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Abstract

This report documents significant achievements in the Enhanced Practical Photosynthetic CO_2 Mitigation project during the period from 10/2/2001 through 10/01/2002. This report marks the end of year 2 of a three-year project as well as the milestone date for completion of Phase I activities. This report includes our current status and defines the steps being taken to ensure that we meet the project goals by the end of year 3. As indicated in the list of accomplishments below our current efforts are focused on evaluating candidate organisms and growth surfaces, preparing to conduct long-term tests in the bench-scale bioreactor test systems, and scaling-up the test facilities from bench scale to pilot scale.

Specific results and accomplishments for the third quarter of 2002 include:

Organisms and Growth Surfaces:

- Test results continue to indicate that thermophilic cyanobacteria have significant advantages as agents for practical photosynthetic CO_2 mitigation before mesophilic forms.
- Additional thermal features with developed cyanobacterial mats, which might be calcium resistant, were found in YNP.
- Back to back tests show that there is no detectable difference in the growth of isolate 1.2 s.c. (2) in standard and Ca-modified BG-11 medium. The doubling time for both cases was about 12 hours.
- The cultivation of cyanobacteria in Ca-BG medium should proceed in the pH range between 7 and 7.4, but this suggestion requires additional experiments.
- Cyanobacteria can be grown in media where sodium is present at trace levels.
- Ca^{2+} enriched medium can be used as a sink for CO_2 under alkaline conditions.
- Cyanobacteria are able to generate cones of filaments on travertine surfaces. [Travertine is a mixture of $CaCO_3$ and $CaSO_4$]. We hypothesize that SO_4^{2-} stimulates the generation of such cones, because they are not almost generated on $CaCO_3$ surface. On the other hand, we know that plant gas contains elevated concentrations of SO_4^{2-}. We may speculate that the introduction of 11.2 isolate in CRF might significantly increase the productivity of such facility. It is possible that a higher colonization potential for the screens may allow a higher surface productivity than some of the other isolates.
- The colonization of Omnisil surface is an auto-inducible and time-requiring process.
- Omnisil coupons should be treated under pH control, preferably using KOH.

Bioreactor support systems and test facilities:

- The pilot-scale bioreactor construction and debugging is continuing on schedule. Tests of the "natural" lighting system have shown acceptable levels of illumination for the bioreactor screens using only collected sunlight.
- Flow control inserts have been designed for the CRF-2 screens, which require header pipes for flow distribution and control. A staggered drilled-hole design and a thick shim design have both shown acceptable performance results (little to no clogging, uniform flow, ability to load algae on to the screen). They will both be tested in the CRF-2 to see which performs the best over long durations, and the best performing design will be used for the pilot scale bioreactor screens.

- Test results indicate that a sampling method can likely be used to reliably estimate the initial algae mass for a CRF-2 test. Work continues in this area to better define all sources of uncertainty for the initial mass estimation and to establish that the method is indeed repeatable and reliable.
- Tests have been run to set up the desired light distribution and intensity for a four screen test in the CRF-2. The light intensity test results indicate that overall light levels are in the desirable range, but a slight modification is necessary to be able to get a more uniform distribution between the center and the outside screens. The placement of the screens relative to the lights will be altered and these light intensity measurements will be redone prior to the first quantitative CRF-2 test.

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Results and Discussion

Task 1.0. Evaluate and rank component and subsystem level alternative design concepts
Subtask 1.1 Investigate critical properties of alternative photosynthetic agents (cyanobacteria)

In July of 2002 we carried out the experiment to determine the doubling time of 3d isolate 1.2 s.c. (3). The culture was traditionally grown in BG-11 medium supplemented with 30 mM HEPES. Initial pH of medium was 7.85. Medium was aerated with 5 % CO_2. The graph on Fig.1 suggests that doubling time for 1.2. s.c. (3) isolate was about 7- 8 hours during exponential phase of growth under mentioned conditions. We have to notice, that some decrease of doubling time of third organism during late logarithmic phase may suggest that there were some limitation factors of cyanobacterial growth, i.e. light and CO_2. Generally, this result confirmed our opinion that thermophilic cyanobacteria have significant advantages as agents for practical photosynthetic CO_2 mitigation before mesophilic forms.

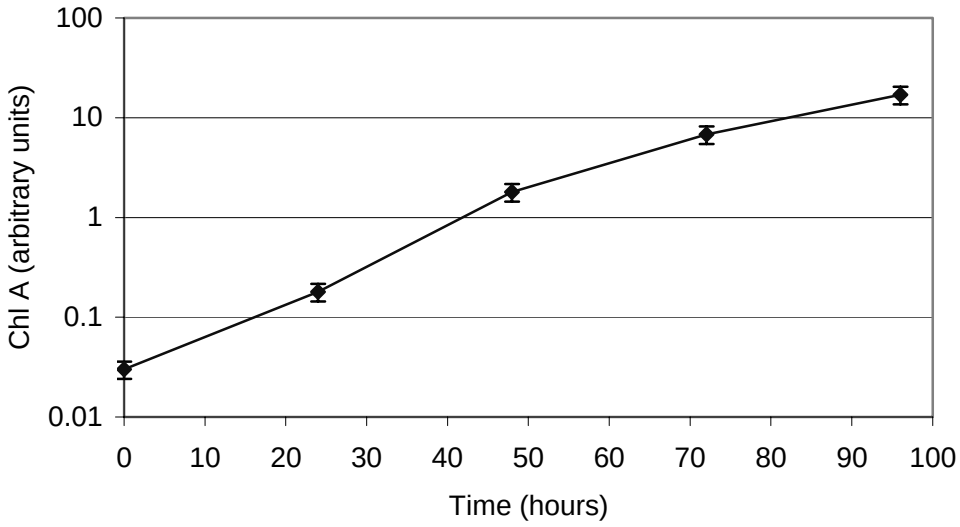

Fig.1. Growth of 1.2 s.c. (3) isolate in BG-11 medium supplemented with 30 mM HEPES aerated with 5% CO_2. Estimated doubling time during exponential growth is 7 hours.

Figure 2 suggests that the growth of 1.2 s.c. isolate proceeded in very optimal pH range - 7.5 – 7.9. It should be noticed that the original pH in the place of this culture isolation was about 7.6. Besides, we can conclude that the CO_2 consumption by cyanobacteria during logarithmic phase exceeded the rate of CO_2 supply (10-15 mL/min), because permanent alkalinization of growth medium was apparent.

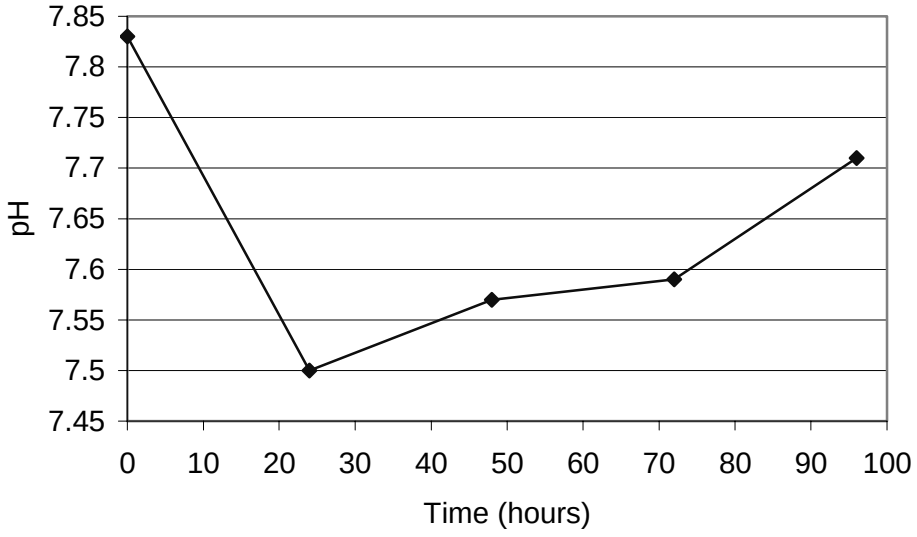

Fig. 2. pH dynamics during cultivation of 1.2. s.c.(3) isolate in BG-11 medium with 30 mM HEPES aerated with 5% CO_2.

We have also carried out an experiment to obtain quantitative data concerning the ability of isolate 1.2 s.c. (2) to grow in so-called Ca^{2+} medium. The initial idea of this experiment was that calcium itself is a good agent for CO_2 binding under alkaline conditions. A secondary idea for this experiment was to find an incubation medium that would provide no significant environmental contamination, as would be the case for sodium –based solutions. From this point of view calcium seems to be a more convenient component of growth media because it can be transformed eventually into insoluble $CaCO_3$. This material can be used in construction and as a soil pH amendment. Last variant seems to be important for this project because calcium carbonate precipitate can be obtained directly from spent growth medium, and consequently such medium will have a lot of biologically active substances having positive effect on plant growth (for review see the book of abstracts of the 1st Congress of the International Society for Applied Algology).

We prepared growth medium using as a basis of the recipe of BG-11, but all Na+ containing salts were replaced with their Ca^{2+} or K^{+} analogs. To check the property of this medium to be a source of calcium for the carbonate binding, we prepared both neutral and alkaline variations of the same medium. Both media were supplemented with Bis-Tris propane buffer, 10 mM, and medium pH was adjusted either to 7 and 9 before autoclaving. However, this process decreased medium pH to 6.7 in neutral variant and to 8.8 in alkaline variant, respectively.

