

• • • • • LARGE ALGAL SYSTEMS

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INTRODUCTION

The Boeing Company began studying the photosynthetic properties of algae in 1959. After a year of experimentation a decision was made to go directly to a man-supporting system by-passing intermediate steps with rodents, primates, or other animals.

Since the objective of this decision was to quickly achieve a workable system for the study of the interactions between algae and man, no attempt was made to develop a zero gravity capability in the design. Instead, the approach was one of "brute force" to supply the necessary oxygen requirement through quantity and the utilization of readily available equipment components. The system was designed with plexiglass algae tanks 6 ft. high, 3 ft. wide and 1½ inches thick. Banks of fluorescent lamps were alternated with algae tanks to give a "sandwich" which would utilize a high percentage of the available light. The thermophilic algae, Chlorella pyrenoidosa, TX 71105, was selected on the basis of the earlier experimental program. The general configuration of the algae unit can be seen in Figure 1.

Based on extrapolation of data from small experimental systems, the first large algal unit consisted of four tanks each containing 55 liters of algae culture. Tests of the system soon showed that the extrapolation had been overly optimistic and the system was increased first to six tanks and then to eight. In retrospect it appears that the difficulty in extrapolation resulted in part from emphasis on oxygen production per unit volume rather than oxygen production as a function of area and light intensity.

To test the capability of the algae unit, a small chamber was prepared for both manned and unmanned runs. The chamber was cylindrical, approximately five feet in diameter and seven feet long with a net volume of 112 cubic feet. There was sufficient room for a man to sit, lay down and move about but not sufficient to stand erect. Provisions were made for temperature and humidity control, food preparation, waste collection and communication.



FIGURE I. SEEDING ALGAE TANKS FOR CLOSED SYSTEM EXPERIMENT.

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Outside the chamber a control panel monitored the flow of air between the chamber and the algae unit. A heat exchanger cooled and removed excess water and the air as it left the algae tanks and an activated charcoal filter removed odors as the air returned to the chamber. A Roots-Connerville blower provided the positive pressure required to circulate the air and force it through the algae tanks against a pressure head of 60 inches of water. A leakage problem was encountered around the blower seals and it became necessary to mount the blower together with a gearbox used for flow rate control inside a small sealed cylinder.

The oxygen level within the chamber was measured externally with the Beckman E-2 paramagnetic oxygen analyzer. Air was circulated with a small diaphragm type "Dyna-pump" from a central location in the chamber to the oxygen and carbon dioxide analyzer and thence to the chamber exit pipe. Carbon dioxide was measured with a gas chromatograph. A special dual column arrangement in the chromatograph made it possible to also measure oxygen and nitrogen and to monitor for carbon monoxide and methane. A Kitigawa Toxic Gas Detector gave additional capability for detecting small concentrations of carbon monoxide, hydrogen sulfide, sulfur dioxide or ammonia.

Physiological instrumentation was provided to monitor the condition of the subject. The available parameters were EKG, EEG, phonocardiograph, respiration, body temperature and GSR. Continuous recording of the biomedical data was made on a Sanborn 6-channel recorder. The Sanborn recorder and other monitoring and control instrumentation are shown in Figure 2.

For the unmanned runs the equivalent respiratory function was simulated in two steps. Oxygen was consumed by a small hydrogen flame to give water vapor, and carbon dioxide was metered into the chamber from gas cylinders. The quantities of hydrogen and carbon dioxide were adjustable to simulate any fractional requirement desires.

Unmanned Runs

The purposes of the simulated man tests were several fold. It was desired to determine the oxygen production capacity of the system, the build-up of toxic gases from the algae over a period of several days, the growth rate and optimum density of the algae culture, the depletion of nutrient components, the susceptibility of the algae to contamination, and the mechanical integrity of the entire system.

