasound sedimentation during dewatering, routes 9-12 has flocculation as harvesting method, routes 13-16 uses DAF and routes 17-20 makes use of filtration. This same order is also used for routes 21-40 and 41-60. HPH and Beadmill are used as disruption method. The Beadmill is used in the routes with an even route number. HPH is used in the routes with an odd route number.

Figure 3-1 shows that route number 51, 53 and 55 have the lowest *y-value*. Route 51 uses flocculation as harvesting method, followed by filtration and HPH. Route 53 start with DAF, followed by filtration and HPH. Route 55 starts with a DAF, uses a centrifuge as dewatering step and HPH as disruption step. A low *y-value* is favourable because the energy requirements per kg of product is low. However, the *y-value* does not say anything about the product recovery or energy use separately. Therefore the energy requirements in $^{J}/_{s}$ and the product recovery in $^{kg}/_{h}$ are determined separately. Figure 3-2 shows the energy requirements for 'wet' routes 1-20. Figure 3-3 gives the product recovery of 'wet' routes 1-20.

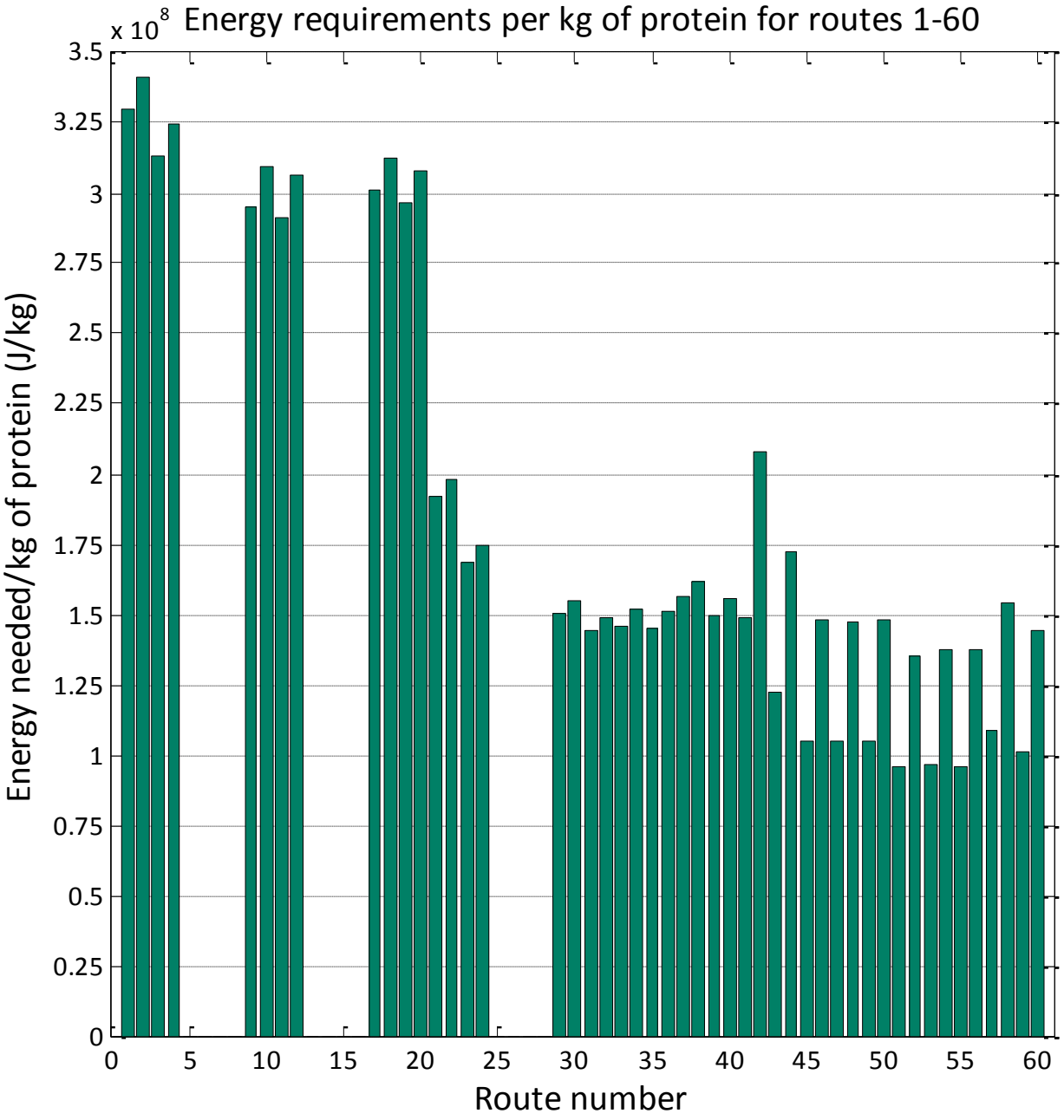


Figure 3-1: Energy needed per kg of protein (J/kg) for routes 1 to 60. The total amount of energy which is used is divided by the total amount of protein mass which is recovered after the last drying step. The y-as gives the y-value and the x-as is the route number. Routes 1-60 only contain the alkaline extraction step to extract proteins. In these routes the hexane extraction step does not take place. Routes 1-60 are described in appendix 7.2. Route 1 to 20 all have a concentration of 50 kg/m^3 when it enters the disruptions step. Routes 21-40 have a start concentration of 100 kg/m^3 when it enters the disruptions step. Routes 41-60 have a concentration of 150 kg/m^3 when it enters the disruptions step. Every four routes another harvesting processing unit is used. Starting with centrifuging (step 1-4), Ultrasound sedimentation (route 5-8), followed by flocculation (route 9-12), Dissolved Air Flotation (route 13-16) and filtration (route 17-20). This order is also used for routes 21-60. Centrifuging and filtration are used as dewatering method. The routes which are an even number contain a Beadmill as disruption unit. The routes which are odd numbers contain an HPH.

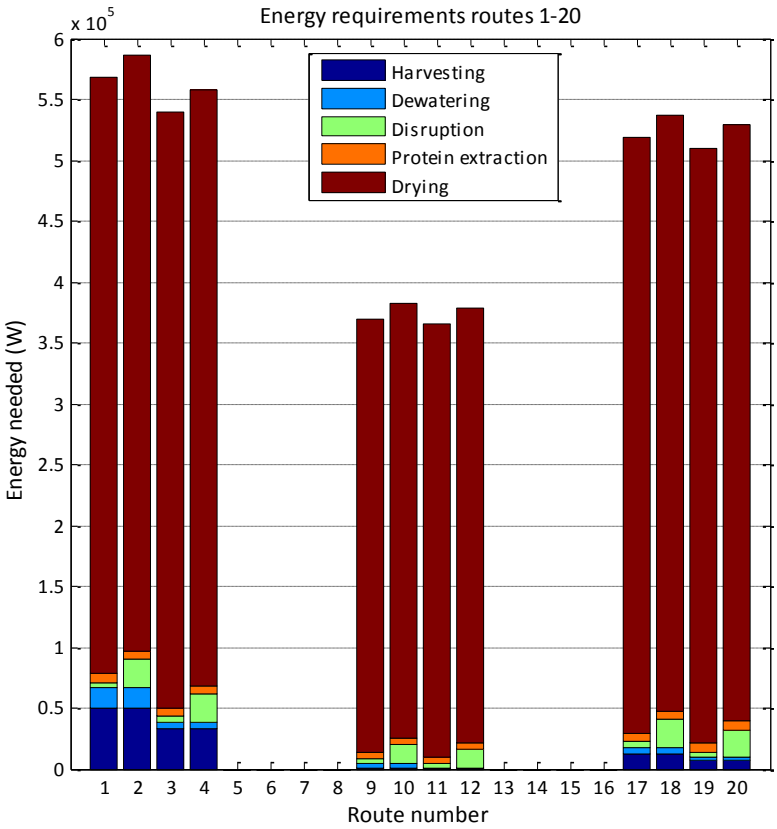


Figure 3-2: Energy requirements in $^{J}/_{s}$ for routes 1 to 20. The different process units are stacked, so it is possible to see how much energy each process unit needs.

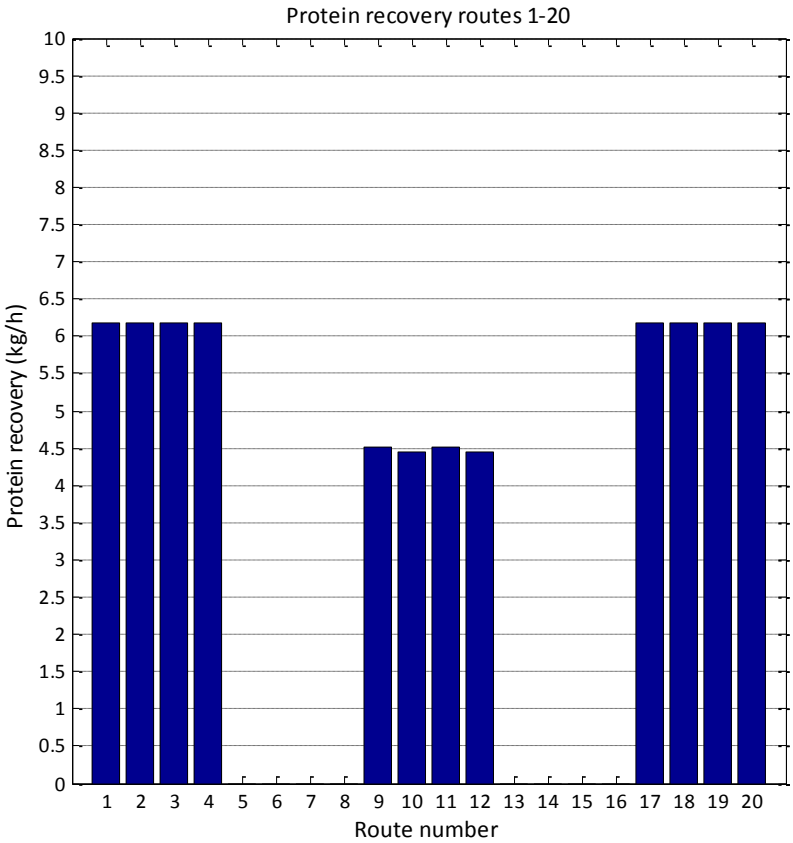


Figure 3-3: Protein recovery in $^{kg}/_{h}$ of routes 1 to 20, with a starting algae mass flow of 25 $^{kg}/_{h.}$

In routes 1-20 the disruption concentration is 50 $^{kg}/_{m3,}$ this means that the harvesting and dewatering step together cannot concentrate more than 10 times, because the start concentration is 5 $^{kg}/_{m3}$. Route 5-8 and routes 13-16 cannot occur, since the harvesting step results in an algae slurry which is more concentrated than 50 $^{kg}/_{m3.}$ In routes 5-8 Ultrasound sedimentation is applied as harvesting step. In routes 13-16 DAF is used to harvest the microalgae. With these two harvesting units it is not possible to control the concentration factor and concentrate more than 10 times, therefore exceeding the disruption concentration of 50 $^{kg}/_{m3}$.

The most important observations from the Figure 3-2 and Figure 3-3 are:

- The drying step requires the most energy. In these routes drying takes only place during the downstream processing after the alkaline extraction. Drying is used to concentrate the stream to the end concentration. In the 'dry' routes the dryer can also be used as a dewatering method, which is not the case in the 'wet' routes. When disruption takes place the concentration is 50 $^{kg}/_{m3,}$ after disruption and extraction the diluted stream must be concentrated to 700 $^{kg\ protein}/_{m3.}$ This change in concentration requires a lot of energy and is done by the dryer. Not only is the drying step very big, the energy requirements of a dryer is considerable larger than any other dewatering/concentrating process unit.
- Figure 4-2 shows that the first four routes uses the most energy. In these routes a centrifuge is used as harvesting step. In route 1 and route 2 two centrifuges are a both as harvesting and dewatering step, which results in the highest energy use. Centrifuging requires a lot of energy which also shows Table 3.
- Route 9 - 12 uses flocculation as a harvesting method, this process unit has the lowest energy use. However the protein recovery is also much lower. It can be concluded that flocculation doesn't require as much energy as filtration and centrifugation do, but also results in a lower algae biomass recovery.
- It is more advantageous to use a high pressure homogenizer (HPH) compared to a Beadmill for disruption. In Figure 3-2 it can be seen that the disruption step requires less energy when a HPH is used. The efficiency is comparable, since the protein recovery between for example route number 1 and 2 is more or less the same.

In Figure 3-4, Figure 3-5, Figure 3-6 and Figure 3-7 the energy requirements and protein recovery for routes 21 to 60 are shown. Routes 21-40 the concentration when disruption takes place is 100 $^{kg}/_{m3}$ and in routes 41-60 the disruption concentration is 150 $^{kg}/_{m3.}$ The order of routes is the same between routes 21-40 and 41-60 (for example route 23 and 43 are the same), only the disruption concentration differs.

From these figures several observations are made:

- Figure 3-4 shows that the overall energy use the route 21-40 are much lower than routes 1-20. This is because the concentration when disruption takes place is twice as high as route 1-20. Also in these routes the drying steps requires a lot of energy because the stream his concentrated from around 100 $^{kg}/_{m3}$ to 700 $^{kg}/_{m3}$. Drying requires a lot of energy, so concentrating more during harvesting and dewatering, which happens in routes 21-40, is favourable. This can also be seen in Figure 3-7, where the disruption concentration in routes 41-60 is 150 $^{kg}/_{m3}$.
- In Figure 3-6 it becomes clear that the protein recovery differs a lot between two almost equal routes due to the disruption step, like route 41 and 42. The protein recovery of route 41 (HPH) is much higher than the recovery in route 42 (Beadmill). The beadmill is less efficient when the algae concentration becomes higher, therefore resulting in a lower protein recovery.
- In route 33-37 DAF is used as harvesting method. Figure 3-5 shows that the DAF method results in a very high protein recovery and average energy use. The routes with the highest recovery contain the DAF as harvesting method.
- In Figure 3-6 Ultrasound sedimentation is used in routes 45-48. This method requires the least amount of energy for its concentration factor compared to the other harvesting units, however the protein recovery is also very low.
- The best routes contain either DAF or flocculation as harvesting step, followed by filtration as dewatering step and HPH as disruption step. These routes have the lowest *y-value.* DAF has a very high recovery and the energy requirements are average. Flocculation has little energy requirements but also a lower protein recovery. Filtration is more favourable than centrifuging when it is applied as dewatering method, because the energy requirements are lower.
- The route which is the least favourable, so with the highest *y-value*, uses two centrifuges as harvesting and dewatering step and beadmill as disruption step.
- The highest losses in algae occurs in routes 46 and 48. Here Ultrasound sedimentation is used as harvesting step and beadmill as disruption step. Ultra sound sedimentation is not efficient, because it has low algae recovery. At low disruption concentration is the disruption efficiency of the Beadmill comparable with HPH, at higher disruption concentrations the Beadmill is less efficient.
- The total amount of needed energy is lower in routes 41 – 60 compared to routes 1-40, because the stream is more concentrated when entering the disruption step and less drying is needed in the last step.

