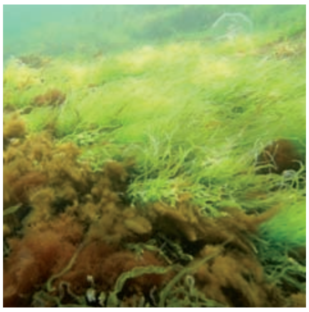
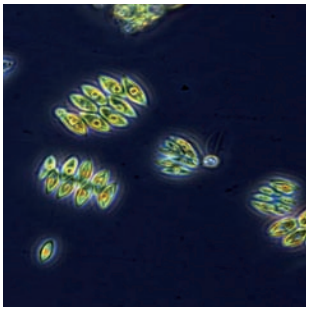
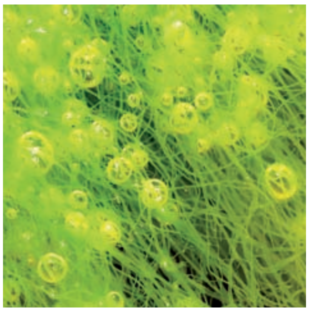
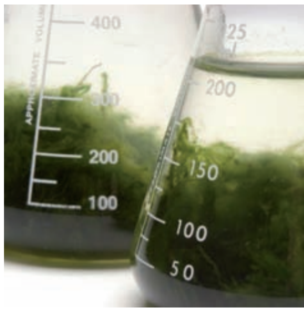


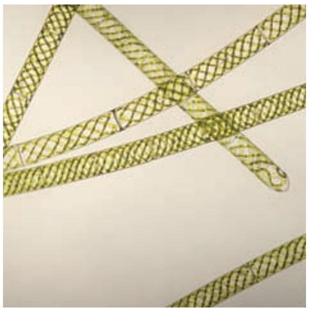
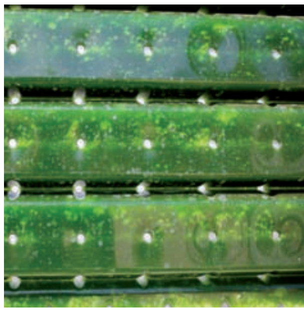




ALGAE: THE SUSTAINABLE BIOMASS FOR THE FUTURE

PERSPECTIVES FROM THE SUBMARINER PROJECT ALGAE COOPERATION EVENT
TRELLEBORG, SWEDEN - SEPTEMBER 28-29, 2011






SUBMARINER

Baltic Sea Region
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DISCLAIMER

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FOREWORD

Global climate change to our earth is rapidly reducing the available land-based food and energy resources. We are all aware of the nescessity to slow down the speed of climate change and to simultaneously find other ways of producing food and energy. In this context, the vulnerable Baltic Sea must be regarded, not as a slowly dying patient, excessively polluted by its catchment area population of about one hundred million inhabitants, but instead as the very valuable sea resource that it is, with a very high potential for increased food and energy production.

Mobilising all modern research forces to transform the excessive release of nutrients into our sea into valuable material for blue technology is a possible future solution to mastering the negative climate trends and saving our sea and its inhabitants from eutrophication before it is too late.

As one of many coastal zone municipalities in the Baltic region, the City of Trelleborg has great hopes for the future, as it learns and gains expertise in the new and very promising uses of algae, both from the seas and grown in freshwater. We are presently concentrating our efforts on the large-scale production of biogas from macroalgae in the southern Swedish coastal region, in combination with agriculture sector residues, and using the gas for transport and energy uses. We also strive, with voluntary stakeholders, to make shipping in the Baltic Sea greener, by using organic wastes from ships as a valuable biogas resource.

By joining the SUBMARINER Project, the City of Trelleborg hopes to share its enthousiasm for this type of potential solution with others around the Baltic Sea. As a platform for this exchange, Trelleborg hosted the first Baltic Sea Region Algae Cooperation Event, held on September 28th and 29th, 2011.

The event brought together distiguished experts as well as stakeholders from the Baltic Sea region and other parts of the world to present the state of the art in the fields of algae-related green energy, food production and other uses. This magazine present the perspectives shared by the speakers during the event.

The Algae Cooperation Event very clearly demonstrated the necessity to move from solemnly launched visions into the real application of large-scale uses of algae, as a vital component of a sustainable future for our Baltic Sea. To start using blue technology at full scale in all Baltic Sea region countries is a must. This cooperation event has shown us the importance of this concept and given us a functioning roadmap for how this can be achieved.

Sten Björk

Environmental Strategist
Trelleborg City Council

INTRODUCTION

The Baltic region is biased towards the sea: the maritime economy is one of the strongest driving forces in our region. For hundred years we have been using our Baltic for various purposes: fishing, mining, transportation, tourism, etc. The pressures on the marine ecosystem have concurrently increased and we now face a situation in which marine resources are limited while their demand remains high.

New scientific and technological developments provide opportunities for innovative uses of marine resources that have the potential to bring positive environmental benefits, offering at the same time new employment possibilities. If we can guide future development toward these uses, we may be able to proactively contribute to the development of a healthy Baltic Sea while at the same time promoting sustainable economies. It is high time for such an approach: without a clear sense of future direction, the region faces the risk of further degrading the marine ecosystem on which it so tightly depends, which would likely result in economic decline.

The SUBMARINER Project is about assessing the environmental impacts of new marine uses, their economic feasibility and regional applicability. We believe that among the new marine uses that are more or less rapidly or intensively developing in other parts of the word, there are some which could benefit both our marine ecosystem and the economy of our region.

We are elaborating a 'Compendium of Innovative Marine Uses', which may serve as a support tool for decision-makers as it will present development pros and cons for each new use analysed as well as the potential combination of them. We also aim to elaborate a first 'Baltic Sea Region Roadmap for Development of New Marine Uses', in which we will present recommendations for how to promote those uses which are most desirable.

SUBMARINER is also taking the first step towards integrating the constituencies behind these new uses: those who already make use of marine resources in innovative ways as well as those who are interested in investing in or implementing these new opportunities. To this end, SUBMARINER is gathering interested actors through a series of thematic cooperation events devoted to the most promising innovative uses. The Algae Cooperation Event in Trelleborg, Sweden is the first in such a series.

The Maritime Institute in Gdansk has the honor of leading representatives from 18 partner organizations comprising environmental ministries and agencies, regional development agencies and scientific institutes. The project has a total budget of €3.6 million and will run until autumn 2013. We invite you to learn more about SUBMARINER and its events on its website: www.submariner-project.eu.

Joanna Przedrzymirska

SUBMARINER Lead Partner
The Maritime Institute in Gdansk

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The Baltic Sea is a basin under great environmental stress. Over time, natural factors that have rendered the sea sensitive to environmental alterations, in combination with human activities, have modified sea conditions. Recently, these altered conditions have led to severe environmental problems. At present, eutrophication against a background of climate change and organic pollution is the most serious environmental threat to the Baltic Sea.

The increased nutrient load stimulates growth of filamentous macroalgae such as the red alga *Polysiphonia fucoids* and the green algae *Enteromorpha* sp. and *Cladophora rupestris*. The algae form a large biomass in shallow waters and also accumulate on beaches, especially in southern Sweden. The algae prevent the beaches from being used for recreation and also release an unpleasant smell. Changes in the local marine ecosystem such as large important fish and bird feeding and reproduction areas becoming non-functional are other problems associated with this macroalgae. In the brackish Baltic Sea proper, noxious blooms of cyanobacteria are a common phenomenon. The toxic cyanobacteria *Nodularia spumigena* in particular has attracted its share of media attention. These bacteria form massive, poisonous blooms on the surface waters, polluting swimming and recreation areas along the coast.

The municipality of Trelleborg, which has Sweden's richest soils and thus very extensive farming, aims to serve as a model for sustainable development in the southern part of the Swedish Baltic Sea region. Together with local farmers and landowners, Trelleborg will establish large-scale biogas production based on new wetlands established along the coastal zone of southern Sweden. Growth and harvesting of reed beds and submerged vegetation for bioenergy producing will take place in these wetlands. Trelleborg will use the biogas for transportation fuel, heating of domestic houses in urban areas and local production of electric power.

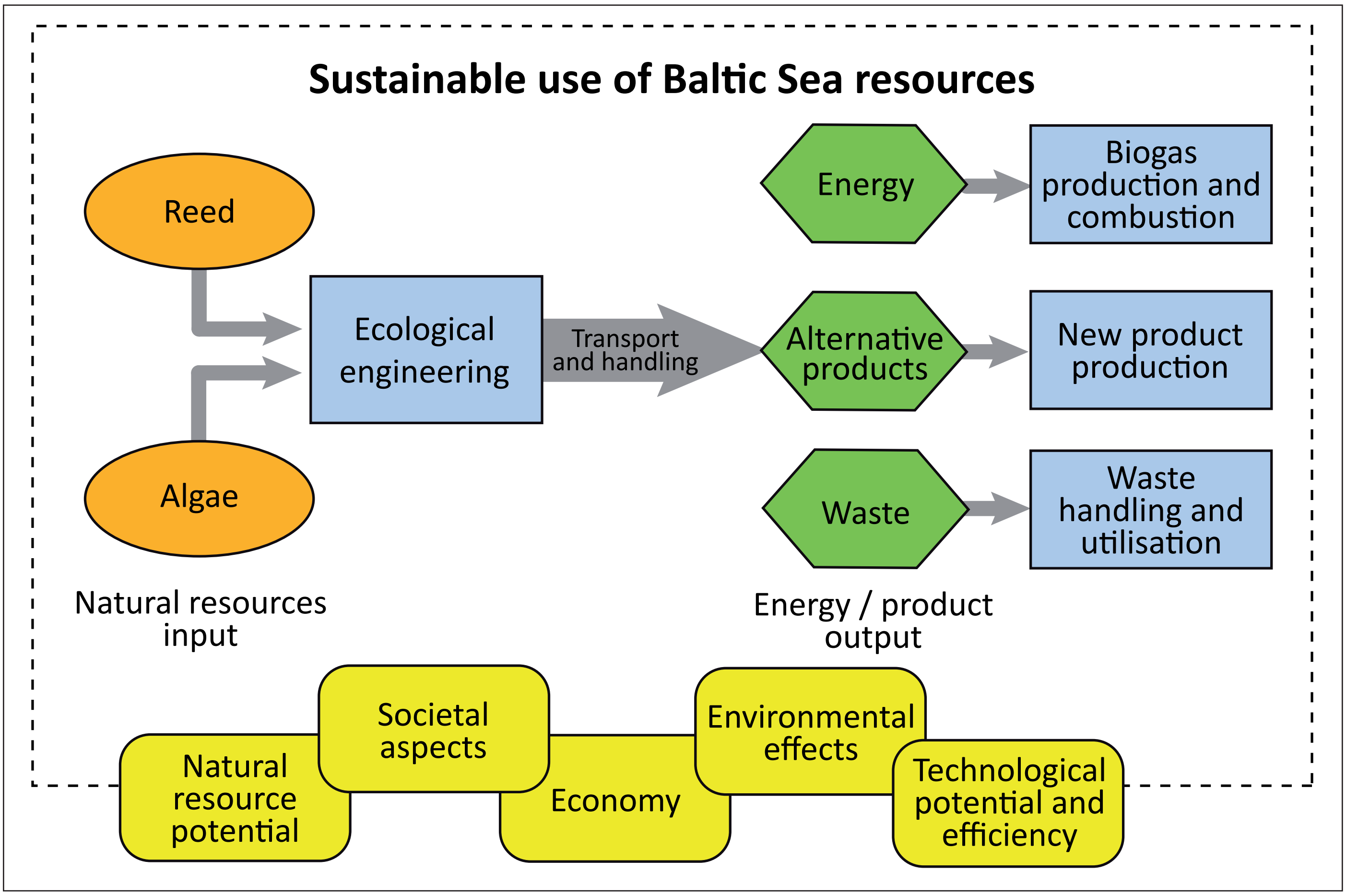
Floating red filamentous algae on the Swedish south coast. Photo: F. Gröndahl

Nutrients reaching the coastal waters from other sources, such as traffic or sewage treatment, and from other geographical areas will be removed with new technology for collecting macroalgae from the sea. The biomass collected will be used for biogas production. During summers with extensive blooms of cyanobacteria, surface accumulations of *Nodularia spumigena* will be harvested and also used for biogas production.

All these new means of removing nutrients from the highly eutrophied Baltic Sea in a low-intensity but steady process could bring about a much needed reversal in nutrient flow if they were to achieve widespread use among the coastal regions. The Trelleborg concept of transforming a problem into a resource by preventing eutrophication through biogas production has a number of benefits for the environment in the region.

A project working with a system analysis of all stages from harvest of algae in the Baltic Sea to biogas production has been initiated in cooperation with the Municipality of Trelleborg. The project deals with the ecological, social and technological aspects in the production chain and includes comparisons with other biomasses such as reed. The project started in summer 2009 and will continue until 2013.

The project will investigate if harvesting of reed and algae as well as mussel farming in the coastal zone and gathering of blue green algae in the open sea may form the basis for sustainable use of natural resources in this area of the Baltic Sea. The project aims to assess sustainability from an environmental, economic and social perspective and analyse both the technology and how much natural resource there is in the area. The project will be done in close cooperation with the municipality and companies operating in the region of Trelleborg. There are few scientific studies that evaluate the sustainability of a project that tries to take a holistic approach. Developing technological solutions to environmental problems is a high priority research field in today's society. If the project is successful, it can serve as a model on how to solve sustainability problems in coastal areas. The project is supported by FORMAS, the Swedish Research Council for Environment, Agricultural Sciences and Spatial Planning.



Sustainable use of Baltic sea biomass resources based on ecological retrieval of biomass (reed and algae) for production of energy and new innovative products (including fertilizers) with an associated waste stream. Blue squares indicate technology-dependent processes and the yellow squares show the sustainability and feasibility aspects.

BEACH-CAST SEAWEED FOR BIOFUEL IN SCOTLAND: ECOLOGICAL CONSIDERATIONS

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Large quantities of seaweed are cast ashore onto Scottish beaches every year, particularly on the west coast of Outer Hebrides after storms. Kelp (*Laminaria* sp.), which forms a major component of the beach-cast seaweed is a proposed source of biomass for biofuel. However, beach-cast seaweed plays a vital role in both terrestrial and marine food webs and its ecological importance needs to be fully assessed before this resource can be sustainably exploited. This project, which is funded by Biomara[1], aims to predict the ecological consequences of removing seaweed for biofuel, using both a field-based and an ecosystem modeling approach.

In order to ask questions about the impacts of removing seaweed for biofuel it is necessary to have a clear quantitative understanding of its ecological role. This involves collecting data on the animals living in or associated with beach-cast seaweed, their predators and all other available food sources. Once this data is collected it is possible to build a mass-balanced ecosystem model - essentially a mathematical representation of an ecological system - using software such as Ecopath with Ecosim. The model can thereafter be used to predict how human activities (such as harvesting seaweed for biofuel) may effect biodiversity and ecosystem functioning.

Fresh beach-cast seaweed in North Uist, Outer Hebrides, November 2010. Photo: K. Alexander (SAMS)

For example, what would be the effects if 50 % of available beach-cast seaweed was removed?

Beach-cast seaweed supports large and diverse communities of consumers by increasing food and habitat availability. Because there is little *in situ* primary production on the beach itself, macrofauna communities inhabiting beaches can be supported almost entirely by the input of organic matter from sea in the form of allochthonous material. To date there have been no studies on the west coast of Scotland that demonstrate the importance of beach-cast seaweed to coastal invertebrates and birds. However, evidence from other countries suggests that its removal would lead to impoverished coastal invertebrate and bird communities.

Decomposition of beach-cast seaweed can supply a vital source of nutrients and particulate organic matter to subtidal and surf-zone communities. Accumulations of detached macroalgae in the surf-zone have been shown to be an important source of food to small crustaceans, which are then preyed upon by juvenile fish and larger crustaceans. Floating macroalgae are also thought to protect fish and possibly crustaceans from larger predators, which would feed less efficiently among patches of weed.

This study has found that the amount of beach-cast seaweed present is highly variable seasonally and is strongly driven by wind direction and strength. This would raise issues for biofuel feedstocks in terms of reliability and security of supply. The fauna inhabiting and feeding off seaweed also varies seasonally. For example the number of birds feeding on beaches is highest during the over-wintering and migration periods. This would raise conservation questions about the best time of year for harvesting seaweed.

As part of this research, beaches both with and without seaweed were compared in a 'natural experiment' in order to make some assumptions about what the beaches would look like (ecologically speaking) if all the seaweed was removed. Beaches were sampled for animals both on the beach (between the high-water and low-water lines) and in the surf-zone where floating detached macroalgae occurs. It was found that there is a direct linear relationship between the abundance of birds on the beach and the amount of beach-cast seaweed. This indicated that beaches with seaweed are highly important feeding grounds for several shorebird species. In the surf-zone the abundance of small crustaceans also increased as the amount of floating macroalgae increased, again indicating the importance of detached seaweed as a food resource for coastal invertebrates. Beaches without cast seaweed were relatively barren and lifeless and it is clear that beach-cast seaweed plays a vital role in maintaining coastal biodiversity and functioning.

In summary, the main ecological concerns related to the harvesting of beach-cast seaweed include: disturbance to vulnerable or threatened shorebirds; the long-term effect of reduced nutrient input to the nearshore environment; and loss of production of other species (such as fish and crustaceans) that depend on the nutrients, detritus or species associated with the seaweed. As outlined above, beach-cast seaweed is very important in coastal, littoral and terrestrial food webs, and facilitates the flow of energy between the sea and land, and vice versa. To date there have been few studies that focus on the disturbances associated with selective harvesting of beach-cast macroalgae, however ecosystem modeling is a promising tool that can be used to predict potential impacts. All these factors need to be kept in mind when considering seaweed for biofuel.

1. EU Funding: ERDF through the Interreg IVA Programme, Crown Estate, Highlands and Islands Enterprise, Irish Government, Northern Irish Government.

