LABORATORY AND FIELD STUDIES OF PHOTOSYNTHESIS IN THE MARINE CROP PLANT *MACROCYSTIS*[1]

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Abstract:

A closed-system water tunnel was designed to allow measurement of photosynthetic rates in benthic marine plants in moving water. This water tunnel, together with a Clark-type oxygen electrode, was used to measure photosynthesis of entire Macrocystis angustifolia blades in the laboratory and in situ in different water velocities. Photosynthetic rates as high as 11 ml $O_2/dm^2/hr$ were recorded in velocities of 4.5 cm/sec. Rates of mature surface blades showed no photoinhibition and continued to rise significantly as the light intensity was increased up to at least 1.8 Ly per minute(350-750 nm). Photosynthetic rates decreased with decreasing water velocity below 4.5 cm/sec. The current velocities in kelp beds fall in this range. The morphology of the blade was demonstrated to create turbulence as water passed over the blade. Such turbulence could greatly increase diffusion rates of nutrients and gases to the blade. The morphology thus may be important in enhancing the transport of nutrients and gases to the plant.

Introduction:

Studies of photosynthetic rates of Macrocystis have typically been done using B.O.D. bottles or Warburg flasks.[1,2] Such approaches necessitate cutting small discs from the blades. The effect of this cutting on metabolism is not yet known. Water motion has not been controlled. Bottles can be stirred or shaken, but the type and magnitude of the water motion generated in this way does not duplicate natural flow past a blade.

Some workers have avoided confinement in B.O.D. bottles. Towle and Pearse[3] used a large plastic bag slipped over the blades of Macrocystis. Whitford and Kim modified an artificial stream apparatus to measure growth of whole algal thalli. Schramm[5] used a large flow-through cuvette with an oxygen electrode for studies of Fucus. Matsumoto[6] measured the growth rates of Porphyra by using rotary spokes to drag attached plants around a circular tank, thus creating water motion relative to the plant. Some limnologists have used chambers to simulate stream flow.[7,8,9,10] However, water motion is still one of the least understood factors affecting the growth of benthic plants.[11] Water motion decreases the diffusion boundary layer and thus enhances diffusion of metabolism into plant cells.[12,13]

We have developed a water tunnel which is suitable for both laboratory and in situ field studies of large marine algae.

With this apparatus, one can study the effects of water motion, light, nutrients, gases, temperature, and the metabolic state of the plant. In this paper, we will emphasize the effects of light and water motion on photosynthesis of Macrocystis angustifolia Bory. The morphology and its influence on the patterns of water-flow over the plant surface are also described. The effects of diffusion-limited processes on algal photosynthesis are discussed.

Materials and Methods:

Photosynthetic rates were measured in a Plexiglas water tunnel(Figure 1). Water was recirculated via polyvinyl chloride (PVC) tubing. Water which flowed into the mixing chamber was forced through flow collimators (plastic drinking straws) and a layer of fine nylon netting to produce a unidirectional laminar flow of constant velocity through the specimen chamber. This chamber was 30 x5 x 50 cm narrowing to a 2.6 diameter PVC pipe. Water leaving the chamber was mixed as it passed through a pump (March 2CMD, 4CMD) then passed over a YSI model 5419 oxygen probe inserted in the PVC pipe. The output of the probe was fed into a YSI model 54 dissolved oxygen meter and thence to a chart recorder. The water-flow was then split into two 1.8 cm diameter pipes and fed back into the mixing chamber. Water, air, and nitrogen were added and removed from the mixing chamber

through a standpipe. Access to the specimen chamber was through a lid which could be closed with an O-ring and four bolts. The entire top of the tunnel (110 x 30 cm) was removable for cleaning and was held in place with bolts against a layer of neoprene sealed with a thin layer of petroleum jelly. Water velocity was controlled with a rheostat; it was measured by collecting the output of the 2.6 cm pipe for a measured time interval.

In the laboratory, the tunnel was submerged in a running seawater bath maintained at ambient sea temperature (± 1/2°C) with a thermostat-regulated water chiller. The light source was a 1500-watt Quartaline (GE Q1500T3/CL) lamp. Light intensity was reduced with neutral density Nitex windo screening. Light intensity and spectral distribution were measured with an ISCO model SR spectroradiometer. Heat from the lamp was dissipated by a circulating water bath placed between the lamp and the filters.

