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Bioconversions at halo-alkaline conditions for methane production from sunlight

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1 Algae as a fuel source

Fossil fuels are a non-renewable fuel source and their combustion results in the emission of the greenhouse gas carbon dioxide, with potential detrimental effects on Earth's ecosystems (IPCC, 2007; Melillo et al., 2014). Biofuels could offer a sustainable alternative for fossil fuels, yet the growth of terrestrial energy crops has severe environmental and socio-economic consequences (Escobar et al., 2009; Fargione et al., 2008; Groom et al., 2008; Kazamia and Smith, 2014; Schenk et al., 2008; Searchinger et al., 2008; Singh et al., 2011). Using aquatic oxygenic microalgae, such as unicellular algae and cyanobacteria, as feedstock for biofuel production eliminates the drawbacks associated with growing terrestrial energy crops. The cultivation of unicellular algae and cyanobacteria does not compete with food or feed crops for arable land and water, since it does not require fertile soil and fresh water (Schenk et al., 2008). Furthermore, the biomass yield ($\frac{ton\ dry\ biomass}{year \cdot ha}$) of aquatic oxygenic phototrophs is about one order of magnitude higher than that of terrestrial crops (Dismukes et al., 2008). For the sake of readability, these aquatic microbial oxygenic phototrophs will be henceforth referred to as microalgae, in accordance with Zamalloa et al. (2011).

Most research on microalgal biofuel focusses on the production of biodiesel, but other types of biofuel can be produced from microalgae as well (Schenk et al., 2008). Anaerobic digestion of algae has been investigated as early as the late 1950s (Golueke et al., 1957). The absence of lignin and the relatively high lipid and carbohydrate content make microalgae well-suited for the production of biogas by means of anaerobic digestion (Schenk et al., 2008; Vergara-Fernández et al., 2008). In contrast to biodiesel and bioethanol, all carbon components of the biomass can be digested which makes it the most efficient option among the different biofuels (De Schamphelaire and Verstraete, 2009; Harun et al., 2011). In particular, when the lipid content of the biomass is lower than 40 %, complete methanisation of the biomass is more efficient than lipid extraction followed by methanisation of the residual biomass (Sialve et al., 2009). From a process point of view, anaerobic digestion of algal biomass partially circumvents the need for biomass concentration (Collet et al., 2011; Klass, 1977). Further, no efforts are required to separate the product of interest (methane) from the culture fluids, because biogas escapes from the liquid spontaneously (Collet et al., 2011; De Schamphelaire and Verstraete, 2009; Harun et al., 2011).

2 Current limits to bio-methane production from microalgae

Currently, several aspects of algal production limit a widespread use. The required input of fossil fuels for the construction and operation of algae growth systems often surpasses the energy content of the produced biofuel, resulting in a negative energy balance (Acién et al., 2012;

Reijnders, 2008; Sawayama et al., 1999; Slade and Bauen, 2013; Uduman et al., 2010; Wijffels, 2008). The monetary costs of growing algae for biofuel production are also too high to make algal biofuel economically competitive with fossil fuel (Acién et al., 2012; Norsker et al., 2011; Reijnders, 2008; Sialve et al., 2009; Stephens et al., 2010; Uduman et al., 2010; Zamalloa et al., 2011). Because of its high cost, the current practice of growing algae mainly aims at high value products such as pharmaceuticals and food additives, instead of biofuels.

The poor technological and economic performance of contemporary algal biofuel production systems has been attributed to a number of factors. Operational costs and energy consumption are high because the gas containing the carbon dioxide needs to be bubbled through the bioreactor filled with diluted algae and the operation of the compressors for the gas bubbling consumes electricity. According to Slade and Bauen (2013), the forced supply of carbon dioxide can make up ca. 50 % of the cost of biomass production in a raceway pond system with a production rate of 3.0 – 3.6 $kg\ m^{-2} d^{-1}$. Acién et al. (2012) estimated the cost of carbon dioxide to be 36.5 % of the total raw materials and utilities cost for the production of dry biomass of *Scenedesmus almeriensis* at a scale of 200 $ton\ y^{-1}$.

The downstream processing of the algae biomass into energy carriers requires an energy-consuming concentration step. It has already been mentioned that this problem is partially alleviated by using the algal biomass for anaerobic digestion, since the latter process requires a less concentrated feedstock than the extraction of lipids for biodiesel production. The biogas resulting from anaerobic digestion of the biomass can be combusted to produce electricity (Oswald and Golueke, 1960; Zamalloa et al., 2011), or it can be upgraded to obtain the same methane content as natural gas, enabling its use as a transport fuel or its injection into the gas grid (Hengeveld et al., 2014; Yang et al., 2014). Obviously, upgrading biogas to a higher methane content entails an energetic and economic cost.

The process concept presented in this contribution overcomes these bottlenecks by conversion of sunlight to biomass and biomass to methane at high pH.

