Recovery and Sequestration of CO₂ from Stationary Combustion Systems by Photosynthesis of Microalgae

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Abstract

Most of the anthropogenic emissions of carbon dioxide result from the combustion of fossil fuels for energy production. Photosynthesis has long been recognized as a means, at least in theory, to sequester anthropogenic carbon dioxide. Aquatic microalgae have been identified as fast growing species whose carbon fixing rates are higher than those of land-based plants by one order of magnitude. Physical Sciences Inc. (PSI), Aquasearch, and the Hawaii Natural Energy Institute at the University of Hawaii are jointly developing technologies for recovery and sequestration of CO₂ from stationary combustion systems by photosynthesis of microalgae. The research is aimed primarily at demonstrating the ability of selected species of microalgae to effectively fix carbon from typical power plant exhaust gases.

This report covers the reporting period 1 July to 30 September 2003 in which PSI, Aquasearch and University of Hawaii conducted their tasks. Based on the work during the previous reporting period, Aquasearch and PSI continued preparation work on direct feeding of coal combustion gas to microalgae. Aquasearch started the first full scale carbon sequestration tests with propane combustion gases. Aquasearch started to model the costs associated with biomass harvest from different microalgal strains. University of Hawaii continued effort on system optimization of the CO₂ sequestration system.

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1. Introduction

Emissions of carbon dioxide are predicted to increase in this century¹ leading to increased concentrations of carbon dioxide in the atmosphere. While there is still much debate on the effects of increased CO₂ levels on global climate, many scientists agree that the projected increases could have a profound effect on the environment. Most of the anthropogenic emissions of carbon dioxide result from the combustion of fossil fuels for energy production. It is the increased demand for energy, particularly in the developing world, which underlies the projected increase in CO₂ emissions. Meeting this demand without huge increases in CO₂ emissions requires more than merely increasing the efficiency of energy production. Carbon sequestration, capturing and storing carbon emitted from the global energy system, could be a major tool for reducing atmospheric CO₂ emissions from fossil fuel usage.

The costs of removing CO₂ from a conventional coal-fired power plant with flue gas desulfurization were estimated to be in the range of \$35 to \$264 per ton of CO₂.² The cost of power was projected to increase by anywhere from 25 to 130 mills/kWh. DoE's goal is to reduce the cost of carbon sequestration to below \$10/ton of avoided net cost.

Photosynthesis has long been recognized as a means, at least in theory, to sequester anthropogenic carbon dioxide. There has been relatively little research aimed at developing the technology to produce a gaseous combustion effluent that can be used for photosynthetic carbon sequestration. However, the photosynthetic reaction process by plants is too slow to significantly offset the point source emissions of CO₂ within a localized area. Aquatic microalgae have been identified as fast growing species whose carbon fixing rates are higher than those of land-based plants by one order of magnitude.

The Department of Energy has been sponsoring development of large-scale photovoltaic power systems for electricity generation. By this analogy, a large-scale microalgae plantation may be viewed as one form of renewable energy utilization. While the PV array converts solar energy to electricity, the microalgae plant converts CO₂ from fossil combustion systems to stable carbon compounds for sequestration and high commercial value products to offset the carbon sequestration cost. The solar utilization efficiency of some microalgae is ~ 5%, as compared to ~ 0.2% for typical land based plants. Furthermore, a dedicated photobioreactor for growth of microalgae may be optimized for high efficiency utilization of solar energy, comparable to those of some photovoltaic cells. It is logical, therefore, that photosynthetic reaction of microalgae be considered as a mean for recovery and sequestration of CO₂ emitted from fossil fuel combustion systems.

Stationary combustion sources, particularly electric utility plants, represent 35% of the carbon dioxide emissions from end-use of energy in the United States.¹ The proposed process addresses this goal through the production of high value products from carbon dioxide emissions. Microalgae can produce high-value pharmaceuticals, fine chemicals, and commodities. In these markets, microalgal carbon can produce revenues of order \$100,000 per kg C. These markets are currently estimated at >\$5 billion per year, and projected to grow to >\$50 billion per year within the next 10 to 15 years. Revenues can offset carbon sequestration costs.

An ideal methodology for photosynthetic sequestration of anthropogenic carbon dioxide has the following attributes:

1. Highest possible rates of CO₂ uptake
2. Mineralization of CO₂, resulting in permanently sequestered carbon
3. Revenues from substances of high economic value
4. Use of concentrated, anthropogenic CO₂ before it is allowed to enter the atmosphere.

In this research program, Physical Sciences Inc. (PSI), Aquasearch, and the Hawaii Natural Energy Institute at the University of Hawaii are jointly developing technologies for recovery and sequestration of CO₂ from stationary combustion systems by photosynthesis of microalgae. The research we propose is aimed primarily at quantifying the efficacy of microalgae-based carbon sequestration at industrial scale. Our principal research activities will be focused on demonstrating the ability of selected species of microalgae to effectively fix carbon from typical power plant exhaust gases. Our final results will be used as the basis to evaluate the technical efficacy and associated economic performance of large-scale carbon sequestration facilities.

Our vision of a viable strategy for carbon sequestration based on photosynthetic microalgae is shown conceptually in Figure 1. In this figure, CO₂ from the fossil fuel combustion system and nutrients are added to a photobioreactor where microalgae photosynthetically convert the CO₂ into compounds for high commercial values or mineralized carbon for sequestration. The advantages of the proposed process include the following:

1. High purity CO₂ gas is not required for algae culture. It is possible that flue gas containing 2~5% CO₂ can be fed directly to the photobioreactor. This will simplify CO₂ separation from flue gas significantly.
2. Some combustion products such as NO_x or SO_x can be effectively used as nutrients for microalgae. This could simplify flue gas scrubbing for the combustion system.
3. Microalgae culturing yields high value commercial products that could offset the capital and the operation costs of the process. Products of the proposed process are:
(a) mineralized carbon for stable sequestration; and (b) compounds of high commercial value. By selecting algae species, either one or combination or two can be produced.
4. The proposed process is a renewable cycle with minimal negative impacts on environment.

