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## Scaling Up Alternative Energy

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### PERSPECTIVE

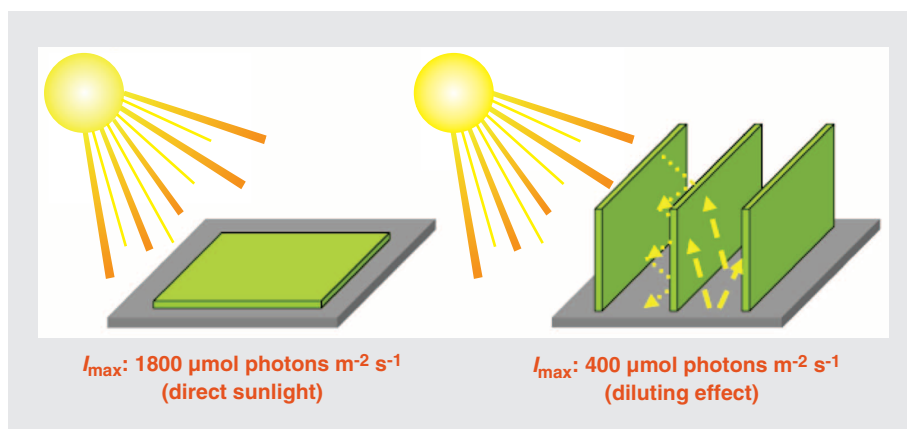
## An Outlook on Microalgal Biofuels

René H. Wijffels<sup>1</sup> and Maria J. Barbosa<sup>2</sup>

Microalgae are considered one of the most promising feedstocks for biofuels. The productivity of these photosynthetic microorganisms in converting carbon dioxide into carbon-rich lipids, only a step or two away from biodiesel, greatly exceeds that of agricultural oleaginous crops, without competing for arable land. Worldwide, research and demonstration programs are being carried out to develop the technology needed to expand algal lipid production from a craft to a major industrial process. Although microalgae are not yet produced at large scale for bulk applications, recent advances—particularly in the methods of systems biology, genetic engineering, and biorefining—present opportunities to develop this process in a sustainable and economical way within the next 10 to 15 years.

The concept of using algae to make fuels was already being discussed 50 years ago (1), but a concerted effort began with the oil crisis in the 1970s. Large research programs in Japan and the United States focused on developing microalgal energy production systems. From 1978 to 1996, the U.S. Department of Energy's Office of Fuels Development funded a program to develop renewable transportation fuels from algae (2). The main focus of the program, known as the Aquatic Species Program (ASP), was the production of biodiesel from high-lipid-content algae grown in ponds, using waste CO<sub>2</sub> from coal-fired

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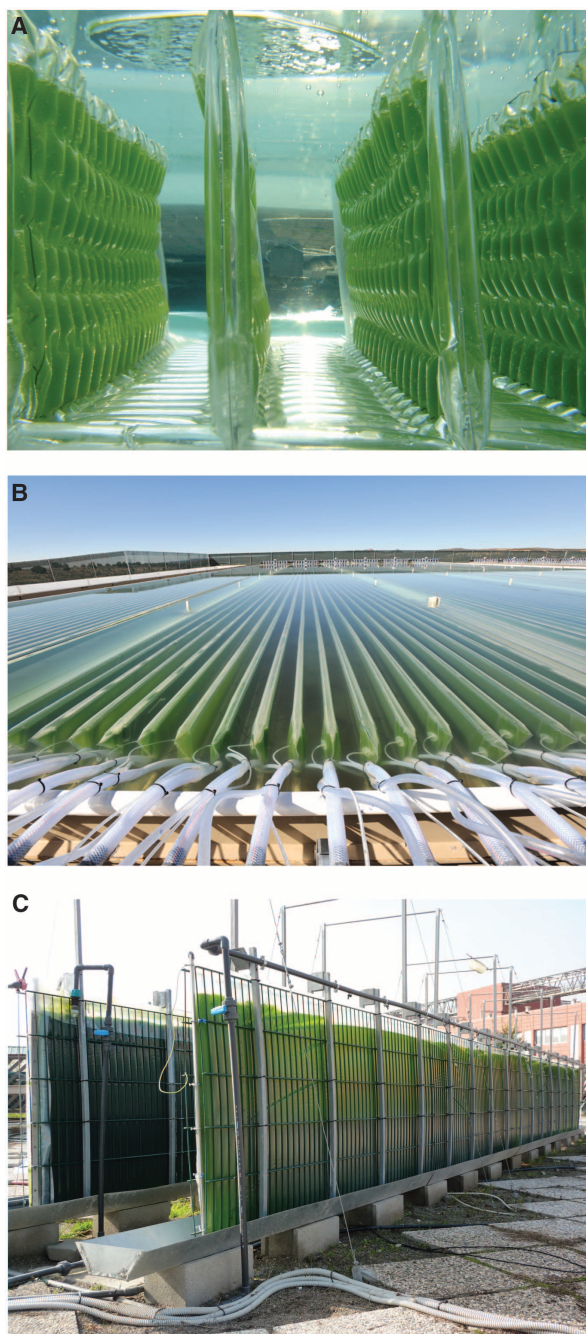
**Fig. 1.** The principle of light dilution. The light intensity ( $I$ ) striking closely spaced vertical panels is much lower than the intensity striking a horizontal reactor on the same surface.