Two multi-day unmanned runs were made, one for two and one-half days and the other three and one-half days. In each case the maximum oxygen production rate was achieved 24 hours after seeding. The average culture density at this time was 1.3 grams/liter dry weight. The peak oxygen production for these runs averaged 43 ml. oxygen/liter/hour. For each tank of 55 liter capacity, the oxygen output was approximately 2.4 liters oxygen/tank/hour. Eight tanks should then provide 19 liters/hour or about the requirement of one man in a resting or low activity environment.

No toxic gases were detected in these runs and there were no serious mechanical failures of the pumps, blowers, tanks or analytical gear. Although large numbers of bacteria grew in the algae culture, there was no evidence that they seriously

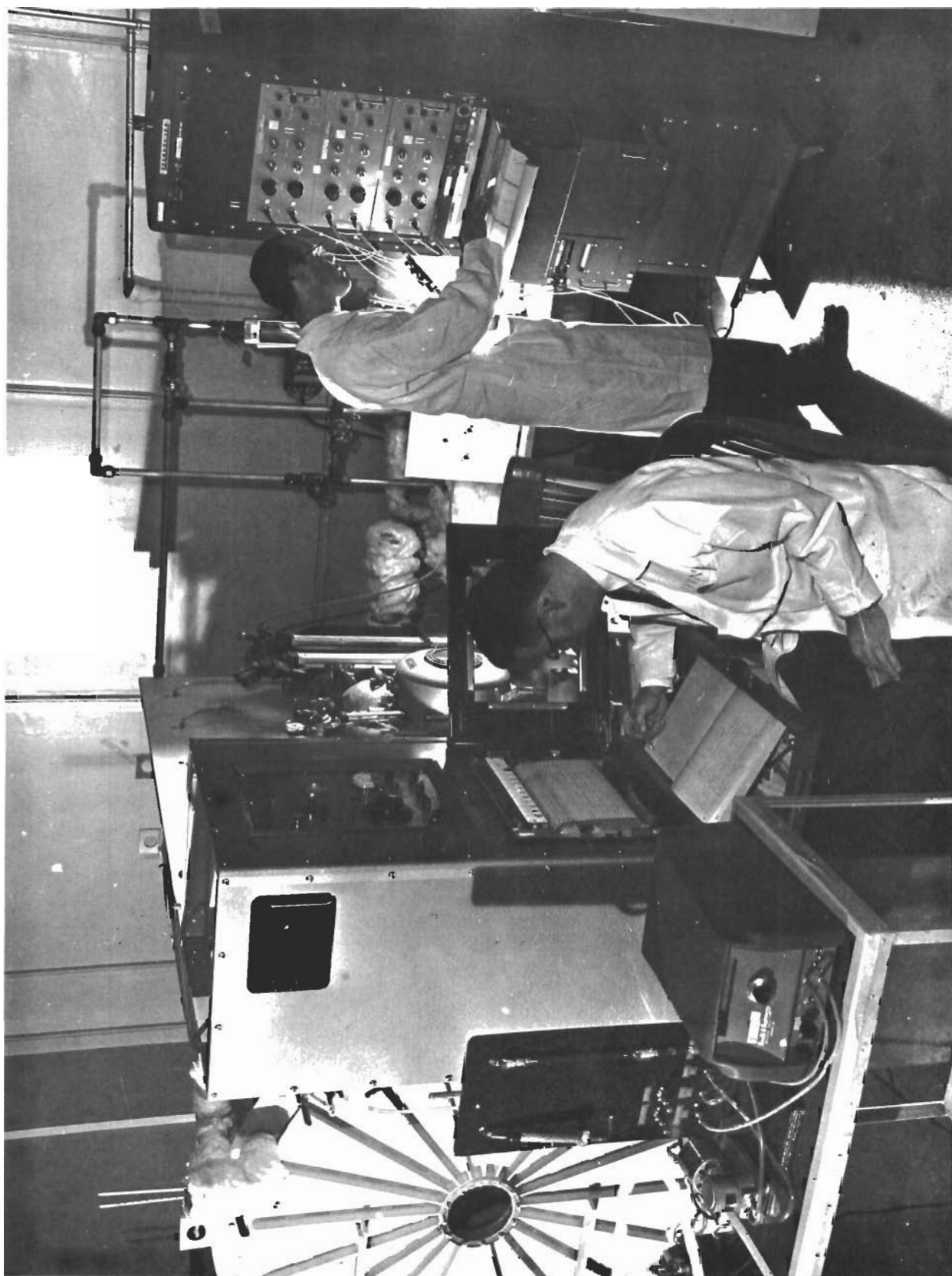


FIG. 2. CHAMBER AND INSTRUMENTATION FOR ALGAE TESTS.

