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THE PROBLEMS OF APPLICATION OF RADIOCARBON METHOD FOR ESTIMATION OF PRIMARY PRODUCTION IN OLIGROTROPHIC WATERS

PROBLEMES METRODOLOGIQUES D'ESTIMATION DE LA PRODUCTION PRIMAIRE PAR LA METHODE ^{14}C DANS LES EAUX OLIGOTROPHES

D.K., KRUPATKINA*, B.R., BERLAND**, S.Y. MAESTRINI***.

* South Seas Biological Institute of South Seas
Academy of Sciences, YCCP, Sevastopol (URSS)
** Centre d'Océanologie de Marseille - U.A. - C.N.R.S. n 41.,
Station Marine d'Endoume, rue Batterie des Lions,
13007 MARSEILLE (FRANCE)
*** Centre de Recherche en Ecologie Marine et Aquaculture de l'Houmeau,
(CREMA), case 5, 17137 NIEUL-SUR-MER, (FRANCE).

In the past several years, the reliability of primary production values obtained by the standard radiocarbon method, has been maintained to the rather arguable. (Grézé, 1982 ; Sorokin, 1971). The data obtained by other methods indicate that the values are somewhat underrated (Eppley *et al.*, 1980 ; Jenkins, 1982 ; Kerr, 1983). The errors in primary production estimation by the radiocarbon method may also differ from one trophic region to another. Such supposition is based on unequal phytoplanktonic structural characteristics of these regions. These errors are likely to appear rather significant in oligotrophic waters predominated by nano- and ultranano- plankton (the so-called picoplankton).

According to scarce evidence, the leading role in the formation of organic substance in the ocean belongs to ultrananoplankton or picoplankton with the size of 0.2 - 2.0 µm. Picoplankton of oligotrophic waters is characterized by high growth rates (1.3 - 2.5 fissions day^{-1}) and high biomass (70 per cent) and chlorophyll content (80 per cent) (Bienfang et Takahashi, 1983 ; Takahashi et Bienfang, 1983). It has been also found that the picoplankton contribution to primary production is rather substantial, totalling 20-80 per cent. Moreover, this contribution is especially high (50-80 per cent) in the lower layers of the euphotic zone (Li *et al.*, 1983 ; Platt *et al.*, 1983).

Since the organic substance of oligotrophic waters is obviously produced by picoplankton for the most part, the need for clarifying possible errors when estimating primary production by the radiocarbon method, which is the purpose of the present work is urgent.

For these topics in oligotrophic waters, we carried out measurements, in two ways : (1) according to the generally accepted pattern (Sorokin, 1956, 1958) and (2) with the use of methodical experiments aimed at modifying that pattern.

Primary production value estimation according to the generally accepted scheme established by Yu. I. Sorokin, has revealed extremely low values both in the surface layer (0.24.50 mg $C.m^{-3}$. day^{-1}) and below 1 m (13.25 mgC. m^{-2} day^{-1}). In situ measurement has also shown extremely low values of the primary production in the surface layer (0.56 mg $C.m^{-3}$. day) and below 1 m (46.3 mg $C.m^{-2}$. day^{-1}).

The experiments revealed a number of factors reducing the total primary production value. For example, the solution of unpurified isotope $NaH^{14}CO3$ produces an inhibiting effect on the primary production (Fig.1). Since the production of oligotrophic waters was considered to be low, then, in order to obtain reliable results, a high isotope concentration was usually employed (20-30 μCu per 0.25 l of water). However, we have found that at such a concentration, primary production was underrated by 30-40 per cent. This result may be explained by the presence of heavy metals in the isotope (Carpenter and Lively, 1980 ; Fitzwater *et al.* 1982). This supposition was substantiated by comparing the primary production values obtained as a result of using both purified and unpurified isotopes (Fig.2) : in the case of the unpurified isotope, primary production was dramatically underestimated. We have also found that isotope impurities unequally affect the production of different size fractions of phytoplankton (Table 1). It is interesting to note, that after a two hour exposure, the highest growth of the primary production was registred in the smallest fraction (0.45-0.85 μm). Obviously, the isotope impurity produces the maximum effect on this fraction within the first hours of exposure.

Absorption of ^{14}C by different size fractions of phytoplankton in the daytime has shown that the highest contribution to the total primary production within the first 1-4 hours corresponds to the smallest fraction (0.45-0.85 μm). The share of larger fractions becomes significant 8 hours after exposure.