Fronds of Pterygophora californica were removed from the kelp bed just off Campus Point (34°21'18" N. Lat., 119°50'36" W. Long.) off the University of California, Santa Barbara during the months March through June. The fronds were placed in a large holding tray in direct sunlight within 15 minutes after collection. Water was circulated through the tray at 60 liters/minute and the water temperature was maintained at ambient (±1°C).

Fronds were kept in this system for a maximum of two days under natural light and photoperiod. A mature surface blade was detached from the frond in a region between the 40th and 100th blade from the meristem, and cleaned of epiphytes. The exposed surface at the base of the vesicle was covered with Parafilm to prevent leakage of metabolites into the surrounding water. The blade was then placed in the water tunnel. Before and after several experiments Winkler[14] were made on samples taken from the mixing chamber to check the calibration of the oxygen electrode.

The water tunnel was also used in the kelp forest during the month of July, 1974. The tunnel was lowered over the side of a small boat. Both the pump and the electrode were submergible and were run off a 12-volt battery with a DC-AC converter. A diver placed the blade in the chamber. Water velocity in the chamber was adjusted to match the *in situ* current velocity which was measured with an Eckman-Merz current meter and dye markers. Duration of Field as well as laboratory experiments varied from 1/2 to 1-1/2 hours.

A hydrodynamic view of the Macrocystis blade (Figure 2) was obtained by using a water tunnel constructed by J. Fletcher in the Engineering Department at UCSB under the direction of Dr. R. S. Hickman. With this tunnel, which is similar to that shown in Figure 1, it is possible to photograph and measure the physical boundary layer and water-flow characteristics over the blade.

For flow visualization, a hydrogen bubble generating wire was placed upstream from the blade. As a pulsed line of bubbles passed over part of the blade, the water-flow patterns were revealed. Values of boundary layer thickness obtained from measurements of photographs were compared with theoretical values for laminar and turbulent boundary layers.

Results:

Under full sunlight or its equivalent lamp intensity, net synthetic rates of Macrocystis blades ranged from 6 to 11 ml $O_2/dm^2/hr$. The average and range of net photosynthetic rates for 8 blades is shown in Figure 3. A water velocity of 4.5 cm/sec was used. These curves increase sharply between 0.0 and 0.2 Ly/min. However, photosynthesis is not saturated at this point. The rates continue to rise an average of 7.3% between 0.4 and 1.8 Ly/min. Each of the eight rates showed significant increases when tested with a self-pairing comparison test. No photoinhibition was noted even at 1.8Ly/min. Photosynthesis to respiration ratios averaged 20/1.

Gross photosynthetic and respiration rates of Macrocystis blades are shown in Figure 4 as a function of water motion. Photosynthetic rates decreased with decreasing water motion from 4.5 cm/sec. Values in still water were positive, but significantly lower than at 4.5 cm/sec.

Values obtained from field measurements of gross photosynthesis during the summer were generally lower than those measured in the laboratory during the spring. Water velocities measured in the kelp bed were also low (Figure 5). Some photosynthetic data are presented along with light intensity, degree of oxygen saturation, temperature, and velocity in Table 1.

Measurements of the physical boundary layer are presented in Table 2. Also included are the theoretical laminar and turbulent boundary layer thicknesses as computed for submerged flat plates.[15] The morphology of the blade 'trips' the flow around the blade from laminar to turbulent in velocities as low as 1 cm/sec.

Discussion:

Photosynthetic rates measured during the present study differ from those presented in earlier papers. Clendenning's rates[1] saturate at 0.2 Ly/min. The rate graphed in Figure 3 has an initial steep increase which slows to an average rate of 7.8%. It does not level off, as to the rates measured by Clendenning. Such increases in photosynthetic rates at high light intensities are characteristic of C_4 plants.[16] Karekar and Joshi[17] found both C_4 and C_3 enzymes and products in brown and green algae. However, it remains to be seen whether Macrocystis is a C_4 plant.

Neither Clendenning's study nor this one show photodestruction at high light intensities. The values recorded under 1.2 Ly/min are slightly higher than Clendenning's for the same time of year, but are much higher than the rates obtained by Towle and Pearse.[3] These workers conducted their experiments during the summer when photosynthetic capacities are much lower than in winter and spring.[1] The values obtained in the field are similar to those recorded by Clendenning and Towle and Pearse for this particular time of year and reflect the general deterioration of the kelp canopy observed during the summer.