UTILIZATION OF CAST SEAWEED FOR BIOGAS PRODUCTION: ASSESSMENT OF ENVIRONMENTAL CONSEQUENCES OF THE SOLRØD PROJECT

ANDERS FREDENSLUND[1], THOMAS B. CHRISTENSEN[1], TYGE KJÆR[1], RASMUS B. HANSEN[2] & STEEN DANIELSEN[2]

[1] ROSKILDE UNIVERSITY, DENMARK
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Solrød Municipality has committed itself to reducing total greenhouse gas emissions by 50 % by 2025. A `Sustainable Energy Action Plan´ has been approved by the City Council, in which construction of a biogas plant fed by locally available organic resources is mentioned as a key action to reach this target.

Screening of available materials suggests a viable basis for constructing a biogas plant using three main sources: cast seaweed collected from the beaches at Køge Bugt (20 000 to 40 000 tons/year), organic waste from the local pectin and carrageenan production facility CPKelco (84 000 tons/year) and pig and cattle manure (50 000 to 70 000 tons/year).

The bay of Køge Bugt is, like many other coastal waters in Denmark, nutrient rich. Eutrophication causes an increased production of sea algae. This leads to a large amount of seaweed washing up on the shores of the bay each year. The thick blanket of decomposing seaweed sometimes extends 50 meters into the bay. At present, this cast seaweed is being removed from the beach, creating large amounts of organic waste.

A preliminary study on the environmental and economical consequences of operating a biogas plant using the aforementioned organic materials suggested that the plant will reduce greenhouse gas emissions by 25 000 to 40 000 tons CO_2/year depending on the type of energy utilization. The following factors were taken into account when assessing net reduction of greenhouse gas emissions:

Cast seaweed at Solrød Strand. Photo: A. Fredenslund

Beach cleaning at Solrød Strand, June 2011. Photo: A. Fredenslund

substitution of fossil fuels, use of electricity at the biogas plant, substitution of fertilizer use (using degassed biomass instead of inorganic fertilizers), transport of biomass and reduction of methane emissions from anaerobic decay (for example rotting seaweed).

The most important factor was substitution of fossil fuels. Reduction of methane emissions as a result of seaweed collection - which will undergo partial anaerobic decay if left on the beach - was found to account for up to 30 % of overall reductions in greenhouse gas emissions. The calculations behind assessment of methane from anaerobic decay are, however, based partly on rough estimates. Transport of biomass was found to have little negative impact on net greenhouse gas reduction.

Reduction of nutrients by removal of seaweed was furthermore found to be of high value. Collection of seaweed from Køge Bugt can remove 120 tons of nitrogen per year. The Danish EPA estimates the entire input of nitrogen to Køge Bugt from both natural and anthropogenic sources to be approximately 1 800 tons per year. An action plan to reduce the nutrient load to Køge Bugt is presently in the hearing process. The action plan lists initiatives that will reduce the nitrogen load to Køge Bugt by approximately 86 tons per year at a yearly cost of € 5.7 million. The cost of collecting and pre-treating cast seaweed for production of biogas is estimated to be approximately 1/10 of that and further result in revenue due to energy production.

The next steps toward realizing a biogas plant in Solrød based partly on cast seaweed are preparation of a detailed plant design, securing permits, securing contracts with biomass suppliers and energy companies and setting up a biogas company responsible for constructing and operating the plant.

CYANOBACTERIA HARVESTING IN THE BALTIC SEA: PRELIMINARY ASSESSMENT OF POTENTIAL BENEFITS

JOSEPH SANTHI PECHSIRI, EMMA RISÉN, MARIA MALMSTRÖM, NILS BRANDT & FREDRIK GRÖNDAHL

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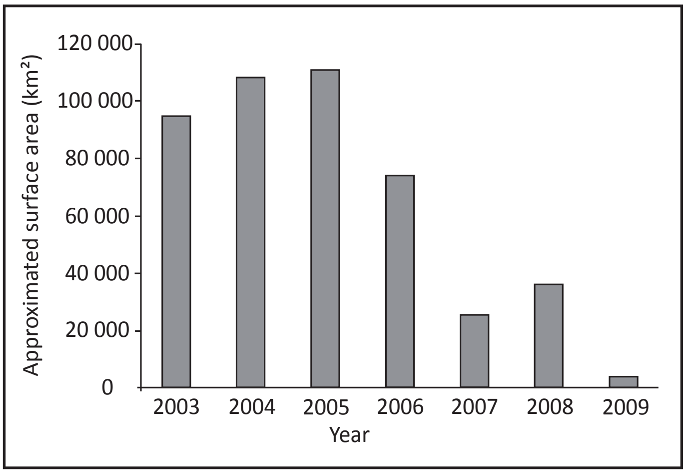
Occurrences of harmful cyanobacteria blooms around the world have caused a variety of environmental and socioeconomic concerns, promoting interest in harvesting wild cyanobacteria. This study investigates the additional benefits in terms of energy production and nutrient removal of harvesting wild cyanobacteria, using the Baltic Sea *Nodularia* sp. and a recently suggested method of harvesting as a case study. The wild cyanobacteria harvesting method involves using modified oil booms to skim over the Baltic Sea surface and harvest floating cyanobacteria flocculent.

The harvesting potential of one boom unit, the total biomass available on the day with most widespread blooms in the period 2003-2009, the biogas production potential of harvesting wild cyanobacteria and the nutrient removal potential of wild cyanobacteria harvesting were quantitatively assessed. The results suggest that the potential of wild cyanobacteria harvesting is substantial (mainly *Nodularia* sp.), with considerable potential for nutrient removal as an additional benefit.

Results suggest that nitrogen removal potential from harvesting the total available wild cyanobacteria during the day of the largest bloom in the Baltic Sea is approximately 1 500 - 45 000 tonnes N depending on the size of cyanobacteria blooms, assuming maximum biomass concentration. However, the biogas production potential was low. Furthermore, the location and duration of powerful blooms were found to be the main limiting factors in harvesting cyanobacteria from the wild.



Surface areas of the largest powerful cyanobacterial blooms in the Baltic Sea.

Testing the modified oil boom. Photo: F. Gröndahl

BIOMASS POTENTIAL OF MACROALGAE HARVESTING ON A REGIONAL SCALE: THE CASE OF TRELLEBORG, SWEDEN

EMMA RISÉN, JOSEPH SANTHI PECHSIRI, MARIA MALMSTRÖM, NILS BRANDT & FREDRIK GRÖNDAHL

DEPARTMENT OF INDUSTRIAL ECOLOGY, THE ROYAL INSTITUTE OF TECHNOLOGY (KTH), SWEDEN

Today, several regional pilot projects aiming at retrieving biomass from the Baltic Sea costal zone have been initiated. The main aim with these projects is to balance municipal nutrient flows in the Baltic Sea coastal area by retrieving nutrients. There are also a number of secondary benefits from the projects, such as biogas production, locally increased water clarity as well as reduced odour problems in recreational areas.

However, there are ecological, economical and social risks with removal of biomass from the ecosystem. There is a lack of scientific studies that consider these risks and it is of utmost importance to address this before any permanent harvesting regimes are set. As part of a sustainability assessment of harvesting macroalgae from the Baltic Sea, we assessed the biomass potential for eutrophication mitigation in a case study area.

The chosen case study area of Trelleborg is a transition zone between the brackish Baltic Sea and the saline Kattegat area. Trelleborg is known for mass occurrences of red filamentous algae during summer. The biomass potential of the summer stock of red filamentous algae was assessed in the case study area with a quantification method based on regional monitoring data as well as photic zone and depth distribution.

The summer algae's nitrogen pool equals about 50 % of the annual nitrogen freshwater runoff from the area, which is a huge amount of nitrogen. If 10-30 % of the summer stocks of red filamentous macroalgae were to be harvested, nitrogen reduction would be in the magnitude of 50-150 tonnes of nitrogen annually. As a secondary benefit of the process, the retrieved biomass has a methane potential that can supply heat to about 170-300 houses annually. The nutrient reduction potential of macroalgae harvesting is considerable and future studies will asses the nutrient reduction with other methods, such as wetland restoration with a comparative cost-benefit approach.

Red macroalgae in Trelleborg. Photo: F. Gröndahl

Nitrogen reduction potential of macroalgae harvesting in Trelleborg

Quantified	Biomass (tonnes)	N (tonnes)
Total red filamentous algae summer stock[1]	19 000	475
Harvesting potential of total stock[2]	1 900 - 5 700	50 - 150
Freshwater runoff**	-	1 000

1. Risén, E. et al. (Manuscript in preparation). *Assessing biomass potential of two red macroalgae species in the Baltic Sea.*
2. Wall, I. 2006. *Hav och åar i Trelleborgs kommun*, Miljöförvaltningen Trelleborgs kommun, Trelleborg, Sweden.

EVALUATION OF MARINE BIOMASS POTENTIALLY USABLE FOR ENERGY PRODUCTION ALONG THE EAST FRISIAN COAST, GERMANY

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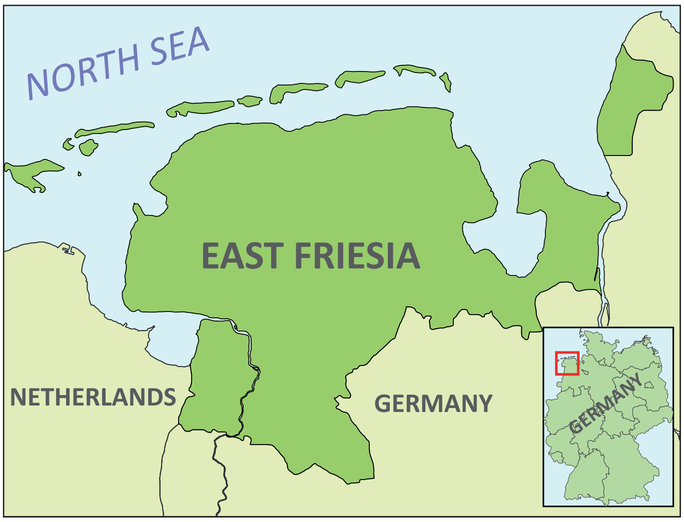
Algae and other marine biomass washed ashore belong to potential sources of bioenergy in coastal areas. However, many open questions and unresolved problems remain in the goal to reach a sustainable and profitable supply chain from raw material to energy production, one of the crucial ones being the evaluation of available biomass. This contribution aims to give an overview of marine biomass potentially usable for bioenergy production along the East Friesian coast of the North Sea (Wadden Sea), as a part of an ongoing study.



Map of the East Frisian coast of the German North Sea (Wadden Sea). Image: Onno/Wikipedia (adapted)

The Wadden Sea area belongs to the most eutrophied seas along the European coast, i.e. there is a surplus of nutrients for growth of algae. Removal of algae biomass with its built-in nutrients would therefore have rather positive consequences for the ecosystem. However, proliferation of both microalgae and macroalgae in the Wadden Sea is strongly limited by light due to the high turbidity of the water column. The growth of macroalgae is also restrained by a lack of hard substrates compared to other coastal areas such as those of the Baltic Sea.

In addition to the constrains posed by natural conditions, large areas of the Wadden Sea coast are excluded from any exploitation by nature protection

Left: Flotsam in front of a dike with the salt meadows in the background. Photo: G. Asche. Right: Flotsam on a dike. Photo: R. Ptacnikova authorities. The Waden Sea coastal area is a unique ecosystem with high biodiversity, providing a feeding base for hundreds of thousands of waterfoul during migration and wintering periods.

Nevertheless, in spite of the above-named limitations, it seems that in the Wadden Sea coastal areas there might be sources of marine biomass potentially usable for bioenergy production, namely flotsam landing on man-made dikes and marine biomass washed ashore on public beaches.

The whole coastal area of East Frisia has been protected from storm floods by a system of dikes built over centuries and reaching now a height of 9-10 m. To slow down the power of incoming waves, there have typically been buffer areas of salty marshlands and salty meadows left between the main dikes and the shoreline, stretching several tens of meters in width.

During storm periods in winter time the plant biomass in these buffer zones gets withered, is torn away by waves and washes up on the dikes. This biomass, called flotsam, must be removed from the dikes in order to maintain the quality of the grass growth on the dikes and therefore the stability of the dikes. The flotsam has been used as fodder or fertilizer. However, given the larger amounts that sometimes land on the dikes under extreme weather conditions, there appears to be a potential for turning flotsam into biogas.

There is, however, a drawback in the flotsam being an irregular and quantitatively hard to predict supply. This source might be supplemented with marine biomass (mainly green and brown algae) collected on several kilometers of public beaches in the area during the summer period. So far, this biomass has been mostly dumped in landfills. Although the amount of algae is typically much smaller than for example in the Baltic Sea coastline, it is potentially still worth considering. The amounts of biomass available in the area have yet to be quantified, the study will then follow with technological and economical assessments.

Algae collected on a public beach. Photo: W. Schuster

GROWING ALGAE IN SCANDINAVIA: UTOPIA OR OPPORTUNITY?

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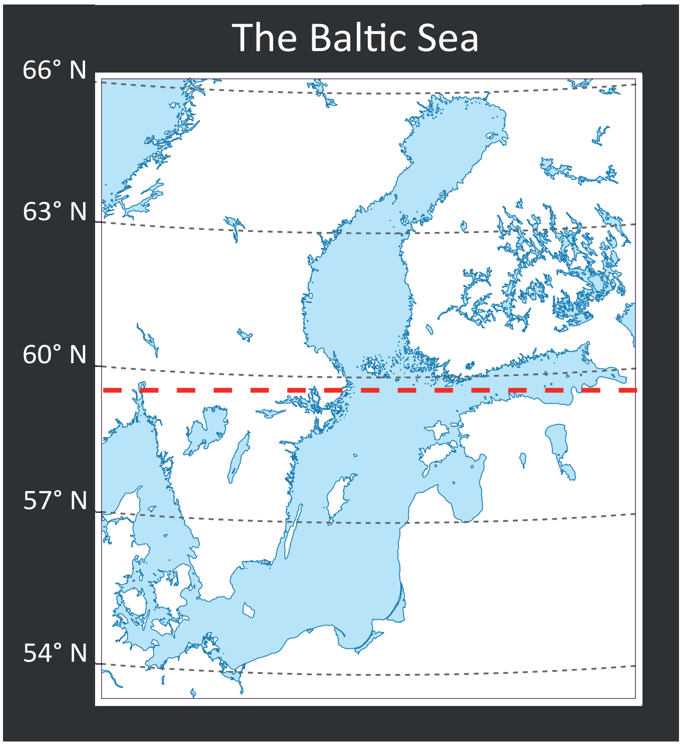
SCHOOL OF NATURAL SCIENCES, LINNÆUS UNIVERSITY, SWEDEN

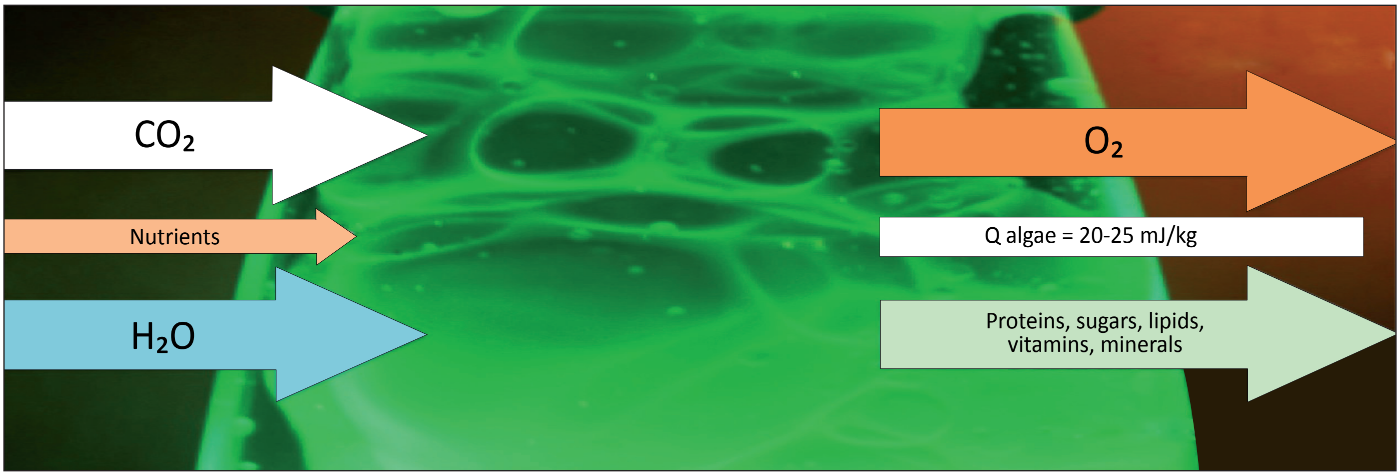
The use of fossil fuels is now accepted as unsustainable due to their depleting supply and the accumulation of greenhouse gases in the environment. Biofuels produced from crop seeds are controversial, as they compete with food for arable land and freshwater. Production of algal biofuels appears to be an alternative.

Algae, like plants, need sunlight, carbon dioxide and water to produce biomass. Algae can be grown rapidly in seawater, are carbon neutral and do not compete with food-production. While the concept of using algae as a renewable and sustainable energy source has been explored for the last few decades, there is no current commercially-viable production system. Researchers and entrepreneurs are interested in algae as a feedstock considering its potential for high productivity in a biorefinery, the non-competition with the global food supply, the potential for use of non-arable land and various water sources (brackish, saline, wastewater), the capture of greenhouse gas released into the atmosphere, the potential for bioremediation of wastewater and the production of both algal biofuels and valuable co-products.