The research and experimentation we propose will examine and quantify the critical underlying processes. To our knowledge, the research we propose represents a radical departure from the large body of science and engineering in the area of gas separation. We believe the proposed research has significant potential to create scientific and engineering breakthroughs in controlled, high-throughput, photosynthetic carbon sequestration systems.

Figure 1. Recovery and sequestration of CO₂ from stationary combustion systems by photosynthesis of microalgae.

2. Executive Summary

This program calls for development of key technologies pertaining to: (1) treatment of effluent gases from the fossil fuel combustion systems; (2) transferring the recovered CO₂ into aquatic media; and (3) converting CO₂ efficiently by photosynthetic reactions to materials to be re-used or sequestered.

Since the inception of the program we have:

- Completed characterization of power plant exhaust gas;
- Identified a number of CO₂ separation processes;
- Analyzed 34 different strains for high value pigments;
- Determined the productivity parameters for over 20 different algae with 5 different simulated flue gases;
- Tested the compatibility of over 20 microalgal species with 5 different simulated flue gases;
- Tested three different strains for carbon sequestration potential into carbonates for long-term storage of carbon;
- Successfully carried out scale up of three microalgal strains to the 2000 liter outdoor photobioreactors;
- Conducted CO₂ mineralization study for *Haematococcus* in laboratory and in open-pond experiment;

- Installed the diagnostic instrumentation for characterization of coal combustion gas at Aquasearch Inc.;
- Delivered to Aquasearch the PSI coal reactor to be used with the Aquasearch 2000 liter outdoor photobioreactor for direct feeding of coal combustion gas to microalgae;
- Continued preparation of coal combustor for pilot scale production runs with coal combustion gases;
- Started the first full scale production run at the 25,000 photobioreactor using propane combustion gas;
- Carried out preliminary work on biomass separation for two microalgal strains grown in 2000 liter outdoor photobioreactors;
- Started to model the costs associated with biomass harvested from different microalgal strains;
- Conducted work on designing key components including: CO₂ removal process; CO₂ injection device; photobioreactor; product algae separation process; and process control devices;
- Developed a photobioreactor design concept for biofixation of CO₂ and photovoltaic power generation.
- Shared the ASPEN model with UH, PSI and Aquasearch for review and discussion;
- UH research staff visited Aquasearch and worked on-site for one week to gather information on the performance of the photobioreactor;
- Photobioreactor data from Aquasearch were analyzed and simple linear relationships for biomass productivity as a function of solar irradiance and CO₂ were developed using multiple regression;
- A review of the technical literature on tubular photobioreactors progressed;
- A literature study progressed to develop the CO₂ flue gas separation subsystem model for both Aspen Plus and Excel models;
- Conducted economic analysis for photobioreactor carbon fixation process; and
- Continued development of economic model to be used in predictions of carbon sequestration cost for a number of scenarios.

In Table 1, current status of each work scope is summarized.

Table 1. Current Status of Each Work Scope

Tasks	Title	% Complete	Milestone/Status Description
Task 1.0	Supply of CO ₂ from Power Plant Flue Gas	85%	Overall status for Tasks 1.1 through 1.3
Task 1.1	Power Plant Exhaust Characterization	100%	Most of pertinent exhaust gases were analyzed
Task 1.2	Selection of CO ₂ Separation and Clean-up Technologies	95%	MEA method identified. Direct injection of exhaust gas into water may be an option.
Task 1.3	Carbon Dissolution Method	75%	Analytical study completed. Direct exhaust gas injection may be studied per our Task 3 outcome.
Task 2.0	Selection of Microalgae	100%	Selection of 6 species out of initial 20
Subtask 2.1	Characterization of Physiology, Metabolism and Requirements of Microalgae	100%	Test compatibility of 20 species with 5 flue gases
Subtask 2.2	Achievable Photosynthetic Rates	100%	Productivity parameters of 20 species with 5 flue gases
Task 3.0	Optimization and Demonstration of Industrial Scale Photobioreactor	15%	Demonstrate viability of CO ₂ with algae at industrial scale
Subtask 3.1	Pilot Evaluation	15%	Evaluation at 2000 L pilot scale. Experimental work with coal reactor to be made.
Subtask 3.2	Full Scale Production Runs	15%	Evaluation at 24,000 L industrial scale
Subtask 3.3	Algae Separation and Final Product	40%	Evaluation of biomass separation
Task 4.0	Carbon Sequestration System Design	50%	Incorporating new system concept
Task 4.1	Component Design and Development	50%	New concept being incorporated
Task 4.2	System Integration and Simulation Analysis	50%	Analyses of new system concept to be made
Task 5.0	Economic Analysis	15%	Economic analysis of commercial microalgal CO ₂ sequestration
Task 5.1	Gas Separation Process	85%	Direct exhaust gas injection option to be assessed
Subtask 5.2	Photobioreactor Carbon Fixation Process	15%	Economic analysis of photobioreactor CO ₂ fixation
Subtask 5.3	Product Processing	5%	Economic analysis of product processing

The work discussed in this report covers the reporting period from 1 July to 30 September 2003.

3. Experimental

3.1 Task 3.1: Pilot Evaluation

During this quarter we have continued preparation of a coal combustor that will allow us to determine the suitability of coal combustion gases for microalgal-based carbon capture and sequestration at pilot scale (up to 2,000 liter photobioreactors). In the Results section, we describe the steps that have been taken to accomplish this goal.

Two 2000 liter photobioreactors growing *Haematococcus pluvialis* were prepared for testing with the coal combustion gases. However, since the coal combustor was not yet functional, a pH experiment was conducted with the cultures. The objective of the test was to estimate changes in the CO₂ capture efficiency at two different pH levels, 7.5 and 8.5. Our previous tests at the chemostat scale (Quarterly Report # 8) indicated that the efficiency of CO₂ capture by the photobioreactors is higher at higher pH levels.

