WASTE STREAMS FOR ALGAE CULTIVATION

LAHTI UNIVERSITY OF APPLIED SCIENCES Degree Programme in Environmental Technology Environmental Engineering Bachelor's Thesis 2011 Antti Kautto Lahden Ammattikorkeakoulu Ympäristöteknologian koulutusohjelma

Kautto, Antti:	Levätuotantoon soveltuvien jätevirtojen kartoitus
Ympäristötekniikan opinnäytetyö, Kevät 2011	32 sivua
TIIVISTELMÄ	

ALDIGA (Algae from Waste for Combined Biodiesel and Biogas Production) on Tekesin rahoittama projekti. Tavoitteena on mahdollisimman suljettu kierto biodieselin ja –kaasun tuottamiseksi. Projekti toteutetaan yhteistyössä mm. VTT:n, Helsingin Yliopiston, Lahden ja Hämeen Ammattikorkeakoulujen, SYKE:n sekä useiden yritysten välillä.

Tässä opinnäytetyössä on kartoitettu levätuotantoon soveltuvia jätevirtoja, joihin kuuluvat kunnallinen jätevesi, elintarvike- ja paperiteollisuuden prosessivedet, karjankasvatuksessa syntyvä jätevesi, biokaasulaitoksen rejektivesi, lajitellusta biojätteestä puristettu neste, kaatopaikkojen suotovesi, kalankasvatusvesi, biokaasulaitoksen jäähdytysvesi sekä mattotuotannon jätevesi. Projektissa käytettävien virtojen valinnan helpottamiseksi rakennettiin työkalu, jonka avulla on mahdollista vertailla virtoja keskenään. Virtojen yhteydessä olevista linkeistä on mahdollista jäljittää lähdetieto, josta kunkin virran parametrit on kerätty. Konsentroituneimpia virtoja on laimennettava kasvatusta varten. Niistä rejektivedessä oli korkein typpi- ja fosforipitoisuus (5150 mg/l N ja 555 mg/l P). Biojäteprässivesi sisälsi 3069 mg/l typpeä ja 39 mg/l fosforia. Tislaamovesi sisälsi 2020 mg/l typpeä ja 240 mg/l fosforia. Sianlantapitoinen vesi sisälsi 569 mg/l typpeä ja 441 mg/l fosforia. Tislaamovesissä oli korkein hapenkulutus (ennen mädätystä 36000 mg/l BOD₇ ja 113000 mg/l COD_{Cr} sekä mädätyksen jälkeen 4000 mg/l BOD₇ ja 39000 mg/l COD_{Cr}). Kiintoainepitoisuus oli korkea biojäteprässivedessä (93000 mg/l) ja tislaamovesissä (98000 mg/l ennen mädätystä ja 43000 mg/l mädätyksen jälkeen). Laimeita ja sellaisenaan mahdollisesti käyttökelpoisia virtoja olivat jäähdytysvesi, kaatopaikkavesi, mattoja paperintuotantojätevesi, pasta- ja jauhontuotantovesi, kalankasvatusvesi ja kunnallinen jätevesi jos niitä on saatavilla kasvatuslaitoksen lähellä. Niistä osaa voidaan myös soveltaa väkevien virtojen laimentamiseen.

Avainsanat: Microalgae, combined, biodiesel, biogas, sewage, leachate, municipal wastewater, organic waste press water, effluent

Lahti University of Applied Sciences Degree Programme in Environmental Engineering

Kautto, Antti:

Waste streams for algae cultivation

Bachelor's Thesis in Environmental Technology Spring 2011 32 pages

ABSTRACT

ALDIGA, short for "Algae from Waste for Combined Biodiesel and Biogas Production", aims to develop a concept for a closed circulation of resources in producing biodiesel and biogas from waste. The project is realized in co-operation between VTT, University of Helsinki, Lahti and Häme Universities of Applied Sciences, SYKE and funded by Tekes.

The project's first work phase ergo this bachelor's thesis covered the mapping of available and suitable streams to be used in the cultivation of algae. These streams included municipal and agricultural sewages, food industry's process waters, land-fill leachates, cooling waters, reject waters, fish farming water and press water from municipal organic waste. In order to choose one or several streams to be used in the project, a spreadsheet tool was built to assist in the comparison of the properties of different streams. The tool features a selection of the streams' parameters and the capability to track the original source of information along the provided links. Several sources were used to gather the information on the waste streams and they ranged from analyses produced for the companies themselves to scientific articles and taking and analyzing samples personally.

From the highly concentrated streams, reject water contained the most nutrients (5150 mg/l of nitrogen and 555 mg/l of phosphorus). Organic press water contained 3069 mg/l of nitrogen and 39 mg/l of phosphorus. Distillery effluent contained 2020 mg/l of nitrogen and both lacked phosphorus compared to reject water (<250mg/l of phosphorus). Swine manure sewage contained 569 mg/l of nitrogen and 441 mg/l of phosphorus. Distillery effluents were by far the richest in oxygen demand (36000 mg/l BOD₇ and 113000 mg/l COD_{Cr} before methanation and 4000 mg/l BOD₇ and 39000 mg/l COD_{Cr} after methanation). The total solid concentration in organic press water was 93000 mg/l. Distillery effluents contained total solids 98000 mg/l before methanation and 43000 mg/l after methanation. Some of the dilute streams, municipal waste water, leachate, cooling water, pasta and flour plant effluent, paper production effluent, carpet production effluent and fish farming water are suitable for diluting the high potential streams, or they can probably be used solely if a sufficient source is available nearby.