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interfered with the algae growth or oxygen production. The nutrient media supported the algae growth at its maximum rate for 72 hours. The growth rate appeared to be logarithmic only at low densities and was essentially linear over the greatest portion of each run.

56-Hour Run

A number of manned tests were made starting with a 6-hour run on May 10, 1960, and culminating in a 56-hour run April 25-27, 1961. For the 56-hour run, eight algae tanks containing a total of 380 liters of culture were used. The illuminated area was 240 square feet and the intensity of illumination averaged 1,000 foot candles. In a baseline test the subject consumed an average of 19 liters oxygen/hour. To insure adequate gas exchange for the algae at the beginning of the run, carbon dioxide was added to the chamber to bring the level to 1 percent. It was planned to maintain the carbon dioxide level at not less than 1 percent by addition of more gas as required during the run. The maximum permissible concentration was set at 2 percent.

To insure maximum comfort of the subject, physiological instrumentation was reduced to a minimum. Respiration and phonocardiograph sensors were used to give a continuous record of breathing rate and heart action. Pulse rate was also determined from the phonocardiograph trace. These parameters together with visual and voice communications completed the medical surveillance link.

The record of oxygen and carbon dioxide levels in the chamber is shown in Figure 3. It is readily apparent that the requirements of the subject and the capacity of the algae system were very closely matched. During waking hours and particularly with activity or excitement, the carbon dioxide level rose while the oxygen level fell indicating a requirement in excess of the algae system capacity. During sleep the subject's requirements decreased and the algae system recouped its losses. During the second day there was little activity or excitement and the oxygen and carbon dioxide concentrations remained relatively steady.

Twenty-eight hours after the start of the experiment, the algae tanks were isolated from the system one by one, partially drained, and refilled with fresh media. The purpose of this test was to simulate the "harvesting" procedure and to determine whether there would be a serious lag in photosynthetic activity. Although there was some increase in carbon dioxide and decrease in oxygen during the changeover period, the system quickly recovered and the total result appears to be due merely to the down time of the tanks rather than a lag effect. The tanks were "harvested" a second time at the 52-hour mark but the resulting effects were obscured by the increased activity and excitement of the subject as the run approached termination.

Average density and pH measurements for the algae culture are shown in Table 1. The increase in density for the 3/4 inch tanks was approximately double the increase in the 1 1/2 inch tanks. The average pH shows a characteristic rise as the media is depleted. Even after harvesting and replenishing the buffered media, the pH remains relatively high.

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TABLE 1

Density and pH of the Algae Culture During 56 Hour Test

Day	Time	Density g/l		Average pH all Tanks
		1 1/2" Tanks	3/4" Tanks	
4-25-61	0900	0.64	0.82	6.4
	1300	0.83	1.32	6.6
	1700	1.13	1.65	6.8
	2300	1.35	1.90	7.4
4-26-61	0500	1.56	2.70	7.4
	0900	1.71	2.95	7.6
	1300	1.73	3.34	8.0
	*			
	1700	1.22	2.19	7.5
4-27-61	0100	1.33	2.65	7.5
	0900	1.60	3.74	7.6

* Part of algae harvested and replaced with fresh media.