The contributions of different size phytoplankton fractions to the total primary production at different depths is shown in figure 3. The contribution of the smallest fraction (0.45-0.85 μm) amounted to 35-80 per cent. The tendency of this contribution to increase with depth was also registered (Fig.3, L). The share of the rest did not exceed 25 per cent. For the samples exposed in the dark (Fig.3, D), the contribution of all fractions was approximately the same (15-25 per cent).

A comparison of the primary production rates measured in vials of different volumes has shown (Table 2) that the production value in large vials (1 l) is about 30 per cent higher than in small (0.25 l) ones.

The above experiments led to the developpement of a modified version of the radiocarbon method. Essentially, the modification included the use of large-volume bottles (2, 3 l), low isotope concentration (30 $\mu Cu.\ l^{-1}$), and short exposure (3-4 h) soon after sampling the water and under conditions of low light intensity and temperature. The measurement of the primary production carried out in accordance with this modified version has made possible the establishment of primary production values of : 200-300 mg $C.m^{-2}$. day^{-1}.

The radiocarbon method of the primary production estimation is known to consist of four stages : (1) water sampling at different depths and filling the incubation bottles, (2) adding to the latter the $NaH\ ^{14}CO_3$ solution, (3) exposing the bottles to light, and (4) filtering the water under low vacuum. In our opinion, the most significant role in the underestimation of primary production is played by the fact that different losses of the smallest phytoplankton fractions occur, due to the use, at the fourth stage, of filters with large pores (0.85 μm). According to our

results, these losses in oligotrophic waters lead to substantial (up to 80 per cent) underestimation of the primary production values. Possibly, these losses increased because high vacuum filtration (up to 0.6- 0.8 atm) was employed.

Furthermore, the errors in primary production estimation are associated with indirect losses caused by the destruction or deterioration of picoplankton. One of the reason consists in the little volume of standard bottles (0.25 l) used. According to our comparisons of primary production values in oligotrophic waters, large bottles (1 l) yielded more production (30 per cent more) than the small ones. This phenomenon is also explained by the fact that the picoplankton is likely to the damaged in a closed, restricted volume (Carpenter, Lively, 1980 ; Gieskes *et al.*, 1979). Next, the standard method requires the sea-water samples to be kept exposed to light for 6 hours and more. But our primary production experiments proved that with an exposure exceeding 3-4.0 h, the primary production value decreases by 20-40 per cent. This fact is explained by the maximum contribution of the smallest fraction (0.45 - 0.85 μm) within the first 4 hours. Finally the standard method normally required a high isotope concentration (20-30 μCu per 0.25 l of water). Our comparisons of the primary production in bottles with different isotope concentrations showed that with the above concentrations the primary production value decreased by 30 - 40 per cent. Most likely, this drop may be explained by the presence of heavy metals in the isotope.

Another source of errors occurring in the estimation of primary production value by the standard radiocarbon method is high temperature, light intensity, and excessive duration of the bottle exposure. Although the natural light used for bottle exposure on the ship's deck is supposed to be repeatedly reduced by the mill gas, the bottles are nevertheless exposed to the effect of 20-30 thou. lux illumination and a flow-through water temperature of 25-27°C for 4, 6, and even 12 hours. When the bottles are exposed for shorter periods (3-4 h) in the laboratory environment with lower light intensity (10-12 thou. luxes) and a temperature of about 20°C, it becomes clear that under the described standard conditions the primary production value was underrated by 20-40 per cent.

Thus, our experiments have demonstrated that the standard radiocarbon method shows underrating at all levels of primary production estimation. The difference in the results obtained by the standard radiocarbon method and our method, which make allowance for all the above-mentionned errors, was significant. In the first case, the primary production value constituted 50 mg $C.m^{-2}.day^{-1}$; in the second one, 200-300 mg $Cm^{-2}.day^{-1}$.