The red line shows the theoretical upper limit for algal cultivation areas. Image: NordNordWest/Wikipedia (adapted)

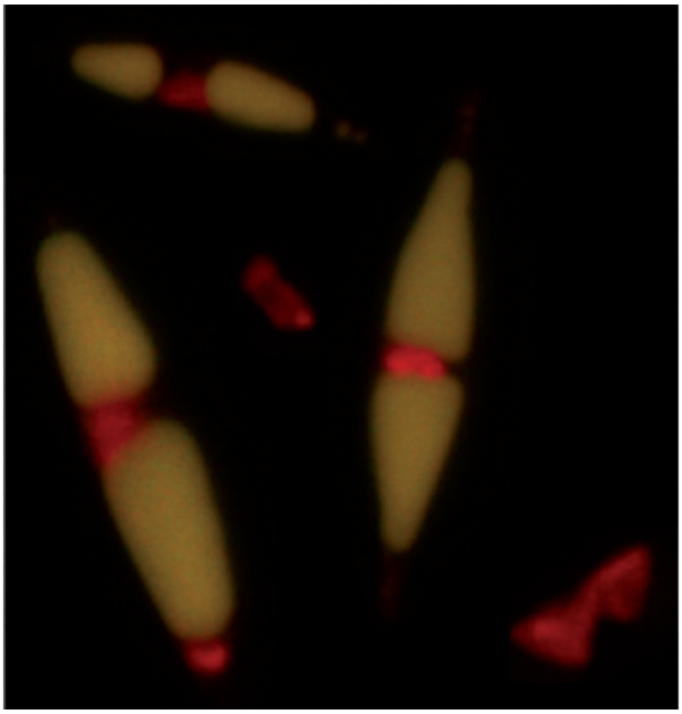
Traditionally, large-scale algal production units are set to be most productive in sub-tropical and warm temperate areas. The idea that algae can grow well in cold temperate regions is often dismissed due to the limitations of the production period (light and



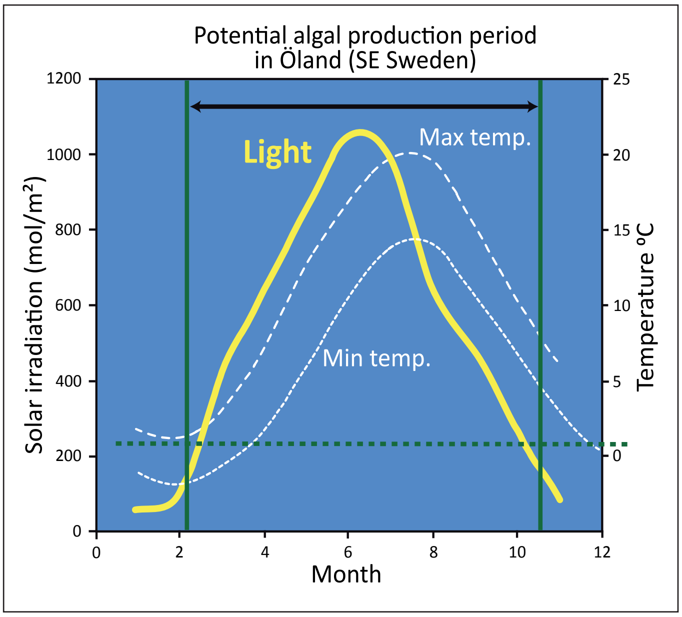
Algae use carbon dioxide and transform it to organic biomass (carbohydrates, lipids, proteins) that can be converted into energy: hydrogen, biodiesel, alcohol, methane, raw biomass and high value commercial products. Photo: C. Legrand temperature). The ordinary range for algal cultivation is 55°S - 55°N. However, in the South Baltic region (< 57 - 60 °N) yearly photosynthetic active radiation (PAR) land temperature conditions are similar to the ones in the center of Germany.

Ice cover, another limiting factor to large scale algal production, is highly variable between years and seldom extends to the Baltic proper. On the other hand, ice can be an advantage if biofuels are the focus of the algal feedstock: many "ice algae" or cold temperate algae are naturally very rich in lipids.

Numerous claims have been made on the biomass productivity based on small-scale experiments and high value algal products in warm temperate regions. It is necessary to know whether these claims can be extrapolated to large-scale production and to other regions. Due to the seasonal dynamics in Scandinavia, the production season is limited by both light and temperature for purely photosynthetic organisms. Realistically, algal production in terms of biomass will not compete with that of warmer regions.



Many "ice algae" or cold temperate algae are rich in lipids. Algal cells stained with Nile Red and observed in blue light. Oils appear yellowish. Photo: M. Olofsson



Yearly sunlight and water temperature in the South of Öland (Southeast Sweden). Data source: Swedish and Norwegian Meteorological Institutes.

If one wants to grow algae in the South Baltic region to capture CO_2 produced by industry or local farms, using nutrients from municipal wastewater, for producing feedstock for various energy sources or high value co-products, or a combination of the latter, there are major aspects to bear in mind. The Baltic Sea is an enclosed sea and a very sensitive brackish sea that has been exposed to massive eutrophication, overfishing, pollution and invasive species. It would then be preferable to work with local isolates, not imported or genetically modified ones. In general, brackish phytoplankton assemblages are very resilient and can withstand extreme variations in temperature, salinity, pH, and CO_2 concentrations over short periods of time. Therefore the effect of diversity on productivity should also be considered.

Is it utopia to grow algae in Scandinavia? No, it is not, as long as we think about both the ecological and economical consequences of using algae in integrated processes.

LARGE-SCALE CULTIVATION OF MICROALGAE IN THE BALTIC SEA REGION

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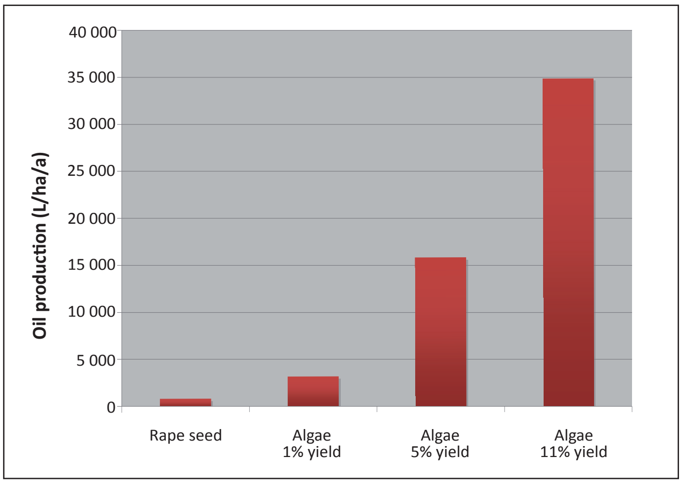
Production of biofuels and other low-cost products from microalgae is not yet economically profitable. Key challenges are related to optimising algal growth, harvesting and processing the biomass and scaling up the production. Some of the difficulties to be solved include a regional component related to climate, available infrastructures or socioeconomic conditions. In this study, we examine regional aspects of large-scale cultivation of microalgae, especially when relevant to the Baltic Sea region (BSR).

Light

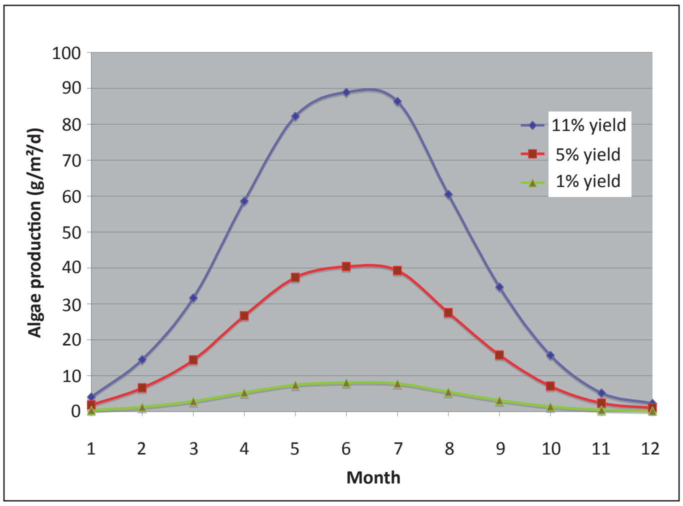
At first glance, areas in high latitudes with long, dark and cold winter seasons, like the BSR, seem rather suboptimal for large-scale microalgae outdoor cultivation. The annual average of sun energy for the BSR under clear sky conditions is 2 500-3 000 kWh m^{-2} d^{-1}, which is approximately 50-66 % of the energy available in the middle of Spain, or 40-50 % of the energy available in the middle of the Sahara desert. Of course, the seasonal variations are much larger in the BSR than in lower latitudes and it is obvious that the dark winter season is not suitable for growing any photosynthetic organism.

Theoretical maximum photosynthetic efficiency, that is, the conversion of sunlight energy to the chemical energy of photosynthetic end products, is around 11 %. In real production systems the photosynthetic efficiency is much smaller, 1-5 % at maximum, as not all light energy absorbed is driving photosynthesis (due to photosaturation and photoinhibition) and as part of the energy is lost during respiration.

The projected areal production of microalgae at latitude 60° N, in the middle of the BSR, ranges from 8-40 g of dry weight m^{-2} d^{-1} during the summer months, using photosynthetic efficiencies of 1 % and 5 %, respectively. Based on literature, comparable productivity levels have been obtained under similar irradiance conditions using closed photobioreactors but not in outdoor ponds. When integrated for the whole season and using caloric values of algae of 25 kJ g^{-1}, an oil content of algae of 30 %, and 80 % conversion from algae oil to biodiesel, the projected annual microalgae oil production at latitude 60° N ranges from 3.2-16 tn of oil per ha. This is considerably larger than the oil yield obtained with rape seed (0.8-1 tn oil per ha). The seasonality in microalgae production in the BSR based on irradiance levels is pronounced and the question remains what fraction of the year the production may be economically feasible.



Potential microalgae oil production at latitude 60° N, in the middle of the BSR, compared to oil production using rape seed. Photosynthetic efficiencies of 1 %, 5 % and 11 % were used in calculations. Algae oil content is assumed to be 30 % of dry weight.



Seasonal algae production at latitude 60°N, in the middle of the BSR. Photosynthetic efficiencies of 1 %, 5 % and 11 % were used in calculations.

Temperature

Temperature is considered another limiting factor for microalgae growth at high latitudes. Some algae species are, however, adapted to temperatures close to the freezing point. For example in the Baltic Sea, the spring algae bloom, with high growth rates, starts when temperature is slightly above 0° C. Maximum growth rates of Baltic Sea cold water species in our culture collections vary from 0.5-0.8 d^{-1}.

The lipid quality affects downstream processing of algae oil and also the properties of oil. Generally, for biodiesel production fatty acids with short chain length and high saturation are preferred. Based on the results from higher plants and macroalgae, it has been expected that saturated fatty acids are not very abundant in cold water microalgae species. Our results with Baltic Sea species do not support this. During the exponential growth phase the examined cold water species contained proportionally much more saturated fatty acids than the warm water species, making them good candidates for biodiesel production.

Nutrient sources for cultivation of microalgae

Fertilizers and CO_2 are major costs when growing phytoplankton at large scale. Using waste streams as a nutrient source for cultivation may thus reduce both the production cost and also provide additional societal services by reducing nutrient fluxes to the sea.

Our tests with various microalgae species show that municipal wastewaters in Finland contain required nutrients for algae growth and they do not contain growth-inhibiting agents. Calculations based on microalgae nutrient stoichiometry in the stationary growth phase (4 % and 0.5 % nitrogen and phosphorus of dry weight, respectively), quantity of wastewater and the assumption that all nutrients in the municipal wastewaters may be converted to algae biomass, results in oil production of approximately 0.1 L oil d^{-1} $person^{-1}$. To have this value in a larger perspective of energy consumption, in Finland 2 % of all liquid transport fuel need may be produced from algae if all nutrients in municipal wastewaters were used in algae cultivation. Evidently nutrient recycling is crucial for large-scale, cost-efficient production of microalgae biomass.

Before algal biofuels can reach the market, years of research and development work must still be carried out, including setting up pilot plants which allow simulation of various processes at industrial scales. The Baltic Sea region has the required infrastructure, motivated and trained scientists and hopefully also the political and economical will to contribute to this development. The location of the future large-scale algae production plants that may have globally significant energy production is not yet on the agenda.

SIMRIS ALG: THE ESTABLISHMENT OF A NEW INDUSTRY IN SWEDEN

FREDRIKA GULLFOT

SIMRIS ALG AB, SWEDEN

Simris Alg AB is the second commercial microalgae operation in Sweden. We are based in Simrishamn, a small coastal town in southeastern Scania, with excellent sun conditions and a well-developed infrastructure for agribusiness and the production of food and energy. Our business model relies on a biorefinery concept and the generation of value throughout the production chain. We are currently in the process of preparing our production plant to be established in 2012. Meanwhile, we are engaged in several greentech projects in which we use microalgae to convert CO_2 emissions and wastewater streams from industrial and agricultural activities into economic value.

Despite Swedish microalgae pioneer BioReal's global success at establishing the antioxidant astaxanthin from *Haematococcus pluvialis* as the business case par excellence for valuable compounds from microalgae, there has been a peculiar lack of activity in the field in the country. Recent developments, in particular concerning closed-system photobioreactors, novel business models and growing markets for high-value compounds, for example in the functional foods area, suggest that farming and processing of microalgae has great potential to establish itself as a thriving new industry and innovative agribusiness in southern Sweden.

Proprietary algal strains in Simris Alg's strain library. Photo: Simris Alg AB

BIOMASS PRODUCTION TO REMOVE NUTRIENT FROM WASTEWATER AND CO_2 FROM A POWER PLANT

FRANCESCO GENTILI

SWEDISH UNIVERSITY OF AGRICULTURE SCIENCES

The goal of the project is to produce biomass and biofuels while reducing CO_2 in an economical and environmentally sustainable way. To achieve this goal we are using microalgae growing on wastewater and flue gases as substrates. We are testing a continuous system for microalgae production using wastewaters, thus recycling important nutrients that represent a hazard for the environment but a nutrient source for algae. The CO_2 from flue gases is used for algal growth and emission reductions.

A synergistic collaboration between researchers and industries is crucial to the realisation of this project. Such a collaboration was put in place a few years ago between the Department of Wildlife, Fish and Environmental Studies at the Swedish University of Agriculture Sciences (SLU Umeå), Umeå Energi AB (a power plant converting municipal wastes into energy) and Umeva AB (a wastewater treatment plant). Recently RagnSells, a company working on waste recycling, has also joined the project.

We have a straightforward approach to achieving our goals: tests at the laboratory level for a quick answer regarding which algae can grow and thrive on our substrates. These tests allow us to identify the most suitable and productive algae strains. Then, based on the laboratory results, we test the selected algae strains in the prototype at the power plant station, where algae grow on wastewater and flue gases to see how they perform in real conditions. Recently we have been focusing on wild algae that performed well in our growing conditions.

The greenhouse built on the roof of the Umeå Energi power plant station located 10 km outside Umeå (northern Sweden). The photobioreactor can be seen inside the greenhouse. Photo: F. Gentili

Dry algae biomass produced in the photobioreactor. The algae were grown on urban wastewater and flue gases. Photo: F. Gentili

In the laboratory, we have been able to reach an algae dry matter production of $1\,g\,l^{-1}\,d^{-1}$, when our two microalgae strains were grown on wastewater with continuous low light ($100\,\mu E\,m^{-2}\,s^{-1}$) intensity. In the bioreactor prototype at the power plant station the biomass production was lower than expected. However we are using only sunlight without addition of artificial light. Furthermore the volume treated is much larger (650 l).

At this point in time, the main focal points of the project are algae productivity and the harvesting system. To reach our goals we are building a pilot plant to enlarge algae production under natural growing conditions. The project has been financed by the Swedish Energy Agency.

BIOENERGY BY CLEANING OF WATER: A GREEN AND SUSTAINABLE APPROACH

HERMAN CARR & TOMMY LANDBERG

CLEAR WATER ENERGY NORDIC AB, SWEDEN

Man-made impacts on aquatic environments are a serious concern around the world. Most solutions, when available, are very costly and produce other environmental problems such as waste and high energy consumption. The new company Clear Water Energy Nordic AB (CWENordic) presents a sustainable solution based on academic research: macroalgae is used to clean a wide range of contaminated waters while simultaneously biomass for bioenergy is produced. This is the vision of CWENordic: a useful green technology applicable in most parts of the world for both cleaning water and producing bioenergy.

At its very start CWENordic received support from Teknikhöjden/Stockholm CleanTech Park and funding from Innovationsbron Stockholm AB. As a result, the technology was able to emerge from the laboratory and be tested, both technically and from a business point of view. The water cleaning market is huge and water cleaning consumes both energy and money. Vast sums of money are also invested in trying to clean the Baltic Sea, which is heavily affected by various contaminants.

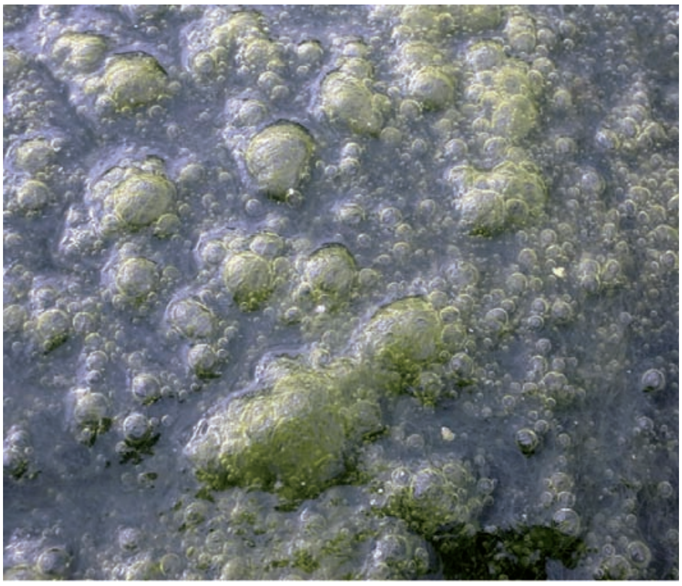
The initial tests in real conditions were performed at a sewage treatment plant at Fors, south of Stockholm, with very promising results. Not long after the first technical tests, the company was contacted by Trelleborg Municipality. CWENordic‘s idea is to catch the excess nutrients and turn them into something valuable, which fits well with the Municipality of Trelleborg‘s vision and environmental action plans and for whom this technology could be very useful. CWENordic is now testing the algae system to clean agricultural runoff water in Trelleborg municipality.

The CWE water cleaning project is financed by Trelleborg and is running on private land. Photo: CWENordic

The testing and development of the system to make it functional in a more complex environment than that of a sewage treatment plant gives very valuable results and experiences. As a result, CWENordic is currently developing the next generation of the water cleaning system, which will be easy to install on large surfaces and be situated close to the surface ,resulting in higher algal yield.