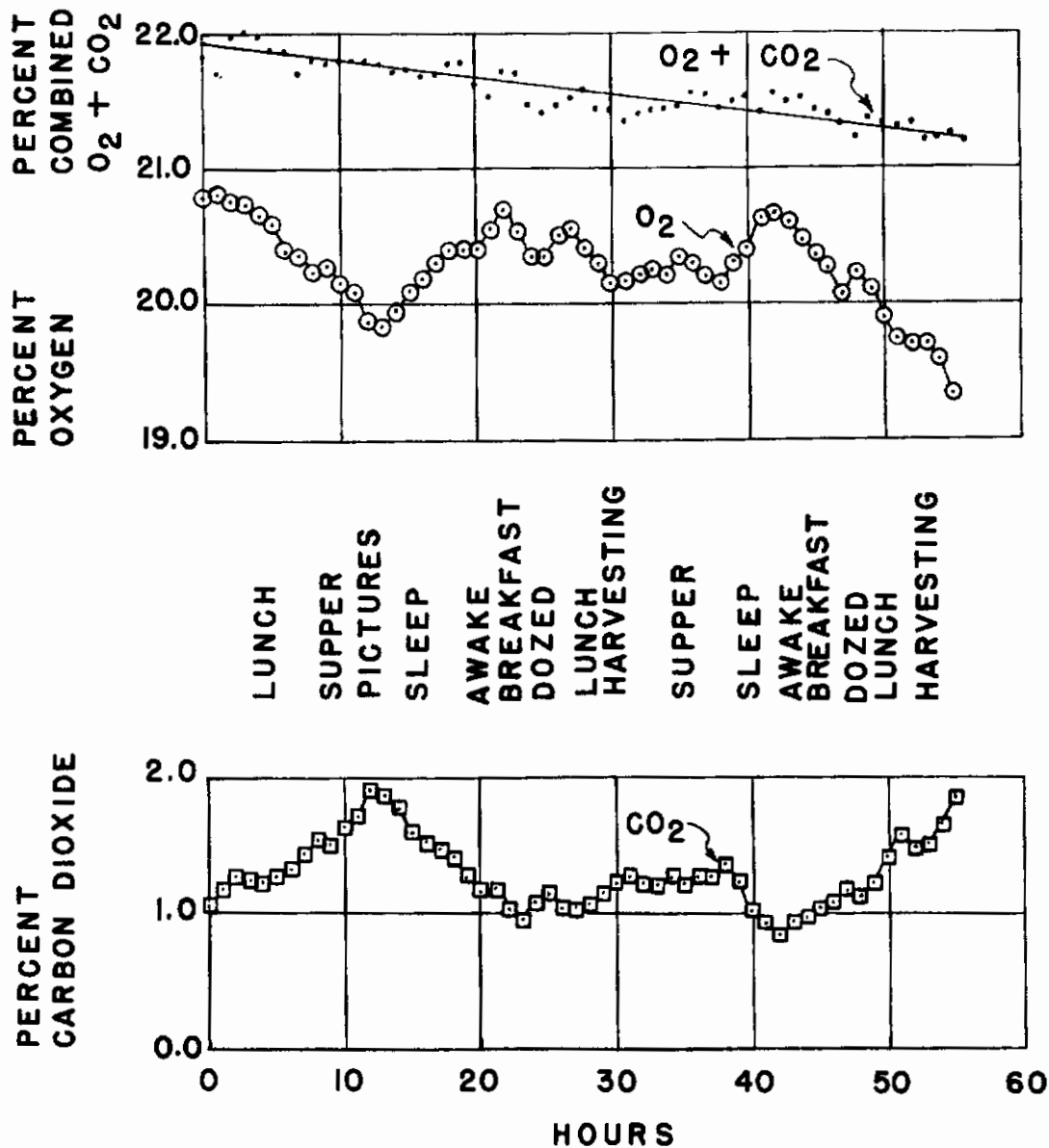


FIGURE 3. OXYGEN AND CARBON DIOXIDE LEVELS DURING 56 HOUR ALGAE TEST.

Discussion

The major objective of our experimental algae studies is to determine the feasibility of developing a closed ecological system for the support of man in future space missions. One of the key factors to this determination is the weight or volume requirement of algae culture per man.

