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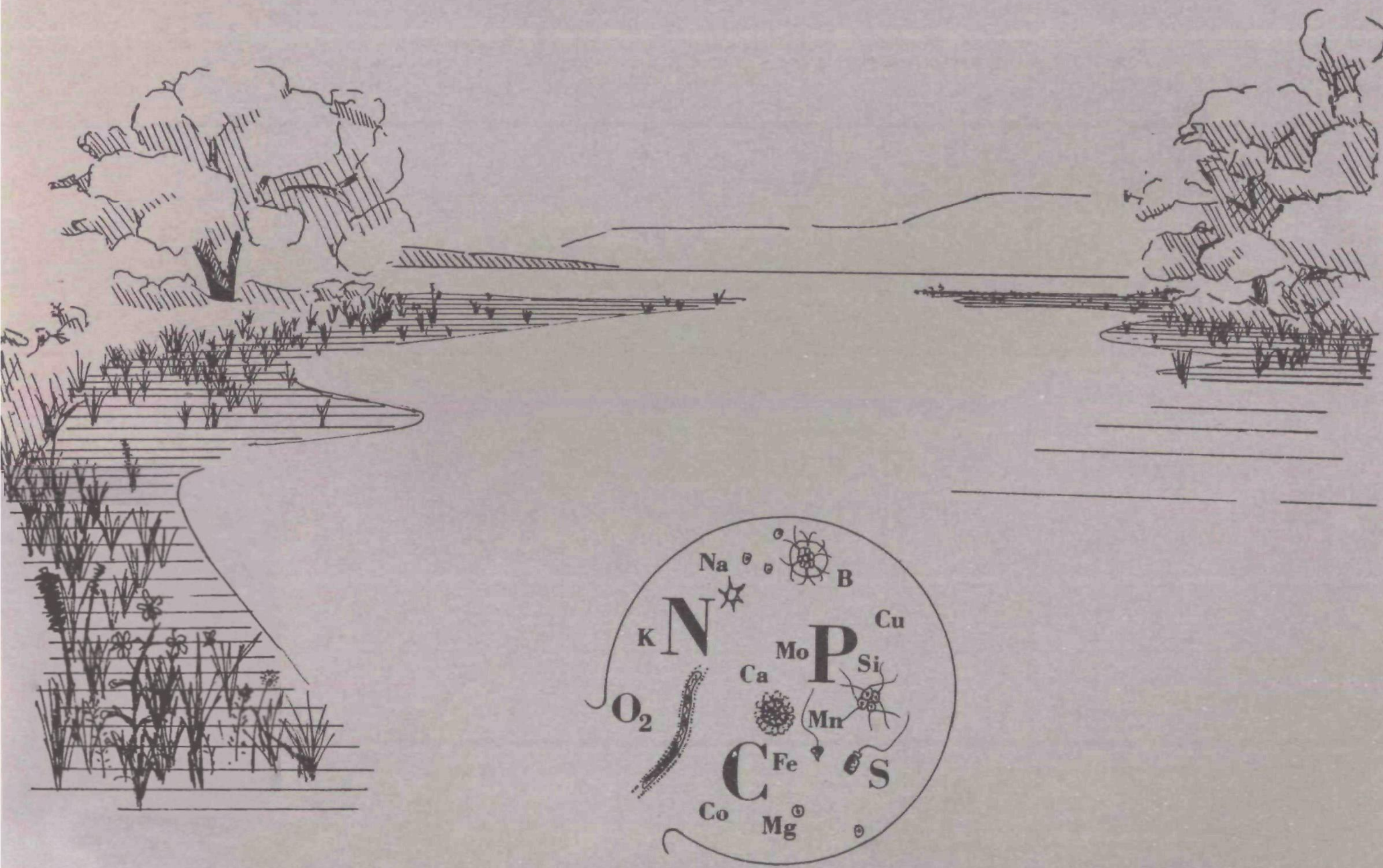
The Carbon Dioxide System and Eutrophication



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The Carbon Dioxide System and Eutrophication

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ABSTRACT

The objective of this research was to determine the feasibility of eutrophication control in natural bodies of water by the control of carbon.

Growth rates of the algae Chlorella, Microcystis, and Anabaena were studied with respect to carbon availability. Algae can utilize dissolved concentrations of CO_2 much lower than those from atmospheric equilibria. Control of algal growth by sweeping the CO_2 out by aeration with air containing very low concentrations of CO_2 is difficult because of atmospheric replenishment of CO_2. Bicarbonate is at least 50% utilized at growth rates as high as 7 mg/l/day. Atmospheric replenishment of CO_2, without any wind mixing, can sustain growth rates of 1.5 - 2 mg/l/day for depths of at least 1.7 meters.

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FIGURES

TABLES

SECTION I

CONCLUSIONS

1) Algae can efficiently utilize carbon dioxide at concentrations much lower than those present from atmospheric equilibria. It is very difficult to control growth by carbon dioxide control in systems open to the atmosphere, even when the carbon dioxide is swept out by aeration with air containing very low concentrations of carbon dioxide.

2) Bicarbonate is a good source of carbon and is at least 50% utilized at growth rates of at least 5 mg/l/d. Many lakes can have massive algal blooms using naturally present bicarbonate as the sole carbon source.

3) The atmosphere, without any vigorous wind mixing, is an adequate source of carbon dioxide for depths of at least 1.7 meters, permitting algal growth rates of up to 2 mg/l/d.

SECTION II

RECOMMENDATIONS

Much research is presently being done on nutrient limitation, such as with phosphorus and nitrogen, to limit algal growth. In addition to this, research is needed on ways to control the type of predominating algal species so that the troublesome blue-greens would be replaced by the less troublesome green algae. Though carbon is not likely to be limiting in most bodies of water, there is evidence that factors such as the ratios of various essential nutrients, including carbon, and physical conditions, can regulate the type of predominating algae, even though the total algal biomass may remain the same.

SECTION III

INTRODUCTION

The role of carbon in eutrophication has been mentioned numerous times over the years (Birge and Juday, 1911; King, 1970). Although it is probably generally agreed that carbon may be limiting for very high algal concentrations, such as in sewage lagoons (Bartsch and Allum, 1957), much controversy exists as to whether carbon may be limiting in eutrophic lakes where the algae concentrations, and therefore carbon needs, are much less. Kerr, Paris and Brockway, 1970; Lange, 1967; and Kuentzel, 1969, claim that bacterial oxidation of organics is necessary to provide the carbon necessary for algal blooms. Wright and Mills, 1967, found carbon to be somewhat limiting in their productivity studies on the Madison River.

This research was originally undertaken to determine whether eutrophication in small areas could be controlled by sweeping the carbon dioxide out of the water with low carbon dioxide air aeration. This would only be a slight modification of aeration methods that are currently being used to improve water quality in lakes and reservoirs (Symons, 1969; Wirth and Dunst, 1966).

A related question arises, namely, is carbon ever limiting in natural bodies of water? Since an algal bloom is of the order of 8 mg/l of algae (dry weight), and grows at most perhaps 2 mg/l/day, about 1 mg/l/day of carbon or about 4 mg/l/day of carbon dioxide are needed. This is the crux of the phosphorus versus carbon limiting nutrient question, that is whether atmospheric replenishment of carbon dioxide is rapid enough to permit algal blooms to occur or whether some other carbon source is needed.

Three major areas were studied. The first was the steady state, in which the growth rates of algae at various constant, maintained dissolved carbon dioxide concentrations were determined. These concentrations were those in equilibrium with atmospheric carbon dioxide concentrations and below, in contrast to much algae growth studies which use 1-5% enriched carbon dioxide air. The second was the non-equilibrium case where natural atmospheric replenishment was the sole carbon source. The third was the growth of algae with inorganic bicarbonate as the sole carbon source.

SECTION IV

METHODS

The algae used were Chlorella pyrenoidosa, Microcystis aeruginosa, and Anabaena circinalis. Allen's medium (Allen, 1952) was used for Chlorella, and ASM medium (McLachlan and Gorham, 1961) for Microcystis and Anabaena, and some of the Chlorella experiments as noted. Neither of these media contain any inorganic or organic carbon. Allen's medium contains 178 ppm nitrogen and 45 ppm phosphorus, while the corresponding values for ASM are 14 ppm and 3.1 ppm.

Growth was followed by spectrophotometric measurements at 600 mμ using a Bausch and Lomb Spectronic 20 with 12 mm diameter sample tubes. Dry weight measurements using 0.45 micron membrane filters were also made as a check on the absorbance measurements. An absorbance of 0.010 was equivalent to 6 mg/l dry weight for all three algae, and was linear over the concentration range of 3-20 mg/l reported here. A Beckman Model G pH Meter was used for pH measurements. All experiments were conducted indoors under continual fluorescent lighting of 90-120 foot candles and repeated three to five times, except as noted.

In the steady state experiments, air containing known quantities of carbon dioxide (15-340 ppm range) was bubbled through the algal suspensions at various flow rates, and the rate of growth determined. In this type of experiment the dissolved carbon dioxide concentration is governed by Henry's Law. This method has the advantage of not being dependent on inadequate and inaccurate methods for measuring the aqueous carbon dioxide at low concentrations. Various carbon dioxide concentrations were obtained by mixing air (340 ppm) and low carbon dioxide (15 ppm) air together in various ratios using flowmeters. Carbon dioxide was removed from air by bubbling through 2 N-NaOH. The air was laboratory air pumped with small aquarium air pumps. The usual supplies of compressed air were not used because of the possibility of contamination with dirt, metal oxide flakes and oil and grease. A Beckman GC-2 gas chromatograph with a thermal conductivity detector was used for measuring the carbon dioxide concentrations in the various air streams. The column was packed with 80-100 mesh silica gel. The column and detector temperatures were 170° C. Helium was the carrier gas. The instrument was standardized using analytical quality gases of known carbon dioxide concentrations. Most experiments were carried out in one-liter flasks containing 600 ml of algal suspension. The flasks were stoppered; a small tube in the stopper permitted the aeration air to escape and minimized the contact of atmospheric air with the algal suspension which would have upset the dissolved carbon dioxide equilibrium.

In the bicarbonate work, sodium bicarbonate was the sole carbon source. The carbon dioxide was first swept out of the suspension for at least two days by aeration with low carbon dioxide air, the bicarbonate added and the flask stoppered, and the growth as a function of time and the final equilibrium total growth were measured.