As discussed above, currents in a kelp bed may be very low. The flow through a kelp bed is not turbulent, but perfect at these velocities. Perfect flow is analogous to laminar flow, and its characteristics can be predicted by the Prandtl mixing length.[18] Turbulent boundary layers are thicker than laminar layers, but have a greater amount of eddy movement, which would intuitively mean greater diffusion rates of material from the passing water to the plant surface. Water velocities less than 4.5 cm/sec cause a decrease in photosynthesis (Figure 4). Thus, it is possible that in slow-moving water kelp photosynthesis may be diffusion-limited. Blade corrugations 'trip' laminar flow to turbulent in slow moving water (Table 2). Blade corrugations on other algal thalli might also have the same adaptive significance. Pelagophycus inhabits quiet waters off Southern California

islands and it too has blade ridges. However, Nereocystis, which has non-corrugated blades, is restricted to the more turbulent outer coast north of Point Conception, California.

In conclusion, a method of measuring the rate of photosynthesis of benthic marine algal macrophytes has been developed. Rate values which are thought to be close to natural were obtained. These rates may be diffusion-limited at the low velocities which frequently occur in the Campus Point kelp bed. Thus, there seems to be a selective pressure on Macrocystis toward increasing diffusion rates. Since 'triping' the flow past the blade from laminar to turbulent greatly increases the the diffusion rate of metabolites to the cell surface, blades which could to this would have a selective advantage to the plant capable of producing them. The observations made here may have implications pertaining to theoretical discussions of kelp adaptation and evolution and to more practical aspects of kelp bed management. Harvesting practices that enhance water flow through kelp beds could enhance the water motion and thus increase productivity.

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Figure Captions:

Figure 1: Plexiglas, recirculating water tunnel and associated laboratory apparatus. The water tunnel, pump and electrode were also used in the sea. CB: cooling bath; CBL: cooling bath for light; CH: water chiller; E: electrode; FC: flow collimating straws; L: light; M: mixing chamber; N: nitrogen; NDS: neutral density screens; OM: oxygen meter; RE: recorder; RH: rheostat; RL: removable lid; S: standpipe; SC: specimen chamber; SP: submersible pump; V: valve. Arrows indicate direction of flow.

Figure 2: Macrocystis blade in water tunnel (Engineering Department, UCSB). Vertical bands of hydrogen bubbles allow visualization of water flow patterns. Physical boundary layer measurements were made where the leading edge of the pulses intersect the blade. Flow is from left to right at 4.5 cm/sec. Bottom notches are one inch.

Figure 3: The effect of light intensity on the oxygen production of entire blades of Macrocystis in a flow of 4.5 cm/sec. Total water temperature range 10-15°C; controlled to $\pm \frac{1}{2}$°C in each experiment. Mean and range for 8 blades are shown. Δ Denotes a significant increase in slope between points.

Figure 4: The effect of low water velocity on photosynthesis. light intensity of 1.? Ly/min (350-750nm) was used.

Figure 5: Current velocity in the Campus Point kelp bed as measured with an Eckman-Merz current meter and dye markers. Ordinate depicts the number of times a particular rate interval was measured.

Table 1: *In situ* gross photosynthetic rates of *Macrocystis* as measured with the water tunnel submerged in the sea during June and July, 1974. Associated environmental parameters are also shown.

Table 2: Measurements of the physical boundary layer thickness (δ) taken from the photograph in Figure 2. Theoretical laminar and turbulent boundary layer thickness $\hat{\delta}^{(es)}$ of submerged flat plates of the same linear dimension (x) are shown for comparison. Water velocity was 4.5 cm/sec.