Key words: Microalgae, combined, biodiesel, biogas, sewage, leachate, municipal wastewater, organic waste press water, effluent

TABLE OF CONTENTS

1	INTRC	DUCTION	1
2	MICRO	DALGAE IN PRODUCTION OF BIODIESEL	3
	2.1	Different energy strategies of algae	3
	2.2	Energy production from algae	4
	2.3	Measures to increase feasibility	5
	2.4	Lipids into biodiesel	6
	2.5	Reactor types	6
	2.6	The selected algae species	9
3	GATH	ERING OF INFORMATION	11
	3.1	Keywords and search criteria	11
	3.2	Search results and selection	12
	3.3	Streams and their source of information	13
	3.4	Analysis of the fish-farming water of Imatra	15
4	COMP	ILING THE SPREADSHEET	17
5	DISCU	USSION AND CONCLUSIONS	21
RE	FEREN	CES	24

1 INTRODUCTION

The diminishing supply of fossil fuels and the much discussed climate change affected by the greenhouse gas emissions have resulted in aiming to increase the production, distribution and eventually consumption of renewable energy. The objective of the new directives and legislations are to simply provide renewable fuels for traffic and other use in the future and to reduce the amount of greenhouse gas emissions (GHG).

The European Union produces and consumes less than 20% of its energy from renewable sources. Approximately 17.6% of total production and 10.3% of total consumption was renewable energy in 2008 (Europe's Energy Portal 2011, Eurostat 2011). In 2010, the European biodiesel production capacity was 22 million tonnes with over 245 facilities while the total production remained at 9 million tonnes. The share of biodiesel was 75 % of renewable fuels in 2009, but it seems wood based fuels were omitted and only biogas and bioethanol remained in the study. Currently, the raw materials used to produce biodiesel require significant areas of arable land, around 3 million hectares in the EU. (EBB 2010)

The Directive 2003/30/EC of the European Parliament and of the Council of 8 May 2003 on the promotion of the use of biofuels or other renewable fuels for transport, along with other legislations, drives the ALDIGA project. More specifically, the directive of 2003/30/EC and The Commission Green Paper "Towards a European strategy for the security of energy supply" set the objective of 20 % substitution of conventional fuels by alternative fuels in the road transport sector by the year 2020. In addition, European Union's climate strategy aims to reduce GHG compared to the 1990 level, to increase energy efficiency and to increase the total use of renewable fuels, each by 20% by the year 2020 (Eur-lex 2010.) The amendment to Finnish legislation of promoting the traffic use of biofuels *HE 197/2010* was verified by the President of Finland, and it came into effect on 1.1.2011. Finland is committed to set the percentage of biofuels in transportation to 10% but ultimately aims at 20% and to reach this goal a legal obligation has been set to fuel distributers to gradually increase the distribution of renewable fuel. At the moment, biofuel is distributed mostly as ethanol in a 10% blend at this point. Also, according to legislation, if the biofuel is produced from waste material, its oil equivalent tonnage is doubled when calculating the production quota.

In principle, algae are able to feed on waste material and increase their biomass. Some species contain a relatively high percentage of lipids in their biomass, which can be utilized in biofuel production. The main objective of the ALDIGA project, short for "Algae from Waste for Combined Biodiesel and Biogas Production", which is also the context of this thesis, is to establish a model for producing biodiesel and biogas from algae grown with waste resources. The process should run as energy independently as possible and utilizing all side streams in addition to main fuels streams (ALDIGA project, 2010. Project plan. Tekes) ALDIGA is funded by Tekes and realized by VTT (Valtion teknillinen tutkimuslaitos) in cooperation with SYKE (Suomen ympäristökeskus), Lahti and Häme Universities of Applied Sciences, Helsinki University, and several bioenergy and waste treatment companies.

The aim of this study is to map suitable wastewaters for cultivating algae and producing biodiesel and gas. This is one of the objectives of work package one. Test culturing and the future large-scale cultivation rely on the data gathered in the first work package (WP1). In order to compare and make a selection of a few waste streams the data was compiled into a spreadsheet tool with visualization and source tracking features.

2 MICROALGAE IN PRODUCTION OF BIODIESEL

2.1 Different energy strategies of algae

Algae have several strategies to obtain energy and carbon and also the production and storing of lipids inside their cells. Phototrophs rely on the energy of sunlight to convert inorganic carbon, the carbon dioxide freely available in the air, for growth whereas heterotrophs use organic carbon for both energy and growth. Mixotrophs are able to use both of these strategies depending on the conditions. A variety of organic carbon sources, such as glucose, acetate, glycerol, fructose, sucrose, lactose, galactose, and mannose have been studied to be suitable as a carbon source for microalgae. Studies also show that mixotrophic species are generally more efficient in growth and lipid storing under heterotrophic conditions as opposed to a lit environment. Algae can use either ammonia (NH4⁺) or nitrate (NO3) for nitrogen and inorganic orthophosphate (PO4⁻³) for phosphorus (Chen et al. 2010).

Algae capable of photosynthesis including photoautotrophic and mixotrophic species can be used in fixation of CO2 in various environments. One application was presented by Sialve and co-workers, in which the emissions of production plants and also biogas produced by anaerobic digestion could be purified by algae resulting in less CO2 released to the atmosphere and higher concentration of methane within the biogas (Sialve et al. 2009). Carbon dioxide is fixed only during the photosynthetic reactions in the presence of light and ideally the algae would use some of the emissions of fossil fuel burned in power plants. However, even if the cultivation of algae fixes carbon dioxide, the processing and transportation should still be taken into account when determining the carbon neutrality. Also, being carbon neutral, algal biofuel production and utilization do not contribute to net reduction of carbon dioxide accumulation that occurs due to the still on-going burning of fossil fuels. (Sawayama et al. 1995; Chisti 2007)

2.2 Energy production from algae

Biodiesel production from algae does not require arable land, and, more importantly, material usable primarily as food as opposed to e.g. palm oil and rapeseeds. This makes the method more ethical. Table 1 presents the oil yields of different crops per hectare and the land area required to satisfy 50% of the total need of transport fuel in the United States, ultimately proving the algal oil to be the only rational alternative to replace petroleum fuels in traffic use. (Chisti 2007.)