The radiocarbon method originated in 1951 and was immediately widely recognized. But today, we have to admit, however, that its application in the case of oligotrophic waters results in the underestimation of the primary production values since it ignores picoplanktonic production. Many of the disadvantages associated with the use of this method were revealed more than 10 years ago by Sorokin (1971). Now, it is quite evident that the radiocarbon method requires substantial modification. Its improvement will provide more reliable results of oligotrophic water production values and allow the correlation of the evidence obtained earlier with the findings of today. However, the radiocarbon method, no matter to what extent it may be modified, would fail to give a comprehensive estimation of the organic substance formation in the ocean, because the principles underlying the structure of the ^{14}C flow are too primitive (Peterson, 1980). The disbalance problem has to be solved not only with the elimination of methodical errors, but rather on the basis of a full-scale consideration of the carbon flow within the oceanic ecosystem. Along with the radiocarbon method, positevely characterized by its re-

latively simple and quick- acting structure and high sensitivity, primary production should also be estimated by other methods involving such characteristics as cellular fission rate, light absorption by chlorophyll, amount of detritus depositing from the euphotic zone, and destruction of organic substance.

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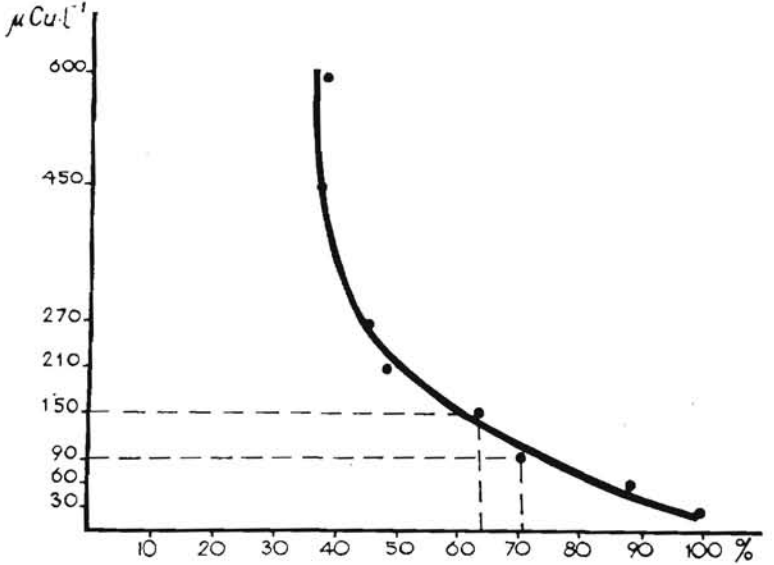


Figure 1 - Effect of 14C (μCu.l-1) on primary production of oligotrophic waters. Dotted line denotes concentration of 14C normally added to glass bottle of 0.25 l volume (20-30 μCu) and percentage of the primary production reduction (30-40 per cent).

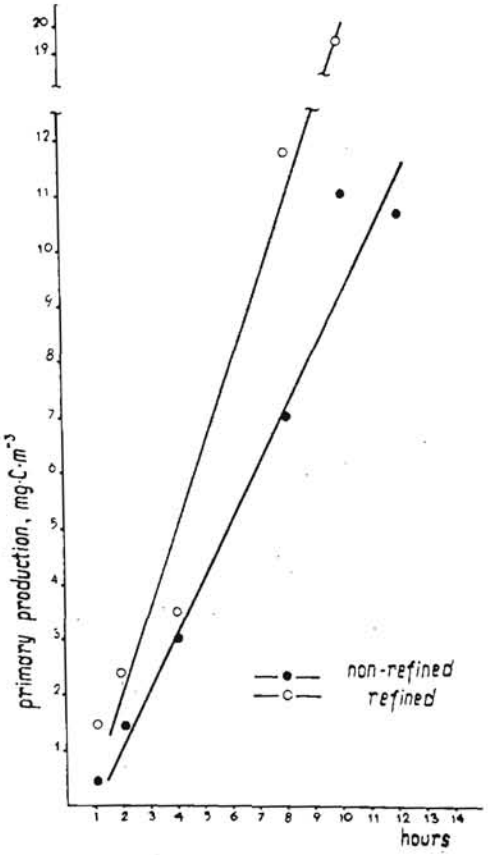


Figure 2 - Primary production of oligotrophic waters (O m) with the use of purified and unpurified isotope $NaH^{14}CO3$.