biofuels, the food versus fuel debate and a virtual explosion in biotechnology resulted in a substantial commitment to the development of algal biofuels by the industrial and governmental sectors. The recent investments in microalgae for fuels are well justified by the potential that these microorganisms offer through their higher lipid productivities per ground area than oleaginous agricultural crops, as well as lack of competition for arable land.

Faced with stresses such as nutrient deprivation, algae store chemical energy in the form of oils such as neutral lipids or triglycerides (4). The algal oil can be extracted from the organisms and converted into biodiesel by transesterification with short-chain alcohols (5) or by hydrogenation of fatty acids into linear hydrocarbons (6). Algae also synthesize other fuel products, such as hydrogen (7), ethanol (8) and long-chain hydrocarbons, that resemble crude oil (9), or the algal biomass can be converted to biogas through anaerobic fermentation (1).

Despite this potential, the production capacity for microalgae is presently limited in comparison to land-based energy crops. The current worldwide microalgal manufacturing infrastructure (producing the equivalent of ~5000 tons of dry algal biomass) is devoted to extraction of high-value products such as carotenoids and  $\omega$ -3 fatty acids used for food and feed ingredients. The total market volume is €1.25 billion, implying an average market price for microalgae of €250/kg dry biomass (10). As an example for comparison with land-based oleaginous crops, the world production of palm oil is nearly 40 million tons, with a market value of ~0.50 €/kg (11).

Production of microalgae for biofuels needs to take place on a much larger scale at much lower costs. If all transport fuels were to be replaced by biodiesel in Europe, there would be an annual need for nearly 0.4 billion  $m^3$  (12). If this biodiesel were to be supplied through microalgae, 9.25 million ha (almost the surface area of Portugal) would be needed to supply the European market, assuming a productivity of 40,000 liters per ha per year. This productivity is based on a 3% solar energy conversion to biomass (theoretical maximum is 9%) and a biomass oil content of 50%, under the solar conditions of Portugal. A leap in the development of microalgae technology is therefore required; on a practical level, the scale of production needs to increase at least 3 orders of magnitude, with a concomitant decrease in the cost of production by a factor of 10. In the past few years, there has been a rather polarized debate



**Fig. 2.** Development of low-cost photobioreactors. Flat-panel reactors from (A) Proviron, Belgium [reproduced with permission from (29)], (B) Solix Biofuels, USA (reproduced with permission from *Biomass Magazine*), and (C) Green Wall Panel reactors from Fotosintetica & Microbiologica S.r.l., Italy.