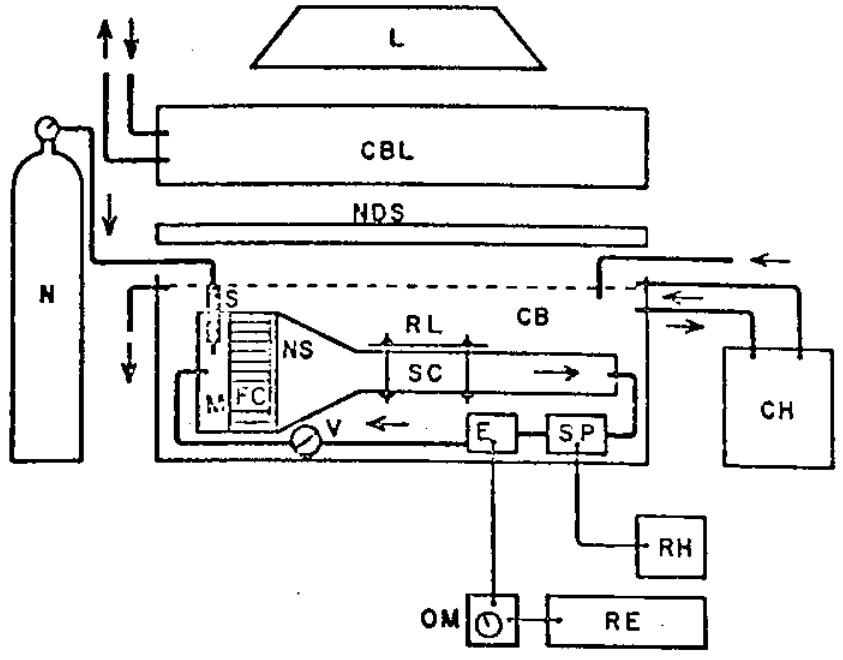

fig 1

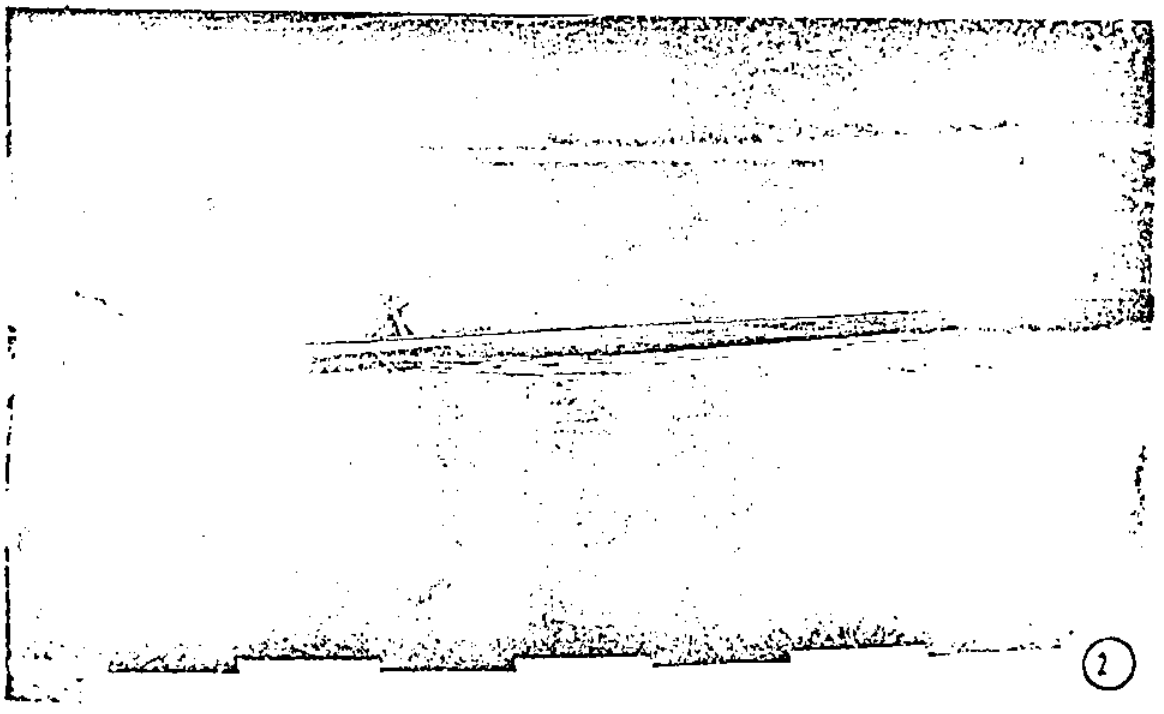

fig 2

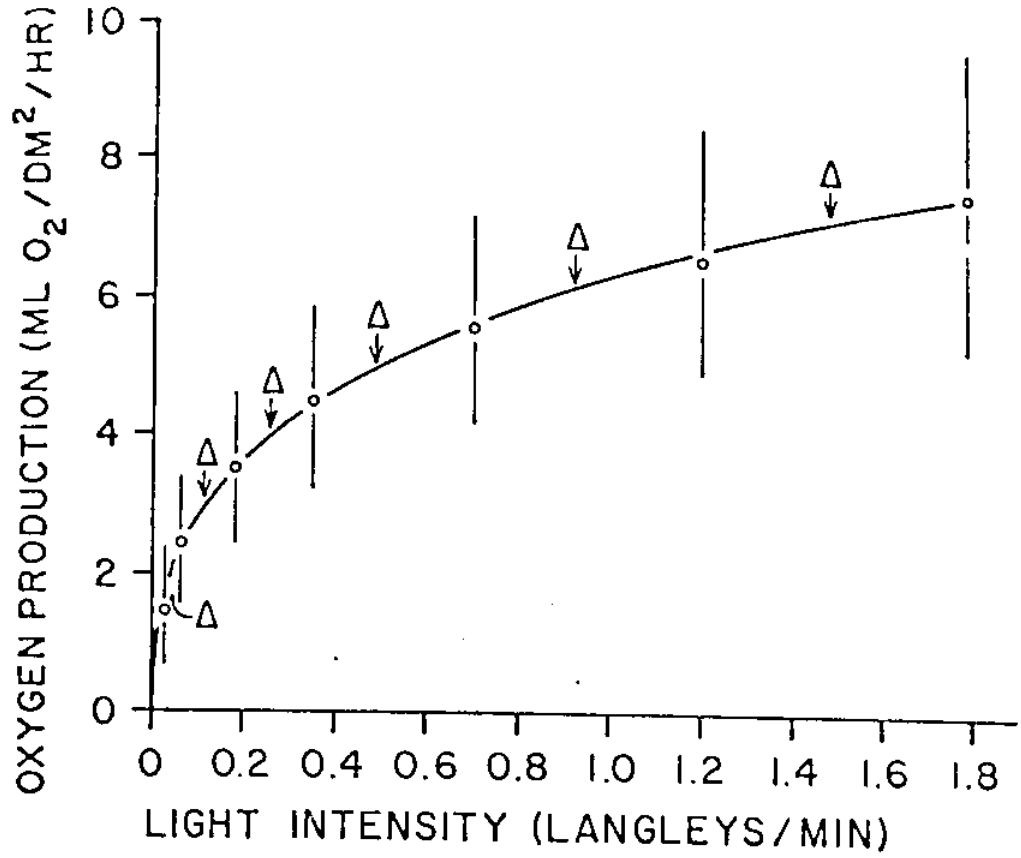
OXYGEN PRODUCTION (ML O_2/DM^2/HR)
10
8
6