Photobioreactors M09 and M10 were inoculated from the same mother culture on September 24. The pH set points for M10 were programmed at 7.4 and 7.6 while the set points for M09 were programmed at 8.4 and 8.6 starting on September 25, one day after inoculation. After 8 days of growth, the pH set points were reversed. Daily, samples were collected to estimate fluorescence, cell counts and pH and alkalinity as described in previous reports. Estimates of dissolved inorganic carbon (DIC) concentration in the medium were calculated from the pH logged every 5 minutes, as described in previous reports, and estimates of alkalinity based on the morning alkalinity measurements. It was assumed that the alkalinity does not change during the night hours since there is no photosynthetic energy available for nitrate assimilation. Then, we estimated changes in alkalinity during the light hours by assuming a linear increase from the alkalinity value measured in the morning on any one day to the value measured the next morning.

An example of the results of the DIC calculation is shown in Figure 2. From the DIC values we, then, calculated the rate of change in DIC with units of mg CO₂/L/minute for 5 min intervals. A decrease in medium DIC is assumed to represent disappearance of DIC from the medium to photosynthesis and/or degassing of CO₂. An increase in medium DIC represents cell respiration and/or an injection of CO₂ into the system (Figure 3). In our calculation, positive values in the rate of DIC change indicate net uptake of CO₂ by the cells and degassing from the medium while negative values indicate injection of CO₂ into the medium and cellular respiration. If we consider only the positive values and compare those obtained during daylight hours from those obtained during the night, the difference is the photosynthetic carbon uptake rate.

Figure 2. Logged pH values, estimated alkalinity and calculated dissolved inorganic carbon (DIC) in an algal culture over an 18 hour period.

Figure 3. Changes in dissolved inorganic carbon (DIC) in the culture medium and the corresponding rates of disappearance in DIC concentration. Positive rates of DIC disappearance represent degassing and/or photosynthesis. Negative rates represent injection of CO_2 into the medium in response to increases in medium pH.

3.2 Task 3.2: Full Scale Production Runs

During this quarter we finished preparation of a propane gas combustor to determine the suitability of propane combustion gases for microalgal-based carbon capture and sequestration at commercial scale (up to 25,000 liter photobioreactors).

The goal of our final set of experiments is to optimize gas delivery systems for photobioreactor performance at present commercial scale. These experiments are conducted in 25,000-L Mera Growth Modules (MGM), the commercial reactors on which current economic models are based. Flue gas is supplied by slipstream from the existing propane combustor to the commercial scale bioreactors. The composition of the flue gas can be modified as needed by the addition of more CO₂ and acid gases in order to simulate the flue gas compositions determined in Task 1 if needed. Based on the results of Task 1.3, we will optimize the gas injection system for maximum dissolution of CO₂. We will conduct experiments in the 25,000-L MGM using propane combustion product flue gas supplied by the system developed and using only the five to six species of microalgae selected for large-scale experiments. Each species will be run at this scale for 6 to 10 weeks, allowing for optimization.

This quarter we have started our first full scale experiment with strain AQ0008 (*Haematococcus pluvialis*). Cultures were grown in our commercial scale MGM photobioreactors. Two cultures were grown in parallel, one was fed pure CO₂ as the microalgal carbon source and pH control (M14), the second one was fed the stack gases from a commercial propane water heater as described in our previous quarterly report (M13). Gas additions, whether pure CO₂ or stack gases, were added to the MGM cultures on demand, i.e., when the pH of the cultures indicated lowering of the concentration of CO₂ in the medium.

Data were collected daily on the concentration of cells in the cultures to estimate growth rate and carbon capture, the fluorescence yield of the cultures (as described above) and the pH and alkalinity of the cultures' medium to estimate the concentration of CO₂, HCO₃⁻ and CO₃⁼ and total dissolved inorganic carbon (DIC). The pH of the cultures was continuously monitored by our computerized monitoring and control system. Changes in pH were used to estimate the rate of carbon assimilation by the microalgal cells as described above.

During this experiment we also used a gas analyzer consisting of a IMR400 gas dryer and a IMR5000 analyzer to measure the concentration of NO_x, and CO₂ in the gas stream from the propane combustor before and after the gas was introduced into the photobioreactor. From these values, we have estimated the relative capture efficiency of the microalgal culture. The analyzer was programmed to alternate between analyzing gases from the combustor smoke stack for a period of 20 min, switch to a purge period for 5 min, switch to the photobioreactor exhaust for 30 min and again to a 5 min purge period. Then, the cycle repeated itself.

3.3 Task 3.3: Algae Separation and Final Product

During the 12th quarter we have analyzed the data obtained during the 11th quarter from a set of bench scale centrifugation tests designed to determine the settling characteristics of different species of microalgae. These tests were designed to make relative estimates of

centrifugation capacity needed for different species of microalgae. It is expected that different species of microalgae will be more or less difficult to separate from the growth medium by centrifugation because of different physical characteristics such a density and particle size. This information will then be used in our economic model to support our cost estimates of algal biomass harvesting and separation form the growth medium.

Essentially, the tests consisted in centrifuging samples of different microalgal cultures in 15 ml tubes for specific amounts of time, noting the volume of pellet formed at those times and estimating the amount of biomass (using fluorescence) left in the supernatant. A digital photograph of the tubes following centrifugation was also be used to record the results.

Microalgal cultures were grown in 250 ml Erlenmeyer flasks under standard conditions (irradiance: $\sim 60 \mu\text{E m}^{-2} \text{s}^{-1}$, 14:10 L:D, temperature: $22 \pm 3^\circ\text{C}$). The flasks were manually shaken three times daily. Culture biomass was estimated from *in vivo* fluorescence. A Pulse Amplitude Modulated (MINI PAM, Walz, Germany) fluorometer was used to measure culture *in vivo* fluorescence. The fluorescence measured is proportional to the amount of chlorophyll, and thus biomass, of the culture. Once grown, the cultures were used in our centrifugation experiments as follows. A 2 to 5 ml sample (approximate) of culture was placed into a glass t-tube and its fluorescence measured. The measurement was repeated twice. The sample was then returned to the Erlenmeyer flask and the flask was thoroughly mixed. Next, six 15 ml samples of the culture were then placed in 15 ml centrifuge tubes. The samples were centrifuged in a Eppendorf 5810R centrifuge programmed for 0, 30", 60", 120", 300" and 600" at 400 rpm. The 0" centrifugation sample was spun up to 400 rpm and immediately stopped. We calculated that the centrifugation force experienced by the samples during speed-up and slow-down was equivalent to 6.5" at 400 rpm so this time was added to all our samples.