The culture was grown in the tubular bioreactor at an illumination level of about 75 $\mu E/m^2/sec^{-1}$ and at a temperature 55^{o}C. Our observations revealed that cyanobacterial biomass in neutral Ca-BG medium increased in 600 times, approximately.

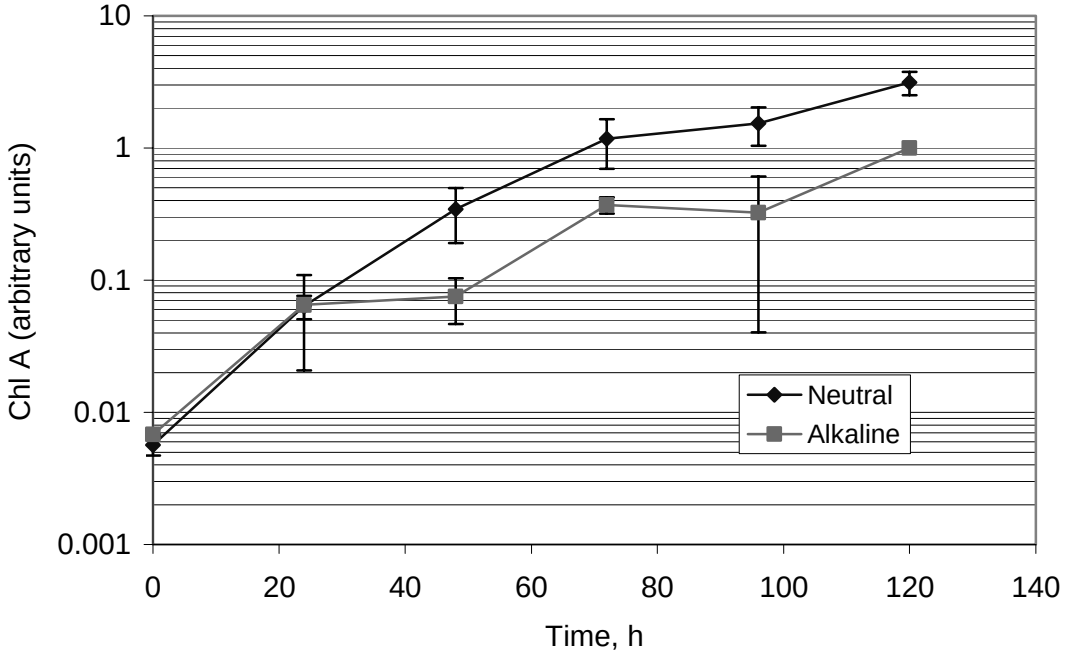

Fig. 3. Growth of 1.2 s.c.(2) in neutral and alkaline Ca-BG media aerated with 5 % CO_2

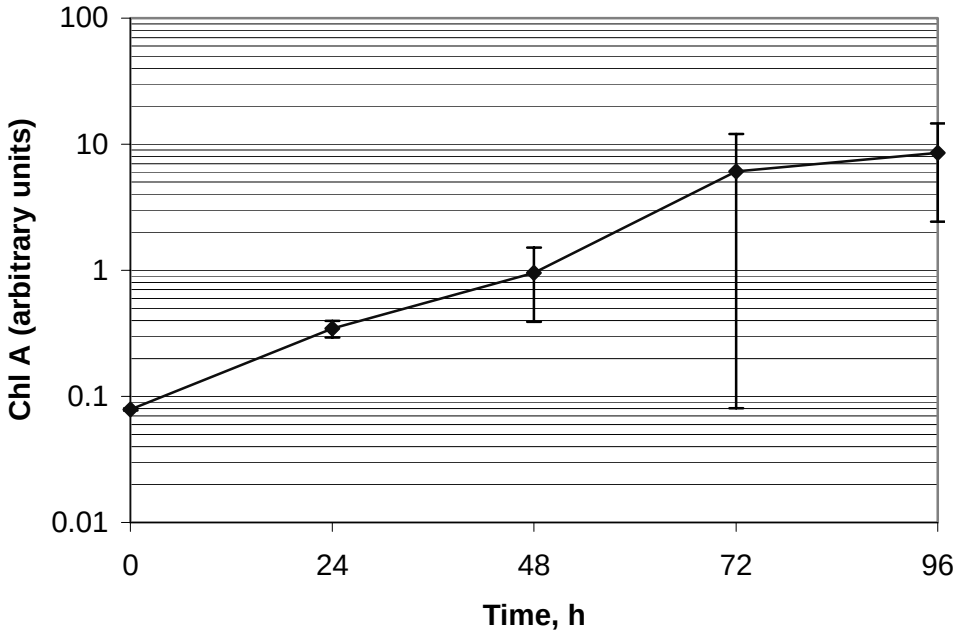

Fig. 4. Growth curve of unialgal isolate 1.2 s.c. (1) in BG-11 medium + 30 mM HEPES, aerated with 5 % CO_2 (Aver. D = 12.5 h) No $NaHCO_3$ added

Comparison of the curves in Fig.3.and 4 suggests that there is no detectable difference in the growth of isolate 1.2 s.c. (2) in standard and Ca-modified BG-11 medium. The doubling time for both cases was about 12 h, although we have observed shorter doubling time (~ 8 h) for

this isolate in standard BG-11. We think further experiments will allow us to find average doubling time for this culture in different media.

Medium pH measurements (Fig.5) showed that the growth of isolate 1.2. s.c. (2) in neutral medium was accompanied with slow alkalinization of growth medium despite permanent aeration of medium with 5% CO_2. This observation points out the possibility of using plant gas with more elevated concentrations of CO_2. Moreover, we can predict that used Ca-BG medium will have alkaline pH, which would be favorable for the precipitation of $CaCO_3$.

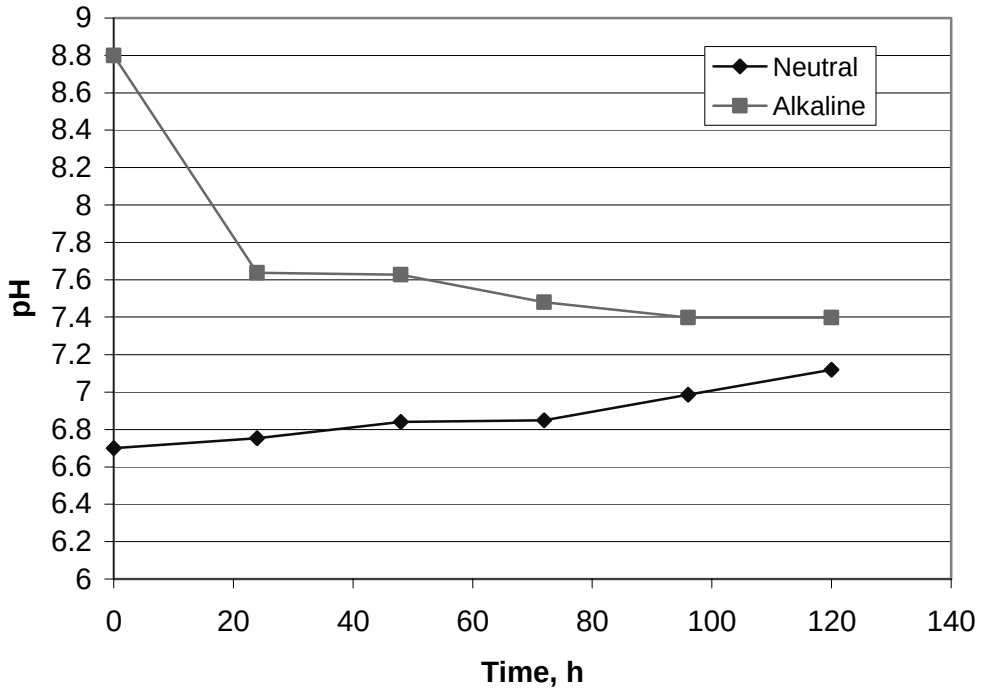

Fig. 5. pH dynamics in neutral and alkaline Ca-BG medium aerated with 5 % CO_2 during growth of isolate 1.2 s.c.(2)

Two points are important were observed:
(1) We observed strong generation of $CaCO_3$ during the first day of CO_2 bubbling. The tubes in the bioreactor were covered with carbonate ooliths.
(2) We also observed a strong affinity of cyanobacterial trichomes to the ooliths. However, the process of $CaCO_3$ precipitation decreased on the third day with parallel increasing of cyanobacterial growth. This may be connected with the reduction of medium pH.

Comparison of data in Figs. 1 and 3 allows us to predict that the cultivation of cyanobacteria in Ca-BG medium should proceed in the pH range between 7 and 7.4, but this suggestion requires additional experiments. We can make the following two conclusions based on these tests:
1) Cyanobacteria can be grown in media where sodium is present at trace levels.; 2) Ca^{2+} enriched medium can be used as a sink for CO_2 under alkaline conditions.

Dr. Igor Brown of Montana State visited Yellowstone National Park (YNP) on August 18-19, 2002. Main goal of this trip was to search additional thermal inventories for the isolation of thermophilic cyanobacteria, which can grow up in media with elevated concentrations of calcium.

