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# Description and Evaluation of Methods for Determining Incident Solar Radiation, Submarine Daylight, Chlorophyll A, and Primary Production

Used by Scripps Tuna Oceanography Research Program  
in the Eastern Tropical Pacific



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UNITED STATES DEPARTMENT OF THE INTERIOR  
FISH AND WILDLIFE SERVICE  
BUREAU OF COMMERCIAL FISHERIES



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By

ROBERT W. HOLMES

Contribution from the Scripps Institution of Oceanography  
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# Description and Evaluation of Methods for Determining Incident Solar Radiation, Submarine Daylight, Chlorophyll A, and Primary Production

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By

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## ABSTRACT

A detailed account and evaluation are presented for certain methods used by the STOR (Scripps Tuna Oceanography Research) Program. This description should assist readers and users of the STOR data and be of general interest to workers in the field.

An Eppley 10-junction 180° pyranometer was used in conjunction with a Speed-omax recorder for the measurement of incident solar radiation. Daily incident radiation was obtained by integration of the daily record with a polar planimeter. Errors due to integration and failure of the gimbals to stabilize the pyranometer completely are estimated to be ± 1 percent and ± 5 percent, respectively. A comparison between observed daily radiation and computed radiation from four standard climatological equations revealed the inaccuracy of using indirect methods for daily totals.

The deck and submarine irradiance meters were equipped with cosine collectors; their construction, calibration, and operation are described. A blue-green Wratten filter (No. 45) was routinely used with both detectors. The relative spectral sensitivity in air and Jerlov ocean water type I was calculated. In air, the maximum transmittance was at 490 mμ and the half-band width was roughly 63 mμ. At 100-m. depth in type I water, this value shifted to 475 mμ with a 50 mμ band width. By assuming the universal validity of the Jerlov sea-water transmission curves, it was possible to obtain estimates of total visible downwelling irradiance at depth from a single attenuation coefficient measurement. Under ideal conditions and with no depth measurement error, a coefficient of variation of ± 10.5 percent was obtained for the attenuation coefficient. This error increased appreciably when vessel motion and errors of depth measurement occurred.

Chlorophyll a measurements were obtained by spectrophotometric method with acetone extracts of membrane-filtered water samples. Corrections for blank values at 750 mμ were applied, and concentrations computed with the equations of Richards with Thompson. The coefficient of variation for surface samples varied between 14 and 27 percent and increased with depth.

Primary production was measured with C<sup>14</sup>; the details of the methodology and standardization are given. Because a number of different incubation techniques were used (i.e., in situ, deck incubator, trailing bottle, and laboratory incubator), an attempt was made to compare these procedures with the in situ method. All of the methods yielded results which were correlated with those obtained by the in situ method. Thus unbiased estimates of in situ production may be obtained from the different incubation techniques.

## INTRODUCTION

The following description of selected methods and their partial evaluation is a detailed account of some of the instrumentation and procedures which have been used by the author in 1956-65 and by several of his colleagues in the STOR (Scripps Tuna Oceanography Research) Program in 1958-62 in the eastern tropical Pacific. Only abbreviated outlines of these methods have been published (Holmes, Schaefer, and Shimada, 1957; Holmes and others, 1958; Holmes and Blackburn, 1960; Blackburn, Griffiths, Holmes, and Thomas, 1962), and no accounts of statistical reliability of the methods have appeared. Because data obtained with these methods are being widely used, and because I have completed a manuscript that deals with the statistical and descriptive interrelations of the variables measured by these methods, an account of the methods is believed to be timely and of scientific value.

## INCIDENT SOLAR RADIATION

Because of the importance of solar energy in the photosynthetic process, incident solar radiation upon a horizontal plane was measured continuously on all STOR cruises. Although the data have not been used for heat-budget studies, they have been analyzed here with a view toward assessing the accuracy with which the daily incident radiation total may be estimated by indirect means.

### Instrumentation

Eppley<sup>1</sup> 10-junction pyranometers, formerly called pyrliometers (The Eppley Laboratory, Inc., Newport, R.I.), have been used throughout this investigation.

The 180° pyranometer (Drummond, 1961) was mounted above most of the vessel's superstructure on a small metal base which was shock-mounted to an identical pendulum-balanced platform. Except in the roughest seas, this gimbal's mounting maintained the instrument in a nearly horizontal plane.

The electromotive force generated by the 10-junction pyranometer, about 2 mv./g. cal./cm.<sup>2</sup>/min. (millivolts per gram calorie per square centimeter per minute), was recorded with a 0- to 10-mv. Speedomax Model G or H recorder (Leeds and Northrup). Interference originating from the vessel's radiotransmitter antenna during transmission was eliminated by using shielded signal leads and appropriate radio-frequency filters.

The response of the Eppley instrument to energy between 400 and 25,000 m $\mu$  is nearly linear. Incident solar radiation active in algal

photosynthesis, 400 to 700 m $\mu$ , was assumed to average roughly 45 percent of the incident energy recorded (Withrow and Withrow, 1956), regardless of the degree of overcast.

Daily radiation totals were obtained by integration of the unsmoothed daily records with a polar planimeter. The accuracy of the daily radiation totals is not precisely known. Sources of error include: (a) calibration error; (b) failure of the pyranometer to maintain its calibration; (c) recorder error; (d) failure to keep the pyranometer oriented in the same direction at all times; (e) failure to keep the pyranometer horizontal under all sea conditions; and (f) integration errors.

A rough estimate of the total error in the daily radiation total, which includes the above-mentioned sources of error, excepting items a and b, was obtained in the following manner: the solar radiation record for February 19, 1959, was integrated twice with a polar planimeter, once by use of the upper limit of the pen excursion and once by use of the lower limit of the pen excursion. The values obtained were 525 and 478 g. cal./cm.<sup>2</sup>/da., respectively.

Qualitatively it has been noted for a constant recorder-amplifier sensitivity that the amount of pen excursion (i.e., the width of the trace) appears to vary with sea conditions. During darkness, regardless of sea state, the width of the pen trace remained virtually constant at 0.9 mm. On calm clear days the trace width rarely exceeded 1 mm., but on rough days it averaged 2 mm., and once it reached an extreme width of 5 mm.

Had the central point of the pen excursions been followed during the above integration, the daily total would have equalled 501 g. cal./cm.<sup>2</sup> and the range would become  $\pm 24$  g. cal./cm.<sup>2</sup>/day, or roughly  $\pm 5$  percent of this value. Since the sea on this particular day was extremely rough, I believe that the values for the daily radiation totals are rarely in error by more than  $\pm 5$  percent.

According to the pyranometer catalog issued by the Eppley Laboratory, the probable error for the pyranometer in the range 0.25 to 1.50 g. cal./cm.<sup>2</sup>/min. is  $\pm 1.5$  percent. This figure presumably covers calibration error and includes changes in pyranometer sensitivity during the calibration period, as well as recorder error.

One of the two pyranometers used during this study was recalibrated by the Eppley Laboratory after 3 years of use at sea. The change in sensitivity amounted to +0.12 mv. (e.g., from 2.53 to 2.65 mv.) per g. cal./cm.<sup>2</sup>/min. Unfortunately, it is not known if this change is typical for this class of instruments or whether the change in sensitivity was gradual or abrupt. As a result of this sensitivity change, however, an undetermined number of the daily radiation totals will be roughly 5 percent too high.

<sup>1</sup>Use of trade names in this publication does not imply endorsement of commercial products.

Although pyranometers are calibrated with the exposed internal and external leads pointing in a northerly direction, the manufacturer states (personal communication) that failure to keep the instrument oriented in this direction should not introduce serious error or inconsistencies. I have no data which will permit rigorous assessment of errors introduced solely by the failure of the gimbals' support to keep the pyranometer in a horizontal plane. As mentioned above, the gimbals seem to stabilize the pyranometer rather well except in very rough seas.

Daily records for clear and overcast conditions were selected to determine the variability due to integration; each trace was integrated 10 times. A comparison of results (table 1) shows that the coefficient of variation was 1.24 percent on the day with abundant high clouds, and 0.37 percent on the uniformly overcast day.

It should be mentioned that for data collected before 1957 and used in the evaluation of indirect methods of predicting daily totals (see table 2), the daily totals have been reduced by 2 percent (Drummond, 1961). This change is necessary because pyranometers used before December 1957 were calibrated according to the Smithsonian scale of 1913 rather than the International Pyrhelometric scale of 1956.

To recapitulate, the daily radiation totals are believed to be accurate on average within  $\pm 7$  to 10 percent of the reported value.

### Comparison of Observed and Computed Values

Although continuous measurements of incident solar radiation falling upon a horizontal plane have been obtained on a majority of cruises and expeditions during this study, the data are not yet sufficient to permit the construction of maps giving seasonal annual insolation values. The data are sufficient, however, to permit a preliminary examination of the relation between observed daily radiation totals and similar totals computed from a selected number of climatological equations which use cloud-cover observations. It will become apparent that these climatological equations do not yield precise estimates of daily insolation and that their use for estimating daily totals should be avoided unless the relatively large errors attendant upon their use can be tolerated.

Four equations were chosen for these comparisons: that of Kimball (1928), the Savino-Ångström formulation (Budyko, 1956), that of Black (1956), and that of Laevastu (1960). In

Table 1.--Variability of 10 successive planimeter integrations of two daily incident solar radiation records with different cloud cover, eastern tropical Pacific, 1958

Weather conditions	Values of planimeter integrations											Mean ( $\bar{x}$ )	Standard deviation ( $\bar{s}$ )	Coefficient of variation
	----- Planimeter units -----													
Abundant high clouds	2,567	2,649	2,606	2,588	2,602	2,660	2,558	2,625	2,598	2,611	2,606	32.4	1.24	
Moderate uniform overcast	1,740	1,762	1,753	1,758	1,757	1,749	1,750	1,755	1,750	1,745	1,752	6.51	0.37	

Table 2.--Statistical results of 104 comparisons between observed daily totals of incident solar radiation falling upon a horizontal surface and values computed from four climatological equations (Kimball, Savino-Ångström, Black, and Laevastu) or taken from Black's monthly maps, 1956-59

Source of value	Observed	Kimball (1928)	Savino- Ångström (Budyko 1956)	Black (1956)		Laevastu (1960)
				Monthly maps	Equation (3)	
Total radiation for 104 days.....	49,423	52,205	53,108	44,355	42,255	70,254
Mean daily total (g.cal./cm. <sup>2</sup> ).....	475	502	511	426	406	676
Correlation coefficient	-	<sup>1</sup> 0.71	<sup>1</sup> 0.72	<sup>1</sup> 0.40	<sup>1</sup> 0.74	<sup>1</sup> 0.78
Regression equation....	-	Y = 0.940X	Y = 0.928X	Y = 21.11X	Y = 1.153X	Y = 55 + 0.621X
Sample standard deviation from regression.	-	94.4	91.2	120	88.9	81.9
Significance of regression coefficient.....	-	t = <sup>1</sup> 11.4	t = <sup>1</sup> 11.1	t = <sup>1</sup> 4.62	t = <sup>1</sup> 14.1	t = <sup>1</sup> 12.6

<sup>1</sup> Significant at the 1-percent level.



addition, the values obtained by the use of Black's (1956) monthly charts were examined.

The equations are as follows:  
Kimball (1928):

$$Q = Q_0 (0.29 + 0.71 [1.0 - C]) \quad (1)$$

where  $Q$  is the calculated average daily total of solar radiation received on a horizontal surface (direct and diffuse),  $Q_0$  is the average daily total of solar radiation with a cloudless sky, and  $C$  is the proportion of sky covered by clouds.  
Savino-Ångström (Budyko, 1956; Burdeki, 1958):

$$(Q + q) = (Q + q)_0 (1 - [1 - K] n) \quad (2)$$

where  $(Q + q)$ ,  $(Q + q)_0$ , and  $n$  are identical with the Kimball quantities  $Q$ ,  $Q_0$ , and  $C$ , and  $K$  is the ratio between the actual radiation under overcast sky conditions and the possible radiation (Budyko, 1956; p. 31).  $K$  varies with the region and with latitude, and diurnally and seasonally. Values of the coefficient  $K$  have been calculated and averaged for various latitudes by Budyko (1956; table 2).

Black (1956):

$$Q = Q_A (0.803 - 0.340C - 0.458C^2) \quad (3)$$

where  $Q = Q_0 = (Q + q)$ ,  $C = n = C$  above, and  $Q_A$  is the maximum possible radiation in the absence of an atmosphere.

Laevastu (1960):

$$Q_s = 0.014 A_N t_d (1 - 0.0006C_3) \quad (4)$$

where  $Q_s = (Q + q) = Q$  above,  $A_N$  is the noon sun altitude,  $t_d$  the day length in minutes, and  $C$  the cloudiness (in tenths).

These equations were developed with different purposes in mind. The Kimball and Savino-Ångström equations have been used by oceanographers in heat-budget studies and for estimating the amount of solar energy available for photosynthesis. The Black equation was developed in a recent study of the distribution of solar energy on the earth's surface at monthly intervals. Laevastu has attempted to devise a formula which is suitable for the computation of insolation on a daily basis. Although more sophisticated equations have been proposed which take into account the cloud cover at different altitudes, water vapor content of the atmosphere, . . . . (see Bortovskii, 1961), they cannot be evaluated here because the necessary data are not available.

Each of the four equations given above is different because somewhat different theoretical considerations attended their development. Kimball's equation is based on the assumption that the amount of solar energy reaching the earth's surface is proportional to cloudiness and that the transmission of the atmosphere under cloudless conditions approaches a constant. In the Savino-Ångström equation the underlying assumptions are similar, except that a coefficient  $K$  is introduced to take into

consideration the seasonal and latitudinal variations in the effect of cloudiness. Black chose to evaluate the effects of clouds by considering the relation between cloudiness and the amount of radiation that would reach the earth's surface in the absence of an atmosphere, and obtained a regression which takes a quadratic form. Laevastu assumed that the amount of daily insolation on the ocean surface under a clear sky depends mainly on day length and the noon altitude of the sun, but also provided a correction for cloudiness.

The summarized data used in these computations and calculated daily radiation totals on the basis of the five methods are presented in table 2. It should be noted that two series of figures are tabulated for the Black method; one series was obtained from the monthly charts prepared by Black, and the other was computed from the Black equation (3) given above. The value of  $Q_0$  in the Kimball equation was taken from the tabulation of  $Q_0$  values (i.e.,  $(Q + q)_0$ ) of Budyko (1956, table 1) rather than from the tabulation of Kimball (1928, table 3).

To obtain a quantitative estimate of the error arising from the use of climatological equations in the eastern Pacific, a comparison by linear regression techniques was made between the measured values and those obtained with the four climatological equations. In each comparison between the measured and computed values the correlation was significant ( $p \leq 0.01$ ). The regression coefficients were highly significant, and the sample standard deviations ranged from 81.9 to 120 g. cal./cm.<sup>2</sup>/da. The intercept differed significantly from zero only in the Laevastu comparison.

Both the Kimball and Savino-Ångström equations yielded estimates that were roughly 6 to 7 percent higher than the observed value, whereas the Black map and equation underestimated the mean value by a somewhat greater amount. The Laevastu equation overestimated the mean value considerably, and the failure of the regression line to pass through the origin indicates additional bias. Of course, neither the Kimball, Savino-Ångström or Black equations, nor the Black map, were designed for the estimation of a given daily radiation total, and their use for such a purpose is not recommended unless a relatively large standard error of estimate can be tolerated.

On the few occasions when cloudiness was essentially lacking during the day, the Kimball, Savino-Ångström, and Black equations yielded daily total estimates that were in excess of the observed daily radiation total. This disagreement suggests that the tropical air mass is somewhat more opaque to solar energy under a clear sky than the equations predict. Unfortunately, the data for clear skies are so few that the correctness of this conclusion must be re-examined after more data become available.

The regression equation computed from the Laevastu formulation yields the lowest standard error of estimate, but it is immediately apparent that the Laevastu equation grossly overestimates the daily total as judged by the mean value of the 104 test observations, as well as the regression equation (table 2). As was true with the other equations, changing the constants in the equations could improve agreement between the observed and predicted values. The tendency of this equation to overestimate incident solar radiation under cloudless skies is also apparent in a comparison of the tabulated values of Budyko (1956) at 0, 10, and 20°N, for the 15th day of each month of the year with that computed by the Laevastu equation. The total of incident solar radiation for the 36 days was 22,941 g. cal. per square centimeter according to Budyko (1956; table 2) and 26,887 g. cal. per square centimeter according to the Laevastu equation. The daily difference amounts to 101 g. cal. per square centimeter. The results of this comparison and the one above suggest that the Laevastu equation has a systematic error which should be evaluated before application in tropical ocean areas.