After each tube was centrifuged, the tubes were photographed and the pellet volume noted. Finally, a 2 to 3 ml of supernatant was taken from each tube and its fluorescence determined using the MINIPAM fluorometer. The change in fluorescence between pre- and post-centrifugation was used as an estimate of centrifugation efficiency. The values thus calculated were fit to a hyperbolic tangent model to estimate a centrifugation efficiency factor that will later be used in estimating centrifugation needs for a commercial sized facility producing the specific strains.

In our previous report we showed that the relative centrifugation efficiencies for the different strains ranged from 0.046 to 357. Using our model we can now estimate the amount of biomass harvested (under the conditions tested) for each strain for a standard set of conditions. From these estimates we, then, can calculate the costs of biomass centrifugation for the different strains. To do this, we have related the % biomass of each individual strain that would be harvested under our standard test conditions (Progress Report 11) within a standard 30 second period using the equation

$$\% Harvest = \% Harvest_{\max} * \left(1 - e^{\left(\frac{-a*time}{\% Harvest_{\max}} \right)} \right) * \left(e^{\left(\frac{-b*time}{\% Harvest_{\max}} \right)} \right) \quad (1)$$

The resulting 30” harvest estimates can then be related to the actual costs of centrifugation (Task 5).

3.4 Task 5.2: Photobioreactor Carbon Fixation Process

In this quarter we have continued to work on our cost modeling efforts of the carbon fixation process. An economic model for industrial scale algal facilities has been designed. The model includes over 500 variables that go into calculating monthly cost of goods, capital equipment requirements, land requirements and cash flow and balance sheets for 15 years of operation.

As a first step in the design process, we have specified the mass flows of the different materials necessary to run Mera’s present microalgal plant in Kona, Hawaii (Figure 20 in Quarterly Report #9). The model has been designed to be flexible and accept parameters for significantly larger microalgal plants that would produce a number of different microalgal products (e.g., astaxanthin from *Haematococcus*, biomass from *Spirulina*, and B-carotene and lutein from *Dunaliella* and *Nannochloropsis* respectively) utilizing open and closed photobioreactors while capturing carbon from smoke stack gases. We are now working on incorporating variables that will reflect differences in cost for centrifuging biomass from different microalgal strains.

4. **Results and Discussion**

Work accomplished in this reporting period is summarized according to the task structure of the program.

4.1 Task 1: Supply of CO₂ from Power Plant Gas to Photobioreactor

Most of the work within the two subtasks (Task 1.1: Power Plant Exhaust Characterization and Task 1.2: Selection of CO₂ Separation and Cleanup Technologies) has been conducted during the previous reporting periods. No additional activity was made during the present reporting period.

4.2 Task 2: Selection of Microalgae

Almost all work in this task was completed in the last reporting period. No additional work was made in this reporting period.

4.3 Task 3: Optimization and Demonstration of Industrial Scale Photobioreactor

The goal for this phase of this research program is to optimize carbon sequestration, high value component production and CO₂ mineralization utilizing microalgal cultures at a commercially significant scale. This is being done in two phases. First, we will conduct a pilot evaluation using 2,000 liter enclosed photobioreactors (pilot scale MGM, Task 3.1) and actual coal combustion gases as the carbon source for the microalgal cultures. Second, we are

conducting full scale production runs using 25,000 liter enclosed photobioreactors (full scale MGM, Task 3.2) and actual combustion gases from a propane burner as the carbon source for the algal cultures. Concurrently, research into the appropriate technologies for harvesting and processing the produced biomass will be conducted (Task 3.3).

As indicated in the 11th quarterly report, a number of technical problems associated with the gas analyzer and the coal combustor have significantly delayed progress in this part of the research program.

4.3.1 Gas Analyzer

With the help of the Tech Department at IMR over the phone and through e-mail, we attempted to work out all the bugs in the gas analyzer at Mera's facility. The problems we were having with the analyzer consisted of readings from the controllers that drifted from the settings and the NO_x sensor was inoperative. We made numerous attempts to put the IMR 5000 Gas Analyzer into service but were unsuccessful. After discussing the situation with Takashi Nakamura and Joe Morency (both from PSI), it was decided that the best course of action would be to return the gas analyzer to IMR and have them troubleshoot the unit and then repair the malfunctions at their facility. We shipped the gas analyzer to IMR for repair in July '03.

The gas analyzer was restored to "as-new condition" by IMR and most of the repairs were covered by the warranty. The repaired IMR 5000 was returned to Mera on 8/12/03 and we were finally able to place it into service on 8/13/03. Once the analyzer was in service, it was noticed that the gas concentrations reported for stack 1 (representing the gas input into the photobioreactor) were lower than those for stack 2! After some trouble shooting it was found that one of the pieces of flexible plastic tubing connecting the gas input adapter to the stack 1 manifold had loosened and was not capturing the gas from stack 1. After correcting this problem, the analyzer was then in continuous operation until 10/15/03, when a vacuum pump failed. A new pump was ordered and the old pump was sent to IMR for warranty inspection. After IMR inspected the failed vacuum pump, they determined it failed prematurely and its replacement would be covered under the warranty. The new vacuum pump was sent to us and the analyzer was repaired and placed back into service 10/23/03. The analyzer has been working continuously since and still continues to operate as expected without the development of any new problems.