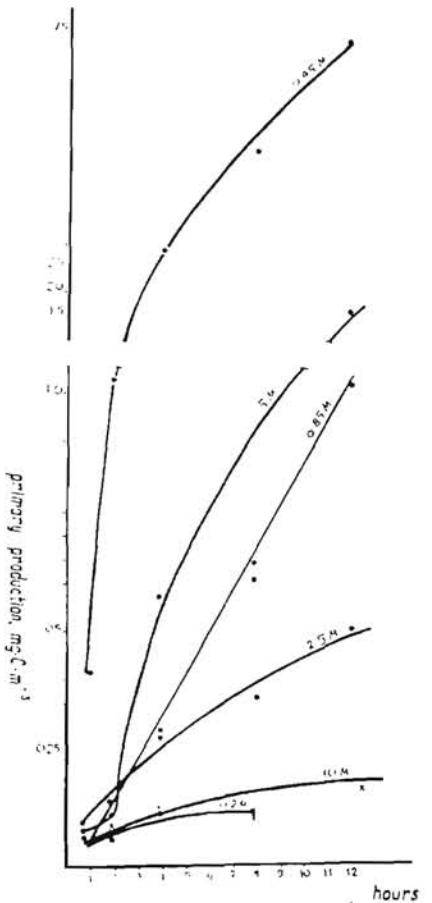
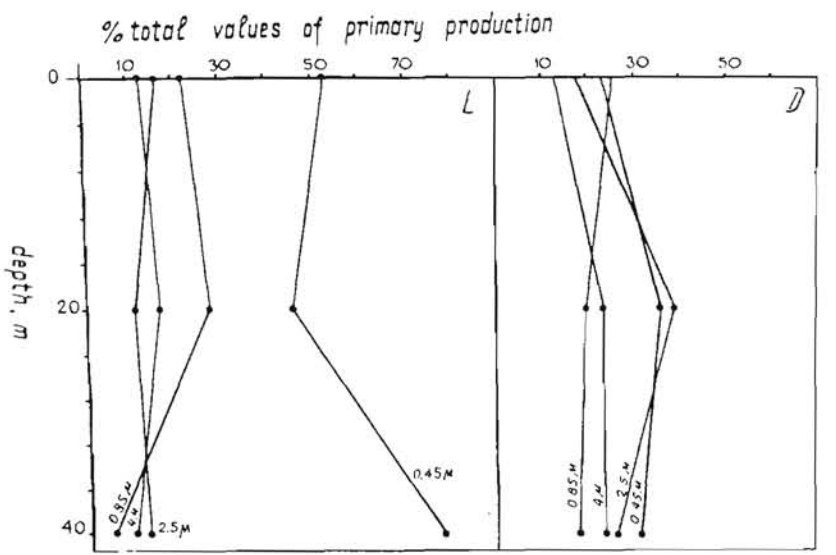


Figure 4 - Contribution comparison of different phytoplankton size fractions to the total primary production value at various depths (Davis Bank). L - bottles exposed in the light, D - bottles exposed in the dark.

Figure 3 - Absorption of 14C by different size fractions of phytoplankton (0 m) of oligotrophic waters during the day-time.



Exposure of samples, h	Size fractions, μm			
	>0.45-<0.85	>0.85-<2.5	>2.5-<4	>4
2	179.6	93.9	113.0	143
4	127.8	240.7	177.6	100.0
8	161.0	166.8	189.2	190.5

* In the case of unpurified isotope, the primary production value is taken as 100 per cent.

TABLE I - EFFECT OF ISOTOPE $NaH^{14}CO_3$ CONTAMINATION (%) ON PRODUCTION OF BASIC SIZE FRACTIONS OF PHYTOPLANKTON (0 m) IN OLIGOTROPHIC WATER (ATLANTIC OCEAN) *

Depth, m	Bottle volume	Primary production
0	Large (L)	1.99
	Small (S)	1.26
	(L/S, %)	62.3
20	Large (L)	0.51
	Small (S)	0.37
	(L/S, %)	72.5
0	Large (L)	1.86
	Small (S)	1.30
	(L/S, %)	58.8

* Large bottle are made of polyethylene; small ones, of polycarbon. Sample volume (1 l) filtered by one filter was equal for large (1 l) and small (0.25 l x 4) bottles.

TABLE II - EFFECT OF LARGE VOLUME (2, 3 l) AND SMALL VOLUME (0.25 l) OF INCUBATION BOTTLES ON PRIMARY PRODUCTION (mg $C.m^{-3}.day^{-1}$) OF OLIGOTROPHIC WATERS (MEDITERRANEAN SEA) *