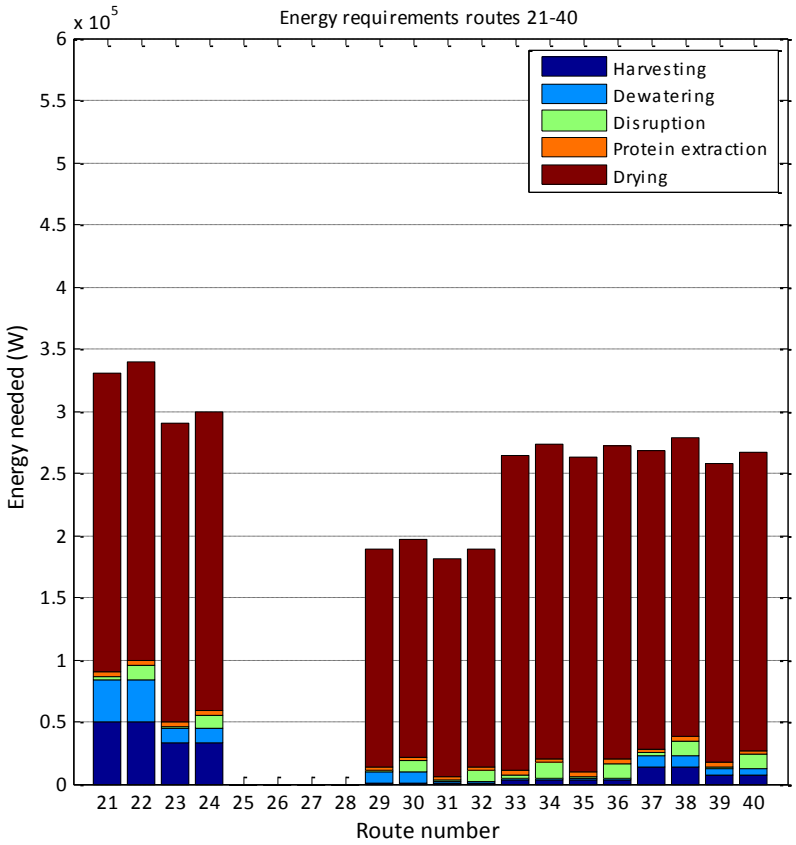


Figure 3-4: Energy requirements in $^{J}/_{s}$ for routes 21 - 40. The different process units are stacked, so it is easy to see how much energy each process unit needs.

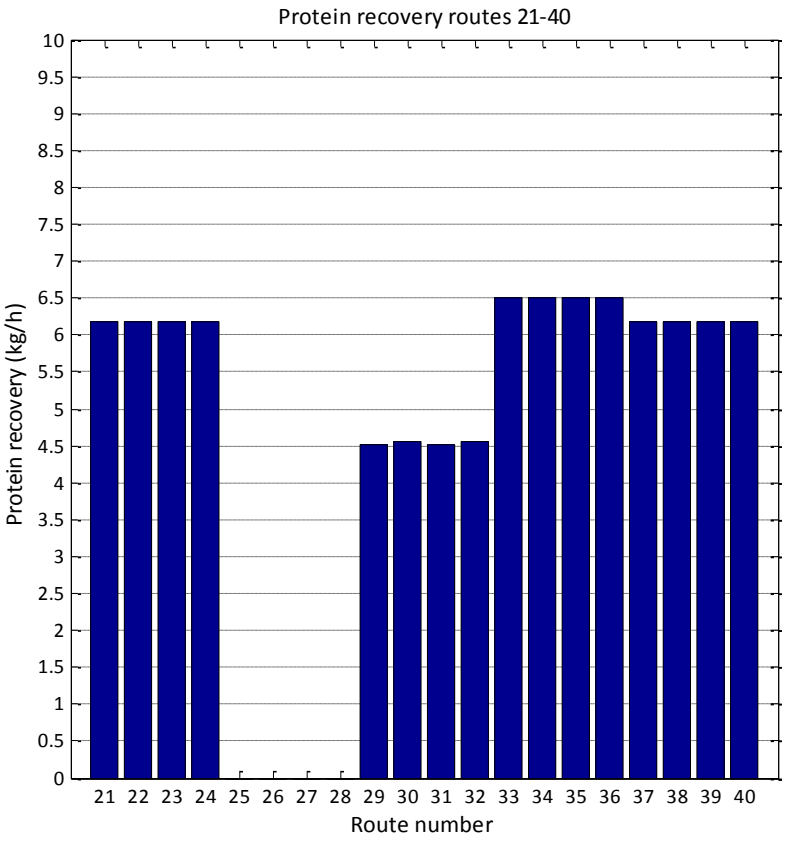


Figure 3-5: Protein recovery in $^{kg}/_{h}$ of routes 21 - 40, with a starting algae mass flow of 25 $^{kg}/_{h}$.

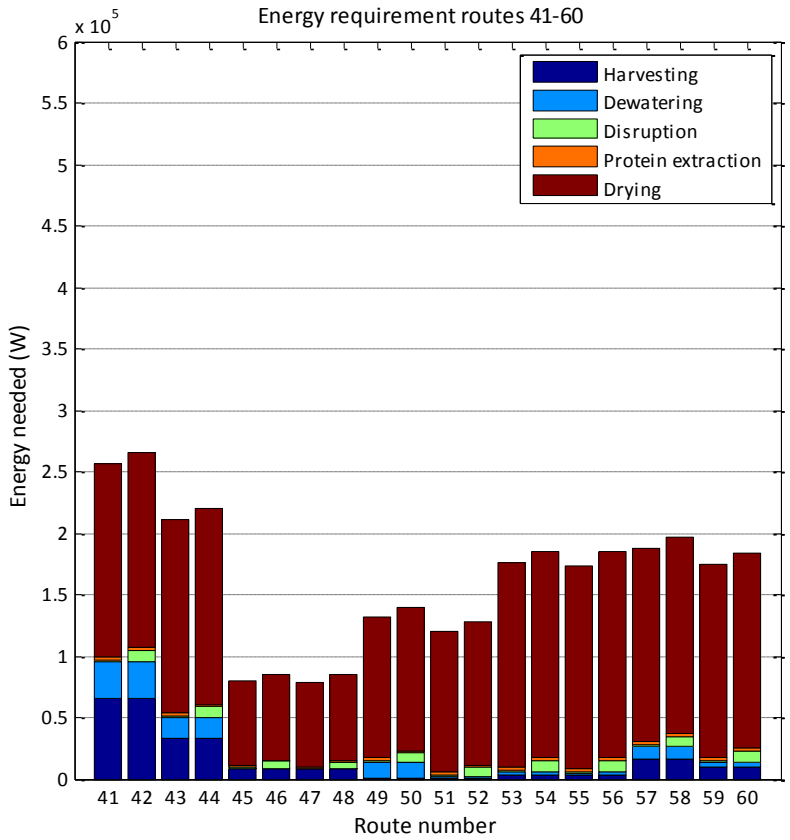


Figure 3-6: Energy requirements in $^{J}/_{s}$ for routes 41 - 60. The different process units are stacked, so it is easy to see how much energy each process unit needs.

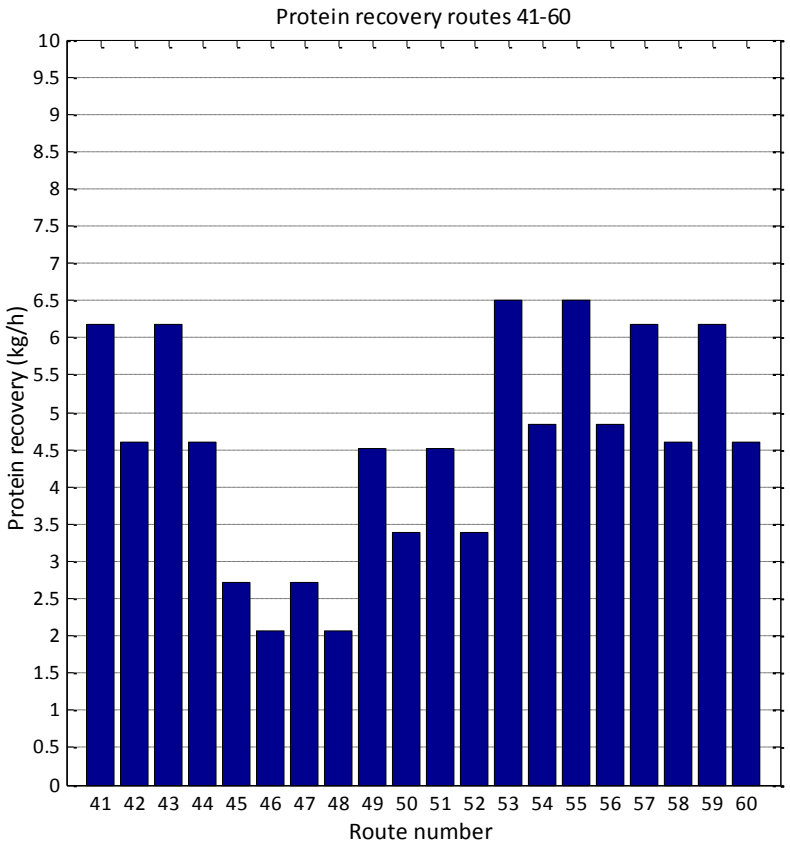


Figure 3-7: Protein recovery in $^{kg}/_{h}$ of routes 41 - 60, with a starting algae mass flow of 25 $^{kg}/_{h}$.

Routes 61-120 are defined in appendix 7.2 and in superstructure Figure 2-2. These routes have the same order of process units as routes 1-60, but there is one big difference: the hexane extraction step is added, to extract lipids from the stream. Routes 61-80 have a concentration of 50 $^{kg}/_{m3}$ when it enters the disruption step. For routes 81-100 the concentration of disruption is 100 $^{kg}/_{m3}$ and for routes 101-120 this is 150 $^{kg}/_{m3.}$. Figure 3-8 shows the *y-values* of routes 61-120. This figure shows that routes 111,113 and 115 are most favourable, since they have the lowest *y-value.* Route 111 uses flocculation as harvesting method, followed by filtration and HPH. Route 113 start with DAF, followed by filtration and HPH. Route 115 starts with a DAF, uses a centrifuge as dewatering step and HPH as disruption step.

In Figure 3-9, Figure 3-11, and Figure 4-13 the energy requirements for routes 61-120 are shown. In Figure 3-10, Figure 3-12 and Figure 3-14 the product recovery (proteins + Lipids) is given. Both proteins and lipids have the same value, no distinction is made between the two products.

Many observations of routes 61-120 are the same as routes 1-60, however also some additional conclusions can be made:

- The *y-values* are lower in routes 61-120 compared to routes 1-60, even though the routes have the same process units composition (like route 1 and route 61). The only difference is the additional step of hexane extraction. The hexane extraction requires a lot of energy thus the total amount of energy for the DSP for each route is higher, but also hexane extraction also provides a significant amount of lipids. When Figure 3-1 and Figure 3-8 are compared it can be concluded that it is more favourable to extract both lipids and proteins from the algae stream. Hexane extraction requires an significant amount of energy and it lowers the protein recovery, but the *y value* is still lower because the product recovery is much higher, due to the extraction of lipids.
- When extracting lipids, the amount of protein that is recovered, is lower. However, the sum of lipid- and protein recovery is higher than the recovery of only proteins. It is favourable to extract both components from the algae stream.
- Figure 4-9 shows that the hexane extraction requires a lot of energy. The amount of energy needed for hexane extraction is coupled to the amount of hexane used. In some routes the energy requirements for hexane extraction are lower. This is because the amount of hexane used is proportional to the amount of available lipids. When Ultrasound sedimentation is used as harvesting step, a lot of algae are lost. This results in a lower amount of disrupted algae and therefore in a lower amount of available lipids.

It is observed that it is most favourable to use a HPH as disruption method and DAF , flocculation and filtration as harvesting method. The figures show that it is more favourable to concentrate as much as possible during harvesting and dewatering, to minimize the

drying step which takes place after the extraction step. Routes 41-60 and routes 101-120 need a lot less energy. This was also expected. Drying requires a lot of energy so it is better to concentrate the stream as much as possible to keep the drying as low as possible.
In the introduction one of the main questions is: "*What provides the lowest y-value, extraction of proteins or extraction of both proteins and lipids?".* When the routes are compared with each other, it can be observed that the (average) *y-value* of routes 61-120 are lower than routes 1-60. Thus it is more favourable to extract both proteins and lipids.