It seems doubtful that equations such as those discussed above will provide adequate estimates of daily insolation on any given date at any location in the ocean. Direct measurement of incident solar radiation is certainly to be desired, although more complicated equations that use data on relative humidity, and type, amount, and height of clouds will probably give better estimates than do the simpler equations.

## SUBMARINE IRRADIANCE

### Definitions

Submarine irradiance was originally measured to determine the rate of attenuation of diffuse downwelling irradiance with depth and its relation to photosynthetic rates. During this study it became evident that the average diffuse attenuation coefficient,  $k$ , was significantly correlated with chlorophyll  $a$ , and, in fact, could be successfully substituted on occasion for chlorophyll  $a$  in certain statistical techniques that lead to the prediction of zooplankton standing crop. Thus the irradiance measurements obtained with the equipment described below have been used in two separate contexts, which I describe elsewhere.

Irradiance at a specified wavelength is defined as follows:

$$H = \int_0^{\pi/2} N \cos \theta \, d\omega$$

where  $H$  is radiance,  $\theta$  is the angle of incidence and  $\omega$  the solid angle measure (Preisendorfer,

1960). The value of the diffuse attenuation coefficient  $k$  is given by

$$\frac{H_{Z_2}}{H_{Z_1}} = e^{-k\Delta Z}$$

or, in the more common form

$$k = \frac{\log_e H_{Z_1} - \log_e H_{Z_2}}{Z_2 - Z_1}$$

where  $H_{Z_1}$  and  $H_{Z_2}$  are irradiance values at the shallower and deeper depths, respectively.  $H$  must be specified as to direction (downwelling or upwelling) and wavelength.

In this paper  $k$  is always calculated from downwelling irradiance values from the third equation given above. The assignment of wavelength specificity is discussed below.

### Instrumentation

The underwater and deck detector units contained a Weston self-generating barrier layer cell (Type 856RR) mounted in watertight housings. Optical coupling between the water and the barrier layer cell was obtained with an abraded translucent plastic collector with cosine collecting properties. The deck unit was mounted in gimbals above most of the vessel's superstructure, to give a nearly uninterrupted view of the sky.

A Wratten No. 45 filter (blue-green) was used in the underwater unit. The half-band width of the complete unit is estimated to be about 63  $m\mu$  and to have peak transmittance centered at 490  $m\mu$  (table 4). In ocean water the transmittance peak and half-band width of the unit changes with depth as a result of the interaction between the changing spectral distribution of the water and the detector's nonlinear spectral response.

Most of the measurements in this study were made in water types I, II, or III (Jerlov, 1951: table 9), and calculations show that the attenuation coefficients measured with the equipment may be assigned to 475  $m\mu$ .

### The Irradiance Meter

A number of different submarine irradiance meters provided with nearly identical detector units have been used to measure relative downwelling irradiance. Diffuse attenuation coefficients have been calculated from irradiance values obtained at different depths in the upper 20 to 120 m.

Each detector unit (fig. 1) consists of a photovoltaic cell housing (D) and a threaded cap (C). The metal or plastic housing (D) contains a photovoltaic cell (G) and an optical filter (E). Silica gel, placed in the cavity



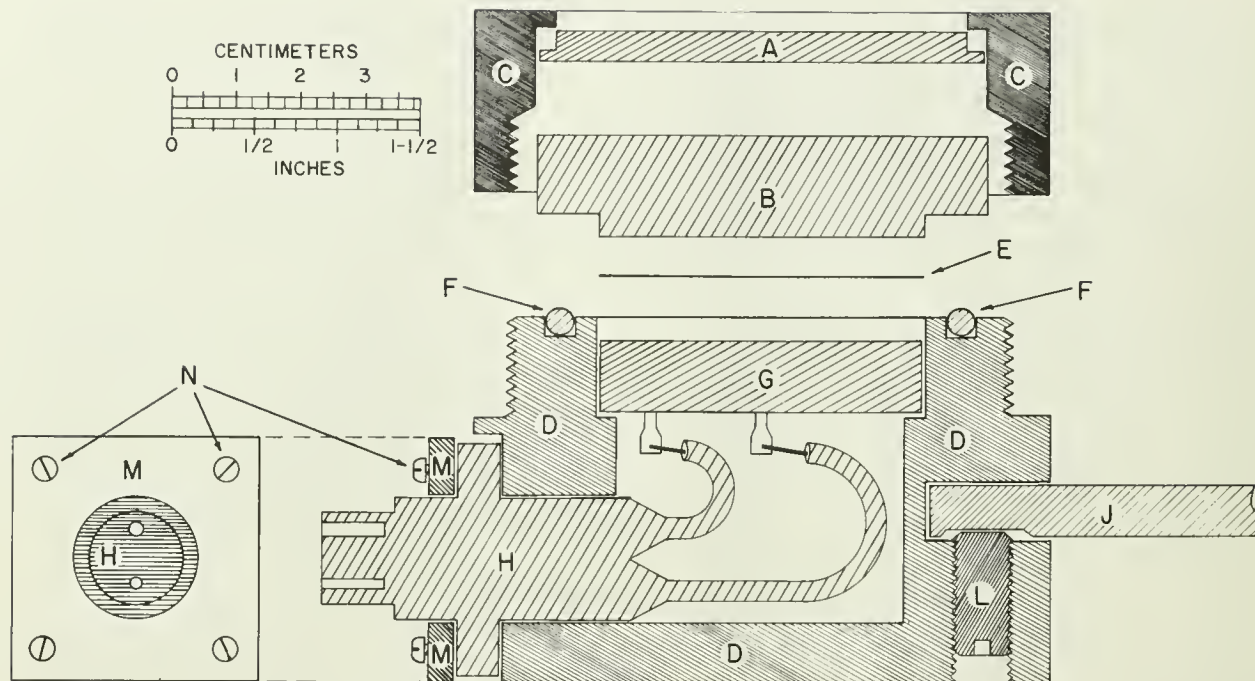


Figure 1.--A typical irradiance meter detector housing: A, cosine collector; B, clear plastic window; C, threaded brass cap; D, brass photovoltaic housing -- base section; E, optical filter; F, O-ring; G, photovoltaic cell; H, Joy plug; J, supporting arm; L, locking screw for supporting arm; M, pressure plates; N, screws for pressure plate.

below the photovoltaic cell (G), prevents possible internal window fogging and internal moisture damage to gelatin-type filters resulting from condensation of water vapor contained in the detector unit. The two conductors of the Joy plug (H) (Joy Manufacturing Co., Compton, Calif.) are connected to the photovoltaic cell terminals. A watertight seal is effected between the housing (D) and the Joy plug (H) by tightening the screws (N) of the pressure plate (M) and thus compressing the external rubber collar of the Joy plug. A recessed shoulder on the bottom of the clear plastic window (B) holds the filter in place. The cosine or Lambert collector (A) is placed above the window and may be fused to it by use of a suitable solvent. The collector (A) is so machined that 1 to 2 mm. protrude above the threaded cap (C) when the cap is tightened. The watertight seal is obtained by tightening the cap with enough force to effect a seal between the silicone-lubricated O-ring (F) and the window (B). One or more holes are drilled part way into the housing (D) to accommodate one or more supporting arms (J) which may be locked into position with set screws (L).

The cosine collector (fig. 1) was fabricated from translucent plastic (Plexiglass No. 2333, Rohm and Hass, Philadelphia, Pa.), the external surface of which had been abraded with No. 180 grit carborundum. No laboratory tests of the collecting properties of this plate were made. The design paralleled that of Foster (1951), who used pot opal glass rather than

plastic for the collector. Foster found that elevating the surface of the collector above the instrument housing permitted flux to pass through the edge of the collector; this arrangement helped to compensate for the losses due to specular reflection at low angles of incidence. Since little of the downwelling flux reaching a horizontal plate suspended in the sea strikes at low angles, this design is believed to provide adequate data for biological studies.

Hermetically sealed barrier layer cells, the self-generating dry-disk type (Model 856RR) manufactured under the name Photronic Photoelectric Cell by the Weston Electrical Instrument Corp., Newark, N.J., have been used as the radiant flux detectors. These photovoltaic cells have proved stable and sufficiently rugged for use in this application.

The signal generated by the photovoltaic cell was conducted to the vessel by a two-conductor (steel wire, No. 18 gage) neoprene-covered cable, which also supported the underwater components of the irradiance meter. Waterproof connections between the support cable and the underwater components of the meter were made with matching polarized Joy connectors (fig. 1, H). This configuration made it possible to detach and store the underwater unit in a protected location on shipboard between lowerings.

The support cable was handled from either a hand-operated or motor-driven winch. During a measurement series, the cable passed over

a block (pulley) located at the end of a 5-m.-long boom--or a 2-m. boom on SCOPE Expedition (see Holmes et al., 1958). The boom was generally secured to the stern quarter of the vessel, although on cruises TO-58-2 and TO-59-1 (see Blackburn et al., 1962) it was located amidships. The cable was marked with colored tapes at spaced intervals which indicated distance above the underwater detector unit. Wire-angle measurements during a lowering were made by visually superimposing the edge of a conventional wire-angle indicator with the support cable and reading the angle.

Electrical connection between the winch end of the support cable and the associated deck equipment was achieved in one of two ways. With the hand-operated winch used on SCOPE Expedition (Holmes et al., 1958) and the motor-driven winch used on SCOT Expedition (Holmes and Blackburn, 1960), electrical connection between the microammeter and detector was made manually by inserting a polarized plug in an appropriate socket on the winch spool. A shorting plug was replaced by the microammeter plug between measurements. After SCOT Expedition, gold-plated slip rings were incorporated into the winch design so that electrical continuity was maintained between the microammeter and the detector at all times. When measurements were not being made, shorting of the photovoltaic cell as recommended by the manufacturer was easily accomplished by using the shorting position on the scale switch incorporated in the Rawson microammeter.

During lowerings of the irradiance meter, the ambient light incident upon the sea surface was monitored by a gimbal-supported waterproof deck cell mounted above the vessel's superstructure. The light-sensitive element and flux collector were the same type as those used in the submarine irradiance meter. A reducing screen of thin stainless steel with equally spaced holes about 2 mm. in diameter was placed between the cosine collector and photovoltaic cell. No filter was used. The current generated by the deck irradiance meter was measured with a 0- to 1-milliampere meter of low resistance (about 50 ohms) or, on TO-59-1 and subsequent cruises, with a Rawson-type 501 meter.

The current generated by the photovoltaic cell in the submerged detector and by the deck cell (see above) was measured with an internally damped Rawson microammeter, type 501 (Rawson Electrical Instrument Co., Cambridge, Mass.). The damping feature (45-second period), incorporated into the meter by the manufacturer at my request, permits measurement of irradiance levels near the sea surface (upper 15-20 m.). In this depth range, vessel surge and the focusing of incident specular energy from surface ripples and waves produce a pulsating DC signal with

a frequency of the order of 50 to 125 cycles per minute (see Tyler, 1960; table 11). This fluctuating signal makes measurements with an undamped meter almost impossible. In the deeper water the damping feature is useful on occasion but is not necessary.

The Rawson-type 501 meter is designed for use with photovoltaic cells and possesses a switch which allows the operator to short-circuit the photovoltaic cell or select any one of 5 scales (50, 100, 200, 500, and 1,000 microammeter), each having an internal resistance of 100 ohms.

These improvements do not affect the reliability of  $k$  measurements in general, and all of the measurements obtained during this investigation are considered comparable. A brief discussion in another section, however, mentions the adequacy of depth determinations calculated from wire length-wire angle relations, which incorporates information obtained on cruise TO-60-1 with the depth-sensing device.

### Characteristics of the Photovoltaic Cells and Filters

The photovoltaic cells and filters used to measure attenuation coefficients possess electrical and physical characteristics that affect the precision and accuracy of the  $k$  values obtained.

The current generated by a Photronic cell in a constant monochromatic light field is a function of its sensitivity, the temperature, and the external circuit resistance. Technical information supplied by the manufacturer shows that the percentage deviation in output at the maximum and minimum light levels and the temperature range (5-30°C.) encountered in these studies does not exceed  $\pm 2$  percent, at the external resistance of the circuitry (meter and photometer cable) used ( $105 \pm 1$  ohm). The present data have not been corrected for this temperature effect.

Although the same microammeters have been used and the detector design has remained essentially constant during these investigations, a number of improvements have been made in the design of the equipment. These improvements principally involved auxiliary equipment which permits the measurement of attenuation coefficients in different spectral regions, and ratios in the same spectral region of upwelling, downwelling, and horizontal irradiance. This additional work may now be accomplished over the same two-conductor cable without the return of the detector units to the surface or changing the orientation of individual detectors. Furthermore, a depth-sensing unit accurate to within  $\pm 2$  percent in the depth range 25 to 150 m. has been developed which telemeters the depth signal over the same two-conductor cable to a deck component, where depth may be read

simultaneously with detector output. These modifications have been reported in detail by Holmes and Snodgrass (1961).

As the light energy level increases, the linearity of response of the photovoltaic cell is affected greatly by the external circuit resistance; the response becomes more and more linear at a given temperature as the external circuit resistance approaches zero. At the external circuit resistance of the order of 100 ohms, as used in the present study, the departure from linearity is generally not

noticeable between 100 and 300  $\mu\text{a}$ . output but is marked at 1,000  $\mu\text{a}$ . The linearity varies considerably between different cells, although a given cell seems to be exceptionally stable in this respect. For this reason, the linearity of response up to 1,000- $\mu\text{a}$ . output has been determined for each cell before and after use on a cruise. This linearity test is made on an optical bench and with the same galvanometer and cable that were used for the actual measurements at sea. The results of such a typical test are illustrated in figure 2.

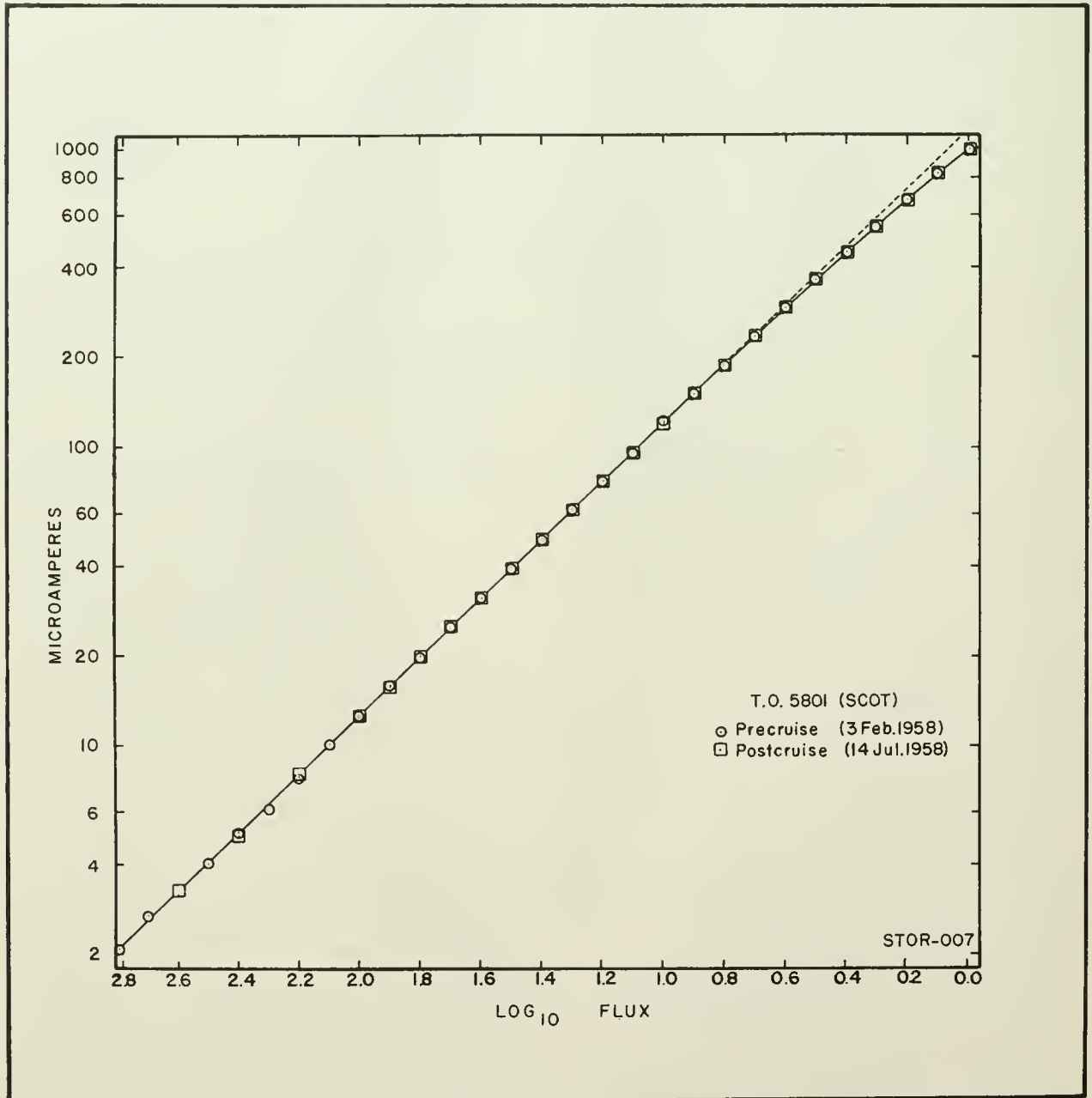


Figure 2.--Linearity of response of a Weston 856RR Photronic cell under a load of  $105 \pm 1$  ohms.