Despite the fact that we had previously carried out the isolation of cyanobacteria from Angel Terrace (Mammoth Hot Springs Area, characterized with elevated concentrations of calcium and CO_2 in thermal waters), further efforts to increase the diversity of this group of cyanobacteria seems to be rational, because obtained cultures were isolated in areas with local temperatures bellow 55^{o}C. To realize this the following areas in YNP were sampled – Midway Basin, Firehole Lake, West Thumb, Angel Terrace and Narrow Gauge.

At Midway Basin, a semidry fumarole attracted attention. This fumarole was not filled with water; however this one permanently releases hot steam (Fig.6).

Fig. 6. A fumarole in Midway Geyser Basin

Despite semidry conditions, there were visible cyanobacterial mats on internal walls of this fumarole. White color of soil around a fumarole indicates that some kind of calcite was expelled from this fumarole together with steam. An analogous fumarole was found in Firehole area (Fig. 7). The walls and bottom of this fumarole were covered with significant layers of cyanobacterial mats. Soil and rock around this fumarole has white color, and we can presume elevated concentration of calcium in this area.

Fig. 7. A fumarole in Firehole area**.**

Strong cyanobacterial mats were also found in Hot Cascades, Firehole area (Fig. 8 and 9). Based on data received from YNP research office, waters at Hot Cascade are between 45 and 65^{o}C and an alkaline pH. The red arrow in Fig. 9 points to a small waterfall covered with cyanobacterial mat. Its appearance can be viewed as a natural analog of CRF facility at Ohio University.

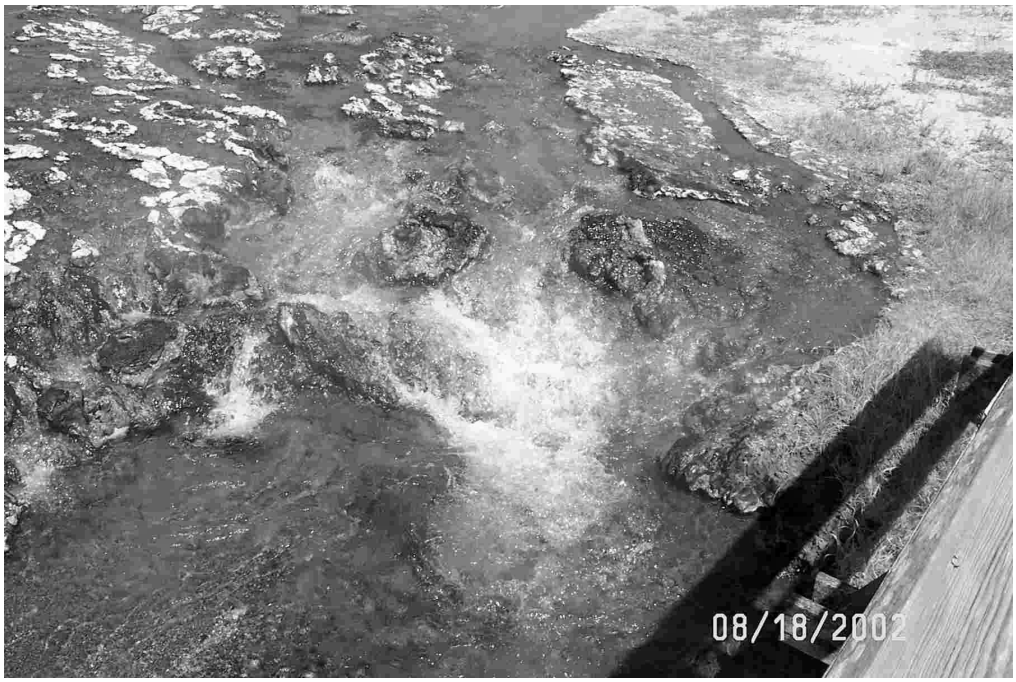

Fig. 8. Surface cyanobacterial mats in Hot Cascade

Fig. 9. Underwater cyanobacterial mats in Hot Cascade

The thermal inventories in West Thumb Geyser may have significant interest for our project. On the base of information obtained from YNP office, thermal waters in West thumb area have an alkaline pH. In Fig. 10, a small thermal feature is shown with diameter about 70 cm saturated with cyanobacteria and precipitated calcium. Another fumarole (unfortunately the picture was lost) that was found in this area has two important properties related to this work. First, it was a small fountain and second, it had lips, similar to a volcano, dripping with hot water. The lips were covered with a thick cyanobacterial mucilinous mat. As it seems to us, exo-polymer substance of this mat could give additional stability for screens in CRF facility.

Fig. 10. A fumarole in West Thumb Geyser Basin area

We were unable to collect samples from mentioned inventories, because a Park Ranger was not available. However, we have discussed sampling in mentioned area with Mrs. C. Hendrix (Office of Research Permits). She believes that the areas we have described should be accessible till the beginning of November. And, if we apply on ranger assistance in advance, she will issue a special permit for the sampling in these areas.

However, we were able to carry out a traditional scraping in a shallow hot stream in Narrow Gauge Area, which is shown in Figure 11. No information is available about the scraping at the time of this report.

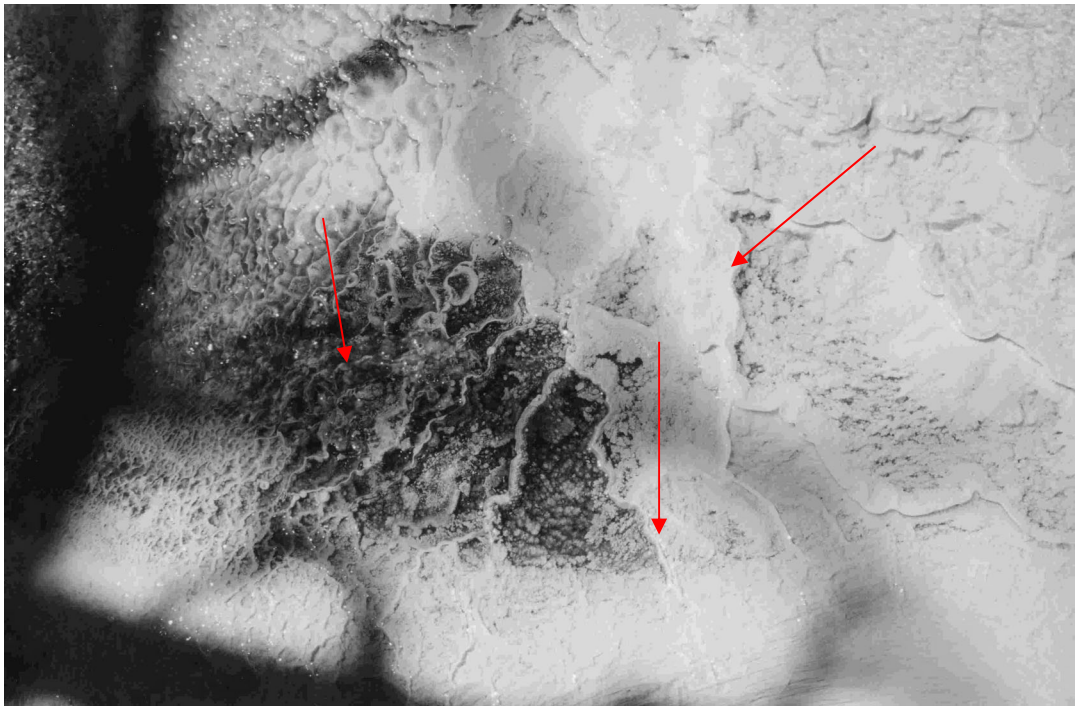

Fig. 11. Places of scraping (red arrows) in a hot spring in Narrow Gauge area

While no solid conclusion can be drawn from this work without sampling, it appears that there are opportunities for discovering additional organisms with features suitable or beneficial for our project in YNP. It is possible that the cyanobacterial mats that were observed by our team might be calcium resistant.

Also, during September 2002 we concentrated on screening an appropriate isolate, which is able to grow in so-called “Ca-BG” medium. The principal peculiarity of this medium is that all salts containing sodium were changed to calcium or potassium. The initial pH of this medium was 7. You can see in Fig.12 that initial inoculum of 1.2 s.c.(2) was at a very low density of cyanobacterial cells.

Fig. 12. Initial inoculum

The next picture was taken after 3 weeks of incubation under temperature 55^{o}C, rate of illumination – 35 μE/m^{2}/sec. Fig. 13 suggests that isolate 1.2 s.c. (2) adapted to the new type of medium and has divided. We noted the adhesion of 1.2 s.c. isolates cells to flask bottom. Such strong adhesion of cells to flask bottom might limit the transport of CO_2 (HCO_3^{2-}) to cells. However, the vortex of culture as well as medium aeration with 5% CO_2 may increase the rate of growth of isolate 1.2 s.c.(2) in Ca-BG medium.