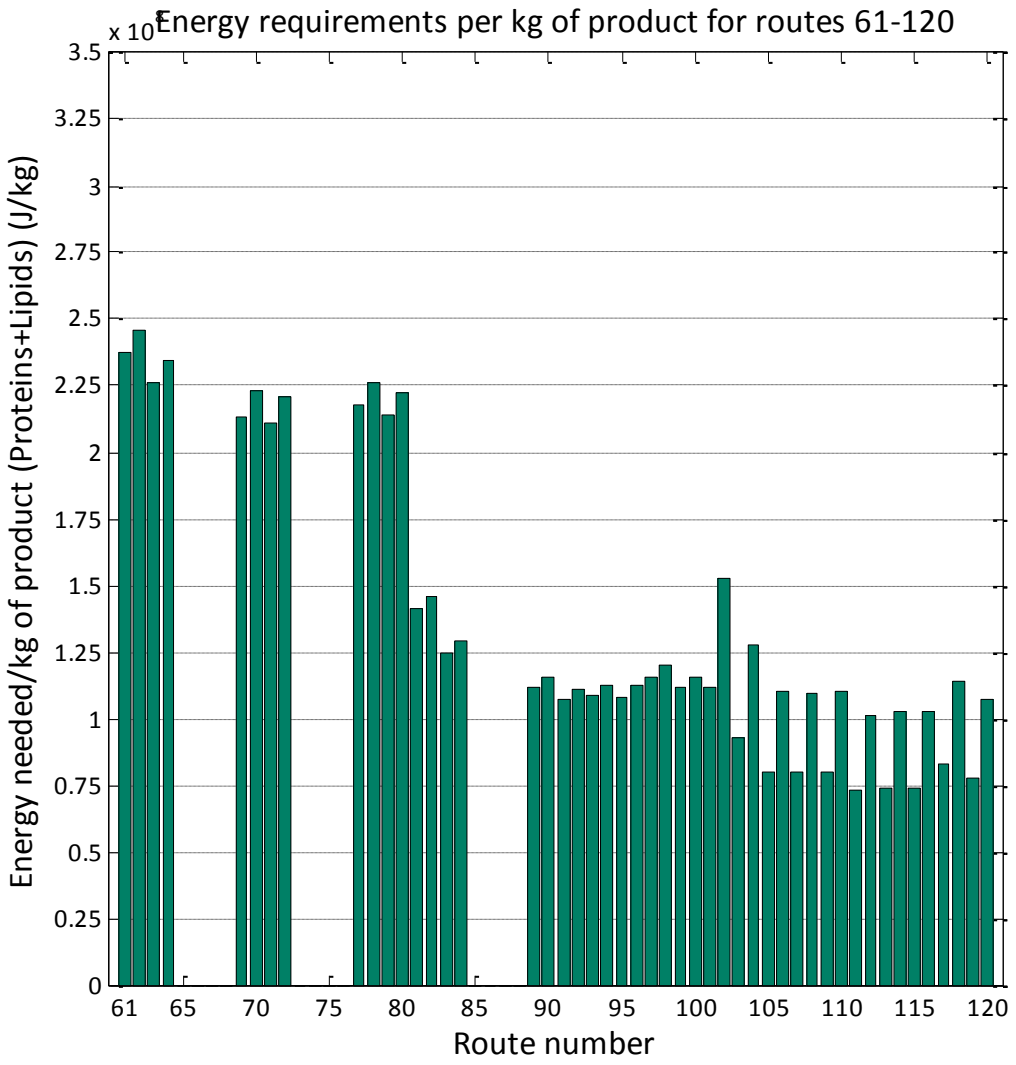


Figure 3-8: Energy needed per kg of product (J/kg) for routes 61 to 120. In these routes both protein and lipids are extracted from the disrupted algae broth. After disruption first the hexane extraction takes place. This is followed by protein extraction using an alkaline solution. In this chart the total amount of energy needed for all processing units is divided by the total amount of protein mass and lipid mass which is recovered, called *y*. Routes 61-120 are described in appendix chapter 7.2 Route 61 to 80 all described routes where the concentration of the algae stream is 50 kg/m^3 when it enters the disruptions step. Routes 81-100 where the concentration of the algae stream is 100 kg/m^3 when it enters the disruption step. In routes 101-120 the disruption concentration of the algae stream is 150 kg/m^3. Every four routes another harvesting processing unit is used. Starting with centrifuging (step 61-64), Ultrasound sedimentation (route 65-68), followed by flocculation (route 69-72), Dissolved Air Flotation (route 73-76) and filtration (route 77-80). This order is also used for routes 81-120. The routes which are an even number contain a Beadmill as disruption unit. The routes which are odd numbers contain an HPH.

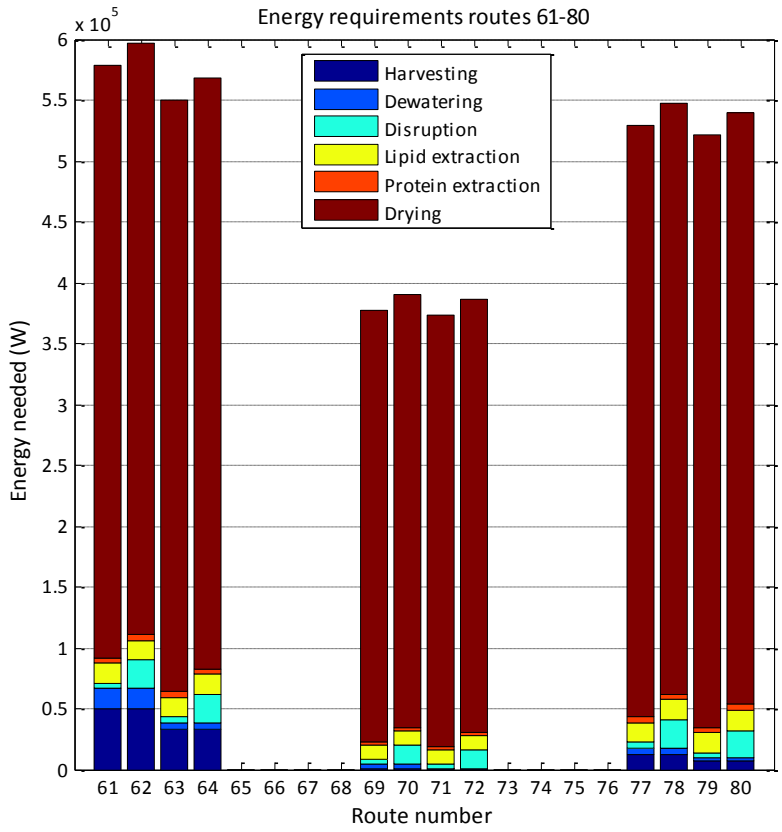


Figure 3-9: Energy requirements in $^{J}/_{s}$ for routes 61 - 80. The different process units are stacked, so it is easy to see how much energy each process unit needs.

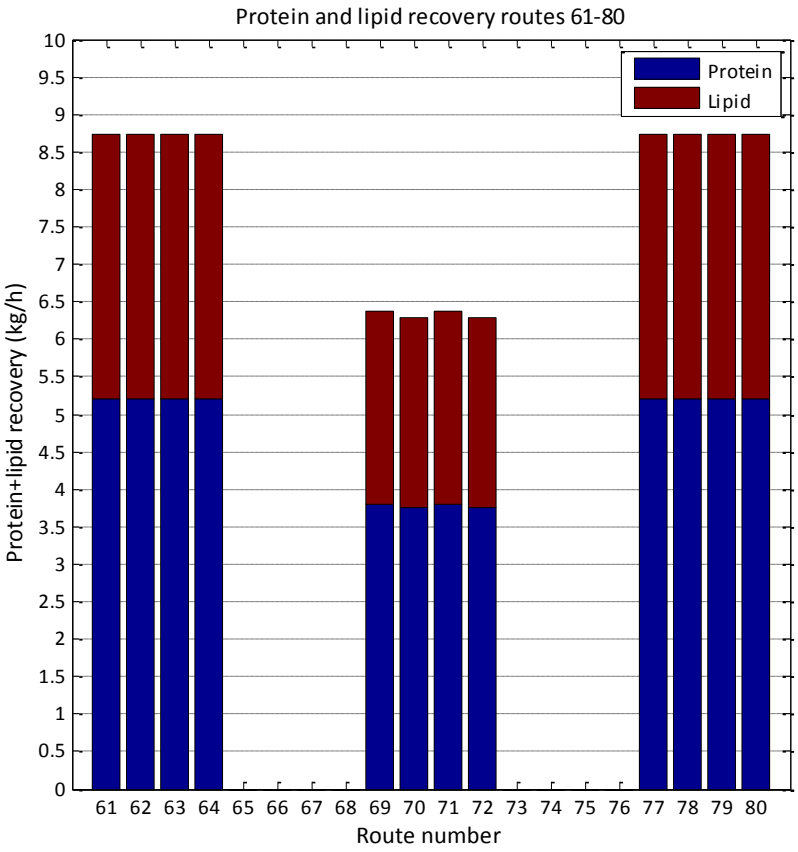


Figure 3-10: Protein and lipid recovery in kg/h of routes 61 - 80, with a starting algae mass flow of 25 kg/h.

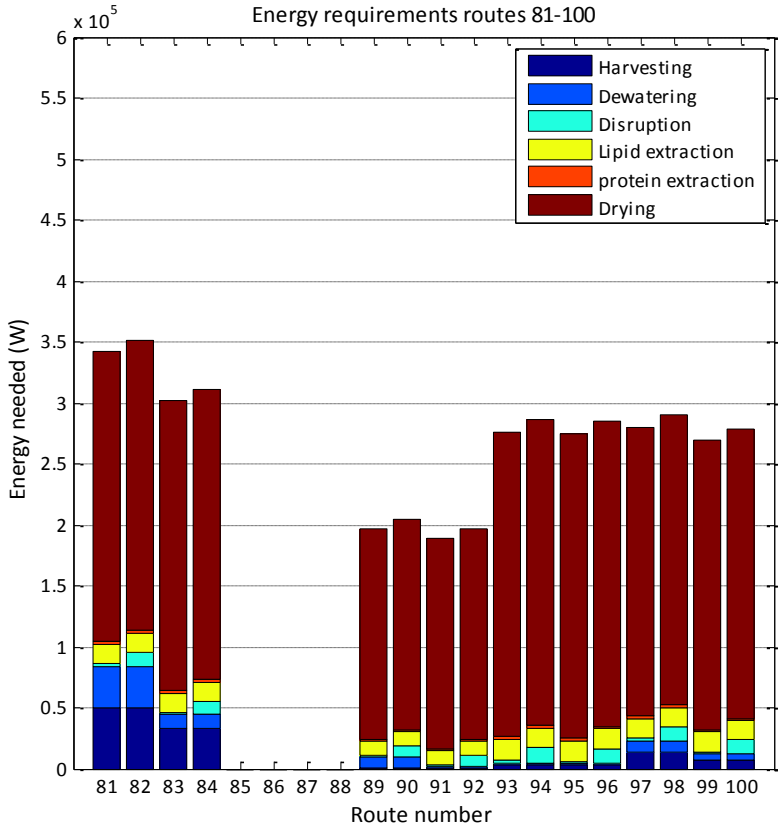


Figure 3-11: Energy requirements in $^{J}/_{s}$ for routes 81 - 100. The different process units are stacked, so it is easy to see how much energy each process unit needs.

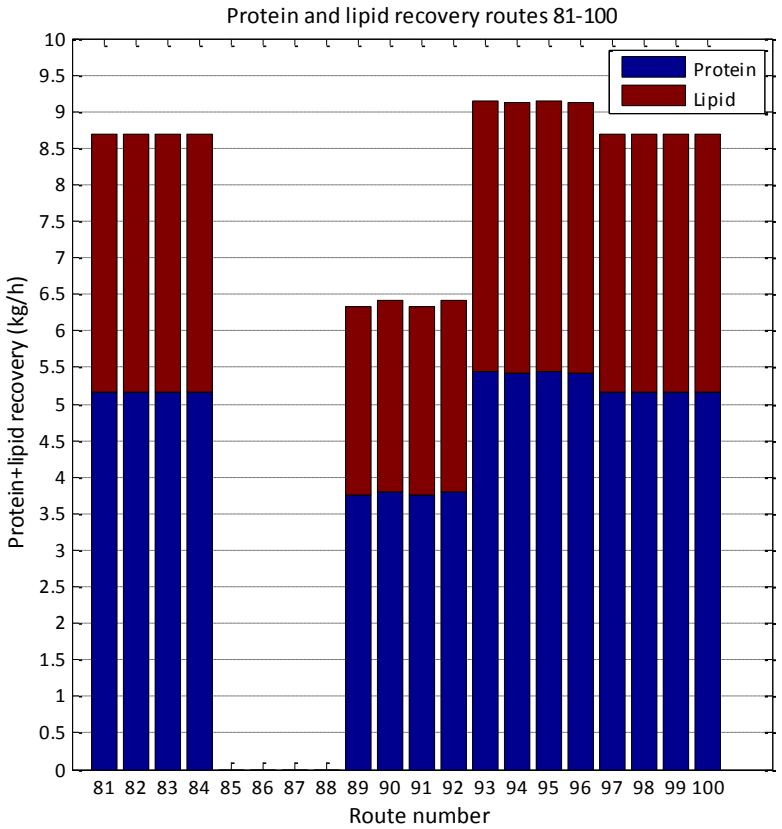


Figure 3-12: Protein and lipid recovery in $^{kg}/_{h}$ of routes 81 - 100, with a starting algae mass flow of 25 kg/h.

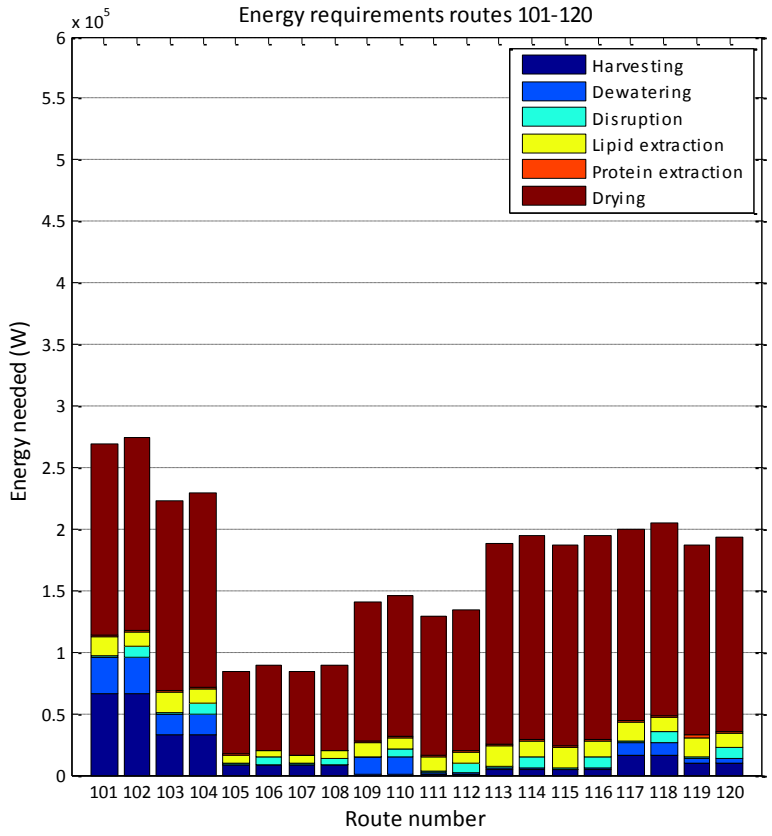


Figure 3-13: Energy requirements in $^{J}/_{s}$ for routes 101 - 120. The different process units are stacked, so it is easy to see how much energy each process unit needs.