3 Alkaline conversions in nature

Both growth of algae and anaerobic digestion of organic matter are known to happen in nature at high pH and high salinity. Several studies of alkaline soda lakes have clearly shown that both microalgae and cyanobacteria are highly active in such lakes (Andreote et al., 2014; Ballot et al., 2004; Ballot et al., 2005; Melack and Kilham, 1974; Samylina et al., 2014; Schagerl and Oduor, 2008; Seckbach, 2007). Soda lakes are saline lakes containing mainly sodium carbonates. They can be found in the East African Rift, Siberia and South-western USA, all with pH above 9. Indeed, these ecosystems are among the most productive in the world (Melack, 1981). Alkaline soda lakes studied in Africa, Siberia and North America have a moderate to very high salt concentration (sodium carbonate concentration up to saturation), a pH range from 9 to 11 and a diverse microbial community that actively performs carbon, nitrogen and sulfur cycling (Kupriyanova and Samylina, 2015; Mesbah et al., 2007; Sorokin et al., 2015b; Sorokin et al., 2014; Sorokin and Kuenen, 2005; Sorokin et al., 2011; Zavarzin et al., 1999).

Also for the feasibility of the second part of the proposed process, the anaerobic digestion of biomass at high pH, less is known but, still, some experimental evidence is available. In the alkaline soda lakes already mentioned, biomass is degraded in the sediments with methane as the end product (Nolla-Ardèvol et al., 2012; Sorokin et al., 2015a; Yoshida et al., 2014). Alkaline pretreatment of lignocellulosic feedstock for anaerobic digestion is known to improve biogas yields by facilitating the enzymatic hydrolysis (Monlau et al., 2013; Taherzadeh and Karimi, 2008; Zheng et al., 2009). Sediments from Central Asian soda lakes were found suitable for the inoculation of high pH methane producing bioreactors (Nolla-Ardèvol et al., 2012).

4 A new approach for algal biogas production at haloalkaline conditions

Microorganisms appear to thrive in several naturally alkaline environments, but also in technical systems microorganisms were successfully applied under haloalkaline conditions (Zhao et al., 2014). Biotechnology at halo-alkaline conditions, therefore, in theory, has great potential for algal growth and subsequent biogas production.

The process put forward in the present contribution aims on the one hand to reduce capital costs by increasing volumetric productivity and on the other hand to reduce operational costs by improved carbon dioxide absorption rates and by the elimination of active phase separations in downstream processing. These benefits could be reaped by growing photosynthetic microorganisms in a mixed culture biofilm at high pH, and by digesting the produced biomass into methane, also at high pH. An overview of the presented process scheme is shown in Figure 1.

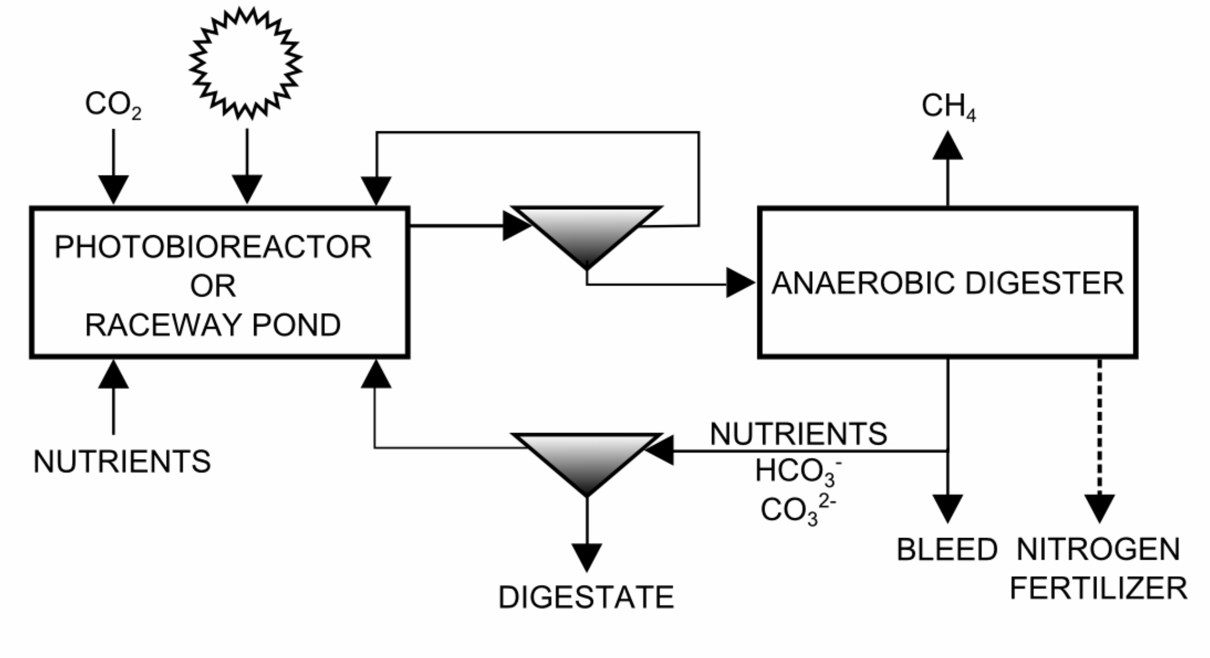

Figure 1. Scheme of the proposed process. Algae are grown either in a photobioreactor or in an open raceway pond. The system's elevated pH enhances the solubility and transfer rate of inorganic carbon. After a contingent dewatering step, the algal suspension is fed to an anaerobic digester that is operated at high pH. Because of the high alkalinity in the digester, the inorganic carbon remains dissolved and the biogas has a high methane concentration. The dissolved

inorganic carbon is recycled to the algae, together with nutrients. The high pH in the anaerobic digester causes an elevated concentration of ammonia, which can potentially be recovered as a fertilizer. A bleed stream will be required to prevent accumulation of precipitates and recalcitrant compounds.