In the non-equilibrium experiments, though some smaller vessels were used, most of the work used 6 foot (1.8 m) high by 1 foot (0.31 m) in diameter plexiglas cylinders that were open to the atmosphere. A fluorescent light parallel to the side provided constant illumination of 250 foot-candles along the entire length of the cylinder.

The work was done in a laboratory which was originally designed for animal studies. The ventilation system brings in three-quarters outside air and recirculates one-quarter of the inside air. The air is changed a minimum of every twenty minutes. Gas chromatographic measurements showed the carbon dioxide concentration of the laboratory air was the same as the outside air.

SECTION V

RESULTS AND DISCUSSION

Most of the experiments, where possible, were carried through an algal concentration of at least 100 mg/l. Occasionally in the later growth stages of an experiment, some, apparently inorganic, precipitation occurred. No data taken when precipitation was occurring are included. Various unseeded control experiments were run using Allen's and ASM media in which the pH and bicarbonate concentrations were varied. No precipitation occurred in the pH ranges of interest in this work.

The original raw data are given in the Appendix while the calculated, summary data are given in this section. In some areas where limited experimental work was done, a few general conclusions are given only. Some other data are not included as they are very similar to other data in the tables.

All growth rates given are average growth rates over their respective algal concentration ranges. In all tables and figures, the aeration rate is the ratio of the volume of air per hour to the volume of algal suspension.

Equilibrium or steady state -- The goals were to, one, obtain basic data that other experiments could be compared with, and two, obtain preliminary data to show whether eutrophication could be controlled by low carbon dioxide aeration. The raw data are shown in Tables 9 through 30 in the Appendix. The calculated, summary data are shown in Figures 1 and 2 and Tables 1 through 5 in this part of the report.

Figure 1 shows the rate of growth for *Chlorella* and *Anabaena* as a function of the amount of air aeration. The rates level out after a certain aeration rate is reached. These growth rates, 15 mg/l/d for *Chlorella* and 7 mg/l/day for *Anabaena*, can be looked at as the rates of growth for maximum wind mixing under these conditions. *Microcystis* data, not shown, gave a maximum growth rate of 11 mg/l/day. Notice that Tables 1 through 5 have Cav/Cused ratios, the theoretically available carbon provided by the carbon dioxide in the aeration air to the carbon that the algae actually used in their growth. The tables also have higher algal concentration data while the figures show only the lower 3-20 mg/l range applicable to lake situations. For Chlorella, C_{av}/C_{used} decreased from 8.3 at the highest aeration rate to one at the steep part of the curve. For Anabaena, it did not decrease to one due to the slower growth which needed a smaller rate of carbon supply.

Figure 2 shows the growth rate of Chlorella as a function of the aeration rate and of the carbon dioxide concentration in the aeration air. As expected it is dependent on both. The C_{av}/C_{used} ratios, not shown in the tables but readily calculated from the figure, were about 1.5 throughout the aeration range where the curves are not flat. This shows the algae are very adept at utilizing very low concentrations of carbon dioxide, that is, no "minimum" concentration of carbon dioxide is necessary for algal uptake. The practical implications of this are apparent. To control eutrophication in a small area of a lake by aeration with low carbon dioxide air would necessitate using a low aeration rate. But a high aeration rate is needed to keep atmospheric replenishment of carbon dioxide from taking place. This will provide enough carbon dioxide for algal growth, unless the carbon dioxide concentration in the aeration air is actually zero, a difficult task on a large scale. This is what was found in swimming pool and other experiments that were open to the atmosphere. We have had no significant success in controlling algal growth with aeration with low carbon dioxide air, unless the atmosphere over the algae was also controlled.

Bicarbonate -- The raw data are shown in Tables 31 through 45 in the Appendix, while the calculated, summary data are shown in Tables 6 and 7 in this section. As the amount of bicarbonate increases, the fraction utilized decreases. This is reasonable since as bicarbonate is converted to carbon dioxide the pH rises; 50% conversion corresponds to conversion to carbonate and a pH of near 11, which we observed. Full conversion to carbon dioxide would produce sodium hydroxide, causing a very high pH that the algae could not tolerate. Jolliffe and Tregunna, 1970, working with marine algae, sea water, and low bicarbonate concentrations, found that the slow down in the growth rate as growth proceeds is more due to bicarbonate limitation than to the high pH. Tables 6 gives Chlorella results for both Allen's and ASM media. The difference in buffering capacities of these media may explain the different final pH's. Though we generally observed a higher growth rate in the middle stages than in the earlier stages of growth, it is clear that as bicarbonate is converted to carbonate, the growth slows. Table 7 indicates Microcystis to be somewhat better than Anabaena in

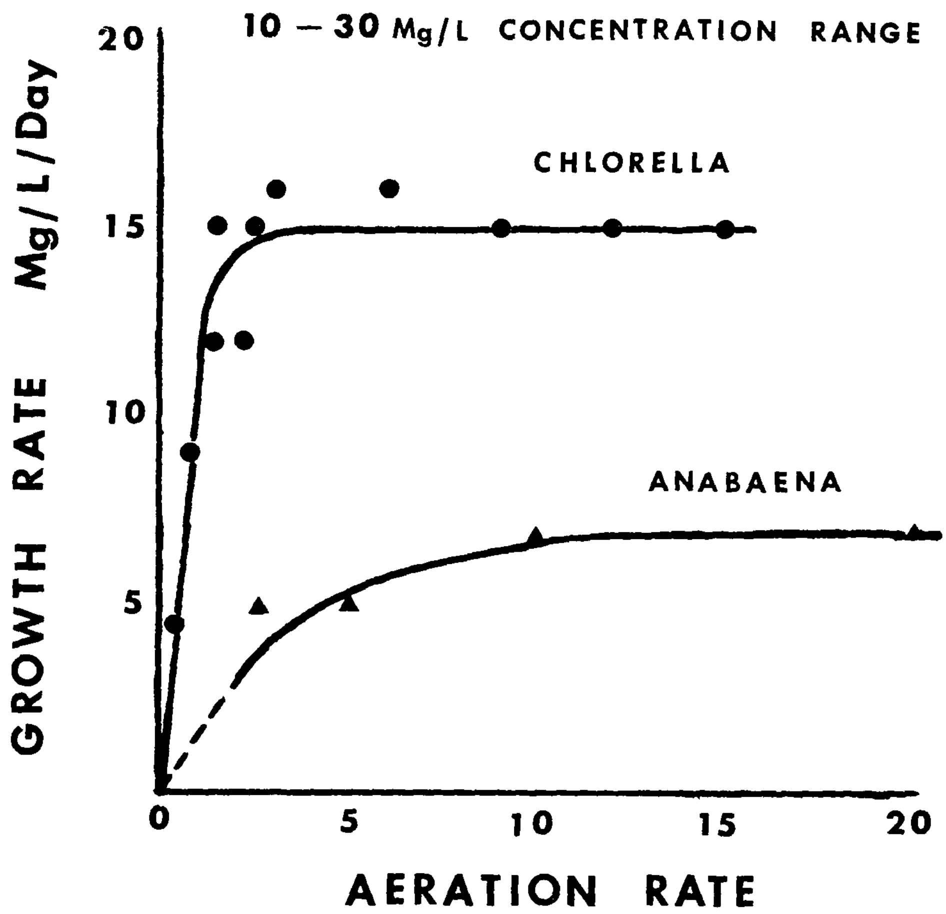

Figure 1. Growth of Chlorella and Anabaena versus air aeration rate -- volume air per hour to volume algal suspension.

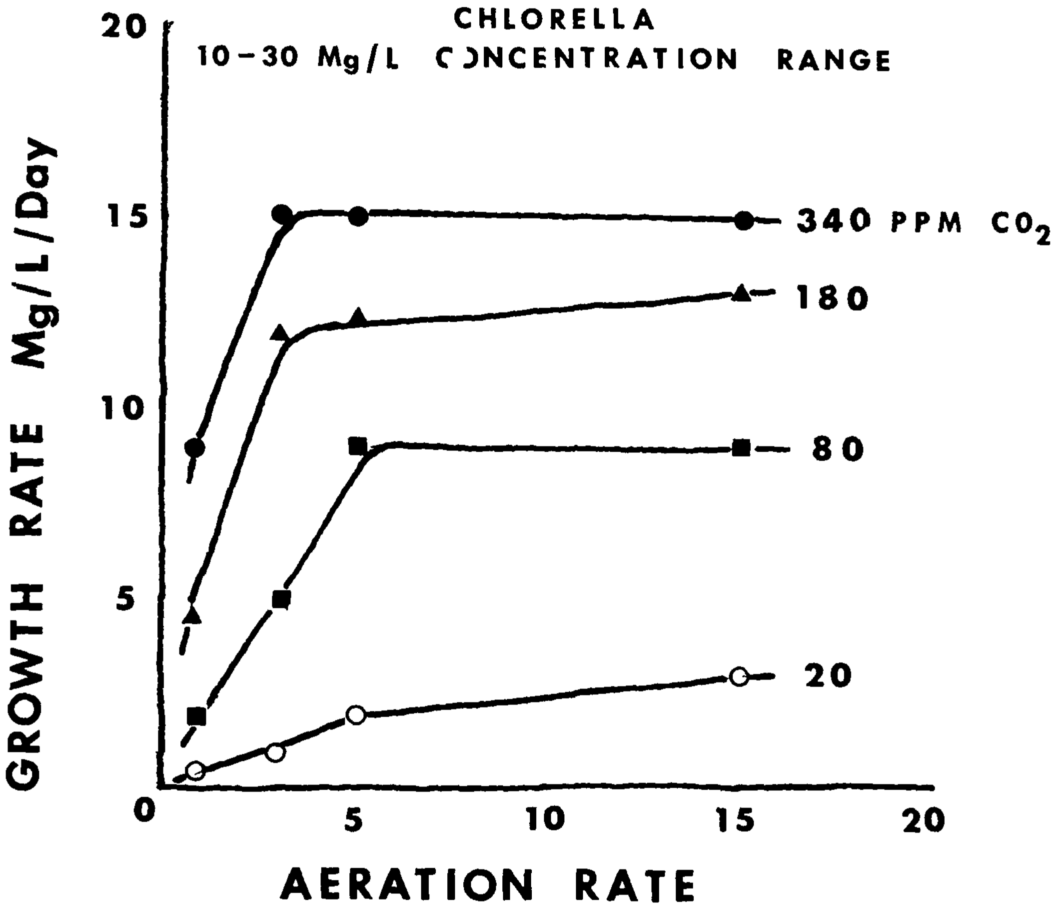

Figure 2. Growth of *Chlorella* versus aeration rate at various CO_2 concentrations in aeration air. Aeration rate is volume air per hour to volume algal suspension.