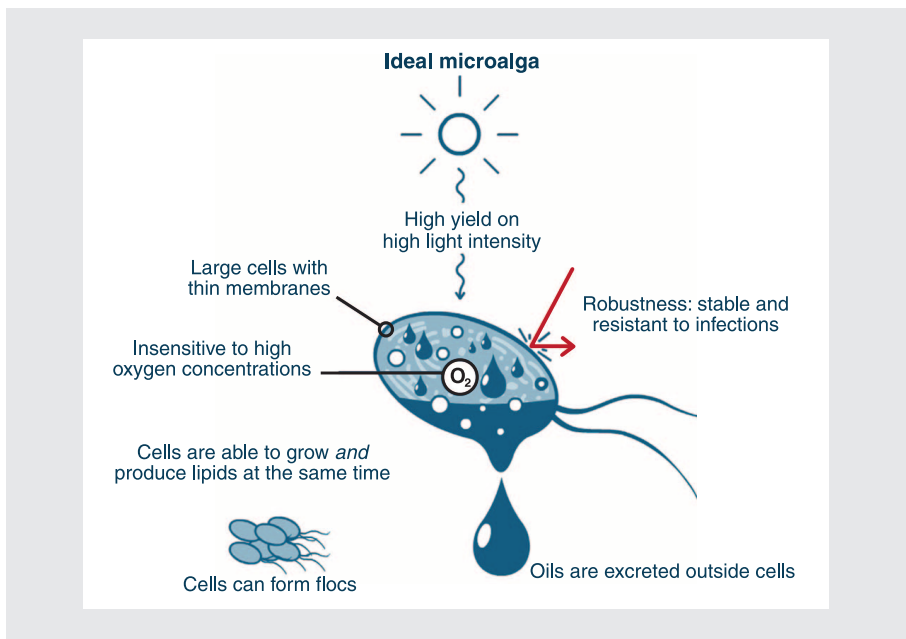
between researchers in the field over technology readiness and the prospects for productivity enhancement, with some parties pressing for scale-up and commercialization now, while others cautiously stress the need for additional research leading to more careful step-by-step development (13).

We believe a multidisciplinary approach will be required to realize the full potential of microalgae as a biofuels feedstock. A comprehensive

research portfolio should cover the whole chain of process development in an integrated and iterative way, including fundamental biology, systems biology, metabolic modeling, strain development, bioprocess engineering, scale-up, biorefineries, integrated production chain, and the whole system design, including logistics (14). The main objective is to reduce production costs and energy requirements while maximizing lipid productivity and to increase the biomass value by making use of all algal biomass components. For cost and energy reduction and maximization of lipid productivity cell properties, bioreactor design, efficiency in supply, and use of nutrients and resources need to be improved, and to make use of all biomass ingredients, a biorefinery infrastructure needs to be established.

In microbial fermentations, substantial improvements in productivity have been obtained through both technological (reactor design, process control, harvesting, and extraction) and strain improvements. As an example, the present productivity of penicillin synthesis by fungi is 5000 times as high as it was 50 years ago (15). Commercial production of microalgae is still based on traditional technologies using a few strains. There are many more species yet to explore; in addition, genetic engineering offers the possibility for strain improvement. High lipid productivity is essential for the commercial production of biodiesel from microalgae. In nonstressed growing algae, lipids are mostly present in the form of phospholipids in the cell membranes. Some microalgae, when exposed to stress conditions (e.g., nutrient deprivation or high light intensities), accumulate lipids in the form of triacylglycerols in so-called oil bodies. This accumulation occurs at the expense of energy used for growth, leading to a decrease in growth rate and a consequent decrease in productivity. Knowledge of the biosynthesis mechanism of triacylglycerols and their accumulation in oil bodies is limited and often based on analogies with higher plants (4). If the mechanism were known, it could open the possibility of inducing lipid accumulation in oil bodies without having to apply a stress factor. More broadly, detailed insight into metabolic pathways may lead to strategies to induce lipid accumulation based on process conditions, defined nutrient regimens, and/or the use of metabolic engineering techniques. For this purpose, well-annotated genomes need to be available.

There are still very few algae for which full or near-full genome sequences have been obtained



**Fig. 3.** The ideal photosynthetic cell factory for production of biofuels.

[e.g., *Chlamydomonas reinhardtii* (16), *Thalassiosira pseudonana* (17), and *Phaeodactylum tricornutum* (18)], and transfection systems have barely been developed. Currently, there are about 10 different algal species that can be transformed (19). However, sophisticated metabolic engineering, whereby several genes are overexpressed or down-regulated in a single organism, is currently only really possible with *C. reinhardtii*. We expect that the genome of more algal strains will be sequenced in the near future, due to the present high interest in the field, enlargement of the scientific community, and the availability of fast and reliable technologies for genome sequencing.

The main inputs required in addition to the algae themselves are sunlight, water, CO<sub>2</sub>, nitrogen, and phosphorus. Large-scale cultivation of microalgae for biofuel production must be based on sunlight as the sole source of light energy. Especially when working in summertime, and/or when working at lower latitudes, sunlight intensities are high and often oversaturate the photosynthetic cycle, limiting growth and leading to a drop in productivity. In recent years, much effort was put into increasing photosynthetic efficiency of microalgae under oversaturating light (the normal condition on a sunny day) by developing new strains with smaller antenna sizes (20) and by decreasing the light path of photobioreactors while increasing mixing (turbulence) in high cell density cultures (21, 22). Turbulence requires high energy input and therefore is not suitable for large-scale production of biofuels from microalgae. One strategy to obtain high photosynthetic efficiencies under bright sunlight in systems with lower energy requirements is to reduce the light intensity at the reactor surface.