4.3.2 Coal Combustor

The coal reactor has presented us with numerous problems during the startup procedure. When the reactor arrived on site at Mera's facility, it was noted during initial setup that the ceramic piece at the reactor's exit and the diffuser at the top entrance to the reactor were in very poor condition. Both pieces were repaired as well as they could be with the available materials and then put into place in the reactor. From the onset, coal could not be fed into the reactor via the supplied coal feeding system without it plugging. In order to encourage the coal to feed down the feeding tube and into the coal reactor without clogging, the feed airflow pattern has been changed, a bin vibrator has been added onto the coal feeding tube, the vibrator's position

has been adjusted for maximum efficiency, and the shape of the coal feeding tube has been redesigned.

It was initially thought that moisture in the coal might be causing the coal-feeding problem. However, we determined through analysis that the Freemont coal we were feeding into the reactor has a moisture content of only 2.5%. From this analysis, we made the assumption that moist coal was not a contributing factor to the feeding problem and after much iteration at feeding coal into the reactor, we have successfully revamped the coal feeding system so that coal continuously feeds into the reactor without clogging the feeding system.

After the above mentioned changes were in place, we were able to run the reactor continuously for over 2 hours without having coal-feeding problems. However, during this extended burning period, only a partial burning of the coal was observed (based on gas concentration data measured by the gas analyzer). We also noticed a significant increase in the furnace pressure. This indicated to us that the reactor's exit was plugged and the reactor was shut down.

Before we disassembled the reactor for inspection and troubleshooting, we contacted Joe Morency to consult with him regarding this new set of problems we were now experiencing. Mr. Morency provided some insights for solutions to the above-mentioned problems, such as adding pure oxygen to the combustion air stream in order to achieve a more efficient burning of the coal and to make certain there are no obstructions to the flue gas stream. Upon disassembly and inspection of the reactor we discovered that the reactor's exit was completely plugged with what appeared to be very hard carbon slag. The ceramic piece at the reactor's exit was cracked and it separated into two pieces when removed. Also, we observed that the alumina retort had cracked and had broken into two pieces. With these two critical parts of the reactor in such poor repair coupled with the ceramic diffuser at the top of the reactor being in marginal condition, it seemed probable that the reactor, as supplied, could not function at the required level without it first being fitted with new parts. It appeared that returning the reactor and all its supporting apparatus to as near the original specifications as possible would give us the best chance to collect sufficient data from flue gases produced by burning coal in the reactor.

From the above noted observations, we decided that reconditioning the reactor would be in the project's best interest and ours and, therefore, we have ordered a new retort and the material to cast a new replacement ceramic piece for the reactor's exit at the bottom of the alumina retort. However, the alumina tube and the ceramic material are not expected to arrive on site until November 15. With the success of this project always in mind, we will continue to troubleshoot the system and repair the reactor as needed until the furnace is operational.

4.3.3 Task 3.1: Pilot Evaluation

While waiting for the coal reactor to be brought on line, we used two pilot scale reactors to test whether culture pH has an effect on the CO₂ capture efficiency of microalgae cultures at large scale. Our previous results obtained from chemostat scale cultures (Quarterly Report #8) indicated that at lower culture pH a larger fraction of the CO₂ in the medium was lost back to the

atmosphere, lowering the CO₂ capture efficiency of the system. Figure 4 shows the pH recorded, at a frequency of every 5 min, for the two modules. Both cultures were started at a nominal pH of 7.5, i.e., the controlling set points were 7.4 and 7.6. On the second day, M09 pH was changed to 8.5, the set points were 8.4 and 8.6. The pH of the cultures was reversed after 8 days of growth.

Figure 4. pH traces obtained from M09 and M10.

Figure 5 shows the alkalinity measured in both photobioreactors during the experiment. Alkalinity increased over time due to nitrate uptake by the microalgal cells, indicating growth. Large decreases in alkalinity occurred when the culture was diluted with freshly made nutrient medium.

Figure 5. Alkalinity values measured in photobioreactors M09 and M10.

Figure 6 shows the fluorescence-based biomass estimates obtained once daily and Figure 7 shows the growth rates accomplished by the cultures during the experimental period, based on cell counts.

Figure 6. Fluorescence-based estimates of daily biomass.

Figure 7. Growth rates for M09 and M10 based on cell counts. Data missing is from days when culture dilutions took place.

The calculated rates of CO₂ disappearance from the medium are shown in Figure 8. It is clearly seen that the rates are larger when the culture pH is maintained near 7.5 than when maintained near 8.5 in both photobioreactors. These results reflected those obtained in our chemostat scale experiments reported on earlier. We have then averaged the CO₂ disappearance rate over each day and night period, eliminating those days during which culture dilutions took place from the calculations (since the alkalinity of the medium was significantly changed). We have further averaged the day and night values for each pH condition for each photobioreactor. Our results indicate that the night-time rate of CO₂ disappearance from the medium averaged 0.76 (M09 at 7.5 pH), 0.03 (M09 at 8.5 pH), 0.82 (M10 at 7.5 pH), and 0.01 (M10 at 8.5 pH) mg CO₂/L/min (Table 2). If we calculate the average concentration of CO₂ in the medium for each individual night period at the appropriate pH levels (7.5 or 8.5) we find a linear relationship between the concentration of CO₂ in the medium and the rate of degassing (Figure 9). According to this relationship, significant amounts of dissolved are lost from the medium at CO₂ concentrations of 1 mg CO₂/L and above.

Figure 8. Rates of dissolved inorganic carbon (DIC) disappearance from the medium (photosynthesis and/or degassing) for M09 and M10. The different symbols indicate the different conditions under which these data were obtained (night vs day, pH 7.5 vs pH 8.5).

Figure 9. Relationship between CO₂ concentration in the culture medium and night-time rate of dissolved inorganic carbon (DIC) disappearance from the medium (degassing) for M09 and M10.

Finally, we consider the difference between the rates of CO₂ disappearance from the medium obtained during the day and night periods. Assuming similar degassing and respiration rates, the difference between night and day can be ascribed to photosynthesis. We have averaged the calculated photosynthetic rates and present those results in Table 2. Based on our limited data set, faster photosynthetic rates were obtained at pH 7.5 than at pH 8.5 in M10 but not in M09.