Estimates based on extrapolation from laboratory data have varied widely. This is not too surprising since many different types of data have been used as a basis for these estimates. For example, one can calculate the algae volume from photosynthetic efficiency, light intensity, and culture density. Another method is to extrapolate from algae growth rates, equating grams of cell tissue to liters of oxygen. The most direct procedure is to measure the volume of oxygen produced per unit volume of algae culture.

The Boeing studies have utilized both of the latter procedures, and have found a close relationship between weight of algae produced and volume of oxygen. In order to increase oxygen production per unit volume of algae culture, tank thickness has been decreased stepwise. The initial tanks for the manned system were 1 1/2 inches thick. This was reduced by a factor of two in making 3/4" tanks. Still later, 1/2" tanks were fabricated for a "multi-man" system. Currently, a modified tank with an equivalent 1/4" thickness is being tested.

The relationship of oxygen production to tank thickness is shown in Figure 4. Based on the value of 0.235 liters oxygen/liter of culture/ hour which has been achieved in the 1/4" tanks, one man's requirement of 600 liters of oxygen/day could be met by 106 liters of algae culture. On the basis of the studies to date it is predicted that the direct relationship between oxygen production and tank thickness will continue to hold for thinner tanks until limited by culture viscosity and gas flow difficulties. Preliminary tests with 1/8" tanks indicates no serious problems with gas flow and it is believed that oxygen production on the order of 0.470 liters of oxygen/liter of algae/hour can be obtained from such a system. This would be the equivalent of 53 liters of algae culture per man.

The technique used for determining oxygen production and carbon dioxide absorption is a measurement of rates of change of the respective parameters in a closed recycling system. A small diaphragm pump draws air from the top of the algae tank and forces it in at the bottom. A jug or other sealed container is used as a "chamber" in the system to provide sufficient air volume to give easily measured rates of change. The concentration of oxygen and carbon dioxide is monitored with a Beckman E-2 Oxygen Analyzer and the carbon dioxide is measured with a Wilkins gas chromatograph.

To initiate a run, the unsealed system is charged with carbon dioxide to a predetermined concentration, generally between 5 and 10 percent. The system is then sealed and recycled until sufficient data has been obtained to determine the desired rate values. In Figure 5 the data from a run with the 1/4 " algae is plotted with the carbon dioxide scale inverted for easy comparison of relative rates. In general, both curves rise linearly until the photosynthetic reaction is limited by inadequate carbon dioxide concentration. The rates are determined by striking an average graphically and measuring the percent change per unit time. The oxygen production and carbon dioxide absorption are then computed on the basis of the air and algae volumes.