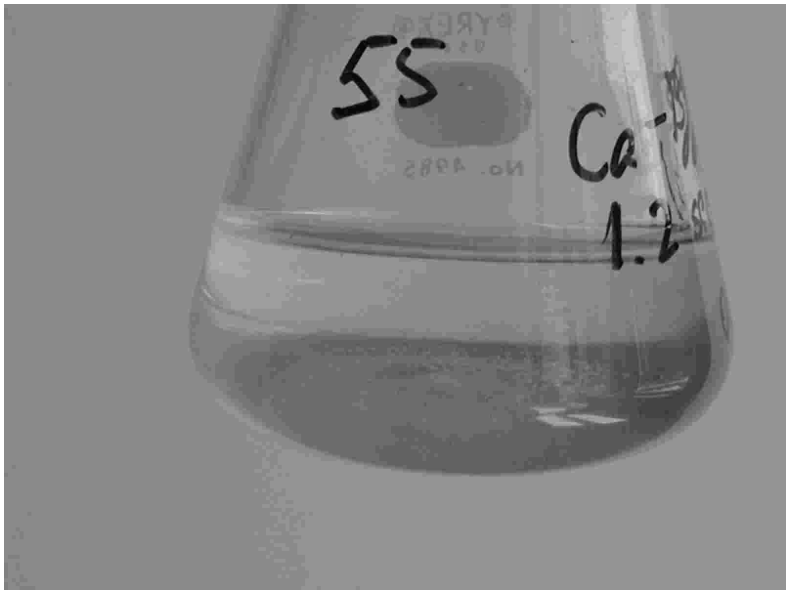

Fig. 13. Inoculum three weeks later

We also tested the growth of unialgal culture 8.2.1 *Lingbia*, isolated from travertine surface of Angel Terrace (the area with highest concentration of CO_2 in underground water) in Ca-BG medium. Fig. 14 and 15 suggest that this culture can also grow in Ca-BG medium. Again, note the adhesion of *Lingbia* cells to flask walls.

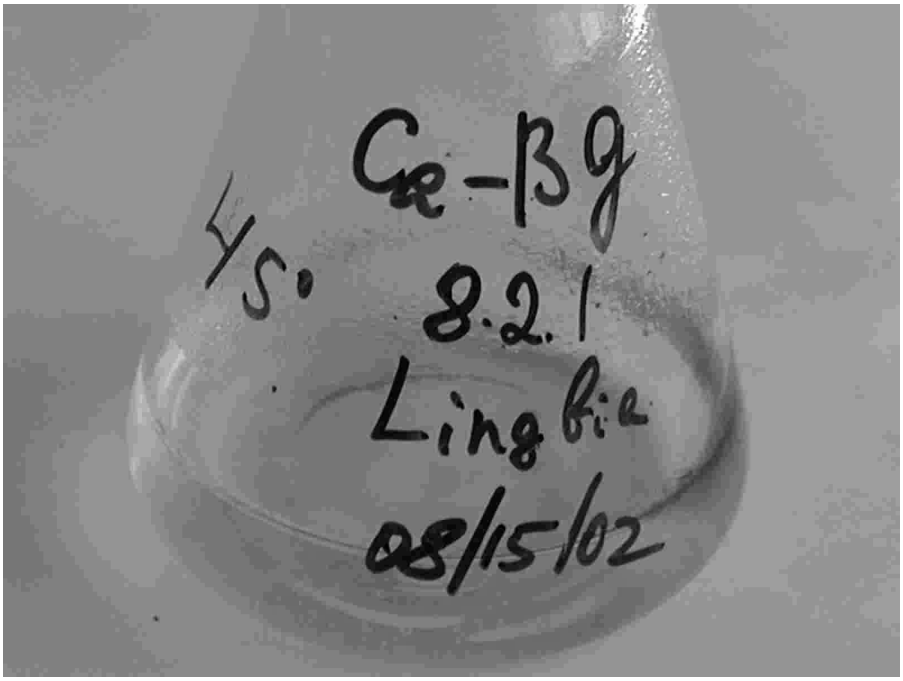

Fig. 14. Unialgal culture 8.2.1 *Lingbia* Ca-BG medium

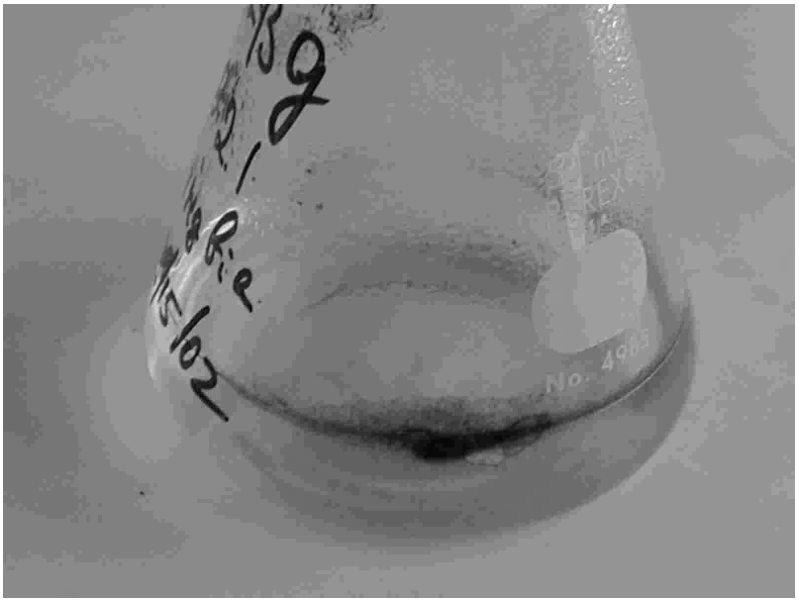

Fig. 15. Unialgal culture 8.2.1 *Lingbia* Ca-BG medium after three weeks

During last 3 weeks we have also continued the initial steps of culturing of new samples, collected in Mammoth Hot Springs area in August 2002. Our attention was attracted to a sample, which we have since named #11.3. Cyanobacteria from this sample are able to generate cones of filaments on the travertine surface, as shown in Figure 16. [Travertine is a mixture of $CaCO_3$ and $CaSO_4$]. We hypothesize that SO_4^{2-} stimulates the generation of such cones, because they are not generated on $CaCO_3$ surface. On the other hand, we know that plant gas contains elevated concentrations of SO_4^{2-}. We may speculate that the introduction of 11.2 isolate in CRF might significantly increase the productivity. It is possible that a higher colonization potential of this organism on membranes (such as what we would find in our bioreactor) may allow a higher surface productivity than some of the other isolates.

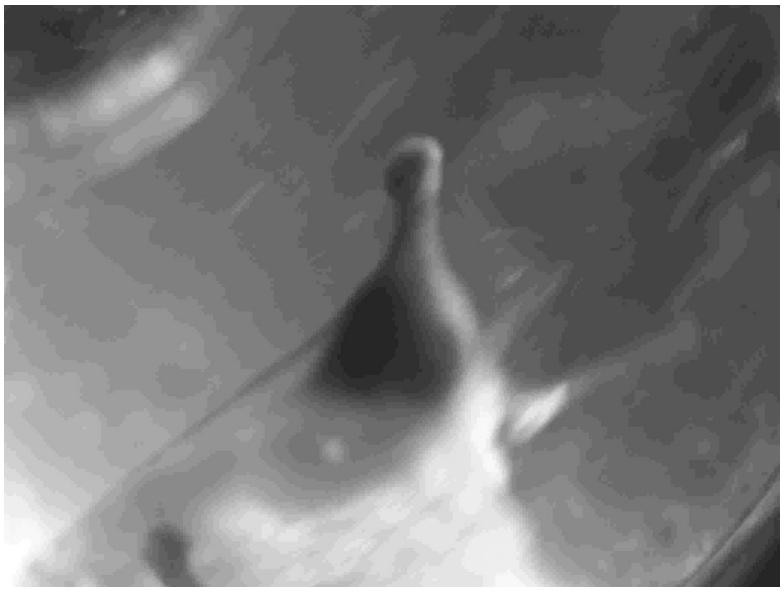

Fig. 16. Sample #11.3 cyanobacteria cone filaments

The organisms, TR9.4 and 1.2 s.c. (2), continue to be grown in large containers (from 5-gallon to 109 gallon) for use in CRF tests, offline harvesting tests, and eventually the pilot-scale bioreactor tests. Additional organisms (currently identified as organism 3 and organism 4 for simplicity) are also being cultured and are in the process of being scaled up to the 5-gallon containers. These organisms will be available soon for preliminary testing in the bioreactor systems.

Large-scale Mass Measurements

One of the high priority activities with respect to subtask 1.1 is the preparation of the test facilities and validation of the test procedures for the CRF-2 bioreactor. CRF-2 is the scaled up version of the original CRF model scale bioreactor, and it serves as the intermediate scale test system as we simultaneously work to develop the pilot scale system. Construction work to improve the CRF-2 pump and water system as well as to expand the bioreactor chamber to accommodate new screen designs (with header pipes for water distribution) has been successfully completed.

A series of tests was run to demonstrate that the mass of algae loaded into the CRF-2 system can be accurately determined based on a statistical sampling procedure. In these tests an initial volume of 10 gallons of algae slurry is loaded in to the CRF-2, then fifteen 150 mL samples are randomly taken from the well-mixed slurry. The average dry algae mass based on the 15 samples is used to establish an estimated initial loading of algae (with a statistically determined uncertainty). The slurry is run through the CRF-2 system for a short time (short enough so that no algae growth is expected) then the entire system is flushed and the system filters are dried to determine the actual total loading of algae for comparison to the estimate based on the sampling procedure. Results for one such test are shown in Table 1. These results are not conclusive but they do indicate that the sampling method can likely be used to reliably estimate the initial algae mass for a CRF-2 test. Work continues in this area to better define all sources of uncertainty for the initial mass estimation and to establish that the method is indeed repeatable and reliable.