Table 1. Rate of growth of Chlorella versus air aeration rate

Aeration rate(1)	Concentration range 10 - 30 mg/l Rate mg/l/d	C_{av}/C_{used}(2)	Concentration range 60 - 150 mg/l Rate mg/l/d	C_{av}/C_{used}(2)
20.0	--	--	42	4.0
15.0	15	8.3	42	3.0
12.0	15	6.7	30	3.3
10.0	--	--	36	2.3
9.0	15	5.0	34	2.2
6.0	16	3.2	24	2.1
5.0	--	--	18	2.3
3.0	16	1.5	10	2.6
3.0	18	1.4	12	2.1
2.5	15	1.4	15	1.4
2.25	12	1.5	16	1.2
1.5	15	0.87	--	--
1.5	12	1.1	8	1.6
0.75	9	0.70	--	--
0.375	4	0.76	--	--

(1) Volume air per hour/volume algal suspension.
(2) Carbon in aeration air/carbon used by algal growth

Table 2. Growth rate of Chlorella versus CO_2 concentration at aeration rate of 15

CO_2 concentration ppm	Concentration range 10 - 30 mg/l Rate mg/l/d	C_{av}/C_{used}	Concentration range 60 - 150 mg/l Rate mg/l/d	C_{av}/C_{used}
344	15	8.3	42	3.0
180	13	5.3	30	2.1
111	--	--	19	2.1
81	9	2.8	21	1.4
56	9	2.2	9	2.2
45	6	2.7	--	--
31	2	--	--	--

Table 3. Growth rate of <u>Anabaena</u> versus CO_2 concentration at aeration rate of 15

CO_2 concentration ppm	Concentration range 10 - 30 mg/l Rate mg/l/d	C_{av}/C_{used}	60 - 150 mg/l Rate mg/l/d	C_{av}/C_{used}
344	6	21	22	5.6
180	7	8.9	18	3.5
111	8	5.3	18	2.3
81	5	5.6	9	3.3
56	5	4.1	9	2.2
45	6	2.7	8	1.9
31	5	2.3	--	--

Table 4. Growth rate of <u>Microcystis</u> versus CO_2 concentration at aeration rate of 15

CO_2 concentration ppm	Concentration range 10 - 30 mg/l Rate mg/l/d	C_{av}/C_{used}	60 - 150 mg/l Rate mg/l/d	C_{av}/C_{used}
344	10	11	33	3.8
180	>6	--	30	2.1
111	>6	--	27	1.5
81	>6	--	19	1.5
56	>6	--	12	1.7
45	7	2.3	10	1.6
31	5	2.3	--	--

Table 5. Rate of growth of <u>Anabaena</u> versus air aeration rate

Aeration rate	Concentration range 10 - 30 mg/l Rate mg/l/d	C_{av}/C_{used}	Concentration range 60 - 150 mg/l Rate mg/l/d	C_{av}/C_{used}
20.0	7	23	8	19
10.0	7	12	8	11
5.0	5	8	11	4
2.5	5	4	9	2.4

utilizing bicarbonate. In another experiment, successive additions of various amounts of bicarbonate, all to total 100 ppm of bicarbonate carbon after two weeks, were added to Allen's medium that had been seeded with Chlorella. The total growth was the same in all cases. For example, 100 ppm added initially had the same final effect as ten daily 10 ppm additions.

Many lakes with calcium carbonate containing sediments, have 10-40 ppm bicarbonate carbon. Since an algal bloom is about 8 mg/l, only about 8 mg/l of bicarbonate carbon is needed if 50% is utilized. The rates of growth we obtained are quite high, averaging around 4 mg/l/d, certainly higher than usually seen in eutrophic lakes.

This is in direct and startling disagreement with Kuentzel who claimed that algae cannot use bicarbonate at any appreciable rate, and is in general agreement with Guyomarch and Villeret, 1965, who found bicarbonate to stimulate growth when carbon dioxide concentrations were low.

<u>Non-equilibrium</u> -- Except under conditions of extreme wind mixing in shallow lakes, lakes are not necessarily in equilibrium with respect to the sediments, water, and atmosphere (Morton and Lee, 1968). Two questions are apparent. One, what is the rate of atmospheric carbon dioxide replenishment, with no stirring or mixing, for depths greater than the typical laboratory glassware? The second is, how well is bicarbonate utilized when the water is open to the atmosphere containing 0.034% carbon dioxide, in comparison to a closed atmosphere over a bicarbonate solution which can theoretically permit a higher gaseous carbon dioxide concentration?

Table 6. Growth of Chlorella with sodium bicarbonate as sole carbon source

	Bicarbonate-C mg/1	Total growth mg/1	Growth rate[1] mg/1/d	% C used	Final pH
A	10	23	4	100	8.5
B	10	16	3	80	10.1
A	25	42	6	84	8.7
B	25	24	3	48	10.4
A	50	48	7	48	8.7
B	50	42	4	42	10.6
A	100	60	10	30	8.9
B	100	72	8	36	10.7

A - Allen's Medium
B - ASM Medium

(1)Average rate over 3-20 mg/1 algal concentration range.

Table 7. Growth of Anabaena and Microcystis in ASM Medium with sodium bicarbonate as sole carbon source

	Bicarbonate-C mg/1	Total growth mg/1	Growth rate[1] mg/1/d	% C used	Final pH
An	5	9	1	90	10.5
M	5	9	2	90	10.6
An	10	11	2	55	10.4
M	10	15	3	75	10.8
An	25	27	4	54	10.7
M	25	36	5	72	10.8
An	50	48	6	48	10.8
M	50	60	7	60	10.9
An	100	72	6	36	10.5
M	100	126	7	62	10.8

An - Anabaena
M - Microcystis

(1) Average rate over 3-20 mg/1 algal concentration range

The 6 ft (1.8 m) high by 1 ft (0.31 m) in diameter open plexiglass cylinders were used in this phase of the research.

Algal growth experiments were conducted to determine whether bacterial degradation of the plexiglass could provide significant amounts of carbon dioxide. No differences between the experiments with and without plexiglass were seen.

The Allen's medium in the cylinders was initially equilibrated by bubbling air through overnight before being seeded with Chlorella. The algal suspensions were stirred for 2 minutes once a day before sampling. To determine whether this stirring may have caused a significant amount of carbon dioxide to go into solution two checks were made. One cylinder was stirred the usual way and another let grow for a week without disturbance. Both grew at the same rate. Another check was that no change in pH could be detected before and after stirring. In some experiments air was blown across the top ("top air ventilation") of the algal suspensions. There was not enough wind to cause any noticeable water movement or waves. Data in this section for "no mixing" should be interpreted as for no air movement or just very gentle air movement over the surface of the algal suspension. A few of the bottom, bubble aeration experiments were not quantitative regarding aeration rates. This is noted in the tables by the absence of any given aeration rate.

The raw data are given in Tables 46 through 53 in the Appendix while the calculated summary data are shown in Table 8 in this section. The algae grew at a surprisingly rapid rate of about 1.5-2 mg/l/d in the 3-15 mg/l algal concentration range. Much of the time the growth was unsteady. This growth rate is an average value of all experiments and is also quite conservative, as we often observed faster rates. With 20 ppm bicarbonate carbon, also open to the atmosphere, the rate increased to about 7 mg/l/d. The pH's during growth were 7.9 with no bicarbonate, and 8.3-8.8 with bicarbonate. One of the cylinders had a stopcock at the bottom which facilitated bottom sampling. The pH's at the top and bottom were the same in all phases of the various experiments. The pH's of the bottom aeration cylinders were lower, as expected, because of the atmospheric carbon dioxide in the aeration air. Considerable amounts of data, not given, for Chlorella, Microcystis, and Anabaena, in shallower vessels of 0.15-0.31 m depth, open to the atmosphere, showed growth rates of 3-7 mg/l/day with no bicarbonate and 4-9 mg/l/d with bicarbonate present. For any given experiment, the bicarbonate growth rate was always higher.

It is not realistic with experiments that are open to the atmosphere to concern ourselves with the percent utilization of bicarbonate carbon, since it will re-equilibrate with the atmospheric or respiratory carbon dioxide and revert to bicarbonate. Thus it becomes a kinetic question of bicarbonate utilization versus atmospheric or respiratory replenishment.

The rates observed for depths up to 5.5 ft (1.7 m), about 1.5-2 mg/l/d without bicarbonate and 7 mg/l/d with, are all greater than seen in eutrophic lakes. King, 1970, feels that bicarbonate and respiratory carbon dioxide are the major sources of photosynthetic carbon, and that atmospheric carbon dioxide is minor. Our data shows that atmospheric replenishment of carbon dioxide provides a sufficient supply for algal blooms, for at least 5.5 ft (1.7 m) of depth and no mixing. No mixing does not mean that the rate of carbon dioxide supply is controlled by true molecular diffusion. The algae themselves cause some mixing by their occasional up and down movements that we observed at numerous times, and furthermore, small scale eddy currents are undoubtedly present.

Table 8. Rate of growth[1] of Chlorella in vessels[2] open to the atmosphere

Depth meters	No bicarbonate mg/l/d	20 ppm bicarbonate-C mg/l/d
0.31	3	6
0.93	2	-
1.5	1.5-2	7
1.7	1.5-2	7

[1] Average rate over 3-15 mg/l algal concentration range

[2] No mixing or stirring except when sampled

These experiments were conducted under rather ideal growth conditions, with the idea that if atmospheric replenishment is sufficient to permit a high growth rate, it is even more likely to be sufficient under lake conditions where the algae grow slower needing a lower rate of carbon dioxide supply, and where there is usually some wind mixing.

Other areas of study -- A few experiments using 16 liter jugs were conducted to determine whether air blown over the top of an algal suspension would speed up growth compared to the algal suspension being open to a calm atmosphere. No differences were seen, even with C_{av}/C_{used} ratios as high as ten. This gives evidence that calm air provides as much useable carbon dioxide as does a gentle wind that does not cause waves or mixing. Growth rates for Chlorella were about 4 mg/l/d for one-foot of depth.

Bottom bubble aeration or mixing will, of course, speed up growth in deep vessels; for shallow vessels such as typical laboratory glassware, it makes little difference. In our 16-liter jug experiments (Tables 54, 55 in the Appendix), the rates of growth in the low (5-20 mg/l) algal concentration range were similar for bubbled and non-aerated open vessels. In the high concentration range (>60 mg/l), the bubbled grew about twice as fast as the non-mixed.