Table 2. Average Rates of DIC Disappearance from the Medium (mg CO₂/L/min) for M09 and M10 at Either 7.5 or 8.5 pH and Either During the Day or Night Periods. The Difference is Assumed to be the Photosynthetic Rate.

PH	7.5			8.5		
	Day	Night	Difference	Day	Night	Difference
M09	0.93	0.76	0.17	0.21	0.03	0.18
M10	1.24	0.82	0.42	0.09	0.01	0.08

4.3.4 Task 3.2: Full Scale Production Run

During this quarter we have carried out the first direct comparison of microalgal performance in full scale reactors being fed either pure CO₂ or actual combustion gases from a propane combustor. Figure 10 shows the pH values measured during the duration of the experiment in both photobioreactors. Both photobioreactors were started at a nominal pH of 7.5, i.e., the controlling set points were 7.4 and 7.6. After several days, the pH was changed to 8.5, the set points were 8.4 and 8.6.

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Figure 10. pH traces obtained from M14, a full scale photobioreactor fed CO₂, and M13, a full scale photobioreactor fed propane combustion gases.

Figure 11 shows the cell concentrations and fluorescence-based biomass estimates obtained during the experimental period for these two photobioreactors. Growth rates were calculated from the changes in cell concentration and averaged for the each photobioreactor for the periods when the reactors were maintained at either 7.5 or 8.5 pH. For M14 (running on CO₂) the average growth rates were 0.28 d⁻¹ and 0.10 d⁻¹ at pH 7.5 and 8.5 respectively. For M13 (running on propane combustion gases) the average growth rates were 0.25 d⁻¹ and 0.12 d⁻¹ at pH 7.5 and 8.5 respectively.

The calculated rates of CO₂ disappearance from the medium are shown in Figure 12. As was the case in the pilot scale experiments (Subtask 3.1), it is clearly seen that the rates are larger when the culture pH is maintained at 7.5 than when maintained at 8.5 in both photobioreactors. These results reflected those obtained in our chemostat scale experiments reported on earlier. We have then averaged the CO₂ disappearance rate over each day and night period, eliminating those during which culture dilutions took place. We have further averaged the day and night values for each pH condition for each photobioreactor.

Figure 11. Cell concentration and fluorescence-based estimates of daily biomass for M14 and M13.

Figure 12. Rates of dissolved inorganic carbon (DIC) disappearance from the medium (photosynthesis and/or degassing) for M14 and M13. The different symbols indicate the different conditions under which these data were obtained (night vs day, pH 7.5 vs pH 8.5).

Finally, we subtracted the disappearance rates obtained during the night periods from those obtained for the daylight periods to calculate average photosynthetic rates. The results are shown in Table 3 and indicate that while the photosynthetic rates are higher for cultures maintained at 7.5 pH, the amount of CO₂ that is lost to the atmosphere from the medium is about 10 times higher at 7.5 pH than at 8.5 pH. These results agree well with those obtained in the pilot scale photobioreactors and in previous chemostat-scale experiments.

Table 3. Average rates of DIC disappearance from the medium (mg CO₂/L/min) for M14 (fed pure CO₂) and M13 (fed propane combustion gases) at either 7.5 or 8.5 pH and either during the day or night periods. The difference is assumed to be the photosynthetic rate.

pH	7.5			8.5		
Period	Day	Night	Difference	Day	Night	Difference
M14 (CO ₂)	0.58	0.24	0.34	0.13	0.02	0.11
M13 (propane)	0.44	0.17	0.27	0.09	0.01	0.07

Finally, we have considered the effect of CO₂ concentration in the culture medium on CO₂ degassing rate during the night periods (i.e., in the absence of photosynthesis). The results are shown in Figure 13 superimposed on the results obtained from the pilot scale photoreactors (M09 and M10, Figure 9). As was the case for the pilot scale experiment, more CO₂ is lost from the medium at higher CO₂ concentrations. Interestingly, at any CO₂ concentration in the medium the rate of degassing is lower for the full scale photobioreactors which translates into higher carbon capture efficiency. Due to technical issues (described earlier), we were able to obtain a limited amount of data on individual gas concentrations in the gas stream, before and after passage through the photobioreactor. These measurements were carried out on the photobioreactor being fed propane combustion gases.

Figure 13. Relationship between CO₂ concentration in the culture medium and night-time rate of dissolved inorganic carbon (DIC) disappearance from the medium (degassing) for M13 and M14 (full scale photobioreactor-25000L-, red squares) superimposed on the values obtained for M09 and M10 (pilot scale-2000L-blue diamonds, Figure 9).

Figure 14 shows an example of the gas concentration data obtained for a 24 hour period. As was noted earlier, the propane combustor produces combustion gases on demand, i.e., when the pH of the culture is rises above 7.6 in this example. The periods during which the combustor is active can be identified by the rapid decreases in pH resulting from the injection of CO₂ into the photobioreactor. During periods of no combustion, the gas concentrations measured reflect ambient air composition.

Figure 14. Example showing the concentration of CO₂ and NO_x in the gas stream supplied from the propane combustor into the photobioreactor (IN) and in the gas stream leaving the photobioreactor (OUT) for a 24 h period.

Figure 15 shows the CO₂ and NO_x gas concentrations measured during several days of photobioreactor operation. The data indicates that the concentration of CO₂ in the combustor exhaust is about 8.7% (v:v) but only about 4.8% in the photobioreactor's exhaust when the combustor is running. Similarly, the concentration of NO_x is about 91 ppm in the combustor exhaust but only about 75 ppm in the photobioreactor's exhaust. This means that about 45% of the CO₂ and about 18% of the NO_x is scrubbed from the flue gas by passage through the photobioreactor.

Figure 15. Concentration of CO_2 and NO_x in the gas stream supplied from the propane combustor into the photobioreactor (IN) and in the gas stream leaving the photobioreactor (OUT) for a 4-day period.