Table 1. CRF-2 sampling test result for validating the estimation of the initial mass of algae

	Dry Filter (g)	Dry Filter with Dry Algae (g)	Net Dry Algae(g)
1	0.1532	0.1715	0.0183
2	0.1526	0.1691	0.0165
3	0.1511	0.1710	0.0199
4	0.1510	0.1663	0.0153
5	0.1534	0.1720	0.0186
6	0.1506	0.1677	0.0171
7	0.1531	0.1694	0.0163
8	0.1506	0.1675	0.0169
9	0.1509	0.1660	0.0151
10	0.1540	0.1711	0.0171
11	0.1527	0.1694	0.0167
12	0.1509	0.1698	0.0189
13	0.1537	0.1719	0.0182
14	0.1513	0.1687	0.0174
15	0.1516	0.1696	0.0180

Average Dry Mass(Xaverage) = 0.0174 g/100ml
Standard Deviation(std) = 0.0013 g/100ml
Initial loading estimate= 6.3249 g $\pm$ 0.2619 g (4.14% uncertainty)

	Dry Weight (g)	Dry Weight With Dry Algae(g)	Algae Dry Weight (g)
Membrane	467.0361 container1=243.9758	468.5003- container1=245.2811	1.4642
Coarse Filter (350 μm)	323.9969- container2=150.4356	328.0311 container2=155.5220	4.1342
Fine Filter(0.5 μm)	340.9335- container3=242.2729	341.8009 container3=245.0114	0.8674

Sum of dry algae= 6.4658(g)

(Note: The actual measurement of algae mass falls within the uncertainty range for the initial loading estimation)

Light Distribution

Additionally, tests have been run to set up the desired light distribution and intensity for a four screen test in the CRF-2. Results of these light intensity tests (Figure 17 and Table 2) indicate that overall light levels are in the desirable range, but a slight modification is necessary to be able to get a more uniform distribution between the center and the outside screens. The placement of the screens relative to the lights will be altered and these light intensity measurements will be redone prior to the first quantitative CRF-2 test.

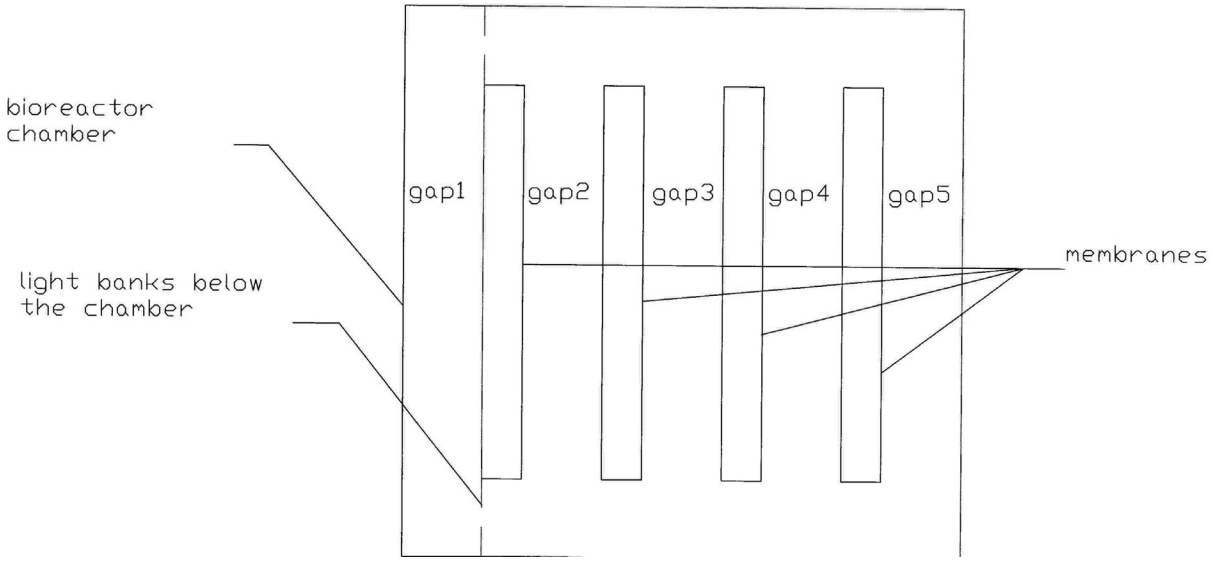

Fig. 17 a: Top view, showing gap numbers

Left	Center	Right
1	1	1
2	2	2
3	3	3

Fig. 17 b: Side view of each membrane, showing measurement locations

Table 2. CRF-2 light intensity measurements
All readings are in (μmols-sec^{-1}m^{-2}).

GAP 1:

	Left			Center					Right		
	Up	Normal	Down	Left	Right	Up	Normal	Down	Up	Normal	Down
1.	7.12	6.37	7.87			11.99	6.37	6.37	5.62	6.37	5.25
2.	7.12	8.62	7.12	7.12	6.00	8.62	6.00	6.75	5.25	8.62	5.25
3.	7.12	9.745	6.00			9.37	5.62	6.37	5.25	10.12	4.50

GAP 2:

	Left			Center					Right		
	Up	Normal	Down	Left	Right	Up	Normal	Down	Up	Normal	Down
1.	23.61	32.98	22.11			22.48	38.98	29.98	22.86	15.74	29.23
2.	26.23	31.11	25.48	26.98	26.98	32.98	37.86	34.48	23.61	23.61	28.86
3.	24.36	29.98	29.61			40.48	38.23	32.98	20.99	21.36	25.48

GAP 3:

	Left			Center					Right		
	Up	Normal	Down	Left	Right	Up	Normal	Down	Up	Normal	Down
1.	43.48	38.60	55.85			53.23	51.35	62.97	53.60	45.35	51.35
2.	50.98	38.23	54.72	38.60	43.10	61.85	53.97	67.84	59.97	45.73	49.48
3.	47.98	39.73	55.47			64.10	54.35	67.47	55.85	44.60	48.73

GAP 4:

	Left			Center					Right		
	Up	Normal	Down	Left	Right	Up	Normal	Down	Up	Normal	Down
1.	37.86	34.86	49.10			43.85	42.73	55.47	44.23	37.48	37.48
2.	41.60	35.23	48.73	39.73	38.61	49.85	46.10	57.72	45.73	38.61	39.73
3.	41.98	36.36	42.73			50.98	50.60	56.22	47.60	39.73	41.23

Conventions:

Up: Pointing the LiCor sensor upwards (towards the header pipe)
Down: Pointing the LiCor sensor downwards (towards the base of the bioreactor chamber)
Normal: Pointing the LiCor sensor facing outwards and normal to the membrane
The readings were taken with all the lights ON.

Subtask 1.2 Design deep-penetration light delivery subsystem

ORNL continues to support the pilot-scale bioreactor development. During the first long duration test the light fibers separated from their containment within the lighting panels. The problem was traced to breakdown of the adhesive used for the mounts at high temperature. ORNL has repaired the light distribution panels using a special adhesive capable of withstanding 150°C sustained temperatures.

The next three photographs (Fig. 18 through Fig. 20) show the panels "in action." In Figure 18, the panels are viewed along the flow path of the simulated flue gas (at least as close as possible without getting inside the bioreactor) Note that the membrane substrates for the microalgae have been removed for this picture. If the substrate panels were not removed, the detail of the lighting panels would not be visible. In Figure 19, the fibers going into the panel can be seen. In Figure 20 presents a clearer view of the top of the panels, including the fibers from the solar collector.