Some data for bottom aeration, top air ventilation, and cotton stoppered flasks are shown in Tables 56 through 61 in the Appendix. These data, though not relevant in lake situations, are useful in interpreting laboratory work. The bottom aeration flow rates are not quantitative, but are in great excess regarding the carbon dioxide needs of the growing algae. Growth rates were similar for top aeration and for the cotton stoppered flasks, and were often similar for bottom aeration in half or less full 250 ml, 500 ml, and 1000 ml erlenmeyer flasks in the lower algal concentration ranges.

Other experiments, also using laboratory sized glassware, were done using intermittent aeration. Here the algae were aerated for a specified time and then the flask was stoppered until the next aeration. These data are shown in Tables 62 through 65 in the Appendix. Generally, for 20 minute aeration periods once a day and once every 3 days, C_{av}/C_{used} was approximately equal to one. For 14, 21, and 40 day intervals, C_{av}/C_{used} was somewhat less than one, reflecting a small amount of leakage of air into the flasks, probably during sampling. Another group of experiments (Tables 66 and 67) used 1, 4, and 8 hour aeration daily. The rates of growth and total amounts of growth were often similar for all three cases. Also, for the blue-green algae, continuous aeration and one hour daily aeration often gave similar growth. It is interesting to speculate from these data, whether continual wind mixing in a lake would permit more algal growth than just occasional wind mixing.

The fraction of dissolved bicarbonate that can be used by algae is dependent on the buffer capacity of the media or lake. This is because as the buffer capacity increases, more hydroxide ion can be accepted by the solution without the pH becoming too high for algal growth. This will permit a greater conversion of bicarbonate and carbonate to carbon dioxide. Some experiments were conducted using Chlorella in ASM medium with the phosphate replaced by a phosphate buffer. For varying amounts of buffer, 7-65 ppm or 5-45 ppm as phosphate, the percent bicarbonate carbon utilized was 60-100% for 10 ppm carbon, 40-80% for 20 ppm carbon, and 40-60% for 40 ppm carbon. The final pH's were all between 10.1 and 10.5. Since lake waters generally have low buffer capacities, we think that in most lakes, the utilization of bicarbonate is about 50%, corresponding to conversion to carbonate with a final pH of 10-11.

SECTION VI

ACKNOWLEDGEMENTS

We thank Dr. George Fitzgerald of the University of Wisconsin, a consultant on this project, for helpful discussions and for providing the algae samples.

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SECTION VII

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SECTION VIII

PUBLICATIONS

1) Morton, S. D., Sernau, R. C., and Derse, P. H. "Carbon and Eutrophication". Presented at the Water Pollution Control Federation Conference in Boston, Massachusetts, October 5, 1971.

2) Morton, S. D., Sernau, R. C., and Derse, P. H. "Natural Carbon Sources and Rate of Replenishment in Lakes". Presented at the American Society of Limnology and Oceanography Symposium on "The Limiting Nutrient Controversy", W. K. Kellogg Biological Station, Michigan State University, Feb. 12, 1971.

3) Morton, S. D., Sernau, R. C., and Derse, P. H. "Natural Carbon Sources and Algal Growth". Presented at the American Chemical Society Meeting, Washington, D. C., September 14, 1971.

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SECTION IX

APPENDIX

Table 9. Growth of *Chlorella* under steady state CO_2 concentrations. Aeration rate of 15

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm	180 ppm	15 ppm
	Optical density		
0 days	0.0010	0.0010	0.0010
6	0.125	0.080	0.015
7	0.170	0.130	0.015
8	0.230	0.175	0.018
9	0.260	0.195	0.015
12	0.365	0.320	0.050
13	0.380	0.350	0.040
14	0.420	0.390	0.055
16	0.460	0.450	0.060
19	0.500	0.520	0.075
0	0.0013	0.0013	0.0013
5	0.010	0.010	0.002
6	0.012	0.012	0.002
7	0.026	0.032	0.002
10	0.130	0.115	0.005
11	0.200	0.175	0.008

Table 10. Growth of *Chlorella* under steady state CO_2 concentrations. Aeration rate of 15

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm	111 ppm	15 ppm
	Optical density		
0 days	0.0010	0.0010	0.0010
6	0.115	0.080	0.005
7	0.180	0.115	0.020
8	0.250	0.155	0.025
9	0.300	0.180	0.020
12	0.500	0.270	0.045
13	0.550	0.305	0.050
14	0.600	0.330	0.050
16	0.700	0.360	--
19	0.800	0.430	--

Table 11. Growth of <u>Chlorella</u> under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm	81 ppm	15 ppm
	Optical density		
0 days	0.0008	0.0008	0.0008
5	0.025	0.019	0.009
6	0.035	0.019	0.009
7	0.075	0.040	0.010
8	0.132	0.055	0.009
9	0.240	0.080	0.010
12	0.375	0.152	0.020
13	0.410	0.200	0.030
14	0.510	0.225	0.035
15	0.570	0.225	0.050
16	0.630	0.290	0.060
19	0.720	0.350	0.075
20	0.750	0.375	0.085
21	0.800	0.390	0.090

Table 12. Growth of <u>Chlorella</u> under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm	56 ppm	15 ppm
	Optical density		
0 days	0.0027	0.0027	0.0027
4	0.045	0.020	0.005
7	0.190	0.068	0.015
9	0.330	0.090	
11	0.410	0.120	0.018
12	0.440	0.120	0.019
13	0.46	0.140	0.022
14	0.47	0.160	0.035
15	0.48	0.18	0.030
16	0.48	0.19	0.030
17	0.48	0.20	0.040
18	0.48	0.23	0.040
22	0.46	0.27	0.050
23	0.48	0.30	0.060

Table 13. Growth of Chlorella under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm	45 ppm	15 ppm
	Optical density		
0 days	0.0010	0.0010	0.0010
5	0.070	0.020	0.005
6	0.125	0.030	0.010
7	0.180	0.050	0.015
8	0.220	0.050	0.015
12	0.355	0.075	0.015
13	0.385	0.095	0.025
14	0.400	0.095	0.020
15	0.430	0.105	0.020
18	0.490	0.130	0.023

Table 14. Growth of Chlorella under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm	31 ppm	15 ppm
	Optical density		
0 days	0.0027	0.0027	0.0027
4	0.040	0.010	0.010
7	0.220	0.020	0.015
9	0.350	0.021	0.015
11	0.43	0.034	0.023
12	0.44	0.038	0.025
13	0.43	0.040	0.021
14	0.48	0.048	0.025
15	0.49	0.050	0.025
16	0.49	0.050	0.035
17	0.49	0.065	0.030
18	0.50	0.070	0.030
22	0.50	0.100	0.045
23	0.50	0.105	0.045

Table 15. Growth of *Chlorella* under steady state CO_2 concentrations. Aeration rate of 5.

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm Average of 4 experiments	180 ppm	111 ppm	81 ppm	56 ppm
	Optical density				
0 days	0.007	0.007	0.007	0.007	0.007
3	0.070	0.045	0.050	0.040	0.040
4	0.15	0.070	0.070	0.060	0.050
7	0.29	0.14	0.12	0.11	0.080
8	0.30	0.16	0.15	0.13	0.10

Growth time	Air-344 ppm Average of 2 experiments	45 ppm	35 ppm	15 ppm
0 days	0.001	0.001	0.001	0.001
4	0.037	0.030	0.020	0.010
5	0.090	0.040	0.030	0.012
6	0.15	0.040	0.030	0.010
7	0.19	0.055	0.040	0.015
8	--	0.060	0.040	0.015
11	--	0.085	0.060	0.020
12	--	0.095	0.060	0.020

Table 16. Growth of Chlorella under steady state CO_2 concentrations. Aeration rate of 3.

Concentration of CO_2 in aeration air

Growth time	Air-344 ppm Average of 4 experiments	180 ppm	111 ppm	81 ppm	56 ppm	15 ppm
	Optical density					
0 days	0.001	0.001	0.001	0.001	0.001	0.001
4	0.015	0.015	0.010	0.005	0.005	0.005
6	0.090	0.055	0.045	0.020	0.030	0.007
7	0.17	0.070	0.060	0.030	0.040	0.009
8	0.25	0.090	0.060	0.040	0.045	0.017
11	--	0.12	0.070	0.055	0.060	0.015

Growth time	Air-344 ppm Average of 2 experiments	45 ppm	35 ppm	15 ppm
0 days	0.001	0.001	0.001	0.001
4	0.075	0.025	0.020	0.015
5	0.14	0.025	0.025	0.015
6	0.18	--	0.025	0.015
7	0.23	0.030	0.030	0.015
8	0.26	0.035	0.030	0.020
11	--	0.040	0.040	0.025

Table 17. Growth of Chlorella under steady state CO_2 concentrations. Aeration rate of 0.75.

Growth time	Concentration of CO_2 in aeration air: Air-344 ppm Average of 4 experiments	180 ppm	111 ppm	81 ppm	56 ppm	15 ppm
	Optical density					
0-1 days	0.010	0.010	0.010	0.010	0.010	0.010
3	0.050	0.030	0.035	0.025	0.030	0.015
4	0.080	0.045	0.035	0.035	0.035	0.020
6	0.14	0.060	0.045	0.040	0.040	0.020
7	0.17	0.065	0.045	0.045	0.040	0.020
11	0.27	0.070	--	0.045	--	--
14	--	0.085	0.050	0.060	--	--
17	--	0.10	0.065	0.070	--	--

Growth time	Air-344 ppm Average of 2 experiments	45 ppm	35 ppm	15 ppm
0 days	0.001	0.001	0.001	0.001
2	0.010	0.005	0.001	0.005
5	0.035	0.010	--	0.015
6	0.050	0.015	0.015	--
7	0.080	0.020	--	--
8	0.11	--	0.015	--
9	0.15	0.020	0.015	--
12	0.21	0.020	0.020	--

Table 18. Growth of Chlorella versus air aeration rate

Optical Density

Growth time	Aeration rate 3.0	1.5	0.75	0.375
0 days	0.001	0.001	0.001	0.001
4	0.010	0.010	0.010	0.010
7	0.030	0.035	0.030	0.030
8	0.060	0.055	0.045	0.035
9	0.080	0.075	0.060	0.050
10	0.115	0.090	0.060	0.050
11	0.140	0.110	0.080	0.055
14	0.210	0.140	0.100	0.065
15	0.240	--	0.105	0.065
16	--	--	0.11	0.070
17	--	--	--	0.075