The amount of carbon dioxide that can be dissolved under a given partial pressure remains essentially constant over the entire pH range, but given the chemical equilibria between the different inorganic carbon species, the solubility of (bi)carbonate in equilibrium with a given carbon dioxide partial pressure and concentration increases exponentially with pH (Figure 2). Both halo-alkaliphilic eukaryotic algae and cyanobacteria can use bicarbonate instead of carbon dioxide when the latter becomes growth-limiting at high pH (Goldman, 1999; Kupriyanova and Samylina, 2015; Maberly et al., 2009; Price et al., 2008; Raven, 2010; Smith, 1983). As a consequence, at high pH such organisms could be grown at a bicarbonate concentration that is not limiting, which would increase the volumetric productivity and as a consequence decrease the capital costs. Also, bicarbonate could be supplied as a solution, alleviating the need to bubble carbon dioxide through the growth medium. This would significantly lower the operational costs and the energy requirement related to the supply of compressed carbon dioxide. The growth of algae using bicarbonate has already been suggested by Yoshida et al. (2014) as a means to sequester carbon that is captured from fossil fuel combustion.

Besides the increased solubility of bicarbonate, the high pH will also augment the absorption rate of atmospheric carbon dioxide, because all absorbed/hydrated carbon dioxide immediately reacts with OH^- to form (bi)carbonate (Ebrahimi et al., 2003). Due to this reactive absorption, the carbon dioxide absorption kinetics will be much faster than at neutral pH (Figure 3). As a result, a significant share of the inorganic carbon requirement of the algae could be met by atmospheric carbon dioxide.

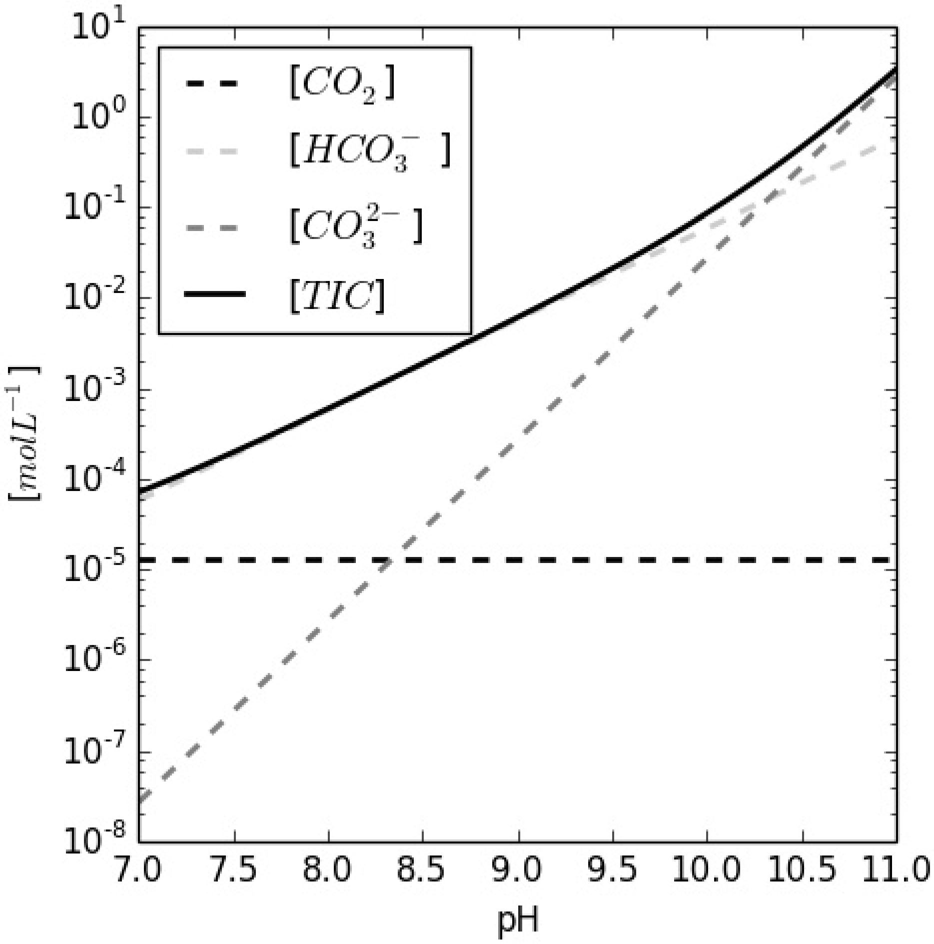

Figure 2. Concentrations of inorganic carbon species carbon dioxide (CO_2), bicarbonate (HCO_3^-), carbonate (CO_3^{2-}) and total inorganic carbon (TIC) as function of pH in equilibrium with an atmospheric carbon dioxide concentration of 380 ppm.

