

# Cyanobacterial Biodiesel: Tubes in the Desert

<http://biofuels.asu.edu/tubes.shtml>

Corn-based ethanol or soy/palm oil-based biodiesel are inherently limited in terms of per-hectare yields and suitable land area for development. However, algae and photosynthetic bacteria overcome these constraints for biofuels by providing a roughly 100-fold advantage in yield (Table 1) and not requiring farmland for production (i.e., they do not compete with food crops). A major ongoing ASU effort, funded in its initial pilot stages by BP and Science Foundation Arizona, generates biodiesel from lipid produced by the photosynthetic cyanobacterium *Synechocystis*. Cyanobacteria are much more amenable to metabolic engineering to improve biofuel productivity than are eukaryotic algae. The genome of *Synechocystis* has been fully sequenced, and the microorganism provides a facile substrate for genetic modification of metabolic pathways to optimize yields of C-16 and C-18 lipids for biodiesel production. In fact, much progress has already been made at ASU in increasing the yields of these lipids through genetic engineering. The current projection is for a proof of concept demonstration of the industrial scale utility of this approach in Calendar Year 2008. Beyond this demonstration, a larger field test bed will be built to refine the approach and enable industrial pilot scale efforts in the 2010 timeframe.

This platform for renewable solar energy-to-biofuels conversion combines innovative metabolic engineering with state-of-the-art, large-scale bioprocess engineering, efficient cell harvesting, cost-effective conversion of lipid to biodiesel, and generation of other valuable byproducts. This is possible because *Synechocystis* is fast growing and robust in accommodating diverse environmental conditions. It can be cultivated over a wide range of salt and fixed-nitrogen concentrations and at CO<sub>2</sub> levels up to 5 percent. The system also requires minimal water consumption. These traits make the microorganism well suited for growth using flue gas effluent from power plants as a carbon source (recapturing the carbon

dioxide from the plant before release into the atmosphere) and using agricultural run-off water contaminated with nitrogenous fertilizer as a fixed-nitrogen source when it is available. When N-contaminated water is not used, fixed nitrogen can be recycled so little new nitrogen will need to be added.

This renewable solar energy-to-biofuels approach is very well suited to arid regions with high levels of sunlight, and Central Arizona is ideal for this purpose. Biofuel production from cyanobacterial photobioreactors should be scalable to a point where it represents a major source of carbon-neutral fuel for the United States, as well as high-quality employment and overall economic growth in the State.

Organism	Lipid production (L ha <sup>-1</sup> yr <sup>-1</sup> )
Photosynthetic microbes	72,000-130,000
Sunflower	570-1,030
Soybean	380-650

**Table 1.** Comparison of lipid production ranges per hectare per year for microalgae and oil-producing plants.

In order to demonstrate the feasibility of biofuel production, our current project involves the production of laboratory-scale photobioreactors, while simultaneously designing and implementing a rooftop photobioreactor, where we will then apply mathematical modeling tools for systems analysis. We plan to address issues associated with bioreactor scale-up prior to introducing the improved strains and equipment into the large-scale field test bed bioreactor for final validation. The current plan involves scaling to a point where, in two years time, we will have designed and fabricated a field-scale bioreactor. This will allow our laboratory-scale organism optimization to be evaluated for suitability in larger scale bioprocess production under "real-world conditions." The test-bed photobioreactor will be located at an APS power plant close to the ASU-Tempe Campus. This location will provide a secure site and will enable engineering assessment of operating the photobioreactor system using flue gas and water recycling from the power plant for biomass production. The proposed research—with its coordination of genetic improvement, testing

at the pilot scale, and industry partners—will create a unique setting in which dramatic advances can be realized in a relatively short time.

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## Algal-Based Biofuels and Biomaterials

<http://biofuels.asu.edu/biomaterials.shtml>

Eukaryotic microalgae also represent a promising alternative renewable source of feedstock for biofuel production. With over 40,000 identified species, microalgae are one of the more diverse groups of organisms on Earth. They naturally produce large quantities of many biomaterials, including lipids/oil. Nature has had ca. 4 billion years to engineer strains with unique abilities to grow robustly in diverse environmental conditions and evolve unique metabolic characteristics such as intracellular lipid storage, with a growth potential an order of magnitude greater than terrestrial crop plants due to their extraordinarily efficient light and nutrient utilization. The exploitation of naturally occurring photosynthetic microalgae, collected and isolated over the past 25 years for production of renewable liquid fuels, initially in collaboration with the Aquatic Species Program of the DOE, and subsequently at Arizona State University, provides a green and renewable resource of feedstock biomass to meet increasing energy needs and especially the demand for liquid fuels. Over the past two decades, our algal-based biofuel research has progressed from screening and evaluation of naturally occurring algal strains that exhibit high growth rate and high oil content to genetic improvement of selected strains for robustness in performance under diverse environmental and culture conditions, to large-scale photobioreactor design and optimization, to outdoor mass culture and downstream processing (i.e., harvesting, dewatering, and drying), to algal oil extraction, pretreatment, and oil conversion to biodiesel and jet fuel, to systems/process scale-up analysis, and life cycle assessment. One of the field demonstration photobioreactors developed for algae feedstock production is shown



Challenges to be addressed include refinement of the cultivation process, downstream processing of biomass, and development of an economic feasibility model for commercialization of algae-based biofuels and biomaterials.

The microalgal research and development effort couples the use of microalgae for biofuels production with environmental bioremediation. Microalgae naturally remove and recycle nutrients (such as nitrogen and phosphorous) from water and wastewater and carbon dioxide from flue-gases emitted from fossil fuel-fired power plants, providing an added environmental benefit. The integration of wastewater bioremediation and carbon sequestration with biofuel production in a novel field-scale bioreactor has been demonstrated. Although algal biomass residues derived from the oil extraction process can be used for animal feed or fertilizer, we are currently exploring, in collaboration with our industrial partners, the opportunity for using biomass residues to produce ethanol, and methane, and high-value biomaterials, such as biopolymers, carotenoids, and very long-chain polyunsaturated fatty acids. Collaboration with industrial partners to provide flue gas (APS), animal wastewater (United Dairyman of Arizona), commercial algal feedstock production capabilities (PetroAlgae), technical assistance with conversion of algae oil to biofuels (UOP and Honeywell Aerospace Division), and assistance with marketing of algal feedstock (Cargill) has either been initiated or is on-going.