Vast numbers of algal strains are kept in banks available for purchase. Most strains were originally discovered and isolated for other purposes than bioenergy. (University of Texas 2010) For example *C. vulgaris*, studied extensively in the production of biodiesel and biogas, is also an ingredient in nutritional supplement products or cosmetics (Cosmetic Analysis 2010, Chlorenergy 2010). The effectiveness of cosmetic and nutritional products is not widely admitted. The strains that have been isolated are usually the ones dominating the samples, which does not necessarily diminish the value of the species in the production of bioenergy since mass cultivation calls for species that will remain dominant despite non-laboratory conditions open to invading organisms (ALDIGA research team 2010).

	Oil		
Crop	yield	Land area	Percent of existing
	(l/ha)	needed (M ha) a	US cropping area <i>a</i>
Corn	172	1540	846
Soybean	446	594	326
Canola	1190	223	122
Jatropha	1892	140	77
Coconut	2689	99	54
Oil palm	5950	45	24
Microalgae b	136900	2	1,1
Microalgae c	58700	4,5	2,5

Table 1: Comparison of some sources of biodiesel (Chisti 2007)

a For meeting 50% of all transport fuel needs of the United States

b 70% oil (by wt) in biomass

c 30% oil (by wt) in biomass

2.3 Measures to increase feasibility

By combining the digestion of the residual biomass of the algae it is possible to increase the feasibility of the algal bio diesel production even further. Furthermore, feeding the cultures CO2 from an adjacent source will contribute to the reducing of flue CO2 released into the atmosphere, and refining the algal residue efficiently into different products eliminates waste. (Sialve et al. 2009; Chisti, Y. 2007)

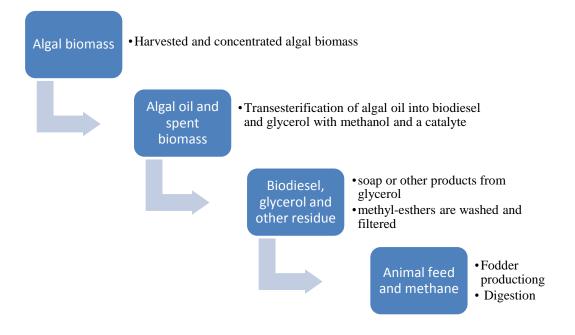


Figure 1: Microalgal biodiesel refinery: producing multiple products (information adapted from Oilgae 2010)

Using the organic carbon, nitrogen and phosphorus present in the waste waters, it is possible to not only maximize the biomass and lipid production of mixo- or heterotrophic microalgae but also to treat the otherwise problematic source of eutrophication and thus oxygen depletion in the natural bodies of water. For example, *Chlorella vulgaris*, removed over 90% of nitrogen and 80% of the phosphorus content from primary treated sewage (Orpez et al. 2008; Pittman et al. 2010). The feasibility for producing biodiesel and biogas from algae is achieved by efficiently using all side streams of the process e.g. as shown in Figure 1.

2.4 Lipids into biodiesel

The harvesting of algae from water is done by filtration, and the mass is concentrated from a dilute suspension into a paste. To mechanically extract the oil, the biomass must be dried because when suspended in water, the algal cells are elastic and very resistant to breaking. Chemical extraction does not require drying but large amounts of e.g. hexane in mass production. The drying of algae can be done either by consuming external energy or by letting it dry, which takes more time. After the biomass is mechanically pressed or chemically processed to break the cells, the raw oil is converted via transesterification into biodiesel with methanol and by alkali-catalyzing the mix. The by-product, glycerol, is pure enough to be utilized as material or burned to produce process steam. (Oilgae 2008, Oilgae 2011.)

2.5 Reactor types

Currently, there are two major methods for cultivating algae, closed and open reactors that are mixed mechanically with a pump or paddlewheel at the starting point. Large-scale cultivation insists on continuous operation, and as economical and simple a process as possible. The challenges of producing biofuel from algae are the energy and time consumption of harvesting the biomass from the reactor, and in some cases drying of the biomass and extracting of lipids from inside the cells. A race-way pond for growing algae as seen in Figure 2 is fundamentally a long looped river mixed with a paddle-wheel between the feeding and harvest points. It is by far the simplest and the most inexpensive method in terms of equipment, installing and maintenance but the productivity is much less compared to closed reactors. Temperature changes and gas exchange between air and water are not controlled outside the start of the loop. The race-way trough is typically lined with plastic to facilitate the flow and make maintenance and cleaning easier. There are baffles in the turns of some race-ways to control the flow of water better in the turns. (Chisti, Y. 2007)

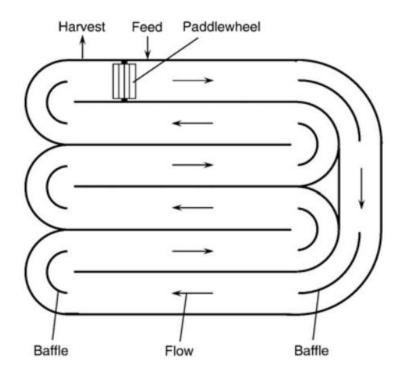


Figure 2: Aerial view of a raceway pond (Chisti, Y. 2007)



Figure 3: Paddle-wheel of a race-way pond (Oilgae 2011)

Photobioreactors consist of transparent tubes usually placed in rows, a degassing column, and a pump as seen in Figure 4. Since the reactor is closed with the exception of the degassing column, the oxygen generated by photosynthesis is not able to dissipate, and equally the CO2 cannot be replenished while the water is traveling inside the tubes. Therefore, the maximum length of continuous tubing for the algae to travel inside before it has to return to the degassing column has been determined to be 80 meters, and due to turbidity and its effect on photosynthesis the common diameter of the tube is 0.1 meters. There are also helical tubular photobioreactors in which the tubing is curved to form a large continuous spring. This kind of reactor is usually used only to grow small amounts of algae because otherwise the optimal diameter and length of a continuous run are easily exceeded. (Molina et al., 2001) The limits set by optical density and CO2 concentration for the cultivation of phototrophic algae could be changed to some extent by cultivating mixotrophic algae.