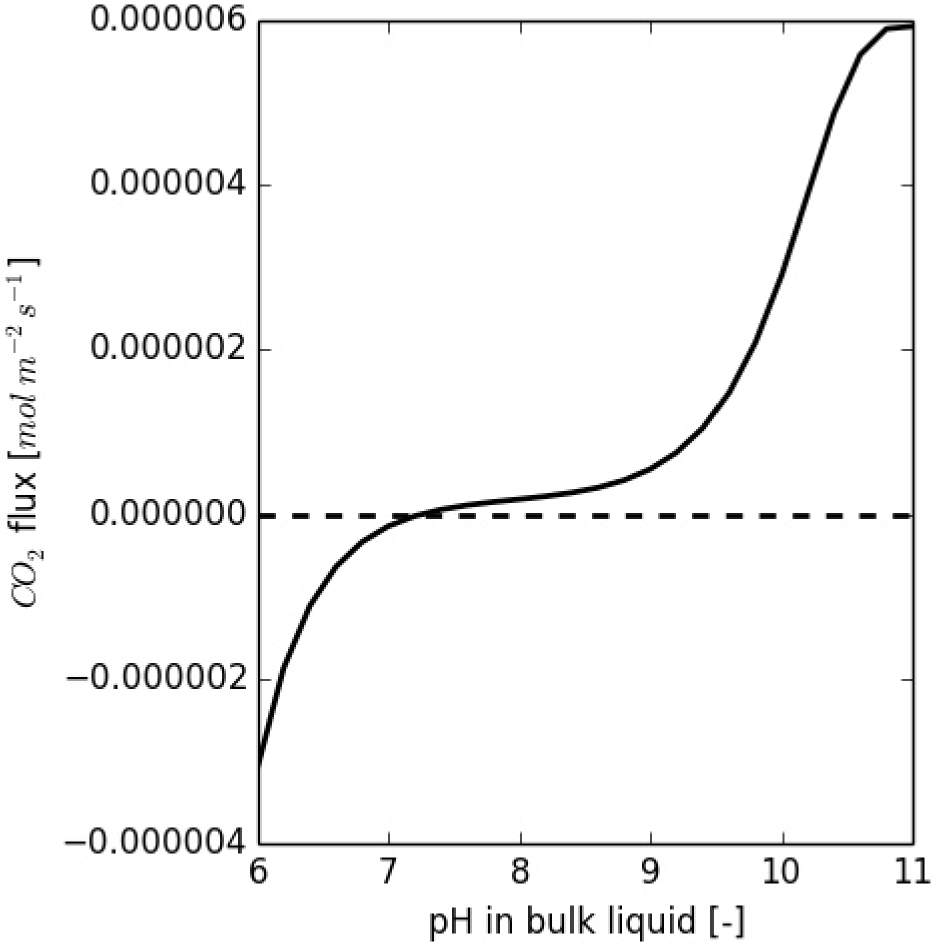

Figure 3. Carbon dioxide transfer rate as function of pH in the bulk liquid. A positive value indicates absorption, a negative value implies desorption. The transfer rate was modelled according to Ebrahimi et al. (2003) using COMSOL. The model assumes a steady state bicarbonate concentration of 0.0001 M in the bulk liquid and an atmospheric carbon dioxide concentration of 380 ppm. The bicarbonate concentration in the bulk is the same order of magnitude as the algae's half-saturation constant (Goldman, 1999).

Since the algal biomass is to be digested, it would be advantageous to have a relatively high C:N ratio in the biomass to avoid process failure due to ammonia accumulation (Weiland, 2010). Microalgae that fulfill this criterion are the ones that are able to produce a large amount of storage polymers (lipids and sugars) or cell wall sugar polymers. The conventional approach to maintain a microalgae culture with a desired property (in this case the ability to produce storage polymers) would be to screen for a specific strain that possesses this characteristic and to grow this in axenic conditions. Yet, maintaining a large scale axenic phototrophic system is expensive and cumbersome (Kupriyanova and Samylina, 2015; Yoshida et al., 2014). Instead, we propose to grow microalgae in an open system and to use selective pressure to enrich algae with a high storage compound production capacity, as described by Mooij et al. (2013). In their approach, Mooij et al. (2013) subjected an open algal culture in a photobioreactor to a cyclical light/dark regime. During the light period carbon dioxide was supplied, while nitrogen was only supplied during the dark period. In the presence of light and carbon dioxide, algae can produce storage polymers but since there is no nitrogen present, they cannot produce any protein. On the other hand, only algae that are able to convert fixed carbon dioxide to storage compounds are able to assimilate nitrogen during the dark period, since nitrogen assimilation consumes energy and carbon (Johnson and Wen, 2010). Since biomass was harvested in every cycle, algae that were not able to store carbon were washed out over time, while carbon storing species were enriched. At steady state, the polyglucose level in the biomass produced by Mooij et al. (2013) was comparable to the highest reported values for pure cultures (57 ± 2 % of volatile suspended solids).