4.3.5 Task 3.3: Algae Separation and Final Product

In our previous report we showed that differences in algal morphology result in large differences in centrifugation efficiency. Using our modeled efficiency results we have now estimated the amount of biomass harvested (under the conditions tested) for each strain for a standard set of conditions. From these estimates we, then, can calculate the costs of biomass centrifugation for the different strains. Using Eq. (1), we estimated the % biomass that would be harvested under our standard conditions within a standard 30 second period. The results are shown in Figure 16 and range from less than 2% for AQ0011 (a very small coccoid organism) to 100% for filamentous forms like AQ0012 and AQ0016.

We are most familiar with the costs associated with the harvesting/centrifugation of *Haematococcus pluvialis* (AQ0008), a green microalga that we produce commercially. For our cost modeling efforts (Task 5) we will relate the centrifugation costs of other microalgal strains to those of *Haematococcus* as shown in Figure 17.

Figure 16. Calculated percent biomass harvested in a standard 30 second period for the different microalgal strains.

Figure 17. Estimates of relative centrifugation costs for the different microalgal strains. Haematococcus (AQ0008) is 1.

4.4 Task 4: Carbon Sequestration System Design

To evaluate the potential for application of photosynthetic sequestration of CO₂ to industrial-scale combustion systems, we are conducting a system-level design study. The purposes of this study are:

- (1) Identify design concepts for components and the integrated system of the proposed concept.
- (2) Optimize and evaluate performance of the components and the system.
- (3) Develop deployment methodologies.
- (4) Identify key technology issues for further development.

This task consists of two sub-tasks: Task 4.1: Component Design and Development, and Task 4.2: System Integration.

4.4.1 Task 4.1: Component Design and Development

The purpose of this subtask is to develop design concepts for each of the key components of the industrial scale photosynthetic sequestration of CO₂. Key components to be designed include: CO₂ removal process; CO₂ injection device; photobioreactor; product algae separation process; and process control devices. As the proposed system depends on the solar energy to photosynthetically convert CO₂ to products compounds, optimization of the photobioreactor is an important part of this task. In the reporting period, PSI continued to focus its effort on preparation of the PSI coal reactor to delivered to Aquasearch, and no significant progress was made in Task 4.1. In the following quarters, we will evaluate the concept to (i) assess feasibility of the concept for large scale photobioreactor, (ii) quantify cost benefit, and (iii) address technical issues for commercial applications.

4.4.2 Task 4.2: System Integration

The integrated process model being developed by UH requires submodels that accurately represent the behavior of key components, notably, the photobioreactor and the CO₂ flue gas separation system. In order to obtain data on the performance characteristics of the Aquasearch photobioreactor, the graduate research assistant (GRA), Mr. Simon Tsang, visited Mera Pharmaceuticals' facilities in Kona to collect data on microalgal cells growing in a photobioreactor fed with combustion gases from a pulverized coal furnace. Cells would be collected over 6 days and these samples would be analyzed to determine cell dry weight, organic carbon content, and other properties relevant to modeling photobioreactor performance. Due to problems with the pulverized coal feeder, the combustion tests were suspended and other experimental tasks were performed by the GRA during his visit.

4.4.2.1 Cell Dry Weight Analysis

The mass of an average microalgal cell and variations over time are needed by the photobioreactor model being developed. During his visit to the Mera Pharmaceutical facility, the GRA obtained data on cell dry weight from a photobioreactor that was being supplied with pure

CO₂ gas during daylight hours. Cell dry weight was calculated by dividing the measured average dry weight per volume of media by the measured average cell count per volume of media. For every hour of solar irradiance, three samples were collected, dried, and weighed to determine the dry weight per volume of media and one of these samples was analyzed three times with a cell counter to determine the number of cells per volume of media. Table 4 provides the sampling times, dry weight per volume of media, and corresponding averages. Data on the individual and average cell counts are shown in Table 5 and the calculated values of cell dry weight are summarized in Table 6 plotted in Figure 18.

Table 4. Dry Weights Per Volume of Media

Date	Time	Dry Weight (g/mL)			
		Sample 1	Sample 2	Sample 3	Average
9/25/03	6:07	6.70E-04	5.42E-04	5.94E-04	6.02E-04
	7:37	3.58E-04	4.00E-04	4.60E-04	4.06E-04
	8:27	4.72E-04	3.66E-04	3.50E-04	3.96E-04
	9:27	4.40E-04	3.48E-04	4.12E-04	4.00E-04
	10:47	4.50E-04	4.74E-04	4.20E-04	4.48E-04
	11:40	4.46E-04	4.32E-04	4.10E-04	4.29E-04
	12:50	4.50E-04	4.50E-04	4.72E-04	4.57E-04
	13:35	4.12E-04	4.72E-04	5.42E-04	4.75E-04
	14:20	5.74E-04	5.56E-04	4.64E-04	5.31E-04
	15:21	5.68E-04	6.06E-04	5.26E-04	5.67E-04
	16:12	5.96E-04	5.32E-04	5.54E-04	5.61E-04
	17:17	5.24E-04	5.42E-04	4.82E-04	5.16E-04
	18:04	4.54E-04	4.56E-04	4.58E-04	4.56E-04
	19:04	5.18E-04	4.78E-04	5.20E-04	5.05E-04

Table 5. Cell Counts per Volume of Media

Date	Time	Count 1 (cells/mL)	Count 2 (cells/mL)	Count 3 (cells/mL)	Average Count (cells/mL)
9/25/03	6:07	320275	319596	321633	320501
	7:37	334534	330576	331352	332154
	8:27	304173	300603	302194	302323
	9:27	321982	322098	319072	321051
	10:47	337288	340392	336532	338071
	11:40	337230	330770	336241	334747
	12:50	322176	323379	316899	320818
	13:35	331391	340897	335717	336002
	14:20	378707	362392	366970	369356
	15:21	360568	362974	358085	360542
	16:12	368037	363498	361441	364325
	17:17	361519	361829	364216	362521
	18:04	350248	349045	349103	349465
	19:04	332652	332012	333467	332710

Table 6. Cell Dry Weight

Date	Time	Cell Dry Weight (g/cell)
9/25/03	6:07	1.88E-09
	7:37	1.22E-09
	8:27	1.31E-09
	9:27	1.25E-09
	10:47	1.33E-09
	11:40	1.28E-09
	12:50	1.43E-09
	13:35	1.41E-09
	14:20	1.44E-09
	15:21	1.57E-09
	16:12	1.54E-09
	17:17	1.42E-09
	18:04	1.30E-09
	19:04	1.52E-09

Figure 18. Cell dry weight.