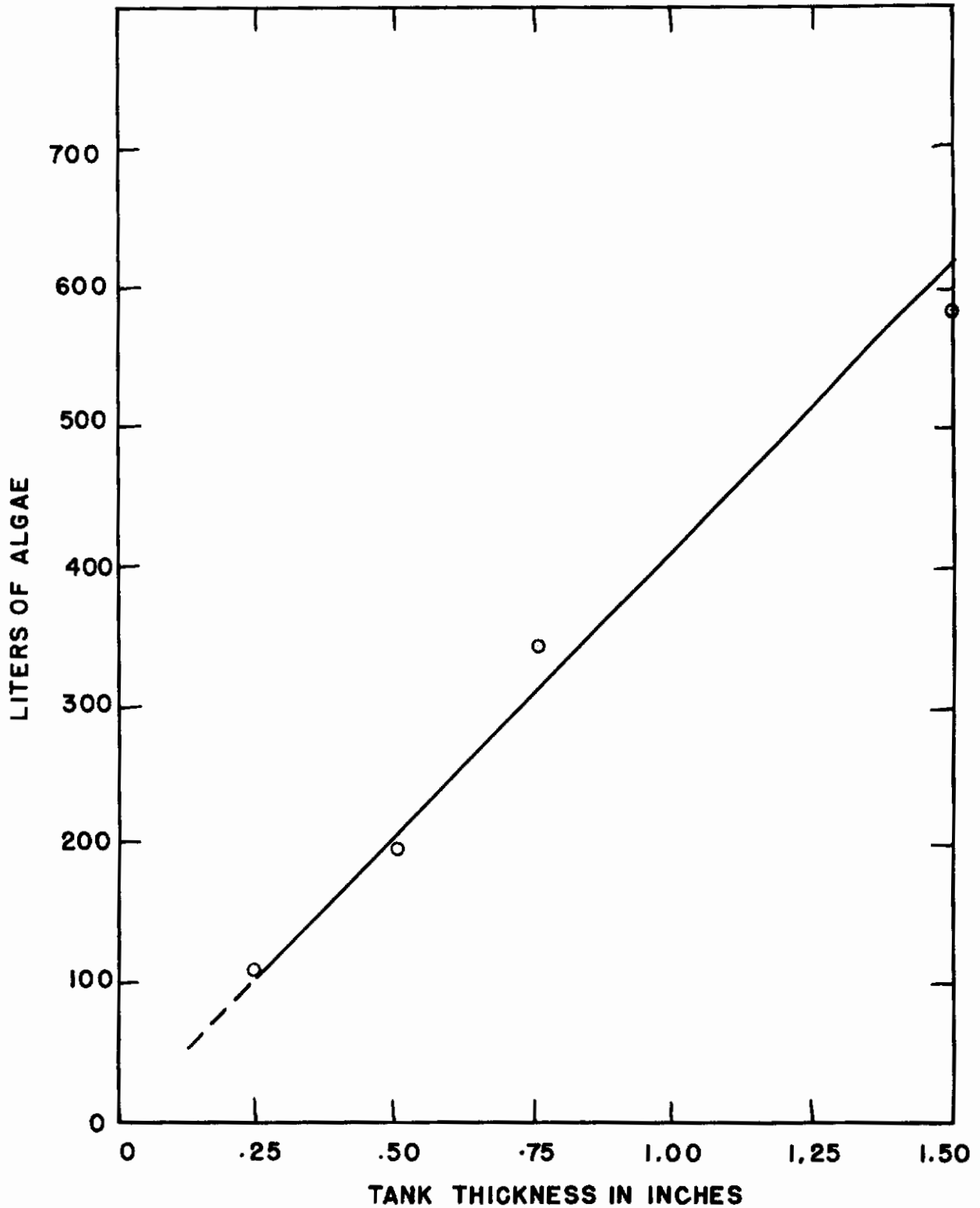


FIG. 4. ALGAE REQUIREMENT PER MAN BASED ON OXYGEN PRODUCTION FROM TANKS OF VARIOUS THICKNESS.