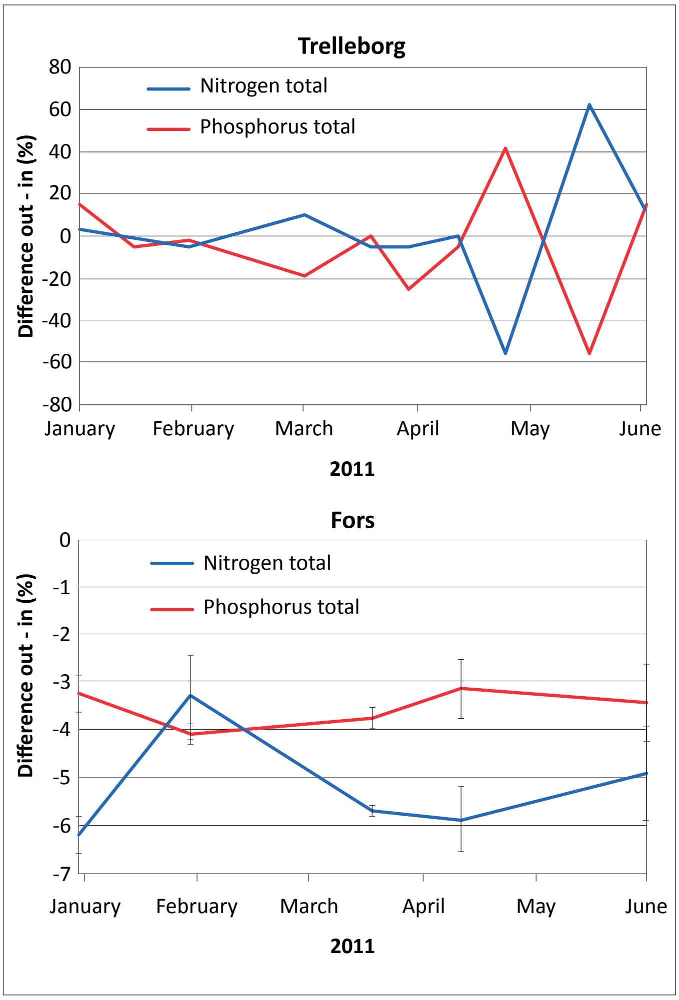
The growth of algae is very good in the sewage treatment plant site in Fors, Sweden. Photo: CWENordic

Results from the sewage treatment plant show that the system indeed reduces contaminants. However, the results from Trelleborg show that the variations are very large.



Algae produce a lot of oxygen. Photo: CWENordic

Furthermore, results show a very good production of oxygen. This is important because the highest cost in a sewage treatment plant is the aeration of the water. In the algae system the energy comes from the sun and the oxygen comes for free!



Differences in the concentrations of total nitrogen and phosphorous between in and out of CWENordics algae-based water cleaning systems at two locations (Trelleborg and Fors).

In short, the use of algae by means of various technologies shows a great potential. For cleaning water and producing biomass for bioenergy the algae technology has so far showed very promising results. Although some development is needed the technology is ready for large-scale water treatments for some types of contaminated waters.

GREEN MICROALGA SCENEDESMUS SP. GROWTH IN LIVESTOCK MANURE TREATMENT EFFLUENT: DEVELOPMENT OF A POTENTIAL BIOREFINERY

CLAUDIO LEDDA, ANDREA SCHIEVANO & FABRIZIO ADANI

GRUPPO RICICLA, DIPARTIMENTO PRODUZIONE VEGETALE, UNIVERSITÀ DEGLI STUDI DI MILANO, ITALY

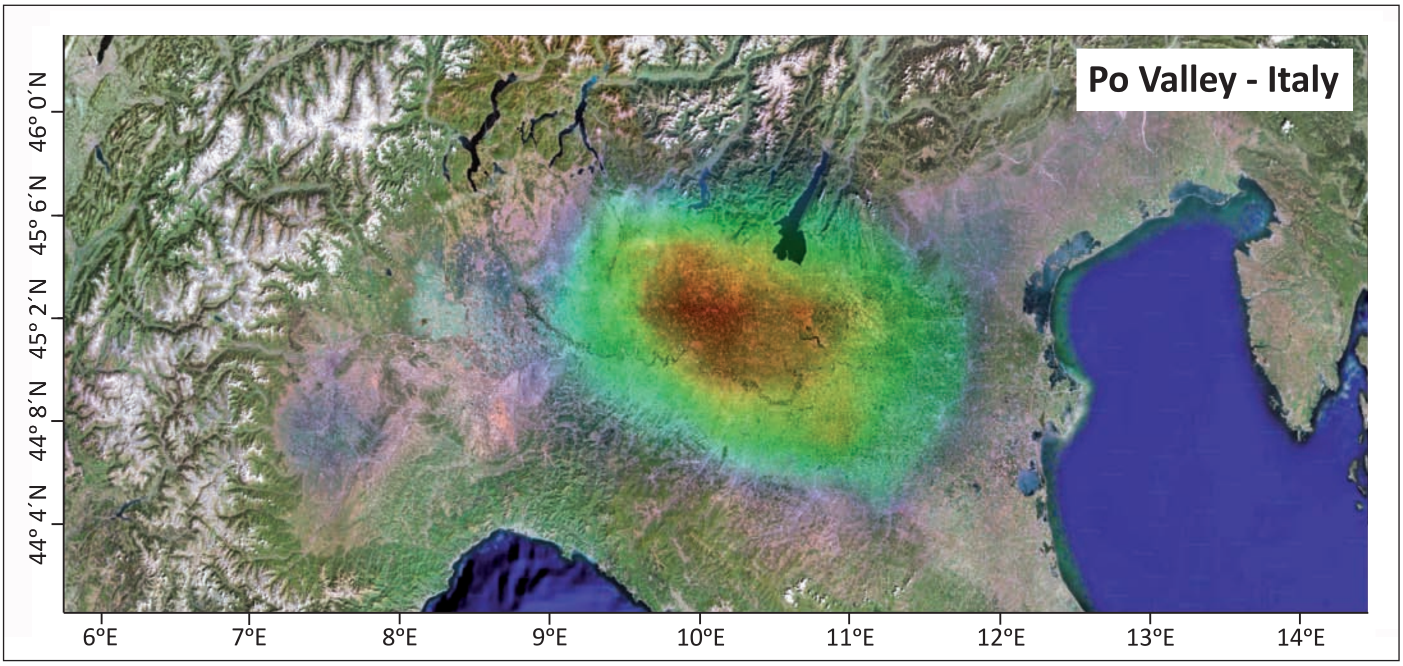
Over the past decades human activities have deeply changed our environment, as in the case of the livestock production chain, responsible for huge volumes of wastewaters characterized by a high organic and mineral load.

As an example, a region like the Po Valley in northern Italy, which encompasses 63 % and 83 % of the total Italian cattle and swine populations respectively (L'Agricoltura Italiana Conta, 2009), is not capable of naturally managing livestock sewage in a sustainable way.

As a result, the EU Nitrates Directive (91/676/CEE) and corresponding national and local rules have been put in place to regulate sewage disposal and its agronomical use, limiting nitrogen and other nutrient loads per hectare of cultivated land.

To deal with this issue, the University of Milan (Gruppo Ricicla) in collaboration with Regione Lombardia and S.A.T.A Aral (Technical Service for Livestock Farming) is evaluating a new technology, called N-Free®, characterised by a series of chemical-physical treatments (ultra-centrifugation with decanter, ultra-filtration in 40 kDa membranes, reverse osmosis and cold ammonia stripping). This process allows depuration of manure, with the ability to discharge the final effluent in surface water bodies and the production of organo-mineral concentrates and ammonium sulfate (30 %), usable as valuable organo-mineral fertilizers (OMF).

The liquid permeate from the ultra-filtration step is proposed here as a potential growth substrate for the microalga *Scenedesmus* sp., in order to increase

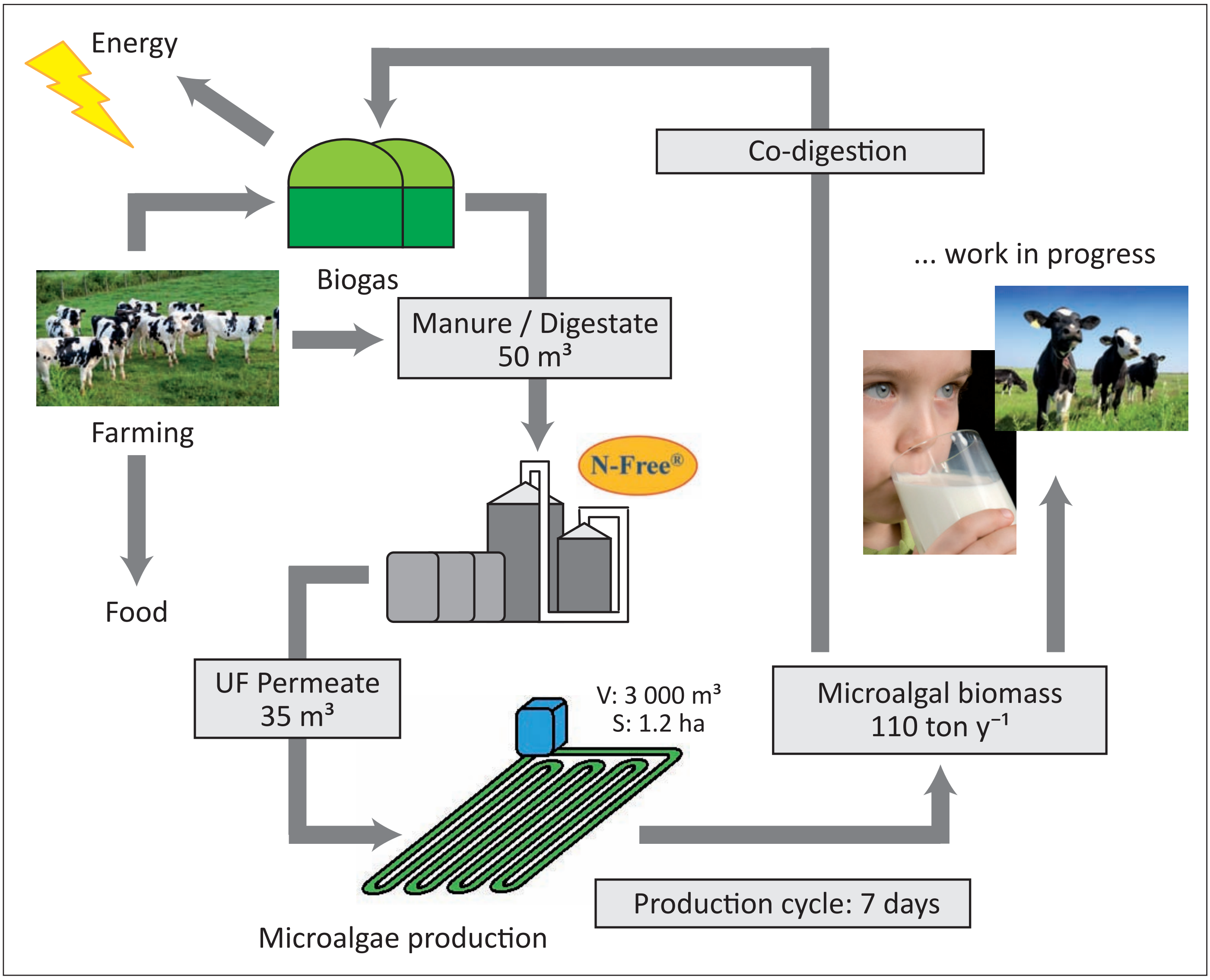


Satellite detection of atmospheric free ammonia (NH_3) due to ground volatilization (from Clarisse, L. et al. 2009. *Global ammonia distribution derived from infrared satellite observations*, Nature Geoscience 2, 479-483.)

the added value of the whole process. Even showing lack of some nutrients, the tested medium gave good microalgal growth performances ($\mu = 0.09\ d^{-1}$, Pd - Daily Volumetric Productivity = $124\ mg\ L^{-1}\ d^{-1}$).

Taking into account that the basic N-free® treatment unit ($50\ m^3\ d^{-1}$ of manure) produces $35\ m^3\ d^{-1}$ of ultra-filtration liquid permeate, we hypothesized a microalgal cultivation unit with a land surface use of nearly 1.2 hectares and a culture volume of $3\,000\ m^3$. The microalgal plant would produce approximately 110 ton y^{-1} of dry matter available for livestock feed, nutraceutics or the energy market (co-digestion for biogas production).

Further studies will better evaluate optimization, economic feasibility and returns of the culture system.



Example of a microalgae / N-Free® system-based biorefinery.

GUIDELINES FOR ENVIRONMENTAL ASSESSMENT OF NEW USES OF BALTIC SEA MARINE RESOURCES: THE MACROALGAE CASE STUDY

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[1] INFORMUS GMBH, GERMANY
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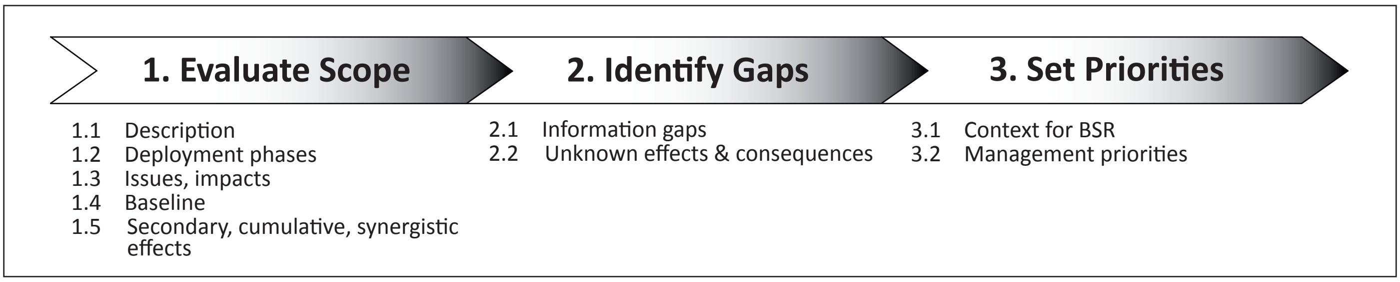
The Baltic Sea is one of the world's largest semi-enclosed bodies of brackish water. It is almost entirely land-locked with very limited water exchange. Its ecosystem is fragile and particularly sensitive to human activities. It faces pressing environmental issues including eutrophication, ocean acidification, introduction of alien species and input of organic pollutants. Eutrophication is a major problem in the region arising from excess nutrient inputs from primarily land-based sources. The HELCOM vision for the Baltic Sea is "a healthy […] environment, with diverse biological components functioning in balance, resulting in a good ecological status and supporting a wide range of sustainable human economic and social activities". In support of the EU Marine Strategy Framework Directive to establish Good Environmental Status by 2021, the HELCOM Baltic Sea Action Plan has set a strategic goal to have a "Baltic Sea unaffected by eutrophication". A set of ecological objectives corresponding to good ecological status are regularly assessed to evaluate progress towards achieving HELCOM's strategic goal.

The SUBMARINER project explores opportunities in the Baltic Sea region for sustainable new uses of marine resources, which have the potential to significantly contribute to solving some of its environmental problems. An important task in developing new uses of marine resources is to identify potential environmental issues that may arise as a result. In this context, a SUBMARINER general framework for assessing positive and negative impacts that a single and/or combined use of marine resources may have on the Baltic Sea natural environment is being developed.

The framework draws on existing good practice, conventions, policy and legal instruments (e.g. HELCOM, EU Marine Strategy Framework Directive and the EU Environmental Impact Assessment and Strategic Environmental Assessment Directives). Its objective is to offer a systematic approach to evaluate the scope of environmental issues that may arise as a result of the new use, highlight gaps in information and establish priorities for research to improve future environmental management of the resource. The goal is to have a general framework that can be applied to a diverse set of marine resources and potential new uses. The approach consists of three phases, which pull together different pieces of information related to the new use of the marine resource.



General environmental assessment framework

The notion of using macroalgae to mitigate against excess nutrient inputs and eutrophication in the Baltic Sea region is attractive. Many coastal areas suffer from chronic blooms of macroalgae and endure long periods when layers of algae dominate the intertidal zone and wash up along coastlines. The smell is unpleasant and negotiating a path to swim can be a nuisance. These alone constitute good reasons to consider removing excess biomass, given their negative impact on marine tourism. And there is an added bonus in the application of harvested biomass to produce biofuels. SUBMARINER has begun to apply the general environmental assessment framework to the exploitation of macroalgae for mitigation against excess nutrients in the Baltic Sea region and to produce biofuel. A preliminary evaluation of environmental issues as applied to natural harvest of living and beach-cast macroalgae is given as an example below. (Macroalgae cultivation, independently or in combination with fish aquaculture or other offshore infrastructures presents a different suite of environmental issues and calls for a separate evaluation).

The next steps in this process are to identify the gaps and unknowns arising from the evaluation of environmental issues. It is important to put the issues into context and understand the local environment. Finally, a set of management priorities for the new use of the resource can be established.

Intertidal and beachcast macroalgae harvest example

1. Evaluate Scope
1.1. Description: provide clear description of the marine resource and its intended use(s). Include specifics concerning the species (if living resource), its availability, location in the Baltic Sea region and its proposed use(s).
Marine resource: Bladder wrack (e.g. *Fucus vesiculosus*) and Red seaweed (e.g. *Furcellaria lumbricalis*) **Proposed use:** Mitigation against excess nutrient inputs, eutrophication, improvement of water clarity and production of biofuel
1.2. Deployment phases: identify the different operational phases involved in the exploitation of the resource.
Natural harvest of living and beach-cast macroalgae: 1) harvest, 2) transport and 3) processing
1.3. Issues and impacts: identify the different potential environmental issues and impacts associated with each of the operational phases.
Sustainability: Will harvest impact future recruitment and supply of biomass? **Biodiversity:** Is it an important habitat for marine and coastal species, food source, shelter, spawning and nursery grounds? What is the impact on the overall balance of the marine ecosystem? Dependent trophic food webs? **Coastal processes:** Are there any important coastal processes to be considered, e.g. erosion, shore currents, wave energy, sand dunes? What is the interplay between sediment dynamics and kelp beds? **Harvesting method:** Manual or mechanical? Implications for marine noise, beach disturbance, water clarity, turbidity and seafloor integrity? **Climate change:** Impact of ocean acidification, sea level rise and temperature rise? Geographical distribution of species? Are they living at the edge of their comfort zone? How are they likely to respond to changes in climate?
1.4. Baseline: a description of what is known about the current status of the environment and all aspects of the environment that may be affected by the new use of the marine resource. If nothing is known, this is also a baseline.
Identifying a baseline provides a reference against which all future monitoring can be assessed.
1.5. Secondary, cumulative, synergistic effects: make a preliminary assessment of any secondary, cumulative or synergistic effects (e.g. is a problem being moved from one location to another? Are any problems compounded? Is it possible to identify long-range impacts? Are there benefits which can result from synergies with other uses?)
Secondary: Does removal/recycling of excess nutrients by macroalgae harvesting from untreated sewage wastewater encourage less preventive sewage treatment measures implemented on land? **Cumulative:** What is the combined effect of several Baltic States exploiting the same resource? **Synergies:** Biofuel production …
Next Steps … 2. Identify Gaps and 3. Set Priorities

THE WAB PROJECT

ELLINOR TJERNSTRÖM & MATILDA GRADIN

CITY OF TRELLEBORG, SWEDEN

Trelleborg Municipality is Lead Beneficiary for Wetlands, Algae and Biogas (the WAB Project), a Southern Baltic Sea Eutrophication Counteract Project. It creates sustainable solutions that benefit the marine and land environment, reduces eutrophication and produces biogas.