4
2
0
0 0.2 0.4 0.6 0.8 1.0 1.2 1.4 1.6 1.8
LIGHT INTENSITY (LANGLEYS/MIN)

fig 3

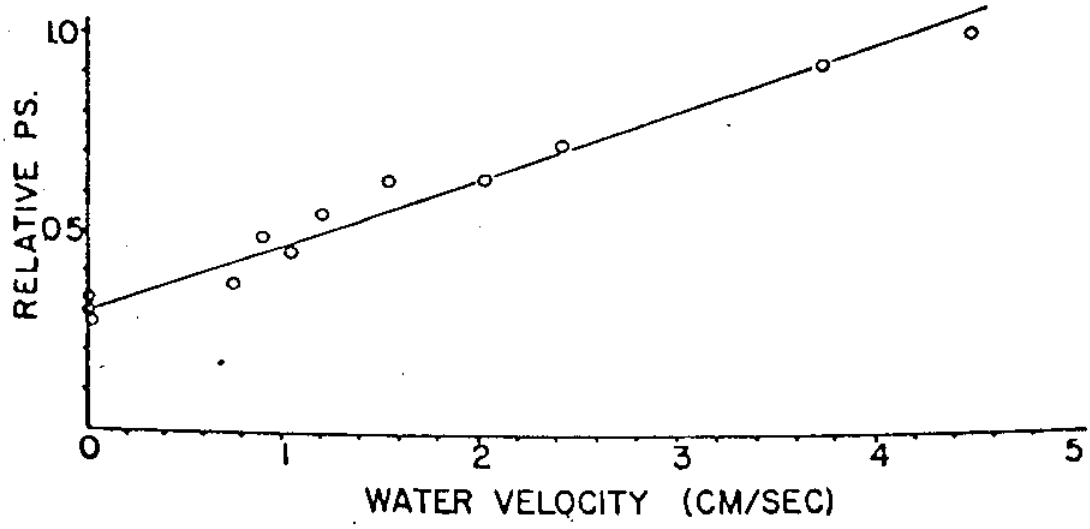
RELATIVE PS.
1.0
0.5
0
1 2 3 4 5
WATER VELOCITY (CM/SEC)