This can be done by stacking the reactor units vertically (Fig. 1): Narrow spacing in the stacks minimizes loss of light to the ground surface (21). However, if not combined with a short light path, this setup leads to voluminous reactor systems with low volumetric productivity and low biomass concentration (23). To reduce investment costs of these systems, vertical panels can be made from thin plastic films such as polyethylene (Fig. 2). There are examples of thin film systems submerged in large water volumes for good temperature control and a lower associated energy requirement for cooling (14). We expect that in the coming years many systems will be developed based on these design principles (Fig. 2). Improvements are to be expected in material lifetime (polyethylene has a lifetime of ~1 year), ease of cleaning, and energy requirements (for example, the energy requirement for cooling can be further reduced by reflecting the near-infrared portion of the light incident on the reactor surface, which otherwise heats the system without contributing to photosynthesis).

Water usage is another important parameter. For the production of 1 liter of biofuel from fuel crops, approximately 10,000 liters of water are needed (24). Microalgae need much less water. For photosynthesis alone, ~0.75 liter of water is needed per kg of biomass produced (25). Per liter of biofuel, assuming a lipid content of 50%, 1.5 liters of water are required. In practice, water use in production systems is much larger because water is also used for cooling closed systems, and fresh water needs to be added to open ponds to compensate for evaporation. If closed systems are used and cooled with a large saltwater buffer via heat exchangers, freshwater usage can

be reduced considerably. Microalgae can also be grown on seawater; even deserts would be suitable if there is access to salt aquifers. Growth could also take place in confined systems on large water surfaces such as lakes or seas, assuming there is adequate protection from the wind.

The production of large quantities of biomass also requires a large amount of CO<sub>2</sub>. A total of 1.8 tons of CO<sub>2</sub> is needed to produce 1 ton of algal biomass (25). This means that 1.3 billion tons of CO<sub>2</sub> would be required for the production of 0.4 billion m<sup>3</sup> of biodiesel to supply the European transportation market. The European Union produces about 4 billion tons of CO<sub>2</sub>, (26) so production of microalgae could go some way toward relieving this CO<sub>2</sub> excess. However, the distance across which CO<sub>2</sub> may need to be transported in this context is a matter of concern.

The main nutrients needed for the production of microalgae are nitrogen and phosphorus. The biomass of the algae consists of 7% nitrogen and 1% phosphorus. Consequently, for the European biofuel market ~25 million tons of nitrogen and 4 million tons of phosphorus are needed. This is about twice the amount that is presently produced as fertilizer in Europe (27). For sustainable production of biodiesel from microalgae, it will be important to make use of residual nutrient sources (about 8 million tons of nitrogen in Europe) and to recycle nutrients as much as possible.

After production, the biomass needs to be harvested, the lipids extracted, and the remaining cell components recovered. Harvesting of microalgae is currently expensive because of the high energy requirements and capital costs involved. Because most microalgae are small individual cells, centrifugation is often used as a preferred harvesting method. However, as the biomass concentration is generally low (<3 g/L), centrifugation of diluted streams requires a large capacity of the centrifuge, which makes the process energy-demanding and expensive. Flocculation, followed by sedimentation and flotation, before centrifugation or filtration will substantially reduce harvesting costs and energy requirements. Ideally, algae would flocculate spontaneously at a certain stage of the process. After harvesting, the oil needs to be extracted. The cells are first disrupted, and then the oil can be extracted with organic solvents or with more environmentally benign, but more expensive, solvents (e.g., supercritical CO<sub>2</sub>). Most microalgae strains are, in general, relatively small and have a thick cell wall. For this reason, very harsh conditions need to be used (e.g., mechanical, chemical, and physical stress) to break the cells for extraction of the products, which may affect the functionality of cell compounds like proteins. Excretion of the oils, in a manner similar to what naturally occurs in the microalgae *Botryococcus braunii*, will lead to a simplified biorefinery and improve the downstream economics. However, it will not provide a complete solution because the remaining

cell components still need to be recovered from the cells. Thin cell membranes, strong enough to prevent shear damage during production, would facilitate cell disruption. Small spherical cells with a thick cell wall, like *Nannochloropsis*, are clearly not ideal fuel sources for this reason.