Except at SCOPE Expedition station 9C, all measurements of irradiance have been made with a blue-green (peak transmission at 480 mμ) gelatin filter (Wratten No. 45). On clear or slightly overcast days, this cell-filter-meter combination permits measurements to a depth where about 5 percent of the incident downwelling blue-green flux is present.

This particular gelatin filter has the disadvantage that it becomes progressively more opaque with exposure to intense light (table 3), although the transmittance does appear to increase somewhat when the filter is stored in darkness between exposures to light. Fortunately, the transmission peak does not shift appreciably, and the increase in opacity is slow enough under in situ measurement conditions so that its effect on any one set of observations from which attenuation coefficients are obtained is negligible.

#### Relative Spectral Sensitivity of the Detector in Air and in Sea Water

The output of the detector when lowered in water will be proportional to the summation of the wavelength-by-wavelength product of

the wavelength-dependent parameters (notation from Tyler, 1959), namely:

- (a) the energy distribution of the light immediately below the water surface ( $E_\lambda$ ),
- (b) the spectral sensitivity of the photovoltaic cells ( $S_\lambda$ ),
- (c) the transmittance of the filter ( $T_F$ ),
- (d) the transmittance of the window ( $T_O$ ),
- (e) the transmittance of the cosine collector ( $T_C$ ),
- (f) the transmittance of the water path ( $T_W$ ).

Thus any output value ( $V$ ) of the detector can be expressed as follows (Tyler, 1959):

$$V = k \sum_{\lambda_1}^{\lambda_2} E_\lambda S_\lambda T_F T_O T_W T_W \Delta \lambda$$

where  $k$  is a constant of proportionality, and the limits  $\lambda_1$  and  $\lambda_2$  are determined by one or more of the wavelength-dependent parameters.

We may compute the spectral response of the detector without initial reference to  $E_\lambda$  and  $T_W$ . In table 4 the relative spectral sensitivity of the Photronic cell ( $S_\lambda$ ), the transmittance values obtained for a Wratten No. 45 filter ( $T_F$ ), a plastic window ( $T_O$ ), and a

Table 3.--The progressive decrease in transmittance (percentage T) of a Wratten No. 45 filter after successive exposures to direct sunlight, 1957

[Measured with a Beckman Model DU spectrophotometer]

Wavelength	New filter (22 May)	Exposures				After 24 hrs. in dark (29 May)
		0815-1610 (25 May)	0805-1610 (26 May)	0815-1615 (27 May)	1830-1630 (28 May)	
----- -Percent T - -----						
400	0.00	0.00	0.00	0.00	0.00	0.00
410	.00	.00	.00	.00	.00	.00
420	.00	.00	.00	.00	.00	.00
430	.10	.00	.00	.00	.00	.11
440	4.00	1.96	1.28	1.10	1.10	2.40
450	18.8	10.9	6.98	5.91	5.40	8.50
460	29.9	19.3	13.1	11.4	10.0	12.6
470	35.5	24.1	16.9	14.4	12.3	14.1
475	36.7	25.3	17.6	15.0	12.7	14.2
480	36.9	25.7	17.8	15.1	12.9	14.2
485	36.6	25.5	17.5	14.9	12.6	14.0
490	35.6	25.0	16.8	14.0	11.9	13.2
500	30.2	20.9	13.8	11.6	9.60	11.1
510	20.3	13.9	8.98	7.42	6.20	7.60
520	9.50	6.39	4.30	3.45	2.90	3.82
530	2.50	1.74	1.07	1.00	.85	1.18
540	.35	.20	.11	.10	.10	.19
550	.00	.00	.00	.00	.00	.02
560	.00	.00	.00	.00	.00	.00
580	.00	.00	.00	.00	.00	.00
600	.00	.00	.00	.00	.00	.00

Table 4.--Values of  $S_{\lambda}$ ,  $T_F$ ,  $T_O$ , and  $T_C$  and the resultant detector sensitivity without reference to the spectral energy distribution of a radiant energy source ( $E_{\lambda}$ ) or properties of an absorbing medium ( $T_W$ )

$\lambda$	$S_{\lambda}^1$ Relative spectral sensitivity of photronic cell	$T_F^2$ Wratten No. 45 filter	$T_O^3$ Plastic window	$T_C^3$ Cosine collector	Sensitivity	
					Relative	Adjusted to 1.00 at 490 $m\mu$
$m\mu$		----- Percent T -----				
400	.52	0.0	90.4	11.0	0	0
410	.56	0.0	90.8	14.8	0	0
420	.62	0.0	90.9	16.7	0	0
430	.66	0.1	91.0	18.4	0.00011	0.0018
440	.71	4.0	91.1	19.1	0.00494	0.0810
450	.75	18.8	91.2	19.7	0.0254	0.416
460	.80	29.9	91.2	20.1	0.0438	0.718
470	.84	35.5	91.2	20.3	0.0552	0.905
475	-	-	91.2	-	-	<sup>4</sup> 0.96
480	.87	36.9	91.2	20.7	0.0607	0.995
490	.90	35.6	91.4	20.9	0.0610	1.00
500	.93	30.2	91.6	21.1	0.0542	0.888
510	.96	20.3	91.8	21.3	0.0381	0.625
520	.98	9.5	91.8	21.5	0.0184	0.302
525	-	-	91.8	-	-	<sup>4</sup> 0.150
530	.99	2.5	91.8	21.6	0.00492	0.0807
540	1.00	0.3	92.0	21.7	0.000736	0.0121
550	1.00	0.0	92.0	21.8	0	0
560	.99	0.0	92.2	21.9	0	0
570	.99	0.0	92.2	22.0	0	0
580	.98	0.0	92.2	22.0	0	0
600	.93	0.0	92.2	22.0	0	0
620	.75	0.0	92.2	21.7	0	0
640	.52	0.0	92.2	22.0	0	0
660	.31	0.0	92.0	21.6	0	0
680	.17	0.0	92.0	21.4	0	0

<sup>1</sup> Data from manufacturer's published specifications.

<sup>2</sup> Data obtained with Beckman Model DU spectrophotometer.

<sup>3</sup> Data obtained with a Gary 14 recording spectrophotometer.

<sup>4</sup> Value obtained by graphical interpolation from plot of normalized values from 420-550  $m\mu$ .

cosine collector ( $T_C$ ) are given. The product at each 10- $m\mu$  interval of the four values has been obtained and normalized to 1.00. The maximum transmittance of the detector unit employed in these studies in air is located at 490  $m\mu$  with a half-band width of roughly 63  $m\mu$ .

Next we may consider the effects of  $E_{\lambda}$  and  $T_W$ . The data of Moon (1940) have been used to compute the percentage of incident visible radiant energy at the sea surface at intervals of 25  $m\mu$  from 387.5 to 712.5  $m\mu$  (table 5). The percentage of energy within each 25  $m\mu$  interval has been assigned to the mean wavelength over that interval. The intervals have been chosen to permit the use of Jerlov's (1951) transmission curves (see below). The particular set of data used in this computation (Moon, 1940) refers to an air mass of two (solar altitude 30°). Moon (1940) recommended

this curve for general use. Reflection at the sea surface, whether selective or nonselective, has been ignored in these calculations.

The optical-transmittance characteristics of different sea waters for downwelling irradiance have been conveniently classified into ocean and coastal water types by Jerlov (1951). Since the Jerlov types most frequently encountered in this investigation may be characterized as ocean water, we shall proceed to examine the effect of the selective absorption of Type I, the most transparent, and Type III, the least transparent of the ocean types, upon the effective detector sensitivity.

From the transmission characteristics of Types I, II, and III ocean water (Jerlov, 1951) (table 9) together with the spectral distribution of visible energy for air-mass-two conditions ( $E_{\lambda}$ ), according to Moon, 1940 (table 5),

Table 5--Solar irradiance at sea level for air mass between 387.5 and 712.5 m $\mu$ , and the percentage of total visible energy within 25-m $\mu$  intervals over this spectral region  
[From data of Moon, 1940]

Wavelength	Energy	Percentage of total visible energy
m $\mu$	$\frac{\text{g. cal./cm.}^2}{\text{min./25 m}\mu}$	$\frac{\text{g. cal./cm.}^2}{\text{min./25 m}\mu}$
387.5	0.533	-
400.0	<sup>1</sup> .658	3.64
412.5	.965	-
425.0	1.06	5.52
437.5	1.24	-
450.0	<sup>1</sup> 1.41	7.20
462.5	1.53	-
475.0	1.62	8.44
487.5	1.69	-
500.0	<sup>1</sup> 1.70	8.78
512.5	1.69	-
525.0	1.67	8.56
537.5	1.67	-
550.0	<sup>1</sup> 1.67	8.56
562.5	1.66	-
575.0	1.65	8.58
587.5	1.63	-
600.0	<sup>1</sup> 1.64	8.19
612.5	1.63	-
625.0	1.64	8.34
637.5	1.64	-
650.0	<sup>1</sup> 1.64	8.29
662.5	1.63	-
675.0	1.63	8.11
687.5	1.40	-
700.0	<sup>1</sup> 1.55	7.79
712.5	1.45	-

<sup>1</sup> Values of Moon (1940) given at 10-m $\mu$  intervals. Values in table not followed by a footnote were obtained by graphical interpolation between 10-m $\mu$  intervals.

the percentage of downwelling visible radiant energy has been computed at six depths by integrating the data for the 11 wavelengths (see table 10) for Types I, II, and III water and intermediate types. The wavelength specificity and half-band width of the irradiance meter under conditions of Type I, II, and III may be readily computed by multiplying, wavelength-by-wavelength, the sensitivity of the detector unit by the relative spectral energy distribution at any depth. These calculations reveal that in Type I ocean water at 100-m. depth, the maximum sensitivity is at 475 m $\mu$ , and the half-band width is of the order of 50 m $\mu$ . At 50-m. depth in Type III water, the peak remains at 475 m $\mu$ , but the half-band width is

somewhat broader--about 70 m $\mu$ . The difference between the spectral sensitivity and half-band width of the detector in air and immersed in water is a natural consequence of a system in which one of the component filters, sea water, is in effect continually changing its spectral transmission characteristics. In the calculation of the percentage of downwelling visible irradiance remaining at various depths (optical depth), the half-band width of the detector remains nearly constant and has a peak sensitivity at 475 m $\mu$ . Thus all attenuation coefficients derived from measurements using the above equipment and filters have been assigned to 475 m $\mu$ .

### Observational Technique and Computations

Measurements of submarine irradiance were always made between 1000 and 1430 hours local time and most frequently between 1230 and 1400 hours. The measurements were made in the following manner:

1. The ship was oriented to reduce the likelihood of interference from the ship's shadow.

2. The irradiance meter was lowered with a bathythermograph that had a 150-m. depth range (or, occasionally, with an electronic depth-sensing element) attached, until the output of the submerged cell was about 10  $\mu$ a. The instrument was then raised or lowered to the nearest 10-m. mark on the photometer cable.

3. After the meter reached equilibrium, at least one and usually three sets of simultaneous readings of the output of the deck and submerged cells were made, one observer reading one meter only. On days of variable cloud cover, it often took as long as 10 minutes to obtain three sets of readings at a given depth.

4. The wire angle was measured with a hand inclinometer by visually superimposing the edge of a conventional wire-angle indicator with the conducting cable and reading the angle. The meter was then raised to the next 10- or 20-m. mark. The outputs of the cells were again read (see 3 above). This procedure was repeated until the photometer was at a depth of 5 or 10 m., or until the output of the submerged cell exceeded 1,000  $\mu$ a.

5. The output of the detector was then corrected for departure from linearity of response. These values have been corrected again to an arbitrary but constant deck cell reading to allow for changes in incident radiation. The irradiance meter depths were computed from the wire angle measurements in combination with the maximum depth recorded by the bathythermograph, or by using the depth indicated by the depth-sensing element.



The diffuse attenuation coefficient of downwelling irradiance was computed from the corrected data with the following formula:

$$k = \frac{\log_e H_{\lambda Z_1} - \log_e H_{\lambda Z_2}}{Z_2 - Z_1}$$

where H is the corrected output ( $\mu a.$ ) of the submerged cell at depths  $Z_1$  and  $Z_2$  (in meters); lambda ( $\lambda$ ) refers to the spectral sensitivity of the immersed detector, 475 m $\mu$  (except for SCOPE Expedition station 9C).

### Errors in Determination of k Values

The errors in determination of a given k value arise from two sources: depth-measurement errors and the irradiance-level measurement errors at two depths.

Some indication of the error in measurements of irradiance level may be seen in the data obtained in two series of experiments in which the depth-measurement error was believed to be negligible. The first series was carried out in Lake Pend Oreille, Idaho, under ideal conditions, namely, a stable platform, absence of high waves, and vertical homogeneity (Tyler, Richardson, and Holmes, 1959). The depth-measurement errors were extremely low--probably of the order of  $\pm 8$  to 10 cm, at the maximum depth (17 m.), and much less nearer the surface. The second experiment was carried out at sea in the California Current some 20 miles west of Oceanside, Calif. Wire angles were less than 5 $^{\circ}$  during these latter measurements, and the sun remained unobscured by clouds. The data were

examined statistically by means of an analysis of variance by ranks (Tate and Clelland, 1957).

The Pend Oreille data on attenuation coefficients obtained between 3 and 17 m. (see table 6), according to the analysis of variance by ranks, reveal that the values obtained on different days and at different depth intervals were not significantly different (table 6). The data were also analyzed by every other depth interval (4-5, 7-9, 11-13, and 15-17 m.) so that the same irradiance values would not enter into the k calculations more than once: the results were similar to that obtained above. From these analyses, I conclude the Pend Oreille data were drawn from the same population and that an appropriate measure of the error is the standard error. The mean k value for the data is 0.1805; the standard deviation(s) equals 0.0189; and the coefficient of variation, 10.5 percent.