fig 4

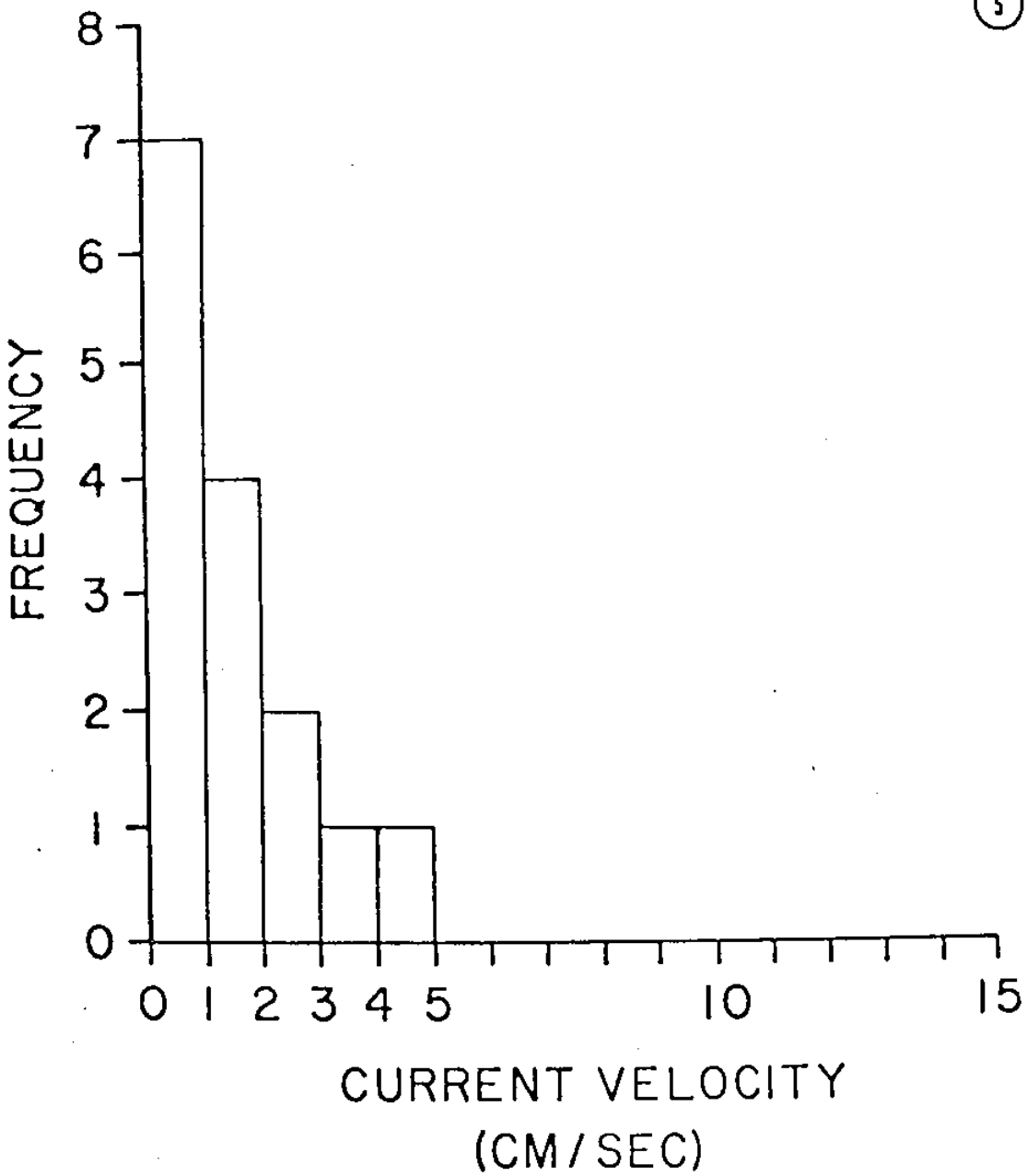

fig 5

(1)

In situ Gross Photosynthetic Rates of Macrocystis angustifolia and Environmental Parameters as Measured During June and July 1974

PS Rates ($mgO_2/dm^2/hr$)	Light Intensity Ly/min	Water Temperature °C	Oxygen Level in Water ppm	Water Velocity in Tunnel cm/sec	Water Velocity in Kelp Bed cm/sec
3.92	0.82-0.89	19	10.85	2	1
2.28	0.52-0.60	18	9.46	2	1.33
3.0	0.60-0.65	17	11.0	2	1.5

table 1

(2)

Theoretical and Measured Boundary Layer Thickness on a Macrocystis angustifolia Blade

Measured		Theoretical	
δ (mm)	Length, (x) (mm)	Laminar (mm)	Turbulent (mm)
1.66	17.80	3.6	1.75
3.12	35.56	5.13	3.0
5.00	111.76	9.09	7.52

table 2