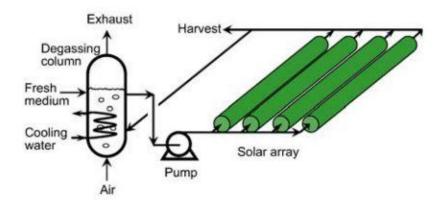


Figure 4: A tubular photobioreactor with parallel run horizontal tubes.

2.6 The selected algae species

Based on the existing information, the ALDIGA research team selected three candidates to be cultivated and tested in the wastewaters; *Euglena gracilis, Chlorella pyrenoidosa, Chlorella protothecoides*. These species have also been widely studied internationally thus making information on similar cases abundant. In fact, some articles provided information on waste streams exclusively from the point of view of algal bioenergy.



Figure 5: Individual E. gracilis cell (Algae resource database 2011)



Figure 6: Multiple C. protothecoides cells (Aquaportail 2011)



Figure 7: Multiple *C. vulgaris* cells as a representation of the appearance of *C. pyrenoidosa* (Aquaportail 2011)

The *C. pyrenoidosa* is extremely similar to *C. vulgaris* in appearance, and there is discussion on whether *C. pyrenoidosa* is factually a separate species or not. (Ullman, J. 2006). This is to some extent confirmed by the fact that information on *C. pyrenoidosa* is especially scarce.

3 GATHERING OF INFORMATION

The focus in this work was to gather mainly already existing information on the waste streams as possible. Only one stream was analyzed personally since sufficient information was not available, and one of the companies participating in the project particularly suggested the stream for the project. The companies involved in the project provided analyses on their waste streams if available. Most streams remained undocumented due to e.g. the early stage of the company's processes and production or because the streams were not analyzed by the company itself for other reasons. The missing information was gathered from scientific articles available to institutions or from environmental impact assessments publicly open to all.

3.1 Keywords and search criteria

From 100 to 150 searches were made by combining the keywords presented in Table 2. The main database used was Science Direct, but a few additional searches were made in the website of Finland's environmental administration and Google to find e.g. environmental impact assessment reports of the paper industry.

	Waste			
Main keyword	water	Effluent	Sewage	Microalgae
Food industry	Х	Х	Х	Х
Paper industry	Х	Х	Х	Х
Industrial	Х	Х	Х	Х
Municipal	Х	Х	Х	Х
Waste water				Х
Washing water				Х
Reject water				Х
Cow manure	Х	Х	Х	Х
Swine manure	Х	Х	Х	Х
Manure	Х	Х	Х	Х
Biowaste	Х	Х	Х	Х

Table 2: Keywords and combinations used in searches

Leachate	Х	х	Х	Х
Fish farming	Х	Х	Х	Х
Aquaculture	Х	Х	Х	Х
Organic waste				Х
Cooling water				Х
Kinetic properties	Х	Х	Х	
Physico-chemical	Х	Х	Х	
Microalgae	Х	Х	Х	
Algal	Х	Х	Х	
Bioenergy	Х	Х	Х	Х
Biodiesel	Х	Х	Х	Х
Digestion	Х	Х	Х	Х
Cultivation	Х	Х	Х	Х
Bioreactor	Х	Х	Х	Х
Algaculture	Х	Х	Х	Х
Nutrients	Х	Х	Х	Х
COD	Х	Х	Х	Х
BOD	Х	Х	Х	Х
Phosphorus	Х	Х	Х	Х
Nitrogen	Х	Х	Х	Х
Ammonium	Х	Х	Х	Х
Total solids	Х	Х	Х	Х
Nitrate	Х	Х	Х	Х

3.2 Search results and selection

Fifty articles were obtained based on their title and abstract, nine of which were processed into the spreadsheet and fifteen were cited in this thesis. Nine other sources were used to complete the spreadsheet parameters including companies' analyses, environmental impact assessment and personal analysis of a single stream. The spreadsheet contains a total of 20 references. The primary targets were the existing studies of producing biodiesel and gas from the same waste streams, and with the same algae used in the ALDIGA project. Each article was first downloaded based on its title and abstract to see if it contained mentions of the waste stream in question, and later the abstract and tables were searched for signs of the desired parameters. Articles with clear tables instead of results written into the paragraphs were favored to save time.

3.3 Streams and their source of information

Manure and manure based waste water was suggested by two companies. They are so high in nutrients and oxygen demand that it is a problem for waste water treatment but also a very potential stream for the ALDIGA project with proper dilution. Cattle manure after digestion at a biogas plant is rich in nutrients and poor in organic carbon. This stream is therefore very useful for the photosynthetic cultivation conditions or a certain phase in mixotrophic cultivation. Studies also show that algae grow well in manure derived waste waters, and particularly *Botryococcus braunii*, grown in piggery wastewater containing 788 mg/l of nitrate, removed 80% of the initial NO3 content. Scientific articles were used in the spreadsheet for parts concerning manure and to some extent municipal sewage. (Travieso, L. 1994, 2006. An, J.Y. 2003)

Municipal sewage was suggested by two companies. It is much lower in biological content than agricultural sewage, and therefore, they are usually mixed in proportions such as 1:60 of manure to sewage. The proportions are in accordance to the production rates of each component. The information was gathered from scientific articles since the ideal source, the waste water treatment plant of Lahti, was not able to release their data. Additional data on municipal sewage was received from another project, and the results correlated well with the ones already included in the finished spreadsheet tool and so this source is not mentioned in the reference list. (Travieso, L. 1994, 2006. Pittman, J.K. 2010)