The data on attenuation coefficient in the California Current (table 7) were analyzed in the same manner as the Pend Oreille data but gave somewhat different results. The probability for the H value indicates that the k values were not drawn from the same population with respect to depth. This finding was not surprising, since the phytoplankton standing crop appeared moderately high, judging from the color of the water, and any nonuniform vertical distribution of the phytoplankton would be reflected in variability of k. On the other hand, the k values obtained from the different lowerings at the same depth showed less variability than would be expected (table 7). The cause of this stability is not understood, and the data may be suspect. Nonetheless, the coefficients of variation have been calculated for each of the depth intervals to gain a rough

Table 6.--Attenuation coefficients (k) per meter observed in Lake Pend Oreille, Idaho, 15 to 20 March, 1957  
[H = 5.5; p < 0.75 > 0.50]

Date	Pacific standard time	Sky condition	Depth interval (in meters)							
			3-4	4-5	5-7	7-9	9-11	11-13	13-15	15-17
<u>March</u>										
15	1413-1446	Overcast	0.1834	0.1850	0.1733	0.1764	0.1786	0.1972	0.1807	0.1828
16	0930-1022	Overcast	.1994	.1618	.2174	.1882	.1862	.1788	.1878	.1900
16	1443-1517	Thin overcast	.1801	.2117	.1506	.1919	.1748	.1750	.1891	.1910
18	0833-0914	Overcast	.1665	.1677	.1812	.1948	.1757	.1915	.1872	.1646
18	1357-1456	Overcast	.2125	.1956	.1910	.1920	.1768	.1812	.1906	.1667
19	0932-1017	Overcast	<sup>1</sup> .1861	<sup>1</sup> .1861	.1878	.1720	.1902	.1787	.1804	.1842
19	1050-1138	High scattered clouds	.2029	.1626	.1643	.1815	.2094	.1745	.1926	.1572
19	1325-1355	High scattered clouds	.1956	.1814	.1318	.1872	.1872	.1954	.1994	.1733
20	0908-1009	Overcast	.1579	.2036	.1704	.1893	.1840	.1986	.1504	.1893
Sum of ranks			379.5	380.0	241.0	378.0	324.0	355.5	369.0	248.0

<sup>1</sup> k was determined between 3 and 5 m.

Table 7.--Attenuation coefficients from photometer lowerings at a station 20 nautical miles west of Oceanside, Calif., 1 May 1959  
 $[H = 0.18; p < 0.975 > 0.99]$

Depth	Lowerings				Sum of ranks
	No. 1	No. 2	No. 3	No. 4	
<u>Meters</u>					
6-11	0.0505	0.0495	0.0460	0.0453	11
11-21	0.0596	.0588	.0555	.0512	30 (H = 12.5)
21-31	0.0502	.0699	.0685	.0722	37 (p < 0.01)
31-41	.1033	.0870	.0914	.0771	58
Total sum of ranks	34	36	34	32	136

idea of the variability encountered. The values obtained (depth intervals in meters in parentheses) were: 0.05 (6-11), 0.07 (11-21), 0.15 (21-31), and 0.11 (31-41).

It appears then that under excellent observational conditions the coefficient of variation is of the order of 10 percent. I estimate that measurements of comparable quality to those discussed above are obtained only 30 to 50 percent of the time in the northeastern tropical Pacific. The remainder of the time, rapid fluctuations in incident radiation resulting from nonuniform cloud cover and fluctuations in underwater irradiance resulting from vessel surge and from the focusing effects of surface waves tend to give a greater scatter to replicate irradiance measurements at a given "depth." These factors reduce the precision and accuracy of the estimates of attenuation coefficients.

Variations in irradiance level caused by vessel surge and focusing effects of surface waves cannot be detected with the equipment used below depths where about 25 percent of the incident energy is found. Although changes in incident radiation are still troublesome below this depth, the largest error in estimating the attenuation coefficient at depths below 30 m. probably results from errors of depth measurement.

I must emphasize that the Pend Oreille data provide error estimates which are minimal. Under conditions of varying cloud cover accompanied by pronounced vessel surge or high wire angles, observations are certainly not as reproducible as those given above. Data from both experiments yield estimates of precision which probably cannot be surpassed with the equipment described.

The analysis of k data thus far has assumed negligible error in depth determination. Unless one has a depth-sensor attached to the irradiance meter, depth is usually computed from wire length and departures in wire angle from the vertical. Computation of instrument depth with angles in excess of 20° and with wire lengths greater than 40 to 50 m. is rather unsatisfactory, although the data available are

not abundant enough to permit a rigorous assessment of this error.

To illustrate the problem, data from two SCOT Expedition stations are considered. These stations were chosen because wire angles were large and wire lengths were as great as 100 m. In these two examples, the depth indicated by the depth sensor is taken to be the true or "observed" instrument depth. Two different but related questions need to be considered. First, how accurately may one compute instrument depth; and secondly, how well do the computed depth intervals agree with the observed intervals? The answer to the first question tells us if we are assigning our k value to the appropriate depth interval, and the answer to the second tells us how much we may expect the k value to vary from the true value.

The computed depth is always equal to or less than the observed depth. This relation results from the fact that the computational method assumes the wire to be straight, whereas it is almost certainly curving.

In Table 8, column 6, the difference between the observed depth and the computed depth is given for the two SCOT stations. The values show that the instrument was often 2 to 5 m. and occasionally 9 to 10 m. below the computed depth. This difference means that the k values will frequently be assigned to incorrect depth intervals. It is reasonable to believe that differences between true and computed depth generally increase with wire length, but the few available observations fail to show any such systemic variation.

Aside from the question of absolute depth, the variation between the observed and computed depth interval also needs to be considered, for it is the depth interval that enters directly into the k calculations. Table 8, column 9, gives the difference between the observed and computed intervals. If we assume that these data are representative of conditions of high wire angles, then errors as great as 3 m. in 14 will give k values that are 22 percent too low. The median value lies between 1 and 2 m. difference, and may be typical.

It should be pointed out that this assessment of errors must be considered preliminary, although there is no reason to suspect the data. Thus these values are probably typical for the equipment used.

### Optical Depth Determinations

In addition to the rate of attenuation of diffuse downwelling irradiance (i.e., the diffuse attenuation coefficient) another related quantity, optical depth, is frequently used by investigators concerned with estimates and measurements of primary production. These investigators frequently wish to determine the

Table 8.--Estimated errors of depth measurement associated with measurement of attenuation coefficient at two SCOT Expedition stations in the eastern tropical Pacific, April 1958

[Station 6: lat. 21°13' N., long. 117°23' W. Station 8: lat. 18°41' N., long. 114°53' W.]

Station	Wire length	Wire angle	Depth			Depth interval		
			Computed	Observed	Difference	Computed	Observed	Difference
	Meters	Degrees	Meters	Meters	Meters	Meters	Meters	Meters
6	20	25	18	20	2	-	-	-
	40	30	35	37	2	17	17	0
	60	40	46	51	5	11	14	3
	80	40	61	70	9	15	19	4
	100	40	77	88	10	16	18	2
8	10	10	10	-	-	-	-	-
	20	20	19	20	1	9	-	-
	40	30	35	37	2	16	17	1
	60	35	49	52	3	14	15	1
	80	40	61	66	5	12	14	2
	100	45	71	76	5	10	10	0

Table 9.--The transmission (percentage T/m.) of different sea water types at 25-m $\mu$  intervals in the visible region of spectrum

[Transmission characteristics of Type I, II, and III from Jerlov (1951).

Intermediate water types obtained by interpolation from Jerlov Types I, II, and III]

Water type	Wavelength (m $\mu$ )										
	400	425	450	475	500	525	550	575	600	625	650
	----- Percent -----										
I	97.2	97.8	98.1	98.2	97.2	96.1	94.2	92.0	85.0	74.0	64.3
IA	96.3	97.1	97.4	97.5	96.6	95.5	93.6	91.2	84.2	73.5	<sup>1</sup> 64.1
IB	95.5	96.4	96.7	96.8	96.0	94.9	93.0	90.5	83.3	73.0	<sup>1</sup> 64.0
IC	94.6	95.6	96.0	96.1	95.4	94.3	92.4	89.8	82.5	72.5	<sup>1</sup> 63.8
ID	93.7	94.9	95.4	95.4	94.7	93.7	91.7	89.0	81.7	72.0	<sup>1</sup> 63.6
IE	92.9	94.2	94.7	94.7	94.1	93.1	91.1	88.2	80.8	71.5	<sup>1</sup> 63.5
II	92.0	93.5	94.0	94.0	93.5	92.5	90.5	87.5	80.0	71.0	<sup>1</sup> 63.3
IIA	90.4	92.2	92.9	93.0	92.6	91.7	89.7	86.5	79.0	70.4	<sup>1</sup> 62.9
IIB	88.8	90.9	91.8	92.0	91.7	90.9	88.9	85.5	78.0	69.8	<sup>1</sup> 62.4
IIC	87.2	89.6	90.7	91.0	90.8	90.1	88.1	84.5	77.0	69.2	<sup>1</sup> 61.8
IID	85.6	88.3	89.6	90.0	89.9	89.3	87.3	83.5	76.0	68.6	<sup>1</sup> 61.3
III	84.0	87.0	88.5	89.0	89.0	88.5	86.5	82.5	75.0	68.0	<sup>1</sup> 60.7

<sup>1</sup> These values are somewhat uncertain; they are based on an empirical extrapolation of the Jerlov (1951) data.



depth at which 1 percent of the visible radiation is found. A simple method for estimating this depth, based on measurements of blue-green downwelling irradiance, is presented below.

An examination of the transmission characteristics of Type I, II, and III ocean waters (Jerlov, 1951; see also table 9) reveals that it is possible to define the type of ocean water on the basis of a single diffuse attenuation coefficient, providing wavelength specificity may be assigned to the coefficient. Furthermore, after the water type has been identified, it becomes possible to determine the percentage of surface radiation at any depth in the water column provided the attenuation coefficient remains constant with depth (Johnson and Kullenberg, 1946). Jerlov (1951) presented such an analysis in graphic and tabular form based on attenuation coefficients obtained at 25-m $\mu$  intervals. The percentage of total incident solar energy was also given as a function of depth by Jerlov (1951) for the three ocean water types.

In the present study, the Jerlov data (1951) have been used, but the treatment differs from that of Jerlov in two respects. First, the percentage of visible (rather than total) downwelling energy at various depths has been computed, by use of the incident energy distribution of visible energy given by Moon (1940) for air-mass-2 conditions (table 5). Secondly, a family of curves has been constructed to yield five subtypes between Jerlov Type I and II, and four subtypes between Type II and III ocean water. As will be seen, these additional subdivisions make the original classification somewhat more useful.

The calculations carried out to derive this family of curves are similar in principle to those used by Jenkin (1937). The transmission

values given by Jerlov, 1951, (see table 9) for ocean water Types I, II, and III were interpolated to yield five intermediate water types between Type I and II, and four types between Type II and III; a linear interpolation was used at each 25-m $\mu$  interval between 400 and 675 m $\mu$ . The percentages of radiation remaining at 5, 10, 25, 50, 75, and 100 m. were computed for each of these water types from the interpolated transmission values, on the assumption of homogeneous water and constant energy between 400 and 675 m $\mu$ . This percentage was in turn multiplied by the percentage of energy contained in each 25 m $\mu$  between 375 and 700 m $\mu$ , as derived from the air-mass-2 data of Moon, 1940 (see table 5). The percentage remaining at each depth was then obtained by summing the percentages at each wave-length interval. No corrections were made for losses at the surface. The results of these calculations are given in table 10 and presented graphically in figure 3.

To determine the percentage of downwelling visible irradiance remaining at depth, it is first necessary to be able to specify the in situ spectral sensitivity of the detector. A k value derived from two or more irradiance measurements may then be assigned to an appropriate peak sensitivity and band width. On the basis of this information, a water type (in the Jerlov sense) may then be selected for which the transmission characteristics are approximately known and the percentage of downwelling visible irradiance obtained as a function of depth. In table 10 the k values at 475 m $\mu$  (this is approximately the sensitivity of the equipment used) in Types I, II, and III, and the intermediate types, are given. A somewhat more useful graphic presentation of this information is given in figure 3.

Table 10.--Percentage of incident visible downwelling irradiance computed at selected depths in ocean water types for a zenith sun possessing the spectral distribution of air mass 2 and the k value at 475 m $\mu$

Water type	k at 475 m $\mu$	Surface	Depth in meters					
			5	10	25	50	75	100
-----Percent-----								
I	0.0182	100	57.1	43.2	25.4	12.7	6.9	3.9
IA	.0253	100	55.1	40.4	21.3	8.9	4.2	1.9
IB	.0325	100	53.2	37.5	17.9	6.3	2.4	0.97
IC	.0398	100	51.3	34.9	14.9	4.4	1.4	.46
ID	.0471	100	49.6	32.4	12.5	3.1	0.82	.23
IE	.0544	100	47.8	30.2	10.4	2.15	.47	.10
II	.0618	100	46.9	28.0	8.7	1.5	.28	.043
IIA	.0725	100	43.7	25.1	6.6	0.86	.12	-
IIB	.0833	100	41.4	22.5	5.0	.50	.04	-
IIC	.0943	100	39.1	20.1	3.8	.32	-	-
IID	.1054	100	37.0	18.0	2.9	.17	-	-
III	.1109	100	35.0	16.1	2.2	< .01	-	-

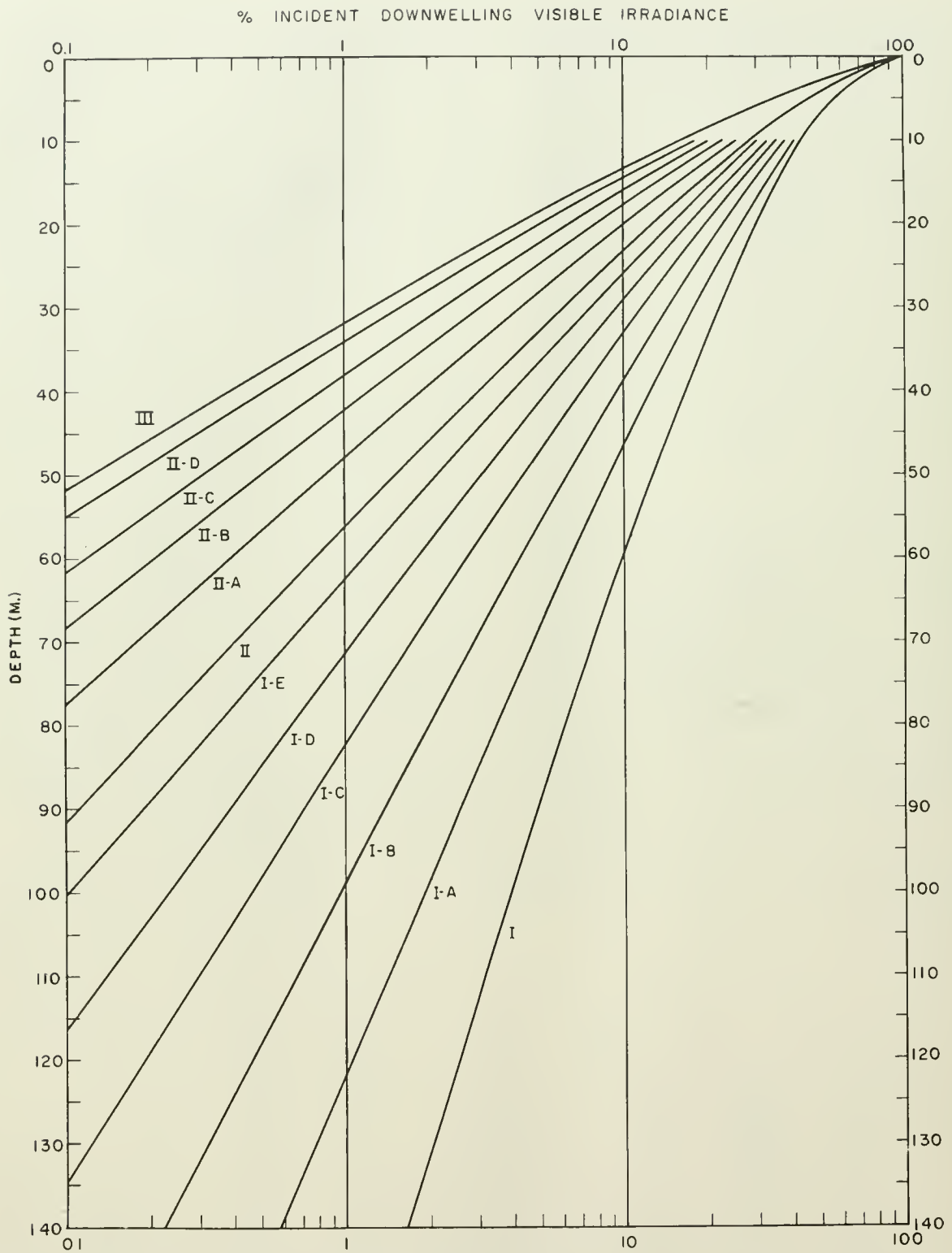


Figure 3.--The percentage of incident visible downwelling irradiance as a function of depth in various oceanic water types for a zenith sun possessing the spectral energy distribution for air-mass-2 conditions.