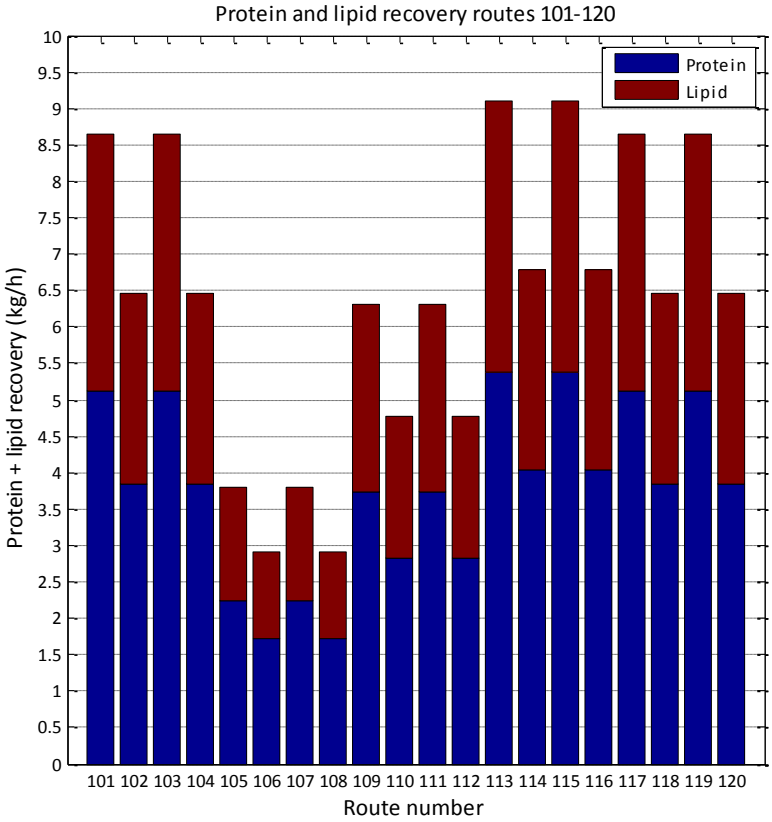


Figure 3-14: Protein and lipid recovery in $^{kg}/_{h}$ of routes 81 - 100, with a starting algae mass flow of 25 kg/h.

3.1.2 'Dry' routes

180 'dry' routes are defined, based on all possible combinations of process units and process conditions. These 180 routes are shown in appendix 7.3. Of these 180 'dry' routes, 34 routes are chosen for the optimization and evaluation. The 34 dry routes are selected based on the results of the wet routes. The dry routes that contain process units which are also used in 'wet' routes with low *y-values,* like DAF and flocculation, are chosen. The list with the 34 routes are shown in appendix 7.3.

In some 'dry' routes the dryer is not only used in the final stage of the downstream processing, to the concentrate the stream to the end concentration of 700 $^{kg}/_{m3,}$ but is also used as second dewatering step. Figure 2-3 and Figure 2-4 show the superstructures from the 'dry' routes. In these figures it can be seen that the dryer can also be used as a dewatering step.

Figure 3-1 show the *y-values* of dry routes 1-23. In these routes only protein is extracted. In routes 1-8 the concentration of the algae stream is 200 $^{kg}/_{m3}$ when it enters the disruption step. Routes 9-13 have a concentration of 300 $^{kg}/_{m3}$ when disruption takes place. Routes 14-18 and routes 19-23 have a disruption concentration of 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3}$ respectively. Routes 1-13 use an HPH as disruption method. Route 4 is an exception, in this route a Beadmill is used. In this route a Beadmill is used to see how efficient the Beadmill functions at this concentration and to compare it with an HPH. Routes 14-23 use a calander as disruption method.

The *y-values* of these dry routes are significantly lower than the *y-values* of most the 'wet' routes. Except from 'dry' routes 1, 4 and 19, all other dry routes are lower than the 'wet' routes. Why the *y-values* of routes 1,4 and 19 are higher, is explained on page 52 .

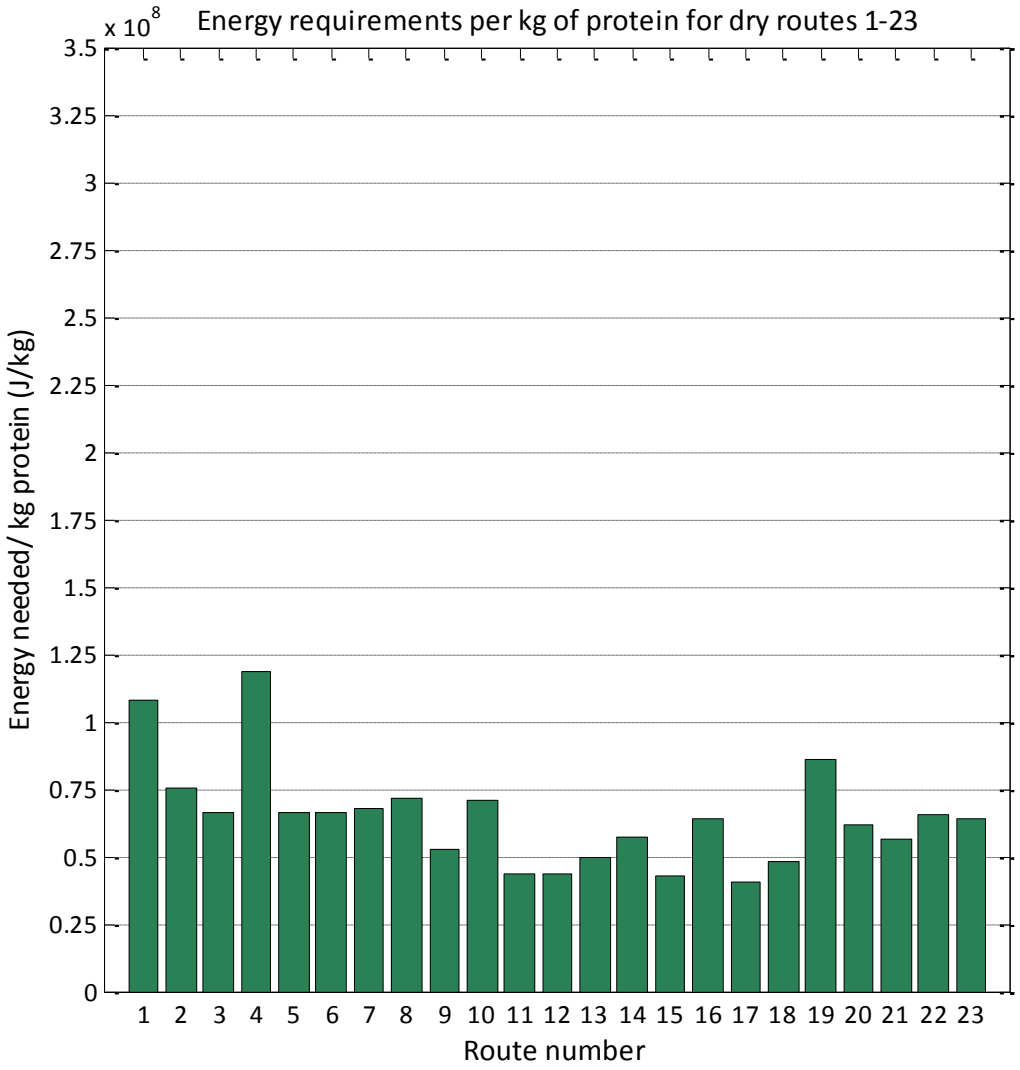


Figure 3-15: Energy needed per kg of product (J/kg) for dry routes 1-23. In these routes only protein is extracted from the disrupted algae broth. In the bar chart the total amount of energy needed for all processing units is divided by the total amount of protein mass which is recovered. Routes 1-23 are described in appendix 7.3. In routes 1 to 8 the concentration of the algae stream is 200 kg/m^3 when it enters the disruptions step. Routes 9-13 all have a concentration of 300 kg/m^3 when disruption takes place Routes 14-18 and routes 19-23 uses a disruption concentration of 400 kg/m^3 and 800 kg/m^3 respectively. Routes 1-13 use an HPH as disruption method. Route 4 is an exception, in this route a Beadmill is used Routes 14-23 use a calander as disruption method. Routes 1-13 use a centrifuge as second dewatering step; route 5, 7 and 10 are exceptions. These routes use a dryer as second dewatering step. Filtration is used as harvesting step in routes 8,13,18,22 and 23. Ultrasound sedimentation is used as harvesting step in routes 2,9,10,14 and 19. Flocculation is used as harvesting step in routes 3,4,5,11, 15,16 and 20. Dissolved Air Flotation is used as harvesting step in routes 6,7,12, 17 and 21. The centrifuge is used as harvesting step in route 1.

From this figure the observation is made that route 17 has the lowest *y-value*. This route uses DAF as harvesting step, followed by filtration and centrifuging. The calander is used for the disruption and the disruption takes place at a concentration of 400 $^{kg}/_{m3.}$ A low *y-value* is favourable, but does not say anything about the product recovery or energy requirements separately. In Figure 3-16 and Figure 3-17 the energy requirements in $^{J}/_{s}$ and the product recovery in $^{kg}/_{h}$ are determined separately.

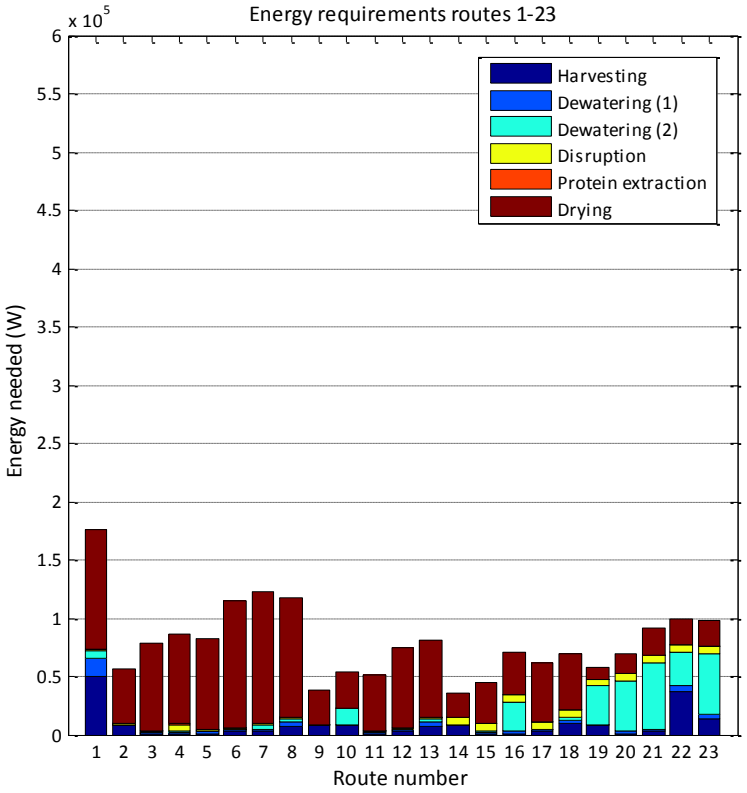


Figure 3-16: Energy requirements in $^{J}/_{s}$ for dry routes 1 - 23. The different process units are stacked, so it is easy to see how much energy each process unit needs.

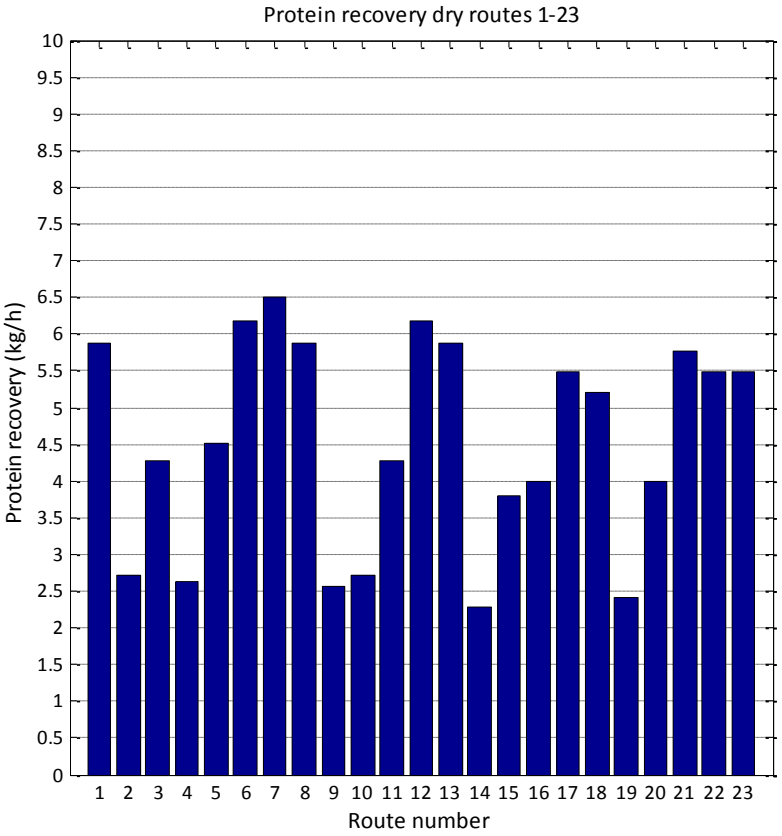


Figure 3-17: Protein recovery in $^{kg}/_{h}$ in routes 1-23, with a starting algae mass flow of 25 kg/h.