Reject water from biogas plants was suggested by three companies. It is extremely high in nutrients, and is a natural extension to the circle of this type of renewable biofuel. The reject water is emitted after the digestion process and being stripped of energy and left abundant with nitrogen and phosphorus. Bioenergy companies Biota Tech Oy, Biovakka Oy and Envor Biotech Oy were contacted, and internal analyses of their reject waters were received from two of them. (Biovakka 2010, Envor Biotech Oy 2010) **Paper industry's waste water** was a personal suggestion to see if locating algae production next to paper industry would also provide a substrate. The paper plant would already generate flue heat as it does in the case of the fish farm in Imatra. However, the nutrition and energy content of paper production waste water was low and it would function only as a diluent. The environmental impact assessment of fiber recycling line of Laminating Papers Oy's was used as the source of information on this stream. (Punta, E. 2010)

Fluid pressed from municipal organic waste was suggested by the ALDIGA research team's professor Martin Romantschuk due to his prior knowledge of the concept. It is very high in nutrients and energy but highly acidic in terms of ideal pH. It should be neutralized with an equally basic stream, or diluted. The information for this stream was received from Irene Bohn at Helsingborg University via e-mail, and from a report of the project. (Bohn, I. 2010, Avfall Sverige 2010)

Fish farm water was suggested by two companies, and it is a safe substrate that could be most likely used without any further handling. This stream was studied personally since insufficient information was available. It was also valuable for comparing the time and the effort spent in producing results from a single stream. Samples were taken from the fish farm in Imatra, in the paper industrial area. Reference data was found in a scientific article, but at first it was considered rather incompatible because of the different type of cultivation (Stephens, W. 2003).

Cooling water was suggested by one company. It is generated in the processes of biogas plants, and would be mainly a source of heat for the algae. The water would be directed out after the stripping and evaporation processes at the biogas plant. Analyses showed that cooling water was in fact richer in nitrogen than e.g. municipal waste water and fish farming water. Biovakka Oy provided analyses also for this stream after they were contacted. (Biovakka Oy 2010)

Distillery effluents fall under food industry waste waters, which were suggested by two companies. They are both before and after methanation the richest in organic carbon, meaning energy for the heterotrophic algae. This can be seen as a high oxygen demand, both chemical and biological. However, the total solids in these streams are extremely high, and they share other characteristics with the press water of municipal organic waste as well. Therefore, solid matter needs to be filtered to not hinder the extraction of the algae. Combining distillery effluent with reject water from biogas plants would produce a mixture with the highest content in all of the parameters studied. Three companies were contacted concerning their effluents but they were unable to produce any data for the project. A scientific article was used for the information on this stream in both forms, before and after methanation. (Biswas, A.K. 2009)

Pasta and flour plant effluent was a personal suggestion to complement the food industry waste waters. Since the local companies were unable to provide any analyses as in the case of distillery effluents, a scientific article was used instead. (El –Salam, M.M. Abd 2010)

Leachate from landfills was suggested by two companies but it is ultimately not a suitable stream for algae cultivation. It showed low nutritional and energetic value and it tends to be cool and it may be toxic. Päijät-Hämeen Jätehuolto Oy (a local waste management company) provided a comprehensive spreadsheet on their leachate upon contacting.

Carpet industry effluent was a personal suggestion that was found during the gathering of information. It seemed like an opportune stream to be used as a reference since an existing article already studied its use as microalgal feed for biodiesel production. According to the article, a selection of 15 different species of algae removed over 96 % of nutrients in the medium in 72 hours, and was able to do so in a steady temperature of 15 °C. The source of information for this stream was a scientific article. (Chinnasamy, S. 2009)

3.4 Analysis of the fish-farming water of Imatra

The water samples were taken from the discharge well after the water had been circulated from inside tanks to outside tanks, and filtered of excess solid matter between the two systems. There was no filtration after the outside tanks. The water contains some fish fodder that was not eaten and excrement that were not re-

- Total solids through GF/C fibre-glass filter.
- Disposable cuvette tests for spectrometric analysis by Lange.
 - LCK414, COD_{Cr} cuvette test measuring range 5-60 mg/l
 - LCK350, Phosphate ortho/total cuvette test measuring range 2.0-20 mg/l PO₄-P
 - LCK349, Phosphate ortho/total cuvette test measuring range 0.05-1.5 mg/l PO₄-P
- OxiTop® for BOD7
- Ammonium, Merckoquant® test strips: 110024 Ammonium Colorimetric Test with test strips and reagent 10 - 30 - 60 - 100 - 200 - 400 mg/l NH4⁺
- Nitrate, Merckoquant® test strips: 110020 Nitrate Colorimetric Test with test strips 10 - 25 - 50 - 100 - 250 - 500 mg/l NO₃⁻

Table 3: The results of the analyses of the spent pool water of the Imatra fish-farmParameter1. mg/l2. mg/l3. mg/l4. mgl/l

Total N	6,61	6,6	n/a	n/a
NO ₃	25	n/a	n/a	n/a
NH_4	0-5	n/a	n/a	n/a
Total P	0,234	0,215	0,313	0,376
PO_4	0,709^*	0,653^	0,948*	1,14*
BOD ₇	4,5	5,9	1,1 ^a	
COD _{Cr}	34,4	n/a	n/a	n/a
Total solids	1,5	2,65	2,4	5,25

* : done with LCK350 – below measuring range

^ : orthophosphate

^a : tap water reference

n/a : an additional analysis was not done

4 COMPILING THE SPREADSHEET

The spreadsheet was made functional as soon as possible, and initially it contained a number of parameters that would be removed later on due to lack of information in most articles. Toxicity, though containing a value for only one stream, remains in the final version of the table because it is one of the main reasons for ruling out leachate as an alternative. For future use of the table, toxicity as a parameter will prove useful if the streams come from sources that are more highly contaminated as opposed to the current streams.