The WAB Project is collecting coastal algae that are produced in large masses as a symptom of the eutrophication in the Baltic Sea as well as vegetation from constructed wetlands and uses them both as additional biomass for biogas production. This results in a reversal of nutrient flows from the Baltic Sea back to the farmland.

Reconstructed wetland in Källstorp, Trelleborg for macroalgae cultivation as a step for retention of nutrients. Photo: M. Gradin

WAB is a project of the South Baltic INTERREG Programme made up of 11 cooperating partners in Sweden and Poland. The core areas for testing and implementation are Trelleborg in Sweden and Sopot in Poland.

The project delivers solutions that are economically and environmentally useful. During the summer and peak tourist season, algae grow and accumulate along the coastline of Trelleborg and Sopot. This has a negative impact on the local economy and recreational values. The removal of algae from the beach will benefit both tourism and recreation. Produced biogas can be used instead of fossil fuels, thus reducing climate impacta. The residue contains nutrients which can be recycled back to agricultural land, reducing the use of commercial fertilizers.

Algae on the coast line in Trelleborg municipality. Photo: M. Gradin

The project offers an innovative holistic approach by constructing a cycle of nutrients previously unseen in the Baltic Sea. The project has the potential to spread the findings throughout the south Baltic region which would bring environmental, social and economical benefits to the area.

Launch of the pilot-scale biogas plant (August 2011), first in Sweden to run purely on algae from Trelleborg's coastline. Photo: P. Finnis

BIOFUEL PRODUCTION FROM MICROALGAE BIOMASS

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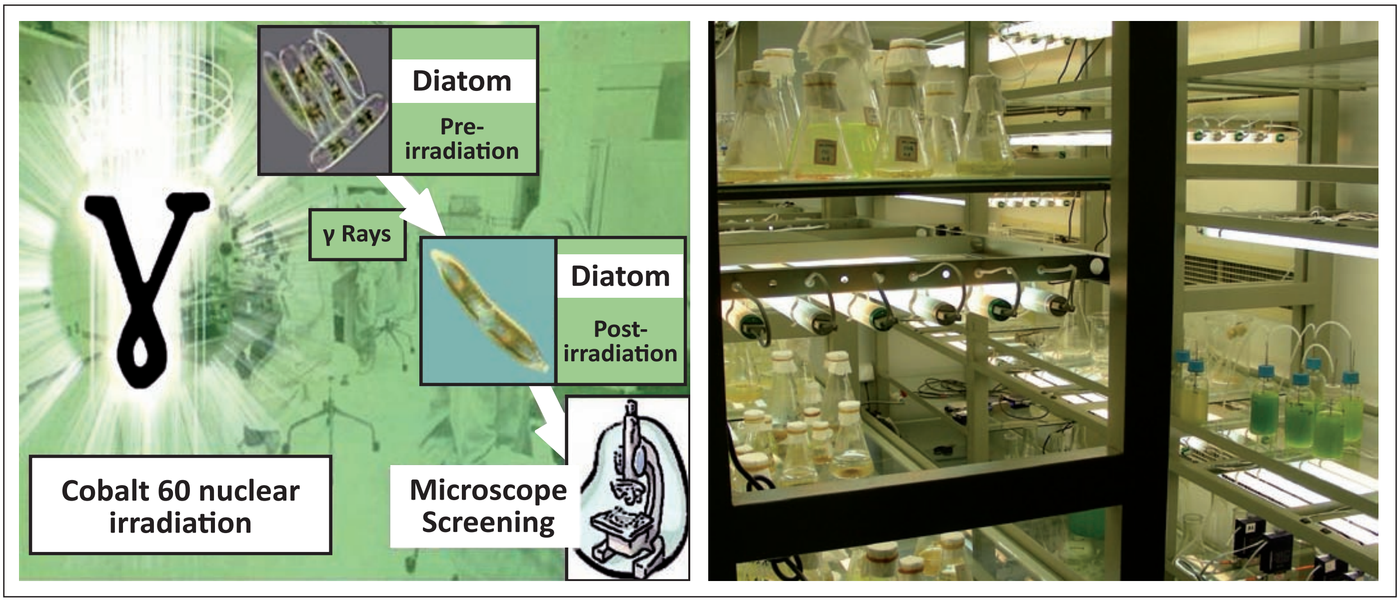
SUSTAINABLE ENERGY INSTITUTE, DEPARTMENT OF ENERGY ENGINEERING, ZHEJIANG UNIVERSITY, CHINA

Biodiesel and biogas generated from renewable sources can potentially replace fossil fuels and address issues of environmental pollution and energy security. Optimal biomass feedstocks to produce biofuels should have high productivity and energy conversion efficiency and be generated sustainably (with minimal competition for resources required for food production). Microalgae have high potential to effectively capture CO_2 emission through photosynthesis and convert it into clean fuels such as biodiesel and biogas. If 3.62 % of China's marine area (3 million km^2) were used to cultivate microalgae, 4.38 billion tons of CO_2 could be fixed in microalgae biomass and converted into 449 million tons of biodiesel annually, which could meet all petroleum consumption requirements in China in 2010.

The main research work on microalgae energy at Zhejiang University of China (ZJU) includes 1) mutation of microalgae to increase biomass yield and lipid content by nuclear irradiation and genetic modification, 2) development of efficient photosynthetic bioreactors to grow microalgae with high density and reduce CO_2 from flue gas, 3) biodiesel production from microalgae lipids through transesterification and hydrothermal reactions, 4) H_2 production from microalgae biomass by dark- and photo- heterofermentation and cogeneration of H_2 and CH_4 by two-stage anaerobic fermentation and 5) a demonstration project of biofuel production from microalgae biomass with CO_2 fixation from flue gas in a coal-fired power plant.

Microalgae mutation by nuclear irradiation

ZJU has built an advanced nuclear irradiation (Cobalt-60) system to mutate microalgae species. When green algae *Chlorella pyrenoidosa* is mutated by nuclear irradiation, the biomass yield increases by 46 % and lipid yield increase by 40 %. The biomass yield of diatom mutants *Hantzschia amphioxys* is increased by 52.7 % after nuclear irradiation. The carbohydrate content in *Spirulina* species is increased from 15 % to 32 % by nuclear irradiation.



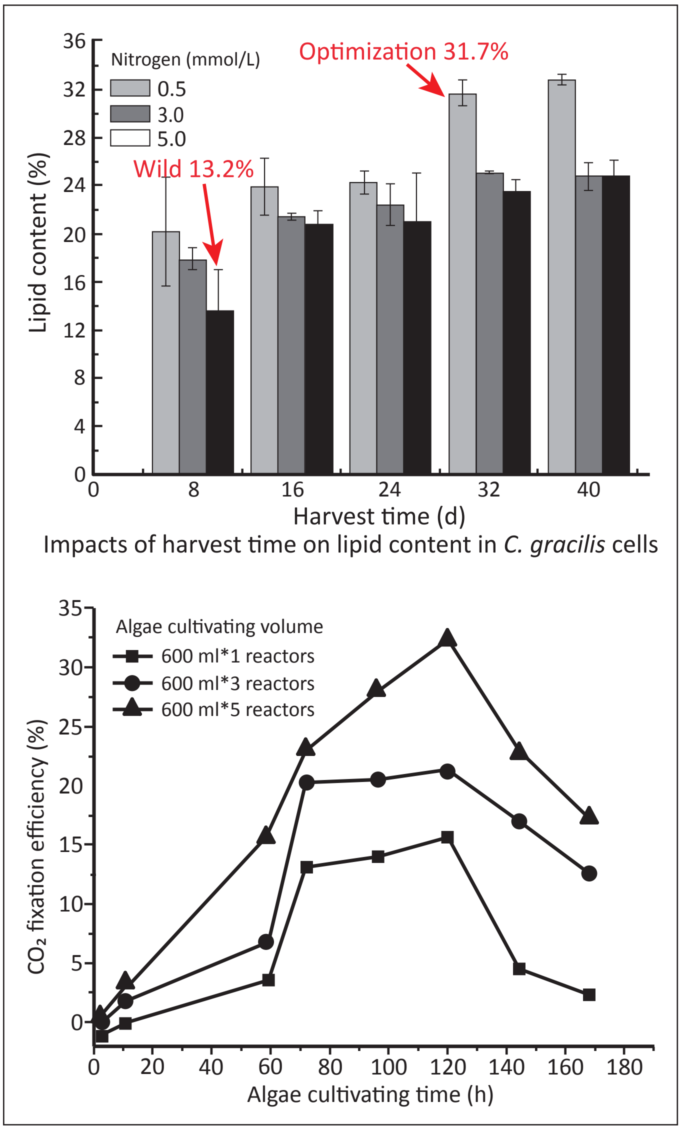
Microalgae mutation by Cobalt-60 nuclear irradiation. Photo: J. Cheng

Growth optimization of microalgae

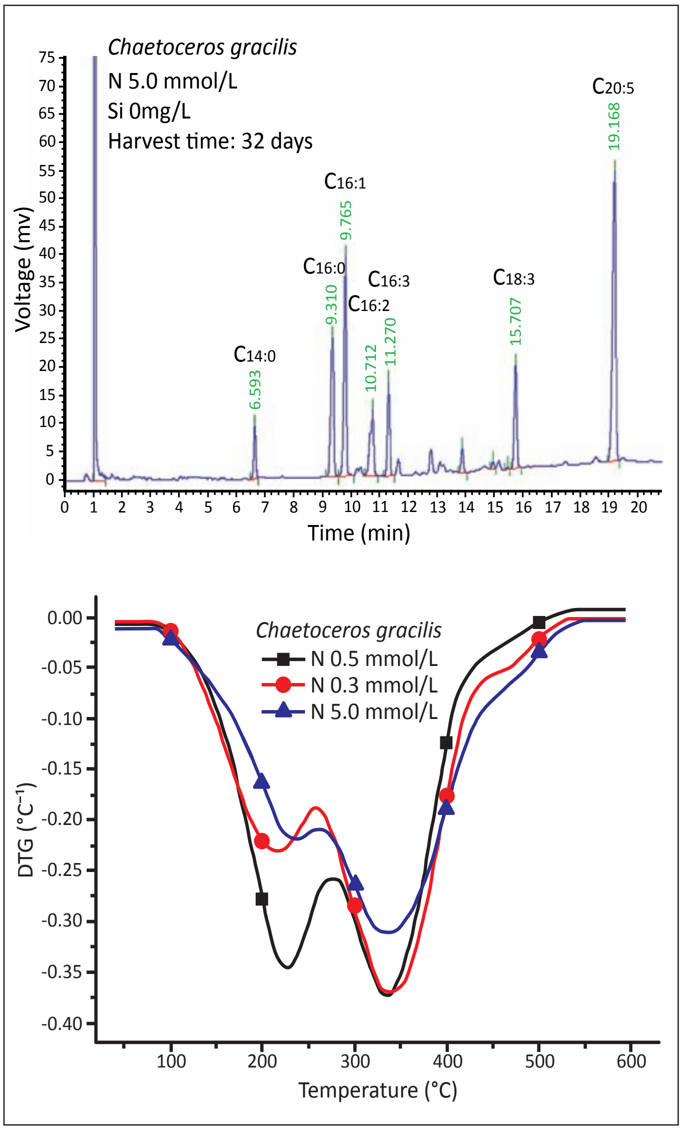
ZJU has developed membrane and spatial light-induction photosynthetic bioreactors to grow microalgae with high density and biofix high-concentration CO_2 from coal-fired flue gas. Growth regulation such as nitrogen deprivation, silicon deprivation and longer harvest time is beneficial to increase lipid content in diatom cells. The lipid content in *Chaetoceros gracilis* increases from 13.2 % to 31.7 % (2.4 times) when the silicon ($Na_2SiO_3 \cdot 9H_2O$) content decreases from 200 to 0 mg/L, nitrogen ($NaNO_3$) content decreases from 12.0 to 0.5 mmol/L, and harvest time increases from 8 to 32 days. When green algae *Chlorella pyrenoidosa* is domesticated with high concentrations (6-15 %) of CO_2, the biomass yield is increased by 51.1 % to 2.41 g/L and CO_2 fixation efficiency is increased to 32.3 %.

Biodiesel production from microalgae

ZJU has developed transesterification and hydrothermal reactors to convert wet microalgae biomass into biodiesel. The main compositions of biodiesel from diatom *Chaetoceros gracilis* are C14:0, C16:0, C16:1, C16:2, C16:3, C18:3 and C20:5. The evaporation starting temperature of biodiesel from *Chaetoceros Gracilis* is 192° C and the activation energy is 46.7 kJ/mol, which are lower than those of biodiesel from Chinese pistache seeds.



Microalgae growth optimization to increase lipid content and CO_2 fixation.



Diatom biodiesel compositions and evaporation thermodynamics.

Apparati for fermentative H_2 production from microalgae. Photos: J. Cheng

H_2 production from microalgae by fermentation

Nutrient deprivation and sodium excess are proven means for shifting cellular compositions in microalgae to obtain more carbohydrates as a precursor to increasing fermentative H_2 yield. Ultrasonic or microwave pre-treatment can effectively break cell walls of wet microalgae to release carbohydrates, which can be fermented by foreign hydrogenogens to produce H_2. The wet biomass of *Arthrospira platensis* gives an experimental H_2 yield of 96 ml H_2/g-DW by heterofermentation with foreign hydrogenogens, which is higher than that of 51.4 ml H_2/g-DW by autofermentation with cellular self-hydrogenase. By combination of dark- and photo-fermentation, the maximum hydrogen yield is greatly enhanced to 300 ml H_2/g-DW.

Demonstration project of CO_2 fixation by microalgae biomass

ZJU is cooperating with a Chinese company to build a large demonstration project to grow microalgae biomass with seawater in 100 000 m^2 of area. A part of flue gas from a 300 MW coal-fired power plant is introduced into the microalgae ponds. About 5 000 tons of CO_2 in flue gas can be fixed by microalgae and converted into biofuels.

Microalgae offer an efficient biological platform to catalyse the production of valuable biofuels from sunlight, water and carbon dioxide on non-arable land. Biofuels from aquatic phototrophs offer to address the need for transportation fuels while minimising the impact on food supplies.

BIOREFINERY OF MICROALGAE

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WAGENINGEN UNIVERSITY AND RESEARCH CENTRE, THE NETHERLANDS

Microalgae are considered one of the most promising feedstocks for sustainable production of commodities such as food, feed, chemicals, materials and biofuels. Microalgae do not need to be grown in agricultural areas, surface areas not suitable for agriculture can be used as well. They can be grown on seawater in addition to freshwater. They can be grown on residual nutrients. They also have a high areal productivity and via biorefinery the algal biomass can be fractionated into valuable products such as proteins, lipids and carbohydrates.

The technology for production is still immature, but if developed it is expected that biomass can be produced at a commercial scale for a cost price less than 0,68 €/kg of dry biomass[1]. If the different biomass components are collected the total value for commodities in algal biomass is higher than 1.65 €/kg of dry biomass[2].

For the development of an economical business model microalgal biomass should be refined into its different components such as lipids, proteins and carbohydrates. The different components should keep their structural characteristics and therefore biorefinery should be mild. In addition, the technology should be scalable and preferably used as a continuous process to handle large amounts of biomass.

To make microalgae really interesting as a source of biofuels the cost price for production needs to be reduced and the scale of production needs to be increased significantly. Technically this will be feasible. However, the development to a commercial process will at least take 10 years[3].

Our research program on the production of microalgae is well developed, both at laboratory scale and at pilot scale. AlgaePARC is a pilot facility with which we intend to bridge the gap between basic research and demonstration projects. In AlgaePARC we will compare state-of-the-art technologies and develop new reactor concepts and production strategies to achieve lower production costs and energy requirements as well as to gain knowledge for the design and process control of large-scale microalgae facilities.

To date, an optimal photobioreactor design for algal bulk products has not been available. Nevertheless, in AlgaePARC pilot units are presently being developed based on the available state-of-the-art technology in order to obtain direct practical experience. The pilot photobioreactors chosen for AlgaePARC reflect the present development of several reactor concepts used by different research groups and companies and will enable a rigorous comparison between systems, the selection and ultimately the development of an improved system. AlgaePARC initially comprises four large (24 m^2) and three small (2.4 m^2) outdoor pilots: a raceway pond, two horizontal tubular photobioreactors, two vertically stacked horizontal tubular photobioreactors and two flat panel reactors. All reactors are fully automated and flexible in order to allow a fast change in photobioreactor type, layout and process control strategies. Productivities in photobioreactors outdoors are expressed per ground surface. At AlgaePARC the large systems have the same ground surface (24 m^2) and receive thus the same amount of light but the volume differs.

The four types of reactor systems at AlgaePARC (left to right, top to bottom): open raceway pond, horizontal tubular reactor, vertically stacked tubular reactor and flat panel reactor. Photos: AlgaePARC

The large systems will allow comparison of different designs and study of the most important fundamental aspects for the successful operation and scale-up of photobioreactors, i.e. light regime, mass transfer and photosynthetic efficiency. They will serve as the basis to build up the knowledge required for the development of new, more competitive systems and strategies for scaling up. The smaller systems will be used to screen species, to test different feedstocks and to gain insight on reactor control and operation. If promising results are obtained in the small systems, we would then be able to scale up these systems immediately to a larger scale (24 m^2).