Table 19. Growth of Chlorella versus air aeration rate

Optical Density

Growth time	Aeration rate 12.0	6.0	3.0	1.5
0 days	0.001	0.001	0.001	0.001
4	0.015	0.015	0.015	0.015
5	0.030	0.030	0.040	0.030
6	0.060	0.070	0.070	0.060
7	0.11	0.11	0.090	0.065
8	0.18	0.16	0.12	0.080
11	0.30	0.25	0.17	0.11
12	0.31	0.29	0.19	0.12
13	0.35	0.32	0.20	0.13

Table 20. Growth of Chlorella versus air aeration rate

Growth time	Optical Density Aeration rate 20.0	 10.0	 5.0	 2.5
0 days	0.001	0.001	0.001	0.001
6	0.13	0.08	0.06	0.045
7	0.21	0.15	0.11	0.07
8	0.27	0.21	0.15	0.09
9	0.37	0.28	0.20	0.13
12	--	0.36	0.29	0.19

Table 21. Growth of Chlorella versus air aeration rate

Growth time	Optical Density Aeration rate 20.0	 10.0	 5.0	 2.5
0 days	0.003	0.003	0.003	0.003
2	0.012	0.008	0.010	0.008
3	0.030	0.018	0.023	0.019
4	0.090	0.070	0.075	0.060
7	0.23	0.22	0.20	0.14
8	0.27	0.27	0.24	0.16
9	0.29	0.31	0.28	0.20
10	0.31	0.34	0.31	0.21

Table 22. Growth of Microcystis under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Concentration of CO_2 in aeration air		
	Air-344 ppm	180 ppm	15 ppm
	Optical density		
0 days	0.0080	0.0080	0.0080
7	0.075	0.075	0.015
8	0.105	0.115	0.035
9	0.145	0.150	--
10	0.215	0.210	0.020
11	0.325	0.280	0.030
14	0.450	0.460	0.040
	Air-344 ppm	111 ppm	15 ppm
0	0.0080	0.0080	0.0080
7	0.080	0.080	0.015
8	0.115	0.120	0.030
9	0.155	0.160	0.030
10	0.245	0.215	0.030
11	0.310	0.260	0.030
14	0.450	0.350	--

Table 23. Growth of *Microcystis* under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Air-344 ppm	81 ppm	15 ppm
	Optical density		
0 days	0.0080	0.0080	0.0080
7	0.075	0.090	0.030
8	0.100	0.125	0.030
9	0.120	0.160	0.035
10	0.190	0.200	0.050
11	0.250	0.230	0.060
14	0.400	0.310	0.060
15	0.440	0.330	0.060
16	0.500	0.350	0.070
17	0.550	0.380	0.070
	Air-344 ppm	56 ppm	15 ppm
0	0.0080	0.0080	0.0080
7	0.110	0.085	0.010
8	0.155	0.110	0.020
9	0.210	0.125	0.015
10	0.260	0.160	0.020
11	0.335	0.180	0.020
14	0.500	0.230	0.020
15	0.525	0.240	--
16	0.560	0.280	0.030
17	0.600	0.300	0.040

Concentration of CO_2 in aeration air (column heading above ppm values).

Table 24. Growth of *Microcystis* under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Concentration of CO_2 in aeration air		
	Air-344 ppm	45 ppm	15 ppm
	Optical density		
0 days	0.0080	0.0080	0.0080
4	0.020	0.035	0.020
5	0.040	0.050	0.023
6	0.050	0.060	0.020
8	0.13	0.088	0.030
10	0.27	0.11	0.035
11	0.34	0.12	0.045
12	0.44	0.14	0.040
13	0.45	0.17	0.045
14	0.50	0.18	0.045
	Air-344 ppm	31 ppm	15 ppm
0	0.0080	0.0080	0.0080
4	0.030	0.030	0.030
5	0.050	0.040	0.040
6	0.070	0.045	0.045
8	0.18	0.065	0.060
10	0.34	0.085	0.075
11	0.42	0.100	0.070
12	0.49	0.100	0.075
13	0.54	0.12	0.090
14	0.59	0.14	0.090

Table 25. Growth of *Microcystis* under steady state CO_2 concentrations. Aeration rate of 0.75.

	Concentration of CO_2 in aeration air			
	Air-344 ppm	56 ppm	35 ppm	15 ppm
Growth time		Optical density		
0 days	0.005	0.005	0.005	0.005
7	0.015	0.015	0.015	0.015
8	0.019	0.015	0.015	0.015
9	0.022	0.015	0.018	0.015
10	0.035	0.015	0.018	0.015
13	0.065	0.023	0.020	0.015
14	0.075	0.023	0.020	0.015
15	0.080	0.027	0.022	0.015
16	0.085	--	--	0.015
17	0.090	0.028	0.024	0.015
20	0.12	0.030	--	0.020

Table 26. Growth of *Anabaena* under steady state CO_2 concentrations. Aeration rate of 15.

	Concentration of CO_2 in aeration air		
	Air-344 ppm	180 ppm	15 ppm
Growth time	Optical density		
0 days	0.0070	0.0070	0.0070
3	0.025	0.020	0.015
4	0.025	0.030	0.015
5	0.050	0.035	0.015
6	0.045	0.055	0.015
7	0.060	0.085	0.025
10	0.130	0.175	0.030
11	0.160	0.220	0.030
12	0.190	0.240	0.030
13	0.210	0.250	0.030
14	0.250	0.300	0.030
	Air-344 ppm	111 ppm	15 ppm
0	0.0070	0.0070	0.0070
3	0.010	0.010	0.010
4	0.030	0.025	0.020
5	0.055	0.030	0.020
6	0.075	0.050	0.025
7	0.120	0.080	0.040
10	0.260	0.180	0.060
11	0.320	0.200	0.060
12	0.350	0.220	0.060
13	0.380	0.250	0.060
14	0.430	0.260	--

Table 27. Growth of *Anabaena* under steady state CO_2 concentrations. Aeration rate of 15.

Growth time	Air-344 ppm	81 ppm	15 ppm
	Concentration of CO_2 in aeration air		
	Optical density		
0 days	0.0070	0.0070	0.0070
4	0.030	0.030	0.020
7	0.055	0.050	0.025
10	0.085	0.088	0.028
11	0.110	0.120	0.030
14	0.200	0.175	0.040
17	0.305	0.220	0.040
18	0.320	0.230	0.045
19	0.350	0.240	0.050
20	0.370	0.280	--
21	0.390	0.270	--
0	0.0070	0.0070	0.0070
7	0.030	0.030	0.020
9	0.055	0.055	0.030
10	0.055	0.060	0.030
11	0.070	0.075	0.030
15	0.110	0.140	0.035
16	0.155	0.160	0.045
18	0.220	0.195	0.050
21	0.365	0.220	0.055
22	0.410	0.240	0.070
25	0.450	0.260	--

Table 28. Growth of *Anabaena* under steady state CO_2 concentrations. Aeration rate of 15.

	Concentration of CO_2 in aeration air		
	Air-344 ppm	56 ppm	15 ppm
Growth time	Optical density		
0 days	0.0070	0.0070	0.0070
4	0.030	0.030	0.020
7	0.048	0.048	0.020
10	0.072	0.072	0.022
11	0.095	0.090	0.025
14	0.190	0.120	0.025
17	0.310	0.155	0.025
18	0.320	0.160	0.025
19	0.350	0.160	0.030
20	--	0.185	0.030
21	--	0.185	--
0	0.0070	0.0070	
7	0.030	0.040	
9	0.040	0.055	
10	0.060	0.080	
11	0.070	0.090	
15	0.080	0.140	
16	0.120	0.160	
18	0.225	0.200	
21	0.310	0.240	
22	0.370	0.240	
25	0.450	0.250	

Table 29. Growth of *Anabaena* under steady state CO_2 concentrations. Aeration rate of 15.

	Concentratian of CO_2 in aeration air		
	Air-344 ppm	45 ppm	15 ppm
Growth time	Optical Density		
0 days	0.0070	0.0070	0.0070
7	0.035	0.030	0.020
9	0.040	0.050	0.025
10	0.060	0.070	0.040
11	0.060	0.080	0.040
15	0.080	0.125	0.040
16	0.100	0.145	0.045
18	0.160	0.180	0.055
21	0.265	0.210	0.065
22	0.320	0.220	--
25	0.370	0.225	--
	Air-344 ppm	31 ppm	15 ppm
0	0.0070	0.0070	0.0070
7	0.025	0.030	0.015
9	0.050	0.050	0.020
10	0.050	0.050	0.025
11	0.050	0.060	0.030
15	0.100	0.080	--
16	0.130	0.100	0.025
18	0.220	0.110	0.030
21	0.320	0.115	0.035

Table 30. Growth of *Anabaena* versus air aeration rate.

Growth time	Optical density			
	Aeration rate			
	20.0	10.0	5.0	2.5
0 days	0.003	0.003	0.003	0.003
4	0.015	0.015	0.015	0.020
5	0.025	0.025	0.025	0.025
6	0.030	0.030	0.030	0.035
7	0.040	0.045	0.035	0.045
8	0.060	0.060	0.050	0.050
11	0.11	0.10	0.080	0.070
12	0.13	0.10	0.090	0.080
13	0.16	0.11	0.11	0.080
15	0.18	0.11	0.11	0.10
18	0.19	0.15	0.18	0.13
19	0.22	0.19	0.21	0.16

Table 31. Growth of Chlorella in Allen's medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flask

Growth time	ppm $NaHCO_3$-C: 0 (control)	50	100
	Optical density		
0 days	0.015	0.015	0.015
3	0.015	0.030	0.030
7	0.015	0.090	0.095
9	0.020	0.105	0.105
13	0.020	0.090	0.105
17	$NaHCO_3$ added again		
		100 ppm cumulative	200 ppm cumulative
17	0.020	0.10	0.12
20	0.020	0.10	0.13
27	0.020	0.16	0.20
31	0.020	0.16	0.22
34	0.020	0.16	0.23
36	0.020	0.16	0.23
37	50 ppm $NaHCO_3$-C added to control		
41	0.070		
42	0.080		
44	0.10		

Table 32. Growth of Chlorella in Allen's medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flask.