As shown in Table 6 and Figure 18, cell dry weight varied from 1.22×10^{-9} g/cell to 1.88×10^{-9} g/cell over the daylight sampling period. The first data point sampled at 6:07 a.m. may be inaccurate, since the sampling protocol was not finalized at that point in time. As seen in Figure 1, the dry cell weight appeared to increase between 7:37 to 15:21 and then decline until sunset.

Descriptive statistics of the data in Table 6 were calculated and are presented in Table 7.

Table 7. Cell Dry Weight Statistics

Mean	1.42147E-09
Standard Error	4.56944E-11
Median	1.41902E-09
Mode	#N/A
Standard Deviation	1.70973E-10
Sample Variance	2.92317E-20
Kurtosis	2.985925354
Skewness	1.462118805
Range	6.55983E-10
Minimum	1.22232E-09
Maximum	1.87831E-09
Sum	1.99005E-08
Count	14
Confidence Level(95.0%)	9.87167E-11

4.4.2.2 Total Inorganic Carbon

Work to determine total inorganic carbon in the photobioreactor from data collected by Mera Pharmaceuticals, Inc. continued during this reporting period. Photosynthetic CO₂ fixation can be used to estimate the quantity of carbon that is metabolized and bound in the cell biomass pool. This information is required to estimate carbon sequestration efficiency. Photosynthetic CO₂ fixation can be inferred from net total inorganic carbon concentration. Assuming that the inorganic carbon system is in equilibrium, the total inorganic carbon is determined by

$$[TCO_2] = [CA] \frac{1 + K_2^1 / H^+ + H^+ / K_{L1}^1}{1 + 2K_2^1 / H^+} \quad (2)$$

where $[TCO_2]$ is the total inorganic carbon concentration in mol/L, $[CA]$ is the carbonate alkalinity in meq/L, $[H^+]$ is the hydrogen ion activity (*i.e.*, 10^{-pH}), K_{L1}^1 is the first apparent dissociation constant for carbonic acid in water, and K_2^1 is the second apparent dissociation constant for carbonic acid in water (Geider and Osborne, 1992).

In order to use Eq. (2), the values of carbonate alkalinity and the first and second apparent dissociation constants must be estimated. These values depend on temperature. Values for the first and second apparent dissociation constants and a relationship for carbonate alkalinity are provided in the literature for a temperature of 25 °C (18th Edition of Standard Methods for the Examination of Water and Wastewater); however, temperature of the media inside the Mera Growth Module varies between 15 °C and 17 °C. A literature search was conducted to locate information to correct for this difference in temperature. References have been requested and calculated total inorganic carbon content results will be presented in the next Quarterly Report.

4.5 Task 5: Economic Analysis

Our aim in the economic analysis is to identify those components of the carbon sequestration process that have the greatest associated costs, given the design based on current data. Subsequent modeling will explore alternative technologies and procedures that might enable significant reduction in both capital and operating costs.

4.5.1 Task 5.1: Gas Separation Process

Much of work pertaining to this subtask has been completed in the previous reporting periods. We will address this issue again after we complete Tasks 3 and 4.

4.5.2 Task 5.2: Photobioreactor Carbon Fixation Process

We have no new results to report here at this time.

4.5.3 Task 5.3: Product Processing

We are continuing the development of an economic model for industrial scale algae facilities, specifically, we are now starting to incorporate costs associated with biomass harvesting (Subtask 3.3, above) into the model (Figure 17). We will continue to fine-tune our modeling efforts as the results from Task 2 and 3 are incorporated for a larger number of microalgal species and microalgal products. We expect to present updated results of our efforts in the next quarterly report.

5. Conclusion and Future Plans

5.1 Task 3: Optimization and Demonstration of Industrial Scale Photobioreactor

In this quarter, we have continued preparations for our pilot scale photobioreactor experiments where we will use combustion gases from coal to produce microalgal biomass and sequester carbon and we have run the first full scale propane combustion experiments. Specifically we have:

- Demonstrated differences in flue gas capture efficiency resulting from differences in culture medium pH and photobioreactor design as had been noted in the earlier bench scale experiments,
- Demonstrated carbon capture by microalgae from actual propane combustion gases,
- Determined that passage of propane flue gas through a microalgal photobioreactor removes about 45% and 18% of the CO₂ and NO_x, respectively, from the flue gas and that this can be improved following photobioreactor changes, and

- Established relative estimates of biomass harvesting costs for a variety of morphologically different microalgal strains.

Within the next quarter we expect to:

- Finalize modification to the coal combustor and start the first experiments with microalgal strains fed carbon exclusively from coal combustion.
- Continue propane-fed, full scale, photobioreactor experiments.

5.2 Task 4: Carbon Sequestration System Design

- Continued carbon balance analyses and estimation of CO₂ sequestration efficiency for the Aquasearch facility.
- Continued total organic carbon content analysis of *Haematococcus pluvialis* samples. A sampling protocol was developed and submitted to Mera Pharmaceuticals, Inc. along with a request for additional samples.

During the next reporting period we plan to conduct the following.

- Evaluate feasibility of large scale photobioreactor with photovoltaic electric power generation.
- Quantify the benefits of the above photobioreactor.

5.3 Task 5: Economic Analysis

In this quarter we have continued the development of an economic model for industrial scale algae facilities. Specifically, we have calculated relative centrifugation costs for individual strains that will be used in our final cost modeling efforts.

Within the next quarter we expect to:

- Continue development of the economic model to be used in predictions of carbon sequestration cost for a number of different scenarios.
- Incorporate productivity parameters for the different microalgal strains as determined during Tasks 2 and 3 of this project.

6. References

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