However, it would obviously be a challenge to achieve such a cyclical uncoupling of carbon fixation and nitrogen assimilation in a full-scale system. Indeed, in a large scale cell suspension it would be challenging to supply sufficient ammonia during night time and implement ammonia deprivation upon sunrise. This hurdle could be overcome by growing algae in attached biofilms instead of as a suspension. In such a system, the biofilm would grow on a wetted substrate (submerged or by trickling). By withdrawing and refilling the medium of the submerged biofilm or by changing the stream used for trickling it should be fairly easy to start and stop supplying nitrogen, and thus imposing the cyclical nitrogen deprivation during the daytime. Growing the algae as a biofilm instead of as suspension would also allow to avoid the biomass concentration step that is required for a diluted algae suspension before the biomass is fed to the digester. Harvesting the biofilm would result in a dense algal biomass product without the need for centrifuges or settling tanks. For a trickling system in particular, biofilms would have the additional advantage that the gas-liquid interphase is relatively high, which would benefit the absorption rate of atmospheric carbon dioxide.

In their review article, Christenson and Sims (2011) compared the phototrophic biomass production for suspended and biofilm systems. As far as suspended cultures are concerned, they reported biomass productivities in the range of 10 – 20 $g\ m^{-2} day^{-1}$ for raceway ponds and 20 – 40 $g\ m^{-2} day^{-1}$ for tubular bioreactors. For biofilm systems, they reported slightly lower biomass productivities in the range of 5 – 20 $g\ m^{-2} day^{-1}$. Yet, Johnson and Wen (2010) compared an attached and a suspended algal culture system and found a higher biomass yield for the attached system.

The produced phototrophic biomass is to be digested at high pH which will result in retention of the produced carbon dioxide dissolved in the liquid phase as (bi)carbonate. As a consequence, the biogas has a methane concentration and caloric value similar to natural gas (Figure 4), which makes it possible to feed it directly to the natural gas grid or to use it as a transport fuel without the need for costly upgrading to green gas. Indeed, Nolla-Ardevol et al. (2015) proved that digestion of the microalga *Spirulina* at alkaline conditions (pH 10, 2.0 M Na^+) into methane rich (96 %) biogas was feasible. In our proposal, the (bi)carbonate that remains dissolved in the liquid is recycled to the phototrophic culture. This recycled inorganic carbon provides a part of the carbon requirement for phototrophic growth, alleviating the need for the supply of external carbon dioxide from the atmosphere or stack gas. If the biogas is combusted on-site, e.g. in a combined heat and power system, also the carbon dioxide in the exhaust gas can be recovered with an alkaline scrubber and recycled to the phototrophic component. In theory, this would result in a virtually closed carbon cycle within the system. An additional advantage of operating the anaerobic reactor at high pH is that also hydrogen sulfide remains dissolved as HS^- (pKa of H_2S = 7.0). Therefore, the resulting biogas will be “sweet” without the need for desulfurization

(Nolla-Ardevol et al., 2015). The sulphur, as well as other nutrients, is recycled to the phototrophic reactor (Uggetti et al., 2014). Finally, by performing both the phototrophic growth and the anaerobic digestion at high pH, there is no need to neutralize the pH of the algae broth before feeding it to the digester, thus avoiding a costly pre-treatment step.

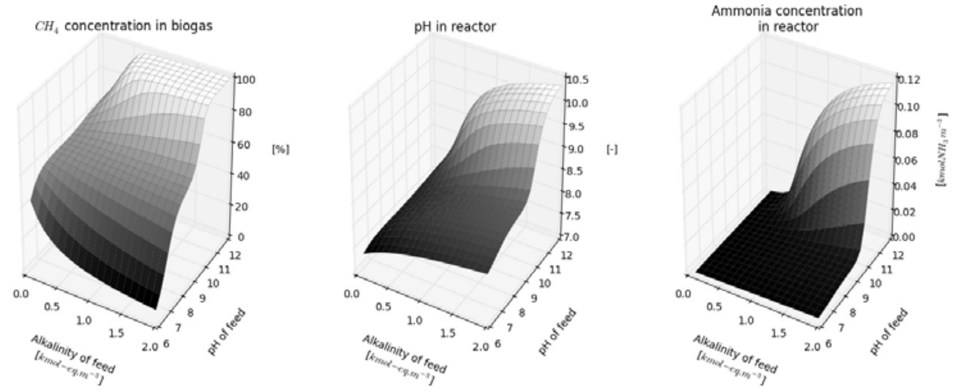

Figure 4. Composition of the biogas, pH in the reactor and ammonia concentration in the reactor resulting from the digestion of algal biomass with composition $C_1H_{2.5}O_{1.0}N_{0.17}$ (Wolf et al., 2007). Digestion was modelled as a first order decay process and stoichiometry was based on elemental balances. The alkalinity in the feed consisted of (bi)carbonate only.

5 Overcoming ammonia toxicity during anaerobic digestion

One of the barriers for the successful application of the proposed process is the elevated ammonia concentration during anaerobic digestion of protein-rich biomass. With an acid dissociation constant of 9.25, most of the ammonium in a digester operated at a pH of 10 – 11 is present as free ammonia (NH_3). Free ammonia is freely membrane-permeable and several intracellular mechanisms have been suggested to explain its toxicity (Gallert et al., 1998). Of all microorganisms involved in anaerobic digestion, the methanogenic organisms are the most susceptible to free ammonia inhibition (Kayhanian, 1994; Koster and Lettinga, 1988).