A rough comparison may be made between these results and those obtained by Jerlov (1951) for Type I, II, and III by assuming that 45 percent of the total incident energy lies in the visible region (Strickland, 1958; Withrow and Withrow, 1956). Agreement for Type II and III is good; it is poorer for Type I.

## CHLOROPHYLL A

In 1957 when the present investigation was initiated, the pigment analysis method of Richards with Thompson (1952) was chosen. This method is a selective and useful ship-board technique. Although the determination of chlorophyll a, b, and c and astacin and non-astacin carotenoids is possible with the method, only chlorophyll a concentrations have been calculated and employed in this study.

Chlorophyll a was selected for several reasons:

(1) It is well known that radiant energy absorbed by chlorophyll c, certain phycobilins, and xanthophylls is transferred to chlorophyll a. Thus it seems likely that radiant energy absorbed flows to the chemical stage of photosynthesis via chlorophyll a. In the pigment complex of the living cell, chlorophyll a concentration might well limit the rate of photosynthesis and also serve as a rough index of plant biomass.

(2) Of all the pigments capable of being determined by the pigment-analysis method of Richards with Thompson, chlorophyll a is subject to the least error (Richards with Thompson, 1952).

(3) The likelihood of interference by phycobillin-type pigments in the open sea is not great, owing to the scarcity of plants bearing these pigments.

Although the validity of the Richards with Thompson (1952) equations have been questioned by Humphrey (1962) and Parsons and Strickland (1963), no set of revised equations has appeared in the literature applicable to dried chlorophyll preparations. Parsons and Strickland pointed out that dried chlorophyll preparations yield equation coefficients which are lower than those obtained with wet preparations and suggested that the Richards with Thompson equations may yield values with dried extracts which are 10 percent too low. In view of the lack of a recent rigorous reappraisal of the equations for dried extracts, however, it seems unnecessary at this time to adopt the 10-percent correction factor suggested by Parsons and Strickland (1963).

These equations yield a quantity called "chlorophyll a"; however, inactive chlorophyll a derivatives (phaeophytin a and others) will be determined as chlorophyll a with this technique. A discussion of this problem appears later in the text.

## Sample Collection

Surface water samples used for the determination of chlorophyll a were collected either with a plastic bucket or with a plastic 3-liter Van Dorn (1956) sampler; subsurface samples were obtained with 3-liter Van Dorn bottles.

In offshore tropical and subtropical waters, two sampler casts were made routinely at each station. Four to 5 liters of water are required to obtain acetone-chlorophyll extracts with sufficient optical density for analysis, namely, a corrected extinction at 665  $m\mu$  of 0.10 or more, with a 5.5- to 6.0-ml. extract volume in a 10-cm. semimicro absorption cell (capacity 5.5 ml.). Thus two separate sampler casts were routinely made at each station; 0.5 to 0.7 liter of water was used in studies of photosynthesis and about 0.25 liter was preserved and fixed for subsequent analysis of species composition and abundance of phytoplankton. The remaining volume of water (about 2.0 liters) was placed in an 8-liter polyethylene bottle and combined with the 3 liters obtained at the same depth (based on wire length and without correction for wire angle) from the second cast.

On two cruises (SCOPE Expedition and TO-58-1) the depth placement of the various samplers was often determined by taking into consideration either diffuse downwelling irradiance or thermal structure or both. During the other three cruises (TO-58-2, TO-59-1, and TO-59-2) the sampler placement was standardized to include the following depths: surface, 10, 25, 50, 75, 100, and 125 or 150 m., and occasionally 200 m.

Except at a very few stations, the results used here were determined from samples collected during the hour preceding local apparent noon.

## Sample Treatment

Before filtration, the volume of each sample was estimated to the nearest 0.1 liter. About 20 to 25 mg. of powdered magnesium carbonate were added to each sample before filtration during SCOPE and SCOT Expeditions; on subsequent cruises magnesium carbonate was not used.

Four, and occasionally eight, samples could be filtered simultaneously with the equipment available; each sample required about 2 hours to filter. Thus the maximum elapsed time between sample collection and complete filtration of the last sample (normally the deepest) rarely exceeded 5 hours. In waters where the standing crop of phytoplankton was relatively high, the sample volume was reduced to 2 or 3 liters, and occasionally the sample was split and two filters were used to reduce filtration time. Owing to the higher density of particulate matter, total filtration time for these samples remained similar to that given above.

Plain white, 47-mm., Type HA Millipore filters (Millipore Filter Corp., Bedford, Mass.) were used for the filtration of chlorophyll samples. Filtration was generally carried out in the laboratory or some other shady, protected place.

The vacuum at the pump was not monitored routinely. On TEMPO cruise, when a vacuum gauge was placed on the vacuum pump, the negative pressure generally ranged from 18 to 25 lbs/in.<sup>2</sup> These values are probably typical of the negative pressures, since the same vacuum pumps have been used throughout the investigation.

### Pigment Extraction and Determination of Chlorophyll a Concentration

Following filtration, the filter edges were trimmed, and the filters were placed in perforated numbered plastic petri dishes in a darkened vacuum desiccator with activated silica gel for at least 24 hours. The dried material on the filter was then extracted in glass-stoppered centrifuge tubes with 3 ml. of 90-percent glass-redistilled acetone for 18 to 24 hours in the dark and cold (ca. 8° C.). The extract was centrifuged for 20 to 30 minutes at about 3,000 r.p.m.; the supernatant was decanted into a glass-stoppered 10-ml. graduated cylinder and placed in the dark. The residue remaining in the centrifuge tube was shaken up in an additional 2 ml. of 90-percent acetone and recentrifuged for 20 to 30 minutes. This supernatant was combined with that previously obtained; if no turbidity was visible in the combined extract, it was diluted with 90-percent acetone to 5.5 or 6 ml. If turbidity was apparent, the extract was recentrifuged until clear.

The spectrophotometric determinations were carried out, by the Richards with Thompson (1952) technique. Semimicro absorption cells of 10 cm. with a volume of about 5.5 ml. were substituted for the 1-cm. cells used by Richards with Thompson. Percentage transmission (against a 90-percent acetone blank), converted to extinction, was measured at 750, 665, 645, and 630 mμ with a modified Model DU Beckman Spectrophotometer (Holmes and Linn, 1961), at a slit width of 0.04 mm. The extinction at 750 mμ was subtracted from that observed at the other wavelengths, and the calculations of chlorophyll a concentration (milligrams per cubic meter) were carried out by the Richards with Thompson formula:

$$\frac{(15.6E_{665} - 2.0E_{645} - 0.8E_{630}) \times \text{vol. extract (ml.)}}{\text{vol. sea-water (l.)} \times \text{absorption cell path length (cm.)}}$$

### Wavelength and Blank Corrections

The wavelength calibration on the spectrophotometer was checked periodically on each expedition or cruise by using the chlorophyll a peak (Richards with Thompson, 1952) as the reference. Occasionally the peak read 2.5 mμ high or low. The wavelength dial reading was shifted accordingly for all wavelengths.

Regardless of the apparent visual clarity of the acetone extracts, there was always some residual extinction at 750 mμ which could not be attributed to chlorophyll a, b, or c absorption. In more than 95 percent of the chlorophyll determinations made, this extinction value fell between 0.03 and 0.08 (about 82-93 percent transmission) for the 10-cm. path length. No apparent relation existed between this value and chlorophyll a concentration, and the extinctions were highest with chlorophyll a concentrations of less than 0.03 mg. per cubic meter. This "blank" value was subtracted from the extinction at the above-mentioned wavelengths under the assumption that the value remains essentially constant over the wavelength range (i.e., 750 - 630 mμ) used in the determinations. This turbidity presumably results from one or more of the following: (1) an insoluble fraction of the filter; (2) the magnesium carbonate; (3) sea salt retained in the filter; (4) material contained in the sample itself.

The possible effects of (1) and (2) above were examined as follows: a prefiltered seawater sample of 400 ml. was divided into four equal aliquots: three containing magnesium carbonate (Y-1, Y-2, Y-3) and one without (Y-4). These samples were filtered through an HA Millipore filter, the filters were dried, the samples extracted in 90-percent acetone, and transmission of the extract measured. The extinction data are presented in table 11. The fact that the extinctions were generally lower than those obtained at sea indicates that some of the absorption at 750 mμ must be due to material contained in the sea water itself. Although the extinction in these

Table 11.--Optical extinction values per 10 cm. of 90 percent acetone extracts of HA Millipore filters, with and without magnesium carbonate, 1958  
[Filters pretreated by passing through 100 ml. of fresh prefiltered sea water]

Sample No.	Magnesium carbonate	Wavelength			
		750 mμ	665 mμ	645 mμ	630 mμ
Y-1	Present	0.0300	0.0315	0.0340	0.0335
Y-2	Present	.0285	.0350	.0375	.0415
Y-3	Present	.0170	.0205	.0225	.0245
Y-4	Absent	.0210	.0300	.0360	.0395



samples at the four wavelengths was not constant, the departure from linearity is not sufficiently great to introduce a large error in the chlorophyll a concentration value. Any nonlinear absorption due to the sample material itself has not, of course, been taken into consideration.

Additional evidence is now available which shows that the sea salt retained on the filter is the major cause of the high blank values at 750 m $\mu$  (J. D. H. Strickland, personal communication).

#### Accuracy and Precision of Chlorophyll a Determinations and Sampling Errors

Richards with Thompson (1952) gave an estimate of total error of 9 percent for chlorophyll a. This estimate was obtained with identical samples and presumably reflects the reproducibility of the method under ideal laboratory conditions when quantities of chlorophyll are large. Humphrey (1960), who analyzed the chlorophyll a concentration in six aliquots of a well-mixed surface sea water sample, found the standard deviation (0.053 mg. per cubic meter) to equal about 6.6 percent of the mean value (mean 0.81 mg. per cubic meter) of the six determinations.

In 1956 and 1957 a number of simple field studies were carried out which gave a rough estimate of the variability in chlorophyll a in repeated sampling. The results have been

tabulated in table 12 together with the mean value, standard deviation, and coefficient of variation for each series at each sample depth. Unfortunately, the interpretation of these data is not as straightforward as I might wish. The difficulty arises from the fact that not all of the data in each series could be collected simultaneously, with the result that the effect of the diurnal rhythm of chlorophyll a (Doty and Oguri, 1957; Holmes and Haxo in Holmes et al., 1958; McAllister, 1963; Shimada, 1958; Yentsch and Ryther, 1957) almost certainly introduces another source of variability distinct from that associated with spatial heterogeneity. Furthermore, in the statistical analysis I have considered the samples to have been collected at random and the values obtained at a given depth to be normally distributed.

Of the data presented in table 12, series B and C best reflect the variability encountered in synoptic sampling and yield coefficients of variation of 14 and 27 percent, respectively. The remaining surface sample series (A, D, and E) yield values below 27 percent even though the sampling series required several hours for completion. A similar analysis of Shimada's (1958) data, which covered a time interval of 44 hours, gives a coefficient of variation of 16.5 percent (mean 0.112; standard deviation 0.0184) in spite of the clear evidence of diurnal periodicity. Thus at the

Table 12.--Variability in field determinations of chlorophyll a, eastern tropical Pacific, 1957

Series <sup>1</sup>	Depth	Sample number									Mean ( $\bar{x}$ )	Standard deviation (s)	Coefficient of variation (s/x)
		1	2	3	4	5	6	7	8	9			
	<u>M.</u>	<u>Mg. chlorophyll a per m.<sup>3</sup></u>											<u>Percent</u>
A.....	0	0.42	0.43	0.36	0.40	0.40	0.33	0.40	0.45	0.46	0.406	0.0353	8.7
B.....	0	.086	.094	.12	.12	.11	.12	-	-	-	.108	.0148	13.7
C.....	1	.18	.18	.16	.13	.20	.28	-	-	-	.188	.0509	27.0
D.....	0	.14	.16	.081	.14	.14	-	-	-	-	.132	.03163	23.9
D.....	20	.18	.20	.28	.13	.13	-	-	-	-	.184	.03616	19.7
D.....	40	.51	.50	.53	.76	.85	-	-	-	-	.630	.04	6.35
E.....	0	.19	.13	.13	.12	.14	-	-	-	-	.142	.0278	19.6
E.....	10	.39	.12	.14	.13	.12	-	-	-	-	.180	.1177	69.2
E.....	20	.79	-	.56	.37	.20	-	-	-	-	.48	.2538	52.9
E.....	40	.45	.56	.35	.50	.74	-	-	-	-	.52	.145	27.9

<sup>1</sup> Series A taken by SCOPE, 22 November 1956: 0915-1202, lat. 9°25' N., long. 89°31' W., nine surface samples collected at and around a surface parachute drogue at 4.8-km. intervals (Holmes, 1958).

Series B taken by Orca, 28 February 1957: 1145, lat. 33° 07.5' N., long. 117° 50' W.; surface samples collected in rapid succession with a plastic bucket at six locations around vessel.

Series C taken by Orca, 26 February 1957: 0848, lat. 32° 39.3' N., long. 117° 12.7' W.; six samples taken at 1-m. depth in rapid succession from the same position on the ship--inshore samples.

Series D taken by Orca, 27 February 1957: 0254-0700, lat. 33° 6.5' N., long. 118° 10.5' W. to lat. 33° 05' N., long. 118° 08.3' W. Samples at 0, 20, and 40 m. collected at five stations at the following times: 0254, 0331, 0440, 0550, and 0700. Ship drifting during entire sampling period.

Series E taken by Orca, 27 February 1957. Samples collected at four depths at about 6.4-km. intervals: E-1 at 1422, lat. 33° 10.4' N., long. 117° 28.4' W.; E-2 at 1518, lat. 33° 10.2' N., long. 117° 32.2' W.; E-3 at 1551, lat. 33° 09.8' N., long. 117° 37' W.; E-4 at 1900, lat. 33° 09.7' N., long. 117° 41.8' W.; E-5 at 1950, lat. 33° 09.3' N., long. 117° 46.5' W.

All times are Pacific standard time.

surface in temperate and tropical areas, the coefficient of variation does not exceed 27 percent and most frequently is much less.

The coefficient of variation for samples collected at depth is greater, except for 40-m. samples in the D series, than that among the surface samples. As a similar trend is apparent in the data of Humphrey (1960), it seems likely that sampling at depth presents problems somewhat different from surface sampling.

The cause of this apparently greater variability in chlorophyll a with depth is not understood completely. Evidence that this increase arises from the failure of the sampler to sample in exactly the same stratum on repeated lowerings is indirect but merits consideration.

The hypothesis that alpha ( $\alpha$ ), the beam transmission coefficient, is related to chlorophyll a in offshore tropical waters, was tested on cruise TO-61-1. In all, some 49 measurements were made at 10 stations at local apparent noon with the beam transmission meter detector covered with a Wratten 45 filter (see table 4). A similar number of observations were made at midnight, with a Wratten No. 61 filter (peak transmission at 520 m $\mu$ ). A nonparametric statistical analysis (Spearman rank correlation coefficient--Siegel, 1956) showed that chlorophyll a and  $\alpha$  were positively correlated at better than the 0.01 level in the upper 20 m. in both sets of data. Below 20 m. and down to 46 m., the maximum depth reached by the  $\alpha$  meter, the observations were not correlated. The seemingly logical assumption, that a causal relation exists between these two variables, suggests that a profile of  $\alpha$  at closely spaced intervals of depth should give a good indication of the vertical distribution of chlorophyll a, at least in the upper 20 m.

Two such detailed  $\alpha$  profiles were obtained at stations 24 and 31 (see fig. 4). At both of these stations  $\alpha$  and chlorophyll a were positively correlated (significance levels: <0.01 and <0.05, respectively). The similarity in trend between chlorophyll a and  $\alpha$  is apparent in both of these sets of data. The major feature to consider in these figures is the manner in which  $\alpha$  can vary between the chlorophyll sampling depths below 10 to 12 m. These variations indicate the existence of considerable gradients in light absorbing and scattering materials which are probably chlorophyll a-bearing. It is quite evident that failure to repeat a sampling depth within 1 or 2 m. may lead to differences in concentration of 50 percent or more between "duplicate" samples. Such sampling faults could easily contribute to an increase in the standard errors of replicate samples taken at depth. The standard error could be further increased by internal waves, patchiness, and diurnal rhythms during any extended sampling series.

## Interpretation of Results

Although an estimate of the total error in field measurements of chlorophyll a is given in the previous section, questions remain as to the exact nature of the entity called chlorophyll a derived from the measurement, and the success of the extraction itself.