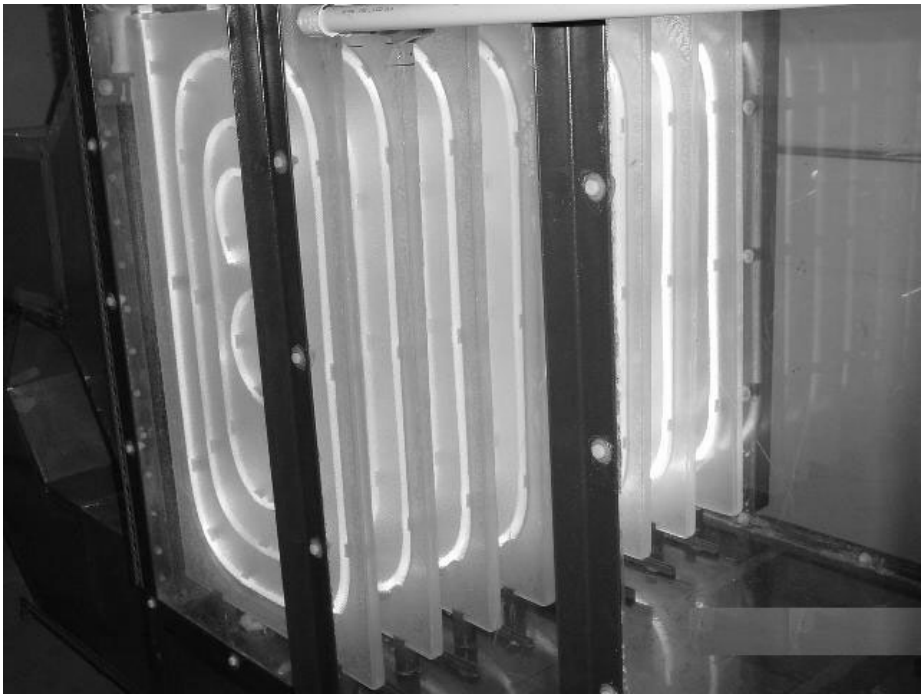

Fig. 18. Lighting panels viewed from direction of gas flow

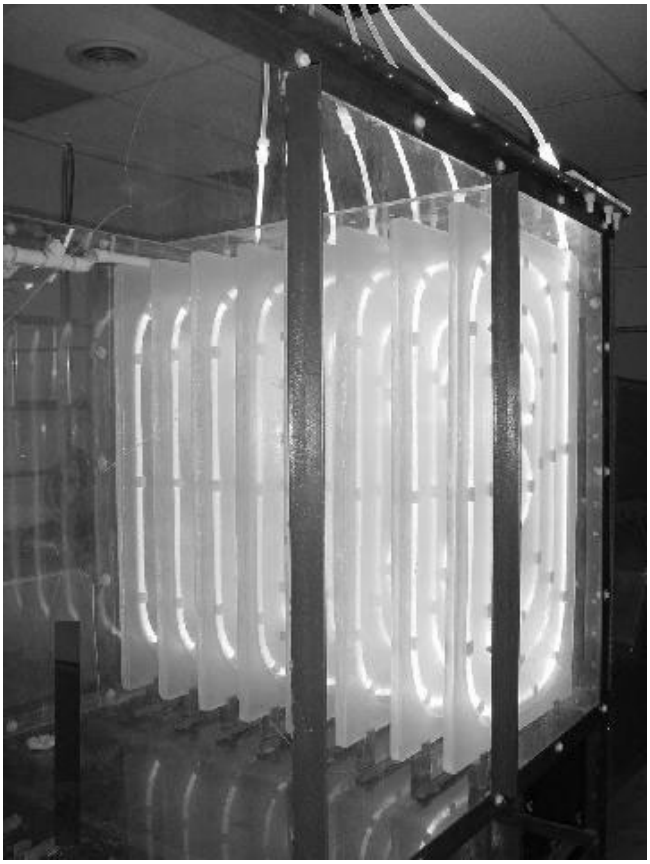

Fig. 19. Lighting panels with fiber optic leads

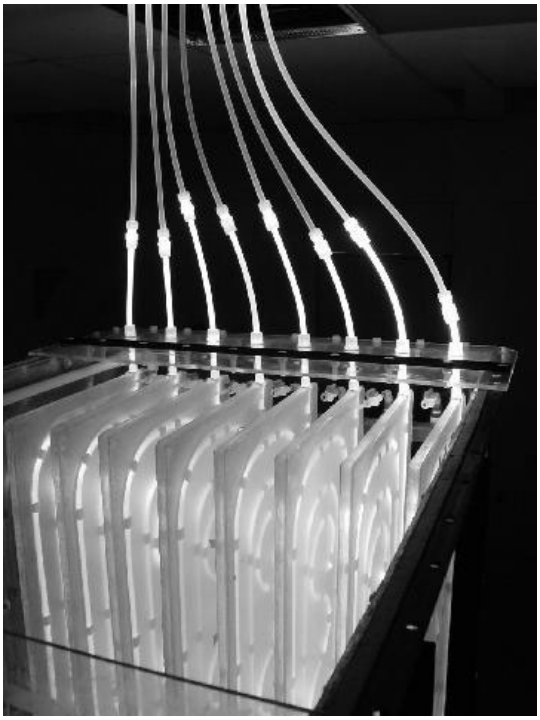

Fig. 20. Top of lighting panels

Subtask 1.3 Investigate growth surface subsystem design

Additional tests of growth surface materials continue, both at Ohio University and Montana State. Tests at Ohio University had identified Ryton material as a good candidate material based on strength and ability to withstand the harsh bioreactor conditions, however its poor wetting properties halted the serious consideration of this material at this time.

Researchers at Montana State studied the effect of Omnisil coupons, prepared in Ohio, on the growth of 1.2.s.c. (2) isolate in BG-11 medium supplemented with 30 mM HEPES and aerated with 5 % CO_2. The general view of fresh coupons is shown in Fig.21.

Note that inoculum density was very low. Nonetheless further cultivation showed good growth of 1.2 s.c.(2) isolate in the presence of Omnisil coupons, treated in Ohio (Fig. 22), although these coupons decreased medium pH to 6.7, while CO_2 bubbling alone decreased medium pH to 7.44. These results are shown in Figure 23. We also checked the effect of Omnisil coupon overnight soaking in BG-11 medium supplemented with 30 mM HEPES (init. pH – 7.8). We found that pH of the medium dropped down from 7.85 to 7.15

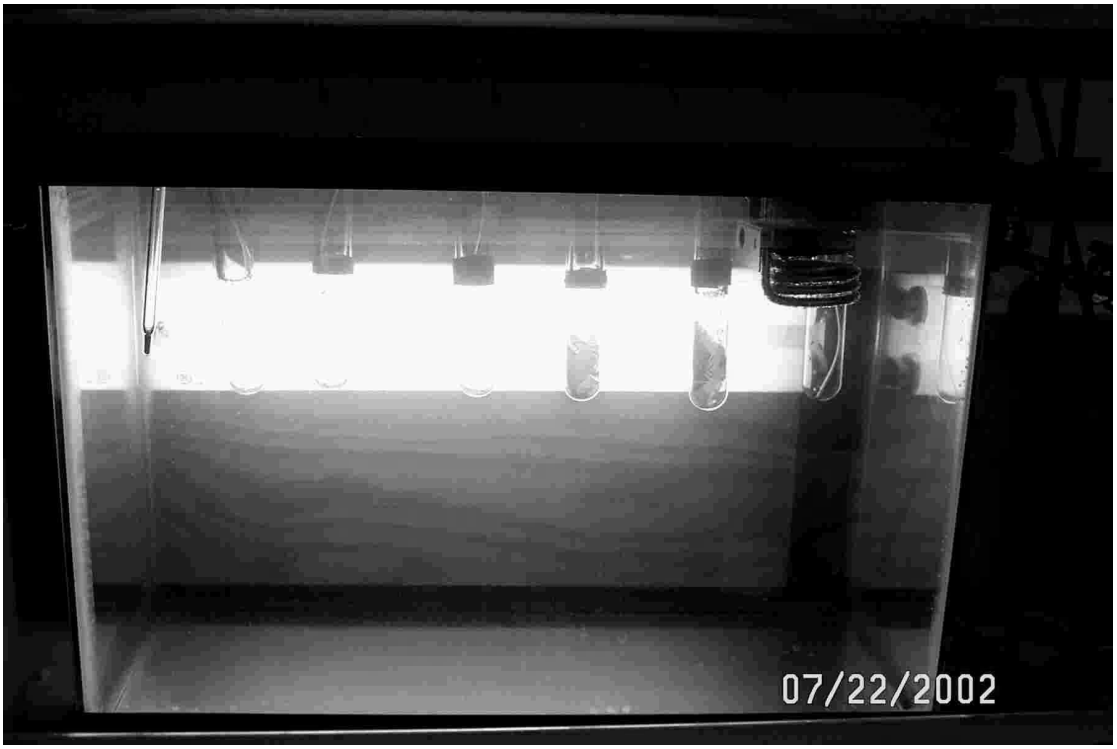

Fig. 21. Tubes with fresh Omnisil coupons

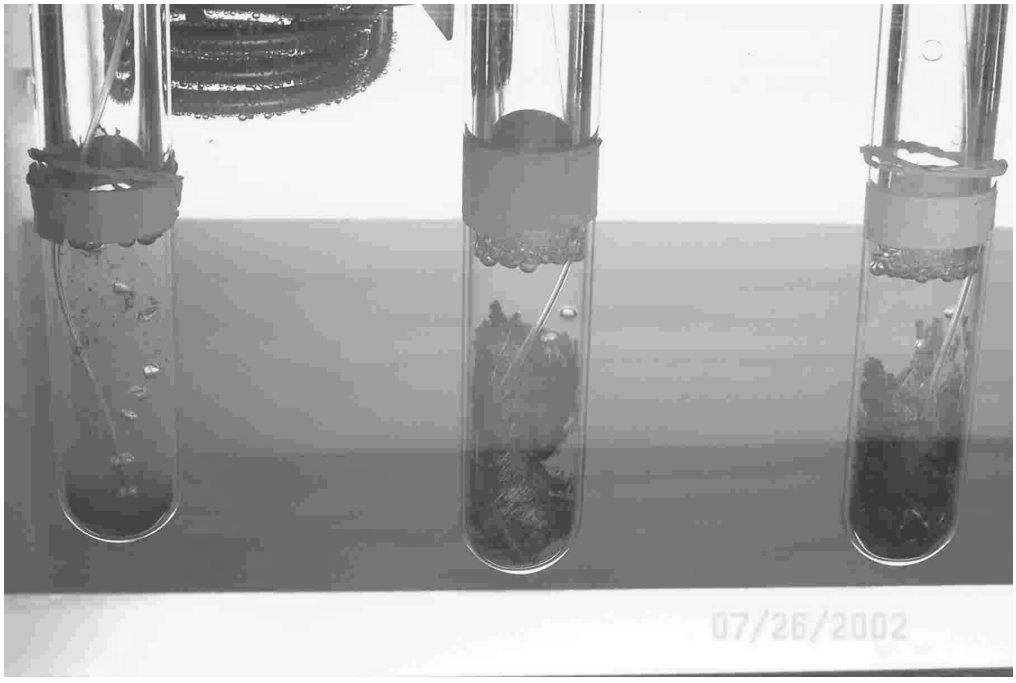

Fig. 22. Colonized Omnisil coupons (last day of experiment; tubes 2 and 3 from left)