	ppm $NaHCO_3$-C				
	0 (control)	25	50	100	Cotton stoppered control
Growth time	Optical density				
0 days	0.001	0.001	0.001	0.001	0.001
2	0.005	0.015	0.015	0.020	0.010
5	0.015	0.080	0.080	0.080	0.040
9	0.015	0.070	0.080	0.090	0.045
12	0.020	0.10	0.12	0.12	0.11
16	0.020	0.10	0.13	0.13	0.10
Same but one-liter flasks full					
0 days	0.001	0.001	0.001	0.001	
2	0.005	0.010	0.010	0.015	
5	0.020	0.050	0.050	0.060	
9	0.015	0.065	0.050	0.065	
12	0.020	0.065	0.075	0.085	
16	0.020	--	0.090	0.090	
20	0.015	--	0.090	0.095	

Table 33. Growth of *Chlorella* in Allen's medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flask.

	ppm $NaHCO_3$-C		
	0 (control)	10	25
Growth time	Optical density, pH		
0 days	0.0025	0.0025	0.0025
4	0.010, 7.6	0.010, 7.9	0.010, 8.0
		0.010	0.015
7	0.020, 8.0	0.020, 8.5	0.040, 8.6
		0.030	0.050
11	0.020, 8.1	0.060, 8.6	0.090, 8.7
		0.055, 8.5	0.090, 8.8
14	0.020, 8.0	0.060, 8.5	0.090, 8.7
		0.055, 8.5	0.080, 8.7

	50	100
0 days	0.0025	0.0025
4	0.020, 8.2	0.020, 8.4
	0.015	0.040
7	0.070, 8.6	0.080, 8.8
	0.060	0.095
11	0.070, 8.7	0.11, 8.9
	0.080, 8.7	0.10
14	--	0.11, 8.9
	0.080, 8.7	0.11

Table 34. Growth of <u>Chlorella</u> in Allen's medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flask

Growth time	ppm $NaHCO_3$ -C 0 (control)	25	50	100	Cotton stoppered control
	Optical density				
0 days	0.001	0.001	0.001	0.001	0.001
2	0.010	0.010	0.020	0.020	0.010
4	0.010	0.045	0.050	0.050	0.025
7	0.010	0.090	0.090	0.12	0.040
11	0.010	0.090	0.095	0.13	0.060
15	0.010	--	--	0.14	0.070
20	0.020	--	--	--	0.10

Same, but one-liter flasks full

Growth time	0 (control)	25	50	100
0 days	0.001	0.001	0.001	0.001
2	0.010	0.010	0.015	0.020
4	0.010	0.045	0.045	0.060
7	0.015	0.060	0.060	0.070
11	0.020	0.080	--	0.10
15	0.015	0.085	0.070	0.10
20	0.015	--	0.070	0.10

Table 35. Growth of Chlorella in Allen's medium with 20 ppm $NaHCO_3$-C as sole carbon source.

Growth time	One liter algal suspension in four liter flask	Two liters algal suspension in four liter flask	Four liter flask full	Control-two liters algal suspension in four liter flask
	Optical density			
0 days	0.001	0.001	0.001	0.001
3	0.030	0.020	0.020	0.005
6	0.080	0.075	0.050	0.010
8	0.080	0.090	0.060	0.015
13	0.090	0.090	0.090	0.020
15	--	--	0.090	0.020

Table 36. Growth of Chlorella in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flask.

	ppm $NaHCO_3$-C		
	0 (control)	5	10
Growth time	Optical density, pH		
0 days	0.001	0.001	0.001
7	0.003, 7.0	0.008, 8.2	0.005, 8.7
8	0.005, 7.2	0.010, 9.3	0.013, 9.5
	0.005, 7.3	0.010, 9.0	0.012, 9.2
9	0.010, 7.2	0.015, 9.5	0.015, 9.6
	0.005, 7.4	0.010, 9.3	0.015, 9.4
10	0.010	0.015, 9.8	0.015, 9.8
	0.005, 7.4	0.010	0.020
11	0.010, 7.6	0.015, 10.0	0.020, 10.0
	0.010, 7.9	0.010	0.015, 9.8
15	0.010, 8.3	0.018, 10.2	0.025, 10.3
	0.008, 9.0	0.020	0.025, 10.3
18	0.010, 8.7	0.020, 10.2	0.025, 10.3
	0.010, 8.8	0.020, 10.2	0.025, 10.3
21	0.012, 8.8	0.020, 10.2	0.025, 10.3
	0.012, 9.1	0.020, 10.2	0.030, 10.3
23	0.010, 8.9	0.020, 10.1	0.030, 10.2
	0.010, 9.3	0.020, 10.0	0.025, 10.1
28	0.010, 8.7	0.020, 10.0	--
	0.010, 9.2	0.020, 10.0	--

Table 36 (continued).

	ppm $NaHCO_3$-C		
	25	50	100
Growth time	Optical density, pH		
0 days	0.001	0.001	0.001
7	0.010, 8.9	0.012, 8.8	0.015, 8.7
	0.005, 8.8	0.010, 8.8	0.015, 8.6
8	0.015, 9.2	0.020, 9.1	0.030, 9.0
	0.015, 9.1	0.020, 9.0	0.025, 8.9
9	0.018, 9.3	0.025, 9.2	0.040, 9.1
	0.015, 9.2	0.020, 9.1	0.030, 9.0
10	0.015, 9.3	0.025, 9.3	0.045, 9.3
	0.020	0.030	0.040
11	0.020, 9.5	0.030, 9.5	0.055, 9.5
	0.020, 9.5	0.025	0.040, 9.3
15	0.030, 9.8	0.047, 9.8	0.085, 9.9
	0.030, 10.1	0.050, 10.0	0.080, 9.8
18	0.038, 10.2	0.065, 10.2	0.11, 10.2
	0.040, 10.3	0.070, 10.3	0.11, 10.2
21	0.042, 10.5	0.075, 10.6	0.13, 10.5
	0.045, 10.4	0.080, 10.6	0.13, 10.5
23	0.040, 10.3	0.090, 10.4	0.14, 10.4
	0.050, 10.3	0.075, 10.4	0.14, 10.4
28	0.040, 10.3	-- ---	0.14, 10.7
	0.045, 10.3	0.075, 10.5	0.13, 10.7

Table 37. Growth of <u>Chlorella</u> in ASM medium with $NaHCO_3$ as sole carbon source, 300 cc algal suspension in 500 cc flasks.

	ppm $NaHCO_3$-C		
	0 (control)	5	10
Growth time	Optical density; pH		
0 days	0.002	0.002	0.002
6	0.010, 7.4	0.020, 9.4	0.020, 9.45
	0.010	0.020	0.020
10	0.010, 8.0	0.025, 10.2	0.040, 10.2
	0.012	0.030	0.045
13	0.020, 8.3	0.030, 10.2	0.040, 10.3
	0.015	0.025	0.045
17	0.015, 8.8	0.030, 10.2	0.045, 10.4
	0.015, 7.9	0.030	0.040, 10.3
20	0.015, 9.4	--	0.045, 10.3
	0.015, 8.6	--	--
	25	50	100
0	0.002	0.002	0.002
6	0.030, 9.6	0.025, 9.4	0.055, 9.6
	0.025	0.030	0.045
10	0.055, 10.3	0.050, 9.8	0.12, 10.0
	0.050	0.070	0.080
13	0.055, 10.4	0,070, 10.1	0.14, 10.3
	0.050	0.085	0.14
17	0.055, 10.5	0.090, 10.8	0.17, 10.8
	0.050	0.085, 10.5	0.14, 10.6
20	---	0.090, 10.8	0.16, 10.7
		0.085, 10.5	0.16, 10.7

Table 38. Growth of *Microcystis* in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flasks

	ppm $NaHCO_3$-C			
	0 (control	10	50	100
Growth time	Optical density			
0 days	0.0007	0.0007	0.0007	0.0007
5	<0.001	0.025	0.050	0.050
6	<0.001	0.040	0.065	0.070
9	<0.001	0.030	--	0.16
12	<0.001	--	0.12	0.23
19	0.010	0.040	0.12	0.24

Table 39. Growth of *Microcystis* in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flasks

	ppm $NaHCO_3$-C			
	0 (control	25	50	100
Growth time	Optical density			
0 days	0.001	0.001	0.001	0.001
7	0.005	0.025	0.030	0.030
12	0.010	0.045	0.095	0.125
19	0.010	0.040	0.080*	0.140*
Same, except flasks filled with algal suspension				
0	0.001	0.001	0.001	0.001
7	--	0.005	0.010	0.015
12	0.010	0.045	0.075	0.080
19	0.010	0.040	0.085*	0.11*

*Algal suspension becoming cloudy and discolored.

Table 40. Growth of *Microcystis* in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one liter flasks

Growth time	ppm $NaHCO_3$-C 0 (control)	5	10
	Optical density, pH		
0 days	0.002	0.002	0.002
9	0.005, 7.0	0.008, 7.4	0.010, 7.9
		0.010	0.010
12	0.010, 7.5	0.010, 7.8	0.020, 9.1
		0.010	0.010
16	0.015, 9.0	0.020, 9.4	0.045, 10.4
		0.020	0.030
19	0.020, 10.5	0.030, 10.6	0.050, 10.8
		0.040, 10.7	0.045, 10.7
21	0.020, 10.2	--*	--*
	25	50	100
0 days	0.002	0.002	0.002
9	0.012, 8.2	0.010, 8.2	0.010, 8.2
	0.015	0.015	0.010
12	0.030, 9.1	0.020, 8.8	0.015, 8.5
	0.025	0.030	0.020
16	0.080, 10.3	0.075, 9.7	0.070, 9.3
	0.095	0.12	0.070
19	0.070, 10.8*	0.16, 11.0	0.20, 11.0
	--*	0.17	0.24, 10.4
21		--*	--*

*Algal suspension becoming cloudy and discolored.