Photosynthetically inactive chlorophyll a derivatives in sea water could, if present in appreciable quantities, affect the interpretation of the chlorophyll a determination. Chlorophyllide a will always interfere because it has the same absorption spectrum and specific extinction coefficients as chlorophyll a (Patterson and Parsons, 1963). Fortunately it does not appear to be common (see below). The two most frequently mentioned compounds of this nature are phaeophytin a and phaeophorbide a. The possible interference of these two pigments with the chlorophyll a measurement can, however, be postulated on the basis of their similar absorption spectra, even though relevant data are for different solvents or different solvent concentrations. Vernon (1960) examined chlorophyll a and phaeophytin a absorption spectra in 80-percent acetone and the red chlorophyll a peak in 90-percent acetone. In 90-percent acetone the peak is 664 m $\mu$ . In 80-percent acetone it is 665 m $\mu$ , whereas the peak for phaeophytin a is 666 to 667 m $\mu$ . If we assume a similar shift in 90-percent acetone, then the two peaks would be separated by only 1 or 2 m $\mu$ . Such a difference cannot be resolved with a DU Spectrophotometer used under field conditions. A similar but greater shift (about 5 m $\mu$ ) toward the red has also been reported by Zscheile and Comar (in Smith and Benitez, 1955, p. 148) in ethyl ether. Thus we must conclude that phaeophytin a can interfere with the determination of chlorophyll a if present in sea water extracts.

Patterson and Parsons (1963) investigated the occurrence of chlorophyllide a, phaeophytin a, and phaeophorbide a in Departure Bay, British Columbia, water samples, in four cultures of different marine species, in littoral mud, and in a net tow in which zooplankton predominated. These authors concluded that most of the water samples and cultures tested, except for *Skeletonema costatum*, contained insufficient amounts of the phaeo-pigments to cause errors in the Richards with Thompson (1952) chlorophyll a method.

Yentsch and Menzel (1963) used a differential fluorescence method to estimate the phaeo-pigments by measuring the change in chlorophyll a fluorescence before and after acidification of the acetone extract. Their results (table 4) on natural populations are limited but show that phaeo-pigments are present in the upper 75 m. in the tropical Sargasso Sea in appreciable amounts. Below a

CHLOROPHYLL (MG./M.<sup>3</sup>)

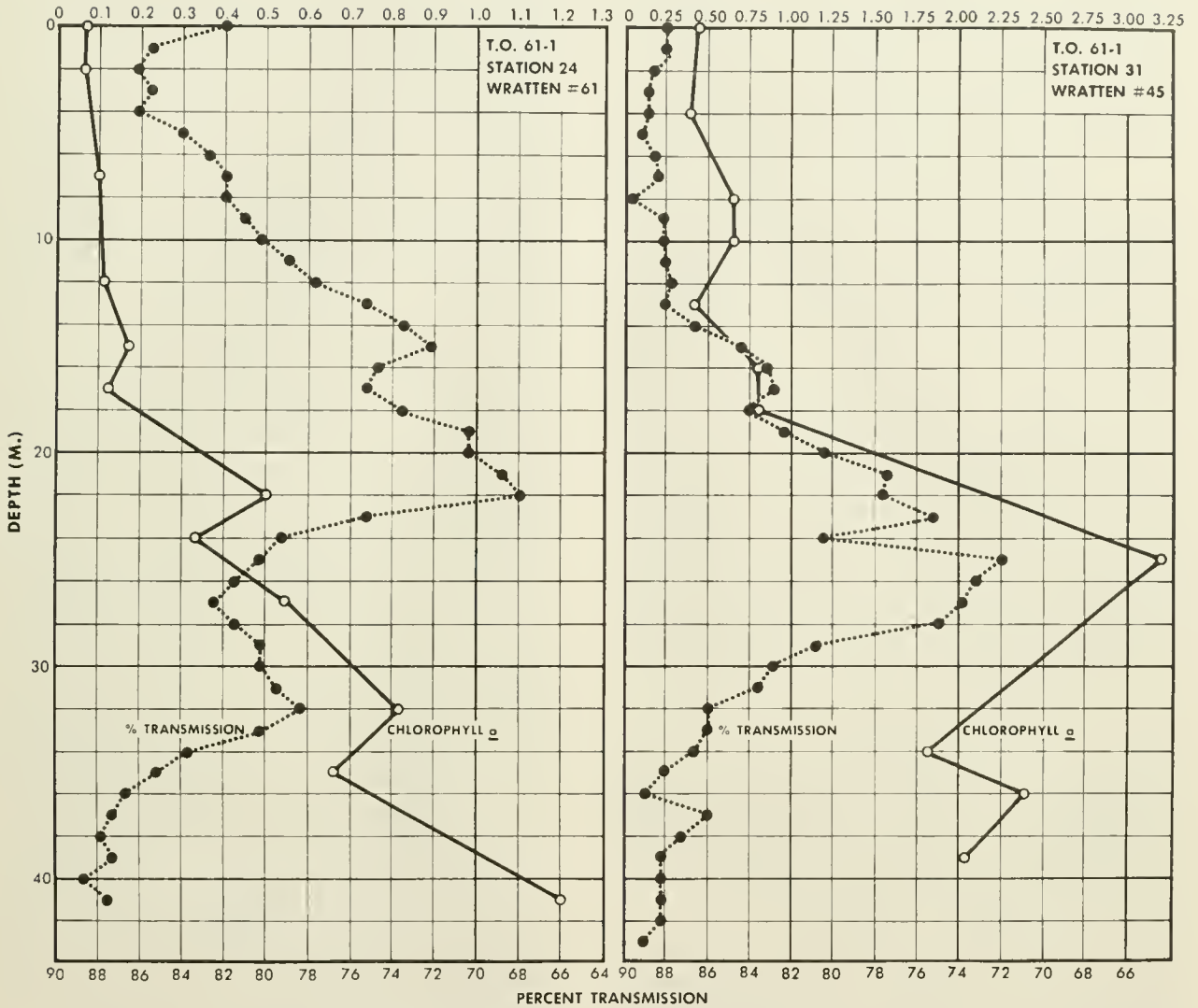


Figure 4.--The vertical distribution of beam transmittance and chlorophyll a in the upper 40 to 50 m, as observed at two stations in the northeastern tropical Pacific.

depth of 100 m. almost all of the chlorophyll fluorescence may be attributed to phaeopigments. In local waters (i.e., Woods Hole, Mass.) Yentsch and Menzel (1963) also observed appreciable quantities of phaeopigments.

The spectrophotometric and fluorometric techniques yield somewhat divergent results. This discrepancy may result from the difference in material analyzed or from differences in technique. The very different results with *Skeletonema costatum* in the papers by Patterson and Parsons (1963) and Yentsch and Menzel (1963) suggest that the two methods are not strictly comparable.

Jeffrey (1961) reported the presence of phaeophytin a in nature with samples containing large amounts of decomposing cells.

Quantitative data were not presented. Humphrey (1962) mentioned that Jeffrey has also observed chlorophyllide (from chlorophyll a) in the presence of decomposing cells. McAllister et al. (1961) failed to detect phaeophytin or phaeophorbide compounds in their plastic-bag experiment, although it is doubtful that their spectrophotometric method was adequate to detect phaeophytin a in small quantities.

Apparently the quantity determined as chlorophyll a by the Richards with Thompson (1952) technique includes inactive chlorophyll a derivatives when they are present. The amount of this inactive material in sea water appears to change with conditions and is probably highly variable in nature with respect to time, space, and depth. Since no data are available on the relative abundance of active



and inactive chlorophyll a in the northeastern tropical Pacific, caution must be used in the interpretation of the data, particularly with respect to production coefficients. In all probability, much of the rather wide range in production coefficients obtained may be partially explained by temporal and spatial variations in these chlorophyll a derivatives.

The degree of success of the extraction of pigments from the organisms on the filter appears somewhat variable. Almost invariably the residue of particulate matter in the bottom of the test tube after centrifugation appears to be pigmented. Reextraction and centrifugation removes only part of the additional pigmented material. Thus, some material on the filter is not successfully extracted, and the amount seems to vary from sample to sample. Since microscopic examination of the residue has not been made, it is not known what percentage is organic. Experience with a limited number of species in culture has shown that the pigments of certain species extract well, whereas others, notably Nannochloris sp., do not (Yentsch and Menzel, 1963). Thus this technique fails to detect an unknown and variable amount of plant pigment. It seems doubtful that the quantity missed ever amounts to more than 10 to 20 percent in the tropical regions sampled, but this assumption is unverified.

## PRIMARY PRODUCTION

Primary production was measured in this study to gain insight into the factors that regulate this process rather than to attempt semi-quantitative assessment of the productivity of the eastern tropical Pacific.

### Sample Collection and Incubation

The samples used in the photosynthesis studies were collected in polyvinyl chloride samplers shortly before local apparent noon, or in some in situ studies, one-half to three-quarters of an hour before dawn. The Pyrex incubation bottles, 125 ml., with ground-glass stoppers, were washed in detergent, rinsed three to five times in tap sea water and soaked in weak HCl-sea water (5-10 percent HCl by volume) for 2 to 3 minutes and rinsed again three to five times in tap sea water. Each incubation bottle was rinsed three times again with water from the sampler before the sample to be incubated was drawn from the plastic sampler.

In the vessel laboratory, 1.25 to 1.50 ml. of the sample was withdrawn from each sample bottle with a nylon syringe and 1.0 ml. of the  $C^{14}$  solution added to the bottle. The  $C^{14}$  was introduced well down into the sample with a nylon syringe and stainless steel hypodermic needle modified to deliver a known and constant volume (about 1.00 ml.) The stoppered bottles

were inverted gently several times to mix the  $C^{14}$  solution and sea water sample. The samples were then incubated.

At the end of the incubation period the samples were placed in a light-tight box and promptly filtered through 25.4 mm.-diameter HA Millipore filters. The filter and filter assembly were then rinsed with three washes (about 10 ml. each) of surface sea water and the filter pads placed in perforated labelled pill boxes. The pill boxes were stored in cans containing silica gel as a drying agent. After the return of the cruise, the sample filters were fumed over concentrated HCl for 15 to 20 minutes, redried in a vacuum desiccator over silica gel, and counted.

Three types of incubation were used to estimate primary production: (1) samples from one or more depths were incubated at sea surface temperatures under constant artificial illumination of  $1,000 \pm 100$  ft.-c. (foot-candles) for 4 hours (called laboratory incubation); (2) samples from one or more depths were incubated at the sea surface by towing the sample bottles astern of the vessel or were incubated at sea surface temperature in a deck incubator (surface or deck incubation); and (3) samples were incubated in situ at the collection depth between sunrise and local apparent noon or from local apparent noon until sunset (called in situ incubation).

The artificially illuminated incubator was patterned after one described by Steemann-Nielsen (1957). Illumination was provided from one direction only. Twelve 20-watt General Electric daylight-type fluorescent lamps provided the light source. Temperature was controlled by circulating surface sea water through the incubator at a rate of 4 to 12 liters per minute. The temperature of the water in the incubator was usually about  $1^{\circ}C$ . above the sea surface temperature, although differences occasionally were as great as  $2.5^{\circ}C$ .

The bottles were placed on a wheel or turntable which rotated slowly (three to six rotations per minute) for the duration of the experiment (generally 4 hours).

The illumination in the incubator was monitored frequently but at irregular intervals during each incubation series. The monitoring was facilitated by mounting an epoxy-resin-sealed barrier layer photocell (Weston YG 856) on the turntable in such a manner that the cell surface was located in the same plane as the half thickness of the incubation bottles. This cell was connected by gold-plated slip rings mounted on the turntable axle to a shunted 0- to 100- $\mu a$ . meter. The meter was installed on the incubator control panel. The cell and meter circuit were calibrated with a Weston Model 756 illumination meter (new candle model), with the incubator lamp bank as a light source.

Although the incubator production measurements are expressed in terms of milligrams

of carbon per hour at 1,000 ft.-c., the use of a foot candle notation is not strictly correct, since the emission characteristics of the lamps are rather different from the relative spectral sensitivity of the human eye. Furthermore, the use of illumination units implies wrongly that the plants themselves have a spectral response comparable to the human eye.

A more appropriate measure is irradiance (i.e., ergs per second per square centimeter). Illumination may be converted to irradiance with the following conversion factor:  $0.33 \times 10^{-5}$  erg/sec./cm.<sup>2</sup> is approximately equivalent to 1 ft.-c. This factor was obtained by making a direct comparison of the foot-candle output (with a Weston Illumination Meter) of the incubator light source, in the range 30 to 90 ft.-c., and the energy as measured with a vacuum thermopile and auxiliary equipment. This value is almost identical with that given by Strickland (1958). Most of the very near infrared (700-800 m $\mu$ ) was filtered out by placing a cell (3.2-cm. path) containing copper sulfate solution (10 g. CuSO<sub>4</sub> · 5H<sub>2</sub>O per liter in 0.5 percent H<sub>2</sub>SO<sub>4</sub>) between the thermopile and source. Longer wavelengths are not emitted in appreciable amounts from fluorescent lamps and thus would not introduce a sizeable error in the thermopile measurements (Withrow and Withrow, 1956, figs. 3-15). This solution was not used in the foot-candle measurement, since the photonic cell of the Weston Illumination Meter is not sensitive to wavelengths greater than 750 m $\mu$ .

The illumination level in the bath was adjusted to 1,000 ft.-c. by varying the lamp ballast voltage, and maintained at nearly this value ( $\pm 2$  percent) during the incubation period. As the fluorescent lamps aged with use, the light output dropped. The output of aging lamps could be raised to something in excess of 1,000 ft.-c. by switching in a second ballast to each lamp. The final adjustment to 1,000 ft.-c. was again made by adjusting a Variac which controlled lamp ballast voltage.

The equipment used during this investigation for surface and deck incubation has undergone several changes, and somewhat different methods have been followed on different cruises.

The first method used was to trail surface samples astern of the vessel immediately below the sea surface. The samples were placed in a brass frame holder designed to shade the bottles as little as possible. This technique was used on Expeditions EASTROPIC and SCOPE.

A more elaborate but similar piece of equipment was used on Expedition SCOT. This trailing incubator consisted of a plastic tube into which the sample bottles were inserted. The tube gave enough clearance to allow neutral photographic filters to be placed around a number of the bottles, which were separated

from each other by opaque spacers. Samples for incubation were collected at optical depths corresponding to the transmittance of the neutral filter series and covered with the appropriate filter during the incubation period. This unit was not entirely satisfactory because condensation of water vapor in the tube during towing damaged the neutral filters and changed their transmission characteristics.

Considerable care was used to prevent the trailing bottle assembly from bouncing on surface waves and in and out of the surface film. This control was achieved on different vessels by different handling methods which consisted of varying the length of towing line, or weighting the towing line with chains to keep the samples submerged about 0.5 to 1 m., or both.

A deck incubator replaced the trailing bottle after the SCOT Expedition. This unit was similar to that used by Bergé (1958). The incubator was cooled with surface sea water, and the compartments in it could be covered with neutral photographic filters of various densities. As previously, irradiance values were determined with the irradiance meter and samples for incubation collected at depths corresponding to the transmittance of the graded series of neutral photographic filters.

In situ productivity measurements were obtained whenever vessel time could be spared. Collection and inoculation techniques were identical with those described above. The sample bottles were fastened on a weighted cotton line at intervals corresponding to their collecting depths and lowered into the sea. The line was supported at the surface by plastic or glass floats, which also supported a bamboo pole bearing a radar reflector and an identification flag. After launching the assembly, the vessel left the immediate vicinity and proceeded with other observations. At the end of the incubation period, either local apparent noon or sunset, depending upon whether the samples had been launched within a half-hour of sunrise or local apparent noon, the buoy was retrieved and the samples were filtered.

Although this incubation technique is supposed to duplicate the natural system closely, certain difficulties prevent attainment of natural in situ photosynthetic rates. Shock from temperature, pressure, or light changes may result from bringing deep samples to the surface for inoculation with C<sup>14</sup> before the in situ incubation can proceed. The effects of these changes have not been evaluated in the present study, nor do other investigators appear to have examined this problem.

The technique as described presents an additional problem. The surface float system, even when glass floats are used, is nearly opaque; it is doubtful that estimates of surface sample rates can be considered reliable.



## C<sup>14</sup> Technique

The carbon 14 method described by Steemann-Nielsen (1952) has been used exclusively in these studies for the measurement of phytoplankton photosynthesis.