From Figure 3-16 and Figure 3-17 the following observations are made:

- Route 1 uses centrifuging both for harvesting and (2 times) dewatering. The disruption concentration is 200 $^{kg}/_{m3,}$ thus the centrifuge has to concentrate 40 times (start concentration of 5 $^{kg}/_{m3}$) . The centrifuge requires a lot of energy, compared to other harvesting methods, which can clearly be seen in figure 4-16.
- Routes 1-8 have a disruption concentration of 200 $^{kg}/_{m3}$ and therefore the energy use for drying to the end concentration of 700$^{kg/}_{m3}$ is high. In routes 9-13 dewatering of the algae broth takes place to a final concentration of 300 $^{kg}/_{m3,}$ the energy use for drying to the end concentration is therefore less. In routes 14-18 and routes 19-23 the concentration when disruption takes place are 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3,}$ respectively. This can be seen in the graph. At higher disruption concentrations, the amount of energy needed for harvesting and dewatering increases and the amount of energy needed for drying to the end concentration decreases.
- The use of Ultrasound sedimentation results in low energy use, but also in low protein recovery, as can be seen in routes 2,9,10,14 and 19. It is not favourable to use Ultrasound sedimentation as a harvesting method.
- The protein recovery difference between route 9 and 10 is caused by the second dewatering step. Both routes are almost identical except from the second dewatering step. For route 9 a centrifuge is used. Route 10 uses a dryer as second dewatering process unit. When a centrifuge is used, there is a 5% loss of algae biomass. When drying is used, there is no loss in algae. Thus, the protein recovery is a bit higher when a dryer is used as second dewatering step.
- Disruption by a calander requires more energy than disruption by an HPH. This can be seen in Figure 3-16 in routes 14-23. Even though the calander requires more energy than an HPH, in some cases it is more advantageous to use the calander. For example: the only difference in route 12 and route 17 is the disruption concentration and the disruption method. The figure shows that it is energetically more favourable to concentrate up to 400 $^{kg}/_{m3}$ and then using a calander, than concentrating up to 300 $^{kg}/_{m3}$ followed by disruption with an HPH. Figure 3-16 show that it requires more energy to disrupt at 300 $^{kg}/_{m3}$ with the use of an HPH and to dry the stream to the end concentration (route 12), than to disrupt the algae cells at a concentration of 400 $^{kg}/_{m3}$ with a calander and to dry the stream to the end concentration (route 17).
- Route 5 and 7 are almost equal except from the harvesting step. In route 5 flocculation is used and in route 7 DAF is used as harvesting method. The energy use of flocculation is lower than DAF, but the algae recovery is also lower. This results in almost equal *y-values.* This is observed from Figure 3-15, Figure 3-16 and Figure 3-17.

Figure 3-18 shows the *y-values* of 'dry' routes 24-34. In these 'dry' routes both protein and lipids are extracted. In routes 24-26 the concentration of the algae stream is 200 $^{kg}/_{m3}$ when it enters the disruption step. Routes 27 and 28 have a concentration of 300 $^{kg}/_{m3}$ when disruption takes place. Routes 29-30 and routes 31-34 have a disruption concentration of 400 $^{kg}/_{m3}$ and 800 $^{kg}/_{m3}$, respectively. The list with the routes 24-34 are shown in appendix 7.3.

Figure 3-19 and Figure 3-20 show the energy requirements and product recovery for 'dry' routes 24 – 34.

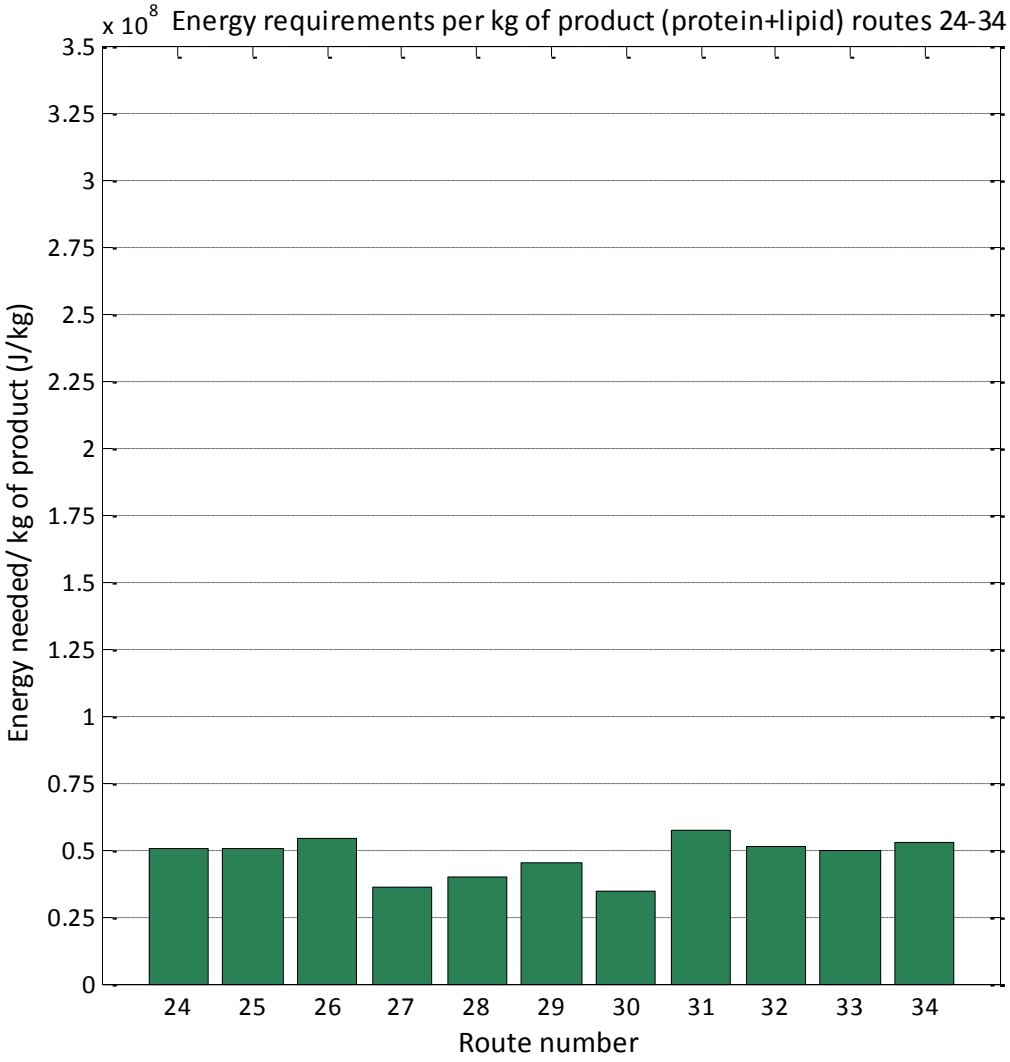


Figure 3-18: : Energy needed per kg of product (J/kg) for dry routes 24-34. In these routes both proteins and lipids are extracted. In the bar chart the total amount of energy needed for all processing units is divided by the total amount of protein and lipid mass which is recovered. Routes 23-34 are described in appendix chapter 7.3. In routes 24 – 26 the concentration of the algae stream is 200 kg/m^3 when it enters the disruptions step. In routes 27 and 28 the concentration of the algae stream is 300 kg/m^3. Routes 29-30 and routes 31-34 use a disruption concentration of 400 kg/m^3 and 800 kg/m^3 respectively. Routes 24 to 30 use filtration as dewatering step 1 and centrifuge as dewatering step 2. Routes 24 to 28 uses an HPH as disruption method and routes 29 to 34 uses a calander as disruption unit. Filtration is used as harvesting step in routes 26 , 28, 33 and 34. Flocculation is used as harvesting step in routes 24,27,31 and 32. Dissolved Air Flotation is used as harvesting step in routes 25 and 30. Ultrasound sedimentation is used as harvesting method in route 29.

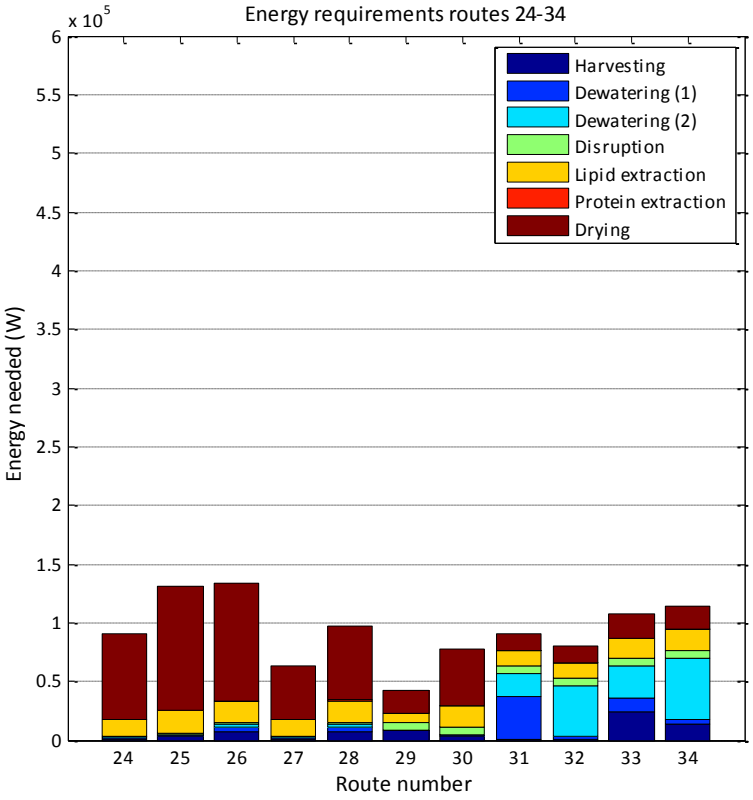


Figure 3-19: Energy requirements in $^{J}/_{s}$ for dry routes 24 - 34. The different process units are stacked, so it is easy to see how much energy each process unit needs.

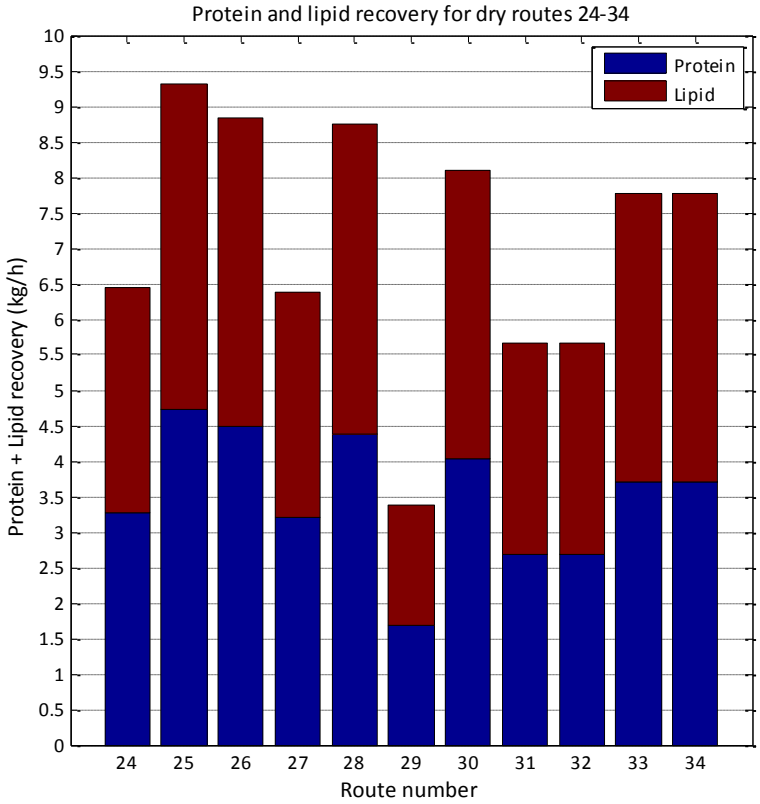


Figure 3-20: Protein and lipid recovery in $^{kg}/_{h}$ of routes 24-34, with a starting algae mass flow of 25 kg/h.

Many observations from Figure 3-19 and Figure 3-20 are the same as the previous mentioned observations in this chapter.

In Figure 3-20 it is observed that the protein recovery is significantly lower than in Figure 3-17. When routes 3 and 24 are compared, which are identical except for the addition of the lipid extraction step in route 24, it becomes clear that the protein recovery is lower in route 24. This is because the extraction of lipid by hexane has an negative effect on the protein recovery. The amount of protein recovered is about 20% lower due to the hexane step. The energy requirements of route 24 are also higher, due to the addition of the hexane extraction step. But the *y-value* of route 24 is lower than route 3 because of the lipid recovery.

In the introduction the following question is defined: "*Which of the two methods, 'wet' or 'dry' downstream processing, is energetically more favourable?".* When Figure 3-2, Figure 3-4, Figure 3-6, and Figure 3-16 are compared with each other, it can be seen that the energy requirements for the 'dry' routes is much lower. This is energetically more favourable because the drying step at the end of the downstream processing is much smaller in the 'dry' routes.

In the introduction one more question is defined: "*Which of the process unit combinations at what conditions results in the lowest y-value, for the downstream processing of microalgae biomass?*". From all the figures shown in this chapter it is observed that 'dry' route 30 has the lowest *y-value*. In this route the disruption concentration is 400 $^{kg}/_{m3}$ and the process units used are DAF, filtration, centrifugation and Calander. The amount of energy needed is 7.7 * 10^{4} $^{J}/_{s}$ and this route provides more than 8 kg of product per hour.

3.1.3 Variables

As described in chapter 2.9, there are five different decision variables: concentration factor, amount of passes in HPH, pressure in homogenizer, bead filling in Beadmill and air flow in DAF. These variables have constraints: an upper and lower border. These constraints can be found in chapter 2.10. During the optimization all the possible variables between the two borders are calculated for each route. For each individual route the best decision variables are defined. The most favourable decision variable is based on the lowest *y-value*.

The routes where an HPH is used the amount of passes used is always one. The corresponding pressure which is used is 420 bar. The best results for all routes which use an HPH is 1 pass in the HPH with an corresponding pressure of 420 bar. The amount of passes increases the disruption percentage, but only with a very small difference. However the amount of energy needed to process the broth several times with the homogenizer (an increased amount of passes) has an big effect on the energy use. Therefore the lowest amount of passes and the highest pressure, results in high disruption and relative low energy use.

For the Beadmill a bead filling of 90% is always used as the optimal percentage. This is because a higher bead filling results in more disruption, but the bead filling has no effect on the energy use. This why the upper boundary for this variable is chosen as most optimal one.