Table 41. Growth of Microcystis in ASM medium with $NaHCO_3$ as sole carbon source. One-liter flasks completely filled with algal suspension

	ppm $NaHCO_3$-C			
	0 (control	10	50	100
Growth time	Optical density			
0 days	0.008	0.008	0.008	0.008
4	0.010	0.045	0.060	0.050
7	0.020	0.040	0.10	0.95
10	0.020	--	0.13	0.14
14	0.020	0.040	0.13	0.21

Table 42. Growth of Anabaena in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flasks

	ppm $NaHCO_3$-C			
	0 (control)	10	50	100
Growth time	Optical density			
0 days	0.0005	0.0005	0.0005	0.0005
7	0.005	0.010	0.025	0.015
10	0.010	0.035	0.070	0.070
14	0.010	0.035	0.10	0.12
17	0.010	0.030	0.090	0.13

Table 43. Growth of *Anabaena* in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flasks

	ppm $NaHCO_3$-C				
	0 (control)	10	25	50	100
Growth time	Optical density				
0 days	0.005		0.005	0.005	0.005
6	0.015		0.020	0.015	0.020
8	0.015		0.040	0.020	0.040
16	0.015		0.040	0.030	0.060
20	0.020		*	0.075	0.085
23	0.020		*	0.11	0.14
26	0.020		*	0.12	0.18
Same except one-liter flasks filled with algal suspension					
0	0.005	0.005	0.005	0.005	0.005
6	0.015	0.015	0.015	0.015	0.015
8	0.015	0.020	0.020	0.020	0.020
16	0.015	--	0.020	0.025	0.040
20	0.015	0.020	0.050	0.080	0.090
23	0.020	0.030	0.055	0.10	0.15
26	0.020	0.040	0.055	0.12	0.17

Note: Discoloration developed after 26 days in all flasks

*Discoloration

Table 44. Growth of *Anabaena* in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flasks.

	ppm $NaHCO_3$-C				
	0 (control)	10	25	50	100
Growth time	Optical density				
0 days	0.003	0.003	0.003	0.003	0.003
3	0.020	0.025	0.025	0.025	0.025
7	0.020	0.040	0.050	0.050	0.045
9	0.020	0.050	0.075	0.11	0.11
15	0.020	*	*	0.11	0.14
17	-	*	*	0.10*	0.13*
Same except flasks filled with algal suspension					
0 days	0.003	0.003	0.003	0.003	0.003
3	0.020	0.030	*	0.015	0.020
7	0.025	0.035	*	0.030	0.030
9	0.025	--	*	0.030	0.045
15	--	0.050	*	0.090	0.090
17	0.020	0.045*	*	0.090*	*

*Discolored and cloudy

Table 45. Growth of *Anabaena* in ASM medium with $NaHCO_3$ as sole carbon source. 600 cc algal suspension in one-liter flasks.

	ppm $NaHCO_3$-C		
	0 (control	5	25
Growth time	Optical density, pH		
0 days	0.003	0.003	0.003
9	0.003, 7.1	0.005, 7.5	0.005, 8.2
12	0.010, 7.6	0.010, 9.1	0.015, 9.0
16	0.020, 10.0	0.020, 10.2	0.040, 9.8
19	0.020, 9.9	0.035, 10.5	0.060, 10.7
21	0.015, 10.0	0.035, 10.4	*
22	0.018, 10.2	0.040, 10.7	*
23	0.018, 10.1	0.040, 10.5	*
26	0.020, 10.1	0.040, 10.5	*

	50	100
0 days	0.003	0.003
9	0.005, 8.1	0.010, 8.4
12	0.010, 8.6	0.025, 8.8
	0.010	0.010
16	0.035, 9.3	0.060, 9.4
	0.035	0.030
19	0.050, 9.9	0.11, 10.1
	0.070, 10.0	0.050, 9.5
21	0.055, 10.0	0.14, 10.2
	0.080, 10.4	0.070, 9.7
22	0.075, 10.4	0.16, 10.7
	0.10, 10.8	0.10, 10.0
23	*	0.16, 10.8
	0.11, 11.0	0.11, 10.0
26	*	*
	*	*

*Discoloration and cloudiness

Table 46. Growth of <u>Chlorella</u> in non-mixed six foot by one foot cylinder open to the atmosphere. Algal suspension three feet deep.

Growth time	Optical density
0 days	0.001
3	0.005
4	0.010
5	0.020
6	0.020
8	0.025
9	0.040
10	0.040

Table 47. Growth of <u>Chlorella</u> in six foot by one foot cylinders open to the atmosphere. Algal suspension five feet deep.

	Optical density, pH	
Growth time	No mixing	Bottom air mixed
0 days	0.001	0.001
2	0.003, 7.5	0.003, 7.5
3	0.005, 7.4	0.005, 7.3
5	0.009	0.009
6	0.015, 7.35	0.010, 7.3
7	0.018	0.011
8	0.022, 7.6	0.015, 7.3
9	0.030, 7.6	0.020, 7.3
10	0.035, 7.5	0.030, 7.4
13	0.045, 7.6	0.030, 7.4
15	0.060, 7.6	0.060
16	0.060, 7.5	0.060, 7.4
20	0.073, 7.5	0.070, 7.2
21	0.071, 7.5	--
22	0.070, 7.5	0.080, 7.2

Table 48. Growth of Chlorella in six foot by one foot cylinders open to the atmosphere. Algal suspension five feet deep.

Growth time	Optical density, pH: No mixing	Optical density, pH: Bottom air mixed at 1.5 l/minute
0 days	0.001	0.001
4	0.012, 7.2	0.012, 7.2
5	0.015, 7.4	0.015, 7.4
6	0.025, 7.6	0.020, 7.5
7	0.030, 7.6	0.020, 7.4
8	0.035, 7.7	0.030, 7.5
11	0.030, 7.8	0.032, 7.6
12	0.032, 7.8	0.040, 7.5
13	0.040, 7.8	0.045, 7.5
14	0.045, 7.7	0.050, 7.4
15	0.055, 7.7	0.055, 7.4
18	0.055	0.065, 7.4
19	0.058	0.068, 7.4
20	0.060	--
21	0.070	--
22	--	0.095, 7.5
25	--	0.105, 7.4

Table 49. Growth of Chlorella in six foot by one foot cylinders open to the atmosphere. Algal suspension five feet deep. Comparison of frequent stirring and weekly stirring.

Growth time	Optical density, pH: Frequent sampling[(1)]	Optical density, pH: Weekly sampling[(1)]
0 days	0.001	0.001
6	0.020, 7.6	0.018, 7.6
7	0.025, 7.9	--
8	0.025, 7.9	--
9	0.028, 7.9	--
10	0.030, 7.9	--
13	0.030, 7.8	0.035, 7.8
15	0.035, 7.8	--
20	0.050, 7.8	0.055, 7.8

[(1)] Stirred only at time of sampling.

Table 50. Growth of Chlorella in six foot by one foot cylinders open to the atmosphere. Algal suspension five feet deep.

Growth time	Optical density, pH: No mixing	Optical density, pH: Same, but 20 ppm $NaHCO_3$-C present
0 days	0.002	0.002
3	0.005	0.005
4	0.010, 7.7	0.012, 7.8
5	0.020, 7.7	0.027, 8.0
6	0.027, 7.8	0.045, 8.2
7	0.027, 7.9	0.055, 8.5
8	0.032, 7.9	--
9	0.041, 7.9	--
11	0.048, 7.8	--
12	0.053, 7.8	--

Table 51. Growth of Chlorella in six foot by one foot cylinders open to the atmosphere. Algal suspension five and one-half feet deep.

Growth time	Optical density, pH: No mixing	Optical density, pH: Top air ventilation, 2 l/minute	Optical density, pH: No mixing, 20 ppm $NaHCO_3$-C added at 5 days
0 days	0.003	0.003	0.003
5	0.012, 7.5	0.010, 7.5	0.009, 7.7
6	0.018, 7.7	0.020, 7.8	0.015, 8.1
7	0.022, 8.0	0.040, 8.2	0.029, 8.5
8	0.028, 8.2	0.053, 8.2	0.055, 8.7

Table 52. Growth of *Chlorella* in six foot by one foot cylinders open to the atmosphere. Algal suspension five and one-half feet deep.

Growth time	Optical density, pH: No mixing	Optical density, pH: Top air ventilation, 2 l/minute
0 days	0.002	0.002
3	0.008, 7.4	0.008, 7.3
4	0.012, 7.6	0.018, 7.5
5	0.015, 7.8	0.023, 7.8
6	0.018, 7.8	0.031, 7.8
7	0.020, 7.9	0.040, 7.9
8	0.022, 7.8	0.040, 7.9
10	0.025, 7.8	0.043, 7.8

Table 53. Growth of *Chlorella* in six foot by one foot cylinders open to the atmosphere. Algal suspension five and one-half feet deep.

Growth time	Optical density, pH: No mixing	Optical density, pH: Same but 20 ppm $NaHCO_3$ added at 5 days
0 days	0.002	0.002
2	0.010	0.010
5	--	0.021, 7.8
6	--	0.043, 8.4
7	0.040, 7.8	0.065, 8.6
8	0.043, 7.8	0.075, 8.6

Table 54. Comparison of growth of *Chlorella* for bottom (344 and 15 ppm CO_2) and top aeration in full 16 liter jugs open to atmosphere.

Growth time	Bottom-344 ppm	Bottom-15 ppm	Top
	Optical density		
0 days	0.001	0.001	0.001
1	0.005	0.005	0.005
6	0.010	0.010	0.025
7	0.015	0.015	0.025
8	0.020	0.015	0.030
9	0.030	0.015	0.040
10	0.030	0.015	0.040
13	0.045	0.025	0.055
14	0.060	0.030	0.060
21	0.11	0.060	0.070
23	0.12	0.080	0.080

Table 55. Comparison of growth for bottom and top aeration in full 16 liter jugs open to atmosphere.

Growth time	Bottom	Top
	Optical density	
	Chlorella	
0 days	0.001	0.001
2	0.005	0.005
3	0.005	0.010
4	0.005	0.010
7	0.015	0.030
8	0.025	0.040
10	0.040	0.050
18	0.13	0.070
21	0.17	0.085
25	0.17	0.10
	Microcystis	
0 days	0.001	0.001
4	0.005	0.005
21	0.010	0.005
28	0.020	0.015
38	0.100	0.080

Table 55 (continued).

Growth time	Bottom	Top
	Optical density	
	Anabaena	
0 days	0.001	0.001
4	0.010	0.010
13	0.020	0.015
24	0.015	0.035
28	0.020	0.060
38	0.070	0.100

Table 56. Comparison of Chlorella growth for bottom (bubble) aeration, top aeration, and cotton plug aeration for different volumes of algal suspensions.

Growth time	Bottom	Top	Cotton plug
	Optical density		
	250 cc in one-liter flask		
0 days	0.007	0.007	0.007
4	0.25	0.10	0.12
5	0.28	0.14	0.14
6	0.32	0.17	0.17
7	0.35	0.19	0.20
8	0.36	0.25	0.23
	600 cc in one-liter flask		
0	0.007	0.007	0.007
4	0.23	0.080	0.080
5	0.28	0.095	0.090
6	0.33	0.10	0.10
7	0.35	0.13	0.12
8	--	0.13	0.12
	One liter flask full		
0	0.007	0.007	0.007
4	0.22	0.060	0.070
5	0.27	0.065	--
6	0.32	0.070	--
7	0.35	0.090	0.080
8	0.39	--	0.080

Table 57. Comparison of *Microcystis* growth for bottom (bubble) aeration, top aeration, and cotton plug aeration for different volumes of algal suspensions.