Several options could be considered to overcome the ammonia inhibition. One possible solution would be to decrease the nitrogen content of the digester feedstock by co-digestion with a cheap feedstock that has a high C:N ratio, such as maize silage, wheat straw or glycerol. Another approach to tackle ammonia inhibition could be to limit the relative nitrogen content of phototrophic biomass itself. In section **Ошибка! Источник ссылки не найден.** it was already explained how Mooij et al. (2013) obtained algal biomass with a high storage polymer content, and consequently a high C:N ratio, using selective pressure. As an alternative to adjusting the C:N ratio of the feedstock, the ammonia could be removed from the digester by stripping and reabsorption into an acidic liquid such as sulfuric acid through a membrane (Lauterböck et al., 2012). The recovered ammonia can be recycled back to the algae cultivation system or it can be used as a fertilizer (ammonium sulfate), which could contribute to the economic feasibility of the entire scheme. A compromised approach, however, may be to operate the digester at a lower pH, somewhere around 9, in order to balance the trade-off between the $CO_2:CH_4$ ratio of the biogas on the one hand, and the concentration of free ammonia in the digester on the other hand.

6 Nutrient supply

The growth of microalgae requires nitrogen and phosphorus. A part of the requirement will be met by the reject medium from the anaerobic digestion process, but still, external sources might be required. Wastewater could be an attractive resource for nutrient supply. Using wastewater would not only decrease the cost of the N and P supply, but it could even have a positive effect on the economic balance since nutrient removal from wastewater in conventional wastewater treatment plants is relatively costly (Zamalloa et al., 2011).

The supply of external nitrogen could be circumvented if the phototrophic organisms would use molecular nitrogen from the atmosphere. Indeed, some cyanobacteria are able to fix molecular nitrogen, using the nitrogenase enzyme complex. Nitrogenase activity in cyanobacteria has been observed in salt marshes (0.55 M salinity) (Carpenter et al., 1978) and alkaline hot spring microbial mats (Steunou et al., 2008). Recent work at the Winogradsky Institute of Microbiology in Moscow revealed nitrogenase activity and actual diazotrophy in vivo in three groups of haloalkaliphilic cyanobacteria dominating in Siberian hypersaline soda lakes. These results suggest that at least part of the nitrogen requirement for algal growth in haloalkaline conditions could potentially be met by photoautotrophic nitrogen fixation. Furthermore, recent work also demonstrated a presence of the nitrogenase in several soda lake primary and secondary anaerobes (Sorokin et al., 2014). Inclusion of the capability for nitrogen fixation in the proposed process could improve the economic feasibility, if the resulting losses in bioreactor productivity would be limited. Besides converting sunlight into biogas, the process would then also convert atmospheric nitrogen to reactive nitrogen that can be recovered as a fertilizer in the digestion process, thus avoiding the economically and energetically costly Haber Bosch process.

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8 References

Acién FG, Fernández JM, Magán JJ, Molina E. Production cost of a real microalgae production plant and strategies to reduce it. Biotechnology Advances 2012; 30: 1344-1353.

Andreote APD, Vaz MGMV, Genuario DB, Barbiero L, Rezende-Filho AT, Fiore MF. NONHETEROCYTOUS CYANOBACTERIA FROM BRAZILIAN SALINE-ALKALINE LAKES. Journal of Phycology 2014; 50: 675-684.

Ballot A, Krienitz L, Kotut K, Wiegand C, Metcalf JS, Codd GA, et al. Cyanobacteria and cyanobacterial toxins in three alkaline rift valley lakes of Kenya - Lakes Bogoria, Nakuru and Elmenteita. Journal of Plankton Research 2004; 26: 925-935.

Ballot A, Krienitz L, Kotut K, Wiegand C, Pflugmacher S. Cyanobacteria and cyanobacterial toxins in the alkaline crater Lakes Sonachi and Simbi, Kenya. Harmful Algae 2005; 4: 139-150.

Christenson L, Sims R. Production and harvesting of microalgae for wastewater treatment, biofuels, and bioproducts. Biotechnology Advances 2011; 29: 686-702.

Collet P, Helias A, Lardon L, Ras M, Goy RA, Steyer JP. Life-cycle assessment of microalgae culture coupled to biogas production. Bioresource Technology 2011; 102: 207-214.

De Schamphelaire L, Verstraete W. Revival of the Biological Sunlight-to-Biogas Energy Conversion System. Biotechnology and Bioengineering 2009; 103: 296-304.

Dismukes GC, Carrieri D, Bennette N, Ananyev GM, Posewitz MC. Aquatic phototrophs: efficient alternatives to land-based crops for biofuels. Current Opinion in Biotechnology 2008; 19: 235-240.

Escobar JC, Lora ES, Venturini OJ, Yanez EE, Castillo EF, Almazan O. Biofuels: Environment, technology and food security. Renewable & Sustainable Energy Reviews 2009; 13: 1275-1287.