For all routes the airflow used in DAF is 0.500 $^{kg}/_{kg.}$ This number results in the highest possible recovery, as can be seen in graph 2. In this graph it is shown that the recovery increases with increased air/solids ratio up to a certain maximum. The upper boundary for the variable is chosen close to this maximum. Consequently, the highest possible A/S ratio of 0.500 results in the highest algae recovery. Only in one route the best decision variable is not 0.500 $^{kg}/_{kg}$ but 0.300 $^{kg}/_{kg.}$ This is explained in the next paragraph.

The last decision variable is the concentration factor. This variable results for each route in a different outcome. A few observations can be made for the wet routes:

- Centrifuging requires more energy for harvesting and/or dewatering than filtration, but the recovery percentage is equal. When both units are used in one route, the concentration factor of the filtration is always chosen higher than that for the centrifuge. The flow rates also effect the overall energy consumption during these steps. The combination of concentration factor and flow rate in both unit operations are optimised in the calculations to yield the lowest *y-value.*
- When two filtration units are used for both harvesting and dewatering, the filtration units both equally concentrate the algae broth. Since the flow is used in the energy use equation, the second filtration step has a bit higher concentration factor, because the flow is much lower, resulting in a lower energy use.
- When Flocculation, DAF or Ultrasound Sedimentation is used as harvesting step, either filtration or centrifuging is used to further concentrate the stream during the dewatering step. The concentration factor is not a variable anymore, since the disruption concentration is already defined. The concentration factor of the centrifuge and filtration unit are purely based on this disruption concentration.

For the 'dry' routes the concentration factor is different than the 'wet' routes. This is because there are two dewatering steps instead of one. This results in the following observations:

- With filtration a maximum concentration of 270 $^{kg}/_{m3}$ can be reached. A centrifuge can concentrate up to 400 $^{kg}/_{m3}$. The results show that when filtration is followed by centrifuging, filtration will take place till the maximum concentration to minimize the concentration factor and subsequent energy consumption of the centrifuge.
- Drying requires a lot of energy. In some routes drying takes place as second dewatering steps. In the results it can be seen that the harvesting step and first dewatering step are than used till the absolute maximum end concentration possible, to minimize the drying.

- The only difference between dry route 31 and 32 is the first dewatering step. In route 31 a centrifuge is used (till 400 $^{kg}/_{m3}$) and in route 32 filtration takes place (till 270 $^{kg}/_{m3}$). For both routes the second dewatering step is done by drying (till 800 $^{kg}/_{m3}$). Normally the energy use is very different between filtration and centrifuging, since centrifuging requires a lot more energy. In this case the energy difference is relatively small. This can be explained by the fact that a centrifuge can concentrate more than a filter. Therefore the drying step of route 32 is bigger (from 270 $^{kg}/_{m3}$ till 800 $^{kg}/_{m3}$), resulting in a small energy requirements difference.
- In route 16 something interesting happens. The DAF always uses an air/solid ratio of 0.500, because this results in the highest recovery of algae. However in this route an air/solids ratio of 0.3 is taken as most optimum one. This can be explained as follows: the harvesting has a lower recovery and therefore a somewhat smaller outgoing stream. Because the stream is smaller, filtration can occur to an maximum of 270 $^{kg}/_{m3}$. The first dewatering step must have a concentration factor that is a whole number. It is more favourable to have some losses in algae recovery in the DAF step and thus a lower outgoing stream, to maximize the concentration in the filtration step. This results in a smaller centrifuging step (second dewatering step).

3.1.4 Additional observations

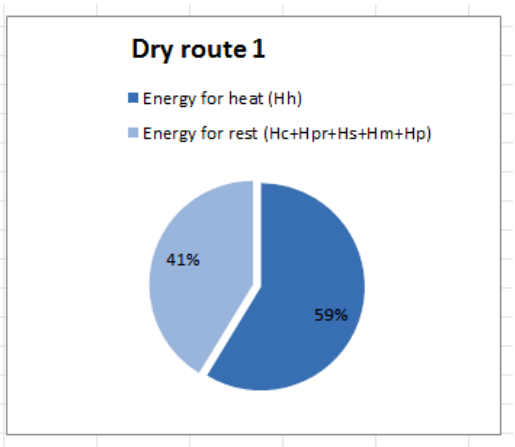
The waste streams of all the routes vary little from one another. The waste stream is defined as a summation of the medium removed from the algae stream and the addition of a costream. Only in a few units there is a costream, as explained in Eq. 2.1. When a route contains only units which do not provide a costream, the waste stream only consist of the removed medium of the mainstream. For example route 1 of the wet routes consist only of downstream processing units which do not contain a costream. In this route only the alkaline solution is added to this stream. The total waste stream is of this route is 5.03 $^{m3}/_{h.}$

The unit operations which provide a costream are flocculation and alkaline extraction. Since alkaline extraction occurs in all routes, it has the same impact on the waste stream for all routes, it will be a bit higher due to the addition of alkaline solution. When flocculation occurs in a route, it is to be expected that the waste stream will be higher. However this is not the case and can be explained as follows: flocculation has a relative low algae recovery and consequently a lower protein recovery. The amount of alkaline solution added to the stream after disruption is based on the amount of algae cells disrupted . Because the amount of algae is lower (and thus the amount of disrupted cells), less alkaline is added, the total waste stream is similar to the other routes.

In some unit operation there is energy needed to heat the system, referred as H_h. The unit operations which need energy for heating are the HPH, dryer, beadmill and calander. The energy needed for heating can be relatively high as can be seen in Figure 3-21 and Figure 3-22. These figures show the percentage of energy needed for heating compared to the

total amount of energy needed, for 'dry' routes 1 and 17. These routes are chosen because route 1 requires the most energy and route 17 has the lowest *y* value.

These figures show that a large amount of energy is needed for heating (and evaporating). Energy needed for heating can be extracted from other sources which produce heat as by-product. Heat as by-product is exergy. Exergy (also called work potential) is the high quality energy that is available to do work and that can be obtained from a system at a given state in a given environment. This energy is still useful and can function as source of energy for the unit operations in this project for heat production. Exergy is used for system optimization. This method is sustainable because it is in fact repurposed 'waste' energy.



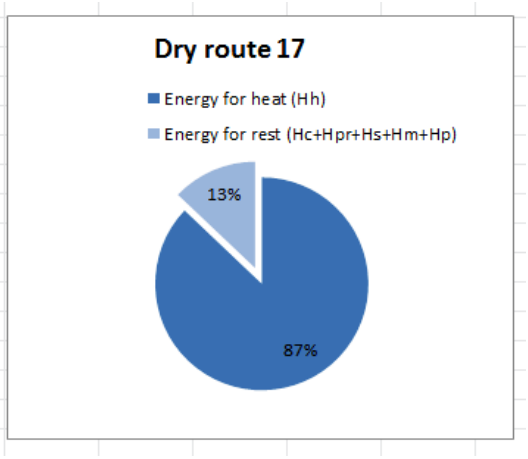


Figure 3-21: Total amount of energy needed for dry route 1 divided in energy needed for heating (H_h) and the sum of energy needed for cooling, pressurizing, mixing, pumping and the mechanical energy.

Figure 3-22: Total amount of energy needed for dry route 17 divided in energy needed for heating (H_h) and the sum of energy needed for cooling, pressurizing, mixing, pumping and the mechanical energy.

Unit operations which produce heat as by-product can be coupled to unit operations which need heat, like for example the dryer. In this project an unit operation which produces heat is the HPH. The HPH produces heat as by-product. In the model of the HPH, cooling takes place internally and the energy requirements for cooling are taken into account, as can be seen in Eq. 2.42. However, if this cooling did not take place and the produced heat in the HPH could be used for the dryer, it would reduce the total amount of energy needed. Less energy is needed for the HPH, because cooling is not needed anymore and less energy is needed for the dryer, because heat is added from the HPH.

3.2 General discussion

The conclusions drawn in this chapter are based on the optimization of the models that are made with overall mass and energy balances. These balances are based on information found in literature. However, the models are sometimes not very detailed due to limited information from experiments or models are more detailed and thus contain assumptions.

In all three disruption models the assumptions is being made that either a cell is disrupted or not. When a cell is disrupted all proteins and lipids are available for extraction. However, in reality cell disruption is far more complex. It depends on the extent of rupture of individual cells as well and on the proportion of cells within the overall population that has been ruptured (Spiden, Yap et al. 2013). When the pressure is increased and the amount of passes increases in the HPH the cell wall of individual cell rupture increases and this results in a higher degree of disruption (Samarasinghe, Fernando et al. 2012). It is estimated that when 25% of the cell wall is disrupted the content of the algae cell becomes available for extraction.(Spiden, Scales et al. 2013). Thus the amount of disrupted cells will probably be lower in reality.

For the flocculation model the energy needed to harvest the algae cells is taken into account. However, the energy needed to produce the flocculants is not. Flocculation has a few shortcomings, because the production of flocculants is expensive and requires a lot of energy (Bosma, van Spronsen et al. 2003). If the amount of energy needed for flocculant production would be taken into account, the energy requirements for flocculation would be much higher.

In the alkaline extraction model it is assumed that only proteins dissolve in the solution and that other components do not dissolve and are removed by centrifuging. In reality many other components will dissolve in the alkaline solution and consequently the end product will not only contain proteins. Consequently, the end product in this project is not pure and more downstream processing steps must be used to obtain an end product which is more pure.

In the optimization of the routes, the lipid extraction with hexane has a negative impact on the protein extraction with alkaline solution. It is not really known how this negative impact occurs and why it happens. The exact impact is not known either. Therefore data is gathered from experts at FBR at Wageningen University and an estimation is made. It would be very interesting if in the future more information would be known about the impact of lipid extraction with an hexane solution on the quality and solubility of the proteins.

The two product streams consists of a highly concentrated not-pure protein solution and a hexane solution with lipids. This hexane solution can be very diluted, since the amount of hexane used depends on the disrupted algae flow. For the ‘wet’ routes this results in a big hexane stream, for ‘dry’ routes the hexane stream is (much) smaller. Whether the hexane stream is big and has a low lipid concentration or the stream is small and the lipid

concentration is high, is not taken into account. In the results, the amount of lipids (flow times concentration) is used. Concentrating the hexane stream to a very high lipid concentration would require a lot of energy. Thus, the 'wet' routes which have a very diluted hexane stream would have an higher energy use, than the 'dry' routes. This would make the difference in *y–values* between 'wet' and 'dry' routes even bigger.

The results in this report show the best *y-value*, which represents the amount of energy needed per kg of extracted product. This *y-value* is used to decide which downstream processing route is best in order to obtain proteins (and lipids) . Unfortunately, it is not possible to compare the results in this report to literature. The *y-value* is a number which is used in this project but is not used in literature. The amount of extracted lipids in this report is calculated in kg of dissolved lipids in hexane per hour. In order to compare the amount of extracted lipids in this project to data given in literature, the lipids need to be converted to FAME. The lipids in algae are triacyl glycerides which can be converted to fatty acid methyl esters (FAME) by transesterification. FAME can be used as biodiesel. FAME are used in literature. To evaluate the results of this project, the conversion of lipids to FAME as to be taken into account. This conversion step requires energy and has a negative impact on the *y-value* (will be higher).

The proteins, which are extracted after the alkaline solution is added, are not pure and are still in solution. These proteins need to be precipitated. During precipitation the solubility of the proteins lowers and the proteins form aggregates. This step is needed to get the proteins out of the alkaline solution. However, this is not taken into account in this project.

Also, in this project no difference is made between proteins and lipids recovery with regards to the *y-value*. When both products are extracted the *y-value* takes only the mass sum of both products (proteins and lipids) into account. There is no additional value added to the proteins, which is the main target in this project. Only the product recovery in $^{kg}/_{h}$ is used. If the protein recovery would have given a higher value than lipid recovery, this would have an effect on the *y-value.* When an additional value is added to the recovered proteins, the *y-values* of all the routes will be lower. The amount of proteins recovered per hour in 'wet' routes 1-60 and 'dry' routes 1-23 is higher than 'wet' routes 61-120 and 'dry' routes 24-34, because hexane extraction has an negative impact on the protein recovery. This result in a larger *y-value* lowering for the routes where only proteins are recovered. The introduction question: *"What provides the lowest y-value, extraction of proteins or extraction of both proteins and lipids?"* could be answered differently. It depends on what value is given to protein recovery compared to lipid recovery.

4 Perspectives

During this project a lot of literature was consulted and many interesting techniques and articles were found. Unfortunately, although exciting, some techniques were too innovative and new to be used in this project. Not enough was found in literature to make a proper model with mass and energy balances. Two techniques that are interesting to keep an eye on in the future, but are not used in this project, are PEF and cell disruption by enzymes. If more information is given in about these two techniques it is very interesting to take this into account in the future when modelling the downstream processing of microalgae biomass.

4.1 PEF

Pulsed Electric Field (PEF) is a technology which causes cells to become permeable and perforated. During the process, the cells are subjected to an electric field with high field strength and this results in cell wall disruption (Goettel, Eing et al. 2013). This method is in theory very interesting for this project since the PEF makes use of intense, but short high frequency pulses, which results in only a slight increase in temperature inside the algae cell. Therefore the intracellular proteins of the algae cell will stay stable. The disruption of algae cells can take place without chemical contamination or degradation of the proteins and PEF can also be scaled up.

PEF seems a promising method to use for disrupting microalgae cells. Unfortunately, available literature on this technique is very limited. This is the main reason why PEF is not used in this project. It is not possible yet to make overall mass and energy balances for PEF. Some numbers that are mentioned in articles give information about the relation between pulse duration, electric field strength or the number of pulses in relation to the amount of intracellular matter (proteins, carbohydrates) released in the medium after treatment. Nothing is found on the actual disruption efficiency and it is not possible to set up a model without this information.

4.2 Cell disruption by enzymes

The cell wall composition of microalgae differs very much from one species to another. The cell wall composition has an influence on the rigidity and strength of the cell wall (Safi, Charton et al. 2013). Enzymes can be used to disrupt the cell wall. It is important when using enzymes to use the appropriate ones, which is chosen based on the chemical composition of the cell wall. The big advantage of enzymatic disruption is its specific and gentle method of disrupting cells. Harsh conditions applied during cell disruption, like the high shear stress of mechanical disruption can be prevented by using enzymes as they gently lyse the cell wall components. Some research has been done about cell disruption of microalgae by enzymes (Zheng, Yin et al. 2011, Ciudad, Rubilar et al. 2014) (Sander and Murthy 2009, Fu, Hung et al. 2010).