AlgaePARC is unique because it is an independent research center covering the whole production chain of microalgal products: systems biology, photobioreactor and production strategy development, biorefinery, scale-up, chain development and systems analysis, thus bridging the gap between fundamental research and industrial applications. AlgaePARC also has a direct link to fundamental research programs at Wageningen UR as well as a wide industrial network: 18 companies participate in this initiative. AlgaePARC is fast in development of technology because of the large critical mass generated in its technology platforms, from which dedicated product platforms can be shaped. AlgaePARC determines its research focus after analysis of the economy and sustainability of the whole production chain.

1. Norsker, N.H. et al. 2011. *Microalgal production – a close look at the economics*. Biotechnol. Adv.
2. Wijffels, R.H. et al. 2010. *Microalgae for production of bulk chemicals and biofuels.* Biofuels, Bioproducts and Biorefining 4
3. Wijffels, R.H.& Barbosa, M.J. 2010. *An .outlook on microalgal biofuels*. Science 329.

THE BIOMARA PROJECT

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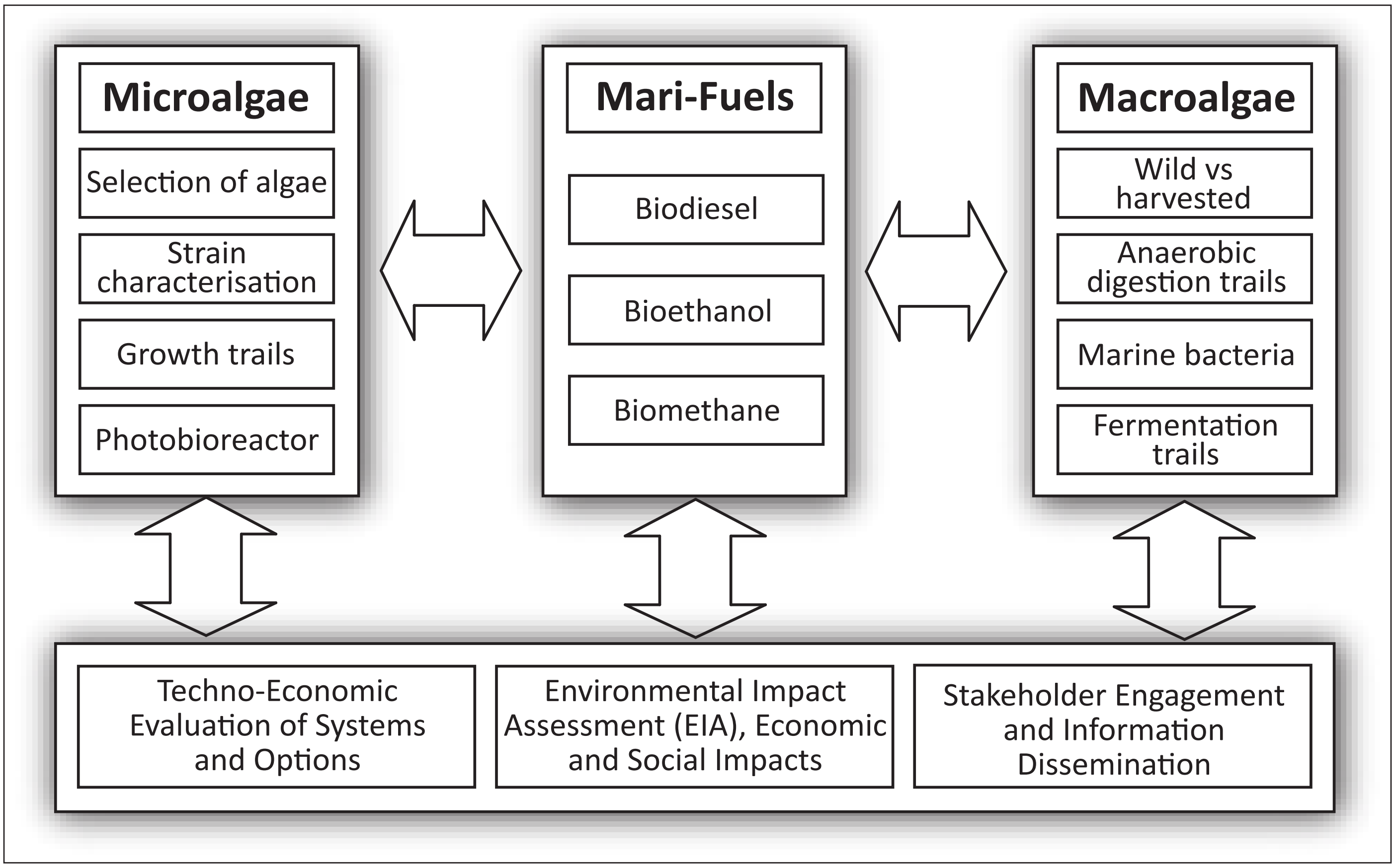
Over the past few years scientists at the Scottish Association for Marine Science (SAMS) have been investigating the potential of the marine environment to provide energy. The BioMara Project, hosted at the institute, is a €6 million joint UK/Irish project funded through the INTERREG IVA Programme, Highlands and Islands Enterprise and the Crown Estate. It kicked off in 2009, within months of the EU's Renewable Energy Directive (2009/28/EC) coming into force, with a requirement that, by 2020, 20 % of Member States' energy consumption and 10 % of transport fuel be from renewable sources.

Along with SAMS, the University of Strathclyde, the University of Ulster, Queen's University in Belfast, the Dundalk Institute of Technology and the Institute of Technology in Sligo are investigating the feasibility and viability of producing third generation biofuels from marine biomass, using both macroalgal (seaweeds) and microalgal (single celled plants) sources as an alternative to agri-fuels production from terrestrial plants. Although the project is focused on the science behind these forms of biomass for biofuel production it is underpinned by stakeholder engagement, economics and the social impacts.

Macroalgae

Macroalgae grow very rapidly and can be fermented to produce alcohols (ethanol, butanol, etc.) or be anaerobically digested (AD) to produce methane.



Overview of the main strands of BioMara.

Because they lack lignin and have a low cellulose content, the biomass is thought to be a better material for complete biological degradation to methane than land plants such as forestry and agricultural wastes. Seaweed to biofuel is not a new idea. As long ago as the 1970s, Americans investigated whether macroalgae could be used as a source of renewable energy. Their data showed that high levels of methane could be readily produced from seaweed. Wild harvest of seaweed for biofuels is unsustainable so cultivated biomass will have to be grown to support this industry as it develops. Cultivation of seaweeds has developed on a massive scale in China, mainly for food and chemical markets. It is now being developed in Europe for biofuels.

Seaweed cultivation has been established in Scotland since 2004. Ripe plants of the type of seaweeds to be cultivated are collected and spore release induced. The spores are then seeded onto special strings, where they germinate to form tiny plants which are ready for transfer to sea after two months. Mature plants are ready for harvest after 6-8 months. The biomass produced is then used to generate methane via anaerobic digestion or fermented to produce ethanol. There have been proven cultivation trials with *Alaria esculenta*, *Saccharina latissima* and *Saccorhiza polyschides*. To produce biofuel from macroalgae at a large scale via either ethanol fermentation or AD there needs to be an improved performance of both. This might be achieved by using marine bacteria for methanisation and bioethanol production, these microbes may be more suitable for conversion of this biomass rather than the currently used terrestrial bugs. The potential effect of seaweed cultivation on ecosystems and biofuel production on the terrestrial environment needs to be investigated further. But a key objective is improvement in crop yields through selective breeding and the expansion of existing culture banks.

Six month old cultivated plant *Sacchoriza polyschides*. Photo: L. Brunner (SAMS)

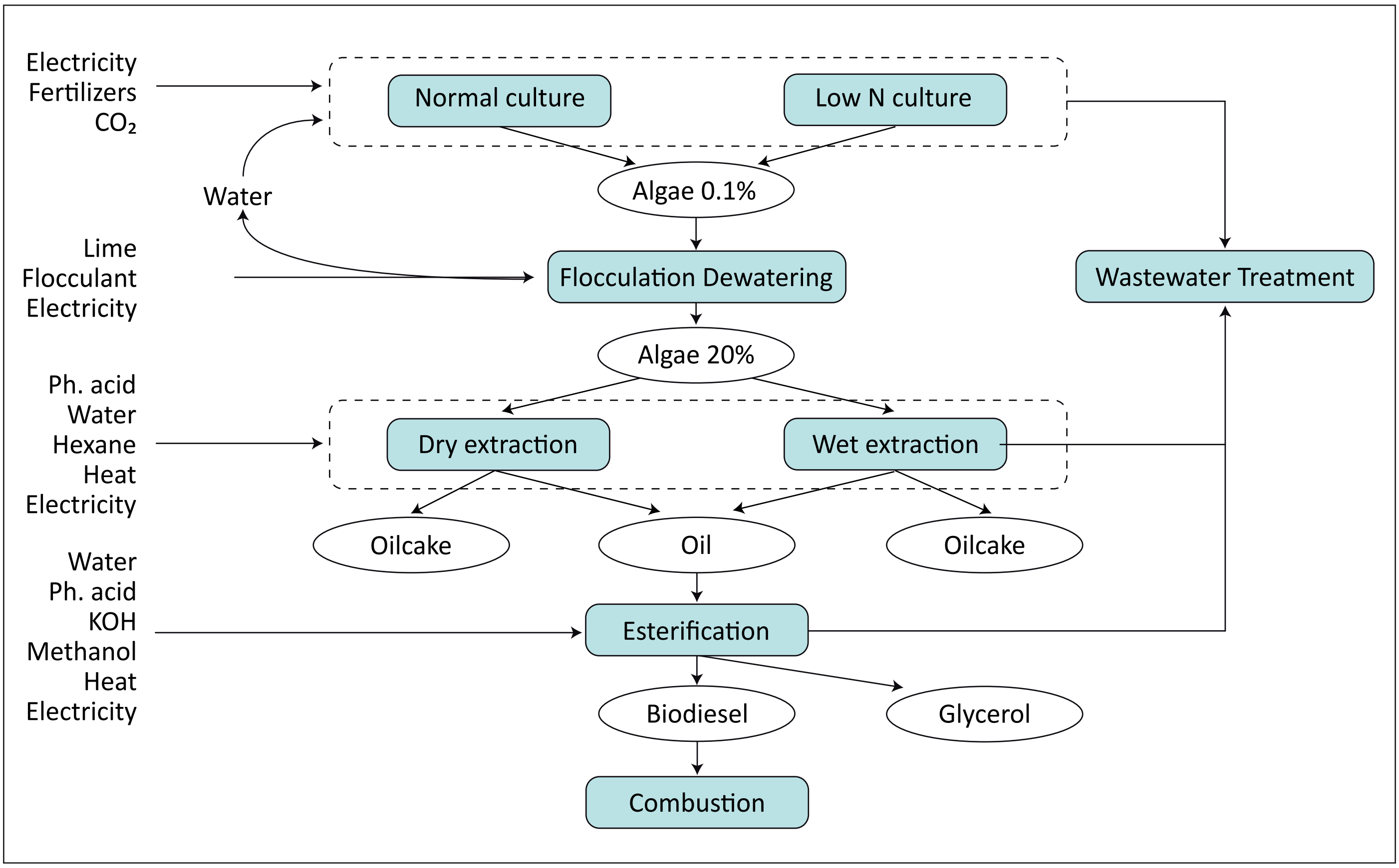
Microalgae

Microalgae are the green colour in your pond (or sometimes on your walls). Biofuel from these organisms has mainly focused on the oils they produce which can then be converted into biodiesel. The biomass they produce can also be used in fermentation and AD. One of the major technology barriers to the development of biodiesel from microalgae is the selection of an appropriate strain and the fact that we don't understand at a very basic level how these organisms actually work. We know they produce oils but not really how they do it and the control mechanisms of this production.

The Culture Collection of Algal and Protoza (CCAP) at SAMS holds the largest algal culture collection in Europe, some 2 700 strains. 500 of these strains are capable of growth under saline conditions, which means they have greater economic potential since they will not impact freshwater supplies for the production of biomass. From these, 200 have been selected for screening for oil production. The project receives starter cultures from CCAP. These are grown in 50 ml of culture media and any of the microalgae growing slowly at this stage they are eliminated from the screening. Slow growth of a microalgae means at the large volumes needed for biofuel production they will not grow fast enough to produce enough oils to be commercially viable. Next the cultures are grown at a large volume with air bubbled through. Again any poor growers are eliminated and the biomass is analysed for quantities and types of oil produced. Not all oils are suitable for biodiesel production and again any cultures which do not have the appropriate characteristics are dropped from the screen. Measurements of proteins and carbohydrates are also made of the biomass to give a more complete picture of the microalgae. Any microalgae with the appropriate characteristics will then be investigated further at a physiological and molecular biology level.

Conclusion

Although both macroalgae and microalgae have potential as sources of biomass for biofuel production, what has become clear is that for both macroalgae and microalgae biomass to be used as a source of biofuels that a biorefinery approach will need to be taken. This is where all aspects of the production are fully integrated and it is probably the only way that these sources of biomass will eventually become economically viable.



A potential biorefinery approach for the production of microalgal biomass.

PHOTOBIOREACTOR DESIGN PRINCIPLES

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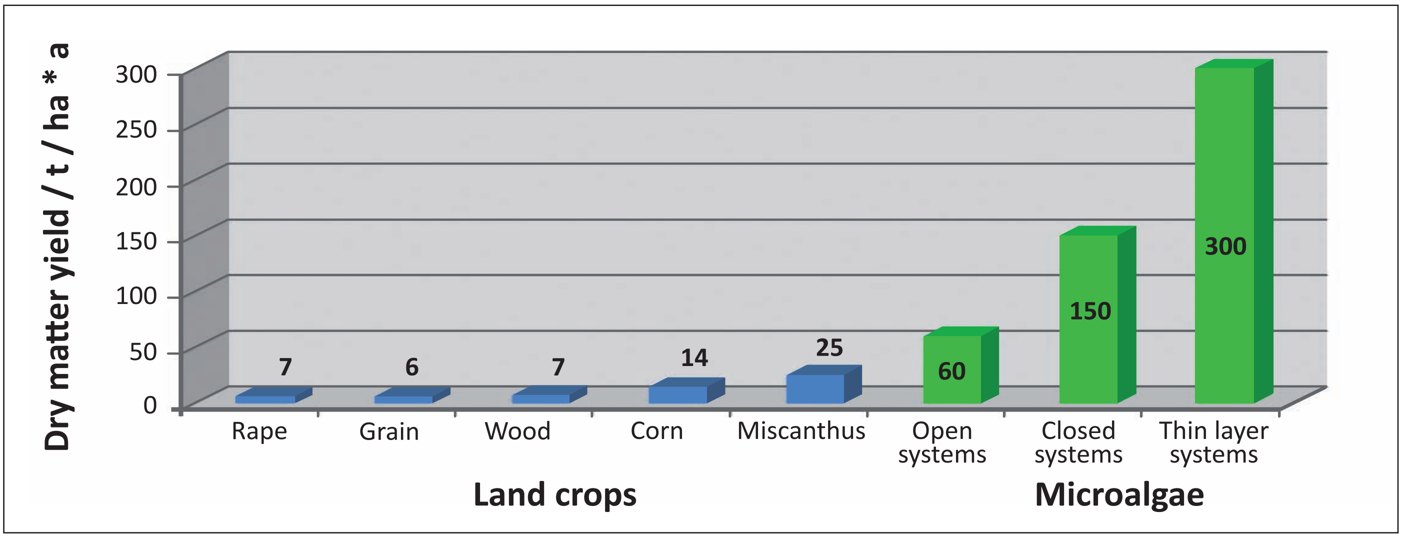
IGV INSTITUT FÜR GETREIDEVERARBEITUNG GMBH, GERMANY

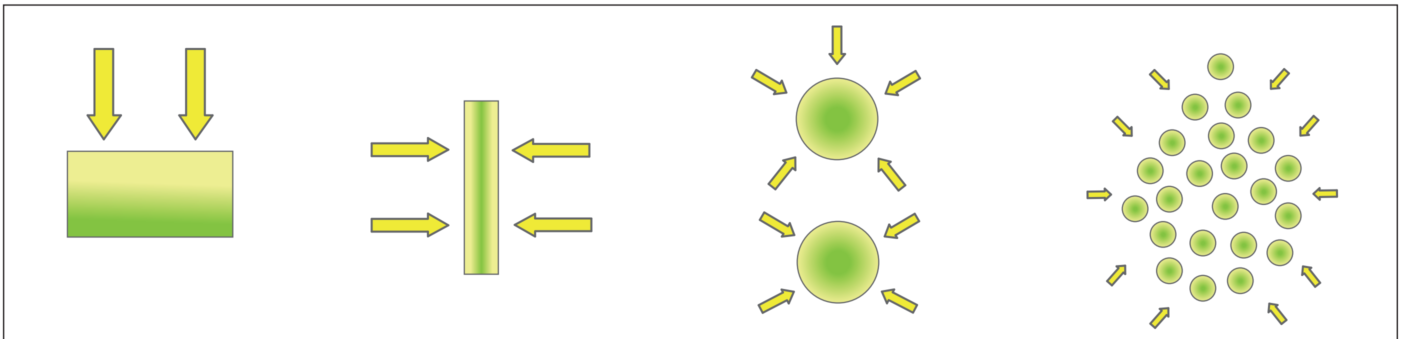
Going along with the discussion about climate change the interest of several companies and governments has been directed to the general replacement of fossil raw materials by renewables. During past years different technologies for general use of renewable resources like biomass, plant oil or solar energy have been developed and implemented all over the world. To date these technologies supply about 20 % of the global energy demand and replace this by renewable resources. The application of renewables for biomass conversion into bio-based raw materials is a central field for future development of a crude oil independent society.

The social aspect in the global use of biogenic resources, especially in the field of bioenergy, requires an extension of the raw material spectrum used for this purpose. To date photobioreactors (PBRs) for production of microalgae were mainly developed for nutritional supplements, cosmetic industry and pharmaceutical ingredients, as was the case for the 2002 Klötze (Germany) plant of closed tubular PBRs (technology by IGV), the largest of its kind with about 10 000 m². Currently, microalgae are undergoing a research and development focus for application to produce crude market raw materials. Many different concepts for cultivation of microalgae have therefore been developed and presented in patents or pilot plants. The core motivation of this development is the utilization of the superior theoretical areal growth, which dissociates the microalgae from the rest of the classic land crops.