The values of the parameters in the analyses provided by the companies were sufficiently similar to the corresponding ones found in the Internet articles, however, the number of reference material was quite small as mostly one to three sources were gathered for each stream. Since the waste streams that will be utilized are obviously not exactly identical to e.g. the ones of an Egyptian pasta plant, it is sufficient for the values to be merely indicative. The main functions of the parameters are explained in Table 4. Altogether, 20 references were used in the spreadsheet.

Table 4: Parameters of the streams explained

Material	Name of the waste stream, sewage, effluent
Source example	A possible location that emits the stream
Total Nitrogen	Total atomic amount of N, not directly bioavailable to algae
Nitrate NO ₃	Primary nutrient, bioavailable to algae
Ammonium NH ₄	Primary nutrient, bioavailable to algae
Total phosphorus	Total atomic amount of P, not directly bioavailable to algae
Phosphate	Primary nutrient, bioavailable to algae
BOD ₇	Biological oxygen demand during 7 days. Indicates energy in the form of organic carbon used in biological activity
COD _{Cr}	Chemical oxygen demand, includes BOD
Potassium	Trace element
Sulphur	Trace element
Total solids	Hinders the harvesting of algae and extraction of lipids
рН	Critical property in the solution to all biological activity
Temperature	Indicates the necessity to heat up the solution with other streams
Conductivity	Indicates salinity, low conductivity is suitable for algae
References	Link to the source of information
Notes	Complementation to the nature of the stream

The dissected version of the spreadsheet tool imported into this thesis is in four parts. The first two parts, Table 5 and Table 6, show the first set of parameters for all streams and the third and fourth part,

Table **7** and Table **8**, show the second set of parameters for all the streams. In the source examples of the spreadsheet tool, WWTF is short for waste water treatment facility. Also in the same column, Helsingborg inside the brackets means municipal organic waste tested by Helsingborg University and Avfall Sverige. However, it can be recreated in recycling centers and landfills from municipal organic waste.

Material	Source ex- ample	Total N (mg/l)	NH ₄ (mg/l)	NO ₃ (mg/l)	Total P (mg/l)	PO ₄ (mg/l)
Cattle manure (digested) Swine manure (undigested)	Bio gas plant Bio gas plant	569	180,5 350		441	97 331
Swine and Sewage mix	Bio gas plant & WWTF	52	18		21	13
Municipal w. water	WWTF	55	13		20	19
Org. W. Press Water	(Helsingborg)	3069	474,3		39,4	
Paper produc- tion w. water	Water facility / paper mill	5,78			0,83	

Table 5: First part of the dissected spreadsheet tool

Table 6: Second part of the dissected spreadsheet tool

Material	Source ex- ample	Total N (mg/l)	NH ₄ (mg/l)	NO ₃ (mg/l)	Total P (mg/l)	PO ₄ (mg/l)
Reject water	Bio gas plant	5 150	3 740	3 740	555	520
Fish farm water	Fish farm	6,61	1	5	0,3762	0,653
Cooling water Leachate	Bio gas plant Landfill	100 165	95		0,1 1,0	
Leachate	facility	105			1,0	
Untreated Car- pet / mun. Ww	Adjacent industry	38,85	21,72	14,175	9,655	27,71
Distillery efflu- ent SW		2020			240	

Distillery efflu- ent PME	1310			130	
Pasta and flour plant effluents	10,15	0,25	0,655	0,658	0,658

					_		Total	
	Κ		BOD_7	COD_{Cr}	Т		Solids	σ
Material	(mg/l)	S (mg/l)	(mg O2/l)	(mg O2/l)	(°C)	pН	(mg/l)	(µS/cm)
Cattle manure (digested)				3317,5		7,915	3969	
Swine manure (undigested)			6414,8	10189		6,5	7210	
Swine and Sewage mix			208,6	316		7,5	298	
Municipal w. water			270,2	292		7,8	313	
Org. W. Press Water						4,3	93000	
Paper produc- tion w. water		242	8,8	118,8			44,2	

Table 7: Third part of the dissected spreadsheet tool

Table 8: Fourth part of the dissected spreadsheet tool

Matarial	V (S	BOD ₇	COD_{Cr}	T		Total Solids	σ
Material Reject water	K (mg/l) 1600	(mg/l) 600	(mg O2/l) 4400	(mg O2/l)	(°C)	<u>рН</u> 8,2	(mg/l) 3300	(µS/cm) 0,5
Reject water Fish farm water	1000	000	5,2	34,4	25,5	8,2 6,9	2,1	0,3
Cooling water				1850		6,6		
Leachate			162,5	575	8,15	7,7	58,25	310
Untreated Carpet / mun. Ww	9,04	114,9	409,5	1413	15	6,865	1416	878
Distillery effluent SW	13000	2820	36000	113200			98000	
Distillery effluent PME	11500	850	4000	39200			43000	
Pasta and flour plant effluents		43,15	31	72,4	33,8	7,9	705,5	544

5 DISCUSSION AND CONCLUSIONS

The most concentrated streams based on the nutrient content is reject water from biogas plants, organic press water and distillery effluents (Table 5 and Table 6). Organic carbon and therefore the energy content determined by the biological and chemical oxygen demand is the highest in distillery effluent, swine manure and reject water (Table 7 and Table 8). The organic carbon is beneficial considering the heterotrophic energy strategy of algae. These streams have to be diluted with the low concentration streams or other streams that were not studied in this work. The transportation of such concentrated streams could also be feasible since they should in any case be diluted into a fraction of the original concentration.

Contrary to expectations, the municipal sewage, pasta plant effluent, fish farming water, paper production waste water and carpet production waste water are the lowest in nutritional and energetic content. This type of waste water could probably support algae cultivation on its own but a source would have to be nearby. Landfill leachate and the cooling waters are actually richer in terms of nutrition and energy content, and they are categorized as semi-potential along with the more obvious one - manure. As the leachate and cooling water might be toxic and because the leachate is generally quite cold, averaging at 8 degrees Celsius, they can not be used to dilute and warm up the solution. Cooling water could be used as a source of heat if no toxicity is present.