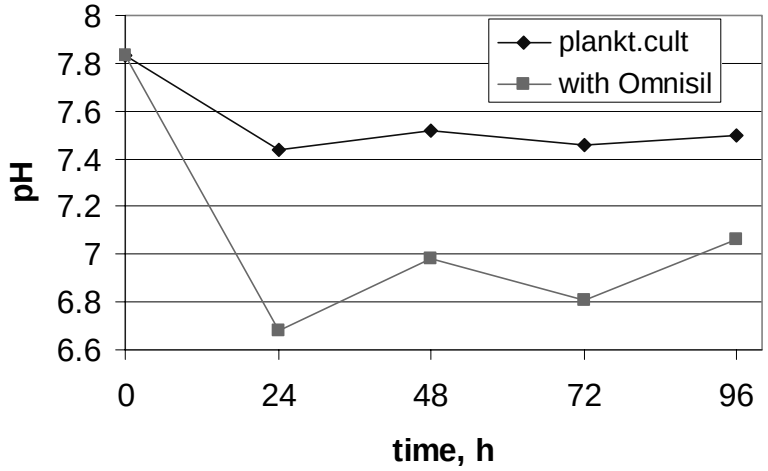

Fig. 23. pH dynamics in cultivation media in tubes with Omnisil coupon (purple curve) and w/o Omnisil (dark blue curve). The aeration with 5 % CO_2 was simultaneously begun.

Macroscopic observation of removed coupons showed satisfied level of colonization by cyanobacteria, especially on the back side (Fig. 24). It is theorized that if coupons would have more flat surface, the rate of colonization would be higher.

Fig. 24a: Coupons captured from face side.

Fig. 24b: Coupons captured from the back side

Figure 25 suggests that there was almost no planktonic fraction of cyanobacteria, when they are grown at the presence of Omnisil. Observed green spots are on internal walls of tubes. Actually, we have observed the same phenomenon for other organisms.

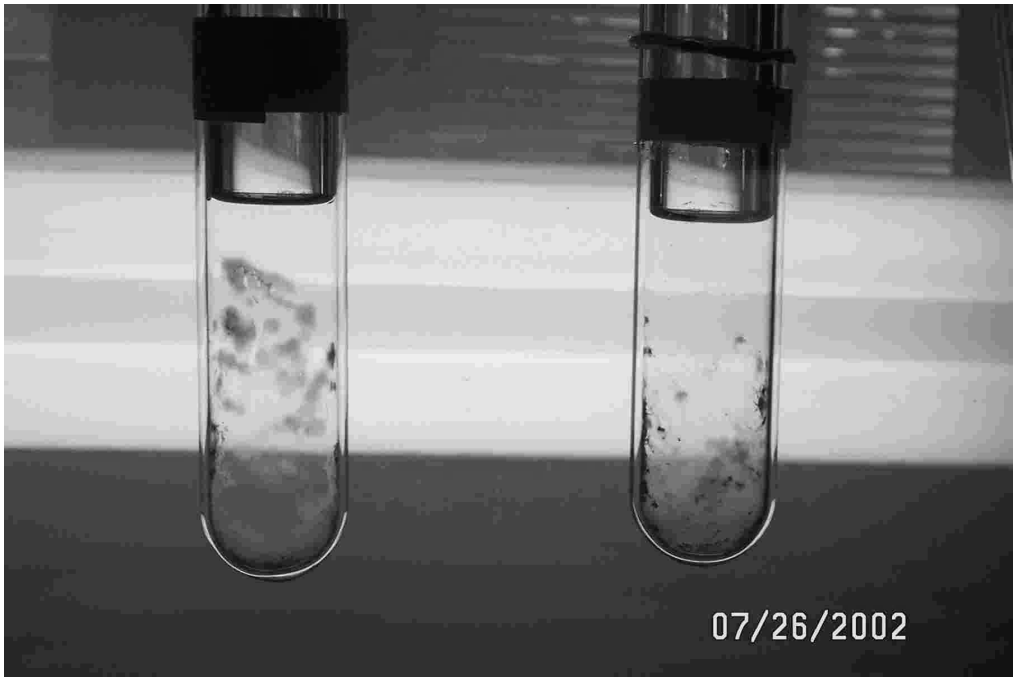

Fig. 25. Photo of tubes after removing Omnisil coupons.

This conclusion was confirmed by separate determination of chlorophyll “a” amount (per 50 mL) on Omnisil coupon and tube walls (Fig. 26). Planktonic chlorophyll was not detectable.

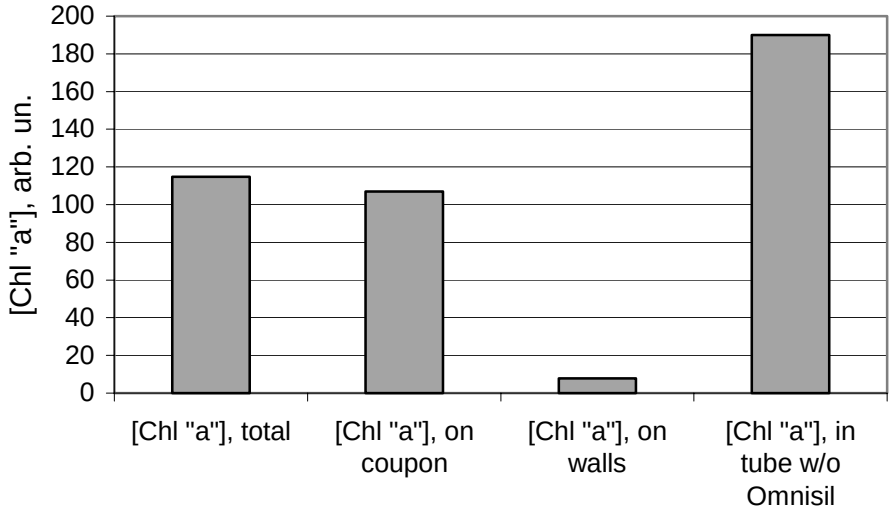

Fig.26. Distribution of chlorophyll (CB biomass) between Omnisil coupon and internal tube walls. Last column on right side represents chlorophyll yield in tubes w/o Omnisil coupon.

Figure 26 suggests that about 90 % of cyanobacterial biomass was concentrated on the Omnisil coupons. At this time, all cyanobacterial biomass has been represented as planktonic form, when cyanobacteria were grown w/o Omnisil coupon. The difference between chlorophyll yield in tubes with and w/o coupons can be explained by lower PAR in tubes with coupons.

Obtained result might suggest that the colonization of Omnisil coupons requires significant time (under our conditions no less than 4 days). Moreover, we assume that Omnisil presence in cultivation vessels induces the production of some exopolymers by cyanobacteria, which mediates the attachment of cyanobacterial cells to artificial substratum. It seems interesting that Omnisil, made also from silica, has so strong an effect on cyanobacterial attachment to its surface, while walls of glass tube do not appear to have such property. In conclusion: 1) the colonization of Omnisil surface is an auto-inducible and time-requiring process; 2) Omnisil coupons should be treated under pH control; 3) we would recommend the use of KOH for Omnisil coupon treatment, because NaOH is already added, when buffer is adjusted to pH about 8, and final sodium concentration may consequently exceed adaptation properties of cyanobacteria.

Subtask 1.4 Investigate the use of a hydraulic jump to improve the system’s overall CO_2 conversion efficiency

Based on the results collected and reported in previous reports, the experiments in this area have been discontinued. The Phase I experimental evaluation of the hydraulic jump is complete.

Subtask 1.5 Design harvesting subsystem

Our main activities this quarter were again focused on design and experimental work for the integrated screen wetting/ harvesting system, working primarily with Omnisil screens and 1.2. s.c.(2) organisms. Previous survivability tests for the 1.2. s.c.(2) organism indicated a need to change the organism loading method for bioreactor tests from pre-colonizing screens in a culturing vat to loading the screens by dripping the algae containing slurry on them while they are in place in the bioreactor.

The need to circulate algae-containing slurry through the system necessitated a design and testing effort to produce a header pipe for the screens that produces uniform flow across the whole membrane, is resistant to clogging, and allows flow control (so the system can be used for both gentle dripping for normal operation and high-flow organism removal for harvesting). The leading designs all use a header pipe with a flow control insert (Figure 27). Insert design 1 uses either holes or slots to set the flow area, while insert design 2 uses a shim/seal to set a pressure-controlled flow area. Both designs have shown acceptable performance results (little to no clogging, uniform flow, ability to load algae on to the screen), but tests have shown that slots have problems with manufacturability and thin shims do not have a low enough flow capability for the drip mode. Therefore, a staggered drilled-hole design and a thick shim design have been selected for additional testing. They will both be tested in the CRF-2 to see which performs the best over long durations, and the best performing design will be used for the pilot scale bioreactor screens.