The first step in planning a microalgae production plant is fixing the final product under consideration of local site conditions and species demands. Even this very basic criterion has strong influence on the plant design. In the case of high value products for cosmetic or pharmaceutical use, closed systems seem to be the optimum solution, though a demand for high sterility requires an increased plant investment. Otherwise some high value products can also be produced in cheaper open systems using a more complex downstream process. Another sub-criterion is the possibility of cultivating under extremophile conditions, i.e. low pH (*Cyanidium* sp.),



Productivity head start of microalgae compared with land crops.



Constructive principles of light supply inside photobioreactors.

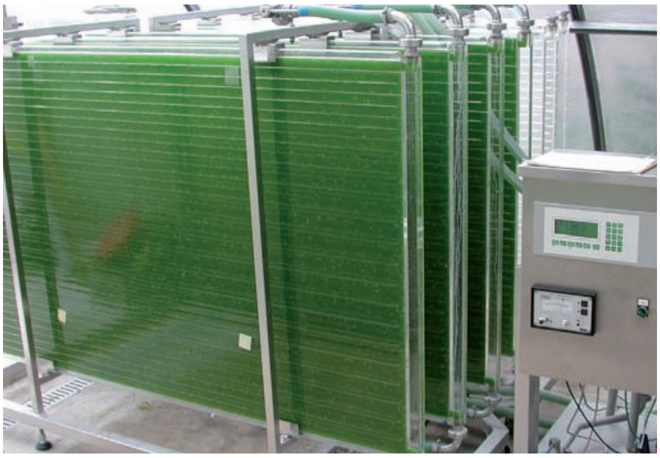
high pH (*Arthrospira* sp.) or high salinity (*Dunaliella* sp.). After determining the mentioned factors within a market study, the technology selection process for a scaled production plant can be performed.

Independent from scaled biomass production processes, different concepts for PBRs have been designed following the second basic design criterion of optimum light supply, which is still under development. During photoautotrophic growth, in addition to nutrients, algae need CO_2 and light for photosynthesis. In high cell densities many algae cells can be shaded in a cultivation suspension by other cells. If the suspension layer thickness reaches a specific level of dry mass dependent light extinction, the growth speed would decrease and leave exponential phase. To avoid this effect various concepts have been developed.

The cheapest systems for microalgae cultivation, the open ponds, are characterized by a 2-dimensional light supply from above. An effect of this construction can be that in large distances to the mixing device the algae cells descend to the dark basin bottom and no longer grow until being lifted up again. This results in daily peak productivities of approximately 20 $g/m^2 d^{-1}$. In contrast, closed PBRs of tubular or plate (glass, plastics or foil) construction design allow 3-dimensional light supply and the adaption of the layer thickness to algae or site specifications like solar irradiation. Compared to open systems, peak productivities can be doubled to approximately 40 $g/m^2 d^{-1}$. Other effects are the improvement of product quality by separation of the suspension from the atmosphere and the reduction of harvesting costs via higher cell densities within the algae culture.

The optimization of the growth-limiting light supply has led to various ideas for realisation. Some solutions have used optical fibers (or similar) to bring light directly into the suspensions, but this concept has not proved convincing yet. Another idea is to build very thin layers of algae suspension to reduce the extinction effect for every single cell. Few companies work on this topic but expected peak growth data of approximately 80 $g/m^2 d^{-1}$ show the potential of those systems. IGV is currently working on the thin layer PBR and the first trials in small technical scale have been successful.

A third central issue of PBR design is the need to mix the culture. Algae cells need to be brought to illuminated zones inside a reactor and are therefore moved around through stirrers, pumps or air bubbles. Another reason to mix the suspension is for oxygen removal and compensation of temperature and nutrient gradients. Finally, mixing devices prevent the decline of algae cells away from light supply and avoid algae cell immobilization inside the PBR wall. The quality and fulfillment of these tasks by the mixing devices is strongly connected to the energy amount put into the medium. The process



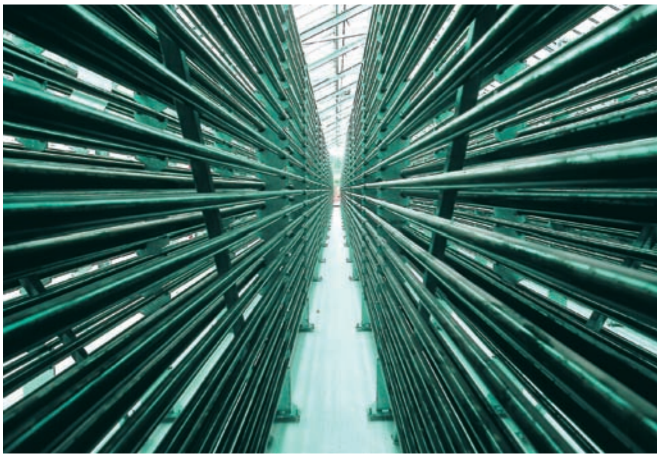


Plate and tubular photobioreactors. Photos: IGV GmbH, Germany

preferences have to be in close contact with the energy consumption profiles, as mixing is one of the main issues in the overall energy balance.

Up to now mainly open ponds and tubular PBRs are used for production of microalgae biomass. Open ponds are advantageous for their low investment costs while tubular PBRs allow for controllable biological and physical process conditions. But neither can reach the biomass production costs of <1 US $ / kg (dry matter) needed for economic feasibility of producing bulk materials for the biofuel market or the chemical industry. By following the path of a biorefinery mass stream, an integrated production process can be generated, in which algae biomass production is directly linked to other downstream and upstream processes, aiming at a complete utilisation of each biomass fraction. This results in a complex network of different industries using waste streams of partners to produce a large variety of possible raw materials for biofuel, chemistry or feed industry. In the end, the sustainability of such a technology cluster has to be checked and proven to be overall balanced, including data on CO_2, energy and water consumption as well as nutrient balances.

Founded in the early 1960s, IGV dedicated its work to applied research and development of customised products, sustainable and effective production processes, practice-oriented technical approaches in food and baking technology, analytic and quality assurance, renewable raw materials and blue biotechnology. The company has a staff of 112, including about 90 scientists, who keep in contact with industrial and scientific partners for optimum application and transfer of technology, resulting in solutions tailor-made to customer demands.

Since the 1980s, IGV Biotech, headed by Prof. Otto Pulz, works in the field of process and application development for cultivation of microalgae. In several research projects the potential of microalgae for food supplements, functional foods, cosmetic and wellness products as well as pharmaceuticals has been examined and developed into marketable product commodities. Photobioreactor development has led to more than 20 international patents which are the foundation for almost 200 units of closed photobioreactors sold worldwide, mainly based on the tubular principle. The largest singular module ever built has an operating volume of 85 000 l. New developments seek the tenfold reduction of production costs, so as to enter the big market of biofuels and CO_2 capture. More information can be found at www.igv-biotech.com.

BIOETHANOL PRODUCTION FROM CARBOHYDRATE-RICH BIOMASS OF MARINE MICROALGAE

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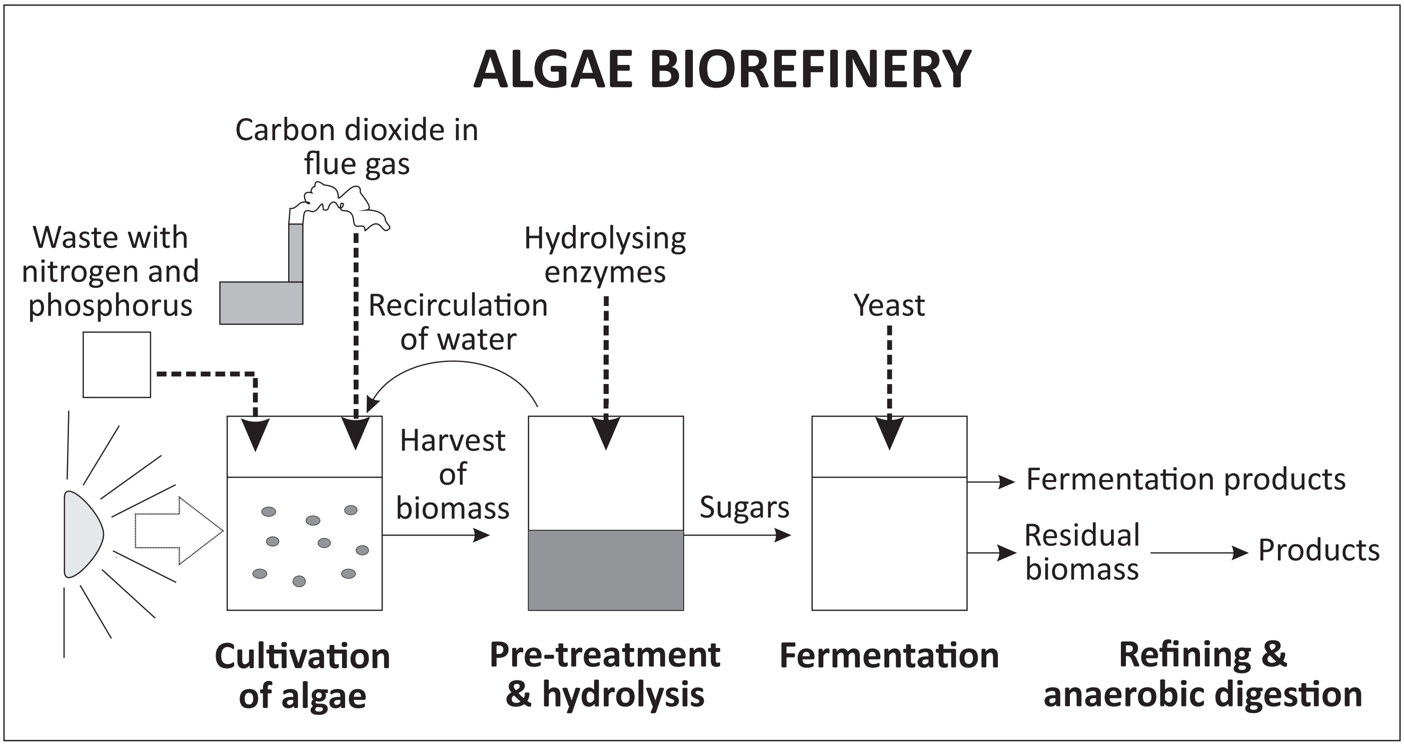
The worldwide demand for renewable energy sources has put focus on the use of biomass as raw material for fuel production. Up until now, plant materials have been used to produce ethanol and biodiesel from carbohydrates and lipids, respectively. However, microalgal biomass also has the potential to provide these compounds for production of fuels.

In this research project we want to use algal biomass as a carbohydrate-rich feed stock for bioethanol production by yeast. To obtain efficient hydrolysis, fermentation and the down-stream processes of ethanol production, the carbohydrates in the algal biomass should preferably be present as starch. Certain groups of algae use starch as a storage compound, which can be accumulated in the cells at nutrient-limiting conditions. Thus, for bioethanol production based on algal biomass, suitable marine microalgal species containing starch need to be identified.

In addition, by optimising culture conditions an algal biomass with optimal cellular composition can be obtained. In our preliminary experiments it was found that some carbohydrate-rich algal species could accumulate carbohydrates at 2-4 times higher levels in nitrogen-limiting conditions compared to balanced medium. The carbohydrates should then be enzymatically degraded to yield fermentable sugars for the ethanol fermentation. In summary, starch from cultured marine algae might be a good alternative source of sugar to be used as substrate in fermentation.



Algae biorefinery concept.

COMPARISON OF TWENTY COLD AND WARM WATER ADAPTED MICROALGAE

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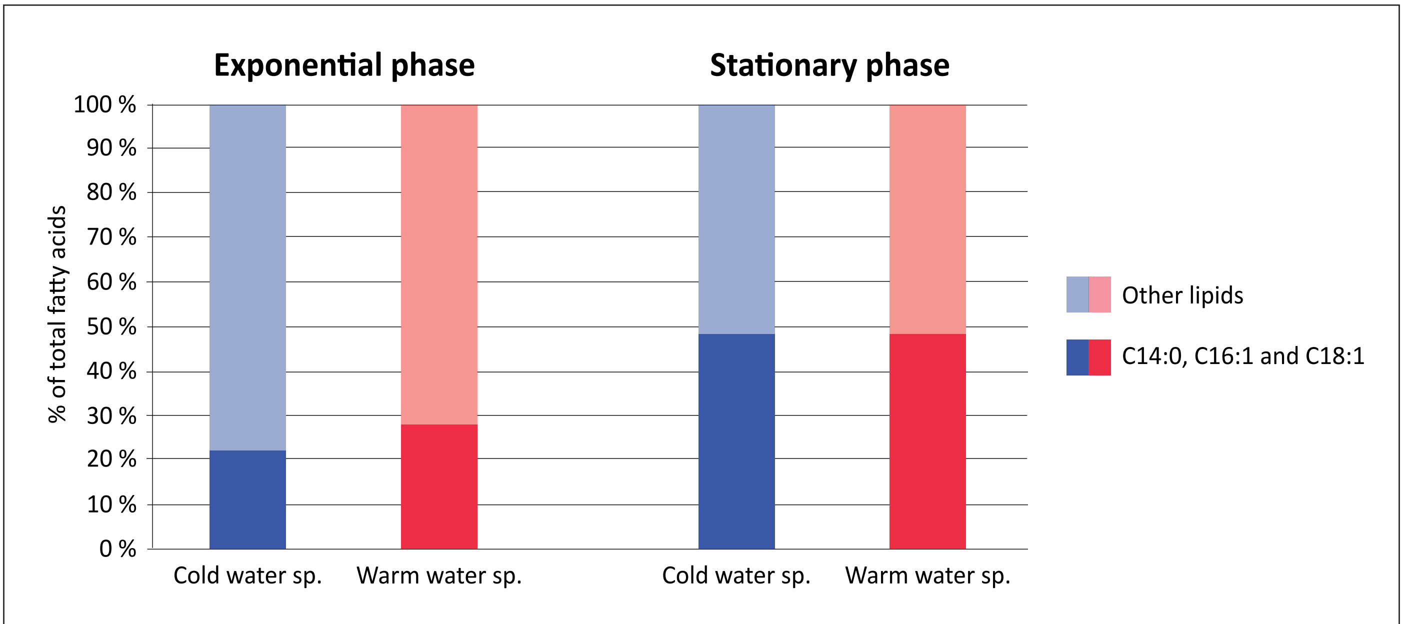
In spite of cold temperatures, polar and sub-polar regions are characterised by high aquatic primary production. Finding microalgae species showing high growth rates and lipid production at low temperatures could allow producing microalgae as a source of biofuel in those countries in which growing terrestrial plants for biofuel production is only possible to a limited extent.

In the presented study we determined lipid contents and profiles of twenty cold and warm water adapted microalgae, mainly from the Baltic Sea, in exponential and stationary growth phase, compared them and evaluated their suitability for biofuel production.

Lipid contents of the investigated cold water species in the stationary growth phase were between 10.9 % and 26.5 % of dry weight, comparable with most of the warm water species. A low ratio of unsaturated fatty acids (UFA) to saturated fatty acids (SAFA) is necessary to ensure high lipid stability. In the exponential growth phase, the investigated cold water species showed a significantly lower UFA/SAFA ratio than the warm water species, while ratios of both groups were approximately similar in the stationary growth phase. Short-chain fatty acids such as C14:0, C16:1 and C18:1 as required for biofuel were comparable between cold and warm water microalgae.

The cold water adapted diatoms *Chaetoceros wighamii* and *Thalassiosira baltica* showed both highest lipid contents and highest lipid quality for biofuel production and are therefore good candidates for further optimisation studies.



Proportions of lipid components C14:0, C16:1 and C18:1 of total fatty acids (%) required for biodiesel production (from Schenk, P. M. et al. 2008. *Second Generation Biofuels: High-Efficiency Microalgae for Biodiesel Production*. BioEnergy Research 1)

OPTIMISING MICROALGAL LIPID PRODUCTION: ENHANCED CULTIVATION METHODS AND EFFECTS OF SPECIES DIVERSITY

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Biofuel is currently produced mostly from terrestrial plant biomass. This may lead to competition for fertile land and water with global food production. An alternative biomass source for biofuel can be the growth of microalgae. The installation of infrastructure for large-scale production of biofuel from microalgae is essential to establish cultivation methods that maximise lipid production but that are also economically viable in terms of energy demand and resource supply.

For this purpose, we isolated several algae that showed both high growth rates and potentially high lipid contents. Population parameters such as 1) algal growth dynamics under different nutrient and light supply levels, 2) biomass yield per unit of limiting nutrients, and 3) biomass lipid content, were estimated. Additionally, we compared different cultivation systems (batch, chemostat) to simultaneously optimise growth and biomass lipid content of different algal species.

Higher light intensities mainly influenced algal growth, while nitrogen limitation mainly influenced algal lipid content (positively). *Botryococcus braunii* was the algal species showing the largest response to nitrogen limitation with highest cell specific lipid content. However, its growth rate was strongly influenced by nitrogen supply, resulting in very low biomass production at low nitrogen levels.

To enhance both biomass accumulation and lipid production simultaneously, we further investigated a two-stage cultivation method to replace one-stage semi-batch cultivation systems. In the first step, a full growth medium allowed an enhancement of biomass accumulation. In the next step, the culture was transferred into nitrogen limited growth medium, where a further accumulation of photosynthetic products in the form of lipids occurred. Two-stage cultivation cultures resulted in higher nutrient specific biomass production and algae lipid content compared to one-stage cultivation. If a continuous cultivation of cultures with high biomass in stage one can be assured, an almost constant supply of huge amounts of algae with high lipid content in the second step could be guaranteed.

Over 40 different algal strains representing the major freshwater algal classes are available for experiments at the University of Munich. Photo: M. Stockenreiter

However, uncertainty remains as to whether monocultures are actually more productive in terms of lipid accumulation than diverse microalgal communities. A potential positive relationship between diversity and productivity within primary producers has already been documented. Hence, we set out to investigate, experimentally, whether diversity may also affect lipid production in microalgae communities.