Most of the streams seemed valuable as a part of the mixture to be used in algae cultivation and a few might be used as the sole substrate. The problematic and increasing amount of municipal and agricultural waste waters would be vital to include in the process of producing fuel as they are currently consuming energy being treated in conventional waste water treatment plants. Especially the concentration of municipal waste water is not optimal to the microbes working in waste water treatment plants but for algae requiring dilute solutions it would be ideal.

The objective was to gather the suggestions of the cooperating companies, and other opportune streams to be used in the project, and to map the parameters of the streams from either existing analyses, or from scientific articles of similar cases. In the end, 14 streams were mapped and only one stream from the suggestions - washing waters - was not mapped due to lack of information. In addition, the compatibility of washing waters for the project was questionable. Problems could arise because of possible toxic substances, and as an unspecific stream the washing waters have an inconsistent content as opposed to e.g. manure.

The Imatra fish farming water analyses proved that producing the desired data yourself from a certain source is by far more resource consuming than searching for a similar case from existing articles. Even if the information in this situation might not be of exact fit, it is practically impossible in terms of time and funds to go and take samples from every source included in the study and do the analyses on them.

The documents sent by the companies had data on exactly the streams that they would provide for the project in the future but they lacked many of the needed parameters. This made it necessary to search for complementary information from outside analyses, and while doing this I noted that the articles' parameter values were quite parallel to the ones provided by the companies.

The work at this point has achieved its purpose and it has been essential to the testing and the to-be-started large scale cultivation. The future use was kept in mind and the spreadsheet tool was planned from the start to be a template for new applications. The broken links to source files in the spreadsheet remained as an unsolved problem. When the spreadsheet is copied to another location, the links have to be manually corrected to refer to the new location. The tool would work best if it were into a website.

As a personal experience, goals for learning teamwork and how large projects function as well as learning techniques for finding information, were reached. Finding the information from scientific articles proved very convenient after trying different keywords and browsing through the keywords used in some articles that were quite similar to the ones that were needed. The majority of the articles used in this work were found from Sciencedirect, a database search service under Elsevier B.V., which is a major publisher of scientific articles. However, a licensed login of an institute is required to read and download complete articles making it necessary to use a computer networked to the institute's systems for the search. Fluent English skills proved essential while studying the articles and trying keywords because a number of synonyms had to be tried to find the articles. Especially the search function of a pdf-reader software for searching multiple files at a time made it convenient to download many promising articles per session and then search through all of them later for the needed data. This made it practical to work at home when it was not necessary to spend a lot of time at an institute library computer.

REFERENCES

Algae Resource Database 2011. NIES-48 Euglena gracilis [Cited 7.3.2011] Available online at <u>http://www.shigen.nig.ac.jp/algae/images/strainsimage/nies-</u> 0048.jpg

Aquaportail 2011. NIES-629 Chlorella protothecoides. [Cited 7.3.2011] Available online at http://www.aquaportail.com/pictures0906/anemone-

clown_1245142448_chlorelle-chlorella-protothecoides.jpg

Aquaportail 2011. NIES-642 Chlorella vulgaris var. vulgaris. [Cited 7.3.2011] Available online at <u>http://www.aquaportail.com/pictures0906/anemone-</u> <u>clown_1245142384_chlorelle-chlorella-vulgaris-var-vulgaris.jpg</u>

An, J.Y. et al. 2003. Hydrocarbon production from secondarily treated piggery wastewater by the green alga Botryococcus braunii. Journal of Applied Phycology. Vol. 15, Iss 2-3, p. 187. Available at Ingentaconnect database of Publishing Technology

http://www.ingentaconnect.com/content/klu/japh/2003/00000015/F0020002/0511 8626

Avfall Sverige 2010. Rapport B2010:01, Utvärdering och optimering av metod för förbehandling av källsorterat hushållsavfall till biogasproduktion. Malmö: Avfall Sverige AB

Bashan L.E. de-. et al. 2003. Microalgae growth-promoting bacteria as helpers for microalgae a novel approach for removing ammonium and phosphorus from municipal wastewater. Elsevier, Water Research Vol. 38, Iss. 2, p. 466-474. Available at Science Direct database of Elsevier <u>doi:10.1016/j.watres.2003.09.022</u>

Biovakka Suomi Oy 2010. TESTAUSSELOSTE 10-3098-1 & 10-3509-1. Turku: Lounaissuomen vesi- ja ympäristötutkimus Oy. Biovakka Suomi Oy 2010. TUTKIMUSRAPORTTI 101310049 & 101310133. Mikkeli: Viljavuuspalvelu Oy. Envor Biotech Oy 2010. TUTKIMUSRAPORTTI N:o K 423/10/6. Karkkila: Novalab Oy.

Biswas, A.K. et al. 2009. Distillery effluents effect on soil organic carbon and aggregate stability of a Vertisol in India. Elsevier, Soil and Tillage Research Vol. 104, Iss. 2, p. 241-246. Available at Science Direct database of Elsevier doi:10.1016/j.still.2009.02.012

Bohn, I. 2010. Re: pressvätska från avfall [email] Recipient Kautto, A. Sent 18.10.10

Chen, C.-Y., et al. 2010. Cultivation, photobioreactor design and harvesting of microalgae for biodiesel production: A critical review. Bioresource Technology. Vol. 102, Iss. 1, p. 72. Available at Science Direct database of Elsevier doi:10.1016/j.biortech.2010.06.159

Chinnasamy, S. et. al. 2009. Microalgae cultivation in a wastewater dominated by carpet mill effluents for biofuel applications. Elsevier, Bioresource Technology Vol. 101, Iss. 9, p. 3097-3105. Available at Science Direct database of Elsevier doi:10.1016/j.biortech.2009.12.026