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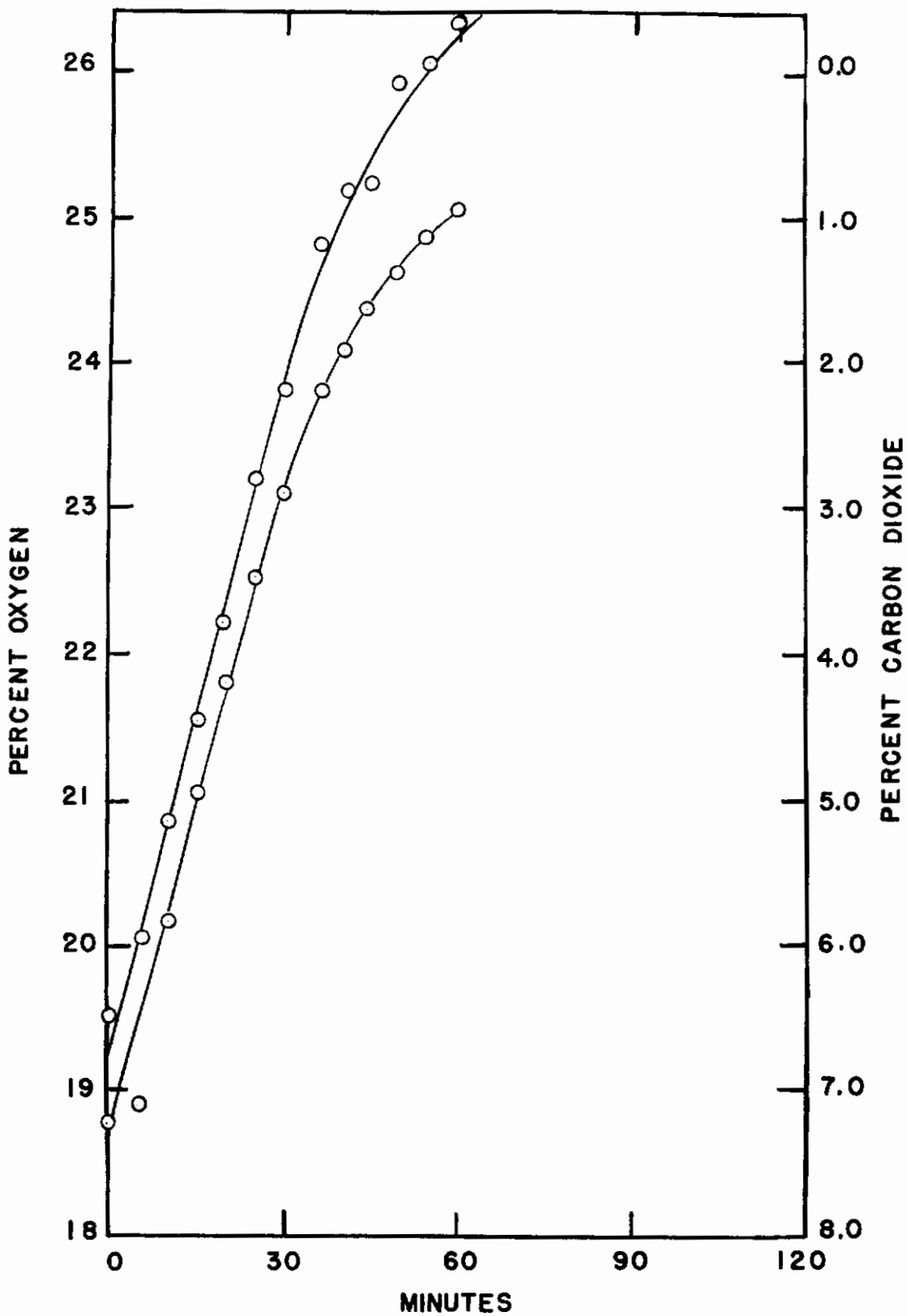


FIGURE 5. PHOTOSYNTHETIC ACTIVITY OF ALGAE CULTURE IN 1/4 INCH TANK.