There are two main reasons why there is no model made for this disruption method. First, because there is no known data available on the use of enzymes as a disruption method on an industrial scale. Second, the biggest energy consuming part isn't the energy needed during the disruption itself (temperature and stirring), but the energy needed to produce the enzymes and to immobilize them. When it is possible to use this disruption method not only on lab scale but also on industrial scale it can become an interesting microalgae cell wall disruption method.

4.3 Protein extraction

Several techniques can be used for the extraction of proteins from the disrupted algae stream, like the addition of an alkaline solution, dialysis and ion exchange chromatography. Almost all articles which provide information about the extraction of proteins from microalgae biomass, have as main goal to determine the protein composition in species and the quantize the amount of proteins. All information provided is based on lab scale. Only alkaline extraction is used on (a somewhat) larger scale. When more techniques in the future are given in literature about the extraction of proteins from microalgae biomass, it is very interesting to look into this .

5 Conclusion

Extraction of proteins from microalgae is an important solution to prevent human malnutrition in the future. The biggest challenge to obtain proteins from microalgae is the downstream processing of a diluted microalgae stream to a concentrated protein stream. The downstream processing can be done with different techniques. The goal of this project is : "*Which of the process unit combinations at what conditions results in the lowest y-value, for the downstream processing of microalgae biomass?*"

For this project a model based optimization approach is done for the downstream processing of microalgae biomass into proteins with different scenarios. The four different scenario's include 'wet' and 'dry' methods and the extraction of only proteins or the extraction of both proteins and lipids. It can be concluded that it is more favourable to extract both proteins and lipids from the microalgae stream, the *y-value* is lower in these routes. The addition of the hexane extraction step results in higher energy requirement and lower protein recovery, but the total recovery of both products (lipid and proteins) is much higher.

It is more favourable to use a 'dry' method to extract proteins from algae, because then a the algae stream is concentrated to at least 200 $^{kg}/_{m3}$ before disruption takes place. This results in less concentrating by drying in the last step of the downstream processing. It is energetically favourable to minimize the drying step in the last stage, because drying is very energy intensive. The routes where most of the dewatering can be done by centrifuging or filtration is requires less energy. The routes where a DAF is used as harvesting method are favourable, since a DAF system does not require much energy and has a high recovery. The route which has a DAF as harvesting technique, filtration and centrifuging as two dewatering steps resulting in a concentration of 400 $^{kg}/_{m3}$, followed by a calander as disruption step is considered best with regards to energy use and product recovery.

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7 Appendix

7.1 Explanation flows in MATLAB

In MATHLAB the going stream is defined as A. The flow of A in m^3/h is named 'AF' and the concentration of algae in flow A is defined as 'AC'. The outgoing algae stream is named C and the waste stream is D. In Figure 7-1 these streams are illustrated.

Figure 7-1: Definition of flows in Matlab

7.2 'Wet' routes

Number	Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
1	wet	50	Centrifuge	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
2	wet	50	Centrifuge	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
3	wet	50	Centrifuge	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
4	wet	50	Centrifuge	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
5	wet	50	USSed	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
6	wet	50	USSed	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
7	wet	50	USSed	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
8	wet	50	USSed	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
9	wet	50	Flocculation	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
10	wet	50	Flocculation	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
11	wet	50	Flocculation	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
12	wet	50	Flocculation	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
13	wet	50	DAF	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
14	wet	50	DAF	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
15	wet	50	DAF	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
16	wet	50	DAF	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
17	wet	50	Filtration	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
18	wet	50	Filtration	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
19	wet	50	Filtration	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
20	wet	50	Filtration	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
21	wet	100	Centrifuge	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
22	wet	100	Centrifuge	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
23	wet	100	Centrifuge	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
24	wet	100	Centrifuge	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
25	wet	100	USSed	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
26	wet	100	USSed	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
27	wet	100	USSed	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
28	wet	100	USSed	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
29	wet	100	Flocculation	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
30	wet	100	Flocculation	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
31	wet	100	Flocculation	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
32	wet	100	Flocculation	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
33	wet	100	DAF	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
34	wet	100	DAF	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
35	wet	100	DAF	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
36	wet	100	DAF	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
37	wet	100	Filtration	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
38	wet	100	Filtration	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
39	wet	100	Filtration	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
40	wet	100	Filtration	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
41	wet	150	Centrifuge	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
42	wet	150	Centrifuge	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
43	wet	150	Centrifuge	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
44	wet	150	Centrifuge	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
45	wet	150	USSed	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
46	wet	150	USSed	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
47	wet	150	USSed	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
48	wet	150	USSed	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
49	wet	150	Flocculation	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
50	wet	150	Flocculation	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
51	wet	150	Flocculation	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
52	wet	150	Flocculation	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
53	wet	150	DAF	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
54	wet	150	DAF	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
55	wet	150	DAF	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
56	wet	150	DAF	Filtration	-	Beadmill	Alkaline_wet	-	Dryer
57	wet	150	Filtration	Centrifuge	-	HPHomo	Alkaline_wet	-	Dryer
58	wet	150	Filtration	Centrifuge	-	Beadmill	Alkaline_wet	-	Dryer
59	wet	150	Filtration	Filtration	-	HPHomo	Alkaline_wet	-	Dryer
60	wet	150	Filtration	Filtration	-	Beadmill	Alkaline_wet	-	Dryer

Number	Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
61	wet	50	Centrifuge	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
62	wet	50	Centrifuge	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
63	wet	50	Centrifuge	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
64	wet	50	Centrifuge	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
65	wet	50	USSed	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
66	wet	50	USSed	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
67	wet	50	USSed	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
68	wet	50	USSed	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
69	wet	50	Flocculation	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
70	wet	50	Flocculation	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
71	wet	50	Flocculation	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
72	wet	50	Flocculation	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
73	wet	50	DAF	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
74	wet	50	DAF	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
75	wet	50	DAF	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
76	wet	50	DAF	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
77	wet	50	Filtration	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
78	wet	50	Filtration	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
79	wet	50	Filtration	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
80	wet	50	Filtration	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
81	wet	100	Centrifuge	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
82	wet	100	Centrifuge	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
83	wet	100	Centrifuge	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
84	wet	100	Centrifuge	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
85	wet	100	USSed	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
86	wet	100	USSed	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
87	wet	100	USSed	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
88	wet	100	USSed	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
89	wet	100	Flocculation	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
90	wet	100	Flocculation	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
91	wet	100	Flocculation	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
92	wet	100	Flocculation	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
93	wet	100	DAF	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
94	wet	100	DAF	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
95	wet	100	DAF	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
96	wet	100	DAF	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
97	wet	100	Filtration	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
98	wet	100	Filtration	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
99	wet	100	Filtration	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
100	wet	100	Filtration	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
101	wet	150	Centrifuge	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
102	wet	150	Centrifuge	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
103	wet	150	Centrifuge	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
104	wet	150	Centrifuge	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
105	wet	150	USSed	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
106	wet	150	USSed	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
107	wet	150	USSed	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
108	wet	150	USSed	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
109	wet	150	Flocculation	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
110	wet	150	Flocculation	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
111	wet	150	Flocculation	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
112	wet	150	Flocculation	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
113	wet	150	DAF	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
114	wet	150	DAF	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
115	wet	150	DAF	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
116	wet	150	DAF	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
117	wet	150	Filtration	Centrifuge	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
118	wet	150	Filtration	Centrifuge	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer
119	wet	150	Filtration	Filtration	-	HPHomo	Hexane_wet	Alkaline_hex_wet	Dryer
120	wet	150	Filtration	Filtration	-	Beadmill	Hexane_wet	Alkaline_hex_wet	Dryer

7.3 'Dry' routes

Number		Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
121	1	dry	200	Centrifuge	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
122		dry	200	Centrifuge	Centrifuge	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
123		dry	200	Centrifuge	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
124		dry	200	Centrifuge	Centrifuge	Dryer	Beadmill	Alkaline_dry	-	Dryer
125		dry	200	Centrifuge	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
126		dry	200	Centrifuge	Filtration	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
127		dry	200	Centrifuge	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
128		dry	200	Centrifuge	Filtration	Dryer	Beadmill	Alkaline_dry	-	Dryer
129		dry	200	USSed	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
130		dry	200	USSed	Centrifuge	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
131		dry	200	USSed	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
132		dry	200	USSed	Centrifuge	Dryer	Beadmill	Alkaline_dry	-	Dryer
133	2	dry	200	USSed	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
134		dry	200	USSed	Filtration	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
135		dry	200	USSed	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
136		dry	200	USSed	Filtration	Dryer	Beadmill	Alkaline_dry	-	Dryer
137		dry	200	Flocculation	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
138		dry	200	Flocculation	Centrifuge	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
139		dry	200	Flocculation	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
140		dry	200	Flocculation	Centrifuge	Dryer	Beadmill	Alkaline_dry	-	Dryer
141	3	dry	200	Flocculation	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
142	4	dry	200	Flocculation	Filtration	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
143	5	dry	200	Flocculation	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
144		dry	200	Flocculation	Filtration	Dryer	Beadmill	Alkaline_dry	-	Dryer
145		dry	200	DAF	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
146		dry	200	DAF	Centrifuge	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
147		dry	200	DAF	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
148		dry	200	DAF	Centrifuge	Dryer	Beadmill	Alkaline_dry	-	Dryer
149	6	dry	200	DAF	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
150		dry	200	DAF	Filtration	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
151	7	dry	200	DAF	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
152		dry	200	DAF	Filtration	Dryer	Beadmill	Alkaline_dry	-	Dryer
153		dry	200	Filtration	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
154		dry	200	Filtration	Centrifuge	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
155		dry	200	Filtration	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
156		dry	200	Filtration	Centrifuge	Dryer	Beadmill	Alkaline_dry	-	Dryer
157	8	dry	200	Filtration	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
158		dry	200	Filtration	Filtration	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
159		dry	200	Filtration	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
160		dry	200	Filtration	Filtration	Dryer	Beadmill	Alkaline_dry	-	Dryer
161		dry	300	Centrifuge	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
162		dry	300	Centrifuge	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
163		dry	300	Centrifuge	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
164		dry	300	Centrifuge	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
165		dry	300	USSed	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
166		dry	300	USSed	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
167	9	dry	300	USSed	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
168	10	dry	300	USSed	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
169		dry	300	Flocculation	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
170		dry	300	Flocculation	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
171	11	dry	300	Flocculation	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
172		dry	300	Flocculation	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
173		dry	300	DAF	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
174		dry	300	DAF	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
175	12	dry	300	DAF	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
176		dry	300	DAF	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
177		dry	300	Filtration	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
178		dry	300	Filtration	Centrifuge	Dryer	HPHomo	Alkaline_dry	-	Dryer
179	13	dry	300	Filtration	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
180		dry	300	Filtration	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer

Number		Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
181		dry	400	Centrifuge	Centrifuge	Centrifuge	Calander	Alkaline_dry	-	Dryer
182		dry	400	Centrifuge	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
183		dry	400	Centrifuge	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
184		dry	400	Centrifuge	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
185		dry	400	USSed	Centrifuge	Centrifuge	Calander	Alkaline_dry	-	Dryer
186		dry	400	USSed	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
187	14	dry	400	USSed	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
188		dry	400	USSed	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
189		dry	400	Flocculation	Centrifuge	Centrifuge	Calander	Alkaline_dry	-	Dryer
190		dry	400	Flocculation	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
191	15	dry	400	Flocculation	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
192	16	dry	400	Flocculation	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
193		dry	400	DAF	Centrifuge	Centrifuge	Calander	Alkaline_dry	-	Dryer
194		dry	400	DAF	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
195	15	dry	400	DAF	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
196		dry	400	DAF	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
197		dry	400	Filtration	Centrifuge	Centrifuge	Calander	Alkaline_dry	-	Dryer
198		dry	400	Filtration	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
199	18	dry	400	Filtration	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
200		dry	400	Filtration	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
201		dry	800	Centrifuge	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
202		dry	800	Centrifuge	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
203		dry	800	USSed	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
204	19	dry	800	USSed	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
205		dry	800	Flocculation	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
206	20	dry	800	Flocculation	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
207		dry	800	DAF	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
208	21	dry	800	DAF	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
209	22	dry	800	Filtration	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
210	23	dry	800	Filtration	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer

Number		Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
211		dry	200	Centrifuge	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
212		dry	200	Centrifuge	Centrifuge	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
213		dry	200	Centrifuge	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
214		dry	200	Centrifuge	Centrifuge	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
215		dry	200	Centrifuge	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
216		dry	200	Centrifuge	Filtration	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
217		dry	200	Centrifuge	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
218		dry	200	Centrifuge	Filtration	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
219		dry	200	USSed	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
220		dry	200	USSed	Centrifuge	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
221		dry	200	USSed	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
222		dry	200	USSed	Centrifuge	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
223		dry	200	USSed	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
224		dry	200	USSed	Filtration	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
225		dry	200	USSed	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
226		dry	200	USSed	Filtration	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
227		dry	200	Flocculation	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
228		dry	200	Flocculation	Centrifuge	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
229		dry	200	Flocculation	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
230		dry	200	Flocculation	Centrifuge	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
231	24	dry	200	Flocculation	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
232		dry	200	Flocculation	Filtration	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
233		dry	200	Flocculation	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
234		dry	200	Flocculation	Filtration	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
235		dry	200	DAF	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
236		dry	200	DAF	Centrifuge	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
237		dry	200	DAF	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
238		dry	200	DAF	Centrifuge	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
239	25	dry	200	DAF	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
240		dry	200	DAF	Filtration	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
241		dry	200	DAF	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
242		dry	200	DAF	Filtration	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
243		dry	200	Filtration	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
244		dry	200	Filtration	Centrifuge	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
245		dry	200	Filtration	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
246		dry	200	Filtration	Centrifuge	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
247	26	dry	200	Filtration	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
248		dry	200	Filtration	Filtration	Centrifuge	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
249		dry	200	Filtration	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
250		dry	200	Filtration	Filtration	Dryer	Beadmill	Hexane_dry	Alkaline_hex_dry	Dryer
251		dry	300	Centrifuge	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
252		dry	300	Centrifuge	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
253		dry	300	Centrifuge	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
254		dry	300	Centrifuge	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
255		dry	300	USSed	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
256		dry	300	USSed	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
257		dry	300	USSed	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
258		dry	300	USSed	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
259		dry	300	Flocculation	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
260		dry	300	Flocculation	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
261	27	dry	300	Flocculation	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
262		dry	300	Flocculation	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
263		dry	300	DAF	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
264		dry	300	DAF	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
265		dry	300	DAF	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
266		dry	300	DAF	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
267		dry	300	Filtration	Centrifuge	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
268		dry	300	Filtration	Centrifuge	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
269	28	dry	300	Filtration	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
270		dry	300	Filtration	Filtration	Dryer	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer

Number		Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
271		dry	400	Centrifuge	Centrifuge	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
272		dry	400	Centrifuge	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
273		dry	400	Centrifuge	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
274		dry	400	Centrifuge	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
275		dry	400	USSed	Centrifuge	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
276		dry	400	USSed	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
277	29	dry	400	USSed	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
278		dry	400	USSed	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
279		dry	400	Flocculation	Centrifuge	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
280		dry	400	Flocculation	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
281		dry	400	Flocculation	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
282		dry	400	Flocculation	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
283		dry	400	DAF	Centrifuge	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
284		dry	400	DAF	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
285	30	dry	400	DAF	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
286		dry	400	DAF	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
287		dry	400	Filtration	Centrifuge	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
288		dry	400	Filtration	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
289		dry	400	Filtration	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
290		dry	400	Filtration	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
291		dry	800	Centrifuge	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
292		dry	800	Centrifuge	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
293		dry	800	USSed	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
294		dry	800	USSed	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
295	31	dry	800	Flocculation	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
296	32	dry	800	Flocculation	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
297		dry	800	DAF	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
298		dry	800	DAF	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
299	33	dry	800	Filtration	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
300	34	dry	800	Filtration	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer

Number		Wet/dry	Condis	Harvesting	Dewatering	Dewatering	Disruption	Extraction (1)	Extraction (2)	Dryer
121	**1**	dry	200	Centrifuge	Centrifuge	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
133	**2**	dry	200	USSed	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
141	**3**	dry	200	Flocculation	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
142	**4**	dry	200	Flocculation	Filtration	Centrifuge	Beadmill	Alkaline_dry	-	Dryer
143	**5**	dry	200	Flocculation	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
149	**6**	dry	200	DAF	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
151	**7**	dry	200	DAF	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
157	**8**	dry	200	Filtration	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
167	**9**	dry	300	USSed	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
168	**10**	dry	300	USSed	Filtration	Dryer	HPHomo	Alkaline_dry	-	Dryer
171	**11**	dry	300	Flocculation	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
175	**12**	dry	300	DAF	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
179	**13**	dry	300	Filtration	Filtration	Centrifuge	HPHomo	Alkaline_dry	-	Dryer
187	**14**	dry	400	USSed	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
191	**15**	dry	400	Flocculation	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
192	**16**	dry	400	Flocculation	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
195	**17**	dry	400	DAF	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
199	**18**	dry	400	Filtration	Filtration	Centrifuge	Calander	Alkaline_dry	-	Dryer
204	**19**	dry	800	USSed	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
206	**20**	dry	800	Flocculation	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
208	**21**	dry	800	DAF	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
209	**22**	dry	800	Filtration	Centrifuge	Dryer	Calander	Alkaline_dry	-	Dryer
210	**23**	dry	800	Filtration	Filtration	Dryer	Calander	Alkaline_dry	-	Dryer
231	**24**	dry	200	Flocculation	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
239	**25**	dry	200	DAF	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
247	**26**	dry	200	Filtration	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
261	**27**	dry	300	Flocculation	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
269	**28**	dry	300	Filtration	Filtration	Centrifuge	HPHomo	Hexane_dry	Alkaline_hex_dry	Dryer
277	**29**	dry	400	USSed	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
285	**30**	dry	400	DAF	Filtration	Centrifuge	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
295	**31**	dry	800	Flocculation	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
296	**32**	dry	800	Flocculation	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
299	**33**	dry	800	Filtration	Centrifuge	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer
300	**34**	dry	800	Filtration	Filtration	Dryer	Calander	Hexane_dry	Alkaline_hex_dry	Dryer

7.4 Example of MATLAB File

In the next two pages an MATLAB file is given. In this file the Beadmill is described.

```
function [ C , D , H ] = Beadmill( A, Bf, par)
%   Beadmill describes the disruption of algae using a bead mill
%   A is the algae flow into the bead mill
%   C is the flow of disrupted algae out of the bead mill
%   There will be no other flow going in or out, so flow A and C have the same size
%   Both flows are of the form [F , F_mass , C1 , C2, C3,  T, p].

global Fprot Flipid

r = par(1);
rhoA = par(2);
cpA = par(3);
rhoM = par(4);
MW_M = par(5);
cpM = par(6);
rhoO = par(7);
MW_O = par(8);
cpO = par(9);
rhoF = par(10);
MW_F = par(11);
rhoW =par(12);
cpW = par(13);
rhoC = par(14);
cpC = par(15);
rhoH = par(16);
cpH = par(17);
R_FO = par(18);
H_M = par(19);
H_H = par(20);

% Disassembly
AF = A(1);                              % Volumetric flow in of desirable stream (algae) (m3/h)
AF_mass = A(2);                         % Mass flow in of desirable stream (algae) (kg/h)
AC1 = A(3);                             % Concentration algae (kg/m3)
AC2 = A(4);                             % Concentration accessable proteins (kg/m3); not applicable
AC3 = A(5);                             % Concentration accessable lipids (kg/m3); not applicable
AT = A(6);                               % Temperature of ingoing stream (K)
Ap = A(7);                              % Pressure of ingoing stream (bar)

%Process parameters
Q_max = 40;                             % Max flow (L/h), [Doucha et al, 2008]
Q_max_ls = 450;                         % Max flow (L/h) in large scale, Doucha 2008
Q = AF * 1000;                          % Mass flow (L/h)

N_units = 1;
N_units(Q>Q_max_ls)=ceil(Q/Q_max_ls);   % Number of bead mills needed
Q_acc = Q;
Q_acc(N_units>1)=Q/N_units;             % Flow in each bead mill (L/h)
Q_acc = Q_acc * Q_max / Q_max_ls;       % Conversion for the disruption formula, which is based on  Q_max (L/h)

BD = 0.5;                               % Bead diameter mm
BF = Bf;                                % Bead filling (% of chamber volume)
PV = 14;                                % Speed of agitation discs (m/s)
DW = AC1;                               % Algae dry weight concentration (kg/m3)
DW(AC1<100)=100;

```

```
a = 1748;
n1 = -0.0356;
n2 = 0.326;
n3 = 0.0768;
n4 = 0.248;
n5 = -0.763;
Disruption = (a * Q_acc^n1 * BD^n2 * BF^n3 * PV^n4 * DW^n5)/ 100; % DynoMill KDL-Pilot A fraction between 0 and 1

% Mass balances
CF = AF;                                % Volumetric flow rate (m3/h)
CF_mass = AF_mass;                      % Mass flowrate out (kg/m3)
CC1 = 0;                                % Biomass algae concentration; living cells (kg/m3)
CC2 = AC1 * Disruption * Fprot;         % Concentration of extractable proteins (kg/m3)
CC3 = AC1 * Disruption * Flipid;        % Concentration of extractable lipids (kg/m3)

DF = 0;                                 % Volumetric flow rate waste (m3/h); not applicable
DF_mass = 0;                            % Mass flow out (kg/h); not applicable
DT =0;                                  % Not applicable
Dp=0;                                   % Not applicable


% Energy balances
CT=AT;                                  % Temperature (K)  But is cooled back to 25 degrees
Cp=Ap;                                  % Pressure (bar)
T_before= 4+273;                        % Temperature of cooled flow before bead milling (K)
T_after = 35+273;                       % Temperature of flow after bead milling (K)

Hc1 = ((AF*1000)*4190*(AT-T_before))/3600;% Power requirement for cooling inflow (J/s)[Doucha et al, 2008]
Hc2 = ((CF*1000)*4190*(T_after-AT))/3600; % Power requirement for cooling outflow (J/s)[Doucha et al, 2008]
Hc= Hc1+Hc2;                             % Total power requiremnet for heating and cooling (J/s)
Hpr = 0;                                 % Power requirement for pressure (J/s)
Hs = 3300 * N_units;                     % Power requirement for mechanical energy; disruption (J/s)
Hm = 0;                                  % Power requirement for mixing (J/s)
L = 25;                                  % Pumping distance (m)
Hp = Pump( CF , CC1 , L ,par);           % Power requirement for pumping product (J/s)
Ht = Hc+Hpr+Hs+Hm+Hp;                     % Total power requirement (J/s)

% Assembly
C = [CF CF_mass CC1 CC2 CC3 CT Cp];
D = [DF  DF_mass  DT  Dp];
H = [Ht Hc];
end

```

7.6 Nomenclature

Symbol	Definition	SI Unit	Specific model/literature
A	Cross-sectional area of the pipe	m^2	*Pump* Wileman et al, 2012
BD	Bead diameter	mm	*Beadmill* Doucha et al, 2008
BF	Bead filling	% chamber volume	*Beadmill* Doucha et al, 2008
C	Concentration	$^{kg}/_{m^3}$	Several models
Cf	Concentration factor	-	Several models
CI	Consistency index for power law fluids	Pa·s	*Pump* Wileman et al, 2012
Cp	Heat capacity	$^{J}/_{kg/K}$	Several models
d	Diameter of the pipe	m	*Pump* Wileman et al, 2012
D	Disruption efficiency	$^{kg}/_{kg}$	Several models
DW	Dry weight	$^{kg}/_{m^3}$	Several models
ΔH	Heat of evaporation	$^{J}/_{kg}$	Dryer
E	Energy requirements for the process	$^{J}/_{m^3}$	Several models
F	Volumetric flow rate	m^3	Several models
f_{prot}	Fraction proteins in algae	$^{kg}/_{kg}$	Several models
f_{lipid}	Fraction lipids in algae	$^{kg}/_{kg}$	Several models
f	Fanning friction factor	-	*Pump* Wileman et al, 2012
H	Power requirements	$^{J}/_{s}$	Several models
K	Decay constant	-	*HPHomo* Spiden et al, 2013
K_{factor}	Consistency factor	$^{Poise}/_{m}$	Pump Wileman *et al*
K_{power}	Power constant stirring	-	Several models
L	Length pipe	m	Several models
MW	Molecular weight	$^{kg}/_{mol}$	Several models
N_{passes}	Amount of passes through homogenizer	-	*HPHomo* Spiden et al, 2013
N_{units}	Amount of units needed	-	*Beadmill* Doucha et al, 2008
n	Behaviour index for power law fluids	-	*Pump* Wileman et al, 2012
p	Pressure	Pa	Several models
r	Rotational radius centrifuge	cm	*Centrifuge*
R	Microalgae recovery	$^{kg}/_{kg}$	Several models
R	Gas constant	8.314 $^{J}/_{mol}$	Several models

Re	Reynolds number	-	*Pump* Wileman et al, 2012
PV	Speed of agitation discs	$^{m}/_{s}$	*Beadmill* Doucha et al, 2008
Q	Feed rate	$^{kg}/_{h}$	*Beadmill* Doucha et al, 2008
RCF	Relative centrifugal force	g	*Centrifuge*
S_a	Air solubility in water		DAF
S_{RPM}	Rotational speed	RPM	*Centrifuge*
S	Stirring Speed	RPM	*Flocculation* Riano et al, 2011
T	Temperature	K	Several models
u	Liquid speed	$^{m}/_{s}$	*Pump* Wileman et al, 2012
u_m	Stirrer speed during mixing	$^{1}/_{s}$	Several models
v	Velocity	$^{m}/_{s}$	Several models
W	Work	$^{J}/_{mol}$	*DAF* Coward et al, 2013
Y	Yield	$^{kg}/_{kg}$	Several models
X_{floc}	Flocculant concentration	$^{g}/_{m^3}$	*Flocculation* Riano et al, 2011
Greek Symbols			
ρ	Density	$^{kg}/_{m^3}$	Several models
τ	Residence time	s	Several models
ϒ	Ratio specific heat of air	1.4	*DAF* Coward et al, 2013
Subscripts			
A	Algae		
algae	Remaining inside algae cell		
Co	Costream		
in	Inflow, A		
l	Lipid		
Main	Mainstream		
out	Outflow, C		
p	Protein		
release	Released proteins/lipids in medium		
rcfl	Recycleflow		*DAF* Coward et al, 2013
X	Component X, undefined		
waste	Waste flow, D		

7.7 Thank you

I would like to write this text in Dutch.

Heel graag wil ik mijn thesis begeleider Ellen bedanken voor haar begeleiding tijdens het schrijven van mijn masterthesis. In dit afgelopen jaar heb ik met ups en downs aan mijn thesis gewerkt. Ups wanneer ik interessante literatuur vond en aan MATLAB mocht klungelen, maar ook downs, toen ik een tijdje niet aan mijn thesis kon werken en later toen ik vast zat met mijn literatuuronderzoek. Ik ben heel blij dat ik mijn thesis heb voortgezet en dat Ellen mij daarbij enorm heeft gemotiveerd en een steuntje in de rug heeft gegeven. Ik ben trots dat ik mijn thesis nu kan inleveren, ondanks alles wat er dit jaar is bij gekomen. Het was een heel leerzaam jaar, zowel op professioneel als op sociaal en emotioneel niveau. Ik heb geleerd om een onderzoeksvraag op te stellen, (alleen) op zoek te gaan naar literatuur en een verslag te schrijven. Dit is met vallen en opstaan gegaan. Aan de andere kant heb ik ook geleerd om eerder mijn grenzen aan te geven en te zeggen wat ik wel en niet kan en wat ik wel en niet leuk vind om te doen. Aangezien ik geen bachelor thesis heb geschreven was dit de eerste keer dat ik aan mijn eigen project heb gewerkt en er een verslag over te schrijven. Dat was een enorme uitdaging, maar het is me gelukt en dat kon niet zonder de hulp van Ellen.

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