Fargione J, Hill J, Tilman D, Polasky S, Hawthorne P. Land clearing and the biofuel carbon debt. Science 2008; 319: 1235-1238.

Gallert C, Bauer S, Winter J. Effect of ammonia on the anaerobic degradation of protein by a mesophilic and thermophilic biowaste population. Applied Microbiology and Biotechnology 1998; 50: 495-501.

Goldman JC. Inorganic carbon availability and the growth of large marine diatoms. Marine Ecology Progress Series 1999; 180: 81-91.

Groom MJ, Gray EM, Townsend PA. Biofuels and biodiversity: Principles for creating better policies for biofuel production. Conservation Biology 2008; 22: 602-609.

Harun R, Davidson M, Doyle M, Gopiraj R, Danquah M, Forde G. Technoeconomic analysis of an integrated microalgae photobioreactor, biodiesel and biogas production facility. Biomass and Bioenergy 2011; 35: 741-747.

Hengeveld EJ, van Gernert WJT, Bekkering J, Broekhuis AA. When does decentralized production of biogas and centralized upgrading and injection into the natural gas grid make sense? Biomass & Bioenergy 2014; 67: 363-371.

IPCC. Climate Change 2007: Impacts, Adaptation and Vulnerability. Contribution of Working Group II to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge, United Kingdom and New York, NY, USA: IPCC, 2007.

Johnson MB, Wen ZY. Development of an attached microalgal growth system for biofuel production. Applied Microbiology and Biotechnology 2010; 85: 525-534.

Kayhanian M. PERFORMANCE OF A HIGH-SOLIDS ANAEROBIC-DIGESTION PROCESS UNDER VARIOUS AMMONIA CONCENTRATIONS. Journal of Chemical Technology and Biotechnology 1994; 59: 349-352.

Kazamia E, Smith AG. Assessing the environmental sustainability of biofuels. Trends in Plant Science 2014; 19: 615-618.

Klass DL. A Long-Range Approach to the Natural Gas Shortage Utilizing Nonfossil Renewable Carbon*. Energy Sources 1977; 3: 177-195.

Koster IW, Lettinga G. ANAEROBIC-DIGESTION AT EXTREME AMMONIA CONCENTRATIONS. Biological Wastes 1988; 25: 51-59.

Kupriyanova EV, Samylina OS. CO2-concentrating mechanism and its traits in haloalkaliphilic cyanobacteria. Microbiology 2015; 84: 112-124.

Maberly SC, Ball LA, Raven JA, Sültemeyer D. INORGANIC CARBON ACQUISITION BY CHRYSOPHYTES1. Journal of Phycology 2009; 45: 1052-1061.

Melack JM, Kilham P. PHOTOSYNTHETIC RATES OF PHYTOPLANKTON IN EAST-AFRICAN ALKALINE, SALINE LAKES. Limnology and Oceanography 1974; 19: 743-755.

Melillo JM, Richmond TC, Yohe GW. Climate Change Impacts in the United States: The Third National Climate Assessment. U.S. Global Change Research Program. 2014: 841.

Mesbah NM, Abou-El-Ela SH, Wiegel J. Novel and unexpected prokaryotic diversity in water and sediments of the alkaline, hypersaline lakes of the wadi an natrun, egypt. Microbial Ecology 2007; 54: 598-617.

Monlau F, Barakat A, Trably E, Dumas C, Steyer J-P, Carrere H. Lignocellulosic Materials Into Biohydrogen and Biomethane: Impact of Structural Features and Pretreatment. Critical Reviews in Environmental Science and Technology 2013; 43: 260-322.

Mooij PR, Stouten GR, Tamis J, van Loosdrecht MCM, Kleerebezem R. Survival of the fattest. Energy & Environmental Science 2013; 6: 3404-3406.

Nolla-Ardèvol V, Strous M, Sorokin DY, Merkel AY, Tegetmeyer HE. Activity and diversity of haloalkaliphilic methanogens in Central Asian soda lakes. Journal of Biotechnology 2012; 161: 167-173.

Nolla-Ardevol V, Strous M, Tegetmeyer HE. Anaerobic digestion of the microalga Spirulina at extreme alkaline conditions: biogas production, metagenome and metatranscriptome. Frontiers in Microbiology 2015; 6.

Norsker N-H, Barbosa MJ, Vermuë MH, Wijffels RH. Microalgal production — A close look at the economics. Biotechnology Advances 2011; 29: 24-27.

Oswald WJ, Golueke CG. Biological transformation of solar energy. Advances in applied microbiology 1960; 2.

Price GD, Badger MR, Woodger FJ, Long BM. Advances in understanding the cyanobacterial CO2-concentrating-mechanism (CCM): functional components, Ci transporters, diversity, genetic regulation and prospects for engineering into plants. Journal of Experimental Botany 2008; 59: 1441-1461.

Raven J. Inorganic carbon acquisition by eukaryotic algae: four current questions. Photosynthesis Research 2010; 106: 123-134.

Reijnders L. Do biofuels from microalgae beat biofuels from terrestrial plants? Trends in Biotechnology 2008; 26: 349-350.