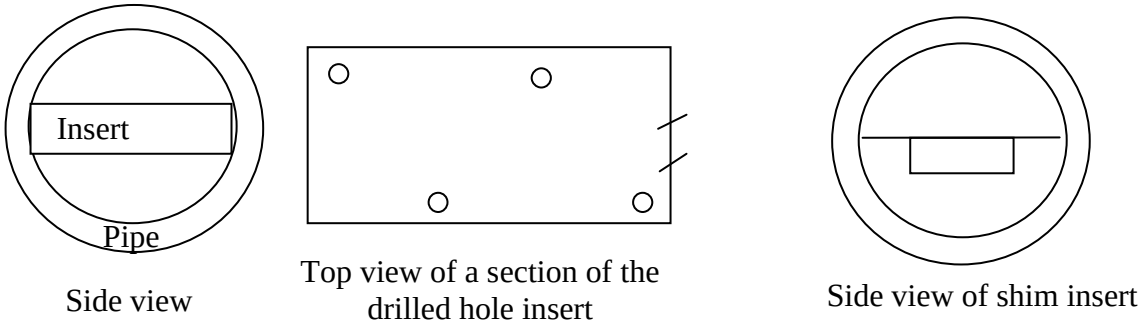

Fig. 27: Header pipe flow control inserts

Both designs do improve the distribution of water, but the results indicate that a more uniform distribution is achieved with the second “seal” or “pressure shim” design. A view of the distribution of the slurry on substrate using the “pressure shim” header is shown in Figure 28.

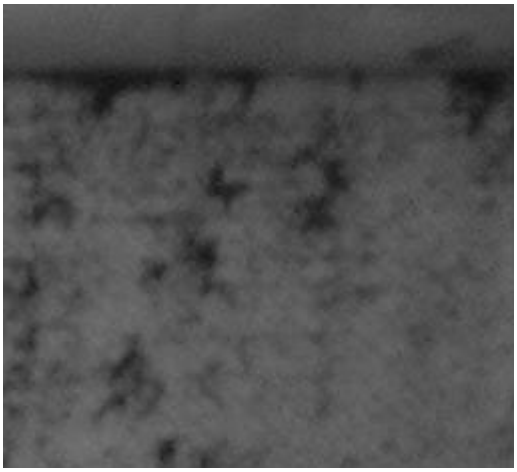

Fig. 28. Deposition of algal mass on substrate from the "pressure shim" header

Subtask 1.6 Quantify properties (higher heating value, elemental composition, volatile content) of dried biomass for potential end-uses.

We continue to search the literature and search for potential end uses for the biomass, but experiments in this area are on hold until we get closer to making a final decision for the organism.

Task 2.0. Evaluate subsystem combinations and select an "optimum" system design

The construction and procedural validation of the larger scale CRF (CRF-2) to be used as the primary test facility for evaluating the subsystem combinations was discussed earlier in the report as an extension of Subtask 1.1. The quantitative tests in the CRF-2 are scheduled to begin in October 2002.

Task 3.0. Implement the optimum system in scaled model

The phase 1 construction of the bioreactor has been completed, including the solar collector and light distribution system. The report from the pilot scale bioreactor group is copied below.

Over the course of Summer 2002, tests were conducted on the auxiliary systems of the pilot-scale unit such as the gas burner, the water pump, the immersion heater, and the fin-strip eater. It was noted that all the subsystems were operational and a weeklong run of the entire pilot-scale bioreactor was successful. However, we experienced a few bottlenecks while performing these test runs. A description of the bottlenecks and the solutions we came up with to clear up these bottlenecks is given next:

1. We came across a leak in the natural gas line (copper pipeline) and shut the gas off. We replaced the copper pipeline by a rigid black pipe gas line. We performed a leak test and found that there were no leaks.
2. The optical fibers used for transmission of light from the solar collector to the inside of the bioreactor chamber came loose (they were glued inside the light sheets in a specific pattern for optimum light distribution) due to high temperatures encountered inside the bioreactor chamber. We contacted Oak Ridge National Laboratories regarding this problem. ORNL replaced the light panels with high temperature resistant light panels. The new light panels were tested at the prescribed temperatures and we found that the light sheets were able to sustain the high temperatures.
3. We installed air filters in the compressed air line to prevent moisture from entering the gas burner.
4. A few minor leaks in the water tank and the bioreactor chamber were fixed.
5. The immersion heater inside the water tank was burned out owing to a fall in the water level during a test run. This immersion heater was replaced by a new one.
6. The main supply of natural gas to the 1005 East State St. facility was turned off. We contacted Guy Philips and arranged to have the gas turned on at all times to avoid burner shut down.

Group accomplishments

1. Regular maintenance of the solar collector and tracker by cleaning the collector surface and keeping the tracker moisture free.
2. We have investigated a Data Acquisition (DA) system and have come up with HOBO data loggers that seen to be a reliable and relatively inexpensive way to record the various parameters required for monitoring the performance of the pilot-scale unit. As of now, we are coming up with an inventory list of the actual loggers and the auxiliaries for setting up the DA system.
3. We conducted light intensity measurements for one light panel with solar illumination. The light distribution over the entire screen is in the acceptable illumination range, with specific results shown in Figure 29.

Action items

1. Set up DA system
2. Replace the water pump by one that will cause less harm to the algae while circulation of the algae through the bioreactor.
3. Repeat the light intensity measurement procedure for all the eight light sheets and compare the results to see if a uniform distribution of light is obtained.
4. Come up with way to keep the algae from settling at the bottom of the water tank once it is decided to introduce the algae into the system.
5. Prepare 7 membranes for the pilot-scale unit using the new shim design.
6. Run a weeklong test of the entire pilot-scale unit with all the subsystems running simultaneously.
7. If the weeklong test is successful, perform a 60-day long test run of the pilot-scale unit.

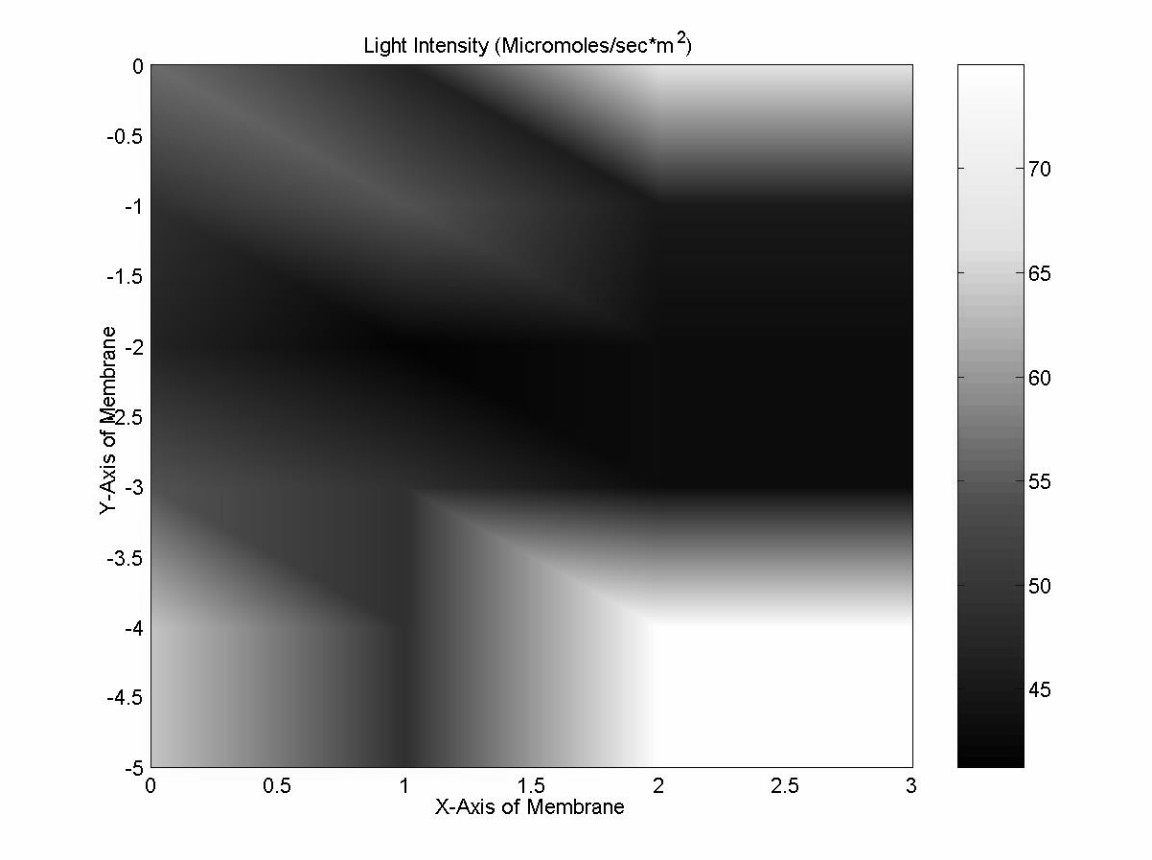

Fig. 29: Pilot scale screen solar illumination levels

Webpage

The web page is running at http://132.235.19.45/DOE . All parties involved in the project have received e-mail instructions and the password to access the information.

Conclusions

This report marks the end of year 2 of a three-year project as well as the milestone date for completion of Phase I activities. As indicated throughout the report, we have made significant progress towards achieving our overall project goals but we have slipped a little behind schedule. Therefore, to ensure that we meet the project goals by the end of year 3 we have made plans for a project "retreat" on October 26th to do a detailed status/review of all work currently being done and how it fits in the critical path for completing the project within the next year. For a detailed list of the specific results and accomplishments for the second quarter of 2002 see the abstract section of this report.