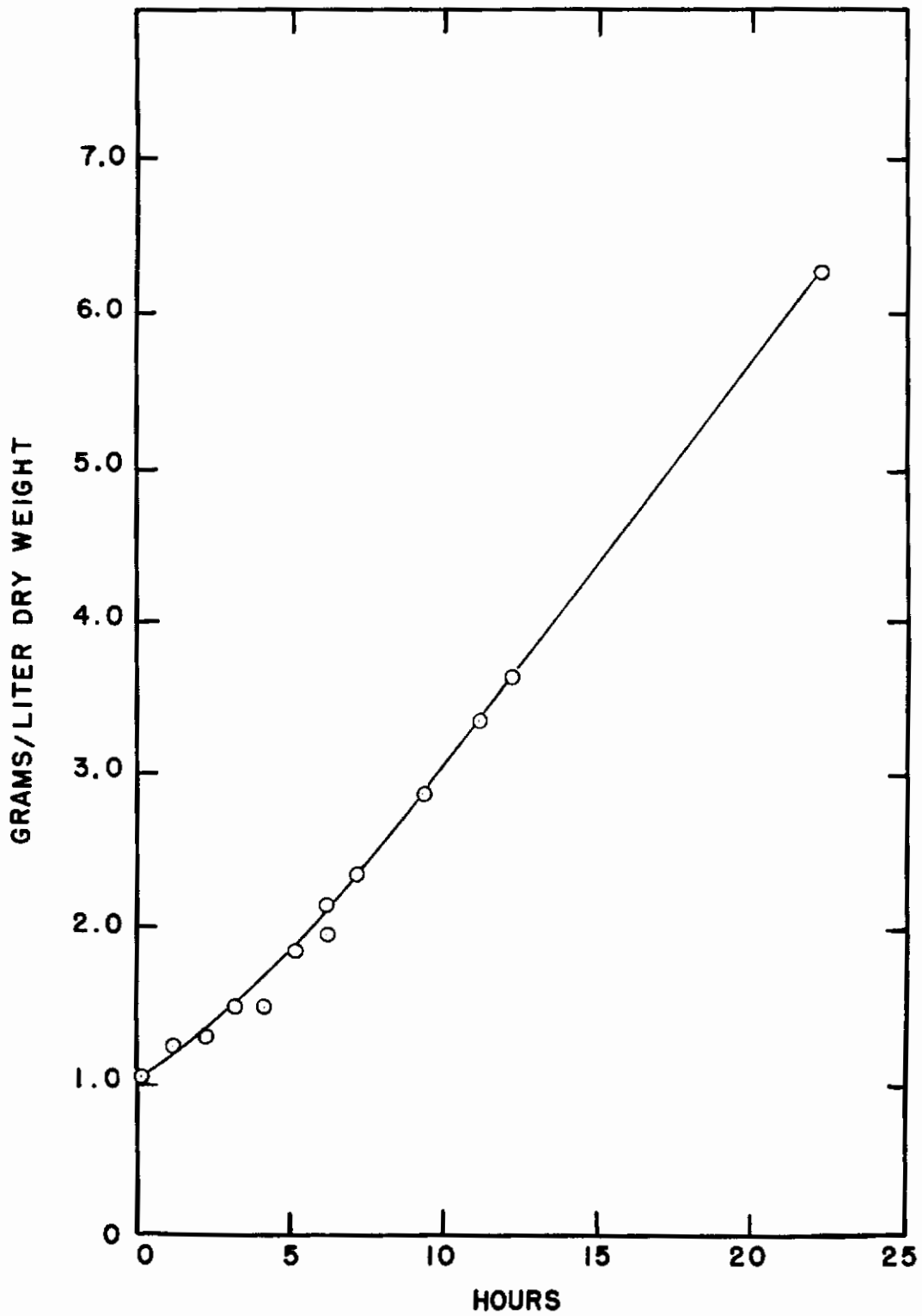


FIGURE 6. ALGAE GROWTH RATE IN 1/4 INCH TANK

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This technique was adopted when direct measurement of concentrations at the inlet and outlet of an algae tank proved too variable for accurate estimation of rates of change. Although it is usually applied to single tanks, this method has also been applied to measuring the capacity of the total system using the 112 cu. ft. chamber for dilution volume. Similarly, one can determine a subject's oxygen requirement and carbon dioxide evolution by sealing him in the chamber and obtaining the required rates from the data slopes.

The growth of cell tissue is determined by plotting the dry weight of the algae in grams per liter as a function of time. As with oxygen production, the rate of change is found by measuring the slope of the line connecting the data. The dry weights are obtained in either of two ways. In one, the algae sample is filtered along with a known weight of celite to prevent clogging. The filter is dried and the algae weight is obtained by difference. In the second method, a known volume of algae sample is centrifuged and the packed cell volume is read. The packed cell volume is converted to a dry weight value by reference to an empirical curve obtained from numerous samples measured by both methods.

Algae growth data for the $\frac{1}{4}$ inch tank is shown in Figure 6. From the changing slope of the curve it appears that the culture is in the lag phase of growth below about 2 grams/liter density, and in a linear phase of growth, above this. With thicker tanks the linear growth phase extends to lower densities.

SUMMARY

The Boeing Company has demonstrated the feasibility of supporting man's respiratory requirements with a photosynthetic algae system and has successfully concluded a 56-hour manned test. Although the system tested required 380 liters of algae culture, tests with thinner tanks have given data indicating that a man can be supported on a much smaller volume.