We investigated the growth and lipid production of microalgae by using species from all major algal groups. Algae were grown in a large number of treatments differing in their diversity level. Additionally, we compared growth and lipid production of laboratory communities with lipid production of natural lake and pond phytoplankton communities along a diversity gradient. Fluorometric analyses were performed using an imaging flow cytometer FlowCAM® to estimate lipid content and save images of each algal cell in diverse communities.

Our results show that diverse algal communities had higher lipid production than monocultures compared under the same growth conditions and resource supply rates. Diversity also influenced the cell specific algal lipid content as a quotient of the total algal lipid content and total algal biovolume. The major underlying mechanism behind our observed diversity-productivity relationship was complementarity. Species complement one another in resource use efficiency or facilitate each other‘s growth. The dominance of a single highly productive species was not responsible for the observed positive effects of diversity on lipid production.

In summary, we observed that 1) there is a link between biodiversity and lipid production in microalgal communities; 2) the existing link is based on resource partitioning and facilitation among algal species and not on the dominance of a single highly productive algal species; and 3) diversity does not only positively influence algal biomass production, thereby increasing lipid yields in more diverse communities, it does influence the cell specific lipid content of microalgae.

The comparison of eight natural lake phytoplankton communities with laboratory microalgal communities showed that the lipid production of selected laboratory monocultures was not significantly higher than that of natural phytoplankton communities. This was an unexpected observation, as natural phytoplankton communities were not habituated to the nutrient-rich growth medium and the environmental conditions of the laboratory, unlike the long established laboratory cultures. Additionally, as species richness increased in the natural communities, lipid production also increased. This supports the diversity-lipid production relationship found in laboratory communities, as well as the diversity-productivity relationships found in natural phytoplankton communities.

This enhancement of the yield efficiency of lipid production in diverse algal communities would be difficult to do only by technical means such as increasing resource supply. In addition, increasing the supply of resources is costly and usually correlated with high energy requirements. In contrast, an increase in biological efficiency (resource use efficiency) of the system is usually self-financing. It is therefore important for biomass production systems to utilise all possible ecological options to increase the efficiency in the use of the supplied resources by integrating basic ecological principles into cultivation systems.

Outdoor facilities at the University of Munich for potential upscaling processes of laboratory experiments. Photo: M. Stockenreiter

ARE ALGAL OIL YIELD ESTIMATIONS DEPENDENT ON SEASONAL VARIATION?

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[1] LINNÆUS UNIVERSITY, SWEDEN
[2] NECTON COMPANY, PORTUGAL
[3] IFREMER, FRANCE

Microalgae as a potential candidate in biofuel production

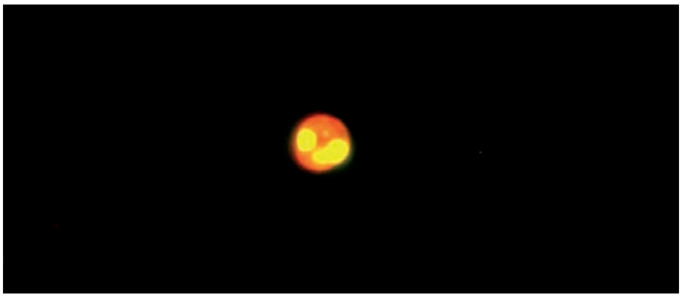
The shortcomings of fossil fuels, among them climate-induced changes due to elevated CO_2 emissions, have create a need for cleaner and renewable fuel. Oil-producing microalgae are a potential feedstock for the biofuel industry. The main advantages of algal biofuel are: 1) the possibility of growing algae on arid or marginal lands, thus not competing with food crops; 2) CO_2 neutrality, since emissions can be captured and used in algal mass cultivation; 3) higher productivity per unit area compared to land grown crops; 4) reduction in the use of freshwater since microalgae can be grown in seawater; 5) recycling of nutrients when using wastewater during the cultivation process; and 6) non-toxic, biodegradable and renewable fuel.

Microalgae, as any other plant, need light, CO_2, water and nutrients to grow and provide a biomass that can be converted into different kinds of fuel such as H_2, biodiesel, bioethanol and biogas. Biodiesels are based on the oil or lipids within the algal cells. The most preferable of these for biodiesel production are the saturated fatty acids (SAFA) and monounsaturated fatty acids (MUFA). Less desirable are the polyunsaturated fatty acids (PUFA). Estimations of the yearly algae-based oil yield in outdoor production are often based on laboratory studies. These projections can be largely overestimated since they do not include environmental or weather fluctuations.

In this study we monitored the seasonal variation of lipid content and composition in relation to light and temperature for the marine microalgae *Nannochloropsis oculata* grown outdoor in large-scale photobioreactors.

Seasonal variations of algal oils can affect a constant biofuel production

Seasonal variation of total lipids (TL) and fatty acid (FA) profiles in *N. oculata* grown outdoors in flat plate photobioreactors were analysed from samples collected monthly from June 2008 to May 2009. Lipid content ranged from 11 % of dry weight (DW) in winter to 30 % of DW in autumn. Lipid accumulation can be visualised in the cells with the lipid stain Nile Red.

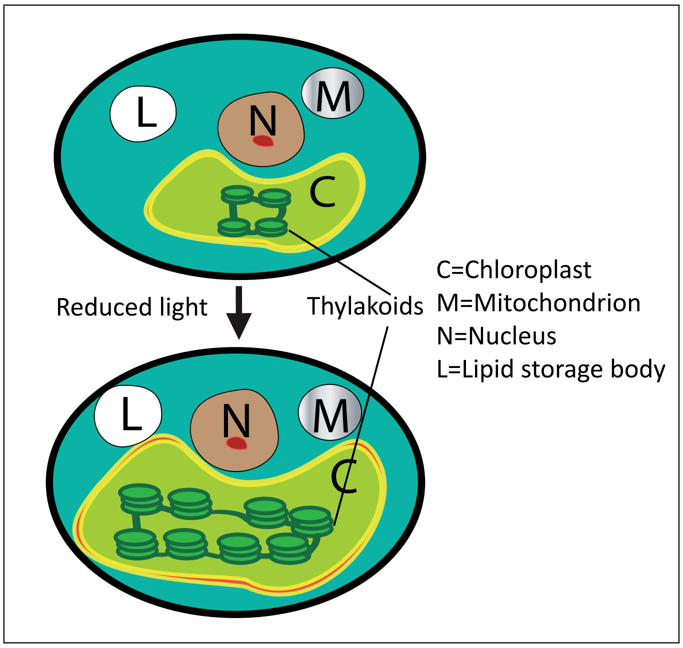


Lipid droplets in *Nannochloropsis oculata* stained with Nile Red appear yellow under blue light. Chloroplasts appear red. Photo: M. Olofsson

Total lipids (TL) and fatty acid (FA) profiles were correlated with light in the form of total global radiation (TGR) (Instituto de Meteorologia, Portugal) and temperature (http://clima.meteored.com). Light and temperature together had a positive significant effect on lipid content ($p < 0.01$) and explained the variation in TL with more than 50 % ($R2 = 0.51$).

The highest lipid content (30% DW) was recorded during autumn when light and temperature started to decrease. It is possible that the photosynthetic apparatus in the cell was rearranged and enlarged at reduced light, resulting in more structural lipids. By doing so the size of the chloroplast in the cell is enhanced and the area of the thylakoid membranes is increased with increased thylakoid stacking.

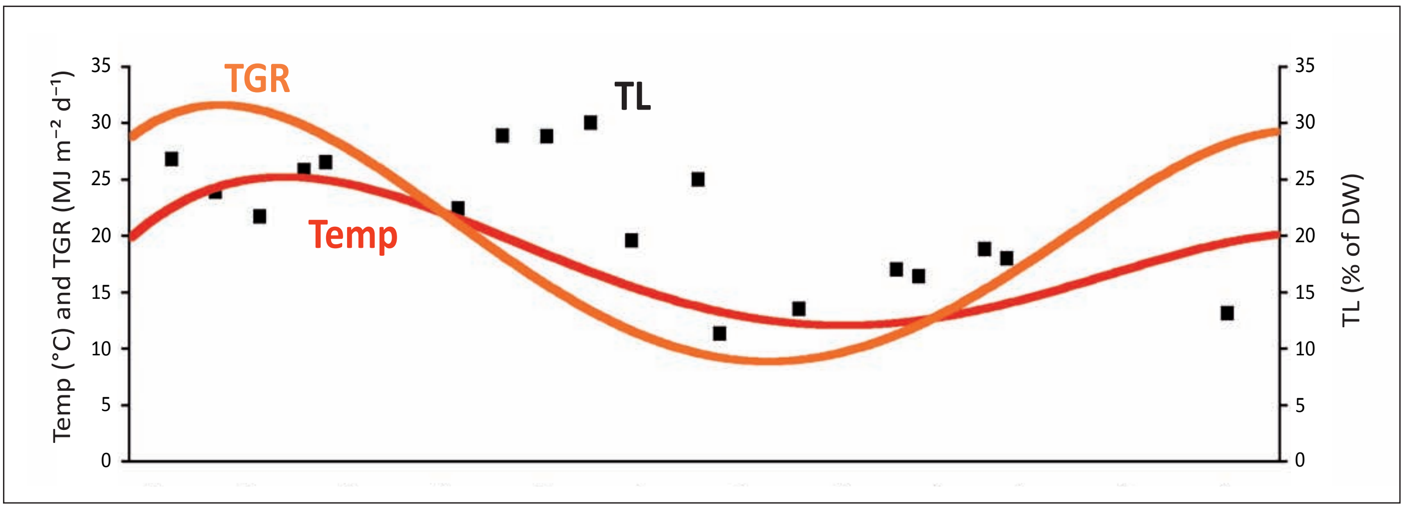
Although FA profiles of *N. oculata* varied over the year, a decrease in light and temperature during autumn coincided with an increased accumulation of monounsaturated fatty acids (MUFA) possibly at the expense of polyunsaturated fatty acids (PUFA). This suggested synthesis of structural fatty acids in the form of MUFA as a consequence of a rearrangement in the photosynthetic apparatus during autumn at lower light.



Seasonal variation of total lipids (TL, squares) in *N. oculata* in relation to temperature (red line) and light in the form of total global radiation (TGR, orange line) during 2008-2009.

Conclusions

N. oculata proved to be a potential candidate for biofuel production due to its relatively high lipid content. Optimal lipid accumulation indicated a complex relationship with light and temperature. Reduced light conditions in autumn may have triggered the lipid accumulation in the form of MUFA through a rearrangement and enlargement of the photosynthetic apparatus leading to increased synthesis of structural lipids. Evidently, our results showed the importance of considering seasonal variation in lipid content and composition when estimating annual oil yield from microalgae. The results in the present study raised a question about the applications of algal cultivation in the Baltic Sea region. Can the drastic decline of light and temperature in autumn around the Baltic Sea help to boost algal lipid production? This remains to be tested in northern latitudes.



Seasonal variation of total lipids (TL, squares) in *N. oculata* in relation to temperature (red line) and light in the form of total global radiation (TGR, orange line) during 2008-2009.

INVESTIGATING THE ENERGY DEMANDS OF MICROALGAL PHOTOBIOREACTORS FOR SUSTAINABLE BIOENERGY

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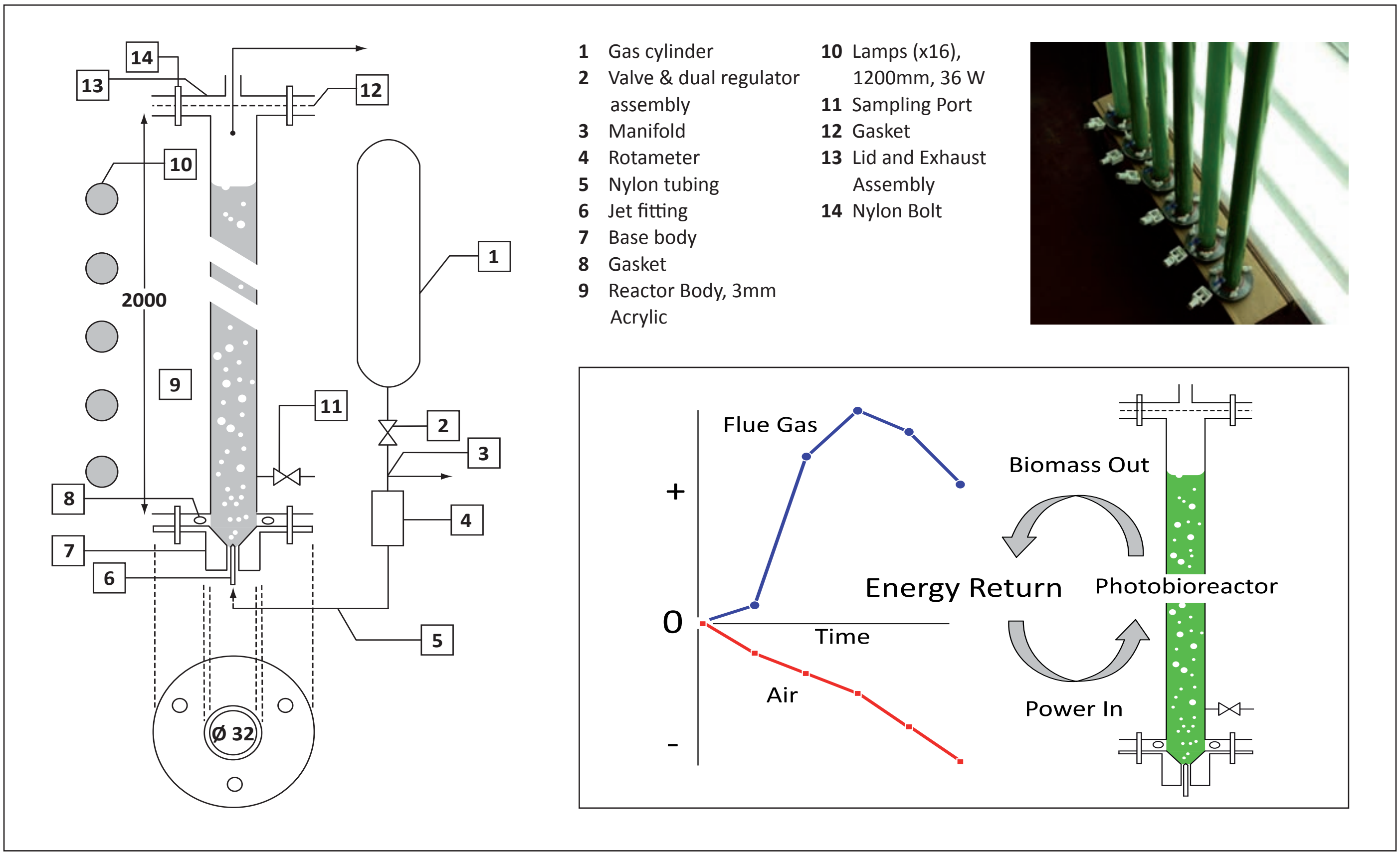
One of the major challenges facing microalgal technologies proposed for bioenergy and CO_2 mitigation purposes is the production of biomass in an environmentally sustainable manner. This work examined the energy return of *Chlorella vulgaris* and *Dunaliella tertiolecta* cultivated in a gas-sparged photobioreactor design where the power input for sparging was manipulated (10, 20, 50 Wm^{-3}). Dry weight, organic carbon and heating values of the biomass were measured, plus variables including Fv/Fm and dissolved oxygen. A model for predicting the higher heating value of microalgal biomass was developed and used to measure the energetic performance of batch cultivations.

High power inputs enhanced maximum biomass yields but did not improve the energy return. Cultivation in 10 Wm^{-3} showed up to a 39 % higher cumulative net energy return than 50 Wm^{-3}, and increased the cumulative net energy ratio up to four-fold. The highest net energy ratio for power input was 19.3 (*Dunaliella tertiolecta*, 12 % CO_2, 10 Wm^{-3}).

These systems may potentially be a sustainable method of biomass production but their effectiveness is sensitive to operational parameters. However, there is considerable potential to further minimise energy inputs into photobioreactors.



Photobioreactors designed to test the effect of power input on the net energy return of microalgal cultivations. Photo: C. Hulatt

ABOUT THE SUBMARINER PROJECT

The SUBMARINER project was initiated in 2010 with partial funding from the EU Baltic Sea Region Programme 2007-2013. Together, the project partners are building the road for furthering environmentally friendly and economically appealing innovative uses within the Baltic Sea region.

ACTIVITIES

Compendium

Describing current and potential future marine uses:

- Inventory of current and new uses
- Strengths, weaknesses, opportunities and threats to the BSR
- Environmental and socioeconomic impacts
- Status and availability of technologies
- Market potential
- Gaps and obstacles in the legal framework

Regional Strategies

Testing new uses in real conditions:

- Feasibility studies for new uses
- Technological and financial needs
- Impacts on environmental and socioeconomic conditions
- Specific legal constraints

BSR Roadmap

Recommending necessary policy steps to promote beneficial uses and mitigate against negative impacts:

- Legal changes (e. g. spatial plans)
- Environmental regulations
- Economic incentives

BSR Network

Bringing relevant players together:

- Business cooperation events
- Network structure
- Virtual information and exchange platform
- Regional, national and BSR-wide roundtables and seminars on new marine uses

PROJECT PARTNERS

Poland

- Maritime Institute in Gdańsk
- Gdańsk Science and Technology Park

Lithuania

- Klaipeda University Coastal Research & Planning Institute
- Klaipeda Science & Technology Park

Denmark

- ScanBalt
- Lolland Energy Holding

Estonia

- Tallinn University of Technology
- Entrepreneurship Development Centre for Biotechnology & Medicine

Finland

- Finnish Environment Institute

Sweden

- Royal Institute of Technology
- The Royal Swedish Academy of Sciences
- Trelleborg Municipality

Germany

- Federal Ministry for Environment, Nature Conservation and Nuclear Safety
- Norgenta North German Life Science Agency
- University Rostock (Inst. of Constitutional & Administrative Law, Environmental Law & Public Economy Law)
- Kieler Wirkstoff-Zentrum am IFM-GEOMAR
- BioCon Valley Mecklenburg-Vorpommern e. V.

Sweden

- Royal Institute of Technology
- The Royal Swedish Academy of Sciences
- Trelleborg Municipality

Latvia

- Ministry of Environmental Protection and Regional Development of the Republic of Latvia
- Environmental Development Association