Samylina OS, Sapozhnikov FV, Gainanova OY, Ryabova AV, Nikitin MA, Sorokin DY. Algo-bacterial communities of the Kulunda steppe (Altai Region, Russia) Soda Lakes. Microbiology 2014; 83: 849-860.

Sawayama S, Minowa T, Yokoyama SY. Possibility of renewable energy production and CO2 mitigation by thermochemical liquefaction of microalgae. Biomass and Bioenergy 1999; 17: 33-39.

Schagerl M, Oduor SO. Phytoplankton community relationship to environmental variables in three Kenyan Rift Valley saline-alkaline lakes. Marine and Freshwater Research 2008; 59: 125-136.

Schenk PM, Thomas-Hall SR, Stephens E, Marx UC, Mussgnug JH, Posten C, et al. Second Generation Biofuels: High-Efficiency Microalgae for Biodiesel Production. Bioenergy Research 2008; 1: 20-43.

Searchinger T, Heimlich R, Houghton RA, Dong F, Elobeid A, Fabiosa J, et al. Use of U.S. Croplands for Biofuels Increases Greenhouse Gases Through Emissions from Land-Use Change. Science 2008; 319: 1238-1240.

Seckbach J. Algae and cyanobacteria in extreme environments. Vol 11: Springer, 2007.

Sialve B, Bernet N, Bernard O. Anaerobic digestion of microalgae as a necessary step to make microalgal biodiesel sustainable. Biotechnology Advances 2009; 27: 409-416.

Singh A, Nigam PS, Murphy JD. Renewable fuels from algae: An answer to debatable land based fuels. Bioresource Technology 2011; 102: 10-16.

Slade R, Bauen A. Micro-algae cultivation for biofuels: Cost, energy balance, environmental impacts and future prospects. Biomass and Bioenergy 2013; 53: 29-38.

Smith AJ. Modes of cyanobacterial carbon metabolism. Annales de l'Institut Pasteur / Microbiologie 1983; 134: 93-113.

Sorokin DY, Abbas B, Geleijnse M, Pimenov NV, Sukhacheva MV, van Loosdrecht MCM. Methanogenesis at extremely haloalkaline conditions in the soda lakes of Kulunda Steppe (Altai, Russia). Fems Microbiology Ecology 2015a; 91.

Sorokin DY, Banciu HL, Muyzer G. Functional microbiology of soda lakes. Current opinion in microbiology 2015b; 25: 88-96.

Sorokin DY, Berben T, Melton ED, Overmars L, Vavourakis CD, Muyzer G. Microbial diversity and biogeochemical cycling in soda lakes. Extremophiles 2014; 18: 791-809.

Sorokin DY, Kuenen JG. Chemolithotrophic halo alkaliphiles from soda lakes. Fems Microbiology Ecology 2005; 52: 287-295.

Sorokin DY, Kuenen JG, Muyzer G. The microbial sulfur cycle at extremely haloalkaline conditions of soda lakes. Frontiers in Microbiology 2011; 2.

Stephens E, Ross IL, King Z, Mussgnug JH, Kruse O, Posten C, et al. An economic and technical evaluation of microalgal biofuels. Nat Biotech 2010; 28: 126-128.

Taherzadeh MJ, Karimi K. Pretreatment of lignocellulosic wastes to improve ethanol and biogas production: A review. International Journal of Molecular Sciences 2008; 9: 1621-1651.

Uduman N, Qi Y, Danquah MK, Forde GM, Hoadley A. Dewatering of microalgal cultures: A major bottleneck to algae-based fuels. Journal of Renewable and Sustainable Energy 2010; 2: -.

Vergara-Fernández A, Vargas G, Alarcón N, Velasco A. Evaluation of marine algae as a source of biogas in a two-stage anaerobic reactor system. Biomass and Bioenergy 2008; 32: 338-344.

Weiland P. Biogas production: current state and perspectives. Applied Microbiology and Biotechnology 2010; 85: 849-860.

Wijffels RH. Potential of sponges and microalgae for marine biotechnology. Trends in Biotechnology 2008; 26: 26-31.

Wolf G, Picioreanu C, van Loosdrecht MCM. Kinetic modeling of phototrophic biofilms: The PHOBIA model. Biotechnology and Bioengineering 2007; 97: 1064-1079.

Yang LC, Ge XM, Wan CX, Yu F, Li YB. Progress and perspectives in converting biogas to transportation fuels. Renewable & Sustainable Energy Reviews 2014; 40: 1133-1152.

Yoshida H, Mønster J, Scheutz C. Plant-integrated measurement of greenhouse gas emissions from a municipal wastewater treatment plant. Water Research 2014; 61: 108-118.

Zamalloa C, Vulsteke E, Albrecht J, Verstraete W. The techno-economic potential of renewable energy through the anaerobic digestion of microalgae. Bioresource Technology 2011; 102: 1149-1158.

Zavarzin GA, Zhilina TN, Kevbrin VV. The alkaliphilic microbial community and its functional diversity. Microbiology 1999; 68: 503-521.

Zheng M, Li X, Li L, Yang X, He Y. Enhancing anaerobic biogasification of corn stover through wet state NaOH pretreatment. Bioresource Technology 2009; 100: 5140-5145.