Currently, some of the desired individual characteristics mentioned above and depicted in Fig. 3 can be found in specific strains but never combined in one ideal strain. In addition, strain characteristics need to be integrated with reactor design principles. For example, if a strain has the capacity to efficiently convert high light intensities into biomass, light dilution will no longer be necessary. If strains are developed that can tolerate high oxygen concentration, the length of tubes in tubular photobioreactors is no longer limited and degassing is no longer required.

Economically feasible production of microalgae for biofuels will only be achieved if combined with production of bulk chemicals, food, and feed ingredients. Despite algae's high suitability for biorefining due to the varied composition of its biomass, the coproduction of multiple products from microalgae remains a challenge. Research is needed to explore mild cell disruption, extraction, and separation technologies that retain the functionality of the different cell components (e.g., proteins, carbohydrates,  $\omega$ -3 fatty acids, pigments, and vitamins). Biorefining is important not only for cost efficiency but also for the supply of food compounds. The algal biomass that could theoretically supply 0.4 billion m<sup>3</sup> of biodiesel to the European market consists of 40% protein; thus, the total amount of protein produced as a by-product would exceed 0.3 billion tons. This is about 40 times as much as the amount of soy protein (18 million tons of soy beans with ~40% of proteins in 2008) presently imported into Europe (11). Therefore, the production of microalgae for fuels would place no pressure on the availability of rich agricultural areas for production of proteins; on the contrary, there is even the possibility of an overproduction of proteins.

In closing, we reiterate our belief that 10 to 15 years is a reasonable projection for the development of a sustainable and economically viable process for the commercial production of biofuels from algal biomass. It is important that further technology development be done in close collaboration with environmental scientists. The production processes should be subjected to life-cycle analysis (28) and monitored for their impact on biodiversity.

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#### REVIEW

## Generating the Option of a Two-Stage Nuclear Renaissance

Robin W. Grimes<sup>1</sup> and William J. Nuttall<sup>2</sup>

Concerns about climate change, security of supply, and depleting fossil fuel reserves have spurred a revival of interest in nuclear power generation in Europe and North America, while other regions continue or initiate an expansion. We suggest that the first stage of this process will include replacing or extending the life of existing nuclear power plants, with continued incremental improvements in efficiency and reliability. After 2030, a large-scale second period of construction would allow nuclear energy to contribute substantially to the decarbonization of electricity generation. For nuclear energy to be sustainable, new large-scale fuel cycles will be required that may include fuel reprocessing. Here, we explore the opportunities and constraints in both time periods and suggests ways in which measures taken today might, at modest cost, provide more options in the decades to come. Careful long-term planning, along with parallel efforts aimed at containing waste products and avoiding diversion of material into weapons production, can ensure that nuclear power generation remains a carbon-neutral option.

In North America and Europe, the development of nuclear power stalled after the March 1979 Three Mile Island accident in Pennsylvania, and until recently the building of additional nuclear reactors was not likely. Yet today, a nuclear renaissance is underway, and globally 52 reactors are under construction (1). How nuclear energy found itself in a state of decline is well documented. Will it continue to move forward and avoid another collapse?

In this article, we assess technological responses and opportunities for nuclear generation

technology on two time scales (Fig. 1): first, those of immediate concern and consequence, and second, matters that will dominate in the longer term (beyond about 2030), when nuclear development could once more stall. The immediate future also indicates continued growth of nuclear energy in the Middle East, East Asia, South Asia, and elsewhere.

If the global electricity system is to be largely decarbonized over the first half of this century, then two key challenges must also be surmounted. One will be to develop civil nuclear programs in all parts of the world without risking the proliferation of nuclear weapons technologies (2). The other will be to deal with nuclear waste in as safe a manner as possible. Settling on policy options has proved extremely difficult in many

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# ERRATUM

*Post date 12 November 2010*

**Perspectives:** "An outlook on microalgal biofuels" by R. H. Wijffels and M. J. Barbosa (13 August, p. 796). The text attributed the following parameter to C. de Fraiture, M. Giordano, Y. Liao, *Water Pol.* **10** (suppl. 1), 67 (2008): "For the production of 1 liter of biofuel from fuel crops, approximately 10,000 liters of water are needed." In fact, the de Fraiture paper states: "It takes on average roughly 2,500 [liters] of crop evapotranspiration and 820 [liters] of irrigation water withdrawn to produce one liter of biofuel. But regional variation is large."