The C<sup>14</sup> was prepared from BaC<sup>14</sup>O<sub>3</sub> in the manner described by Steemann-Nielsen, except that sulfuric acid was the acidifying agent and that glass-redistilled water rather than artificial sea water was the solvent for the Na<sub>2</sub>C<sup>14</sup>O<sub>3</sub>.

Two strengths of C<sup>14</sup> were used. One contained about 8 to 9  $\mu$ c. (microcurie) per milliliter and the other about 1 to 2  $\mu$ c. per milliliter. These solutions were adjusted to a pH of 10.0 to 10.5, filtered through an HA or PH Millipore filter, and placed in 1-, 5-, and 10-ml. glass ampules. The ampules were sealed with cross-fire burners, inverted in a solution of dye, and autoclaved for 30 minutes at 15 lbs./in.<sup>2</sup> pressure. Any ampules with faulty seals were detected by the presence of dye in the C<sup>14</sup> solution and were discarded.

Sample activity was measured by a Nuclear Chicago Scaler (Model 161A) with an automatic sample changer (Nuclear Chicago, Model C110B) and Model D47 gas flow chamber equipped with a Micromil window. Each sample was counted until at least 1,280 disintegrations (usually 2,560) had been detected, including background (which averaged about 18 counts per minute). Two carbon 14 standards and at least two backgrounds were counted with each series of 30 samples. The activity of the standards was followed closely, and the samples re-counted if the instrument showed any mechanical aberration. The instrument was judged to have malfunctioned whenever the disintegrations per minute of one of the standard samples exceeded two standard deviations of the mean count of the standard. The mean value of the standard was determined from 50 separate measurements of the standard during which at least 5,120 disintegrations were detected. One would expect on the basis of chance alone to have this limit exceeded in about 5 percent of the cases. Nonetheless, samples were rerun if this 2-standard-deviation limit was exceeded, for such a deviation might indicate instrument malfunction.

Computation of carbon uptake from the tracer uptake was made in the conventional manner (Steemann-Nielsen, 1952). The carbon dioxide content of the water was assumed to be constant (90 milligrams per liter) at all stations. Corrections for the isotope effect were made on all samples but the respiration correction was not made. Generally the dark-bottle uptake was 10 percent or less than the light-bottle uptake, although the ratio of dark- to light-bottle uptake was sometimes appreciably higher, occasionally equaling 0.8 to 1.0. For instance, on SCOT Expedition (Blackburn et al., 1962), where this condition was particularly evident,

8 of 31 productivity stations showed a high ratio. The rather erratic appearance of such results and the fact that the uptake in surface and deep samples was somewhat high suggests that on some occasions washing and rinsing of the glassware left appreciable quantities of bacteria in the incubation bottles. In other situations SCOT Expedition stations 30, 74, 76, and 79) the ratio in the deepest samples was reasonable, and only the surface samples exhibited a high ratio. This erratic finding suggests that the occasional C<sup>14</sup> ampule contained particulate radioactive material.

Whenever the productivity index (light bottle C<sup>14</sup> uptake/chlorophyll a) fell in the range 2-8 per hour, the light-bottle data were used even if the dark-bottle uptake was unreasonably high.

The standardization of the C<sup>14</sup> was carried out by the method of Steemann-Nielsen (personal communication).

Standard solutions:

- (1) 0.1 M NaOH and 0.1 M NaNO<sub>3</sub>
- (2) 0.05 M Na<sub>2</sub>CO<sub>3</sub>
- (3) 0.1 M BaCl<sub>2</sub> · 2H<sub>2</sub>O

One to three ampules from the same batch of carbon 14 solution were taken at random and the contents mixed together. Generally 1 ml. of this solution was diluted to 100 ml. with distilled water, although the exact volume was varied somewhat according to the activity of the carbon 14 solution. Between 20 and 40 ml. of this diluted carbon 14 stock solution was placed in a 200-ml. volumetric flask. CO<sub>2</sub>-free distilled water was added to give a total volume of 60 ml. Next, 20 ml. of Standard Solution 1 were added, followed by 40 ml. of Standard Solution 2. The mixture was well shaken, and finally 80 ml. of Standard Solution 3 were added. The resultant precipitate was kept in constant motion with a magnetic stirrer during the preparation of the filters, and the flask kept tightly stoppered except during withdrawal of aliquots.

Known volumes of the precipitate suspension were withdrawn with wide-bore pipettes and added to weighed 25.4-mm.-diameter HA Millipore filters. The filter assembly was agitated by hand while suction was applied to help insure an even distribution of the precipitate on the filter surface. The precipitate was rinsed with 10 ml. of 0.001 M BaCl<sub>2</sub> · 2H<sub>2</sub>O while the filter assembly was again agitated. The filters were placed in labelled perforated cardboard pill boxes, and the boxes in turn were placed in a vacuum desiccator over freshly dehydrated silica gel. After drying (24-36 hrs.), the filters were weighed and the activity was measured with the counting equipment described above.

The volume of precipitate used in the standardization varied somewhat, but generally each of the 0.1-, 0.2-, 0.6-, and 0.8-ml. aliquots was duplicated, whereas 1.0-, 2.0-, 3.0-, and 5.0-ml.



samples were prepared in triplicate. The weight of the precipitate was calculated for volumes less than 1.0 ml. on the basis of the mean weight per milliliter of the precipitate as determined from weighing the heavier (i.e., >1 ml.) pads with an analytical balance.

The diameter of the filter assembly bore was measured, and the weight of precipitate per square centimeter of precipitate calculated. The activity (counts per minute) of the precipitate per milligram was also computed, and the results plotted on semilogarithmic paper, with the counts per minute per milligram as the ordinate (see fig. 5).

An eye-fitted curve was drawn through the points, and the counts per minute at zero weight defined by the point at which the extrapolated smooth curve intersected the ordinate (i.e., zero weight). Duplicate and triplicate determinations always differ, especially at low weights, but disagreement between zero-weight activity with different samples of the same batch of  $C^{14}$  solution has not exceeded  $\pm 12$  percent or been less than  $\pm 8$  percent of the mean value for a particular batch.

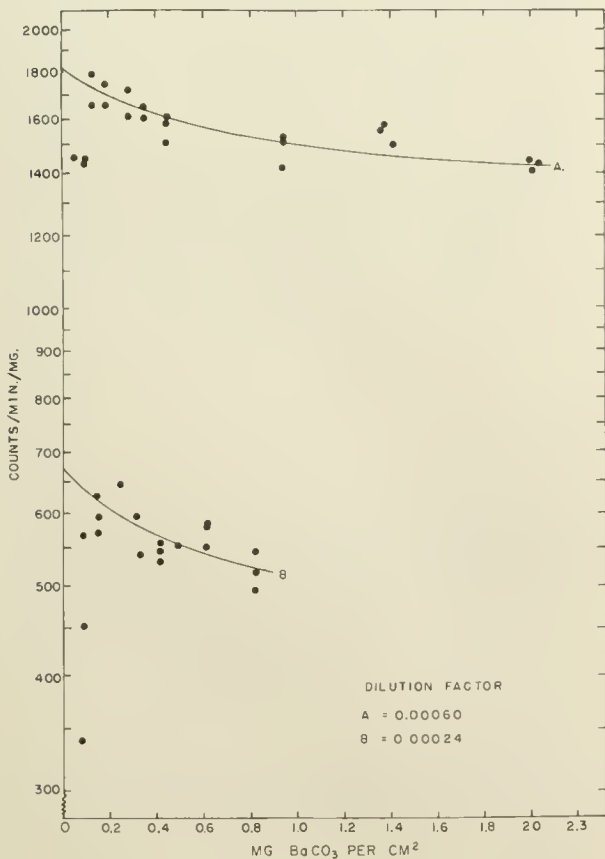


Figure 5.--Self-absorption curves of  $BaC^{14}O_3$  at two dilutions of  $C^{14}$ .

## Evaluation of Incubation Techniques

The purpose of trailing bottle, deck, and laboratory incubation was to obtain unbiased estimates of in situ production without using large amounts of vessel time. The final reference in these comparisons is, then, the in situ measurement itself. The unproven assumption has been made that the in situ observations yield rates which are similar to rates in nature; whether or not this assumption is correct remains to be shown.

Data for a comparison of these techniques are not abundant, and the inherent incompatibility of several of the techniques makes the following analysis somewhat indirect. The analysis is in the following order: (1) a comparison between in situ and laboratory incubator rates as applied to the isothermal layer and upper 100 m.; (2) a comparison between surface sample rates obtained by trailing bottle and deck incubation; and (3) a comparison between rates for surface in situ and deck incubated samples.

The data used to assess the agreement between in situ and laboratory incubator rates have been published elsewhere (Holmes et al., 1957; Holmes et al., 1958; Holmes and Blackburn, 1960, Blackburn et al., 1962). These data are considered from two points of view: the agreement between measurements in the isothermal layer; and the agreement between water column (i.e., 0-100 m.) production values.

Graphs were prepared of observed in situ and laboratory incubator production rates as a function of depth, and the points were joined with a smooth curve. The production values for the water column were obtained by integration with a polar planimeter from a depth of 100 m. to the surface. Isothermal layer rates were integrated in the same manner, on the same graphs; the depth of the thermocline was determined subjectively by visual inspection of the station bathythermogram. The results of these integrations are given in table 13.

The primary production rates, in situ and incubator, for both the isothermal layer and water column are positively correlated at a highly significant level (see table 13 for Spearman rank-difference coefficients,  $r_d$ ). Since the incubator rates are expressed on an hourly basis, they are not directly convertible to in situ rates. Laboratory incubator rates may be converted to approximations of in situ rates by multiplying laboratory rates by the average of the individual laboratory incubator in situ quotients, or by the quotient of the median ratio. The success of this manipulation may be judged by the Mann-Whitney U test (Siegel, 1956); in both series the smaller of the U values calculated, 170 and 177 respectively, have associated probabilities of greater

Table 13.--Comparison of the integrated rate of primary production in the isothermal layer and water column to a depth of 100 m. obtained in situ and with the laboratory incubator, 1961

Expedition and station	Isothermal layer <sup>1</sup>			Water column (0-100 m.) <sup>2</sup>		
	Incubator (at 1,000 foot candles)		In situ	Incubator (at 1,000 foot candles)		In situ
	Observed	Adjusted		Observed	Adjusted	
	Mg.C/m. <sup>2</sup> /hr.		Mg.C/m. <sup>2</sup> /day	Mg.C/m. <sup>2</sup> /day		Mg.C/m. <sup>2</sup> /day
SCOPE: 9C	10.0	100.0	150.0	29.0	270	330
9D	9.0	93.0	69.0	40.0	380	400
9F	6.8	70.0	69.0	31.0	280	380
20A	11.0	115.0	220.0	-	-	-
25B	20.0	210.0	180.0	24.0	220	200
SCOT: 23	3.8	39.0	36.0	12.0	110	78
49	10.0	100.0	66.0	28.0	260	240
56	3.0	31.0	21.0	11.0	100	130
62	20.0	210.0	210.0	31.0	280	290
88	0.9	93.0	8.0	-	-	-
EASTROPIC: 24	4.1	42.0	75.0	70.0	640	460
31-11	0.6	6.2	4.0	2.1	19	12
33	18.0	180.0	130.0	39.0	360	160
35	25.0	260.0	350.0	45.0	400	460
37	42.0	430.0	620.0	68.0	630	850
59-1: 37	9.0	93.0	55.0	14.0	130	140
42	17.0	180.0	60.0	34.0	310	100
46	69.0	710.0	780.0	79.0	730	950
51	30.0	310.0	280.0	34.0	310	314
Sum of ranks		381	360		301	294

<sup>1</sup> Isothermal layer:  $r_d = 0.90$ ,  $p < 0.01$

$U = 170$ ;  $p > 0.20$  (two-sided test)

<sup>2</sup> Water column:  $r_d = 0.82$ ,  $p < 0.01$

$U = 148$ ;  $p > 0.20$  (two-sided test)

than 0.20 for a 2-sided test; this result indicates that the transformed rates have essentially the same distribution as the in situ rates. A similar result (table 13) was obtained with the water column values and a conversion factor of 9.2 to convert laboratory incubator rates into situ estimates ( $U = 148$ ;  $p > 0.20$ --2-sided test).

Although this result may seem somewhat surprising, a closer examination of the data shows that any prediction of in situ production (water column or isothermal layer) based on laboratory incubator measurements would have a relatively large potential error of estimate. Indeed, this poor predictability is what one would expect. The samples in the laboratory incubator are exposed to constant light flux of a color and intensity different from the natural flux field at the respective in situ

depths. Furthermore, at the intensities used in the incubator, deep samples may experience saturation and perhaps inhibition effects that could further complicate the relation between in situ and laboratory incubator rates. These latter effects may explain the somewhat higher degree of correlation obtained between the isothermal layer data than between the water column data.

The correlation between the results from the different incubation techniques probably is not a chance event, and the scatter in the relation may reflect the difference in response of the plants to the different conditions of the two techniques.

Because of the inherent incompatibility of the trailing bottle and in situ incubation methods as practiced on our cruises, it is impossible to compare them closely. Trailing bottle and

deck incubation results may be compared, however, as well as deck incubation and in situ results.

The data used in this analysis (table 14) were collected by Forsbergh and others (Scripps Institution of Oceanography, University of California, 1961, STEP I Expedition, II) in the eastern tropical Pacific, by methods and equipment believed to be comparable to those of the present investigation. The data were selected from those of Forsbergh; the criterion for selection was that duplicate values be available for each type of incubation at each station. This criterion was selected because data from the present study were based on the mean of two sample values. The results of a

Table 14.--Primary production rates of deck incubated and trailed surface samples in the eastern tropical Pacific Ocean, STEP-I Expedition, 1960<sup>1,2</sup>

Date or station <sup>3</sup>	Trailing bottle	Deck incubator
September:	<u>mg.C/m<sup>3</sup>/day</u>	<u>mg.C/m<sup>3</sup>/day</u>
18	2.4	1.6
18	1.6	1.4
19	5.2	6.7
20	5.5	5.0
21	4.5	6.5
21	1.2	1.9
23	4.8	6.4
23	18.0	24.0
24	8.0	9.8
25	3.9	6.1
26	10.0	13.0
Station:		
10	7.6	11.0
15	3.5	4.4
16	0.6	0.4
19A	0.7	3.0
25	4.8	12.0
49	7.0	44.0
54	1.0	3.7
63A	0.5	0.6
68A	0.2	0.6
72A	0.4	2.8
76A	1.1	4.2
81A	4.5	16.0
Sum of ranks	473	608

<sup>1</sup> Scripps Institution of Oceanography, University of California, 1961, STEP I Expedition, Part II.

<sup>2</sup>  $r_d = 0.86, p < 0.01$

$Z = \frac{68}{\sqrt{2,071}} = 1.49, p = 0.14$  (two-sided test)

<sup>3</sup> Observations made in the period September 18-26 were not given station numbers.

statistical analysis of the data (table 14) were similar to those of the previous example, namely, the two types of measurements are significantly correlated ( $r_d = 0.86, p < 0.01$ ). The results of the Mann-Whitney U test (calculated as Z in table 14) indicate that the values were probably drawn from the same population.

As in the preceding example, results from these two techniques differ. The trailed samples received more agitation than the deck incubated samples. Although the spectral distribution of the radiant flux is probably nearly identical in the region of photosynthetic activity, the trailed samples received alternating or pulsating flux as a result of focusing of radiant flux by the surface waves and ripples. It was different in the deck incubator.

The comparison between deck incubator and in situ methods is drawn from six sets of data only. This number probably is not adequate, but since no other comparative material is available, the data have been used. The STEP-I data of Forsbergh and others and results of the statistical analysis are presented in table 15. These data show, as was true with the laboratory incubator-in situ comparisons (table 13), that deck incubator and in situ water column rates are significantly correlated and that no difference exists between the sampled populations ( $p = 0.82$ ).