Chisti, Y. 2007. Biodiesel from microalgae. Elsevier, Biotechnology Advances Vol. 25, Iss. 3, p. 294–306. Available at Science Direct database of Elsevier doi:10.1016/j.biotechadv.2007.02.001

Chlorenergy 2011. About Chlorenergy Chlorella [Cited 4.4.2011] Available online at <u>http://www.bestchlorella.com/product.htm</u>

Cosmetic Analysis 2011. Chlorella Vulgaris Extract [Cited 4.4.2011] Available online at http://www.cosmeticanalysis.com/cosmetic-ingredients/chlorellavulgaris-extract.html Directive of the European parliament and of the council of 8 May 2003 on the promotion of the use of biofuels or other renewable fuels for transport 2003/30/EC. Available online at <u>http://eur-</u>

lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2003:123:0042:0046:EN:PD <u>F</u>

El-Salam M. M. Abd. et al. 2010. In-plant control for water minimization and wastewater reuse a case study in pasta plants of alexandria flour mills and bakeries company, Egypt. Elsevier, Journal of Cleaner Production Vol. 18, Iss. 14, p. 1403-1412. Available at Science Direct database of Elsevier doi:10.1016/j.jclepro.2010.05.006

European Biodiesel Board 2010. 2009-2010: EU biodiesel industry restrained growth in challenging times. Press Release. [Cited 26.3.2011] Available online at http://www.biofuelstp.eu/news/EBB_2009_prod_2010_capacity.pdf

European Biodiesel Board 2010. About biodiesel [Cited 26.3.2011] Available at http://www.ebb-eu.org/biodiesel.php#eupro

European Commission Eurostat 2010. Energy production and imports. [Cited 15.2.2010] Available online at http://epp.eurostat.ec.europa.eu/statistics_explained/index.php/Energy_production _and_imports

Europe's Energy Portal 2011. [Cited 30.3.2011] Available online at <u>http://www.energy.eu</u>

Finlex 2010. Laki biopolttoaineiden käytön edistämisestä liikenteessä annetun lain muuttamisesta HE 197/2010. Available online at http://www.finlex.fi/fi/esitykset/he/2010/20100197

Lin, L. et al. 2007. Use of ammoniacal nitrogen tolerant microalgae in landfill leachate treatment. Elsevier, Waste Management Vol. 27, Iss. 10, p. 1376-1382.

Available at Science Direct database of Elsevier doi:10.1016/j.wasman.2006.09.001

Molina, E. et al. 2001. Tubular photobioreactor design for algal cultures. Elsevier, Journal of Biotechnology Vol. 92, Iss. 2, p. 113–131. Available at Science Direct database of Elsevier <u>doi:10.1016/S0168-1656(01)00353-4</u>

Oilgae 2008. Biodiesel producers can burn glycerol in Combined Heat and Power (CHP) generators. Oilgae Blog [Cited 3.3.2011] Available online at http://www.oilgae.com/blog/2008/12/biodiesel-producers-can-burn-glycerol.html

Oilgae 2010. Transesterification. Oilgae [Cited 1.12.2010] Available online at http://www.oilgae.com/algae/oil/biod/tra/tra.html

Oilgae 2010. Algae Oil Extraction. Oilgae [Cited 1.12.2010] Available online at http://www.oilgae.com/algae/oil/extract/extract.html

Orpez, R. et al. 2008. Growth of the microalga Botryococcus braunii in secondarily treated sewage. Elsevier, Desalination. Vol. 246, Iss. 1-3, p. 625-630. Available at Science Direct database of Elsevier <u>doi:10.1016/j.desal.2008.07.016</u>

Pittman, J.K., et al. 2010. The potential of sustainable algal biofuel production using wastewater resources. Elsevier, Bioresource Technology Vol. 102, Iss. 1, p. 17-25. Available at Science Direct database of Elsevier doi:10.1016/j.biortech.2010.06.035

Punta, E. 2010. YVA-arviointiohjelma Kierrätyskuitulinja Laminating Papers Oy. Joensuu: Linnunmaa Oy.

Päijät-Hämeen Jätehuolto Oy 2010. Käyttötarkkailu_2009.xls. Lahti.

Queensland Government 2010. Ecosystem health indicators. [Cited 15.12.2010] Available online at http://www.derm.qld.gov.au/environmental_management/water/water_quality_mo nitoring/assessing_water_quality/water_quality_indicators.html

Sialve, B et al. 2009. Anaerobic digestion of microalgae as a necessary step to make microalgal biodiesel sustainable. Elsevier, Biotechnology Advances Vol. 27, Iss. 4, p. 409-416. Available at Science Direct database of Elsevier doi:10.1016/j.biotechadv.2009.03.001

Stephens, W. 2003. Instream community assessment of aquaculture effluents. Elsevier, Aquaculture Vol. 231, Iss. 1-4, p. 149-162. Available at Science Direct database of Elsevier <u>doi:10.1016/j.aquaculture.2003.08.009</u>

Travieso Cordoba, L. et. al. 1994. Final treatment for cattle manure using immobilized microalgae II Influence of the recirculation. Elsevier, Resources, Conservation and Recycling Vol. 13, Iss. 3-4, p. 177-182. Available at Science Direct database of Elsevier <u>doi:10.1016/0921-3449(94)00019-2</u>

Travieso, L. et al. 2006. Production of Biomass (Algae-Bacteria) by Using a Mixture of Settled Swine and Sewage as Substrate. Journal of Environmental Science and Health Part A, 41:415–429 Available at informaworld database of Taylor & Francis Group

http://www.informaworld.com/smpp/content~db=all?content=10.1080/109345205 00428302

Ullman, J. 2006. The difference between Chlorella vulgaris and Chlorella pyrenoidosa. Algomed. Available at <u>http://www.algomed.de/media/publikationen/algomed10eng.pdf</u>

University of Texas in Austin 2011. The Culture Collection of Algae [Cited 4.4.2011] Available online at <u>http://web.biosci.utexas.edu/utex/</u>