Growth time	Bottom	Top	Cotton plug
	250 cc in one-liter flask		
	Optical density		
0 days	0.007	0.007	0.007
6	0.10	0.040	0.070
7	0.18	0.070	0.095
9	0.35	0.14	0.17
10	0.48	0.23	0.24
13	0.55	0.42	0.34
14	0.55	0.48	0.38
	600 cc in one-liter flask		
0	0.007	0.007	0.007
6	0.070	0.050	0.050
7	0.11	0.070	0.070
9	0.24	0.14	0.11
10	0.35	0.19	0.16
13	0.53	0.29	0.21
14	0.58	0.34	0.25
	One-liter flask full		
0	0.007	0.007	0.007
6	0.080	0.030	0.020
7	0.12	0.030	0.030
9	0.25	0.040	0.030
10	0.32	0.060	0.050
13	0.46	0.070	0.050
14	0.54	--	0.060
15	0.58	0.085	0.060

Table 58. Comparison of <u>Anabaena</u> growth for bottom (bubble) aeration, top aeration, and cotton plug aeration for different volumes of algal suspensions.

Growth time	Bottom	Top	Cotton plug
	250 cc in one liter flask		
	Optical density		
0 days	0.0070	0.0070	0.0070
8	0.10	0.060	No good
9	0.13	0.085	"
13	0.13	0.10	"
15	0.18	0.11	"
19	0.29	0.12	"
20	0.32	0.13	"
	600 cc in one-liter flask		
0	0.0070	0.0070	0.0070
8	0.080	0.050	0.025
9	0.11	0.055	0.045
13	0.20	0.080	0.055
15	0.33	0.11	0.075
19	0.45	0.15	0.12
20	--	--	0.13
	One-liter flask full		
0	0.0070	0.0070	0.0070
8	0.060	0.030	0.020
9	0.080	0.035	0.025
13	0.11	0.040	0.030
15	0.19	0.050	0.040
19	0.27	0.060	0.040
20	0.30	0.065	0.040

Table 59. Comparison of growth for bottom (bubble) aeration, top aeration, and cotton plug aeration. All 325 cc algal suspension in 500 cc erlenmeyer flasks.

Growth time	Bottom	Top	Cotton plug
	Optical density		
		Chlorella	
0 days	0.0040	0.0040	0.0040
7	0.11	0.065	0.060
10	0.16	0.088	0.088
13	0.26	0.12	0.11
14	0.28	0.12	0.12
16	0.32	0.14	0.14
21	0.54	0.18	0.18
		Microcystis	
0	0.0040	0.0040	0.0040
7	0.075	0.065	0.075
10	0.18	0.14	0.15
13	0.33	0.24	0.26
14	0.38	0.25	0.28
16	0.47	0.30	0.33
21	0.78	0.41	0.48
		Anabaena	
0	0.0040	0.0040	0.0040
7	0.040	0.020	0.020
10	0.085	0.052	0.038
13	0.25	0.10	0.070
14	0.28	0.11	0.070
16	0.38	0.15	0.090
21	0.53	0.26	0.16

Table 60. Comparison of growth for bottom (bubble) aeration and cotton plug aeration. 600 cc algal suspension in one liter flasks.

	Chlorella - Allen's medium	
	Bottom	Cotton plug
Growth time	Optical density	
6 days	0.060	0.030, 0.040
15	0.32	0.070, 0.060
21	0.33	0.070, 0.10
29	0.35	0.10, 0.12
37	0.39	0.10, 0.13
	Chlorella - ASM medium	
6	0.095	0.015
15	0.33	0.040
21	0.43	0.070
29	0.49	0.13
37	0.54	0.18

	Microcystis		*Anabaena*	
	Bottom	Cotton plug	Bottom	Cotton plug
15	0.030	0.010	0.060	--
21	0.070	0.025	0.13	--
29	0.21	0.060	0.16	--
37	0.33	0.15	0.24	--

Table 61. Comparison of growth for bottom (bubble) aeration, top aeration, and cotton plug aeration. All 800 cc algal suspension in one liter erlenmeyer flasks

Growth time	Bottom	Top	Cotton plug
		Optical density	
		Chlorella	
0 days	0.012	0.012	0.012
5	0.045	0.030	0.030
7	0.12	0.060	0.060
8	0.16	0.060	--
12	0.24	0.060	0.060
15	0.33	0.090	0.075
16	0.33	0.090	0.075
19	0.36	0.12	0.075
20	0.42	0.12	0.090
21	0.42	0.12	0.090
		Microcystis	
0	0.12	0.012	0.012
5	0.027	0.027	0.015
7	0.030	0.030	0.015
8	0.045	0.045	0.015
12	0.17	0.11	0.015
14	0.36	0.21	0.054
15	0.47	0.24	0.060
16	0.60	0.29	0.075
19	--	0.39	0.11
20	--	0.39	0.11
		Anabaena	
0	0.012	0.012	0.012
5	0.045	0.030	0.045
7	0.075	0.045	--
8	0.11	0.045	0.036
12	0.23	0.075	0.045
14	0.52	0.12	0.084
15	0.60	0.14	0.075
16	--	0.15	0.10
19	--	0.18	0.10
20	--	--	0.10

Table 62. Growth of Chlorella under intermittent aeration (344 ppm CO_2) in various media. 600 cc algal suspension in one-liter flasks.

Growth time	Allen's 2 ppm $N-NO_3$	Allen's 15 ppm $N-NO_3$	Allen's 178 ppm $N-NO_3$	ASM 14 ppm $N-NO_3$
	Optical density			
20 minutes aeration per day				
1 week	0.045	0.050	0.050	0.030
2	0.080	0.090	0.080	0.053
3	0.10	0.11	0.085	0.070
5	0.15	0.15	0.13	0.10
6	0.18	0.18	0.15	0.12
8	0.22	0.20	0.19	0.12
20 minutes aeration every 3 days				
1 week	0.040	0.020	0.035	0.015
2	0.070	0.065	0.055	0.040
3	0.070	0.070	0.050	0.035
5	0.090	0.090	0.070	0.042
6	0.090	0.10	0.085	0.055
8	0.11	0.13	0.090	0.060
20 minutes aeration every 7 days				
1 week	0.040	0.035	0.030	0.020
2	0.045	0.055	0.045	0.020
3	0.050	0.060	0.050	0.020
5	0.070	0.070	0.060	0.020
6	0.070	0.075	0.070	0.025
8	0.080	0.085	0.080	0.040
20 minutes aeration every 14 days				
1 week	0.040	0.030	0.030	0.020
2	0.050	--	0.040	0.020
3	0.060	--	0.040	0.020
5	0.070	--	0.050	0.020
6	0.070	--	0.040	0.020
8	0.070	--	0.050	0.020

Table 63. Growth of *Chlorella* under intermittent aeration (344 ppm CO_2). One-liter flasks filled with algal suspension.

Growth time	20 minutes aeration every day	20 min/ 4 days	20 min/ 10 days	20 min/ 20 days	20 min/ 40 days
			Optical density		
10 days	0.030	0.025	0.015	0.020	0.020
4 weeks	0.070	0.040	0.030	0.035	0.020
7 weeks	0.130	0.060	0.050	0.030	0.030

Table 64. Growth of *Microcystis* under intermittent aeration (344 ppm CO_2). 600 cc algal suspension in one-liter flasks.

Growth time	20 minutes aeration every 2 days	20 min/ 4 days	20 min/ 7 days	20 min/ 14 days	20 min/ 21 days
			Optical density		
2 weeks	0.020	0.020	0.010	0.010	0.010
3	0.050	0.040	0.025	0.025	0.020
4	0.052	0.042	0.025	0.025	--
6	--	0.060	0.030	0.025	0.020

Table 65. Growth of *Anabaena* under intermittent aeration (344 ppm CO_2). 600 cc algal suspension in one-liter flasks.

Growth time	20 minutes aeration every day	20 min/ 3 days	20 min/ 7 days	20 min/ 14 days	20 min/ 21 days
			Optical density		
2 weeks	0.035	0.025	0.020	0.010	0.010
3	0.035	0.020	0.020	0.012	0.010
5	0.080	0.035	0.020	0.020	0.010

Table 66. Growth of *Chlorella* under intermittent aeration (344 ppm CO_2). 600 cc algal suspension in one-liter flasks.

Growth time	8 hr aeration/day	4 hr/day	1 hr/day
	Optical density		
	Allen's medium		
6 days	0.060, 0.095	0.070, 0:090	0.050, 0.060
15	0.20, 0.23	0.17, 0.20	0.12, 0.12
21	0.27, 0.33	0.23, 0.27	0.15, 0.19
29	0.35, 0.44	0.29, 0.30	0.21, 0.22
37	0.40, 0.50	0.31, 0.36	0.20, 0.24
	ASM medium		
6	0.035	0.035	0.025
15	0.11	0.080	0.040
21	0.19	0.14	0.085
29	0.25	0.20	0.12
37	0.31	0.27	0.15

Table 67. Growth of *Microcystis* and *Anabaena* under intermittent aeration (344 ppm CO_2). 600 cc algal suspension in one-liter flasks.

Growth time	8 hr aeration/day	4 hr/day	1 hr/day
	Optical density		
	Microcystis		
15 days	0.020	0.030	0.020
21	0.045	0.075	0.060
29	0.19	0.25	0.19
37	0.43	0.50	0.37
	Anabaena		
15	0.037	0.050	0.030
21	0.025	0.070	0.065
29	0.25	0.18	0.13
37	0.34	0.27	0.20

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Growth rates of the algae Chlorella, Microcystis, and Anabaena were studied with respect to carbon availability. Algae can utilize dissolved concentrations of carbon dioxide much lower than those from atmospheric equilibria. Control of algal growth by sweeping the carbon dioxide out of water by aeration with air containing very low concentrations of carbon dioxide is difficult because of atmospheric replenishment of carbon dioxide. Bicarbonate is at least 50% utilized at growth rates as high as 7 mg per liter per day (dry weight). Atmospheric replenishment of carbon dioxide, without any wind mixing, can sustain growth rates of 1.5-2 mg per liter per day for depths of at least 1.7 m. (Morton - WARF Institute)

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