Notwithstanding the difficulties that may be experienced occasionally with the C<sup>14</sup> method (such as high dark-bottle counts), the above data and analysis show that either the laboratory or deck incubator method provides data which may be used to estimate in situ productivity. Errors of estimate from these indirect methods are probably somewhat variable in time and space. Nonetheless, the two indirect methods provide useful information on

Table 15.--Simultaneously observed deck incubator and in situ production rates at stations, in the eastern tropical Pacific, STEP I Expedition, 1960<sup>1,2</sup>

Deck incubator	In situ
<u>Mg.C/m<sup>3</sup>/day</u>	<u>Mg.C/m<sup>3</sup>/day</u>
50	80
175	185
320	280
220	260
150	212
140	120

<sup>1</sup> Scripps Institution of Oceanography, University of California, 1961, STEP I Expedition, Part II.

<sup>2</sup>  $r_d = 0.95 < 0.02 > 0.01$

$U = 16, p = 0.82$  (two-sided test)



in situ rates and are useful substitutes for the time-consuming and costly in situ method.

In the present investigation deck incubation soon replaced the trailing bottle method of surface sample incubation. This change was a matter of convenience, since trailing bottle assemblies of the type employed required attention whenever the vessel slowed down or stopped. These two methods appear to yield comparable results (table 14), although a comparison of these methods with simultaneous in situ measurements would be worthwhile.

## SUMMARY

1. Incident solar radiation was measured on shipboard with a gimbals-mounted 10-junction Eppley pyranometer and potentiometric recorder. Daily radiation totals were obtained by integrating the unsmoothed daily records with a polar planimeter. The daily totals are believed to be accurate to within  $\pm 7$  to 10 percent. One hundred and four of the daily radiation totals were compared with estimates obtained from the climatological equations of Kimball, Savino-Ångström, Black, and Laevastu, by using multiple linear regression techniques. All these indirect methods yielded daily means which were significantly correlated with the measured values. The regression coefficients were all significant at the one percent level and only the Laevastu equation yielded an intercept value significantly different from zero. On cloudless days all of the climatological equations except that of Laevastu overestimated the daily total.

2. Submarine irradiance  $H$ ,

$$\text{where } H = \int_0^{\pi/2} N \cos \theta \, d\omega,$$

$\theta$  is the angle of incidence, and  $\omega$  the solid angle measure, was measured with a simple waterproof housing containing a Weston photo-voltaic cell, Wratten No. 45 filter, and a cosine collector. The attenuation or extinction coefficient ( $k$ ) was calculated from downwelling irradiance values at two depths  $z_1$  and  $z_2$  (both in m.;  $z_2$  deeper than  $z_1$ ) according to the following expression:

$$k = \frac{\log_e H_{z_1} - \log_e H_{z_2}}{z_2 - z_1}.$$

The outputs of the deck and submerged irradiance meter were read simultaneously at a given depth on electrically damped low resistance microampere meters. Corrections for departures in linearity of response of the detector and ambient irradiance were made at each depth at each station before calculating  $k$ .

Calculations show the detector has a maximum sensitivity at 490  $m\mu$  with a half-band width of approximately 63  $m\mu$ . These characteristics shift to 475  $m\mu$  and 50  $m\mu$  at 100 m. depth in Jerlov Type I ocean water. In Type III ocean water the peak remains at 475  $m\mu$  while the half-band width increases to 70  $m\mu$ .

A method is described for obtaining the percentage of incident "visible" energy at any depth in the upper 100 to 200 m., based on measurements of the attenuation coefficient at 475  $m\mu$  in Jerlov ocean water Types I, II, III, and intermediate types.

A statistical treatment of replicate determinations of attenuation coefficient under ideal conditions with negligible depth error yielded a coefficient of variation of 10 percent ( $k = 0.1805$ , standard deviation 0.0189). It is doubtful that this precision can be exceeded with the equipment described. Under adverse field conditions the precision is obviously much worse although measurements of this type have not been evaluated.

Errors in depth measurement cause irradiances to be assigned to incorrect depths;  $k$  values likewise will be in error and be assigned to incorrect depth intervals. Comparisons of depths computed from wire angle and length and depths measured with a precision depth sensor show the computed depth to be almost invariably less than the measured depth when wire angles exceed about 25 degrees. In the two examples discussed, these differences amounted to 5 and 8 m. with wire length 100 m. and wire angles of 40 to 45 degrees. Such differences become less as wire length is decreased but may amount to 1 or 2 meters with only 20 meters of wire. Depth intervals derived from indirect depth measurements can cause a  $k$  value to be 22 percent too low, as well as being referred to an incorrect depth interval.

3. Chlorophyll a was measured spectrophotometrically with dried extracts by using the equations of Richards with Thompson (1952). Pigment-bearing materials were collected on HA Millipore filters. Absorption at 750  $m\mu$  was subtracted from that at 665  $m\mu$  before calculating chlorophyll a concentration. Wavelength settings on the spectrophotometer were periodically checked by using chlorophyll a extracts or occasionally with didymium glass.

The variability in chlorophyll a under different sampling conditions was investigated. When certain statistical assumptions were used, it was found that the coefficient of variation for surface samples never exceeded 27 percent and most frequently was less. At depths between 10 and 40 meters the coefficient of variation increased appreciably. It is suggested on the basis of indirect evidence that a significant part of this variability results from the failure of the samplers to

collect water in exactly the same stratum on successive casts.

The significance of phaeophytin a and phaeophorbide and their influence upon "chlorophyll a" concentrations was discussed but could not be satisfactorily resolved with the available data.

4. The technique and equipment used to measure primary production with  $C^{14}$  are discussed in some detail. They follow rather closely those used by Steemann-Nielsen (1952). Between 1 and 20  $\mu$ c. per ml. of sterile  $C^{14}$  was stored in 5- to 15-ml. glass ampules. One or more ampules of appropriate  $C^{14}$  strength was used at each station; about 1 ml. of the  $C^{14}$  solution was added to the 125-ml. sea water sample with a constant delivery syringe. The activity of the  $C^{14}$  solution was measured by barium precipitation of the radioactive carbonate on tared Millipore filters. The standards, samples, and background were counted until at least 1,280 disintegrations had been obtained.

One of the main difficulties encountered during these investigations was the relatively frequent occurrence of high dark-bottle uptake. No completely satisfactory explanation could be found.

The following methods of sample incubation were employed: an artificially illuminated incubator maintained at temperatures slightly above sea surface temperatures; deck incubators; trailing incubators; and finally in situ incubations. Because two different techniques were generally used simultaneously, comparisons of the rates of  $C^{14}$  uptake between different methods could be made. Non-parametric statistical methods were used to show that the results of the different paired incubation techniques were significantly correlated. This correlation demonstrated that the different incubation techniques yielded unbiased estimates of in situ productivity. However, because the degree of correlation, even though statistically significant, varied appreciably, the accuracy of the extrapolation from incubator measurements to in situ productivity can be expected to vary with the method of incubation.

#### LITERATURE CITED

- BERGE, GRIM.  
1958. The primary production in the Norwegian Sea in June 1954, measured by an adapted  $C^{14}$  technique. In measurements of primary production, Cons. Perm. Int. Explor. Mer, Rapp. Proc.-Verb. Reün. 144: 85-91.
- BLACK, J. N.  
1956. The distribution of solar radiation over the earth's surface. Arch. Meteorol. Geophys. Bioklimatol. 7, Ser. B: 165-189.
- BLACKBURN, MAURICE, RAYMOND C. GRIFFITHS, ROBERT W. HOLMES, and WILLIAM H. THOMAS.  
1962. Physical, chemical, and biological observations in the eastern tropical Pacific Ocean: three cruises to the Gulf of Tehuantepec, 1958-59. U.S. Fish Wildl. Serv., Spec. Sci. Rep. Fish. 420, iii + 170 pp.
- BORTKOVSKII, R. S.  
1961. K metodike raschetov elementov teplovogo balansa poverkhnosti okeanov. (Concerning the method of calculating the constituents of the heat balance in the surface of the oceans.) Okeanologiiia, 1(3): 512-521. [English translation in Deep-Sea Res. 9: 362-370.]
- BUDYKO, M. I.  
1956. Teplovoi balans zemnoi poverkhnosti. (The heat balance of the earth's surface.) Gidrometeorologicheskoe Izdatel'stvo, Leningrad, 255 pp. [English translation by U.S. Department of Commerce, Weather Bureau, Washington, D.C., 1959.]
- BURDECKI, F.  
1958. Remarks on the distribution of solar radiation over the surface of the earth. Arch. Meteorol., Geophys. Bioklimatol., 8, Ser. B: 326-335.
- DOTY, MAXWELL S., and MIKIHICO OGURI.  
1957. Evidence for a photosynthetic daily periodicity. Limnol. Oceanogr. 2(1): 37-40.
- DRUMMOND, A. J.  
1961. Current developments in pyrhelio-metric techniques. Solar Energy 5(1): 19-23.
- FOSTER, NORMAN B.  
1951. A recording daylight illuminometer. Illum. Eng. 46(2): 59-62.
- HOLMES, ROBERT W., and MAURICE BLACKBURN.  
1960. Physical, chemical, and biological observations in the eastern tropical Pacific Ocean, SCOT Expedition, April-June 1958. U.S. Fish Wildl. Serv., Spec. Sci. Rep. Fish. 345, iii + 106 pp.
- HOLMES, ROBERT W., and ROBERT J. LINN.  
1961. A modified Beckman Model DU spectrophotometer for seagoing use. U.S. Fish Wildl. Serv., Spec. Sci. Rep. Fish. 382, iv + 4 pp.
- HOLMES, ROBERT W., MILNER B. SCHAEFER, and BELL M. SHIMADA.  
1957. Primary production, chlorophyll, and zooplankton volumes in the tropical eastern Pacific Ocean. Inter-Amer. Trop. Tuna Comm., Bull. 2(4): 128-169. [English and Spanish.]
- HOLMES, ROBERT W., and JAMES M. SNODGRASS.  
1961. A multiple-detector irradiance meter and electronic depth-sensing unit for

- use in biological oceanography. *J. Mar. Res.* 19(1): 40-56.
- HOLMES, ROBERT W., and others.  
1958. Physical, chemical, and biological oceanographic observations obtained on Expedition SCOPE in the eastern tropical Pacific, November-December 1956. U.S. Fish Wildl. Serv., Spec. Sci. Rep. Fish. 279, vi + 117 pp.
- HUMPHREY, GEORGE F.  
1960. The concentration of plankton pigments in Australian waters. C.S.I.R.O. Div. Fish. Oceanogr. Tech. Paper 9: 1-27.  
1962. Phytoplankton pigments in the Pacific Ocean. In Proceedings of the conference on primary productivity measurement, marine and freshwater, University of Hawaii, August 21-September 6, 1961. U.S. At. Energy Comm., Div. Tech. Inform., TID 7633: 121-141.
- JEFFREY, S. W.  
1961. Paper-chromatographic separations of chlorophylls and carotenoids from marine algae. *Biochem. J.* 80: 336-342.
- JENKIN, PENELOPE M.  
1937. Oxygen production by the diatom *Coscinodiscus excentricus* Ehr. in relation to submarine illumination in the English Channel. *J. Mar. Biol. Ass. U.K.* 22: 301-343.
- JERLOV, NILS G.  
1951. Optical studies of ocean waters. Repts. Swed. Deep-Sea Exped. 1947-48, vol. 3, Physics and Chemistry, 1: 1-59.
- JOHNSON, NILS G., and B. KULLENBERG.  
1946. On radiant energy measurements in the sea. *Svensk. Hydrogr.-Biol. Komm. Skr., Tredje Ser.: Hydrogr.* 1(1): 1-27.
- KIMBALL, HERBERT H.  
1928. Amount of solar radiation that reaches the surface of the earth on the land and on the sea, and methods by which it is measured. *Mon. Weather Rev.* 56: 393-399.
- LAEVASTU, T.  
1960. Factors affecting the temperature of the surface layer of the sea. *Soc. Sci. Fenn., Commentat. Phys.-Math.* XXV-1, 136 pp.
- McALLISTER, C. D.  
1963. Measurements of diurnal variation in productivity at ocean Station "P". *Limnol. Oceanogr.* 8(2): 289-292.
- McALLISTER, C. D., T. R. PARSONS, K. STEPHENS, and J. D. H. STRICKLAND.  
1961. Measurements of primary production in coastal seawater using a large-volume plastic sphere. *Limnol. Oceanogr.* 6(3): 237-258.
- MOON, PARRY.  
1940. Proposed standard solar-radiation curves for engineering use. *J. Franklin Inst.* 230: 583-618.
- PARSONS, T. R., and J. D. H. STRICKLAND.  
1963. Discussion of spectrophotometric determination of marine-plant pigments, with revised equations for ascertaining chlorophylls and carotenoids. *J. Mar. Res.* 21(3): 155-163.
- PATTERSON, J., and T. R. PARSONS.  
1963. Distribution of chlorophyll *a* and degradation products in various marine materials. *Limnol. Oceanogr.* 8(3): 355-356.
- PREISENDORFER, R. W.  
1960. Application of radiative transfer theory to light measurements in the sea. In Nils G. Jerlov (editor), Symposium on radiant energy in the sea. International Union of Geodesy and Geophysics, Int. Ass. Phys. Oceanogr. Monogr. 10: 11-30.
- RICHARDS, FRANCIS A., with THOMAS G. THOMPSON.  
1952. The estimation and characterization of plankton populations by pigment analyses. II. A spectrophotometric method for the estimation of plankton pigments. *J. Mar. Res.* 11(2): 156-172.
- SCRIPPS INSTITUTION OF OCEANOGRAPHY, UNIVERSITY OF CALIFORNIA.  
1961. STEP-I Expedition, preliminary report, II. Biological photometric and additional chemical data. *Progr. Rep., SIO Ref.* 61-18, 66 pp.
- SHIMADA, BELL M.  
1958. Diurnal fluctuation in photosynthetic rate and chlorophyll "a" content of phytoplankton from the eastern Pacific waters. *Limnol. Oceanogr.* 3(3): 336-339.
- SIEGEL, SIDNEY.  
1956. Nonparametric statistics for the behavioral sciences. McGraw-Hill, New York, 312 pp.
- SMITH, JAMES H. C., and ALLEN BENITEZ.  
1955. Chlorophyll *a*: analysis in plant materials. In K. Paech and M. V. Tracey (editors), *Moderne Methoden der Pflanzenanalyse* 4: 143-196. Springer-Verlag, Berlin.
- STEEMANN-NIELSEN, E.  
1952. The use of radioactive carbon ( $C^{14}$ ) for measuring organic production in the sea. *J. Cons.* 18(2): 117-140.  
1957. The chlorophyll content and the light utilization in communities of plankton algae and terrestrial higher plants. *Physiol. Plant.* 10: 1009-1021.
- STRICKLAND, JOHN D. H.  
1958. Solar radiation penetrating the ocean. A review of requirements, data and methods of measurement, with particular reference to photosynthetic productivity. *J. Fish. Res. Bd. Can.* 15(3): 453-493.



- TATE, MERLE W., and RICHARD C. CLELLAND.  
 1957. Nonparametric and shortcut statistics in the social, biological, and medical sciences. Interstate Printers and Publishers, Inc., Danville, Ill., 171 pp.
- TYLER, JOHN E.  
 1959. Natural water as a monochromator. *Limnol. Oceanogr.* 4(1): 102-105.  
 1960. Radiance distribution as a function of depth in an underwater environment. *Scripps Inst. Oceanogr., Bull.* 7(5): 363-412.
- TYLER, JOHN E., W. H. RICHARDSON, and R. W. HOLMES.  
 1959. Method for obtaining the optical properties of large bodies of water. *J. Geophys. Res.* 64(6): 667-673.
- VAN DORN, WILLIAM G.  
 1956. Large-volume water samplers. *Trans. Amer. Geophys. Un.* 37(6): 682-684.
- VERNON, LEO P.  
 1960. Spectrophotometric determination of chlorophylls and phaeophytins in plant extracts. *Anal. Chem.* 32(9): 1144-1150.
- WITHROW, ROBERT B., and ALICE B. WITHROW.  
 1956. Generation, control, and measurement of visible and near-visible radiant energy. In Alexander Hollander (editor), *Radiation biology, visible and near-visible light 3*: 125-258. McGraw-Hill, Inc., New York.
- YENTSCH, CHARLES S., and DAVID W. MENZEL.  
 1963. A method for the determination of phytoplankton chlorophyll and phaeophytin by fluorescence. *Deep-Sea Res.* 10: 221-231.
- YENTSCH, CHARLES S., and JOHN H. RYTHER.  
 1957. Short-term variations in phytoplankton chlorophyll and their significance. *Limnol. Oceanogr.* 2(2): 140-142.

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